The influence of the food matrix on allergenicity*

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April 11, 2012

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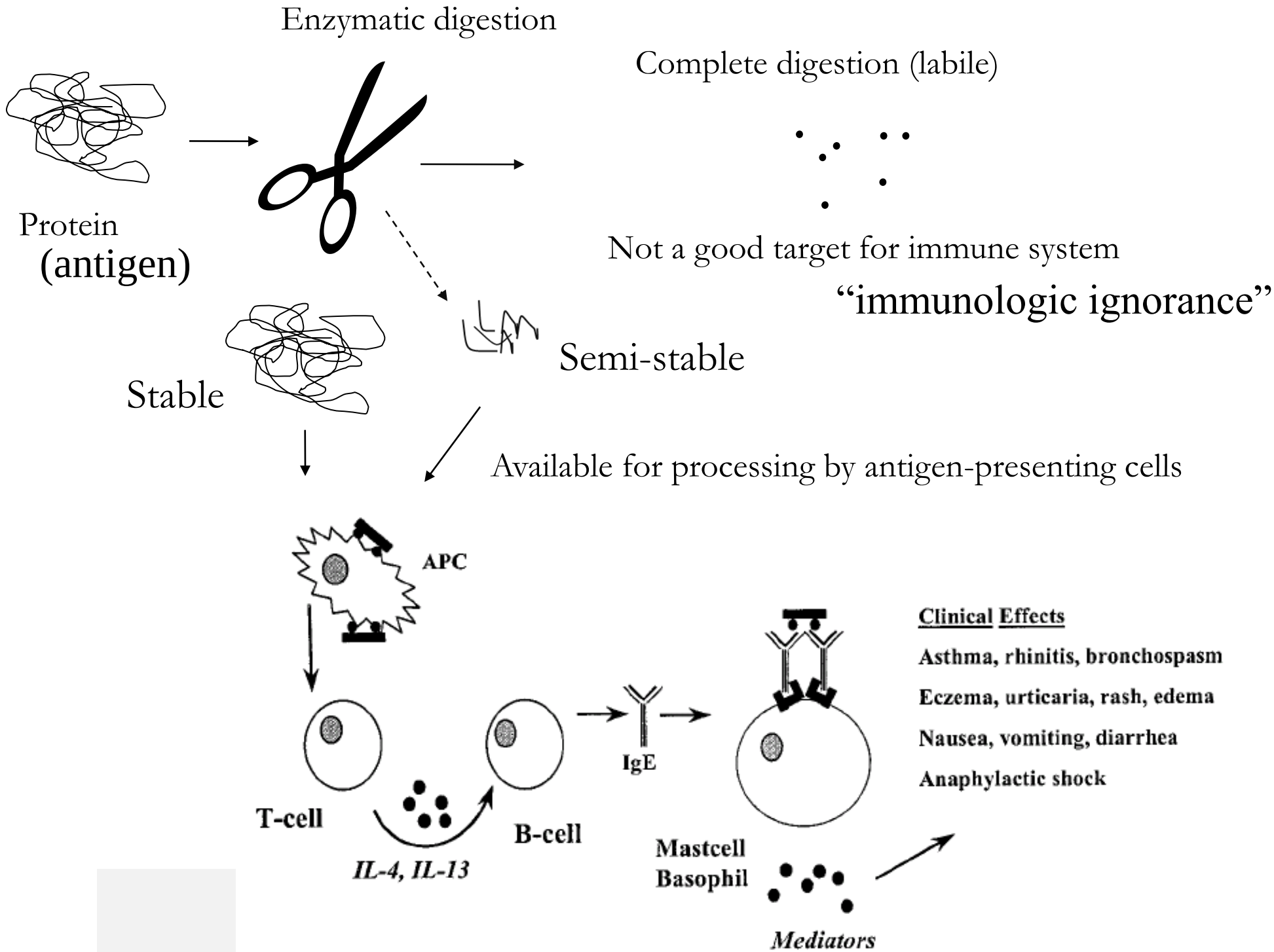
Food matrix issues can affect:

- Detection and quantitation of proteins
- Processing alterations
- Digestibility assays in vitro
- Digestion and absorption in vivo
- Interactions with the immune system

Matrix effects: Digestibility



Dong et. al, TOXICOLOGICAL SCIENCES 73, 8–16 (2003)

Protein content

- High protein meals can overwhelm digestive enzyme levels
- Matrix may contain proteins with specific enzyme inhibition activity

Plant structures

- Cell walls may not be broken down in the upper GI tract
- Nut protein bodies – dry, relatively insoluble – hydration status may be deciding factor in allergenicity of some of these

Lipids

- In vitro pepsin digestion of α-lactalbumin slowed by phosphatidylcholine (abundant surfactant in milk and produced by the stomach) – Moreno et al. 2005 J Agric Food Chem
- Phosphatidylcholine did not affect pepsinolysis of β-lactoglobulin, but protected from duodenal enzymes – Mandalari 2009 Mol. Nutr. Food Res.

Polysaccharides

- Gum arabic, pectin, or xylan at 50 wt% enhanced peptic digestion of β-lactoglobulin. Interaction with polysaccharides reduced protein aggregates/increased solubility – Mouecoucou et al. 2004, BBA
- Affects the size of peanut peptides obtained by two-step hydrolysis – Mouecoucou et al. 2004, Clin Exp Allergy

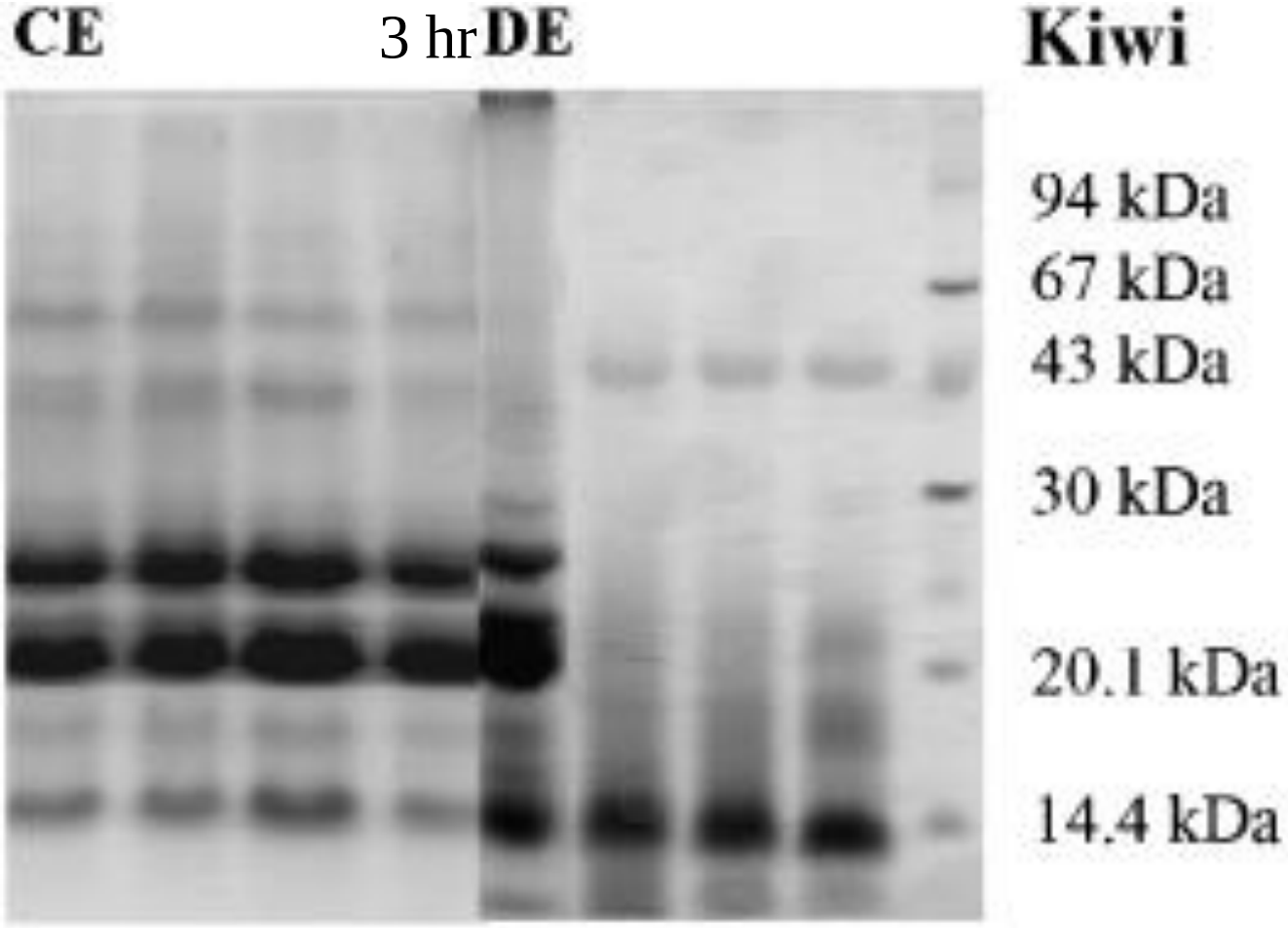
Polysaccharides cont'd

Pectin forms a gel at low pH

Effects of 3% or 6% pectin on fruit protein digestibility: -- Polovic et al. 2007 Clin. and Exp. Allergy.

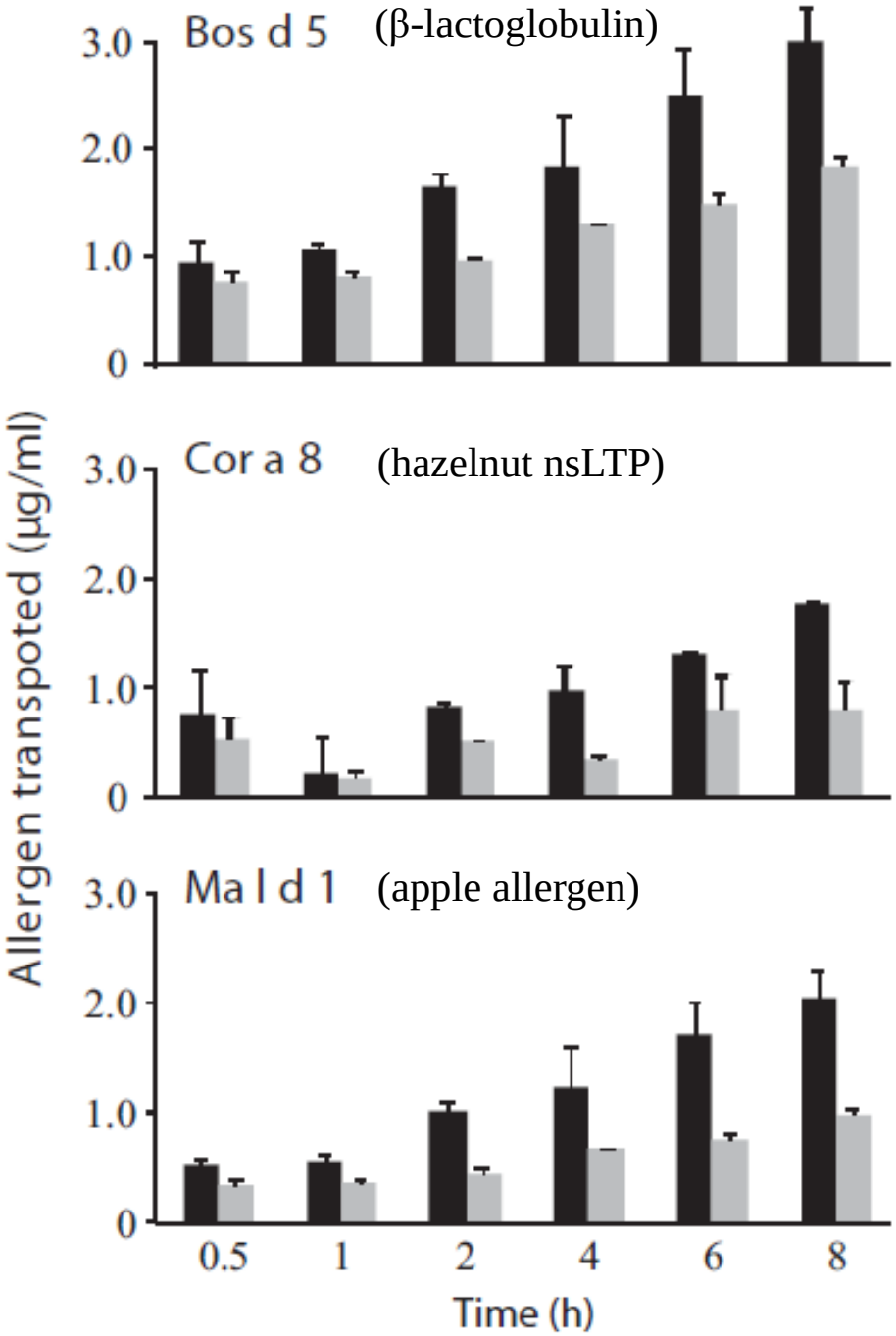
In vitro digestion of crude and dialyzed kiwi extracts

Crude extract (CE) and extensively dialyzed extract (DE) of kiwi fruit in simulated gastric fluid



Polovic et al. 2007 Clinical and Experimental Allergy

The presence of a food matrix significantly impedes the transepithelial transport of allergens in vitro



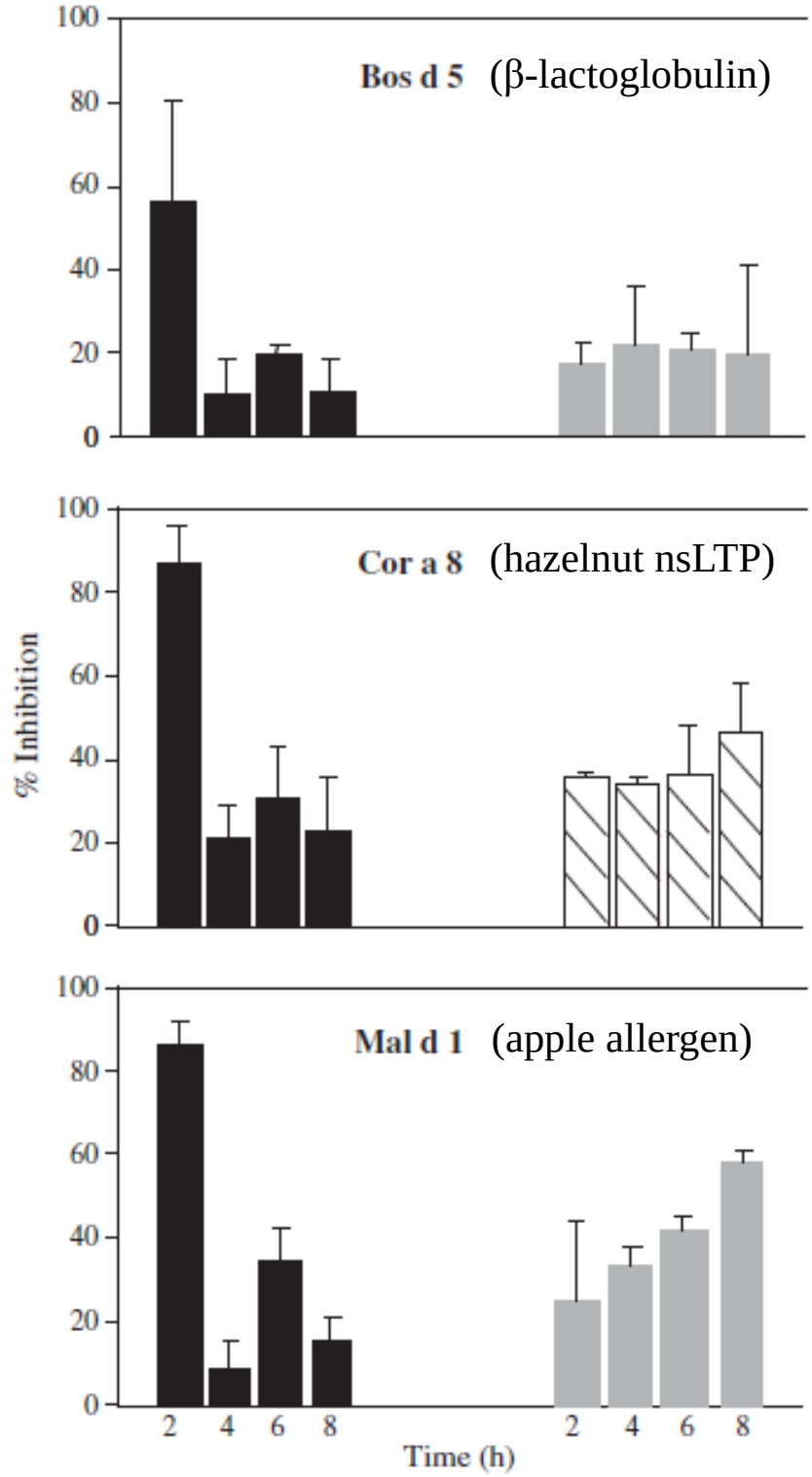
no hazelnut extract

with hazelnut extract

(aqueous extract)

Schulten et al. 2011, Mol. Nutr. Food Res., 55: 1484–1491.

Proteins in blood of rats after feed with and without hazelnut extract



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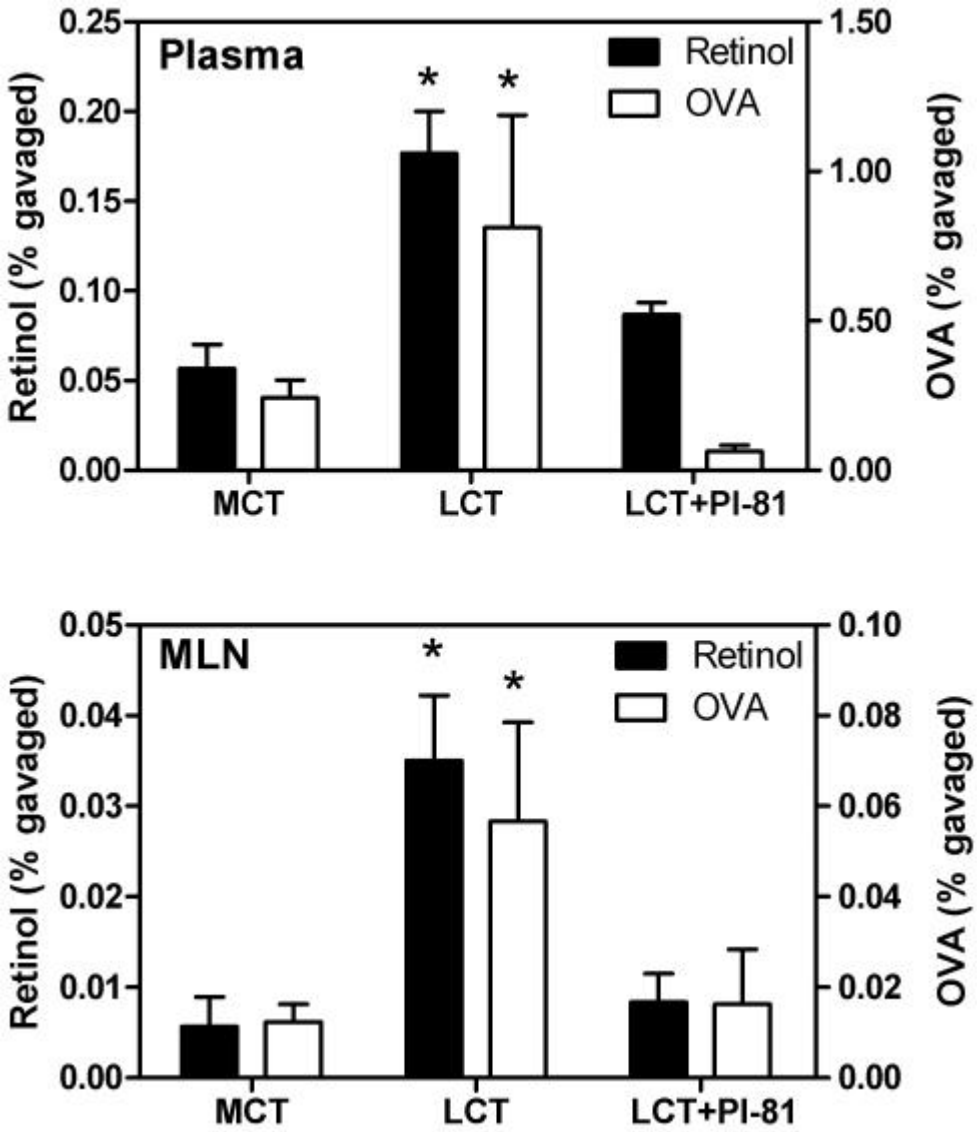
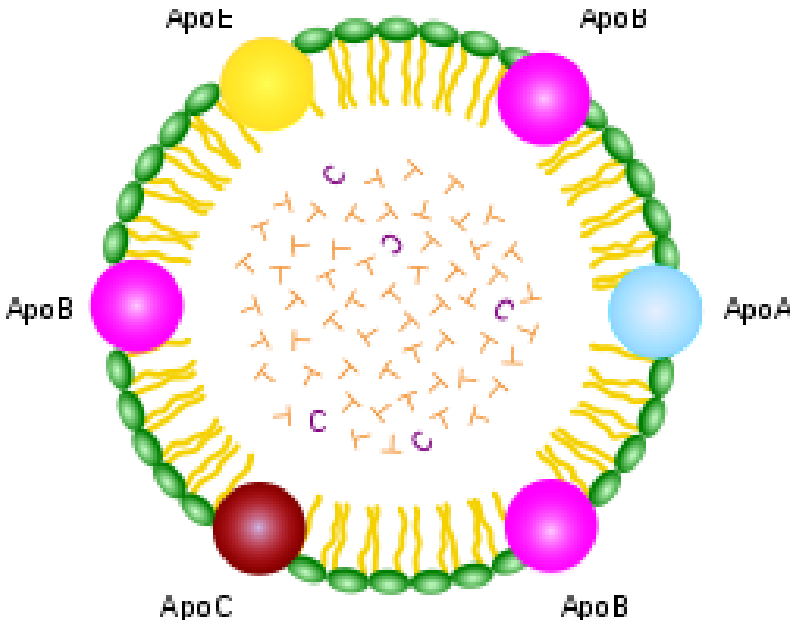
Lipids: long chain fatty acids enhance antigen absorption

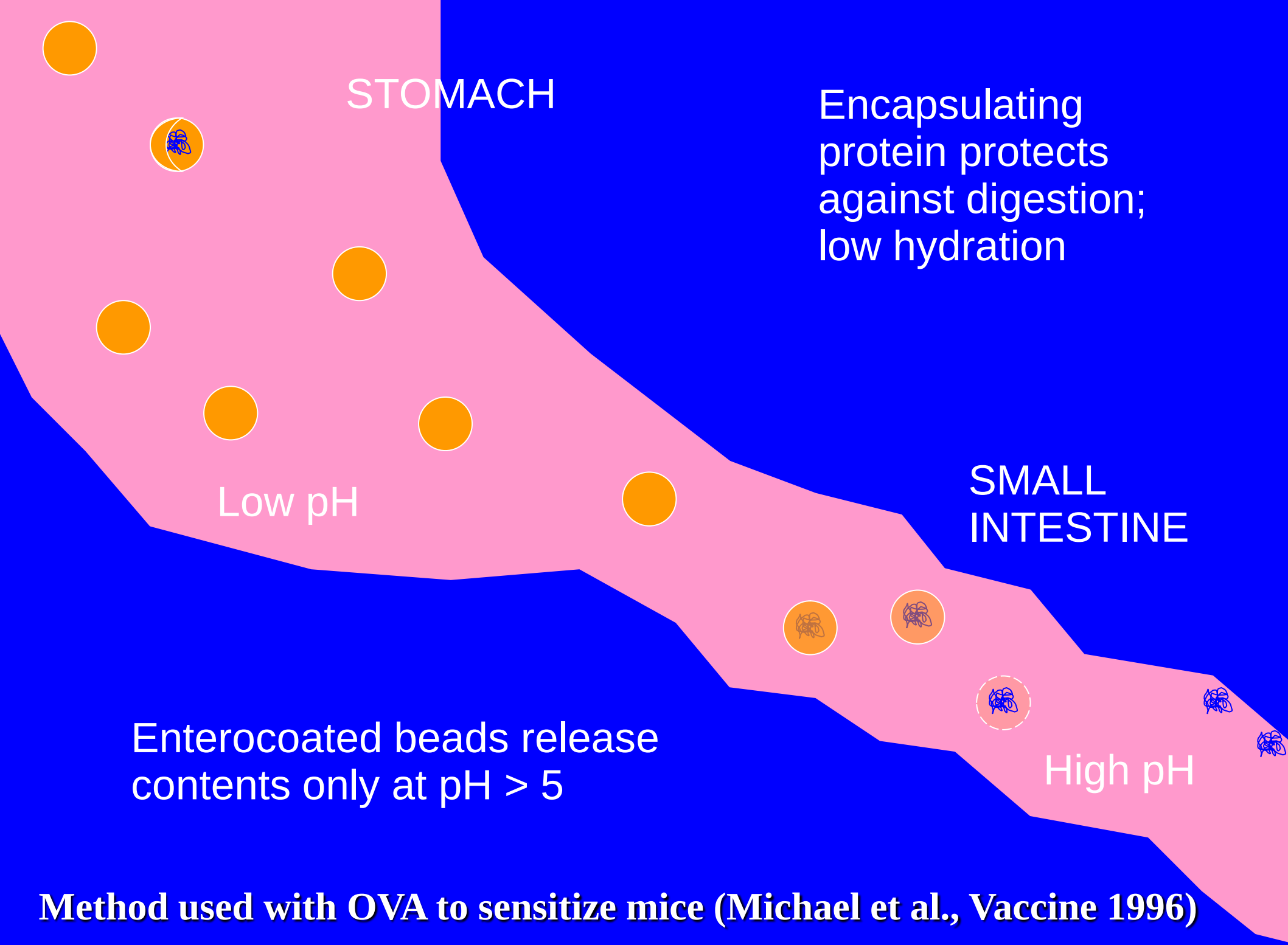




chylomicron

Wang et al. 2009 PLoS One

Palmer et al. 2012 Brit Med J. epi study



Method used with OVA to sensitize mice (Michael et al., Vaccine 1996)

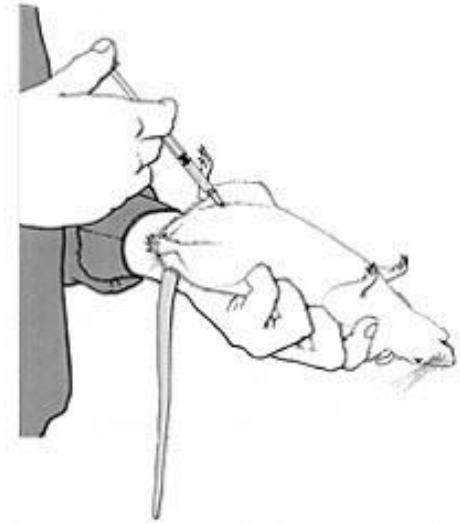
Oral tolerance

- Immunologic hyporesponsiveness to specific antigen encountered via feeding
- Inhibits allergic responses to food
- Generally thought that soluble protein/antigen is better for inducing tolerance

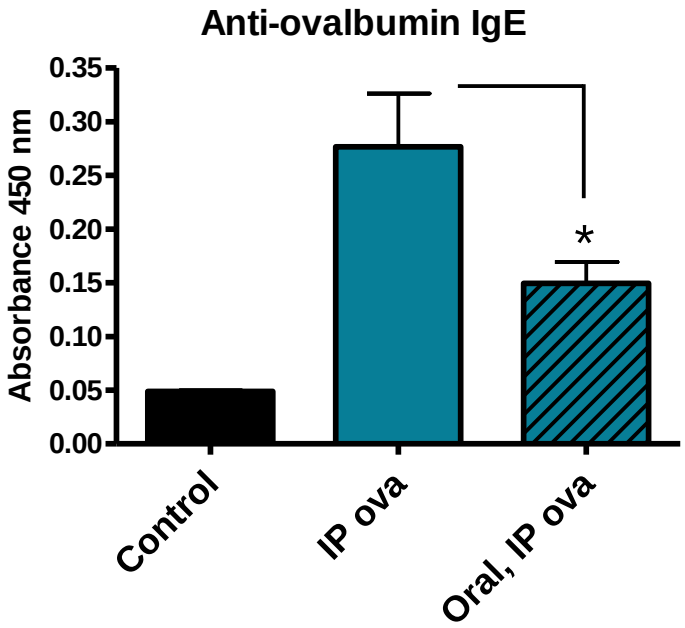
Oral tolerance

1 week

Tolerance generated



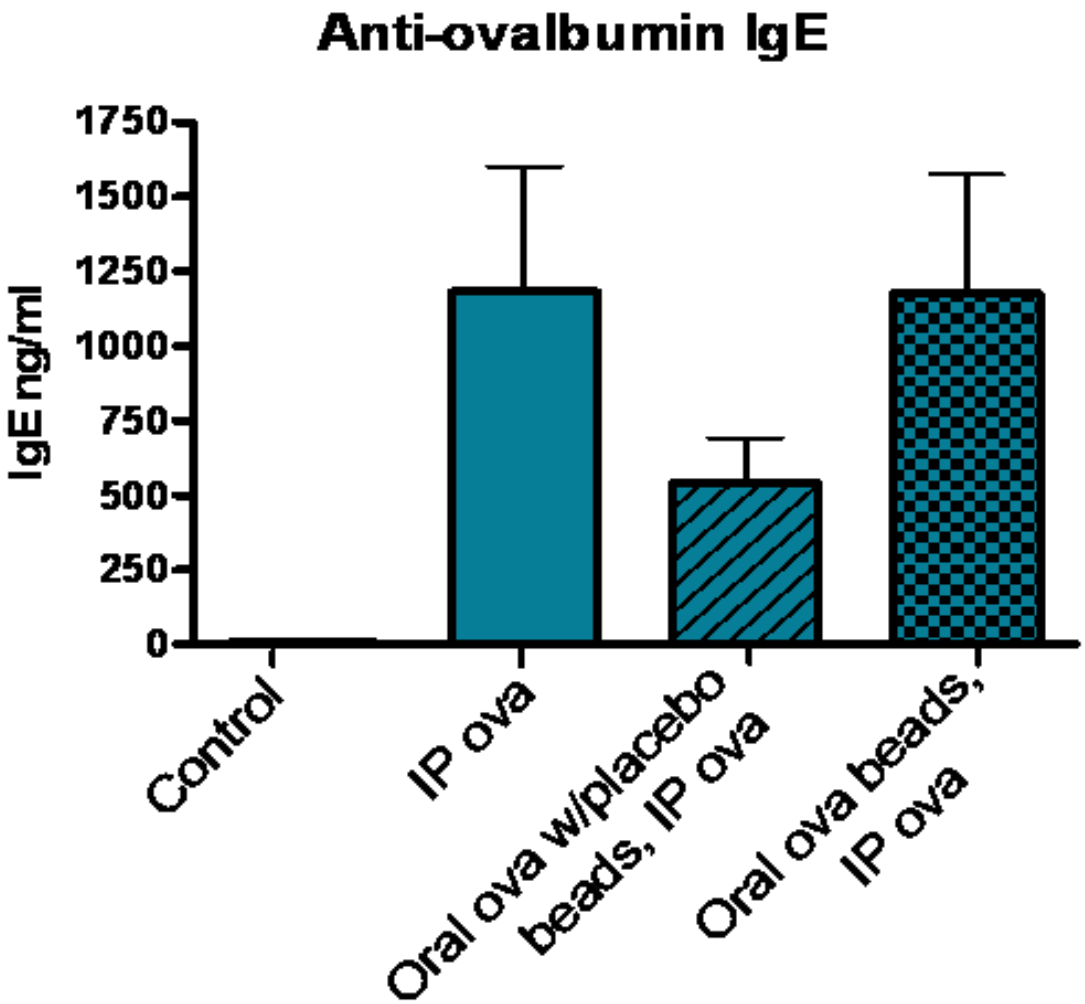
reduced IgE



Anergy or deletion = less T cell help for IgE response
Generation of Tregs = active suppression of Th and B cells

Bowman and Selgrade 2008 Tox Sci

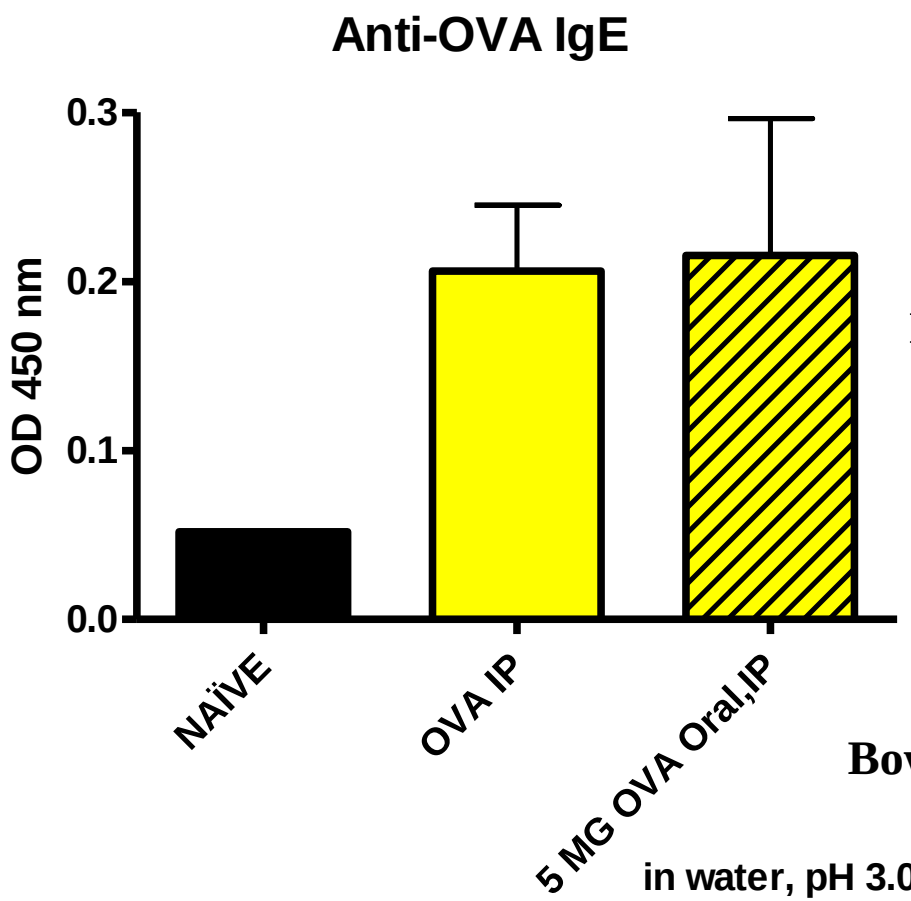
Oral tolerance is no longer induced when ovalbumin is encapsulated



Bowman and Selgrade 2008 Tox Sci

Also demonstrated by JG Michael et al.1996 Vaccine

Matrix pH may affect protein characteristics and therefore tolerance induction



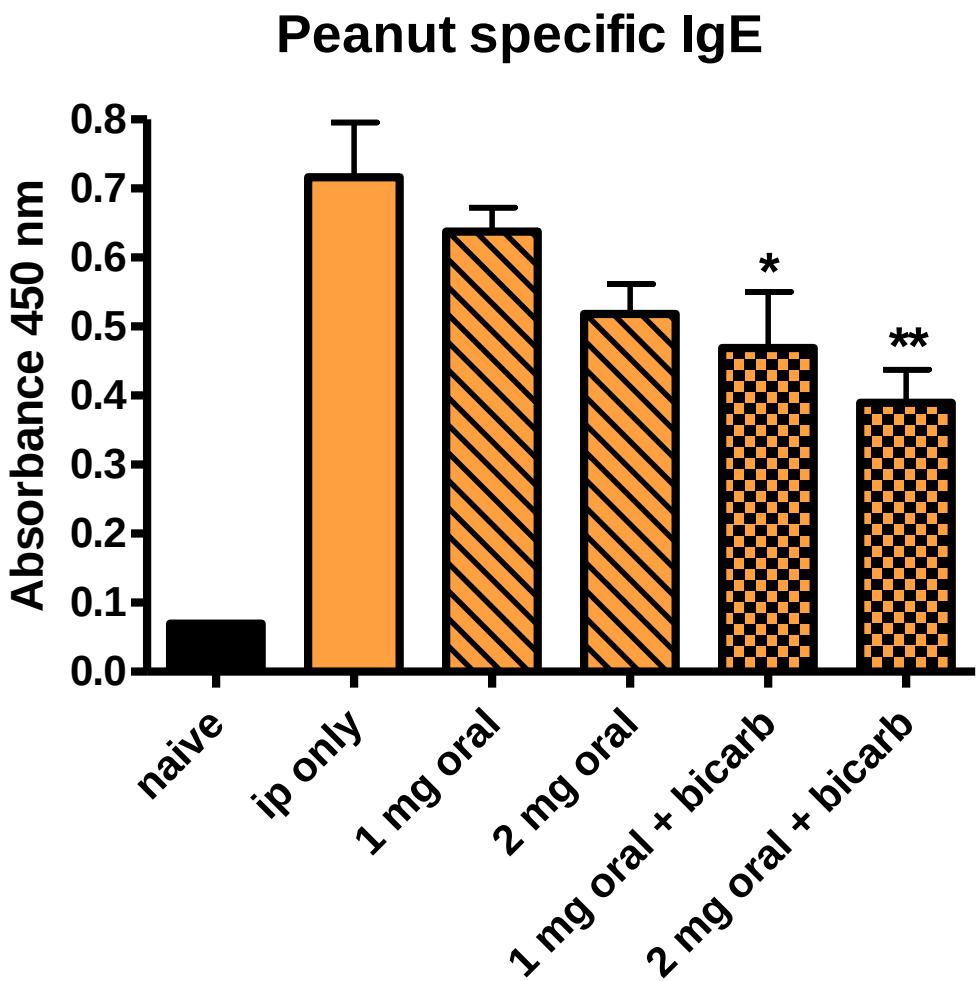
no tolerance

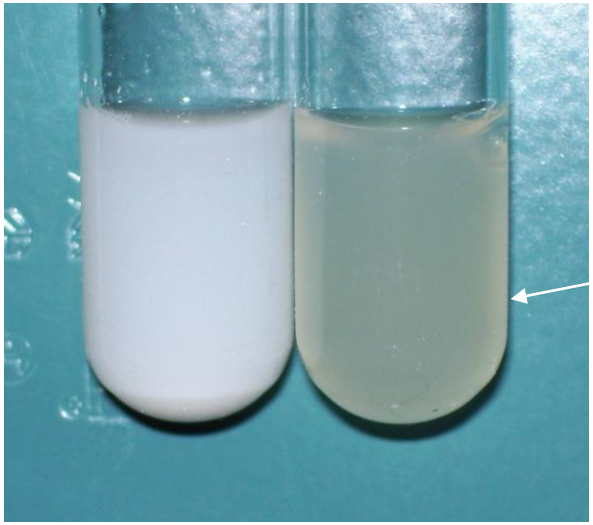
Chemical and heat denaturation also abrogate oral tolerance (Peng 1998).

Bowman and Selgrade 2008 Tox Sci

Ovalbumin was aggregated and did not induce oral tolerance

Peanut becomes orally tolerizing with sodium bicarbonate treatment





increased solubility

untreated

sodium bicarbonate added

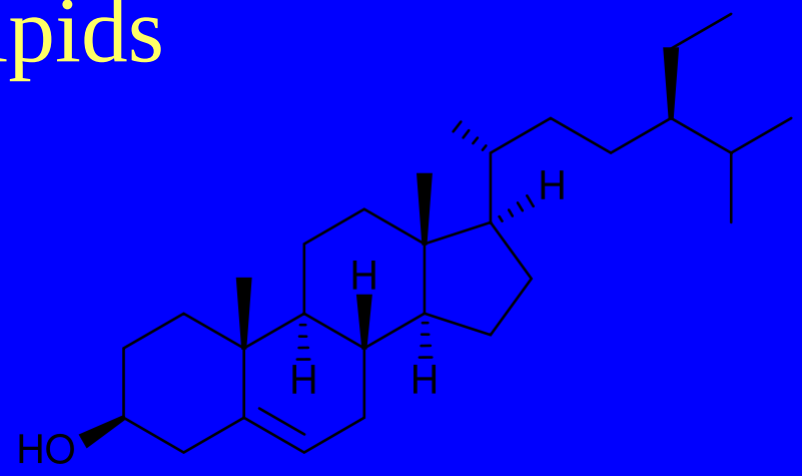
Increased solubility?

or digestion inhibition?

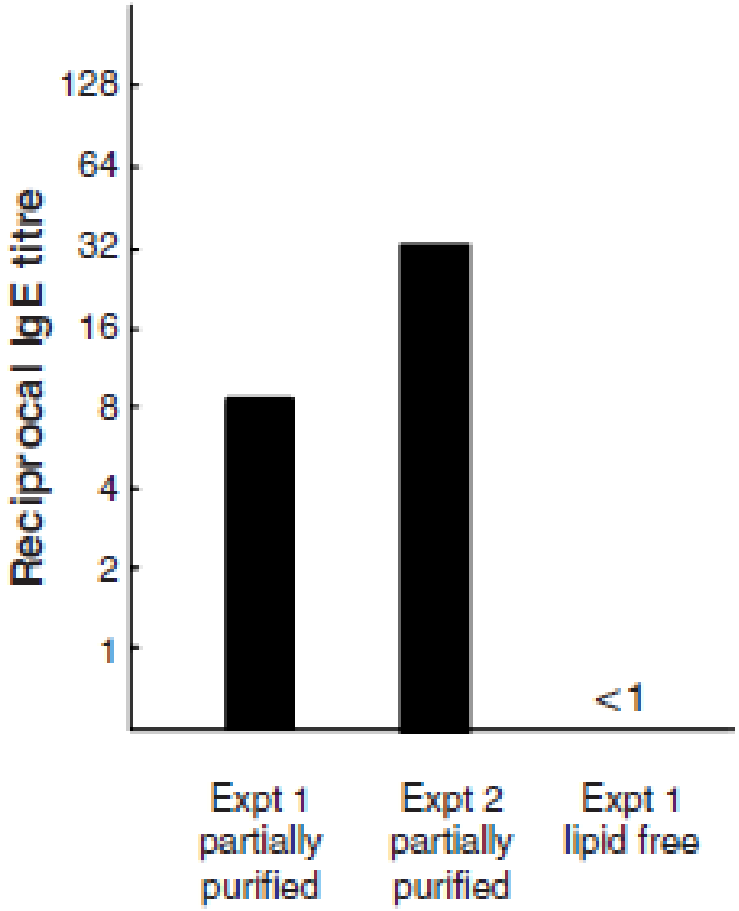
Bowman and Selgrade 2008 Tox Sci

Matrix effects: stimulation of the immune system

- Saponin adjuvants; increased antigen uptake – Maharaj et al., 1986 but also act at a distance
- Plant sterols
- Liposomes, LPS, other lipids
- Lectins



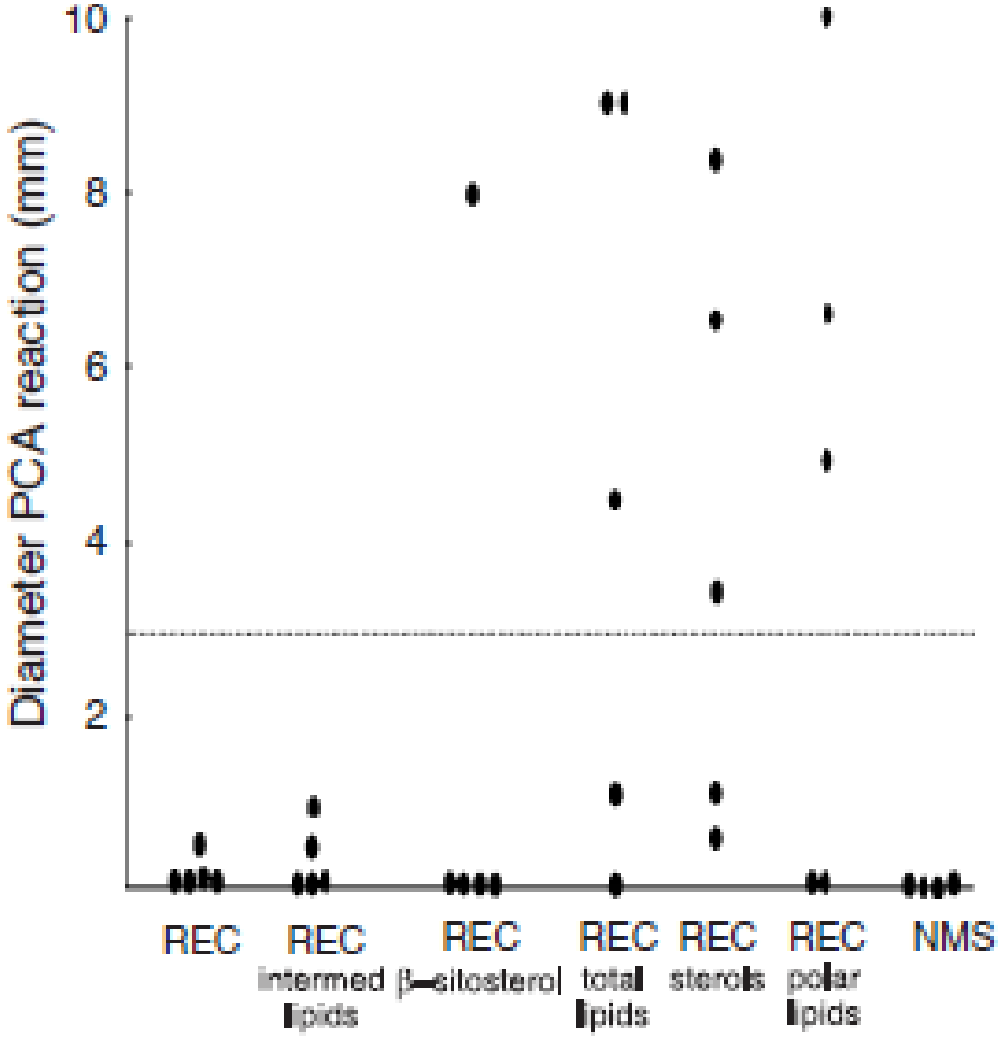
The influence of lipids on IgE production in an injection hazelnut allergy mouse model



Mice exposed ip to partially purified hazelnut extract or recombinant (lipid-free) Ber e 1

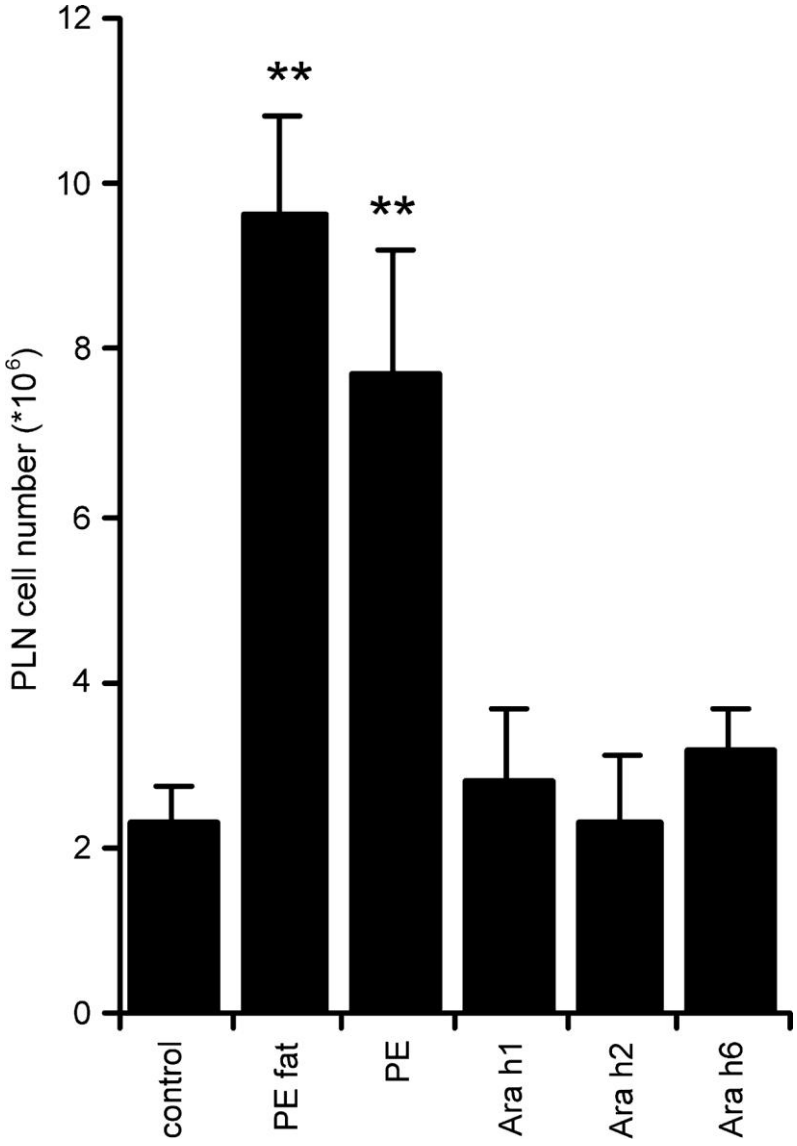
Dearman et al. 2007; Clin and Exp Allergy

The influence of lipid fractions on IgE production in an injection hazelnut allergy mouse model



Dearman et al. 2007; Clin and Exp Allergy

Whole peanut extract stimulates an increase in cell number after footpad injection in mice



van Wijk F et al. Toxicol. Sci. 2005;86:333-341

Processing

- Protein aggregates are generally more immunogenic
- Glycosylation via heating with matrix-supplied sugars

Detection

Need to understand exposure; relies on accurate determination of specific protein content

- Compatibility with standardized reference materials
- Matrix ‘quenching influence’ in immunoassays – varies with protein of interest and matrix tested -- Van den Bulcke 2007 Eur Food Res Technol
- Potential cross-reactivity between assay antibody and other proteins

Recap of food matrix considerations

- Detection and quantitation of proteins
- Processing alterations
- Digestibility assays in vitro
- Digestion and absorption/handling in vivo
- Interactions with the immune system

Discussion

sugars pH fats structure other proteins adjuvants