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Plant Derived Vaccines – New Door to Periodontal Vaccine

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Abstract:

Periodontitis is an immuno-inflammatory disease with multifactorial origin and have high association with systemic disorders such as cardiovascular disease, diabetes mellitus. The widespread nature of the periodontal disease initiated the need for its control and prevention. Plant-based vaccine is an emerging concept in field of vaccination. PubMed, Embase, Web of science, Cochrane, Medline and Google Scholar search engines were used to obtain relevant publications in medical and dental research and appropriate papers were reviewed and findings were briefly summarized. Preliminary studies in the field of medicine and veterinary sciences showed beneficial effect of these plant derived vaccines in a wide array of diseases. This comprehensive review enlightens their scope of application in periodontal therapy.

Key words: Periodontal disease, Plant derived vaccines, Plantibodies, Prevention.

INTRODUCTION:

In general vaccines following active immunization concept, involve stimulation of the antibody production in host by injecting attenuated whole bacterial cell, sub-unit vaccines and synthetic peptides and there by providing immune protection against diseases.[1] Genetic immunization or DNA vaccination are novel techniques in this domain, where genetically engineered DNA is used to elicit host response, eg: plasmid vaccines and live, viral or vector vaccines. [2] Traditionally, passive immunization is a process of supplementation of immunoglobulins such as monoclonal antibodies, to "at risk" individuals to interfere with microbial pathogenesis.[3] Plant based vaccines are an evolving thought of immunization, where plants are infected with genetically engineered DNA to express specific antigens or antibodies. In medicine, the use of transgenic plants for the expression of molecules with diagnostic and therapeutic applications has been very well documented during the last 20 years. In periodontics, they are still an emerging concept of vaccine preparation. This review narrates the scope of plant-based vaccines in periodontal therapy.

PLANT DERIVED VACCINES:

Plant based vaccines involve application of molecular biologic techniques to create transgenic plants which will be used to produce antigens or antibodies. In transgenic plants the DNA is modified by artificial insertion of desired genes using genetic engineering techniques. The inserted gene sequence is known as TRANS GENE.[4] The transgene can then be expressed in the plants either by a stable transformation system or by transient transformation system, depending on the location where the transgene has been inserted into the cells.[4] The first transgenic plant was reported in the year 1983.

- Stable transformation method:

Stable transformation can be done through nuclear integration. It occurs due to the permanent changes caused in the recipient cell genetics, where the target transgene is integrated into the genome of the host plant cells. Laere et al ., 2016 stated that stably transgenic plant cells produce a lower amount of subunit antigen, in the range of 0.01 to 0.30% of total soluble plant protein.[4]

- Transient transformation method:

Transient transformation method involves the production of desired protein (Plantibody) soon after the heterologous gene resides transiently in the host cells. The transgene is not incorporated into the genome of the plant cells. In this method, the regeneration of whole plant is not required and the frequency of its occurrence is higher. Plants use their endomembrane and secretory systems to produce large amounts of clinically viable proteins which can later be purified from plant tissue. The expressed plantibodies can be any kind of antibody molecule, ranging from the smallest antigen-binding domains/fragments to full length or even multimeric antibodies.

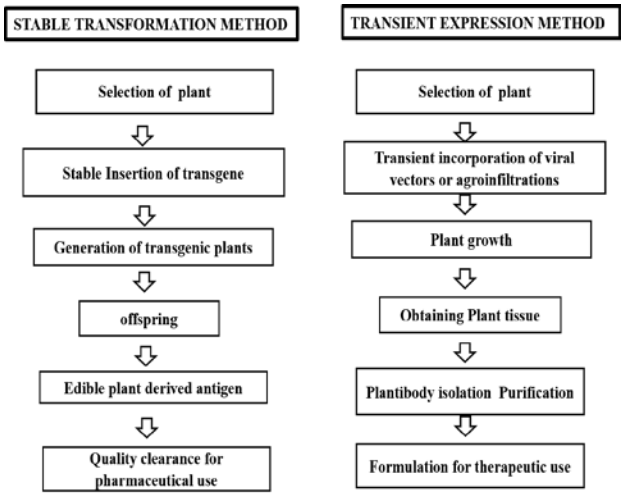

Figure 1: Steps in procuring the plant derived vaccines

Applying the above principles, many recombinant proteins have been expressed in several agronomic species of plants such as tobacco, corn, tomato, potato, banana, alfalfa and canola (Hammond et al.,1999).[5]

ADVANTAGES: Plant based vaccines are highly stable with higher degree of functionality. Application of plants as transgenes account for a large amount of preparation of pharmaceutical compounds with increased therapeutic value. Plants may not have or produce human pathogens.[6] As a result, the resulting vaccines will not have major safety issues for humans as compared to other vaccines such as heat killed vaccines or whole cell antigen which may transmit the pathogenicity.

DISADVANTAGES: Compared to other vaccines, the foremost disadvantage of plantibodies are, that edible vaccines are very likely to be mistaken with normal fruits, and thus, may be consumed more than formulated. Thus, education regarding the consumption of such products are a necessity to ensure adequate safety. Edible vaccines are flushed through a person's system relatively quickly, in a matter of hours or days, before the host's immune system has adapted to producing antibodies.

DISCUSSION:

Plants being a bio-Factories in the production of recombinant proteins offers an exciting alternative for pharmaceutical production of antibodies in large scale. Transgenic plants that express antigens in their edible tissue might be used as an inexpensive oral vaccine production and delivery system. Thus, immunization might be possible through consumption of an "edible vaccine to provide active immunization (Mason et al.1995).[7]

SCOPE IN MEDICINE: In medicine, these vaccines were explored in numerous conceivable ways and the plant-based vaccines were developed towards Bacteria (New castle diseases)[8], Viruses (Human papilloma virus)[9]. Further, vaccines for malignancies such as ovarian, testicular, colon cancer as well as B-cell lymphoma have been under research[10]. A few of the formulated Plant derived vaccines are listed in table 1

SCOPE IN DENTISTRY: The need for dental vaccines becomes vital because of the epidemic nature of these diseases. Further vaccines may be beneficial for individuals at risk of advanced periodontal destruction namely, immunocompromised patients by improving the host resistance. Plant based vaccines i.e Plantibodies opened the new door in the dental vaccination. Very few studies are available regarding the application of plant based vaccines towards dental diseases (table 3)

DENTAL CARIES: At current, two plant based dental caries vaccines were studied, namely **CaroRx** [19[and **Guy's 13 Plantibody.**[20] They were prepared from transgenic tobacco plants and these plantibodies bind specifically to Streptococcus mutans, inhibiting the bacterial adherence to salivary agglutinin. (Robinette et al. 2011) [20]

PERIODONTITIS: The concept of periodontal vaccination was initiated in 1980's. Among these virulent pathogens, Porphyomonas gingivalis is considered as the key stone pathogen. Hence, mainstream of research in periodontal vaccination was focussed on controlling this candidate pathogen and the various vaccines were developed from the virulence factors of P. gingivalis using outer membrane proteins, gingipains, fimbriae, heat shock proteins, lipopolysaccharides (LPS) as sub-unit vaccines or as a whole cell antigen. Table-2 gives the literature studies towards the available periodontal vaccine from 1980's till date.

PLANT DERIVED VACCINES IN MEDICINE			
Author	**Year**	**Plant**	**Vaccine**
Zeitlin [11]	1998	Soybean	Herpes simplex virus glycoprotein B
Kapusta [12]	1999	Lupin	Plant derived edible vaccine against Hepatitis B virus
Franconi [9]	2002	Tobacco	Plant-derived human papillomavirus 16 E7 on coprotein
Hull [13]	2005	Tobacco	Antibodies engineered to bind to *Bacillus anthracis*
Berinstein [14]	2005	Tobacco	New castle disease virus (NDV) vaccine
Broadzik [15]	2006	Tobacco	Antibodies against Lewis Y antigen in cancer
Shaaltiel [16]	2007	Carrot	Production of glucocerebrosidase as enzyme replacement therapy in Gaucher's disease
Lai [17]	2010	Nicotiana benthamiana	Monoclonal antibody produced in plants efficiently treats West Nile virus infection
Langreth [18]	2014	Tobacco	Ebola Vaccine

Table 1: Plant derived vaccines in medicine

YEAR	AUTHOR	PERIODONTAL VACCINES
1989	Mouton et al [21]	OMP of P. gingivalis
1990	Ebersole et al [22]	A.A Leukotoxin
1991	Yamashitha et al [23] Ebersole et al [24]	A.A specific Th cloneA3 Whole cell antigen of P.gingivalis and P.intermedia
1992	Kesavalu et al [25] Evans et al [26] Kesavalu et al [25]	Whole cell antigen of P. gingivalis Fimbriae of P. gingivalis LPS
1994	Person et al [27]	Formalin-killed whole cell P.gingivalis
1995	Holt et al [28] Chen et al [29]	Cell envelope Ag of P.gingivalis,P.intermedia, C.rectus, F.nucleatum & Cellwall Ag of A.viscus LPS of P. gingivalis
1998	Genco et al [30] Moritz et al [31]	Gingipain R Gingipain
1999	Katz et al [32]	Recombinant Hagb + Freunds adjuvant
2000	Simpson et al [33]	Rgp-Kgp Proteinase.

YEAR	AUTHOR	PERIODONTAL VACCINES
	Choi et al [34]	F.nucleatum ATCC +P.gingivalis specific T cell clone.
2001	Sharma et al [35] Gison & Genco et al [36]	Fim A of P.gingivalis Heat killed P.gingivalis + Freunds adjuvant.
2002	Rajapakse et al [37]	Arg X & Lys-x specific cysteine proteinases
2003	Decarlo et al [38] Gonzalez et al [39]	Recombinant HA2 +Freunds adjuvant. CPS of P.gingivalis.
2004	Robertz et al [40] Gemmell et al [41] Ross et al [42]	Formalin+ killed P.gingivalis in SAF adjuvant. F.nucleatum + P.gingivalis viable bacteria Genetic vaccine derived from P.gingivalis
2005	Simpson et al [43]	RgpA-Kgp + synthetic ABM
2006	Lee et al [44]	P.gingivalis r HSP 60
2007	Miyachi et al [45] Takahashi et al [46] Page et al [47]	Rgp A DNA Vaccine Fimbriae of P.gingivalis Cysteine proteinase
2008	Momoi et al [48]	OMP of P.gingivalis+ Nontoxix chimeric adjuvant
2009	Zhag et al [49]	OMP of P.gingivalis
2011	Muramatsu et al [50]	Rgp A DNA vaccine
2013	Yamamoto et al [51] Fan et al [52]	P.Gingivalis Ag Hgp 44 RgpA DNA vaccine in experimentalPeri-implantitis
2014	Han et al [53]	Whole genomic pg DNA in alum
2016	Simpson et al [54] Wilensky et al [55] Choi et al [56]	P.Gingivalis Gingipain Rh RapA Peptide vaccine P.Gingivalis Fim A plantibody
2017	Puth et al [57]	Hgp 44 domain polypeptide of Arg - gingipain(RgpA)

*OMP- Outer membrane protein, A.A-aggregatibacter actinomycetomocomitans, LPS-Lipopolysacchride, HagB-Haemeagglutinin, Rgp-Kgp- Arginine gingipains- Protease gingipains.

Table 2: Periodononta l vaccines

Al the above mention studies primarily focused on active immunization protocol for treating periodontal disease and so far passive immunization is not well explored in the field of periodontics. In the mission of appropriate periodontal vaccine development, Plantibodies attracted the focus in this era.

Choi et al. 2016 [56] studied **Anti-Fim A plantibody** as a potential vaccine candidate to control P. gingivalis – induced periodontal disease. They stated that anti-FimA plantibody produced in the rice cell suspension culture inhibits the biological activities of P. gingivalis through interactions with native fimbriae on the bacterial cells. Further the author suggested that, anti-FimA plantibody may be used as a mouthwash or topically applied into the subgingival area of teeth after periodontal scaling and root planing and intervention for periodontal disease.

PLANT DERIVED VACCINES IN DENTISTRY			
Author	**Year**	**Plant**	**Vaccine**
Larrick [19]	1998	Tobacco	CaroRx (Dental caries vaccine against streptococcus mutans)
Robinette [20]	2011	Tobacco	Guy's 13 Plantibody (Dental caries vaccine against streptococcus mutans)
Choi [56]	2016	Rice	Anti-Fim A plantibody (Periodontitis vaccine Antibodies against fim A of Porphyromonas gingivalis)

Table 3: Plant derived vaccines in dentistry

Limitations:

Invitro and animal studies showed that plant derived vaccines have appreciable therapeutic value to treat various diseases. However, there are many hurdles in their production i.e., such as selection of appropriate plant, selection of virulent antigen, formulation of therapeutic dosage and manufacturing by fulfilling the Good manufacturing Practices Procedures guidelines by WHO.[4] Though preliminary research is demonstrating their benefits, plantibodies are yet to be approved for human consumption due to the above-mentioned challenges and difficulties. Even then, development of a better and widely acceptable plant-based vaccine among researchers still remains intact.

FUTURE DIRECTIONS

1. Studies employing P. gingivalis strains with different types of fimbriae and other virulence factors are also required to investigate the plantibody and its usefulness for active or passive immunization to control and prevent the P. gingivalis-induced periodontal disease.
2. Further studies are needed to determine whether anti-FimA plantibody has any modulatory effect on the host immune response to P.gingivalis in vivo and to define an optimal antigen/antibody ratio for eliciting the beneficial immunomodulatory effect.
3. Since, periodontal disease is a poly microbial disease, production of plant derived vaccines towards multiple pathogens from a single plant variety would help in ease of therapeutic application.

CONCLUSION:

Periodontitis is stated as a multi-factorial and poly microbial disease. Hence, improving the host immune system in form of vaccination plays a protective role and

enable host to defend against periodontal pathogens. Utilizing the path of plantibodies in the periodontal vaccine may lighten up the way towards host immune modulation and in therapeutic management of periodontal disease.

Conflicts of interest: Nil

Acknowledgements: Nil

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