A novel allergen-encoding immunotherapeutic vaccine in the treatment of peanut allergy

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Peanut allergy: high prevalence in Australia
3% of children are peanut allergic
Of which, 80% remain allergic for life

By 2025:
- Projected 8 million peanut allergic individuals in the 7 major markets
- Projected market size of 11.3 billion USD
“This incredible growth stems from the projected entry of four new peanut immunotherapy products into a previously empty marketplace. These new therapies include three oral immunotherapy (OIT) products—Aimmune Therapeutics’ AR-101, Prota Therapeutics’ PPOIT, and Camallergy’s CA-002—and one epicutaneous immunotherapy product—DBV Technologies’ Viaskin Peanut.”
Current treatment: total avoidance
If you are exposed: EpiPen adrenaline

**EPIPen Price Under Mylan**

- **Source:** Truven Health Analytics

The more specific the prescription, the greater the likelihood the pharmacist will fill it as intended.

Prescribe an EpiPen 2-Pak® for each location where your patient may need immediate access:

- **HOME**
- **SCHOOL**
- **WORK**
- **GYM BAG**
Desensitization-oral immune therapy

- Intense and costly administration of allergen
- Requires maintenance
- Benefit not a long term solution
- This treatment has safety concerns and is not approved by the FDA
- Sub lingual oral immunotherapy at home, 3 years duration (dust mite and pollen) compliance as low as 7%
Permanent desensitization by re-educating the allergic T cell response with a vaccine?

Allergic status

Th1 Th2

Non allergic status

Th1 Th2

Desensitization
tolerance

Mast cells and basophils

Antigen-presenting cell → CD4 T cell

Th2 molecules

IL-4

IFN-γ

B cells

Peanut-specific IgE

Allergic response
The propriety ‘Sementis Copenhagen Vector’ platform technology developed with Sementis Ltd.

**Vaccine Delivery Vehicle (SCV Vector):**

“Genetically crippled smallpox vaccine that can be engineered to make ANTIGENS from disease targets to raise immunity to that disease”

**Totally attenuated vaccine vector system**

**Manufacturing Cell Substrate:**

“The CHO biotechnology friendly cell substrate engineered to produce the SCV vector”

**A first for the production of vectored vaccines!**

Declared COI: JD Hayball holds shares in Sementis Ltd and sits on the SciAdBrd
How does the SCV platform work?

**Smallpox Vaccine**
(Live replication competent)

- Vaccination
- Viral genome replication (gene amplification)
- HIGH level antigen production
- Viral assembly
- Progeny virus
- Amplified infection cycle
- Vaccine complications (death rate 1-in-million)

**SCV Vaccine**
(Live NON-replication competent)

- Vaccination
- Viral genome replication (gene amplification)
- High level antigen production
- Arrested progeny virus production

*Note: Aim to preserve genome replication*  
*That leads to gene amplification*  
*That gives excessive antigen production!*
SCV does not multiply in human and mammalian cells lines

<table>
<thead>
<tr>
<th></th>
<th>Vaccinia</th>
<th>SCV</th>
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<tr>
<td>143B</td>
<td>Human Bone Cells</td>
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Safety and biodistribution of SCV in immunocompromised SCID mice

Average Body Weights ± SEM
(n=9 per treatment group)

In the absence of an antiviral immune response SCV was unable to cause productive disease.

Survival Plot
(n=9 per treatment group)

In the absence of an antiviral immune response SCV is not pathogenic.
Sementis’ SCV-cell substrate for manufacturing was derived from GMP produced CHO-S cell line:

- Sourced as a GMP produced batch of CHO-S from Life Technologies (ThermoFisher Scientific), Cat # A1136401, royalty free, one off licence fee per field, ie, infectious diseases, immunotherapeutics
- Suspension cell line – suitable bioreactor production
- Cultured in serum-free chemically defined medium, eg, CD-CHO medium from Life Technologies, Cat # 10743029
An effective single shot SCV-CHIKV+ZIKV vaccine

Vaccination

Viral genome replication (gene amplification)

High level CHIKV-antigen production
High level ZIKV-antigen production

STIMULATION of anti-ZIKV Immune response
Enhancing of anti-ZIKV Immune response
Non-infectious ZIKV-VLP

Arrested progeny Virus production

STIMULATION of anti-CHIKV Immune response
Enhancing of anti-CHIKV Immune response
Non-infectious CHIKV-VLP
Single-shot SCV-CHIKV+ZIKV vaccination protects against infection with both diseases

- C57BL/6 mice
  - Protection against viraemia

![Graph showing protection against viraemia for C57BL/6 mice.]

- IFNAR mice
  - Protection against viraemia

![Graph showing protection against viraemia for IFNAR mice.]

Protection against CHIKV arthritis

![Graph showing protection against CHIKV arthritis.]

Protection against lethal ZIKV_{MR766}

![Graph showing protection against lethal ZIKV_{MR766}.]
A single shot vaccination protects against ZIKV detrimental foetal outcomes

**a**

<table>
<thead>
<tr>
<th>SCV vaccination</th>
<th>Anti-ZIKV ELISA responses (b)</th>
<th>Mating initiation</th>
<th>Plug detection</th>
<th>ZIKV&lt;sub&gt;Natal&lt;/sub&gt; challenge determinations (c)</th>
<th>Viremia determinations (c)</th>
<th>Fetal/placental assays (d)</th>
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<tr>
<td>Week 0</td>
<td>Week 4</td>
<td>Week 6</td>
<td>E0.5</td>
<td>Days 1-5 post infection</td>
<td>E17.5</td>
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**b**

Anti-ZIKV

**c**

Reciprocal end point ELISA titer x1000

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<th>Day post-infection</th>
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<td>4</td>
</tr>
<tr>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>6</td>
<td>ND</td>
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**d**

SCV-ZIKA/CHIK

SCV-cont

*
A single shot vaccination protects against ZIKV testicular damage
• Implavax technology for SCV delivery
  ▫ Enhanced stability?
  ▫ Enhanced immune responses?

• Stage 1: Formulation of SCV into solid doses (Enesi)
• Stage 2: Immunogenicity study (Sementis)
  ▫ CHIKV/ZIKV vaccine
  ▫ Peanut vaccine
Summary I

- SCV is replication-defective *in vitro* and safe *in vivo*
- Commercially-proven CHO cells for scalable vaccine production
- SCV-CHIKV+ZIKAV vaccination elicits single shot and long term protective immune responses
  - Protects pregnant mice and their offspring from ZIKAV infection
  - Protects testis of male mice from ZIKAV mediated damage
  - No immune interference between CHIKV and ZIKV vaccine antigen expression nor booster responses (*data not shown*)

- Eldi *et al*, Molecular Therapy, 2017
- Prow et al, Expert Review Vaccines, 2019
Permanent desensitization by re-educating the allergic T cell response with a vaccine?

1. Induction of Th2 biased peanut-specific T cells
2. IgE production

- Low Th1 cytokine (IFN-γ)
- Increased Th2 cytokine (IL-4, IL-5, IL-13)

3. Mast cell priming with IgE
4. Mediator release leading to allergic symptoms

Peanut exposure
Can an SCV-based vaccine skew a peanut allergy-specific Th2 responses to a Th1 bias?

SCV-PHAV expresses a ubiquitinated, multi-peanut antigen fusion protein expressing:
Ara h 1, 2, 3, 5, 6, 7, 8, 8.1, 9, 10, 11
Confirming vaccine mechanism of action using APC and total T-cells from blood of a peanut allergic individual

<table>
<thead>
<tr>
<th>Patient ID</th>
<th>Age (yrs)</th>
<th>Skin prick test results</th>
<th>sIgE (peanut)</th>
<th>Basophil activation test</th>
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<tr>
<td>1</td>
<td>25</td>
<td>6mm</td>
<td>6</td>
<td>positive</td>
</tr>
<tr>
<td>2</td>
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<td>0.85</td>
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</tr>
<tr>
<td>3</td>
<td>33</td>
<td>6mm</td>
<td>4.9</td>
<td>positive</td>
</tr>
<tr>
<td>4</td>
<td>23</td>
<td>Strong +</td>
<td>71.3</td>
<td>positive</td>
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<td>25</td>
<td>7mm</td>
<td>0.9</td>
<td>positive</td>
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</table>

Ratio of secretion is a measure of Th1/Th2 bias induced by peanut allergen exposure to vaccinated-APC activated T-cells
Th1/Th2 profiles for SIX peanut allergic volunteers

Peanut Allergic Volunteer: MD (female)
Ex Vivo Th1/Th2 Profile

Peanut Allergic Volunteer: JD (female)
Ex Vivo Th1/Th2 Profile

Peanut Allergic Volunteer: AB (female)
Ex Vivo Th1/Th2 Profile

Peanut Allergic Volunteer: MA (male)
Ex Vivo Th1/Th2 Profile

Peanut Allergic Volunteer: SN (male)
Ex Vivo Th1/Th2 Profile

Peanut Allergic Volunteer: KD (female)
Ex Vivo Th1/Th2 Profile
Vaccine inducing Th1 efficiency in a sample population of 6 peanut allergic volunteers

The Mean Ex Vivo Vaccination Induced Th1/Th2 Profile of 6 peanut allergic Volunteers

**Conclusion:**
1. The **peanut hypoallergy vaccine** treated DCs induces a significant increase in a peanut-specific Th1 response over and above the T-cells treated with **PNE-treated DCs** (peanut protein extract).

2. The **peanut hypoallergy vaccine** treated DCs also induced a significant increase in a peanut-specific Th1 response over and above the T-cells treated with **SCV-vector only DC**.

*Significance was determined using one-tailed Mann-Whitney T-test
Significance = p<0.05*
A mouse model for food allergy

- C3H/HeOuJ mice
- Oral (gavage) dosing of allergen: Peanut, casein or whey with the mucosal adjuvant cholera toxin (CT)

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<tr>
<th>Day</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>7</th>
<th>14</th>
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- Oral dosing allergen with or without CT

- Early parameters like allergen-specific cytokine production by spleen
- Weekly blood collection, serum ELISA’s (antibodies against allergen)
- Allergen-specific cytokine production in the spleen

- Oral challenge
- Sacrifice

Clinical parameters upon oral challenge with allergen:
- Acute allergic skin response
- Intestinal permeability
- Mast cell degranulation
- Diarrhea
- Intestinal motility
Peanut sensitization

vaccination

boost

Antibody analysis

Absorbance units

Reciprocal of serum dilution

Peanut-specific endpoint titres (x1000 + SE)

IgG2c IgG1 IgE

Peanut-specific endpoint titres (x1000 + SE)

IgG2c IgG1

Sensitized Prophylactic vaccination Sensitized Prophylactic vaccination

Total IgE levels (ng/ml)

Sensitized Prophylactic vaccination
Ex-vivo stimulation antigen specific CD4 T cell cytokine analysis

Gated on live proliferating CD4+ cells

IFN-γ

IL-4

% of proliferating cells (peanut specific)

Th1 / Th2 ratio

# IL-4 producing cells (x10⁴)
Summary II

• SCV-PHA *ex vivo* vaccination induces Th1-skewed response in a sample population of 6 peanut allergic volunteers.

• The same Th1-skewed response pattern is observed in the same donor when tested three times over a 12-month period.

• SCV-PHA *in vivo* vaccination vaccination delivers a skewed Th1 vsTh2 response in peanut allergic mice.

• Eldi *et al.*, manuscript in preparation.
Experimental Therapeutics Laboratory

Dr Paul Howley, Sementis Ltd., Vic.
Prof Andreas Suhrbier, QIMR Berghofer, Qld.
Dr William Smith, Royal Adelaide Hospital, SA.
Dr George Lovrecz. CSIRO, Clayton, Vic.
What happens to the peanut-specific antibody levels post-sensitization in vaccinated mice?

**Th1 response:** Sensitization boosts Th1 response in vaccinated mice

**Th2 response:** Sensitization does not induce IgE in vaccinated mice

IgG1 response similar to sensitized mice
Does vaccination prevent mast cell degranulation following peanut exposure?