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Potential of plant-derived vaccines for treatment of human infections in view of COVID-19 pandemic

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Abstract

Vaccines have always been helpful to mankind to battle against various diseases. Plants provide good amount of therapeutic proteins and that too cost effective, safe and more efficient way to combat infections. Plants are not only used as vectors but they also give humoral immunity. Genetic transformation through *Agrobacterium* and genetically altered plant virus are used to obtain the desired antigens. New advancements in science and technology have allowed us to use various other methods like biolistics, polyethylene glycol treatments and ultrasonication. Plant vaccines are more advantageous than earlier methods of forming vaccines, it's production utilizes transgenic, transplastomic and transient expression approaches for obtaining the desired proteins. The present paper portrays the principle and working mechanism of plant-derived vaccines. It presents a general idea of the key plants used in the plant vaccine technology especially for treatment of human infections. This study is relevant and much needed in the current Covid-19 pandemic time. It aims to present the ability of vaccines sourced from plants that can be explored further to direct against corona disease. The paper also highlights the present-day challenges faced by this technology. It is recommended that more targeted and result-oriented studies pertaining to this proficient and harmless technology must be carried out. Further, investigation and advancement is required to explore potential of plant-derived vaccines in treatment of Corona disease is emphasized.

Key Words: Biopharming, Corona, Plant vaccines, Transient expression system

Introduction:

The term vaccine was first developed by Edward Jenner against the disease smallpox. He isolated poxvirus from cows which resulted in protection against the virus (Jenner 1798, 1801). After almost 100 years, Louis Pasteur developed a vaccine against rabies which was live but attenuated and introduced some basic steps for vaccine advancement: isolation, inactivation, and injection of the causative organism. These were directed to provide defense in opposition to many lethal infectious diseases (Fraser and Rappuoli 2005). Inactivation and attenuation of the causative organism were the only prevalent processes for the creation of vaccines for a very long time. Those were effective but sometimes led to unwanted consequences. Attenuation developed non desirable immune reactions and even diseases. It necessitated to think about alternative methods for vaccine development.

Types of vaccines

Vaccines which are available till today are of four types; (1) whole-celled killed vaccines, where causative organism is inactivated by chemical/physical methods; (2) whole-celled live weakened vaccines,where causative agent is alive but is not able to cause the disease; (3) subunit vaccines, which contain extremely defined ingredients made from purified antigen(s) obtained from the causal organism; and (4) conjugate vaccines, which contain poorly immunogenic polysaccharide component of the causative organism which is chemically attached to a protein.

Subunit vaccines are regarded to be more safer as no pathogen is involved and are easily available since a large scale production can be attained. Several expression systems like *E.coli*, yeast, mammalian cells and insect cells have been obtained through recombinant DNA technology. This is called as biopharming which means producing biological drugs using living host. The first form of biopharming was the production of insulin using *E. coli* (bacterial host) in 1978 by Genentech. This led to production of insulin from natural biological sources such as dog and calf pancreases.

Development of plant-derived vaccines

Cholera toxin subunit B (CT-B) is a frequently used immunogen used for the liberation of plant-sourced vaccines. To achieve a stronger immune response, proteins with weak immunogenic properties can be coupled to CT-B. Roy Curtiss, III and Guy A. Cardineau have successfully developed genetically modified tobacco plants which were able to express antigen of *Streptococcus* mutants. Their historical work initiated the idea of plant-based vaccines. Their patent application filed in 1990 became the first one of its kind which was filed for plant-based vaccine technology (Curtiss and Cardineau 1997).

The biggest advantage of using plants as vaccines is that they act both as an antigen and a vector to the mucosal immune organization simultaneously preventing the antigen from degradation during its passage through the abdominal tract. For expressing vaccine epitopes and full therapeutic proteins in plant tissue, several transgenic plants, plant virus expression vectors and transplastomic plants have been obtained. Mason and Arntzen in 1995 explained in depth method for producing plant vaccines (Figure 1).

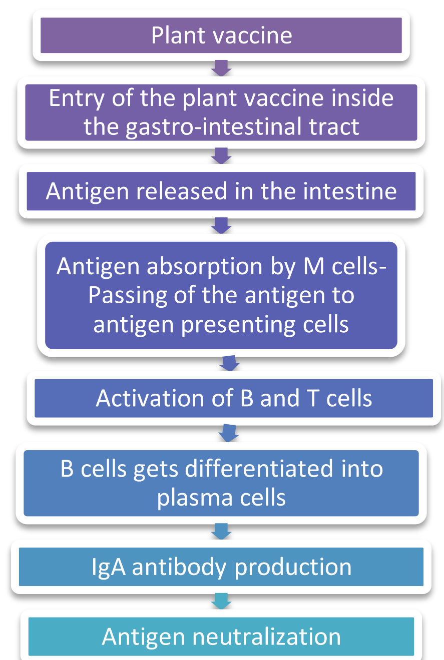

Figure1: Mode of action of plant-based vaccines (Modified from Mishra et al. 2008)

A vaccine obtained from plants is a great opportunity to treat the 'orphan' diseases like dengue, rabies and hookworm treatments in the west. The expansion of plant-derived immunogens has provided relief to the society especially to the people of Third World countries. Furthermore, purification of vaccine proteins from plants requires only few steps and is cost-effective too as compared to the ones obtained from mammalian structures. It may even need only partial decontamination based upon specific purpose and procedure. Besides providing vaccines and therapeutic proteins to the people of developing nations, plant production platforms present other opportunities as well. There is always a challenge regarding the expense of vaccine production globally in spite of having excellent vaccine production technologies and methods.

Plant viruses have been utilized to find vaccines for personalized medical treatment and for some specific ailments. These have the ability to be used against possible biological warfare agents and causes of global pandemics like influenza virus, swine flu or corona.

How plant-derived vaccines work?

A recombinant antigen developed in a low-cost host that can serve as agent of defensive immunity is the basic requisite for developing plant based vaccine. Their biomass or purified fractions assist in providing protective immunity which is administered through various ways (Salyaev et al. 2010). This directed the advancement of oral vaccines or mucosal-derived vaccines as they need less processing of plant raw biomass. Mucosa remains the first major entry point for pathogens through various body surfaces leading to secretion of immunoglobins. The faces of the membrane are linked with set of ordered lymphoid tissue arrangements called as mucosa-associated lymphoid tissue (MALT) segmented anatomically into gut-associated lymphoid tissue (GALT) containing Peyer's patches, the nasopharynx-associated lymphoid tissue (NALT), and the bronchi-associated lymphoid tissue (BALT). Follicle-associated epithelium (FAE) which covers PPs contains the microfold (M) cells (Staats et al. 1994). To achieve immunoprotection, these vaccines stimulate systemic as well as mucosal immune responses. Mishra and co-workers described the working mechanism (Figure 2). This is a cost effective method for global vaccination programmes as it doesn't require any syringes and needles though it suffers from some drawbacks. In relation to dosage and stability, it is important to deal with plant biomass to achieve proper measure in the freeze dried powder, stored properly for long duration (Alvarez et al. 2006). The most desirable method of their introduction into markets is to process the plant material into tablets or fill into capsules.

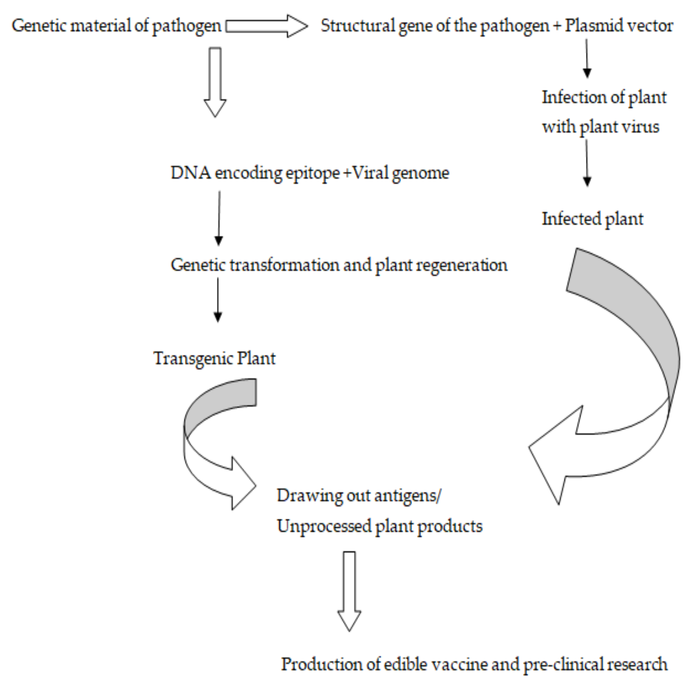

Figure2: Production of plant-based vaccines (Modified from Mason and Arntzen 1995)

Methods of production of plant-derived vaccines

Classical methods: This process involves incorporation of the target sequence of the designated antigen into the vector ahead of its transfer into the expression assembly. Transient transformation system is used to express transgene in plants achieved through nuclear or plastid integration. Direct and indirect gene delivery methods are employed to form plant-based vaccines. Using *Agrobacterium tumefaciens* chromosomal integration or by microprojectile bombardment techniques, the vector is introduced. The microbial DNA encoding for the epitope is inserted in a plant pathogenic virus resulting in the formation of a genetically engineered virus which infects the plant cells and starts replication. The viral genes along with the microbial genes get expressed and produce proteins that get accumulated inside these cells. Chemical stimulation and sonication are the methods employed to increase the efficiency of the transgene delivery.

5.2 Modern methods: Polyethylene glycol is utilized as a chemical stimulant. It enhances DNA uptake by protoplasts through the process of endocytosis and allows the transitory expression of desired genes. This method is cost-effective and is complemented with other gene delivery systems for increased efficacy. But simultaneously, it suffers from certain limitations. The method requires expertise to carry out the procedures (Craig et al. 2005). Due to its toxicity on protoplasts, it results in low survival rate and ceased cell division (Hassanein et al. 2009). Therefore, it is not widely used in plant-derived vaccine production.

Sonication develops microwounds by using soundwaves on plant cells for augmenting the liberation of unsheathed DNA inside the plant protoplast in plant-transformation systems (Santarem et al. 1998). This has an adverse impact on plant tissues and consequently low down transformation competence (Santarem et al. 1998, Liu et al. 2005). In a number of experiments, sonication assisted *Agrobacterium*-mediated transformation (SAAT) is used to cause mechanical interference and microwound formation on plant cells (Rybicki et al. 2014). *Agrobacterium* will penetrate deep into the tissues through wounded cells, thus, accelerating chances of infection (Liu et al. 2005).

Another method called combination of sonication plus vacuum infiltration assisted *Agrobacterium*-mediated transformation magnificently transmuted the seedlings of *Fraxinus pennsylvanica*. This contains binary vector pq35GR having neomycin phosphate transferase (*nptII*) and β- glucuronidase (GUS) fusion gene and an enriched green fluorescent protein gene (Du et al. 2009) that improves the efficacy of *Agrobacterium*-mediated transfer of gene (Liu et al. 2005).

Potential of plant-derived vaccines against COVID-19

The global pandemic caused by SARS-CoV-2 has caused more than 38.2 lakh deaths worldwide till today (at the time of writing this paper). In the light of the information discussed above in this article, we suggest that plant vaccines can be employed as an important manoeuvre to fight and provide a promising therapy against the corona disease. Plants used in treating various infectious diseases are enumerated in Table 1. Phytochemicals like polyphenols, gallates, flavanoids have already been indicated to be putative agents for the therapy of this deadly disease caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) (Chojnacka et al. 2020). *Artemisia kermanensis*, *Satureja hortensis*, *Eucalyptus*

Table 1: Major plants used as vaccine models used against specific ailments			
S.No.	**Model Plants**	**Used against**	**References**
1.	Tobacco	Gastroenteritis, Chicken infectious anaemia, Hepatitis B, Coccidiosis	Yang et al.(2007); Gómez et al.(2009); Pérez Filgueira et al. (2002)
2.	Alfalfa	Hog pest virus,*Echinococcus ganulosus*	Pérez Filgueira et al. (2002)
3.	Carrot	HIV, *E.coli,Helicobacter pylori*	Shin et al.(2015);Yan-Ju et al.(2010); Zhang et al. (2010)
4.	Lettuce	*E. coli*, Classical swine fear hog pest virus , Hepatitis B virus	Srinivas et al.(2008); Kim et al. (2007)
5.	Tomato	SARS , Norwalk virus, Alzheimer's disease, Pneumonia, Septicaemia, and Bubonic plague	Chanand et al.(2015);Estes et al.(2006);Lou et al. (2007)
6.	Rice	*E. coli*	Mason et al.(1996);Oszvald et al. (2007)
7.	Potato	Tetanus, Diphtheria, Hepatitis B and Norwalk virus	Concha et al.(2017); Mason et al. (1996)

caesia, *Rosmarinus officinalis*, *Mentha* spp., *Thymus* spp. and *Zataria multiflora* are some of the plants which are found to be rich in phenolic compounds (Tohidi et al. 2017; Ma and Yao 2020).

According to Chen et al. (2008), SARS-CoV can be restrained by using tender leaves of *Toona sinensis*. The best two herbal remedies used by Chinese management for combating the transmission of COVID-19 are *Radix astragali* (Huangqi) and *Glycyrrhizae radix* (Gancao) (Luo et al. 2020). Sugar moieties present in glycyrrhizic acid, a triterpenoid obtained from *Glycyrrhiza glabra,* have shown anti-SARS activity (Bailly et al. 2020). One of the traditional Chinese medication called Lianhuaqingwen (LH) made from a mixture of 13 herbs including *Forsythia suspensa*, *Ephedra sinica* and *Lonicera japonica* has been demonstrated to curb SARS-CoV-2 replication, lower cytokine production prior to inflammation, and significantly alter the nature of SARS-CoV-2 (Panyod et al. 2020; Runfeng et al. 2020).

Medicago has commenced Virus-like particles (VLP) centered plant vaccines for SARS-CoV-2 (Phillip Morris International, 2020). An American company, iBio (Bryan, TX, USA) is also developing COVID-19 vaccine based on VLP by using tobacco plants (iBio, 2020). *Nicotiana benthamiana* genome is being sequenced at the Queensland University of Technology for developing the COVID-19 vaccine (ISAAA, 2020).

Current challenges

Plant-derived vaccines have been formed in various transgenic plants. There is no human consumed vaccine which is derived from plant till today. The current status of these plant-sourced vaccines is that many of them are in their Phase-I trials and some have completed or are in their Phase-II and Phase-III trials, so that means that there exists definite challenges in the construction of plant-derived vaccines till today, though its production started almost 20 years ago.

According to Good Manufacturing Practice (GMP) course of action, plant-based vaccines confronts three major challenges:

1. Selection of antigen and host – In real scenario, all antigens are not well matched with the particular hosts, therefore selecting antigen

with their compatible plant host is the very first upstream objection. Though, various genomic approaches and proteomic methods can be applied to characterise such compatibility.

2. Dosage consistency – Due to difference in dosage make up in different plants and plant parts belonging to same plant, uniformity of the dosage pose another challenge for its production. Non-specific integration of transgene and difficulty in generalising dosage requirement specific to each patient leads to this challenge. Allergic responses and small reactions like effect on cerebrospinal nervous system, cytokine stimulated ailments, and auto-immune disorders (Sharma et al. 2007) may be induced by consuming plant-based vaccines due to gene silencing.

3. Vaccine built up – Safe, trustworthy and low-cost should be the priority in using plant based vaccines. It is mandatory to follow Good Agricultural Practices (GAP) and Good Manufacturing Practices (GMP) for the downstream manufacturing of plant-derived vaccines. It is challenging to maintain the GMP guidelines throughout in their production. Availability and recruitement of skilled staff, specialized infrastructure for separate seed storage are a few of the pre-requisites. Other mandates such as correct labelling, lot dispensing, development operations, accurate safeguarding of allotment files, guaranteed assurance of the quality and constant monitoring of superiority should be followed.

4. Regulatory issues – United States Department of Agriculture (USDA), Food and Drug Administration (FDA) and/or Environmental Protection Agency (EPA) approval is also applied to plant based vaccines which may create regulatory issues. For rating effectiveness, security and trustworthiness, followed by FDA approvals, any plant-based vaccine need to go through Phase I-1V trials. Unfortunately, they are not considered for proposals instead of their high potency to fulfil the demand of vaccine.

Conclusion

There is immense potential of plant-derived vaccines waiting to be discovered and exploited. This technology can offer an assuring treatment to combat various human infections. There is an urgent need to do further research and development to explore prospective plant-derived vaccines for the management of corona disease that has crippled the world today.

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References

1.Bailly, C., & Vergoten, G.(2020). Glycyrrhizin: An alternative drug for the treatment of COVID-19 infection and the associated respiratory syndrome? *Pharmacol Ther,* 214.

2. Chen, C.J., Michaelis, M., Hsu., H.K, Tsai, C.C, Yang, K.D., Wu, Y.C., Cinatl, J.Jr., & Doerr, H.W. (2008). *Toona sinensis* Roem tender leaf extract inhibits SARS coronavirus replication.*J Ethnopharmacol,* 120(1), 108–111.

3.Chojnacka, K., Witek-Krowiak, A., Skrzypczak, D., Mikula, K., & Młynarz, P. (2020). Phytochemicals containing biologically active polyphenols as an effective agent against Covid-19-inducing coronavirus. *Funct Foods*, 73.

4.Craig, W., Gargano, D., & Scotti, N. (2005). Direct gene transfer in potato: a comparison of particle bombardment of leaf explants and peg-mediated transformation of protoplasts. *Plant Cell Reports*, 24(10), 603–611.

5.Curtiss, R. III, & Cardineau, G.A. (1997). Oral immunization by transgenic plants. *United*

States Patent, 5,654,184.

6.Dania, O., Govea-Alonso., Cardineau, G. A., & Rosales-Mendoza, S. (2014). Principles of Plant-Based Vaccines, In S. Rosales-Mendoza (Ed.), *Genetically Engineered Plants as a Source of Vaccines Against Wide Spread Diseases* (pp. 1-5). Springer.

7.Du, N., & Piju, P.M. (2009). *Agrobacterium*-mediated transformation of *Fraxinus pnnsylvanica* hypocotyls and plant regeneration. *Plant Cell Reports*, 28(6), 915–923.

8.Laere, E., Ling, A.P.K. & Ying Poei, W. (2016). Plant-Based Vaccines: Production and Challenges, *Journal of Botany*, 1–11.

9.Estes, M.K., Zhang, X., Buehner, N.A., Hutson, A.M., Estes, M.K., & Mason, H. S. (2006). Tomato is a highly effective vehicle for expression and oral immunization with Norwalk virus capsid protein. *Plant Biotechnology Journal*, 4(4), 419–432.

10.Fraser. C.M., & Rappuoli, R. (2005). Application of microbial genomic science to advanced therapeutics. *Annu Rev Med*, 56, 459–474.

11.Kumar, G., Loganathan, K., & Rao, K.V.B. (2018). Plant Vaccines: An Overview. In J. Singh et al. (Eds.), *Microbial Bioprospecting for Sustainable Development* (pp. 249-263).

12.Gómez, E., Zoth, S.C., Asurmendi, S., Rovere, C.V., & Berinstein, A. (2009). Expression of hemagglutinin neuraminidase glycoprotein of Newcastle disease virus in agro infiltrated *Nicotiana benthamiana* plants. *Journal of Biotechnology*, 144, 337–340.

13.Hassanein, A., Hamama, L., Loridon, K., & Dorion, N. (2009). Direct gene transfer study and transgenic plant regeneration after electroporation into mesophyll protoplasts of *Pelargonium × hortorum* Panache Sud. *Plant Cell Reports*, 28(10), 1521–1530.

14.Jenner, E. (1798). An enquiry into the causes and effects of the variolae vaccinae, a disease discovered in some of the western counties of England, particularly Gloucestershire, and known by the name of the cow pox. Sampson Low, London 1 Principles of Plant-Based Vaccines. 13.

15.Jenner E (1801). The origin of the vaccines inoculation. Shury, London.

16.Karasev, AV., Foulke, S., Wellens, C., Rich, A., Shon, K.J., Zwierzynski, I., Hone, D., Koprowski, H., & Reitz, M . (2005). Plant based HIV-1 vaccine candidate: Tat protein produced in spinach. *Vaccine*, 23(15), 1875–1880.

17.Kim , TG., Kim,M.Y., Kim, B.G., Kang, T.J., Kim, Y.S., Jang, Y.S., Arntzen,C.J., & Yang, M.S . (2007).Synthesis and assembly of *Escherichia coli* heat-labile enterotoxin B subunit in transgenic lettuce (*Lactuca sativa*). *Protein Expression and Purification,* 51(1), 22–27.

18.Kumar, GBS., Ganapathi, T.R., Revathi, C.J., Srinivas, L., & Bapat, V.A. (2005). Expression of hepatitis B surface antigen in transgenic banana plants. *Plant,* 222(3), 484–493.

19.Liu, Z., Park, B.J., Kanno, A., & Kameya,T. (2005). The novel use of a combination of sonication and vacuum infiltration in Agrobacterium-mediated transformation of kidney bean (Phaseolus vulgaris L.) with lea gene. *Molecular Breeding*, 16(3), 189–197.

20.Lou , XM., Yao, Q.H., Zhang, Z., Peng Z., Xiong, A.S., & Wang, H.K. (2007). Expression of the human hepatitis B virus large surface antigen gene in transgenic tomato plants. *Clinical and Vaccine Immunology*, 14(4), 464–469.

21.Luo, .H, Tang, QL., Shang, Y.X., Liang, S.B., Yang, M., Robinson ,N., & Liu ,JP.C. (2020). *Toona sinensis* Roem tender leaf extract inhibits SARS coronavirus replication. *J Integr Med,* 26(4), 243–250.

22.Ma, L., & Yao, L. (2020). Essential oil composition, total phenolic, flavonoid contents, and antioxidant activity of Thymus species

collected from different regions of Iran. *Molecules,* 25(11).
23.Mason, HS., & Arntzen, CJ. (1995). Transgenic plants as vaccine production systems. *Trends Biotechnol,* 13, 388–392.
24.Mason, HS. (1996). Expression of Norwlak virus capsid protein in transgenic tobacco and potato and its oral immunogenicity in mice. *Proceedings of the National Academy of Sciences USA,* 93, 5335–5340.
25.Mishra, N., Gupta, PN., Khatri, K., Goyal, AK., & Vyas, S.P. (2008). Edible vaccines: a new approach to oral immunization. *Indian J Biotechnol,* 7, 283–294.
26.Oszvald, M., Kang, T.J ., Tomoskozi, S., Tamas, C., Tamas, L., Kim, T.G., & Yang , M.S. (2007). Expression of a synthetic neutralizing epitope of porcine epidemi c diarrhea virus fused with synthetic b subunit of Escherichia coli heat labile enterotoxin in rice endosperm. *Molecular Biotechnology*, 35, 215.
27.Panyod , S., Ho, C.T, & Sheen, LY. (2020). Dietary therapy and herbal medicine for COVID-19 prevention: A review and perspective. *J Tradit Complement Med,* 10(4), 420–427.
28.Filgueira, P.DM., Robertson, N., Carrillo, C., Sadir, A.M., Morris, T.J., Borca, M.V., & Wigdorovitz, A. (2002). Protection of mice against challenge with foot and mouth disease virus (FMDV) by immunization with foliar extracts from plants infected with recombinant tobacco mosaic virus expressing the FMDV structural protein VP1. *Virology*, 264(1), 85–91.
29.Qian , B., Shen , H., & Liang, W. (2008). Immunogenicity of recombinant hepatitis B virus surface antigen fused with preS1 epitope sex pressed in rice seeds. *Transgenic Research,* 17, 621–631.
30. Runfeng, .L., Yunlong, H., Jicheng, H., Weiqi, P., Qinhai ,M., Yongxia .S., Chufang, L., Jin, Z., Zhenhua .J., Haiming, J., Kui, Z., Shuxiang, H., Jun, D., Xiaobo ,L., Xiaota, H., Lin,W., Nanshan, Z., & Zifeng, Y. Lianhuaqingwen exerts anti-viral and anti-inflammatory activity against novel coronavirus (SARS-CoV-2). *Pharmacol Res, 156.*
31.Rybicki, E.P. (2014).Plant-based vaccines against viruses, *Virology Journal*, 11.
32.Salyaev, R.K., Rigano, M.M., & Rekoslavskaya, N.I. (2010). Development of plant-based mucosal vaccines against wide spread infectious diseases. *Expert Rev Vaccines*, 9, 937–946.
33.Santarem, E.R., Trick, H.N., Essig, J.S., & Finer, J.J. (1998). Sonication-assisted *Agrobacterium*-mediated transformation of soybean immature cotyledons: Optimization of transient expression. *Plant Cell Reports*, 17(10), 752–759.
34.Sharma, M., & Sood, B. (2011). A banana or a syringe: journey to edible vaccines. *World Journal of Microbiology and Biotechnology*, 27(3), 471–477.
35.Srinivas, L., Sunil. S.G., Ganapathi, T.G., & Revathi, C. (2008). Transient and stable expression of hepatitis B surface antigen in tomato (*Lycopersicon esculentum* L.). *Plant Biotechnology Reports*, 2, 1–6.
36.Tohidi, B., Rahimmalek, M, & Arzani, A. (2017). Essential oil composition, total phenolic, flavonoid contents, and antioxidant activity of Thymus species collected from different regions of Iran. *Food Chem,* 220, 153–161.
37.Yang, JS., Hahn, B., Jeon, I., Jung, Y., Kim, J.B., Park, J., Ha, S., Kim, K.H., Kim, H.M., & Kim ,Y.H. (2007). Expression of hemagglutinin-neuraminidase protein of Newcastle disease virus in transgenic tobacco. *Plant Biotechnology Reports*, 1, 85–92.
38.Yan-Ju, YE., & Wen-Gui, LI. (2010). Immunoprotection of transgenic alfalfa (*Medicago sativa*) containing Eg95-EgA31 fusion

gene of *Echinococcus granulosus* against Eg protoscoleces. *Journal of Tropical Medicine*, 3, 10–13.
39.Zhang, H., Liu, M., Li, Y., Zhao, Y., He, H., Yang, G., & Zheng, C. (2010). Oral immunogenicity and protective efficacy in mice of a carrot-derived vaccine candidate expressing UreB subunit against *Helicobacter pylori*. *Protein Expression and Purification*, 69, 127–131.
40.iBio. (2020). iBio announces development of proprietary COVID-19 vaccine candidates. https://wwwglobenewswirecom/news-release/2020/03/18/2002546/0/en/iBio-Announces-Development-of-Proprietary-COVID-19-Vaccine-Candidateshtml
41.ISAAA. (2020). Native australian plant paves way for vaccine development against COVID. http://wwwisaaaorg/kc/cropbiotechupdate/article/defaultasp?ID=18054
42.Phillip Morris International. (2020). Medicago develops a plant-based vaccine for Coronavirus. https://www.pmi.com/media-center/news/medicago-develops-a-plant-based-vaccine-for-coronavirus