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Edible Vaccines and Oral Immunization against Viruses: Prospects, Promises and Pitfalls

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Abstract

The lethality of infectious diseases can be largely minimized *via* implementation of crucial sanitary procedures such as vaccination. Vaccine is a biological preparation used to establish or improve immunity to a particular disease. Vaccines, made of killed or attenuated forms of microbes, are the main modes of defense and protection against several bacterial, viral and parasitic diseases. However, the process of production and purification makes them expensive and unaffordable to many developing nations. The usual traditional vaccines are generally microsomal preparations which can be sometimes harmful and not tolerated easily by newborn babies. Another major hurdle to oral immunization is the digestion of macromolecular antigenic protein within the stomach due to extremely acidic pH. Dr. Charles Arntzen of Arizona State University first put forward the idea of edible vaccines which pose an interesting alternative in overcoming some of the constraints of traditional vaccines. They offer cost-effective, easy to administer, storable, widely-acceptable and bio-friendly vaccine delivery system, particularly in developing countries. Edible vaccines are obtained by incorporating a particular gene of interest into the edible parts of plant, which produces the desirable encoded protein. They are specific to provide mucosal activity along with systemic immunity against various viral diseases like measles, hepatitis B, hepatitis C, anthrax, Human Immunodeficiency Virus (HIV), etc, as well bacterial diseases like cholera and diarrhoea. Various foods that are genetically engineered to serve as alternatives to injectable vaccines include cereals (wheat, rice, corn), fruits (banana) and vegetables (lettuce, potato, tomato). Plant-based oral vaccines can be grown locally, reducing the cost and complications of transportation, while the stability of proteins in intact plants removes the need for refrigeration. The edible nature of vaccines eliminates the need for syringe-based delivery, saving money and reducing the risk of infection. However, various technical obstacles, regulatory and non-scientific challenges, including the control of antigen dosage seem surmountable and need to be addressed or overcome. The aim of this review is to present and critically examine the potentiality of edible vaccines as an option for global immunization against pathogenic diseases with special emphasis on viral infections.

Keywords: Edible vaccines; Virus; Antigen; Infectious disease; Immunization

Introduction

Application of vaccines stands out as an effective measure to prevent infectious diseases, accounting for more than 54% of total mortality in developing countries. More than one million people die annually from infectious diseases, with 50% of these diseases caused by pathogens infecting the mucosal membrane of the mammalian host. Outbreaks of pathogens in different parts of the globe are chiefly connected with increase in population density and overcrowding, and relaxation to follow proper hygiene controls, leading to perpetuation of several infectious diseases, increasing morbidity and jeopardizing the global health systems [1]. This can be very well felt in the present scenario when the entire world is essentially crippled due to life-threatening pandemic infection caused by Severe Acute Respiratory Syndrome-Coronavirus 2 (SARS-CoV2). The incidence of this pandemic has very well pointed out that infectious diseases can cause devastations in not only countries lacking health infrastructural facilities, but also in those with strengthened and consolidated health systems, demonstrating the need of sound protective measures to retard the global emergence of newer pandemics, without forgetting the fact that till date, only smallpox virus has been managed to be completely eradicated. Incidentally, the concept of vaccination was first put forth by Edward Jenner in 1796 for small pox. Vaccination technique was introduced almost 200 years back with the purpose of delivering a minimum dose of a disease-causing pathogen to induce a humoral or cellular immune reaction without actually subjecting the individual to the

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risk of true infection. Vaccination is the process by which the body is made ready to face and fight off new infections by improving immunity for a long duration through antibody production, so that future infections can be warded off [2]. The immune system gets ready to combat the invading pathogens very quickly, before they get the chance to spread throughout the body while causing discomfort to the host. Several diseases like tuberculosis, cholera, typhoid and especially poliomyelitis have been successfully regulated through massive vaccination campaigns. Vaccine development involves an extremely dynamic phase, and routine immunization programs through vaccination now go beyond the traditional six childhood vaccines against diphtheria, tetanus, whooping cough, measles, polio and tuberculosis, and include additional vaccines to prevent hepatitis B, rubella, pneumococcal disease and rotavirus [3]. With regard to the present situations, it appears that almost every country all over the globe urgently feels the need of designing a new vaccine against SARS-CoV2 as fast as possible to save human life. The challenge in the present world is therefore to find unique and innovative vaccines that can target various newly-evolving pathogens and infections at various stages.

The evolution of vaccines has led to the discovery of new forms of vaccination that are effective and cover a wider array of diseases. Live-attenuated vaccines are considered as the original vaccines, where a living infectious pathogen is directly used as vaccine. In case of inactivated vaccines, the debris of the dead pathogen is used as vaccine. Toxoid vaccines involve utilization of the toxin produced by the organism as vaccines. Biosynthetic vaccines are synthetically designed having similar shape and properties to the infectious organism. The process of using DNA vaccines to prevent or slow down the spread of disease is also known as polynucleotide immunization. DNA that is injected into the subject undergoes transcription and translation which yield protein, making specific T and B cells to differentiate and proliferate. Recombinant vaccines involve cloning the gene encoding the antigen in a recombinant plasmid, expressing it in bacteria and purifying the protein to be used as vaccine [4].

Challenges Imposed by Conventional Vaccines

Despite worldwide immunization of children against the six devastating diseases, 20% of infants are still left un-immunized; responsible for approximately two million unnecessary deaths every year, especially in the remote and impoverished parts of the globe. Despite the advantages of vaccination, there are certain flip-sides and limitations of traditional vaccines that restrict their use [5]. The main problem to be solved is the boundaries on the production of vaccine, supply and distribution. One hundred percent coverage is desirable, because un-immunized populations in remote areas can spread infections and epidemics in the immunized safe areas, which have comparatively low herd immunity. For some infectious diseases, immunizations either do not exist or they are unreliable or very expensive [6]. The standard oral vaccines should fulfill some criteria like (i) desired antigens should be present in sufficient quantities; (ii) expressed antigens should be stable at room temperature for a long time; (iii) protective immunity must be induced by the vaccine; (iv) vaccines should withstand enzymatic degradation in the stomach [7]. Industrial production of vaccines makes them expensive commodities and inaccessible in developing countries. All these processes involve the use of sophisticated and expensive sterile fermentation technology followed by purification processes. Immunization *via* DNA vaccines is quite expensive with some undesirable immune responses. All

pathogenic agents cannot be cultivated in an exogenous medium. Conventional vaccines produced from attenuated pathogens involve the synthesis of antigenic proteins via mammalian cell culture which is easily prone to contamination with harmful pathogens. Vaccine production through microbial system invites possibility of endotoxin contamination. When cell culture and transgenic animals are used to make vaccines, the contamination possibly arises with viruses, prions and oncogenic DNA. Hence, biosecurity and biosafety issues need to be addressed to cultivate these pathogenic agents [8]. Although bacterial or viral attenuation for vaccine production is a controlled process, reversion of these attenuated micro-organisms to their virulence or pathogenic forms also restricts their use. Vaccine construction in several cases has been hampered because of varying or mutating strains of the pathogen, antigen drift, antigenic shift and other unrevealed mechanisms. Such regulations make it difficult to select the actual peptide sequence to prime the immune system, since the peptide sequences of the individual strains will all be different. Even if successfully produced, these commercial vaccines have specific expiry period and need refrigeration facilities, thereby enhancing control, storage, transportation and distribution costs. Vaccine degradation after acid digestion in the stomach is another concern [9,10]. In the present-day world, due to the existing and continuously emerging diseases, together with the safety and efficacy problems, associated with the available vaccines, researchers have tried to find an alternative approach for production of widely affordable, economic and reliable vaccines that are safe, acceptable and can be easily administered by simple feeding without the requirement of any trained personnel. Moreover, vaccines should be such that there are no refrigeration requirements, are easily transportable and could be smoothly delivered in the developing countries. Most of the currently available vaccines are associated with several problems including safety due to vaccine-associated side effects in human subjects. Edible vaccines are hence considered as ideal replacements for conventional vaccines [10].

Edible Vaccines and Their Advantages

The term 'edible vaccine' was coined in 1992 by Charles Arntzen and co-workers by introducing the concept of transgenic plants as a production and delivery system for subunit vaccines. However, the development of edible vaccines still remains in its infancy due to the emergence of various medical, legal, ethical and environmental uncertainties [11]. The World Health Organization in 1990's presented a task of finding cheap methods of oral vaccine production that do not need refrigeration requirement. Edible vaccines are generally antigen-expressing plants, where the edible part of a plant is genetically modified to express the antigens, thereby eliciting an immune response upon consumption, and serving as a factory for vaccine manufacturing [12]. The process of purification and downstream processing, which make conventional vaccines costly, are eliminated in case of edible vaccines, so that they are inexpensive and more affordable and accessible. Plant-derived vaccines can be designed to contain numerous antigens for various diseases. These multi-component vaccines are called 'second generation vaccines' that provide immunization against many diseases in a single dose. Plant-based systems only need greenhouses and not fermentation tanks for cell culture. Besides, purification from plant extract is simpler because plants are not carriers of viruses that could possibly be detrimental to humans [13]. The advantages of edible vaccines include lack of contamination occurring during traditional mammalian culture cells, no involvement of syringes or needles and easy to deliver to

the body. The specific antigen is protected by the cell wall of plant cells, cannot be damaged by gastric enzymes and can easily reach the blood stream where it activates the mucosal and systemic immunity. Edible vaccines reduce the need for skilled personnel to administer injections and negate the concerns regarding the reuse of syringes or needles. Any recipient of plant-derived vaccine is only exposed to non-infectious and non-toxic bit of protein. Vaccines generated in plants have an element of plant sugars that markedly elicits greater immune response. Though sugars also get attached to antibodies in case of animal-sourced vaccines, that is not reckoned to be beneficial [14]. Heat stability eliminates the need for refrigeration. Plant cells are capable to correctly fold and assemble not only antibody fragments and single chain peptides, but also full-length multimeric proteins. An edible vaccine provides higher safety of individual as compared to traditional vaccine since they are subunit preparation and do not involve attenuated pathogens which sometimes causes disease when administered as vaccine. They can be ingested by eating the plant/part of the plant. So, the need to process and purify does not arise. Expression of antigen in plant seeds provides a convenient system of vaccine storage for long duration of time, thus reducing storage and shipping costs under ambient conditions. If the local/native crop of a particular area is engineered to produce the vaccine, then the need for transportation and distribution can be eliminated [15]. Therefore, plants are considered for efficient production systems for vaccines as an alternative to circumvent the problems of traditional vaccines. Transgenic plants are also convenient for producing antigens in large amounts (kg) for using in parenteral and oral applications. Immunization by directly eating the transgenic plant parts (fruit, tuber, seed) which produce vaccine antigens are thought to reduce the high production costs like purification, storing and transportation costs. As plants are a safe biological system and harbor no pathogen of human infections, there are no ethical problems associated with plant edible vaccines, unlike the vaccines produced from animal cell cultures and transgenic animals.

Mechanism of action

The antigens in transgenic plants are delivered through bioencapsulation, i.e., tough outer plant cell wall that ensure protection against gastric secretions. Administration of edible vaccines lead to the degradation of majority of the plant cells in the intestine by the action of digestive and bacterial enzymes, resulting in the release of antigens present in the plant product. Edible vaccines are mucosal-targeted vaccines that stimulate both the systematic and mucosal immune network, activating the first line of defense of human body through mucosa. The mucosal surfaces are found lining the digestive tract, respiratory tract and urinoreproductive tract. Mucosal immune system is induced after recognition of an antigen by specialized cells called M-cells, which are localized in the mucosal membranes of lymphoid tissues such as Peyer's patches within the small intestines. Upon oral administration, the released antigens are taken up by the intestinal M cells, passed on to macrophages, other antigen-presenting cells and local lymphocyte population which generate antiserum responses. The M cells especially present the antigen to the Antigen Presenting Cell (APC) surface and activate B cells with the co-operation of helper T cells to secrete immunoglobulin A (IgA) [16]. On passing through the mucosal epithelial layer towards the lumen, the IgA molecules form complex with membrane-bound secretory components to form secretory IgA (sIgA), which in turn interacts with specific antigenic epitopes and neutralize the invading pathogen. The common problem of most oral vaccines/ therapeutics

is the tolerance towards the vaccine in the gut. This problem can be overcome by immune suppression using triamcinolone in small amounts, increasing the dosage of the vaccine significantly and applying multiple doses over a specific period of time.

Production of edible vaccines

There are a number of factors that make plants an ideal candidate for edible vaccine production. They should have a long shelf life so that they can be stored for a sufficient period without degradation (e.g., rice, wheat), should have fast luxuriant growth (e.g., tomato, tobacco) and should be easily transformable. Edible vaccines can be produced by incorporation of transgene in the edible parts of the selected plant species, where the antigen producing gene(s) are introduced in plant genome through different transformation techniques like direct gene delivery method (biolistic or gene gun), cloning in a vector by indirect gene delivery method (*Agrobacterium tumefaciens*-mediated transformation) or utilizing chimeric plant viral expression systems [17]. New or multiple transgenes can be introduced by sexual crossing of plants, thus creating novel vaccines against multiple diseases. Moreover, plant cells are able to perform complex posttranslational modification of recombinant proteins, such as glycosylation and disulfide bridging that are often essential for biological activity of many mammalian proteins, allowing for the retention of native biological activity. Potato tubers are widely consumed all around the world and quite affordable. A large amount of data on genetic manipulation is available in potato, thus making optimized protocols available. However, one major disadvantage of using potato is that it requires cooking before consumption which can denature the antigen [1]. The main reason why rice and maize are attractive as candidate edible vaccines is because they can be stored without refrigeration for a very long period of time. However, the problem with the cereals is that they take relatively long periods of time and require perfect conditions to grow. The major disadvantage with tomato is that it undergoes spoilage rapidly after ripening.

Edible Vaccines Against Viruses

Edible vaccines targeted against viruses have been a large and exciting field of research almost from its inception, for viral pathogens ranging from hepatitis B or C to foot and mouth disease viruses, and human papillomavirus, to mention just a few. Successful expression of antigens in plants was achieved for rabies virus G-protein in tomato, Norwalk virus capsid protein in tobacco and potato, hepatitis B virus surface antigen in tobacco and potato, etc [8, 18,19]. Table 1 summarizes the edible vaccines generated against different human viruses.

Can Edible Vaccine Approach be Targeted Against SARS-CoV2?

The 2019-2020 pandemic caused by SARS-CoV2 all over the globe has raised severe concerns and is evidencing the lack of substantial clinical treatments till date. Thus, there is an urgent need for having scalable production platforms to generate SARS-CoV2 vaccines. Plant-made vaccines offer the possibility of performing the oral delivery of formulations made with freeze-dried plant biomass, which avoids the costs derived from purification and parenteral delivery. As an initial step toward provision of an oral vaccine against SARS-CoV, which resulted in a worldwide outbreak in 2003 spanning almost 20 countries, Li et al. (2006) [20] expressed a partial spike (S) protein in the cytosol of nuclear-transformed and in the chloroplast of plastid-transformed tobacco and lettuce plants. The S protein is

Table 1: Edible vaccines generated against different human viruses.

Virus	Antigen targeted	Transgenic plant	Effect
Hepatitis B	S form of HbsAg	Potato, Tomato	HEV-E2 gene correctly expressed and antigen showed normal immune-reactivity in mice
Hepatitis C	Synthetic hypervariable region 1 (HVR1)-derived peptide called R9, a potential neutralizing epitope of HCV derived from the envelope protein E2	<i>Nicotiana benthamiana</i>	Intranasal immunization of mice elicited anti-HVR1 serum antibody
Influenza	Haemagglutinin (HA) protein	<i>N. benthamiana</i>	Grams of cGMP-grade plant-made H7N9 vaccine were produced in the form of HA-only VLPs, in response to the viral outbreak in humans
Cervical Human Papilloma Virus like particles (HPV)	HPV type 11 L1 major capsid protein	Potato	Oral immunization induced anti-VLP immune response in mice
Human rotavirus	HRV-VP7	Potato	Elicited serum IgG and mucosal IgA by oral delivery to mice
Measles	Loop forming B cell epitope (H386-400)	Carrot	Intraperitoneal immunization of mice with carrot extracts induced high titers of antibodies
Respiratory Syncytial Virus (RSV)	RSV-F protein	Tomato	Oral immunization of mice induced both serum and mucosal RSV-F specific antibodies
Gastroenteritis	Glycoprotein S (N-gS) from transmissible gastroenteritis coronavirus (TGEV)	Potato	Intraperitoneal immunization in mice resulted in serum IgG specific for TGEV, oral immunization through direct feeding developed serum antibodies specific for gS protein
Foot and mouth disease virus	VP1 capsid protein	<i>Arabidopsis thaliana</i>	Mice injected intraperitoneally with the partially purified VP1 protein were totally resistant to a challenge with a virulent strain of the virus
HIV-induced Acquired Immunodeficiency Syndrome (AIDS)	Stable chimeric CPMV particles that express epitopes derived from human rhinovirus 14 and HIV-1	Tomato	Expressed protein was detected in different parts of the plant, including the ripe fruit
Norwalk virus	Capsid protein (NVCP)	Tobacco and potato	Stimulated serum IgG and gut IgA specific for NVCP when fed to mice
Rabies	Rabies Virus (RV) glycoprotein	Tomato	Induction of neutralizing antibodies in mice that were parenterally vaccinated with a peptide of RV glycoprotein fused to a plant virus coat protein; antigen-capsid fusion.

a major virion structural protein. S (471-503), a peptide located at the Receptor Binding Domain (RBD) of SARS-CoV S1 subunit, could specifically block the binding between the RBD and angiotensin-converting enzyme 2, resulting in the inhibition of SARS-CoV entrance into host cells *in vitro*. This study provided the possibility of establishing a safe and inexpensive vaccination strategy against SARS-CoV. In another study, the N-terminal fragment of SARS-CoV S protein (S1) was expressed in tomato and low-nicotine tobacco plants and systemic and mucosal immune response was evaluated in mice after oral ingestion of tomato fruits. Sera of mice parenterally primed with tobacco-derived S1 protein revealed the presence of SARS-CoV-specific IgG as detected by Western blot and ELISA analysis [21]. The 2014-2015 Ebola virus outbreak in West Africa (Liberia, Sierra Leone, Mali, Nigeria and Senegal) created an urge among scientists to find out a novel approved vaccine against this virus. Monreal-Escalante et al. [22] successfully overexpressed the VP40 antigen from the Ebola virus in tobacco plants, reaching accumulation levels up to $2.6 \mu\text{g g}^{-1}$ fresh weight of leaf tissues. The antigenicity of the plant-made VP40 (viral protein) antigen was evidenced by Western blot and an initial immunogenicity assessment in test animals, that revealed the induction of immune responses in mice, following three weekly oral or subcutaneous immunizations at very low doses (125 and 25 ng respectively) without the inclusion of accessory adjuvants. Therefore, this plant-based vaccination prototype is proposed as an attractive platform for the production of vaccines in the fight against Ebola virus disease outbreaks. These success stories make us to believe that identification of antigen(s), through genomic, proteomic and bioinformatic tools, most likely to induce an immune response will enable us to overexpress the antigen in plants. Within the COVID-19 vaccine race, the strategy of molecular farming in generating edible vaccines has already been initiated by the Canadian Company, Medicago, which claims to produce 10 million doses per month if their production method and clinical trials obtain approval from US Food and Drug Administration (FDA). Another American company,

Kentucky Bioprocessing which has raised transgenic tobacco and is conducting pre-clinical tests has claimed to manufacture up to three million doses per week. The University of California, San Diego, is working on an innovative collaborative project to develop a microneedle patch-vaccine that uses proteins grown in genetically-modified plants. The Center for Research in Agricultural Genomics (CRAG) of Spain, will also develop antigens for SARS-CoV2 in genetically modified lettuce and tobacco, and the international project NEWCOTIANA, which works on the development of medicines and vaccines in plants with funding from the European Union, has released the complete genetic sequence of *Nicotiana benthamiana* in order to accelerate the development of a plant-based vaccine.

Pitfalls of Edible Vaccines

The three main challenges of edible vaccine production are: (i) the selection of antigen and plant expression host, which will ensure the safeness of the vaccine produced and its thermostability, (ii) consistency of dosage, and (iii) manufacturing of vaccines according to Good Manufacturing Practice (GMP) procedures [23]. Therefore, the challenges facing plant-based-vaccine development include technical, regulatory and economic aspects, and public perception. Among technical challenges to be considered, the crop should provide ample biomass for accumulation of a sufficient quantity of the antigenic protein. The expression of antigens in plants is a major regulatory concern. Targeting transgene expression via a tissue-specific promoter may reduce regulatory concerns. Not all vaccine candidate proteins are highly immunogenic in plant tissues and secondary metabolites found in plants may compromise the ability of the candidate vaccine protein to induce immunity [24]. Among regulatory challenges, issues relevant to any genetically modified crop that have to gain regulatory approval from the USDA, FDA and/or EPA apply equally to vaccines generated in edible plant parts. Allergic reactions to plant protein glycans and other plant antigens are a challenging issue. It has been suggested that plant-

derived recombinant proteins or antibodies may have increased immunogenicity or allergenicity, as compared to mammalian counterparts, along with few side effects such as toxicity on central nervous system, cytokine-induced sickness and autoimmune diseases. Another problem is that the glycosylation of trans-proteins in plants differs slightly from those produced in transgenic animals or animal cells *in vitro*. A significant difference with trans-protein production in plants is their inability to add sialic acid to glycoproteins [24]. There are chances of contamination of recombinant proteins by pesticides, herbicides, mycotoxins or potentially toxic factors due to the fact that some plant species contain numerous toxic alkaloids and other secondary metabolites. All these factors invite regulatory constraints and uncertainties for approval as human drug. The stability of vaccines in fruits is uncharacterized. Moreover, there is a possibility of weakening of the medicinal property and denaturation of vaccine proteins in case of cooked foods. Evaluation of dosage requirement and proper maintenance of dosage are difficult, since consistency of dosage may vary within the plants of the same species, from fruit to fruit and from generation to generation due to the size and ripeness of the fruits or plants [25]. Fruits like tomato and banana do not appear in market in fixed or standard sizes, so that it is quite difficult to optimize the dosage. It is also quite difficult to evaluate the required dosage for every patient. The levels of innate and adaptive immune responses generated in different individuals may vary, based on the types of antigens being exposed in the body. Between two patients with different body weight as well as their age, the dosage of plant-based vaccine required will be different. If this issue is not monitored carefully, an immunological tolerance will be induced when the patient is overdosed, while reduction in antibody production will occur when the patient is under dosed. Development of immune-tolerance to the vaccine protein or peptide is therefore a major concern [1]. Fruit vaccines should be easily identifiable to avoid the misadministration of the vaccine, which may lead to complications such as immune-tolerance. The oral vaccines are complicated by the need to protect the antigen from the effects of the acidic and proteolytic environment of the gut. Hence, to be effective, oral subunit vaccines generally require higher doses than oral replicating vaccines. Variable conditions for edible vaccine are also a major problem. Potato-containing vaccine to be stored at 4°C could be stored for longer time, while a tomato does not last long. Thus, these vaccines need to be properly stored to avoid infection through microbial spoilage [26,27]. Though the plant edible vaccines are a lucrative option in the field of vaccination, several potential issues need to be addressed via exhaustive research and development to use this area of health care for greater benefits.

Conclusion and Future Prospects

Vaccines are capable of reducing the use of antibiotic and can play a vital role in an era where antibiotic resistance is gradually becoming a major challenge. Edible vaccine discovery can be considered as one of the major breakthroughs in the area of biotechnology, which successfully embraced the obstacles encountered in rising vaccine technology. When compared to the traditional vaccines, edible vaccines do not require sophisticated equipments and machines for vaccine production in order to stimulate both systemic and mucosal responses, without any requirement for sterile injection conditions and adequate cold-storage facilities, and may be derived directly by simply consuming a fruit [28]. However, relevant technical, social, environmental, ethical and policy issues concerning plant-based edible vaccines need to be addressed before they become product-ready. Future researches should overcome several limitations like low

expression, immune-tolerance, glycosylation, immunogenicity and stability of the trans-proteins. The development and improvement of suitable gene delivery methods for efficient and optimum vaccine production should continue. More insights into the heterologous gene regulation and expression patterns in transgenic plants could answer some of the unknown mechanisms that make heterologous gene expression a tough task [24]. It is very difficult to establish a stable antigenic protein concentration in plant tissues, and there is no certainty that the expressed antigen will produce an immune response. Further studies are anticipated in overcoming the problem, related to dose variability in the transgenic plants. Some food-processing techniques (batch-processing and freeze drying) could maintain the normal conformation and native antigenicity in transgenic plants such as tomato, potato and Arabidopsis, thus standardizing the concentration of antigens in the plants. Plant-made oral vaccines might induce allergenicity during post-translational modifications and oral tolerance when co-administered with oral adjuvants. This may activate the mucosal immune system by provoking hypersensitive responses to other proteins contained in the daily food. In other words, recurrent delivery of plant-made edible oral vaccines can boost regulatory T-cell stimulation, contrary to vaccine antigen, causing hypersensitivity in case of pollen allergy or food allergy [17]. As a drug that is contained in a plant or its fruit, it should be stringently evaluated, authorized and supervised by Public Health Institutes or a similar Human Health Organization in each country. It is anticipated that regulatory approval will be granted ultimately to help in the wake of global disease control. One very important point is the proper coordination between academia and industry to help these vaccines ultimately reach people. It will also be a challenge to create a positive public perception regarding safety and efficacy of these vaccines, in the midst of enormous fuss created over the safety issues of transgenic crops. The societal acceptance will largely depend on awareness campaigns on the use and benefits of edible vaccines [29,30]. In the face of global SARS-CoV2 pandemic ravaging the entire world at present, edible vaccines represent a ray of hope and valuable bio-friendly alternative, having undeniable potential to mitigate infection and supporting public health programs to reduce the COVID-19 outbreak.

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