Recombinant DNA vaccine technology and their applications in cancer immunotherapy

Arun Kumar T

*Assistant Professor, Department of Chemistry, SNS College of Engineering, Coimbatore-641107, Tamil Nadu, India
*For correspondence e-mail: sakrunt@gmail.com

Article Info: Received 18 June 2016; Revised: 08 Sep 2016; Accepted 09 Sep 2016.

ABSTRACT

Activation of immune system against cancer is believed to be a decent strategy to counter this deadly disease. DNA vaccine platform have emerged with a lot of hope in scientific community and considered to be worth to explore against cancer. Practically these vaccines are easy to develop, cost effective and capable in generating long lasting immune response. Advancement in molecular biology and recombinant DNA technology with the identification of tumor-associated antigens has opened the uncharted territory for DNA vaccine strategies. DNA vaccine can be manipulated to express multiple antigens in single plasmid vector that can generate broad range of immune response. In addition, DNA vaccines allow inducing the immune response in different organelle, and their safety issues are explored in multiple studies. This article focuses on the background of DNA vaccine technology and recent studies performed in this area. This article will provide a brief overview on the development of DNA based vaccine to target cancer.

Keywords: Cancer, DNA vaccine, immunotherapy, cancer antigen.

1. INTRODUCTION

Cancer is one of the leading causes of death in the modern world and believed to surpass the heart diseases in coming years [1, 2]. Finding novel targets and ways to counter cancer has been priority of worldwide researchers. A normal cell converts into the cancerous cell by multiple ways including altered signaling pathways, mutation in genes, activation of oncogenic pathways [3–7], DNA damage [8–10], compromised DNA repair [11, 12] and chemotherapy or radiotherapy induced bystander effects [13–16]. Several, therapeutic strategies including microRNA based therapies [6, 17], chemotherapy [18, 19] and application of toxins obtained from pathogen [20–25] have shown limited success against cancer. Thus, there is an urgent need for novel therapeutic interventions that can selectively target and eliminate tumor cells and induce long lasting protection. Tumor cell adopted the strategy to corrupt and hijack the host immune system by secreting growth factors and cytokines that helps in unchecked growth. The immune system of host is the natural defense mechanism that holds the capacity to combat diseases. Antitumor properties of the host immune system have been demonstrated by intratumoral pathogenic infections that promote the spontaneous tumor regression [26]. This study motivated researchers to explore the possibilities of cancer
immunotherapies including therapeutic cancer vaccines. Vaccines have been used as a preventive measure against diseases that activates immune system to produce antibodies that can neutralize the antigens. It has been demonstrated that eukaryotic cells can express the genes encoded on plasmid DNA after transfection [27]. These findings led the foundation for DNA based vaccine which is considered as third generation vaccines [28]. It has been suggested that the cancer antigens that specifically presented on tumor cells can be utilized in the development of DNA vaccines against cancer [29]. In this article, we explored the background of DNA vaccine technology and its mechanism of action. Further, we have described the several DNA vaccines that have been analyzed in laboratory and in clinical trials to provide the strong rational to researches that may encourage the development of novel DNA vaccine based immunotherapeutic.

2. Background of DNA vaccine technology

DNA vaccine consist of a plasmid backbone having eukaryotic or a viral promoter that promotes the expression of recombinant antigen within a targeted or mammalian host cells. The concept of these recombinant vaccines was introduced by Wolff et al. 1990s by delivering plasmid DNA intramuscularly into the animal and confirming direct protein expression in vivo [27]. However DNA vaccine attains scientific limelight in early 1990s when Tang and Johnston demonstrated the induction of humoral immune responses after DNA vaccine delivery [30]. Further DNA vaccine was shown to protect mice from the lethal influenza challenge [28, 31] by generating CD8+ CTL responses [28]. Moreover, administration of DNA vaccine into the neonates found to stimulates strong humoral and cellular immune responses [32, 33]. These were the early examples that created a buzz in scientific community. DNA vaccines have several advantages over conventional vaccines, including the ease of design alteration in plasmid vector, optimization of antigen and modification of the sub-cellular localization [34, 35]. Easy production and low cost makes DNA superior over other conventional vaccines currently used in the field. Further the application of immunoregulatory genes can improves the immune response against target antigen. The initial findings in the area of DNA vaccine are promising and have been proven effective against a number viral and bacterial infection in preclinical models [36, 37]. These vaccines are believed to be third generation vaccines. The USA Food and Drug Administration (FDA) and the World Health Organizations recommendations are optimistic in this area.

3. Route of immunization

DNA immunization can be performed by administration into intramuscular, epidermal, mucosal, and intravenous routes [31]. Transfer of gene into of muscle fibers was demonstrated with the help of reporter genes such as β-galactosidase or firefly luciferase. Further the β-galactosidase activity was confirmed up to 5% after intramuscular inoculation of plasmid [27]. Studies performed on direct and particle-mediated delivery of this vaccine demonstrated that the particle mediated delivery in epidermics of mice requires 5000 times less DNA than the delivery of DNA vaccine with saline solution in muscular or dermis [38]. Different routes such as intramuscular, intravenous, intranasal, oral, intradermal, intraperitoneal, vaginal, and direct application onto the skin or via scarification were applied to introduce DNA vaccine into animal [39, 40]. Interestingly the delivery method is found to be crucial to determine the amount of DNA require to induce immune response in animals. For example intramuscular injection needs DNA, from 10 μg to 1 mg, whereas gene-gun deliveries require 100 to 1000 time less DNA than intramuscular immunization [41]. However, different species need variable quantity of DNA vaccine to induce protection from disease or pathogen [42]. Further, gene-gun mediated introduction of DNA is believed to be the most efficient mode of delivery [31]. Growing body of evidence suggest that the route of administration and the nature of the adjuvant significantly affect the immune response induced by DNA vaccine [31, 40, 43]. Alternative delivery methods which are greatly appreciated recently are aerosol instillation and mucosal surfaces administration by nasal, lung mucosa and vaginal route. The advantage of the mucosal surface administration is the mucosal surface area of the gastrointestinal, genitourinary and respiratory tract is more than 200 times greater than that of the skin.

4. DNA vaccine mechanism of action

Hypothetically after the DNA administration, cells presumably take up DNA from the extracellular spaces or during injection and produce the encoded protein in the plasmid. Removing the muscle after 10 minutes of DNA shown to be ineffective to induce immune response suggesting that transfection of somatic cells at the site of injection is an essential step in DNA induced immune response [44]. Studies suggest that the host lymphocytes (antigen presenting cells) recognize antigen expressed by DNA vaccine is [45,46] and present in to CD8+ cytolytic T lymphocytes. The CD8+ cytolytic T cells recognize a
non-covalent trimeric complex consisting of major histocompatibility complex (MHC) class I heavy chain allele. Contrary to this β2-microglobulin and 8-12 amino acid long peptide antigen presented by dimeric major histocompatibility complex (MHC) class II molecule. Protein that produced endogenously processed in the cytosol of the cell by multicatalytic proteasomes and presented by MHC class I and become accessible to CD8+ T cells. However the exogenous antigens are presented by MHC class II molecule and become accessible to CD4+ T cells. After recognition MHC, priming of T-cell requires the presentation of antigen by host’s professional antigen presenting cells (APC) that provide co-stimulation. Dendritic cells (DCs) are known to be most potent APCs in animals [47]. Further the role of APCs in the induction of the immune response following DNA vaccination has been determined by Iwasaki [48] and Corr [49]. Further the DNA vaccine studies suggest that APCs can directly induce immune response although, transfected somatic cells can serve as a reservoir for antigens.

5. DNA vaccine and cancer immunotherapy

The foundation of the development of DNA based vaccine to target cancer is based on the introduction of tumor antigens into the host to induce immune response that may clearance of tumor cells. The immune response induced against tumor cells can be useful and effective due to its specificity and effectiveness. Several immunotherapy approaches such as adoptive T cell therapies, cytokine therapies, and efficacy of cancer vaccines have been analyzed in pre-clinical and clinical settings. However, the DNA based vaccines seems promising strategy to immune responses as these vaccines can also elicit antigen specific humoral and cell mediated immune responses [50]. The backbone of plasmids used in DNA vaccines is usually bacterial origin that stimulates innate immune responses [50]. Further the bacterial plasmid serve as a ligand and stimulates Toll-like receptors (TLRs) which are membrane-spanning proteins on DCs, crucial for recognizing pathogen-associated molecular patterns. Activation of TLRs induces a cascade of pro-inflammatory responses that leads to the production of various cytokines which attract other immune cells and induces specific immune responses [51].

Efficacy of several DNA vaccines against cancer have been explored in recent past. To target malignant melanoma several DNA based vaccine that encodes tumor associated antigens (TAAs) were developed and tested in clinical trials. Administration of DNA vaccine encoding Melan-A (MART-1) and tyrosinase have been demonstrated to induce humoral and cell-mediated immune responses [52]. Further, delivery of DNA vaccine encoding human gp100 or xenogeneic mouse gp100 [53] showed mild effect on immune response. Though, no difference in progression-free survival was observed in patients. Another study that compared the immune responses generated by intramuscular or particle mediated epidermal delivery of the xenogeneic gp100 DNA [54] suggest that particle mediated epidermal delivery induce higher IFNγ+ CD8+ T cell production with lower dose of DNA. Although no significant clinical outcomes were observed as only 30% of the vaccinated patients displayed immune responses. To target breast cancer, Her2/neu; an oncoprotein that overexpressed on breast cancer cells is used as antigen for DNA vaccines. This DNA vaccine were delivered with IL-2 and GM-CSF in HER2-expressing breast cancer patients and shown to generate long-term antibody responses [55] one more DNA vaccine developed to target DCs by encoded chimeric rat/human HER2 were able to induce T cell immune responses and decreased tumor growth. Further, the vaccine inhibited the immune suppressive activity of regulatory T cells and TGF-β [56]. DNA vaccine encoding magmoglobin-A (Mam-A); that is overexpressed in breast cancer generated mam-A-specific CD8+ T cell immune responses. Further, this vaccine induced the production of IFN-γ and promoted the lysis of breast cancer cells [57].

As for as application of DNA vaccine against other cancers are concerned, the effects of DNA based vaccine were analyzed in colorectal cancer, prostate cancer and cervical cancer. Modified human carcinoembryonic antigen (CEA) gene fused with T helper epitope of the tetanus toxoid, demonstrated the immunogenic potential in mice. However, few patients show cancer recurrence, after DNA vaccination treatment [58].-Targeted prostate cancer DNA vaccine containing prostatic acid phosphatase (PAP), shown to induce PAP-specific CD8+ T cell immune responses [59]. Further, multiple boosting of DNA vaccine found to induce the immune responses [60]. Human papillomavirus virus (HPV) infection is believed to be the major cause of cervical cancer. Targeting HPV E6 and E7 antigens by DNA vaccine demonstrated to induce potent humoral and cell-mediated immune responses in mice. A phase I trial of this vaccine demonstrated histologic tumor regressions in 33% of the patients [61]. Another study shows that DNA vaccine was able to induce
HPV specific CD8+ T cells with higher levels of IFN-y production [62].

6. CONCLUSION

DNA based vaccine strategy seems lucrative approach to target several diseases and especially the cancer which express antigens exclusively limited to the tumor cells. The ability of DNA vaccine to induce long term immune response makes it valuable asset as cancer have the ability for recurrence that can be countered by immunological memory created against specific antigens by DNA vaccine. Further, the ongoing studies and positive results obtained from multiple clinical trials tells the success story of this platform. Interestingly, genes expressing multiple antigens can be inserted single plasmid DNA and broad range of immune response can be induced. Moreover, clinical trials performed with DNA vaccine have confirmed the safety and tolerability of several DNA vaccine platforms. Further, administration of plasmid DNA is well tolerated by cancer patients with minimal to no toxicities and integration of DNA plasmid into the host genomes is also not a concern. Considering the advantages of DNA vaccine over other conventional vaccine, it can be the decent immunotherapeutic approach against cancer.

Conflict of Interest

The authors declare that they have no conflicts of interest.

References


