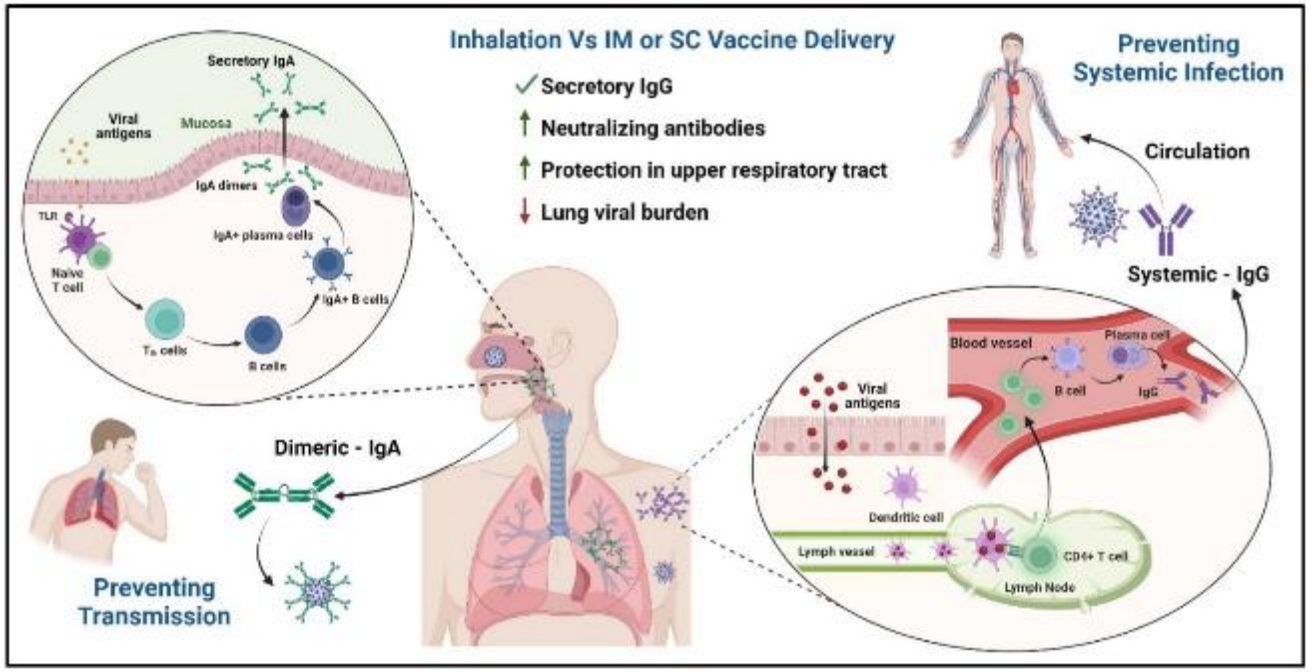
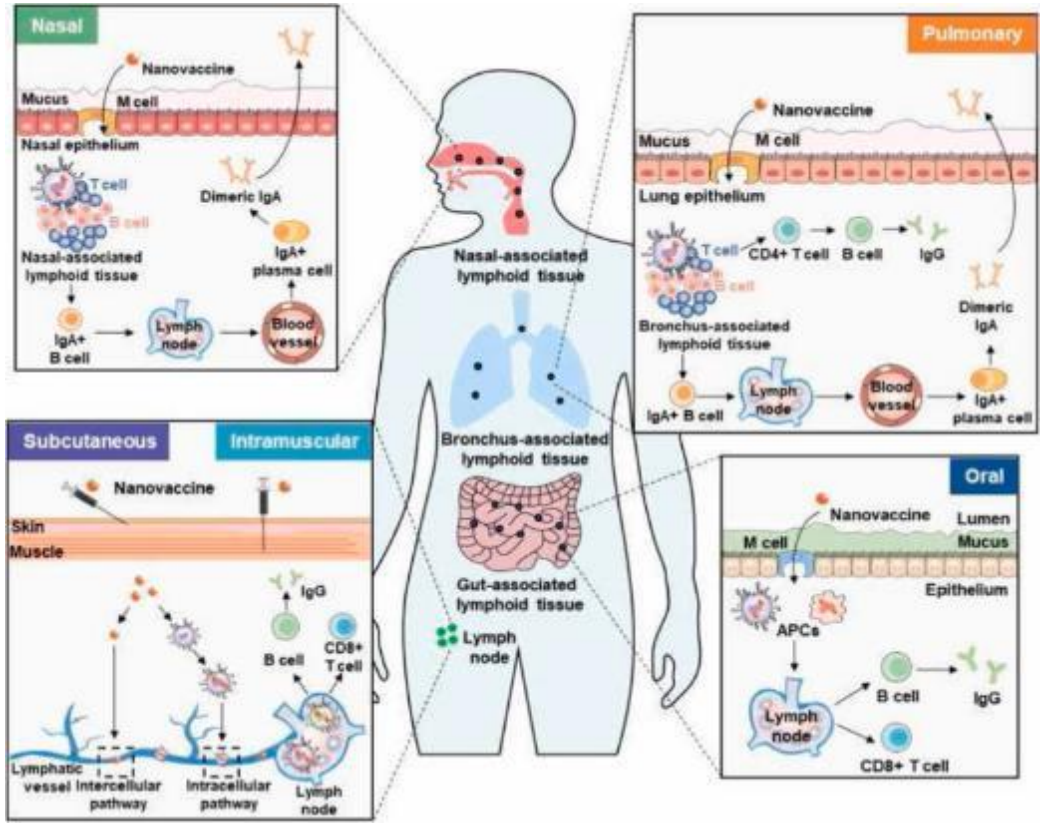
Les enjeux de la vaccination muqueuse

Pr. Stéphane PAUL (GIMAP/CIRI)

Quels sont les enjeux de la vaccination muqueuse?





Les vaccins IM/SC n'induisent pas de réponse muqueuse (site d'infection)

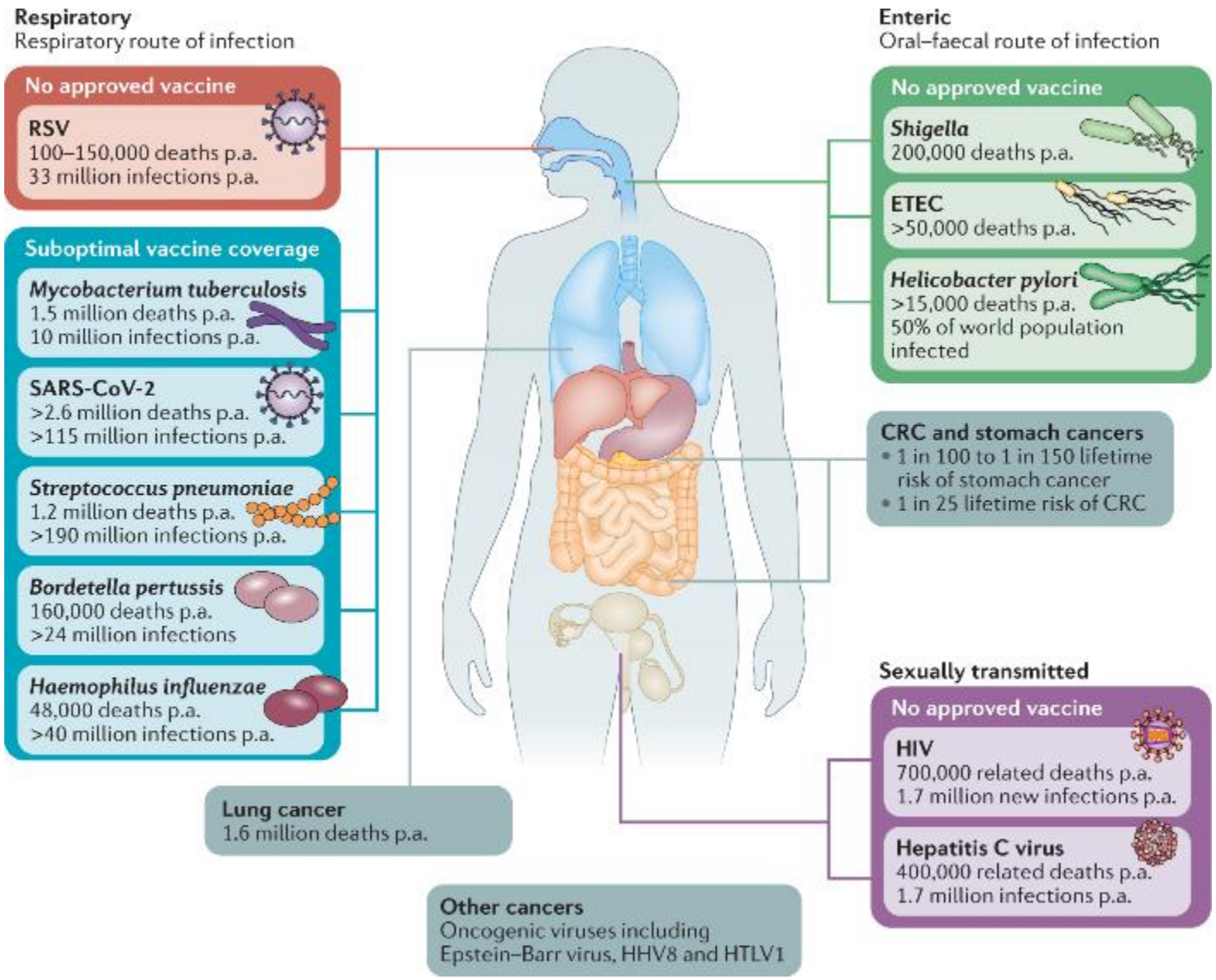
L'émergence de virus respiratoires pandémiques est un challenge

Les vaccins COVID-19 réduisent trop faiblement la transmission

Les vaccins muqueux sont trop faiblement immunogènes

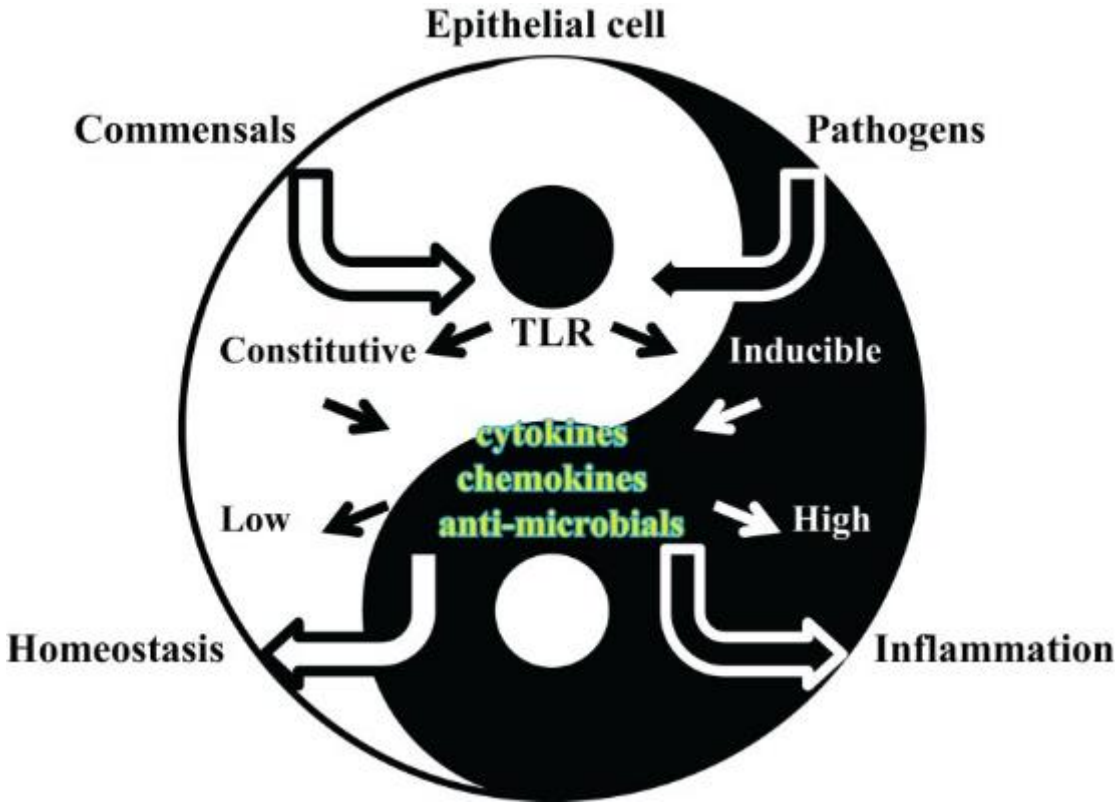
L'acceptabilité des vaccins est assez faible (enfant..)

Les muqueuses sont un site majeur d'entrée des pathogènes



Lavelle and Ward, 2022

Les muqueuses sont uniques d'un point de vue immunologique



- Major entry site of pathogens
  - ✓ Surface (400 m^2)
- Septic medium: Commensal flora
  - ✓ High antigen rate
- Tolerance maintenance / Protection against pathogens
- Ability to discriminate pathogens / commensal

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4129373/

Les muqueuses sont uniques d'un point de vue immunologique

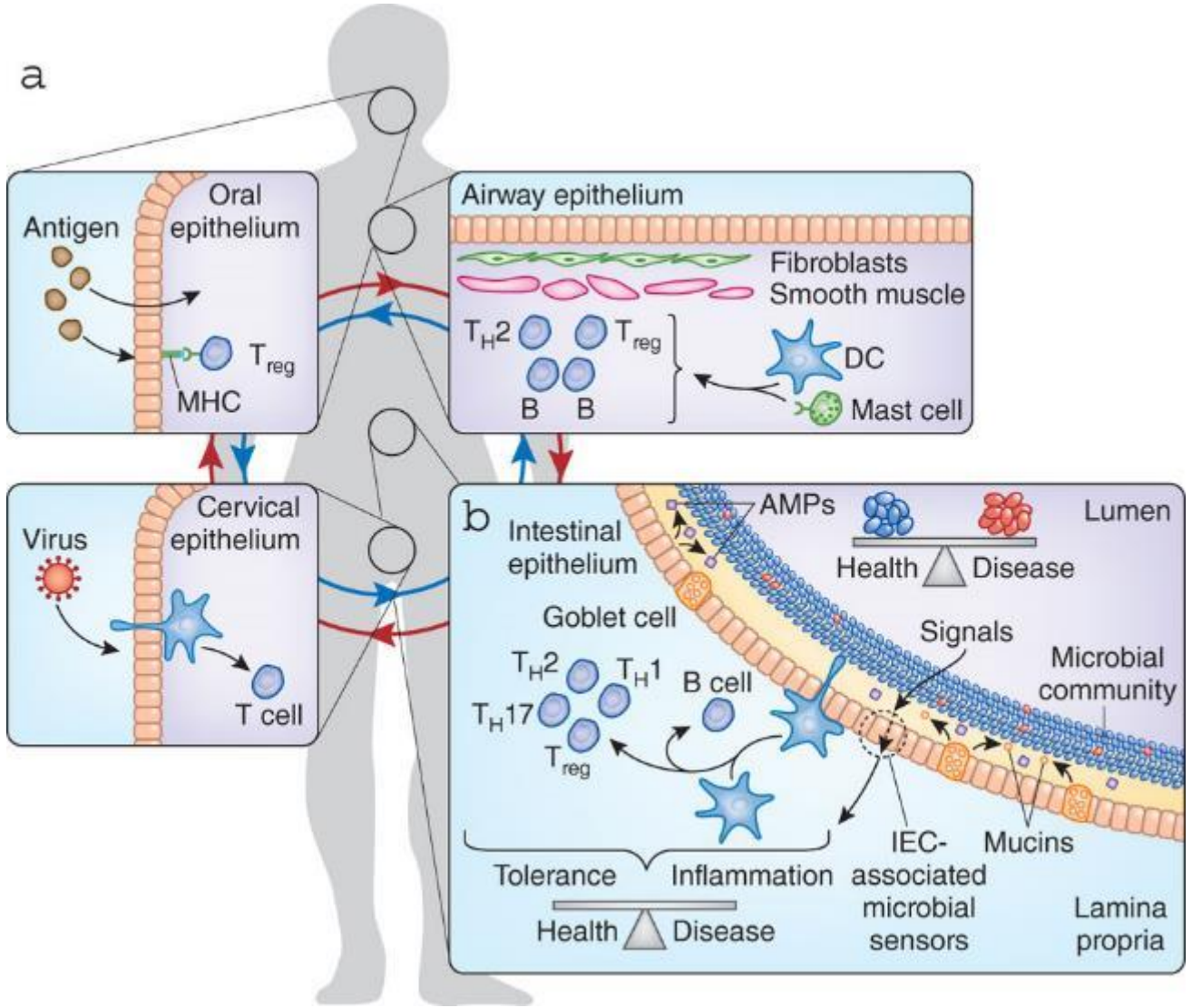
Mucosal-associated lymphoid tissues (MALT) contain around 80 % of immune cells : GALT, NALT, BALT, CALT, SALT

Specific immune features :

- non inflammatory DC and macrophages are abundant
- IgA plasma cells
- Treg et Th17
- Inductive and effector sites

Functions :

- First defense line against antigens and pathogens
- Regulation of immune responses to pathogens
- Prevention of immune responses against commensal bacteria (**tolerance**)



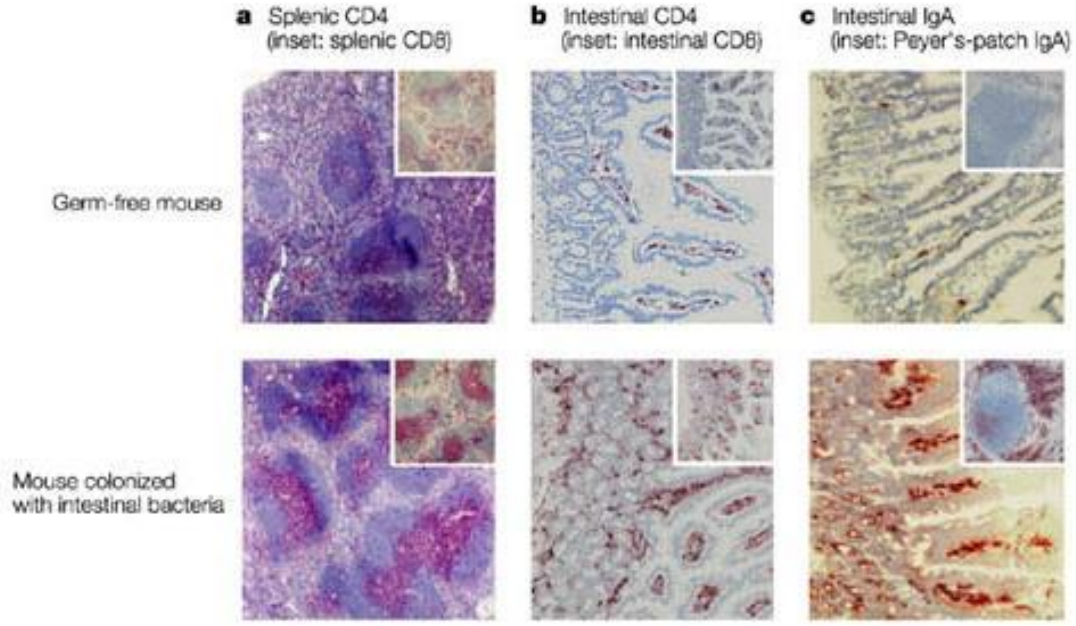
Rôle important de la flore et du microbiote dans la réponse muqueuse

*Colonisation at Birth
*10^{14} bacteria (mainly anaerobic)
*Density gradient Colon >>> Small intestine
*Metabolic activity
*Activity on the epithelial barrier
*Competition with pathogens
*Required for MALT development

*In absence of bacteria
- -Lymphopenia (mainly CD4)
- -No Germinal Centers / No Ig
- -No ILFs
- -Immunodeficiency
- -Homeostasy defect of the mucosa

*Mechanisms:
- TLR
- Nod1 etc….

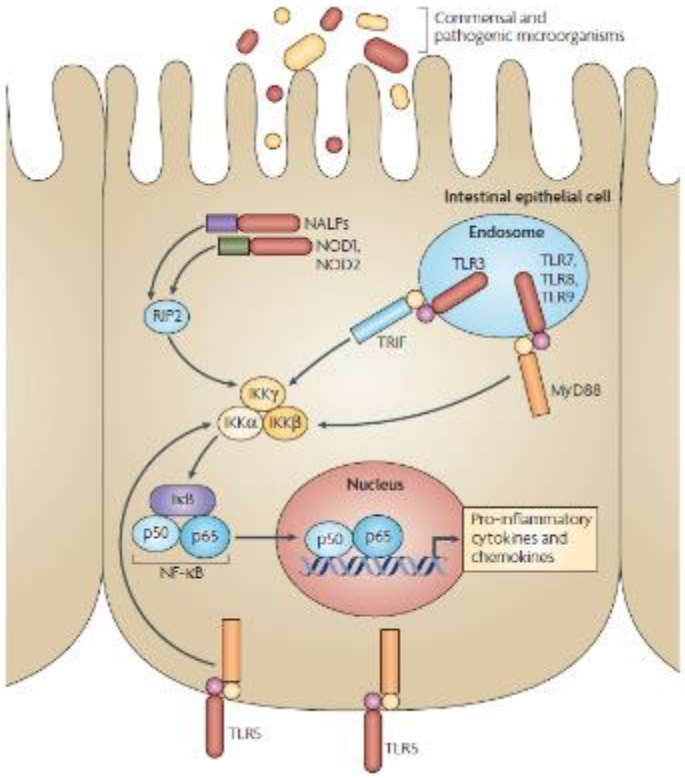
Le microbiote éduque le système immunitaire



Une vaccination muqueuse doit stimuler et traverser l'épithélium

Epithelial cells

- Expression of Pathogen-Associated Molecular Patterns (PAMPs) such as ***Toll-Like Receptors*** (TLR), ***NOD-like receptor*** (NLR)

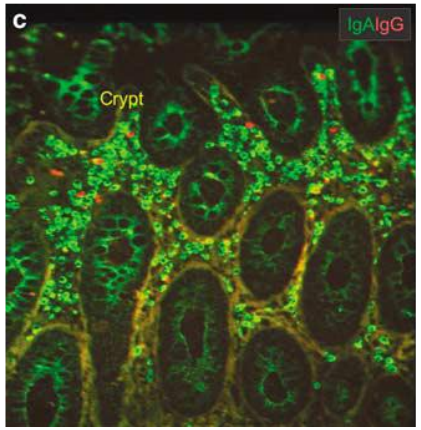


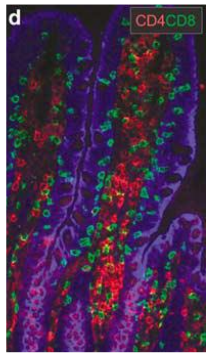
TLR mRNA and protein expression in mucosal epithelial cells.

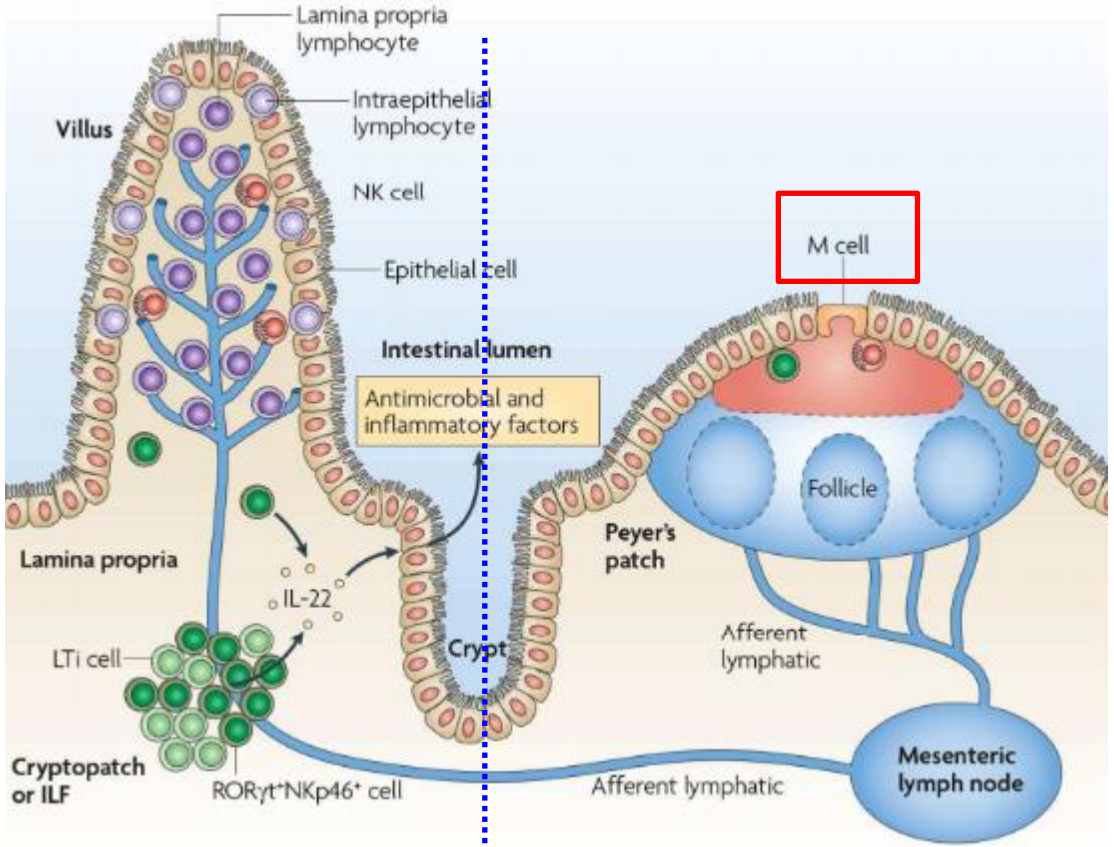
Tissue	TLR	
	mRNA	Protein
ORAL EPITHELIA		
Gingival	TLR1 (71, 72), TLR2 (73–75), TLR3 (73), TLR4 (73, 75, 76), TLR5 (71, 72), TLR6 (71, 72), TLR7 (73), TLR8 (71–73), TLR9 (71, 72)	TLR1 (71), TLR2 (71–73, 75–78), TLR3 (71, 73, 78), TLR4 (71–76), TLR5 (71, 77, 78), TLR6 (71, 72, 78), TLR7 (71, 73), TLR8 (71), TLR9 (71, 74, 75, 79)
Salivary	TLR1–TLR10 (80, 81)	TLR1–TLR4, TLR7 (80)
Tonsillar	TLR1–TLR6, TLR9, TLR10 (80, 82)	TLR2, TLR3 (82)
Ear epithelia	TLR2–TLR4, TLR9 (83–86)	TLR2–TLR4, TLR9 (83–86)
OCULAR EPITHELIA		
Corneal	TLR1 (87), TLR2 (6, 87–90), TLR3 (87–89), TLR4 (6, 87–89), TLR5 (87, 91), TLR6 (87), TLR7 (87, 88), TLR9 (87–89), TLR10 (87)	TLR1 (92), TLR2 (6, 87, 90, 92–96), TLR3 (6, 87–89, 92, 93), TLR4 (6, 88, 90, 93, 96–98), TLR5 (87, 91–95, 97), TLR6 (92), TLR9 (87, 89)
Conjunctival	TLR1 (87), TLR2, TLR3 (87, 88), TLR4 (87, 88, 99), TLR7 (87, 88), TLR9 (87, 88, 99), TLR10 (87)	TLR3 (88), TLR4 (88, 99), TLR9 (99)
Retinal	TLR1–TLR7, TLR9 (100)	TLR2–TLR4 (100)
Iris	TLR4 (98)	TLR4 (98)
AIRWAY EPITHELIA		
Nasal	TLR1–TLR10 (101, 102)	TLR2 (102, 103), TLR3 (102), TLR4 (103)
Tracheal/bronchial	TLR1 (7, 81, 104), TLR2 (7, 81, 104, 105), TLR3 (7, 81, 104), TLR4 (7, 81,	TLR1, TLR2 (7, 104, 105, 107), TLR3 (7, 104, 107), TLR4 (7, 104, 106, 107),

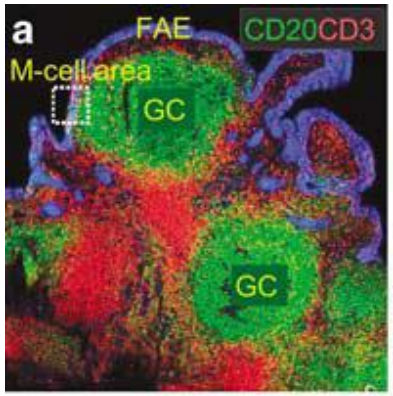
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4129373/

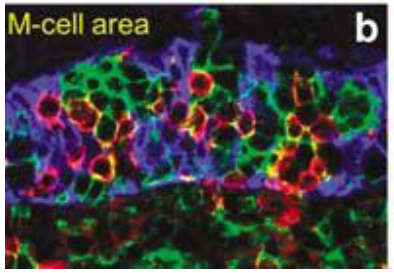
Une vaccination muqueuse doit stimuler l'immunité adaptative











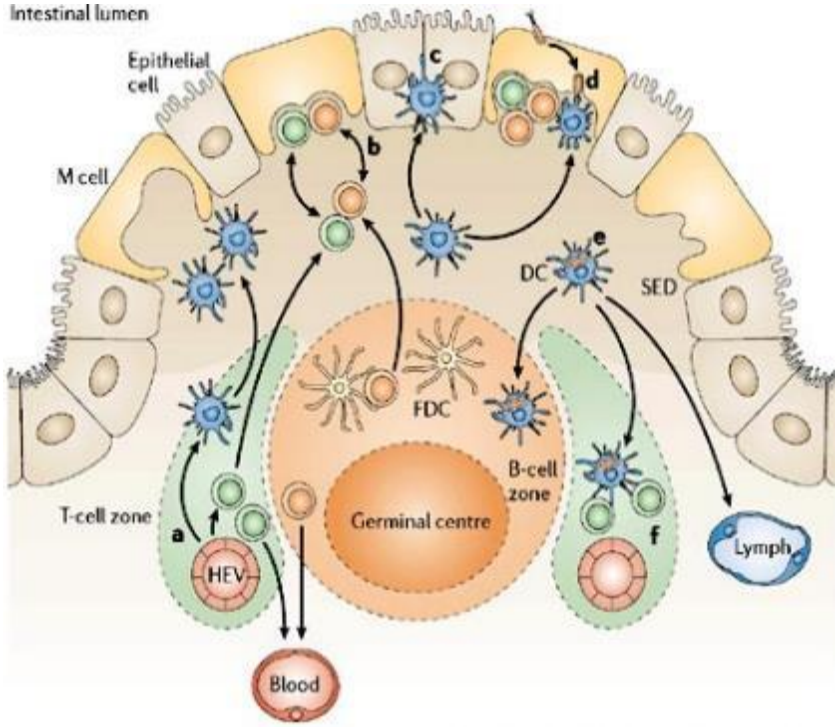
Lamina propria (Effector site)

- IgA plasma cells (green)
- Intraepithelial lymphocytes
- CD4 and CD8 T cells
- Macrophages, polymorphonuclear leukocytes

Peyer's patches (Inductive site)

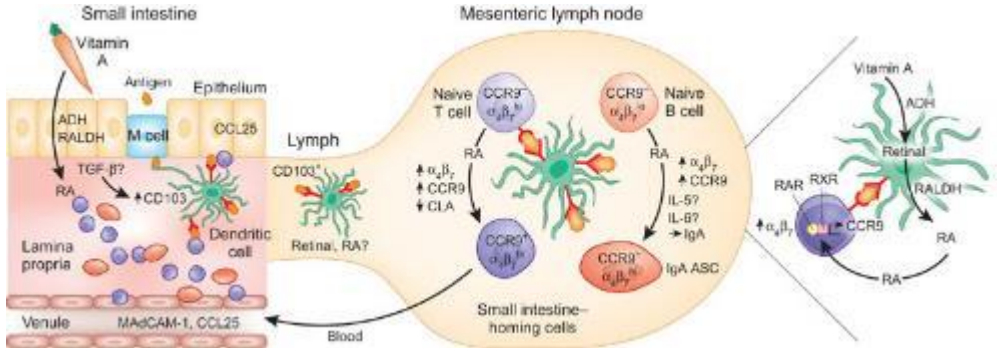
- Subepithelial dome with M cell (blue)
- Antigen-presenting cells (e.g. dendritic cells)
- Interfolicular regions enriched in naive T cells (red)
- Follicles enriched in naive B cells (green)

L’immunité muqueuse est compartimentée



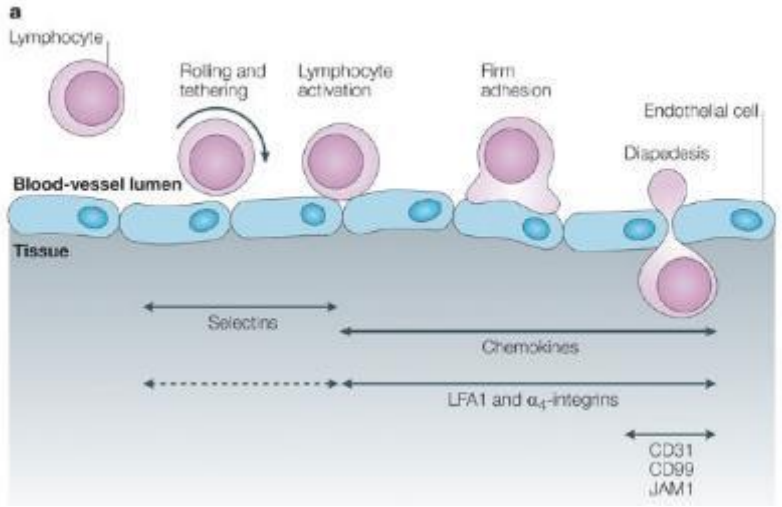
Cellular traffic in organized MALT

A) Naive/memory LT/LB as iDCs enter the mucosa through HEVs.
B) Some LB/LT cells migrate into microfold (M)-cell pockets
C) iDCs remain in the SED but a few migrate into the FAE.
D) Ags/microbes transported by M cells are captured by DCs.
E) Ag induces DC maturation and movement into interfollicular T-cell areas, the B-cell zone, and perhaps into DLNs as MLN



CCR9 et CCR10

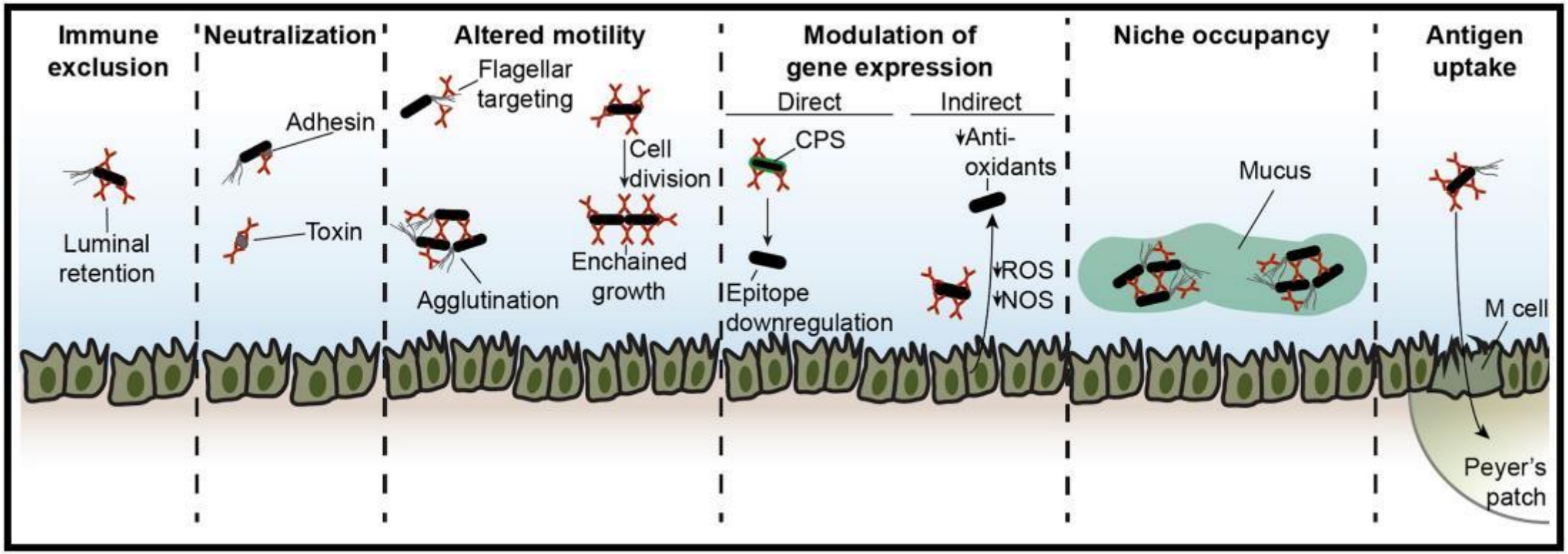
- LyT/B CCR9: intestin grêle
- Ly B CCR10: colon



Adhesion: $\alpha 4\beta 7$/MadCam, $\alpha E\beta 7$/E-Cadherine

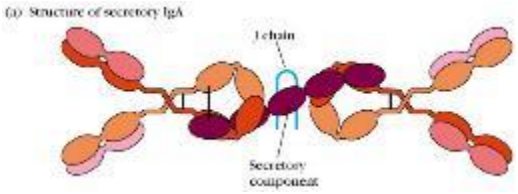
Chimiokines: CCL25 et CCL27

Les IgA sont des anticorps pluripotents des muqueuses indispensables à une réponse vaccinale efficace



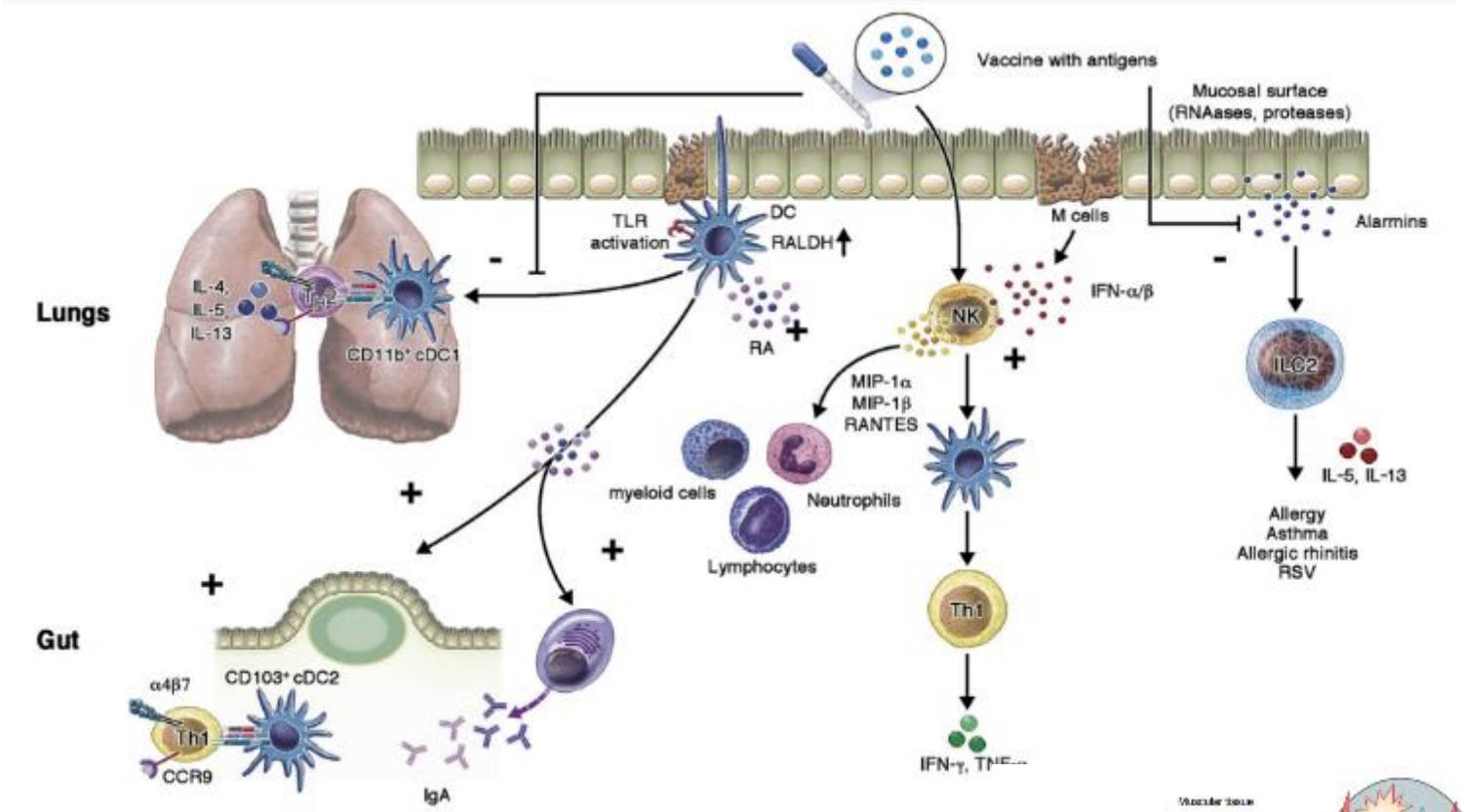
Potential Functions of IgA Antibodies

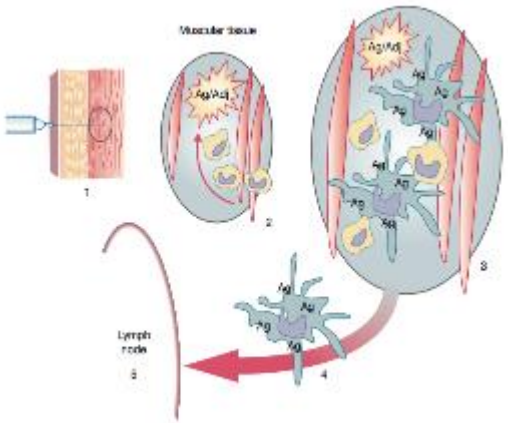
Numerous potential functions for IgA antibodies have been suggested that may or may not play a role *in vivo* in the context of homeostatic interactions with the commensal microbiota. These include immune exclusion, neutralization, altered motility, modulation of gene expression, niche occupancy, and enhanced Ag uptake.



Caractéristiques d’un vaccin muqueux efficace?

*Block adherence of microorganism to host
*Facilitate clearance from host
*Neutralize toxin
*Must recognize “virulence” epitopes
*Must be immunogenic
*Must not induce autoimmune disease
*Should induce long-lasting immunity
*Activate mucosal APCs or epithelial cells
*Must induce the type of response that is effective to eliminate pathogen (eg. Th1 or Th2)





Quels sont les bénéfices d'une vaccination muqueuse?

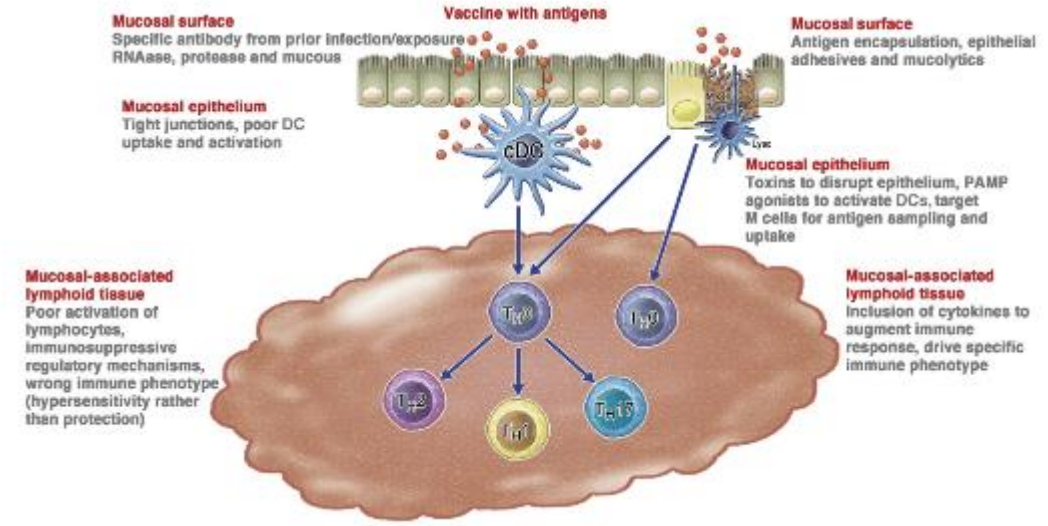
1) Immune responses: induction of local immune responses (mucosal sIgA, mucosal cellular responses; mucosal memory immune responses)

2) Needle-free strategies:

(i) Ease of administration
(ii) Non-invasiveness
(iii) High-patient compliance
(iv) Suitability for mass vaccination
(v) High safety

TABLE I. Theoretical advantages of mucosal immunization

Result of mucosal immunization	Specific immune mechanisms	Result of immune activation	Diseases where potentially valuable	Examples
Immune response homes to mucosal surfaces	Induction of immune cells with mucosal-targeting molecules	Focusing of the immune response to areas of need	• Respiratory and genital viral infections • Enteric infections • Mucosal epithelial cancers	• Rotavirus, COVID-19[16,18,21] • Enteropathogenic *E. coli*[16,17] • Lung and colon cancer[88]
Unique types of immunity produced on mucosal surfaces	IgA, IL-17, and CD8 immunity	Sterile immunity at mucosal surfaces, clearance of virally infected or transformed cells	• Respiratory viral and bacterial infections • Sexually transmitted disease • Mucosal epithelial cancers	• Influenza, COVID-19[14,19] • Herpes simplex 2[11,26] • Pertussis[22] • Lung and colon cancer[88]
Induction of long-term immunity	T_{RM} cells	Long-term protection against recurrent disease	• Recurrent respiratory infections • Enteric infections • Urological infections	• Influenza, COVID-19[13,21,26] • HIV[17] • Urinary tract infections[25]
Regulatory immune responses	Tissue-resident regulatory T cells	Downregulation of immune inflammation and shifting to protective immunity	• Allergies • Autoimmune disease	• Allergic asthma, food allergy[30] • Type I diabetes, multiple sclerosis[29,79]

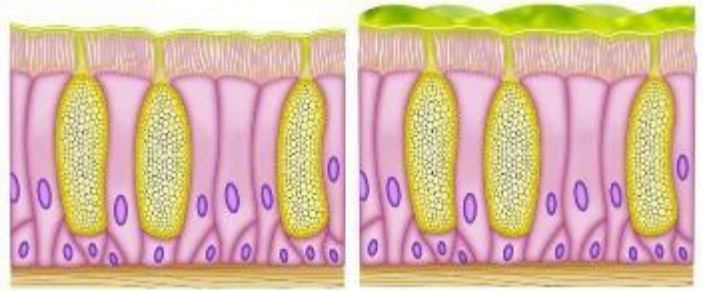


Quels sont les challenges pour la vaccination muqueuse?

1) Vaccine uptake (delivery of antigens) gastric acid, mucus, enzymes, device

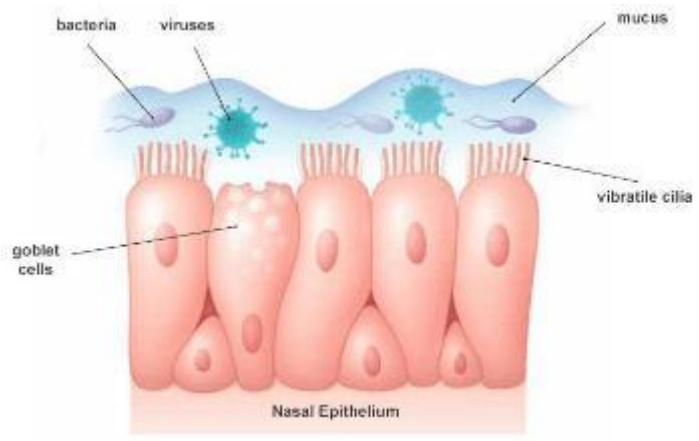
2) Local tolerance

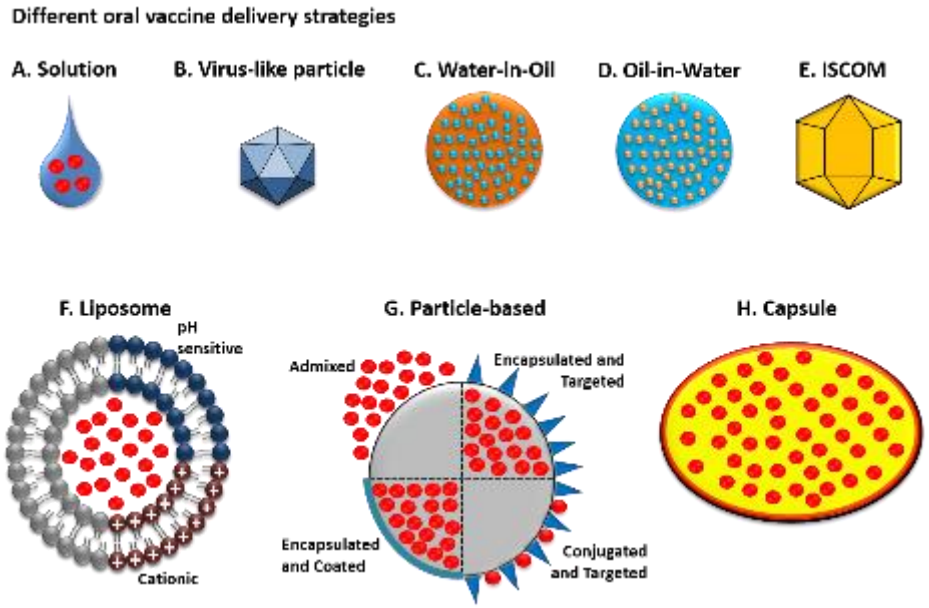
3) Induction of long-term responses and long-term memory



4) Criteria to approve a vaccine are currently based on systemic immune responses

=> Only few licensed mucosal vaccines





Vaccination muqueuse

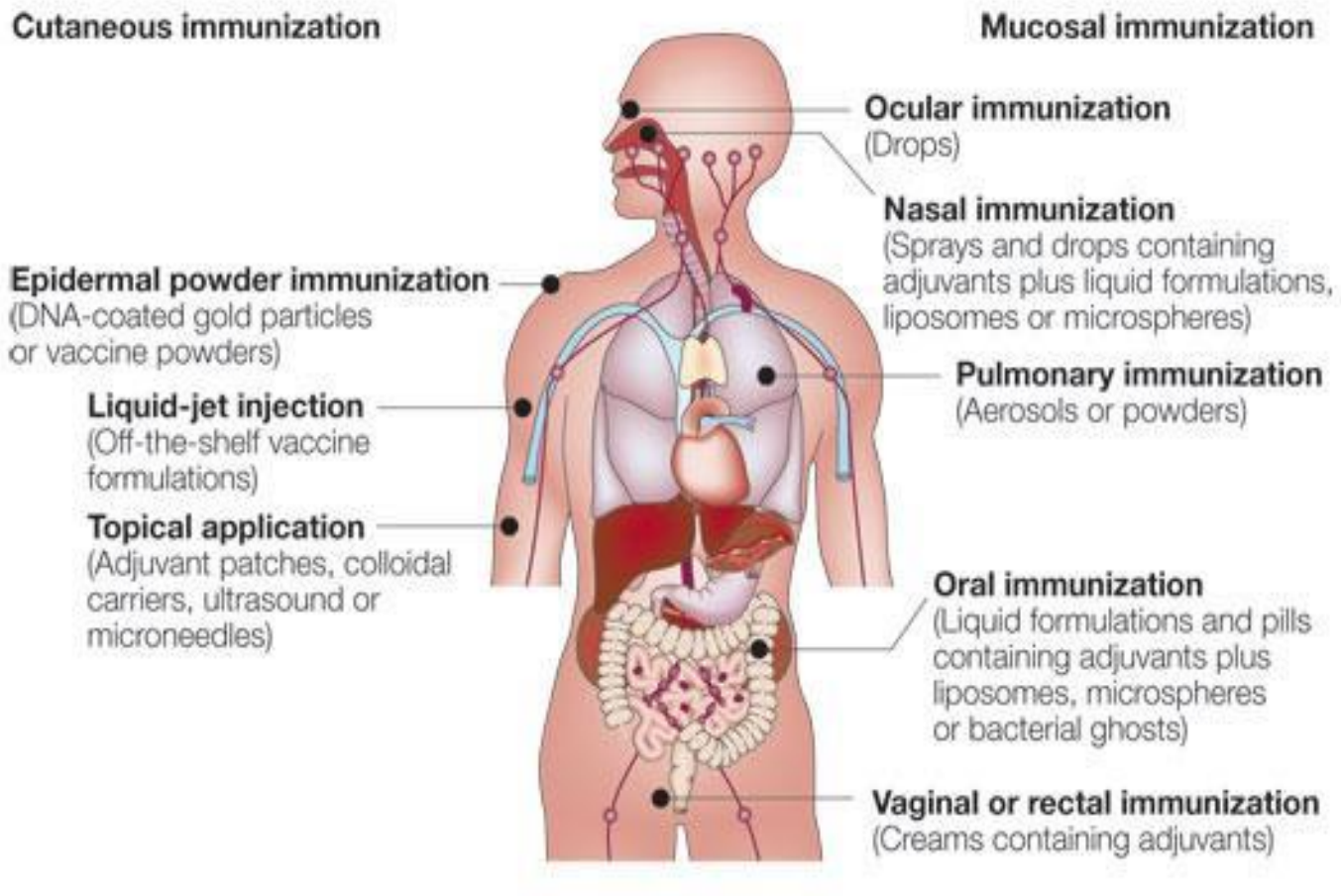
*Oral (buccal, sublingual and gingival)

*Nasal

*Pulmonary

*Vaginal

*Rectal

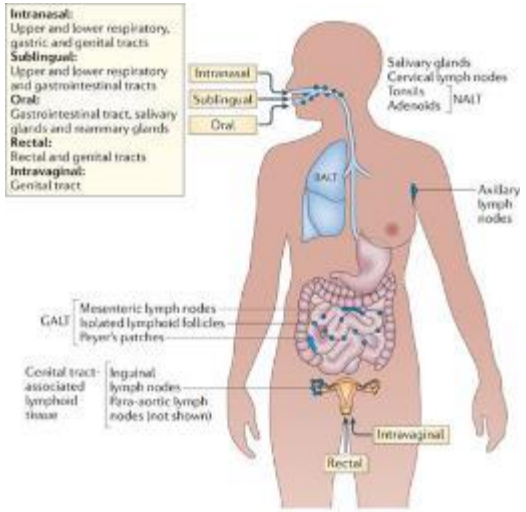


Mucosal immunization, is further classified into ocular, nasal, oral, pulmonary, and vaginal or rectal routes.
Ocular immunization can be carried out using eye drops.
Nasal immunization is carried out using sprays that comprise liquid formulations, liposomes or microspheres.
Vaccines can be delivered orally in the form of liquid doses or pills, both of which can consist of various formulations: for example, microspheres.
Vaccines can also be delivered to the vaginal or rectal mucosal membrane, using topical creams, or to the lungs, using aerosols or powders.

Compartimentation de la réponse immunitaire muqueuse….

	Nasal	Subling	Oral	Rectal	Vaginal	Trans-dermal
Upper respiratory	+ + +	+ + +	–	–	–	+ + +
Lower respiratory	+ to + + +	+ + +	–	–	–	+ + +
Stomach	–	+/+ + +	+/+ + +	–	–	?
Small intestine	–	+ + +	+ + +	–	–	+
Colon	–	?	+	+ +	–	+
Rectum	–	?	(+)	+ + +	–	?
Reproductive tract	+ + +	+ + +	–	–	+ +/+ + +	?
Blood	+ + +	+ + +	+(+)	+(+)	+(+)	+ + +

Nasal and Sublingual vaccination are highly attractive routes of vaccination

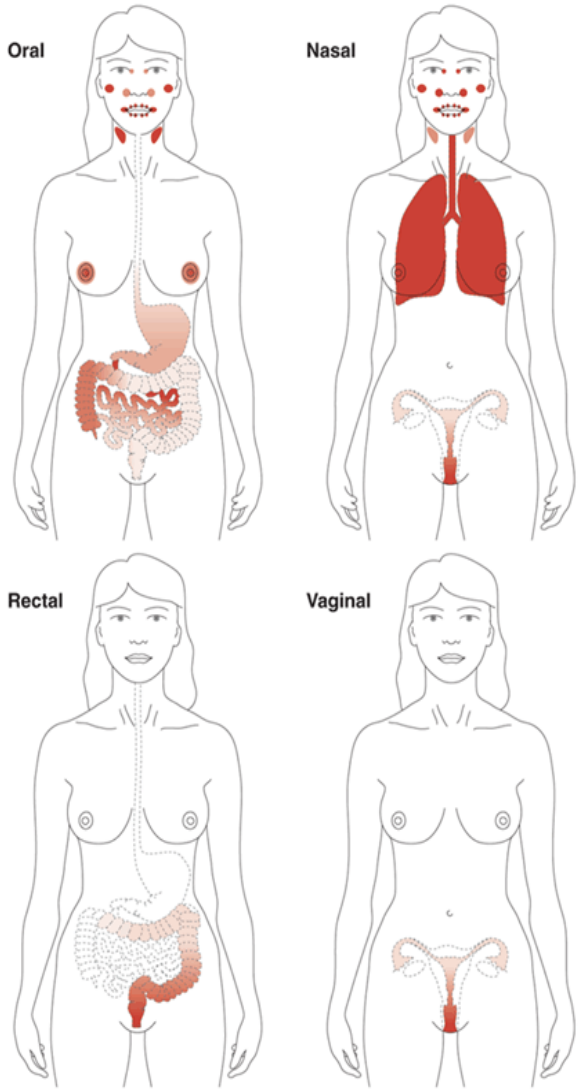


Intranasal vaccination is preferred for targeting the respiratory, gastric and genital tracts

Oral vaccination is effective in the gut and for induction of mammary gland Abs (secreted in milk)

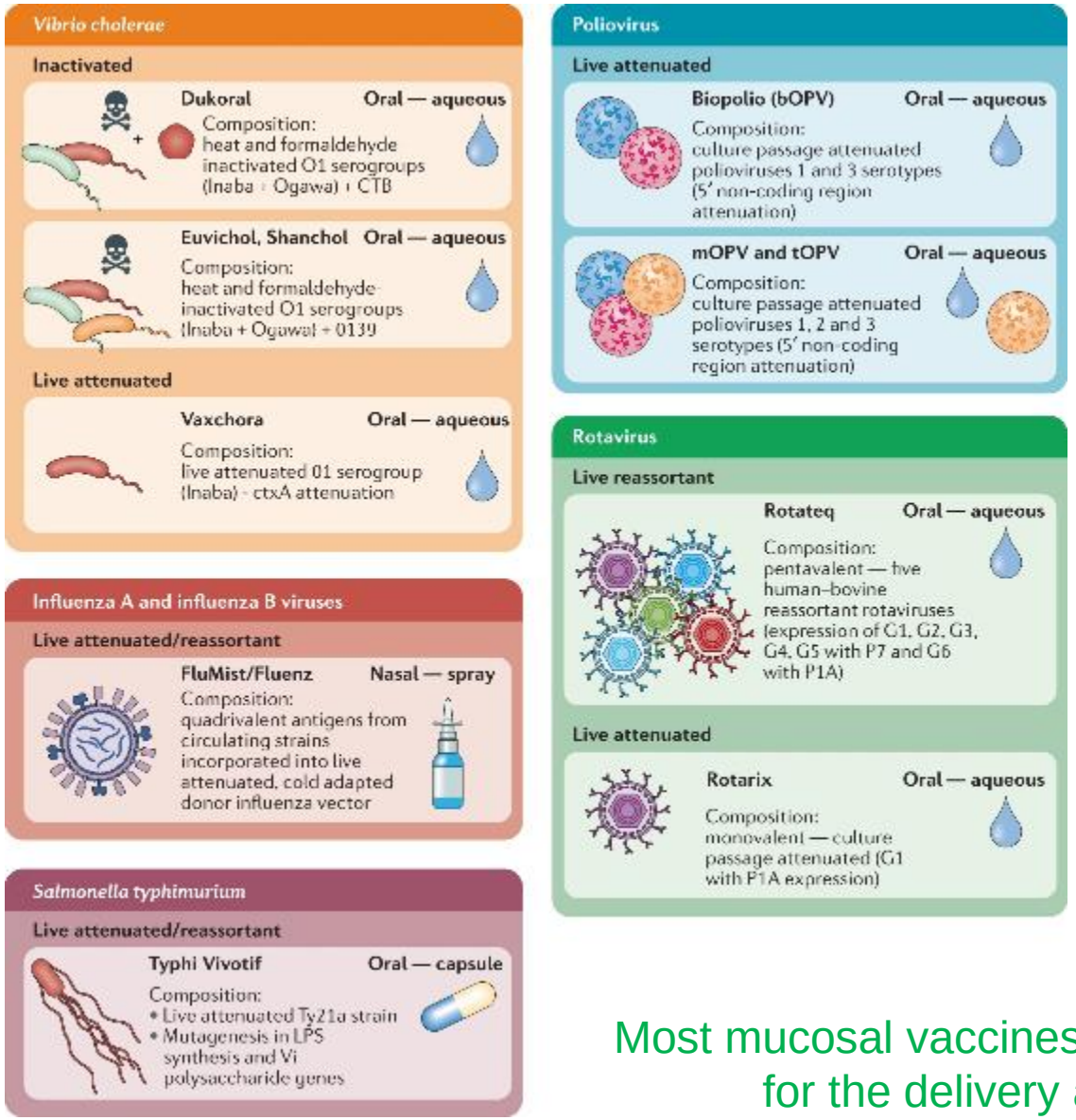
Rectal immunization is best for the induction of colon and rectal immunity and to some extent genital tract immunity

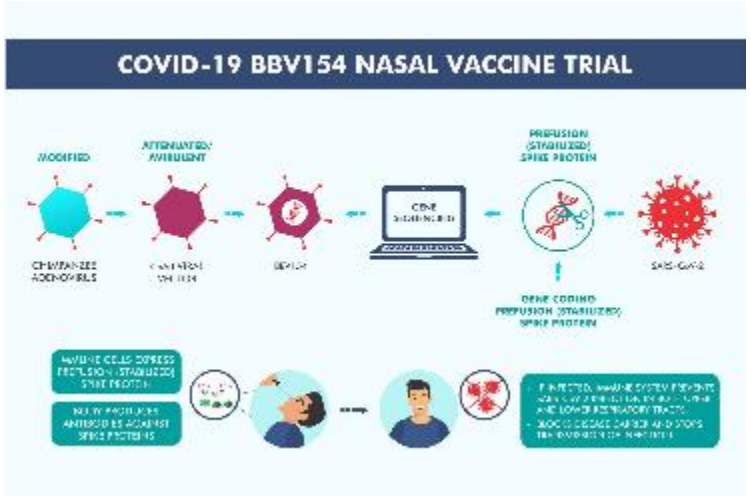
Intravaginal vaccination is the most effective for antibody and T cell immunity in the genital tract.



Playing with compartimentation

Quels sont les vaccins muqueux licenciés?





Bharat Biotech: Chimpanzee adenovirus-based vaccine approved in India for emergency use (2022)

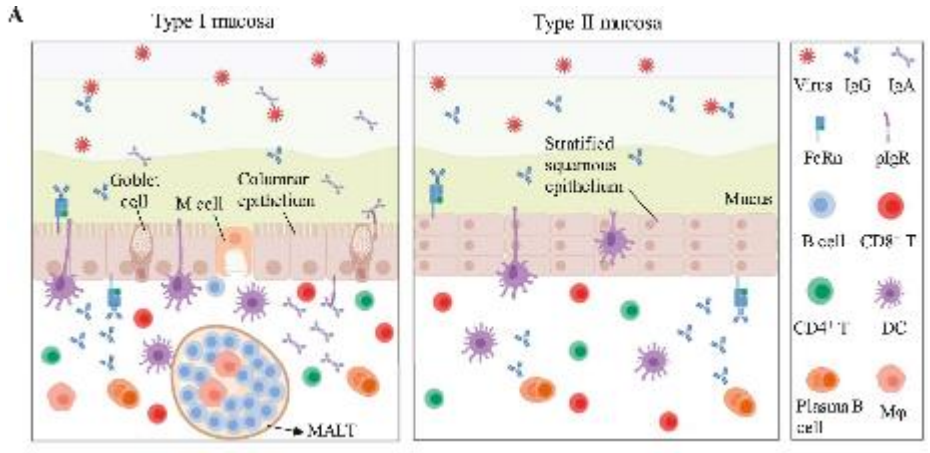
CanSino: Inhaled vaccine against COVID-19 based on Adenovirus 5 approved in China (2022)

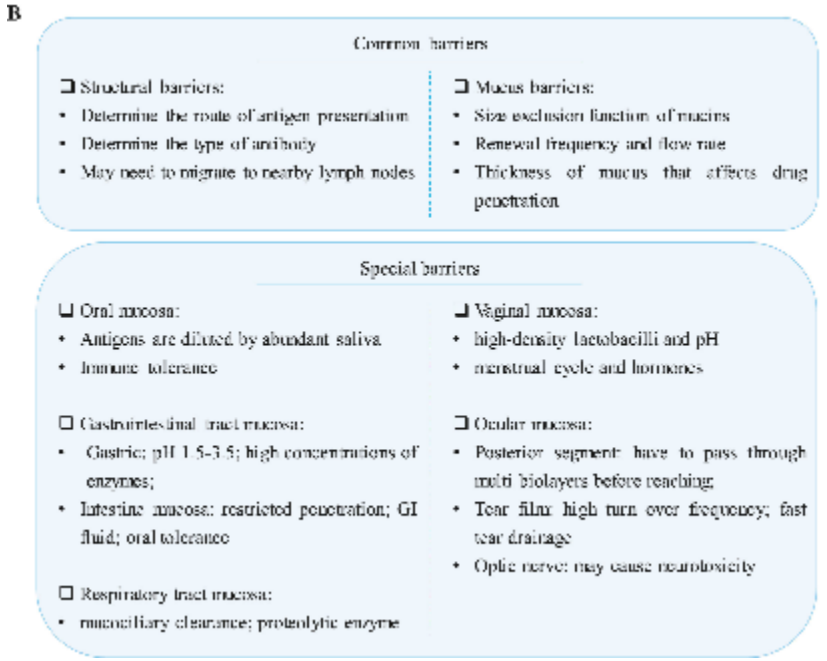
Most mucosal vaccines are based on live attenuated viruses/bacteria: advantages for the delivery and immunogenicity but not suitable for everybody !

Lavelle and Ward, 2022

Comment améliorer la délivrance des antigènes par voie muqueuse?

Challenge: cross the epithelium barrier for inactivated vaccines, subunit vaccines or acid nucleic-based vaccines

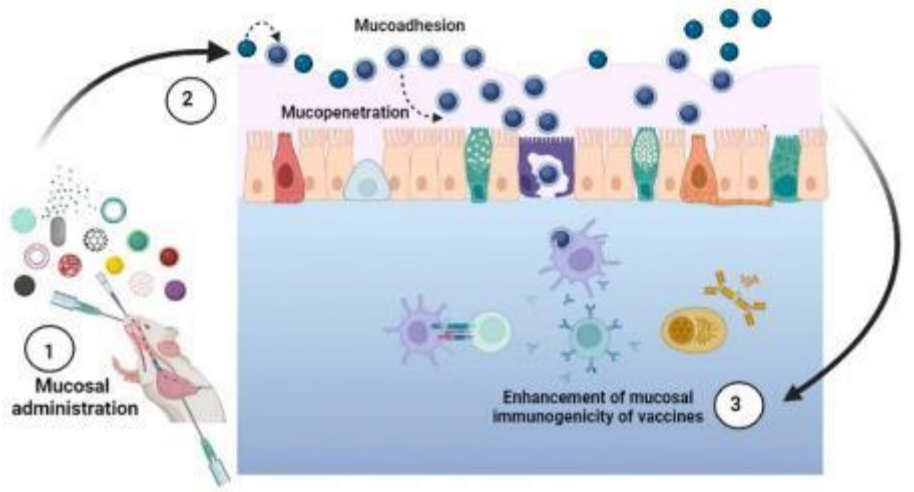




pH, mucus, proteases at the mucosal surfaces (e.g. intestinal surfaces, pH differs based on intestinal region)

- Avoid damaging antigen
- Make sure the antigen will be delivered
- Make sure the antigen will target APCs and that there will be an activation of immune responses in lymph nodes

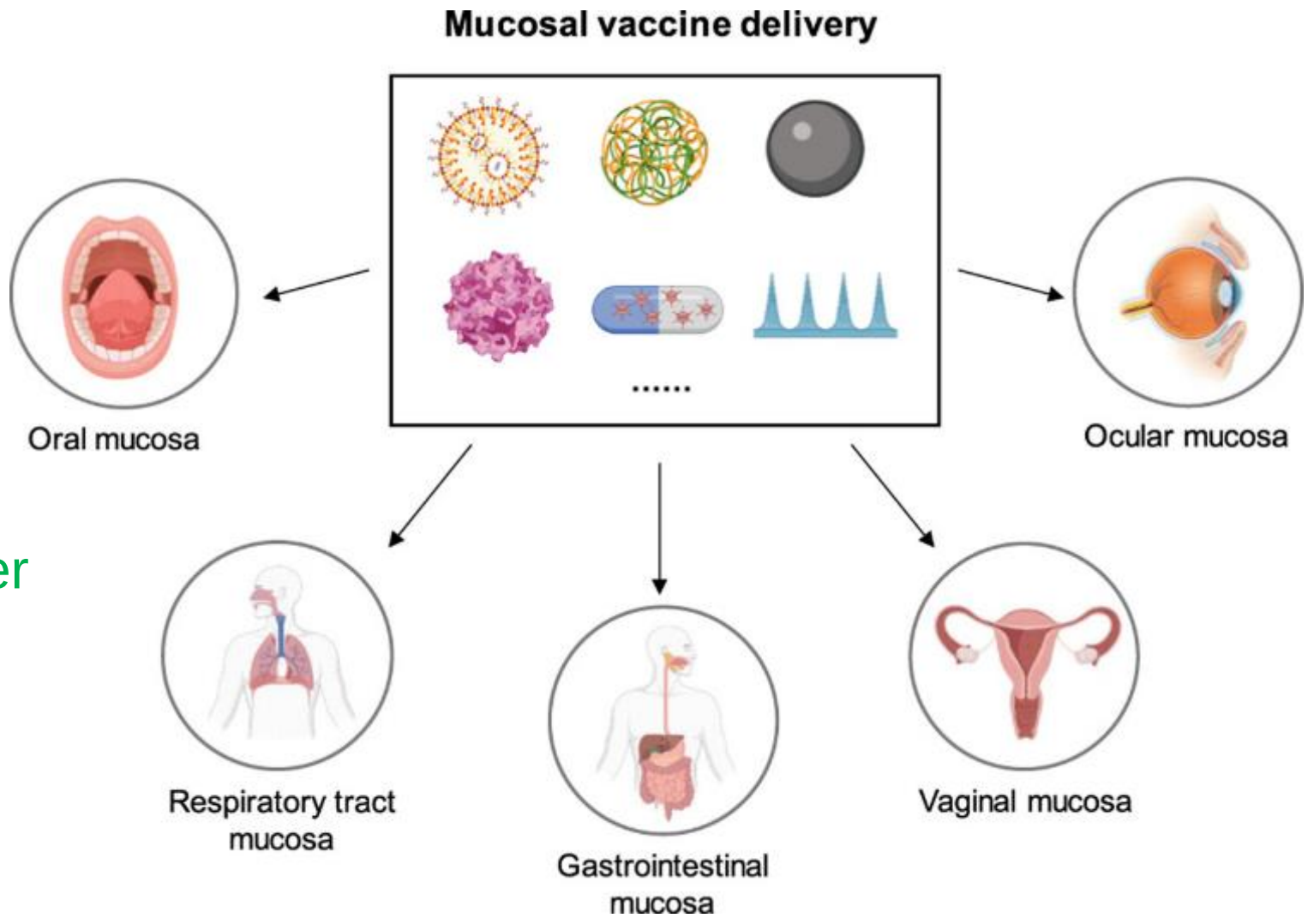
Mucoadhésion ++++



https://www.sciencedirect.com/science/article/pii/S2211383522003136#fig2

Comment améliorer la délivrance des antigènes par voie muqueuse?

Il est nécessaire de designer des nouveaux vecteurs ou systèmes de délivrance physiologique voir physiopathologique (LAV)



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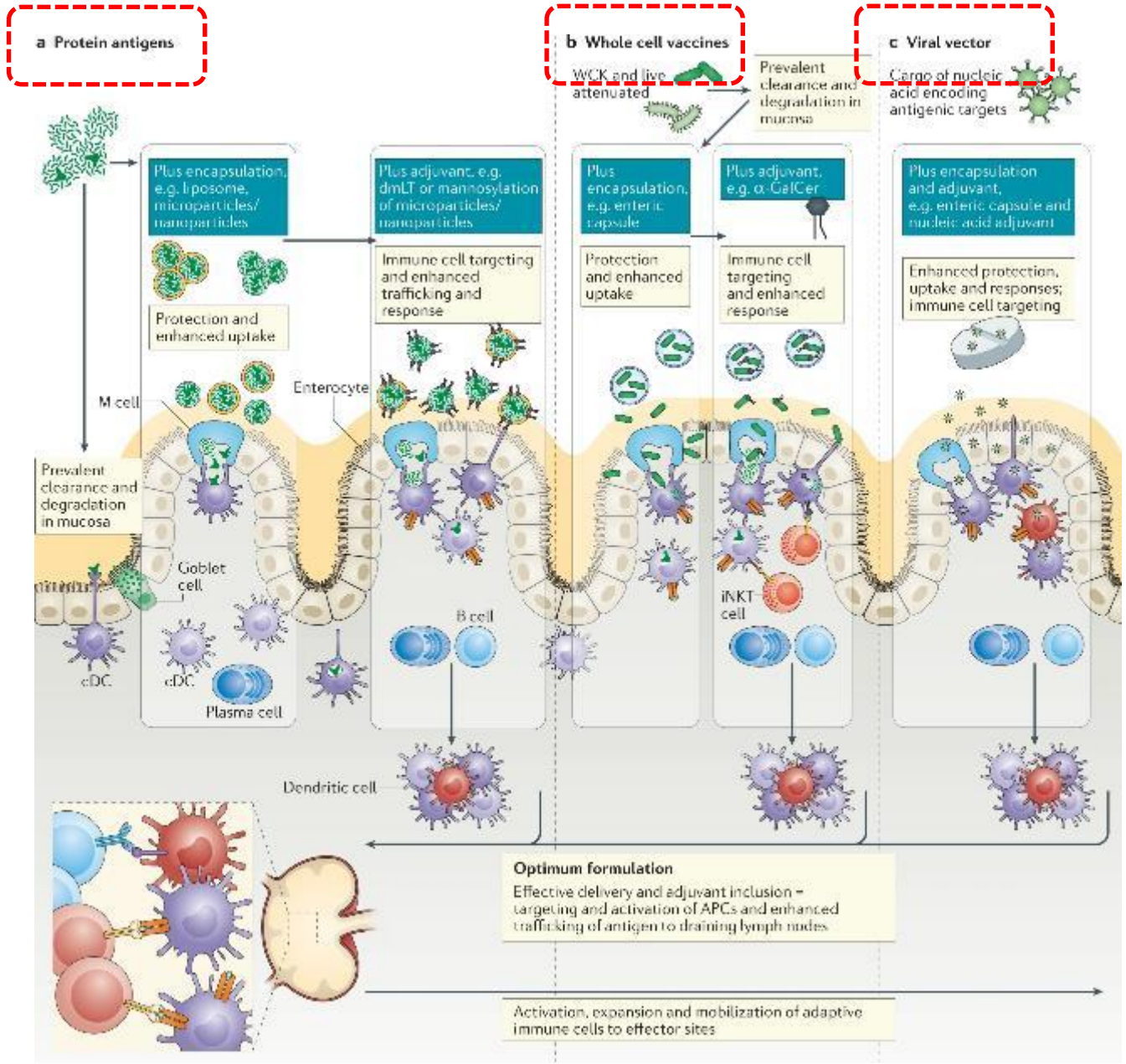
Comment améliorer la délivrance des antigènes par voie muqueuse?



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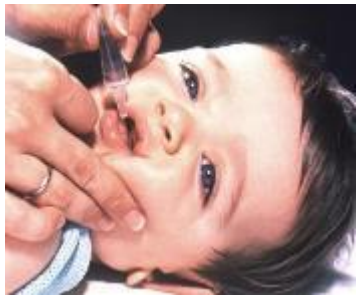
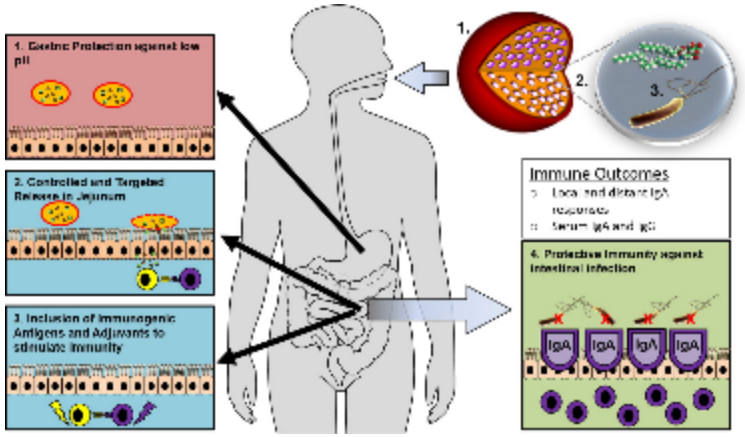
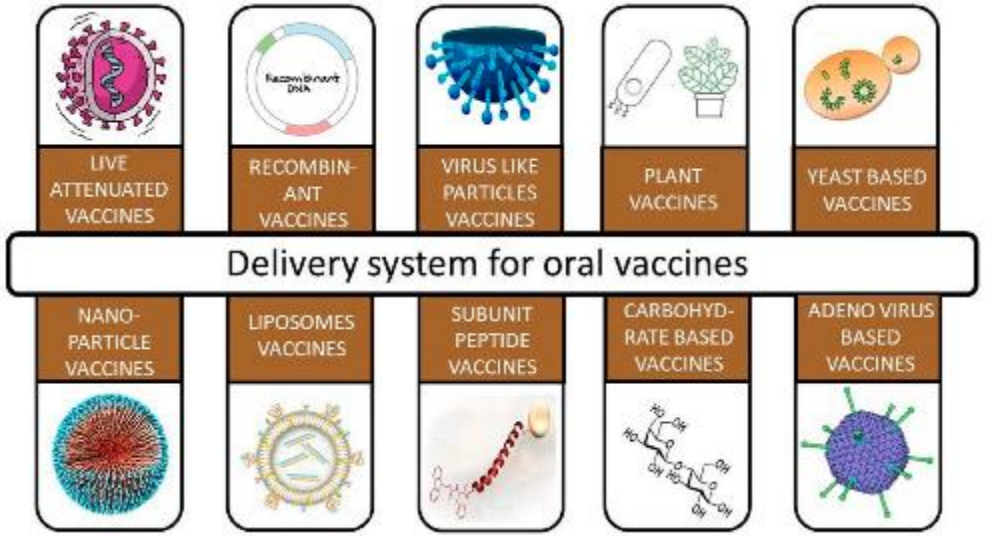
Intérêts de la voie orale

Delivery strategies	Examples	Responses	Model
PLA or PLGA nanoparticles	PLG-encapsulated CS6 antigen from *E. coli*	– IgA antibody-secreting cell responses ↗ – Serum IgG responses ↗	Human
	Lotus tetragonolobus from Winged or Asparagus pea anchored PLGA nanoparticles	– Mucosal and systemic response ↗	Mouse
Liposomes	Plasmid DNA pRc/CMV HBS encoding the small region of hepatitis B surface antigen (HBsAg) into Lipodine liposomes	– IgA responses ↗	Mouse
	L-BLP25 (liposomal formulation of BP25 lipopeptide, MPL® and three lipids)	– Median survival time of 4.4 months longer	Human
Bacterial ghost	*Escherichia coli* O157:H7 bacteria ghosts	– Cellular and humoral immunity ↗	Mouse
Plant lectins/adjuvants	Mistletoe lectin 1, tomato lectin, *Phaseolus vulgaris*, wheat germ agglutinin (WGA), and *Ulex europaeus 1*	– Serum and mucosal antibody responses ↗	Mouse
	Rice-based vaccine that expressed cholera toxin B subunit	– Production of specific serum IgG and IgA antibody after three intra nasal or oral doses ↗	Mouse
Transgenic plants (bioreactors)	Plant-based rotavirus VP6	– High titers of anti-VP6 mucosal IgA and serum IgG ↗	Mouse
	Recombinant LT-B in transgenic corn	– Both serum IgG anti-LT and numbers of specific antibody secreting cells ↗	Human



Infant receiving an oral polio vaccine
(Image courtesy of PAHO/WHO)





*Strong gut IR (IgA, IgG, feces..)

*Particulate Ags or Ag delivery systems which target the GALT

*Highly acceptable but with low immunogenicity.

*Needs of strong mucosal adjuvant.

*Needs to target M cells.

Intérêts de la voie nasale

Delivery strategies	Examples	Responses	Model
Chitosan	Influenza, pertussis, and diphtheria	– Serum IgG responses similar to and secretory IgA levels ↗	Animals/Human (influenza)
	N-trimethyl chitosan (TMC) nanoparticles	– Protection against the appropriate challenge	Mouse
	The nasal diphtheria vaccine with chitosan	– Systemic T cell responses ↗ – Antigen-specific IFN-gamma production ↗ – Th2-type responses ↗ – Protective levels of toxin-neutralizing antibodies ↗	Human
	Aminated gelatin microspheres (AGMS)	– Nasal absorption of insulin ↗	Rat
Cyclodextrins	Dimethyl-beta-cyclodextrin as adjuvants for nasally applied DT and TT	– Specific serum IgG titres ↗	Mouse
Liposomes	Liposomes incorporated insulin and coated with chitosan and carbapol	– Plasma glucose level up to 2 days ↘	Rat
Nanoparticles	Coated poly(anhydride) nanoparticles with either flagellin from *Salmonella enteritidis* or mannosamine	– Serum titers of IgG2a and IgG1 ↗ – TH1 and Th2 response ↗	Mouse
	Nanoencapsulated reporter plasmid (encoding β-galactosidase protein)	– Antibody levels ↗	Mouse
Modified vaccinia virus Ankara (MVA) vector	MVA expressing HIV-1 Env IIIB Ag	– Immune response to the HIV Ag ↗ – Mucosal CD8(+) T cell response in genital tissue and draining lymph nodes ↗ – Mucosal IgA and IgG Abs in vaginal washings ↗ – Specific secretion of beta-chemokines ↗	Mouse

Box 1. Advantages versus disadvantages of nasal vaccines

Advantages

Simple to administer

Appropriate for self-administration

- No need for the involvement of healthcare professionals
- No need for a sterile environment

Suitable for mass vaccination

Needle-free and noninvasive

- No injection-associated pain or fear

Induce both mucosal and systemic immunity

Activate lymphocyte-homing system

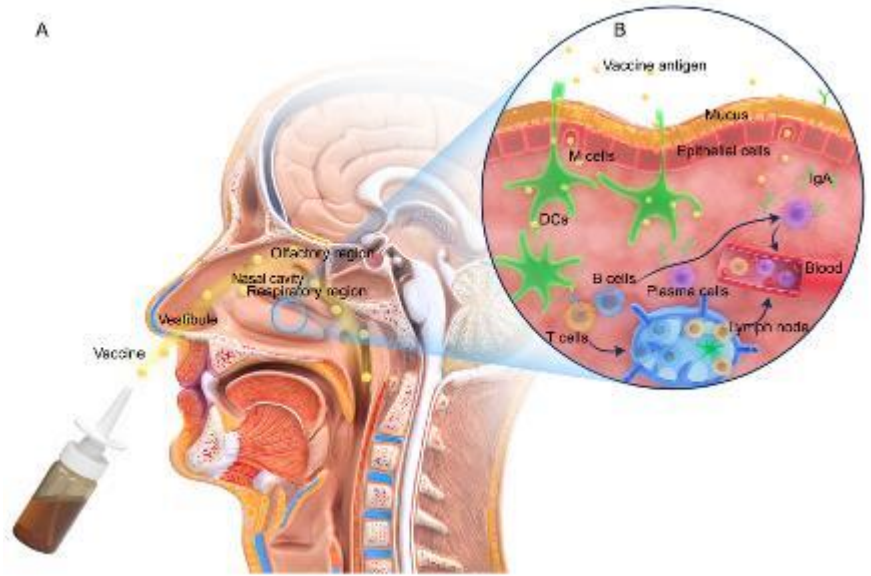
Disadvantages

Rapid clearance by mucociliary barrier and proteases

Limited inoculation volume

Adjuvant and/or other delivery system may be required

Potential side-effects on the central nervous system via dissemination through the olfactory bulb/epithelium



*Nasal vaccine strategies focussed on particulate Ags or Ag delivery systems which target the NALT.

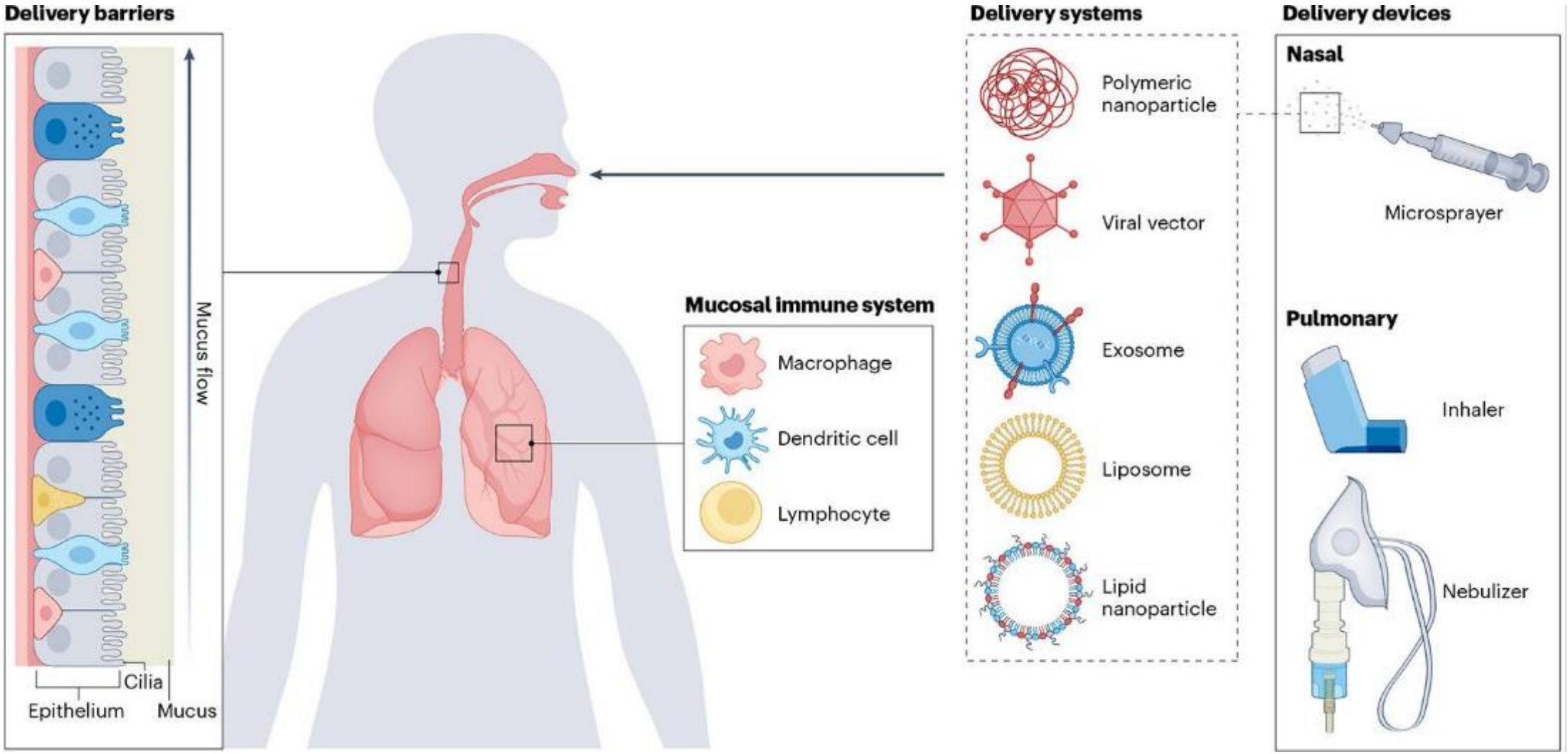
*Nasal vaccines induces both respiratory & vaginal immunity

*Possibility to pass the blood-brain barrier? Toxicity?

*Needs of strong mucosal adjuvant

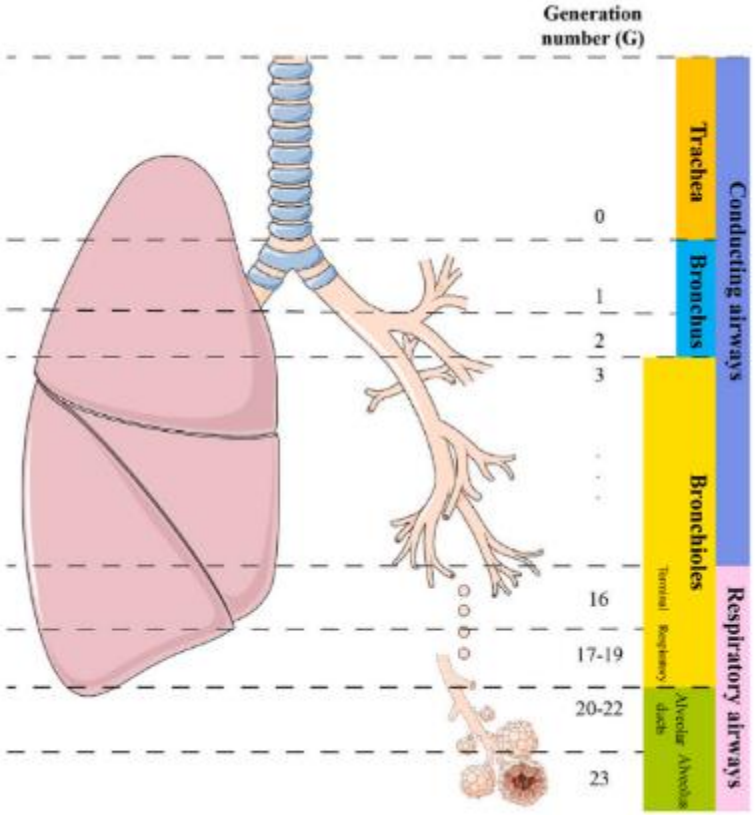
*Acceptability

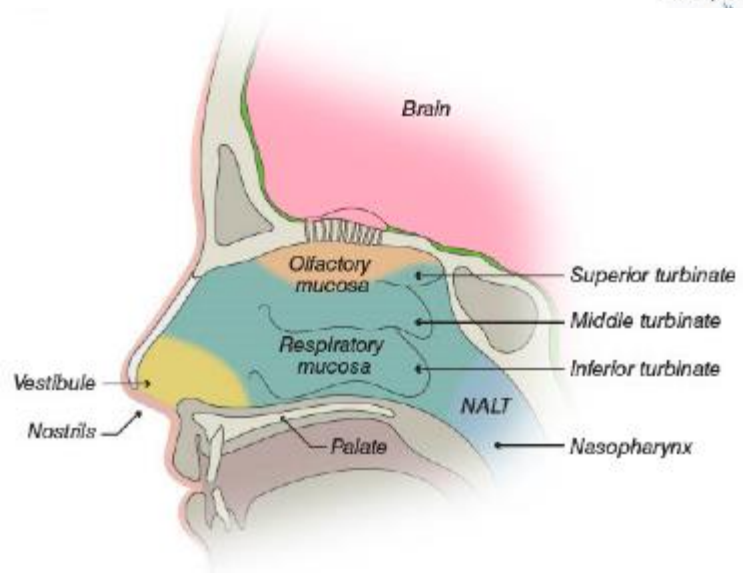
Vaccins par voie nasale et aerosol (besoin de devices+++)

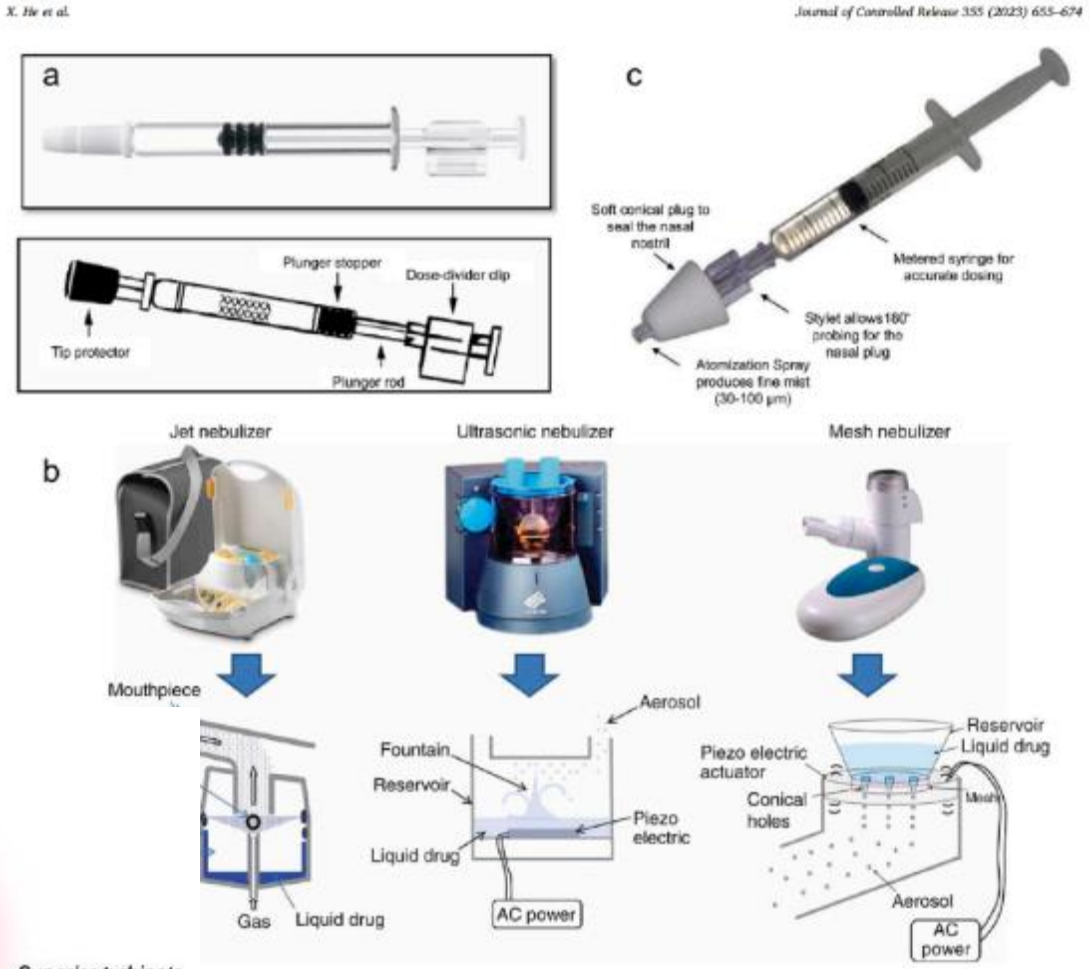


Cahan et al., Nat. Rev. Bioeng. 2023

Spray ou nebulization ? Tout depend ce qu'on cible…



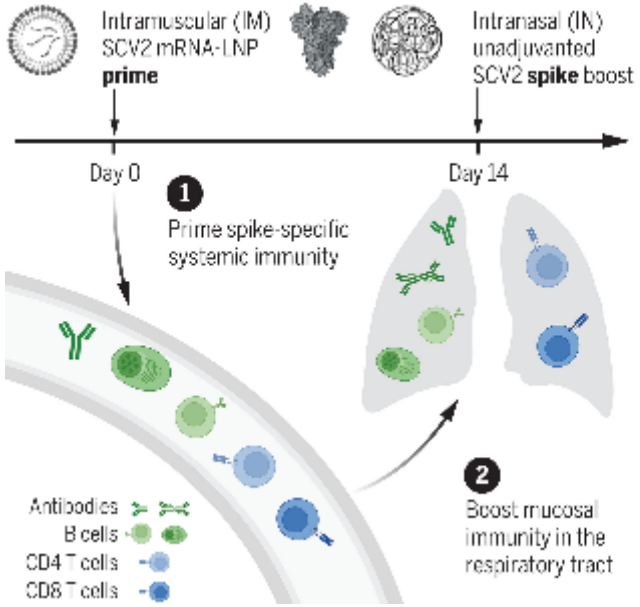


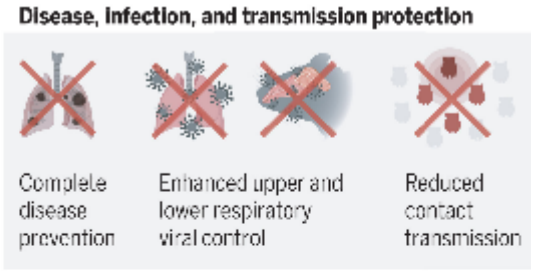


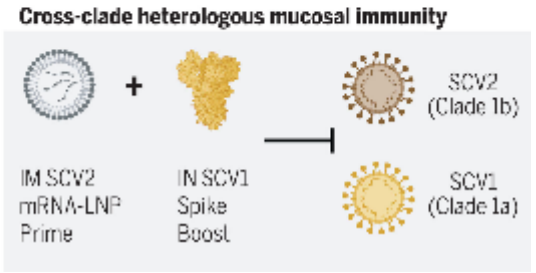
Les devices actuels ne ciblent pas le tissus lymphoide NALT….

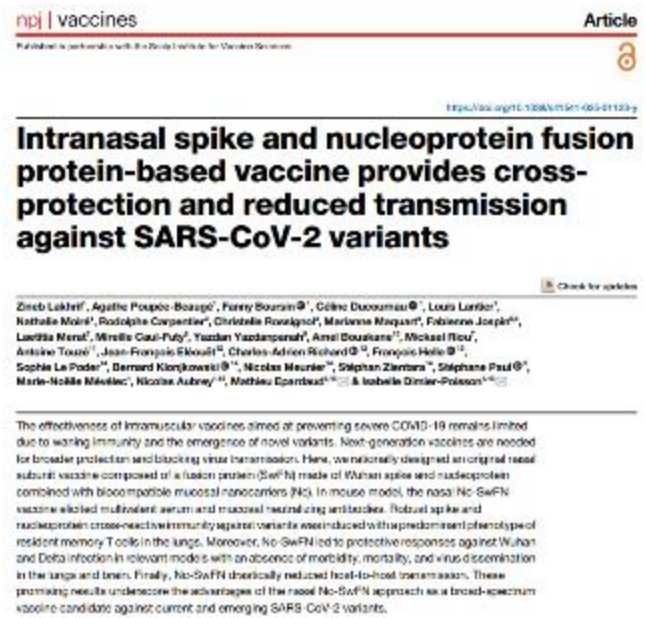
Recipharm
medspray
Nemera
we put patients first
Aptar
pharma

Intranasal vaccines for COVID-19 better as a booster (hybrid)

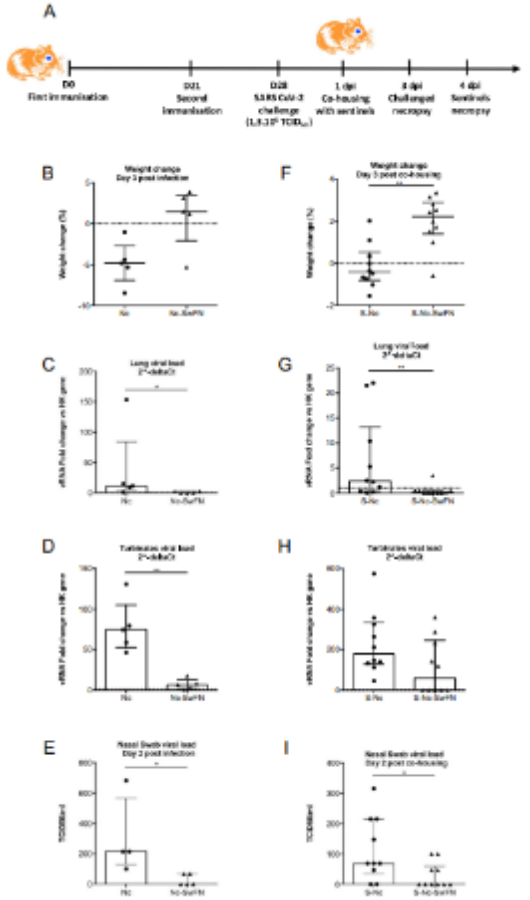


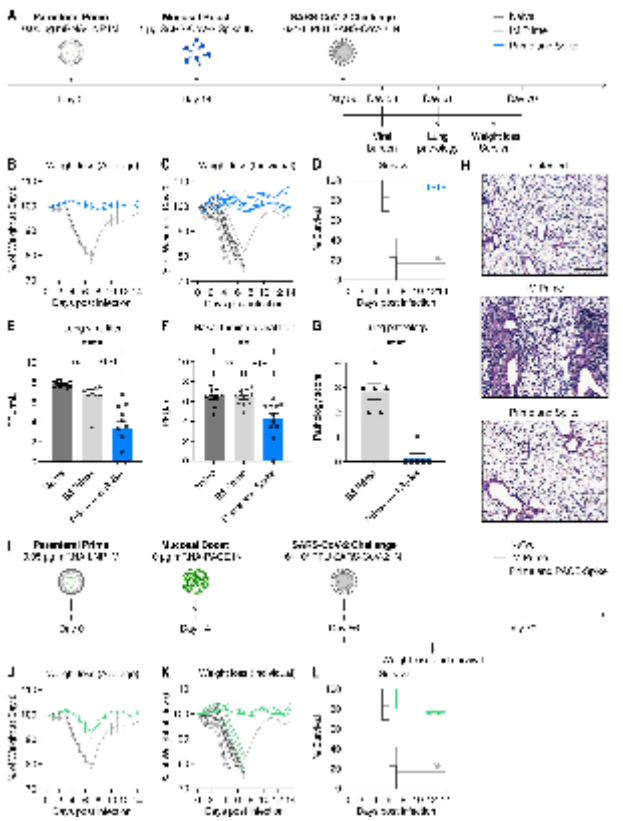





npj | vaccines Article

Intranasal spike and nucleoprotein fusion protein-based vaccine provides cross-protection and reduced transmission against SARS-CoV-2 variants





Higher IgA response
Higher cross-class Nabs
Higher Spike-specific CD4 and CD8
Better memory

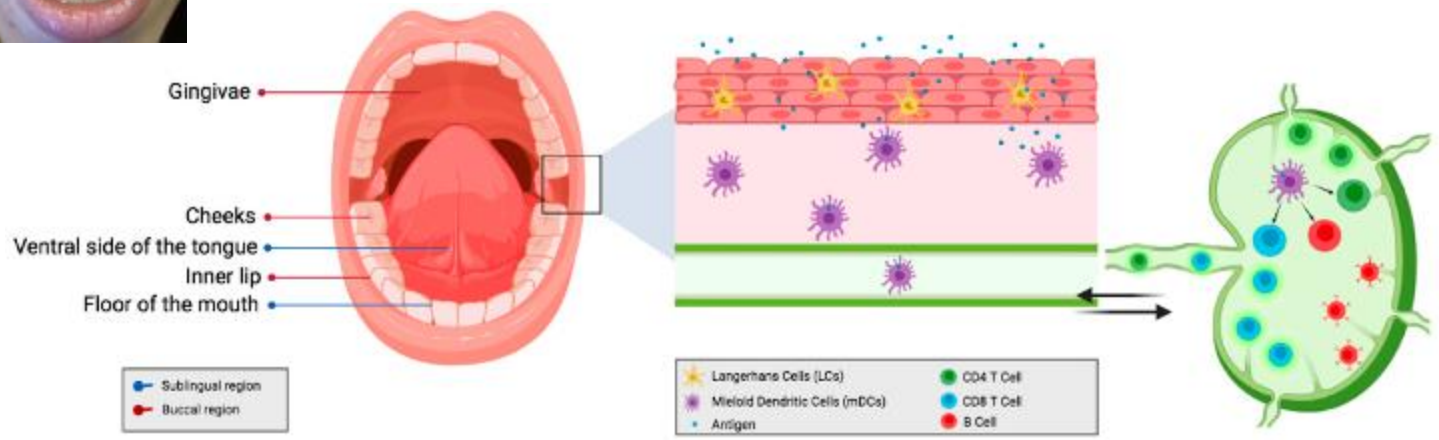
Mao et al., Science 2023
Lakhrif et al., NPJ Vaccine 2025

Meilleure protection
Bloque la transmission

Premier vaccin nasal français

LoValTech
For a pandemic-free world

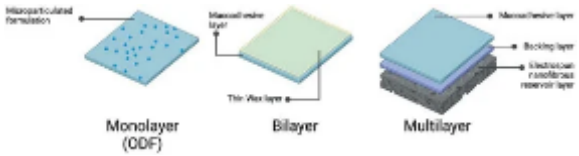
Intérêt de la voie sublinguale?

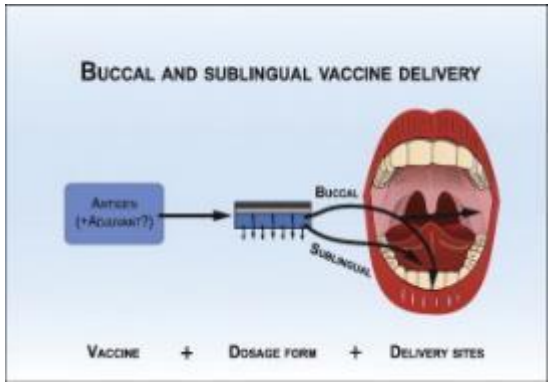


Ag delivery and presentation by sublingual vaccination.
*Epithelial cells express high levels of chemokines.
*Langerhans cells and submucosal DCs are abundant
*DCs migrate into cervical LN to present Ag

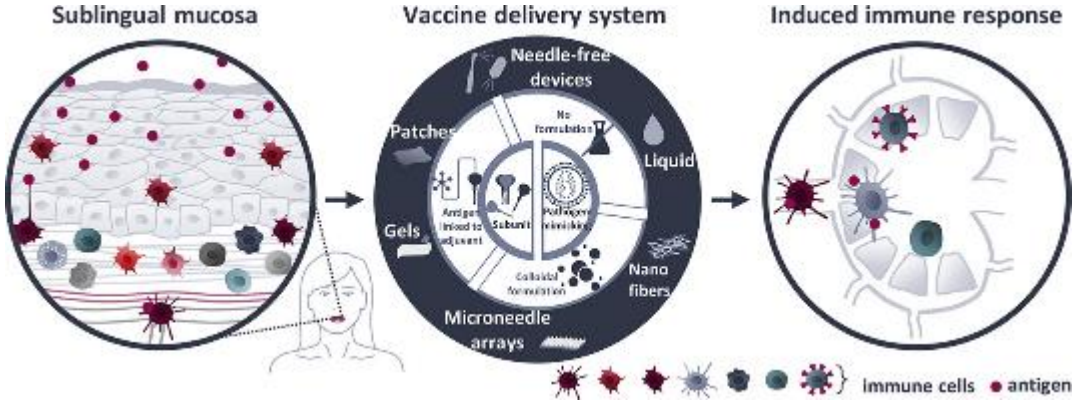
Administration Route	Dosage Form	Main Feature	Antigen/Model Antigen Used
Buccal	Film	Orally disintegrating film loaded with microparticulate vaccine	Live attenuated Measles microparticulate vaccine
Buccal	Film	Bilayer mucoadhesive film	β-galactosidase/plasmid DNA-expressing β-galactosidase
Buccal	Film	Electrospun nanofibrous reservoir multilayer film	Green fluorescent protein loaded nanoparticle and liposomes
Inner lip/ Tongue	Microneedle	Solid stainless steel coated microneedle array	HIV and Ovalbumin Antigens
Buccal	Microneedle	CMC coated solid PLA micromeedle array	Ovalbumin
Oral mucosa	Microneedle	Liposome loaded dissolving microneedle array	BSA

Patch vaccinal





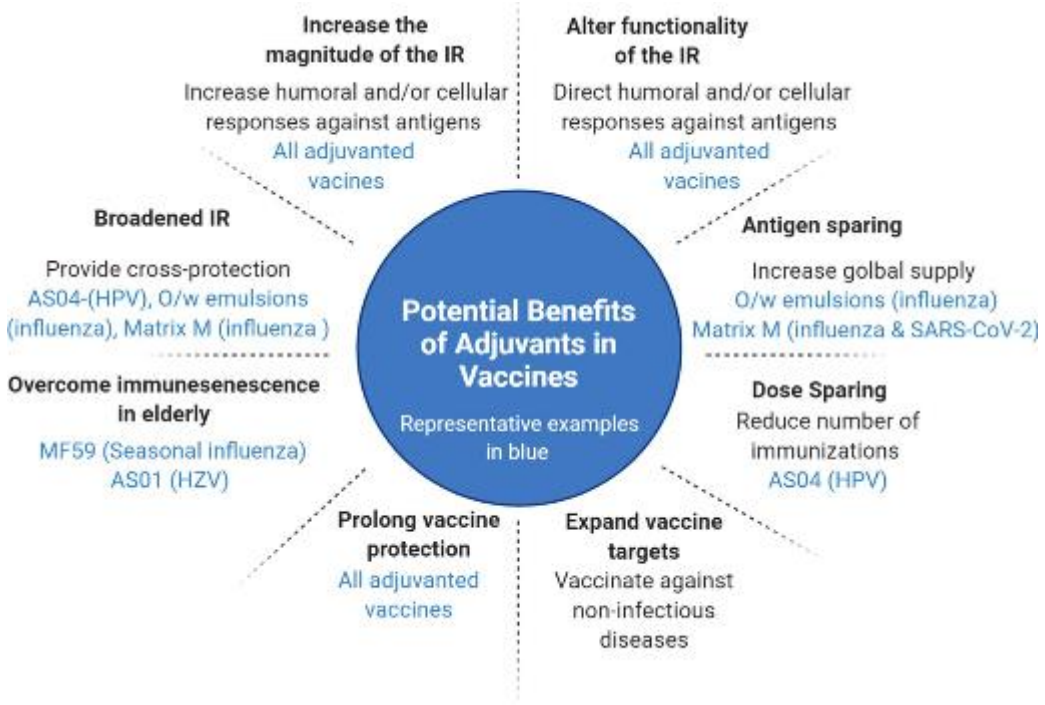
Sublingual vaccination induces both systemic and mucosal Ab responses
Sublingual administration is useful for induction of antibody against Flu
Sublingual administration does not redirect antigens to the CNS
Sublingual vaccination induces genital T/B cell activation
Sublingual vaccination protects mice against genital HPV



Fonctionne bien pour l'allergie ou on recherche de la tolérance

Trincado et al., Vaccines 2021

Intérêt des adjuvants pour augmenter l'immunogénicité



Challenge très important car aucun adjuvant n'est autorisé par voie muqueuse

Table 1

Mode of adjuvant action in vaccines against infectious diseases.

Adjuvant	Description	Mechanism of action	Immune response	Licensed use
Alum	Insoluble particulates of aluminium hydroxide, phosphate or hydroxyphosphate sulphate salts	**Partially defined** Depot effect [15–17] and several DAMPS and PRRs have been implicated, including uric acid [18], host DNA, NETs [19,20], IL-1α [21], IL-33, NLRP3 [22–25] and STING [19]	Abs (functionality can vary depending on the antigen e.g. purified protein [26] vs. inactivated virus [5,27,28])	Routine childhood vaccines and many others
MF59	Oil-dispersed nanoemulsions (mainly squalene), stabilised with polysorbate 80, sorbitan trioleate and citrate buffer	**Partially defined** ATP release from skeletal muscle cells and antigen retention by SCSMΦs in dLNs are involved [29,30]. Systems biology suggests a role for early type-I IFN production [31]. May engage B cells to increase avidity and breadth of the Ab response	Tfh cells and NAbs [32]	Seasonal, pandemic and avian influenza
AS03	Oil-dispersed nanoemulsions (mainly squalene) containing polysorbate 80, α-tocopherol and phosphate-buffered saline	**Partially defined** ER stress in monocytes and NF-kB activation have been implicated [33,34]. α-Tocopherol targets monocytes and macrophages, increasing antigen uptake and cytokine production [33]. May engage naive and memory B cells directly to increase avidity and breadth of the Ab response [35,36]	Tfh cells and NAbs [36,37]	Pandemic and avian influenza [a]SARS-CoV-2
AF03	Oil-dispersed nanoemulsions (mainly squalene) polyoxyethylene-cetostearyl ether, mannitol, sorbitan and phosphate-buffered saline	**Unknown.** Likely triggers cell injury or death that leads to DAMP release	NAbs [37,38]	Pandemic influenza (never marketed)
Virosomes (often referred to as liposomes)	Dispersed lipid vesicles	**Unknown.** Presence of viral factors enhances antigen uptake	NAbs and Th1/Th2 CD4⁺ T cells [39]	Pandemic influenza HAV
AS01 (liposome-based)	Dispersed lipid vesicles containing cholesterol, TLR4 ligand and saponin QS-21	**Partially defined** — MPL triggers TLR4 signalling. QS-21 accumulates SCSMΦs where it promotes caspase-1 activation and HMGB1 release [40]. Synergistic signalling between MPL and QS-21, promoting IL-12, IL-18 and SCSMΦs drives IFN-γ secretion from LN-resident NK and CD8⁺ T cells [41]. This early production of IFN-γ is essential for optimal DC activation. Multiple APC populations are responsible for T-cell activation in dLN [40,42]	Nab and Th1-biased CD4⁺ T cells [43]	Malaria HZV
AS04	Synthetic TLR4 ligand adsorbed to aluminium hydroxide	**Partially defined** In terms of the alum component (see above), MPL triggers TLR4 signalling. Alum prolongs the duration of MPL signalling [44]	Nabs- and Th1-biased CD4⁺ T cells [45]	HBV HPV
CpG ODN (1018)	Soluble TLR9 ligand (oligonucleotide)	TLR9 signalling	NAb- and Th1-biased CD4⁺ T cells [46,47]	HBV SARS-CoV-2

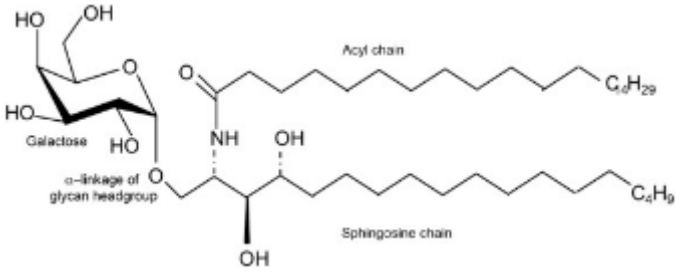
Table 1 *(continued)*

Adjuvant	Description	Mechanism of action	Immune response	Licensed use
Lipid nanoparticles (LNP) containing ionisable lipids	Spherical vesicles comprising helper lipids, cholesterol, polyethylene glycol (PEG) and ionisable lipids	**Unclear** The LNP containing SM-102-ionisable lipids in the mRNA-1273 vaccine activates signals 1 and 2 of the NLRP3 inflammasome in monocytes [48]. In contrast, the LNP containing MC3-ionisable lipids only provides signal 2 for NLRP3 activation [48,49]	Tfh cells and NAbs [48,49]. Unclear whether LNPs contribute to SARS-CoV-2 mRNA-induced CD8⁺ T-cell responses [50]	SARS-CoV-2
Matrix M	Particles comprising Quillaja saponins, cholesterol and phospholipids	**Unknown**	NAbs, Th1- biased CD4⁺ T cells [12,51]	SARS-CoV-2
Alhydroxiquim-II	**Imidazoquinoline** (IMQ) **adsorbed on alum**	**Partially defined** In terms of the alum component (see above), IMQ is a small molecule targeting TLR7 and TLR8 signalling	NAbs, Th1 CD4 + T cells [13,52]	SARS-CoV-2

[a] Under emergency authorisation. Abbreviations: IL, interleukin; NETs, neutrophil extracellular traps; HMGB1, high-mobility-group box 1; IRE1α, inositol-requiring enzyme-1a; Abs, antibodies; Nabs, neutralising Ab; HAV, hepatitis-A virus; HBV, hepatitis-B virus; HPV, human papilloma virus; ER, endoplasmic reticulum; imidazoquinoline, IMQ.

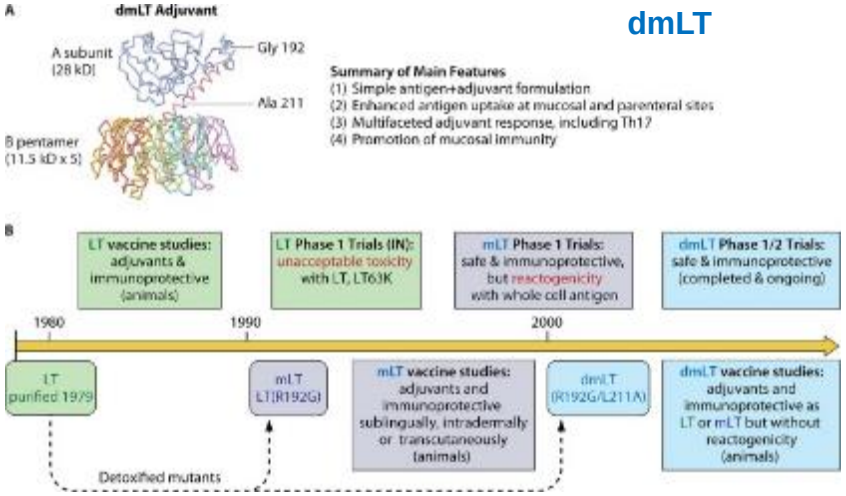
Quels sont les candidats adjuvants muqueux?

Classification	Composition	Target
Bacterial adjuvants	Cholera toxin (CT)	GM1 gangliosides
	CTA1-DD	B cell receptor
	Escherichia coli heat-labile toxin (LT)	GM1 and GM2 asialo-GM1 gangliosides
	Bacillus anthracis Edema Toxin (EdTx)	Anthrax toxin receptors
	Monophosphoryl lipid A (MPL)	TLR4
	Pertussis toxin (PT)	Unknown
	Lipoprotein/lipopeptide	Various
	Probiotics	Various
Nucleic acid adjuvants	CpG oligodeoxynucleorides (ODN)	TLR9
	Polynosinic acid-polyeytridylic acid, polyI:C	TLR3
Cytokine adjuvants	IL-1, IL-2, IL-4, IL-12, IL-18 IFN-α, and IFN-γ	Various
Particles adjuvants	Chitosan	Tight Junctions
	Polyacrylic acid, PAA	Unknown
	Virus-like particle (VLPs)	Unknown
	ISCOMs	DCs



A synthetic glycolipid derived from structure-activity relationship studies of galactosylceramides isolated from the marine sponge *Agelas mauritianus*.

Activation of unconventional T cells:
Alpha-Galactosylceramide which is an iNKT cell activator (oral)

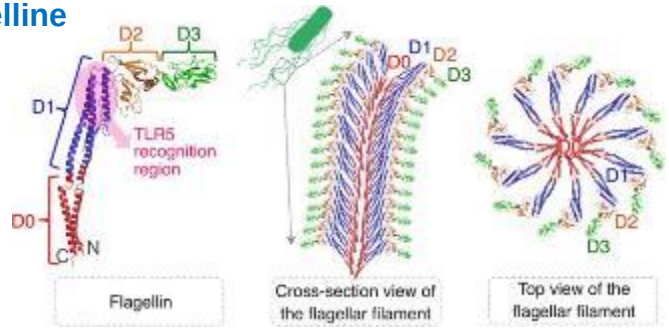


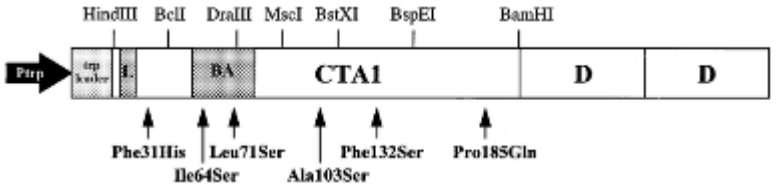
dmLT

Completed and ongoing clinical trials with dmLT adjuvant*

Pathogen: antigen	Route	Study design (Clinicaltrials.gov ID [reference (if available)])	Population; status; results
None; none	p.o.	Phase 1, escalating dose safety study [NCT01167465 [79]]	U.S. adults; completed; no detectable SAE
ETEC whole-killed (ETVAX)	p.o.	Phase 1, whole cells + 10 or 25 μg dmLT (EudraCT no. 2011-003238-11 [71])	Swedish adults; completed; 10 μg dmLT enhanced responses to less immunogenic antigens
ETEC live-attenuated (ACE527)	p.o.	Phase 1 and 2b, 25 μg dmLT with live-attenuated ETEC (NCT01720251 [72])	U.S. adults; completed; dmLT enhanced protective efficacy from oral challenge 6–7 mo postimmunization
None; none	s.l.	Phase 1, escalating dose safety study [NCT03252249]	U.S. adults; completed
None; none	i.d.	Phase 1, escalating dose of 0.1, 0.3, 1, or 3 μg dmLT [NCT02511405]	U.S. adults; recruiting
ETEC whole killed (ETVAX)	p.o.	Phase 1 and 2 escalating dose of ETVAX + 25, 5, or 10 μg dmLT [NCT02531932]	Bangladesh infants, toddlers, children, adults; completed
ETEC subunit (CSe)	i.m.	Phase 1, escalating dose safety study [NCT02494674]	U.S. adults; recruiting
ETEC whole-killed (ETVAX)	p.o.	Phase 2b, ETVAX plus 10 μg dmLT (EudraCT no. 2016-002690-35)	Finnish adult travelers to Benin; ongoing

Flagelline



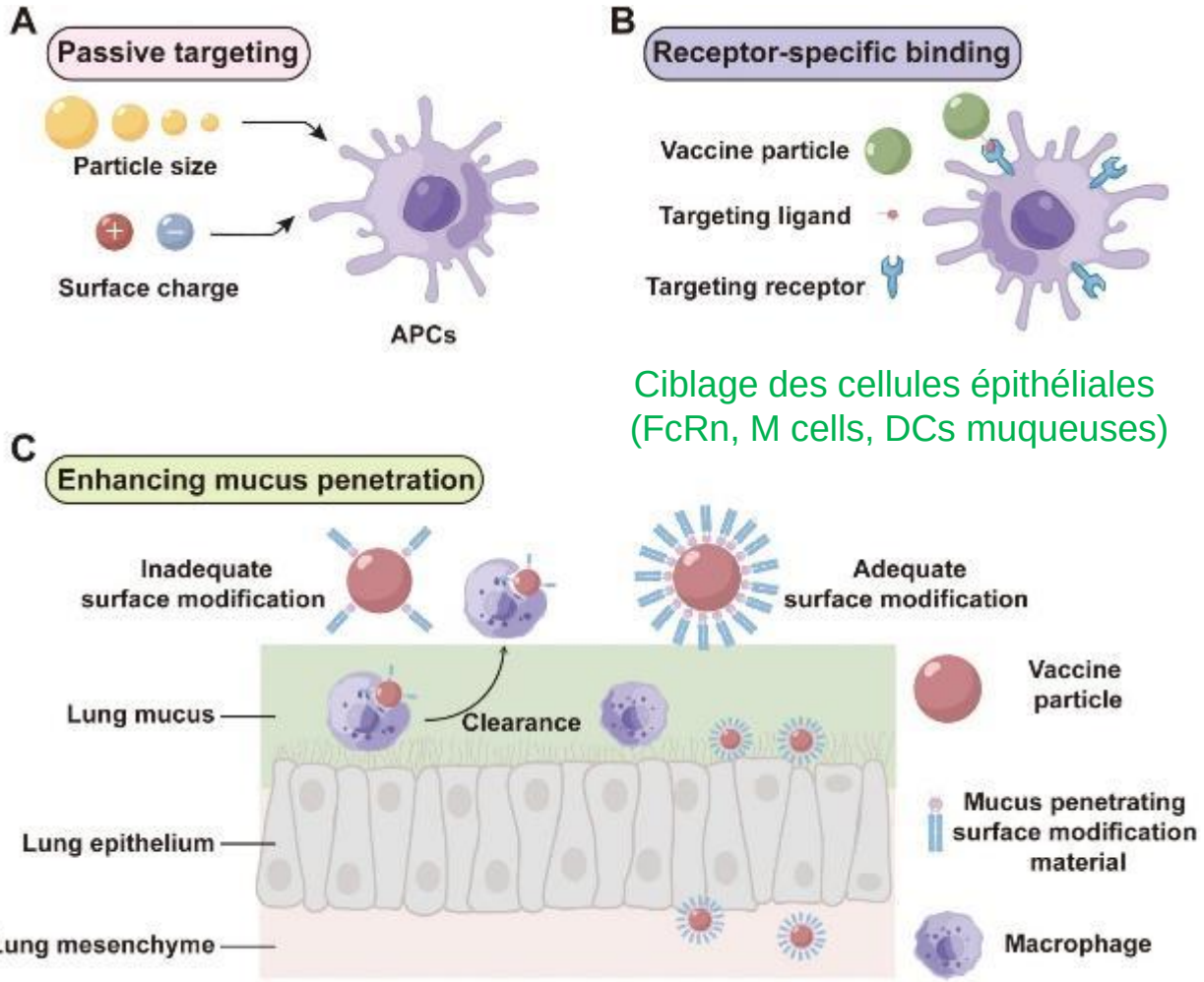


CTA1-DD: non-toxic CT derivative was also developed as CTA1-DD which is a fusion between the A subunit of CT and the D-fragment of the *Staphylococcus aureus* protein A

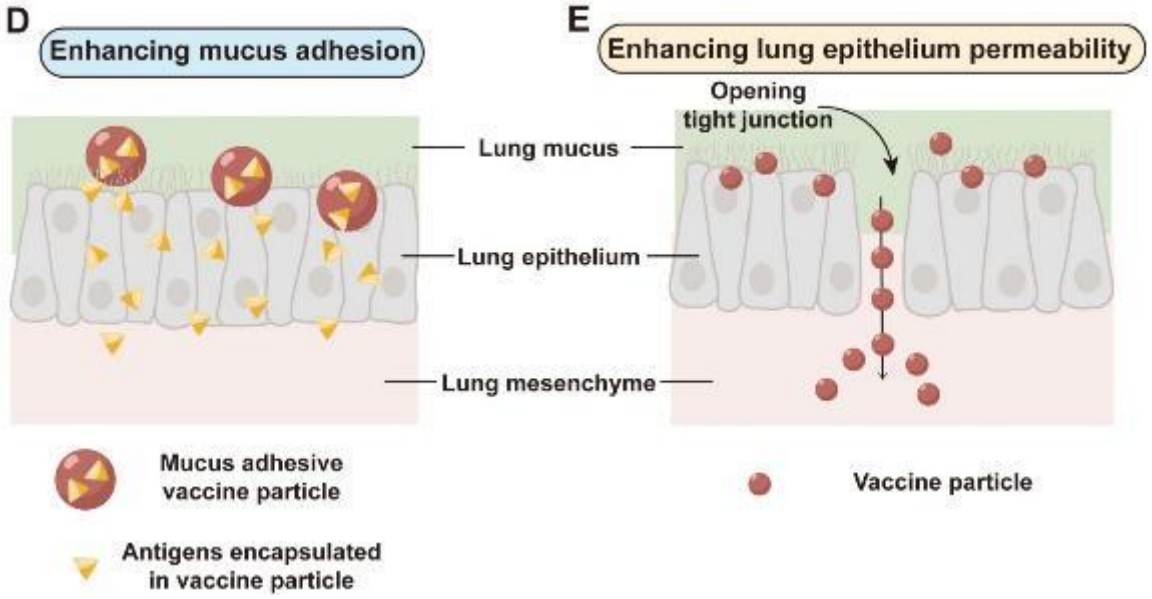
Agren et al 2017, J Immunol

Clements et al, 2018, mSphere. 2018 Jul-Aug; 3(4): e00215-18.

Intérêt du ciblage muqueux pour augmenter l'immunogénicité



Ciblage des cellules épithéliales
(FcRn, M cells, DCs muqueuses)



Ciblage par anticorps
Utilisation de sucres types lectines
Utilisation de mucine / mucoadhésifs

Comment augmenter la durée de la réponse muqueuse? example Dukoral and Sanchol (oral vaccines against cholera)

Measures of Immune Response and Duration of Immunity/Protection

Dukoral: The primary immunization series provides short-term protection (6 months) against cholera, with an overall protective efficacy of 85–90% and 50–60% for 2 years.[23] For adults and children >6 years of age, protective efficacy averages 63% over a 3-year period without a booster dose, but drops to <50% after the first year. The protective efficacy against classic and El Tor cholera is similar in the first 6 months. A large oral inoculum of bacteria can overwhelm even an optimal response to vaccine. Travelers should be advised to observe careful food and water precautions, regardless of vaccination status.

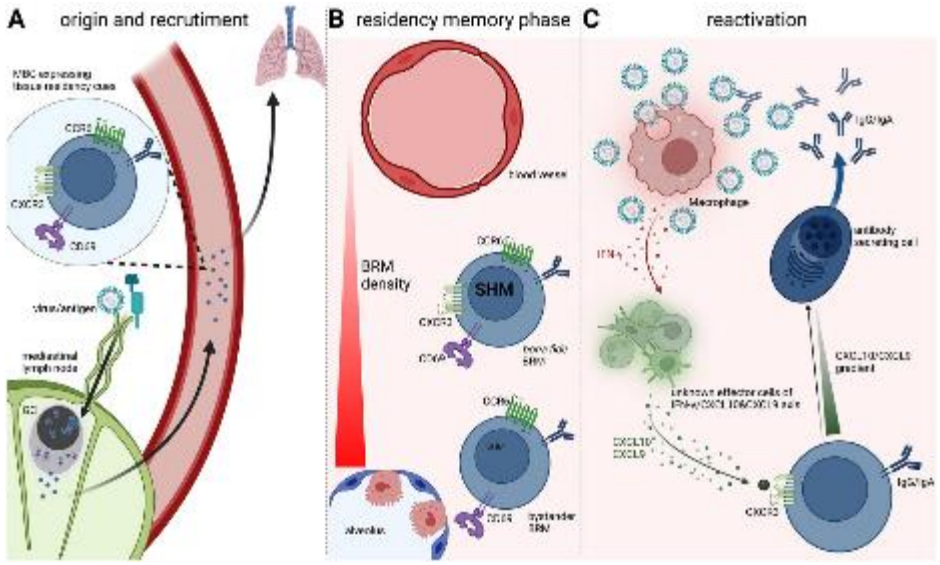
Shanchol: Results of clinical trials show a good safety profile, and a large Phase III efficacy trial in India confirmed a clinical efficacy of 67% after 2 years. An ongoing investigation will provide data on 5-year efficacy.

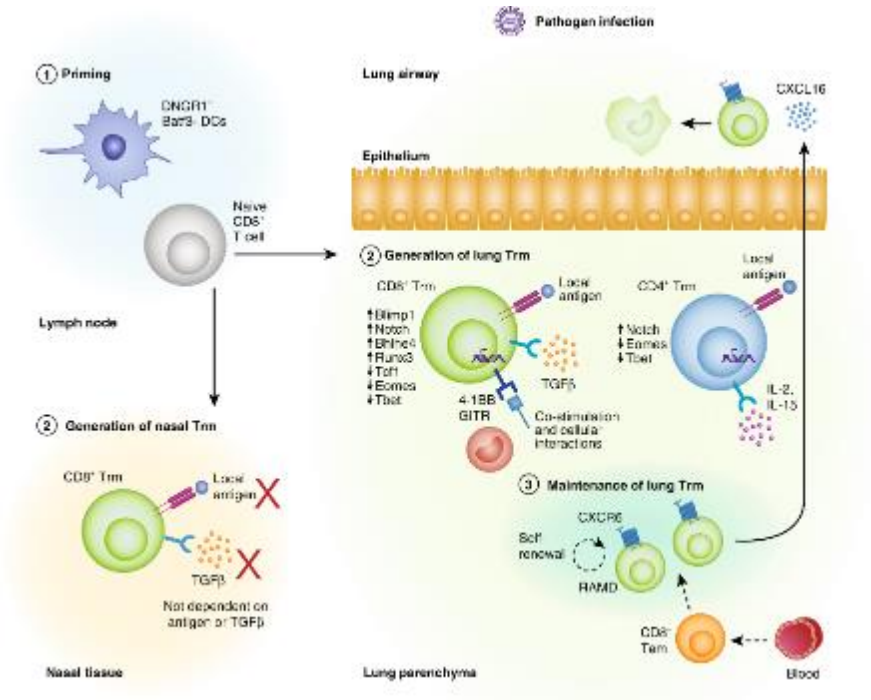
Special Adult Travel Vaccines. Joseph Torresi, Herwig Kollaritsch, in Travel Medicine (Third Edition), 2013

Induire une réponse muqueuse mémoire et notamment par l'induction des cellules residentes mémoires (adjuvants, strategies de prime boost mucosal et systémique?)

Comment augmenter la durée de la réponse muqueuse?

	Trade Name	Approved	Antigen	Type of vaccine	Form/Adm	Immunological mechanism
era	Dukoral®	2003 CANADA	*Vibrio cholerae* +CTB	Live attenuated	Aqueous/Oral	Antibacteria, toxin -specific and LPS-specific IgA
	Euvichol Shanchol	2013 WHO	*Vibrio cholerae*	Inactivated	Aqueous/Oral	
	Vaxchora™	2015 FDA	*Vibrio cholerae*	Live attenuated	Aqueous/Oral	
Acute gastroenteritis	Vivotif®	2013 FDA	*Salmonella typhimurium*	Live attenuated	Capsule/Oral	Mucosal IgA, systemic IgG and CTL responses
Influenza	Fluenz™/FluMist®	2003 FDA	Influenza A and B viruses	Live attenuated	Spray/Nasal	Mucosal IgA and CTL responses
Polio	OPV (b/m/tOPv)	1961 FDA	Poliovirus	Live attenuated	Aqueous/Oral	Mucosal IgA, systemic IgG
nfant liarrhea	RotaTeq®	2006 FDA	Rotavirus	Live reassortant	Aqueous/Oral	Mucosal IgA, systemic neutralizing IgG
	Rotarix	2008FDA	*RIX4414 strain*	Live attenuated	Aqueous/Oral	
respiratory disease	Adenovirus Type 4 and 7 Vaccine	2015 FDA	Adenovirus Type 4 and 7	Adenovirus vector vaccine	Aqueous/Oral	





Etude de la réponse cellulaire muqueuse toujours difficile à faire (quel type de prélèvement etc..)
Corrélats de durée de protection? Trm? Brm?

Isticato, R. Bacterial Spore-Based Delivery System: 20 Years of a Versatile Approach for Innovative Vaccines. *Biomolecules* **2023**, *13*, 947.

Quels sont les enjeux de la vaccination muqueuse?

1) La vaccination systémique n'induit pas de réponse muqueuse (sauf chez les préinfectés, *Gorochov et al., JAMA Net 2024*)

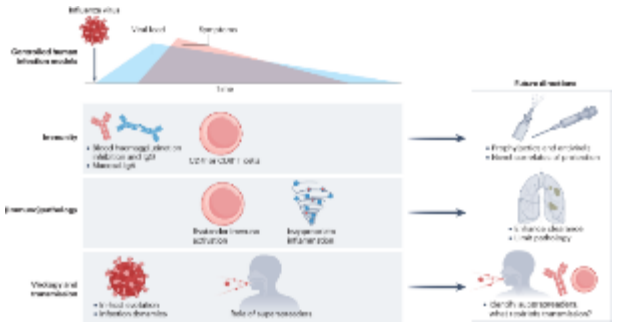
2) Améliorer les vaccins muqueux (devices, adjuvants, formulation, stratégie, rôle du microbiote)

3) Améliorer l'immunomonitoring des réponses muqueuses et en particulier des réponses cellulaires (Trm, Tfh)

4) Identifier des corrélats de protection spécifiques des vaccins muqueux (IgA, Trm, Brm, Tfh etc..)

5) Disposer de modèles d'évaluation *in vitro* et *in vivo* notamment pour évaluer l'efficacité sur la transmission et sur la mémoire immunitaire muqueuse

Intérêt du CHIM



Echantillons

In vitro models