CellPress

Review

Food-Grade Organisms as Vaccine Biofactories and Oral Delivery Vehicles

Sergio Rosales-Mendoza,[1,*] Carlos Angulo,[2,*] and Beatriz Meza[2]

The use of food-grade organisms as recombinant vaccine expression hosts and delivery vehicles has been explored during the past 25 years, opening new avenues for vaccinology. Considering that oral immunization is a beneficial approach in terms of costs, patient comfort, and protection of mucosal tissues, the use of food-grade organisms can lead to highly advantageous vaccines in terms of costs, easy administration, and safety. The organisms currently used for this purpose are bacteria (*Lactobacillus* and *Bacillus*), yeasts, algae, plants, and insect species. Herein, a comparative and updated scenario on the production of oral vaccines in food-grade organisms is provided and placed in perspective. The status of clinical evaluations and the adoption of this technology by the industry are highlighted.

Trends

Recombinant food-grade organisms are being used for vaccine production and delivery as food-grade vaccines (FGVs).

The concept of FGVs is highly advantageous in terms of costs, administration, and safety.

FGVs are currently produced in some bacteria, yeast, algae, plants, and insect species.

FGVs with improved immunogenicity are being successfully explored for painless mucosal administration.

Several FGVs are under clinical evaluation, and the current adoption of this technology by the industry indicates a potential to benefit global healthcare systems.

Vaccine Production and Delivery Using Food-Grade Organisms

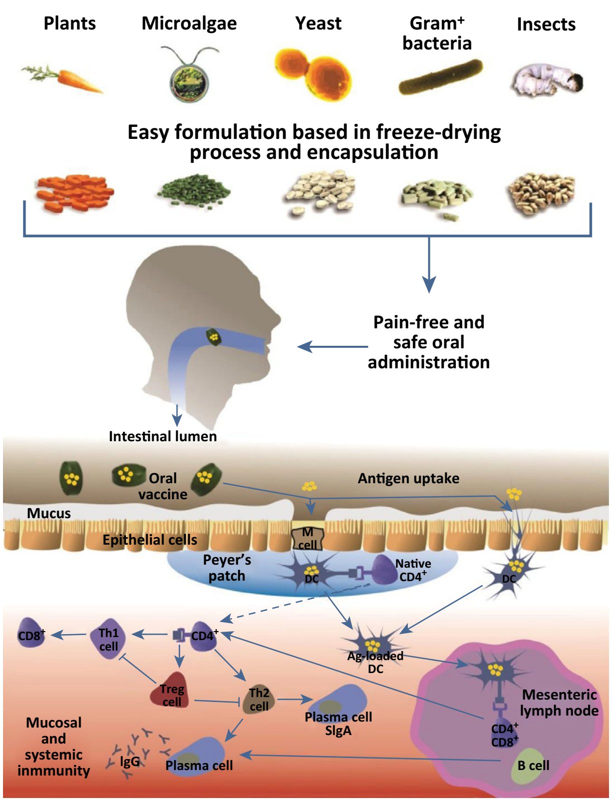
Vaccination has greatly reduced the burden of infectious diseases and has led to eradication of smallpox, near eradication of polio, and the prevention of billions of deaths [1]. Despite the success of vaccination programs against formerly fearsome diseases, many efficacious vaccines are still needed, especially in developing countries. While distinct platforms for subunit vaccine synthesis are well established, there are some clear limitations to their applicability in global vaccination. Although *Escherichia coli* shows very high productivity, the antigens must be purified to eliminate endotoxins before safe use in humans and this host often produces insoluble, incorrectly folded, nonfunctional proteins [2]. Production systems using insect or animal cell lines require expensive culture media, and have longer production times than those required for microbial systems [3]. Besides production costs, the vaccination programs should contemplate the cost for distribution (cold chain) and administration (trained personnel) that hamper vaccination coverage [4]. In terms of easy administration and immune protection at local mucosal tissues, oral vaccination is an attractive approach to fight against several diseases, particularly in poor countries where vaccines are more needed [5]. Thus far several oral vaccine formulations have been developed; however, these are composed of attenuated forms of disease-causing pathogens [6], and thus the risk of reversion into pathogenic forms exists [7]. In addition, the production of these vaccines usually requires high-level biosafety facilities that are not needed for producing subunit vaccines. Under this scenario, new approaches for the development of subunit vaccines that can surmount the above-mentioned limitations are necessary.

During the past two decades, food-grade organisms have attracted the attention in vaccinology as both production hosts and delivery vehicles. Looking to minimize costs, the production of subunit vaccines becomes advantageous if an innocuous organism is used as the expression host for the recombinant vaccine and, moreover, if its biomass can support a straightforward

[1]Laboratorio de Biofarmacéuticos Recombinantes, Facultad de Ciencias Químicas, Universidad Autónoma de San Luis Potosí, Avenida Dr. Manuel Nava 6, SLP, 78210, México
[2]Grupo de Inmunología y Vacunología, Centro de Investigaciones Biológicas del Noroeste, SC, Instituto Politécnico Nacional 195, Playa Palo de Santa Rita Sur, La Paz, Baja California Sur, CP 23096, México

*Correspondence: rosales.s@fcq.uaslp.mx (S. Rosales-Mendoza) and eangulo@cibnor.mx (C. Angulo).

 http://dx.doi.org/10.1016/j.tibtech.2015.11.007



Trends in Biotechnology

Figure 1. Schematic Description of the Rationale of Food-Grade Vaccines. This type of vaccine constitutes a low-cost, easy to administer, and potentially efficacious vaccine. Mucosal and systemic immune responses are induced as consequence of antigen (Ag) translocation by M (microfold) cells or dendritic cells (DC) which mediate antigen presentation to lymphocytes for the induction of adaptive immune responses whose effector mechanisms involve IgA and IgG production and the recruitment of cytotoxic lymphocytes. Abbreviation: SIgA, secretory IgA.

formulation of an oral vaccine. Thus, food-grade organisms can serve at the same time as biofactory and oral delivery vehicle for subunit vaccines, avoiding costly purification processes. We term this immunization approach **food-grade vaccines** (FGVs, see Glossary), which represent an outstanding means to generate low-cost and easy to administer vaccines with the potential to facilitate universal coverage. Under this notion, several systems have been adopted for the production of FGVs, including bacterial species, yeasts, algae, plants, and insects (Figure 1). Each system possesses particular attributes and limitations that should be evaluated in selecting the most appropriate platform for vaccine development (Table 1).

Description of FGVs

Lactic Acid Bacteria-Based Vaccines

Lactic acid bacteria (LAB) are Gram-positive, non-spore-forming cocci, coccobacilli or rods, lacking catalase activity, which ferment glucose primarily to lactic acid or to lactic acid, CO_2, and

Table 1. Comparison of Food-Grade Organisms Used in Oral Vaccine Development

Organism	Diversity of Genetic Tools	Growth Rate	Post-Translational Modification Capacity	Glycoengineering Tools	Adjuvant Producer	Industrial Production Experience
Plants	++++	+	++++	++++	++++	++++
Yeast	++	+++	++	++++	+++	++++
B. subtilis/lactic acid bacteria	+++	++++	+	+	++	++++
Silkworm	++	++	++++	+++	++	++++
Algae	++	++++	++++	+	++++	++++

Glossary

***Bacillus subtilis*:** a Gram-positive, catalase-positive bacterium which holds GRAS status; it can form a tough, protective endospore that allows it to tolerate extreme environmental conditions, features that are exploited in the development of convenient oral vaccines.

***Bombyx mori*:** a silkworm species with GRAS status, currently used for the production of FGVs with promising prototypes.

***Chlamydomonas reinhartdii*:** a GRAS eukaryotic algae widely used for the production of biopharmaceuticals; in particular, several vaccine prototypes based in this species are under investigation.

ELELYSO® (taliglucerase alfa): recombinant glucocerebrosidase produced in carrot and indicated for long-term enzyme replacement therapy in patients with type 1 Gaucher disease. The first plant-based biopharmaceutical approved for human use.

Food-grade vaccines (FGVs): recombinant subunit vaccines produced in safe organisms that may also serve as the delivery vehicle.

Lactic acid bacteria (LAB): a group of Gram-positive bacterial species characterized by the production of lactic acid by glucose fermentation.

Plant-based vaccine: vaccine formulation produced in plants as a convenient source of recombinant antigens.

***Saccharomyces cerevisiae*:** a GRAS yeast species that, in addition to serving for injectable vaccine production, is being used for the production of FGVs based on whole cells expressing a variety of target antigens.