Polymeric hydrogel based systems for vaccine delivery: A review

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ABSTRACT

The appropriate delivery of vaccines is a significant factor for the proper immunization. The proper delivery of the cargo vaccine/antigen along with stimulation of high antigen mediated immune response are the prime factors of an efficient vaccine deliver system. Different delivery systems have been explored for vaccine delivery and immunization. However, significant limitations like inefficient immunogenicity and undesired inflammatory immunogenic reactions are also notable. Thus, the development of an efficient vaccine delivery system is challenging task. Polymer based systems have also been utilized for vaccine delivery. Research also have indicated that a polymeric hydrogel can become an efficient delivery system of foreign antigens and vaccines. These systems can harbor and deliver the cargo vaccine/antigen in desired target organ and also facilitate the antigen mediated immunogenicity. Keeping in view the above perspectives, an attempt has been made to review the significance of polymeric hydrogel based systems for vaccine delivery.

Keywords: Polymer, Hydrogel, Vaccine delivery system Vaccine, Immunogenicity, Vaccination

1. Introduction

Vaccination is one of the remarkable achievements of biomedical science **[1-4]**. Development of a vaccine had equipped human population from deadly infectious diseases like influenza. The importance of immunization through vaccination relies on the fact that, it not only equips the immune system to recognize and protect from an infectious disease but also provide a passive protection in the community level by facilitating herd immunity **[5,6]**.

Modern biotechnological approaches have notably developed different vaccines like DNA vaccines, recombinant subunit vaccines, synthetic peptides, nano-vaccines **[3,7-10]**. The major objective of these vaccines to develop antigen mediated immune response against the pathogen. Several vaccine delivery systems have been developed to deliver the vaccine to the target site and to enhance the efficiency of the system by increasing the immunogenicity **[11]**. Various researches reported the efficiency of different systems like polymeric systems, nanoparticle based systems, three dimensional scaffold based systems have been demonstrated their efficiency in vaccine delivery **[12-15]**. These systems facilitate the slow release and delivery of antigen molecule in a way where booster doses will

not require. In addition, it also ensures the significant presentation of pathogenic antigen molecules to the immune cells **[16].** On the other way, an efficient vaccine delivery system has also shown their ability to work as an "adjuvant". As an adjuvant, it interacts the human immune system and elicits the immunogenic response. Earlier research reported several limitations regarding these delivery systems which involve: problem of vaccine delivery to the target site, inability of the sustain release of the vaccine molecule from the delivery system; problem in delivery of 'chemically diverse antigen molecule' **[4,17]**. In addition, other significant factors like biosafety, degradability of the delivery system are also notable concerns **[15].** Thus a new improved safe delivery system is needed for vaccine delivery and efficient immunization. This need of improved vaccine delivery systems and vaccine administration strategy are thus prominent requirements for the significant global immunization coverage **[18]**.

Polymeric hydrogel based vaccine delivery systems were explored in different researches for their ability and efficiency for vaccine delivery. The hydrogel based systems are unique in the sense that they exhibit the efficiency of the system through delivering the antigen/vaccine significantly to the target anatomical/physiological region, could also act like an adjuvant and could facilitate antigen mediated immune response **[4, 15,19-29]**. The advantage of hydrogel based delivery systems over other are: (i) this system can become an alternative method of needle based parental vaccine conventional administration, and (ii) it can stimulate the antigen mediated immune response of the delivery system compared to free antigen inside the immune system, (iii) polymers within the hydrogels can undergo degradation within the physiological system, (iv) these hydrogel based systems can be prepared with biocompatible polymers, which lowers the possibility of tissue cytotoxicity, however; they can stimulate significant immunogenicity **[4,18,30-33]**. Keeping in view the above perspectives, an attempt has been made to review the development of hydrogel based vaccine delivery systems.

Table 1 Nucleic acid delivery systems.

Delivery systems	Descriptions	Ref
Virus based delivery systems	This system utilizes a viral gene carrier like adenovirus, lentivirus, retrovirus. In this process, the genetic material which was transferred is integrated within the host genome. This finally expressed through protein synthesis. The safety risk is high for this process.	[48–52]
Non-viral delivery systems		
Gene gun	This method is also called 'DNA-coated particle bombardment'. This process employs heavy metal microparticles like gold, silver to carry nucleic acid and administration to the target cells. Different advantages like high efficiency, easy to generate make this method exciting; however, pressure of the applied gas and dose frequency are limitation of the method.	[48,51,53, 54]
Electroporation	This is also a 'physical gene delivery systems. This method utilizes significant pulse of electric current to develop cellular pores (10 nm) for the transfer of the foreign genetic material. The significant limitation of this method involves high cell death due to the application of high voltage.	[51,53, 55]
Ultrasound	This method is an efficient method for the successful nucleic acid i.e. DNA transfer to the target cells. Ultrasound mediated DNA transfection is in pre-clinical trials and has considered as less efficient than virus based delivery systems.	[55–57]

The objective of this review involves to explore and evaluate the recent progresses regarding the significance of different hydrogel based delivery systems specific for efficient vaccine delivery and generation of antigen/vaccine specific immunogenic response.

2. Vaccine delivery systems and vaccine administration

Efficient vaccine delivery systems must elicit appropriate immune response and facilitate the establishment of immunological memory to prevent the future risk of infection **[34,35]**. The outcome of vaccine delivery and immunization depends significantly on the path of vaccine administration. An inappropriate administration of vaccine could enhance the chances of disabled effectivity of the vaccines [**36,37**]. Theoretically, the closer the vaccine is delivered near the lymph node or lymphatic vessel, the stronger immune response is developed. However, this is also influenced by the different factors like appropriate antigenic epitopes and functionality of adjuvants [**37**]. Two primary strategies were considered for the development of efficient vaccine delivery systems: parenteral vaccine delivery systems and non-parenteral vaccine delivery systems.

The parenteral vaccine delivery system follows the administration of vaccine through intradermal, subcutaneous and intramuscular routes through application of hypodermic needles [**38,39**]. This system is widely used systems for vaccine delivery. The vaccine administered to the body according to the anatomical features of the injection site-from outside to inside: epidermis, hypodermis, dermis [**39,40**]. The body site for the administration of parenteral vaccine systems is also crucial. For the efficient administration of this vaccine system in the adults, the deltoid region is selected for the intramuscular and the intradermal delivery; on the other hand, for the subcutaneous delivery, outer triceps area is usually considered. However, the anterolateral thigh region is considered for the toddlers and infants [**39,41,42**].

Table 2 Adjuvant based vaccine delivery systems.

Adjuvant based vaccine delivery System	Description	Ref
First generation Adjuvants		
Emulsion based delivery system	Different oil in water based formulations have been developed for efficient vaccine delivery systems. Emulsion based system like MF59 used to administer for vaccines delivery like influenza, herpes simplex virus etc. This system stimulate cytokine, chemokine production. However, emulsion based systems often exhibit toxicity.	[58, 59]
Liposome based delivery system	Liposome based delivery systems are found to elicit significant cell-mediated and human immune response. Thus they are considered as exciting material for vaccine delivery. Unilamellar liposomes have been utilized for hydrophilic antigen and multilamellar liposomes are utilized for hydrophobic antigens. However, this process has limitation like rapid removal of liposome by mononuclear phagocyte system (MPS).	[60, 61]
Second generation Adjuvants		
Toll-like receptor (TLR) agonists	TLR agonist have considered as effective adjuvant. TLR based system can stimulate APC maturation and immune response.	[35, 62]
Molecular adjuvant	Molecular adjuvant like Bcl-xL anti-apoptotic protein is also utilized as molecular adjuvant for DNA vaccine delivery. This adjuvant facilitate $CD8^+$ T cell immune response for the vaccine delivery of antigens like *T. gondii.*	[48]

In this system, the needle needs to be selected on the basis of muscle size, tissue thickness and other factors which vary with age, sex, body mass for each individual person [**39**].

Albeit, the needle based injection delivery device has been considered as an exciting method for vaccine delivery, however, significant limitations like needle stick injuries, expensive for using are the major drawbacks for this approach. Thus, needle free devices developed for making the process efficient than needle based devices. Furthermore, the pain sensation during the parenteral vaccine administration is considered as a notable disadvantage [**18**]. To address this problems, different non-parenteral, needle free vaccine delivery systems were developed. Different needle free devices like powder, liquid, projectile systems have been utilized. On the other hand, different jet injectors like spring load, battery powered, gas powered jet injectors have been used to develop needle free device for vaccine administration [**43,44**]. How-ever, these methods have limitations for the development of mucosal immunity **[38,45,46**]. However, on the other hand, non-parenteral vaccine system also utilizes the administration of live, killed antigen molecule to elicit antigen mediated immune response. The non-parenteral route of administration of vaccines involve oral route, intranasal, transcutaneous routes [**38**]. The various approaches of this delivery strategies involve nucleic acid delivery system (DNA vaccines) (Table 1), adjuvant based delivery systems (Table 2), microparticle based vaccine delivery system [**35,47,48**].

3. Polymers in vaccine delivery systems

Polymers have been used for the matrix antigen delivery system [**16, 63-66**]. Different biodegradable and biocompatible polymers enhances the efficiency of the vaccine by facilitating the betterment of the targeting ability specific to an organ [**67**] Different delivery systems are prepared with polymers for vaccine delivery systems like liposomes, microsphere, etc. [Fig. 1., Table 3]. Synthetic and natural polymer fabricated vaccine delivery systems facilitate the releasing environment through 'site-specific degradation' and created suitable platform for the efficient delivery of vaccine [**32**]. The polymer based vaccine systems exhibit significant stability and high antigen loading capacity [**32,68, 69**].

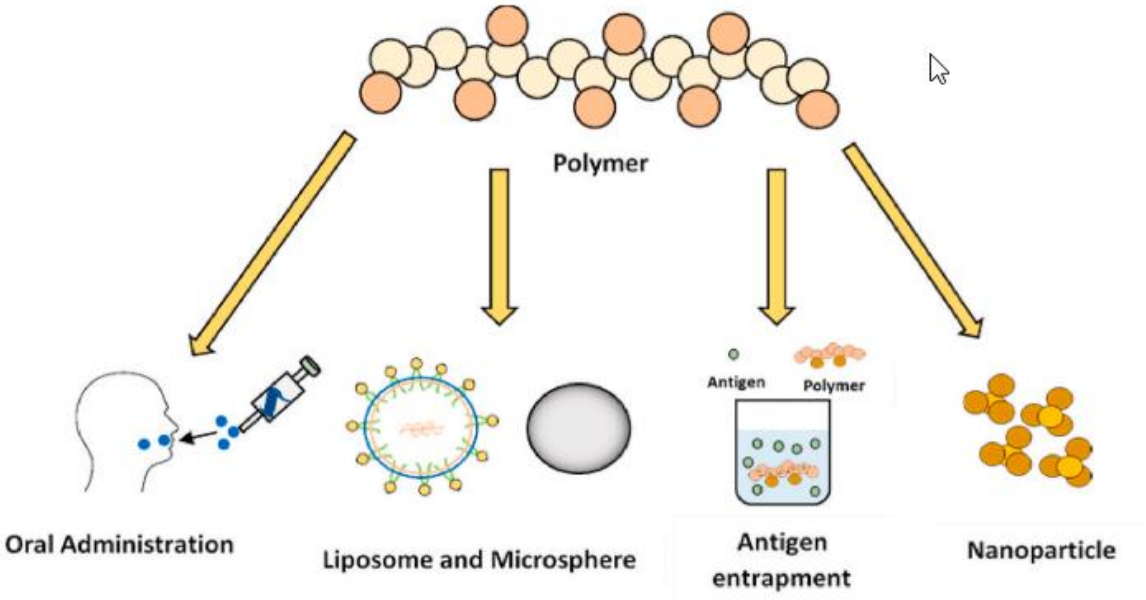

Fig. 1. Application of polymer in vaccine delivery (own concept).

Table 3 List of polymer used in vaccine delivery systems.

Polymer based systems	Application	Ref
Synthetic polymer based systems		
PLGA microsphere	Recombinant tuberculosis antigen, recombinant protein of influenza	[48, 74]
PEG based system	Nanoparticle loaded DNA vaccine administration	[48, 75]
Polylactide based system	Recombinant malaria antigen	[16, 76]
PCL based system	Antigen loaded liposphere for vaccine delivery	[16]
Poly (phosphazenes)	Antigen entrapment at low temperature	[16]
PLG based system	DNA, protein and micro-particle based vaccines administration	[70]
PLG based systems	Nanoparticle loaded vaccine administration	[70]
Poly(oxazoline) based system	Bacterial lipopolysaccharide based antigen administration	[35, 77]
Natural polymer based systems		
Chitosan/hyaluronic acid based systems	Oral vaccine administration, nanoparticle based vaccine delivery	[32, 73]
Acetylated dextran based system	Anti-melioidosis vaccine delivery	[35, 78]

Synthetic polymers like poly(lactic-co-glycolic acid) [PLGA], poly (lactic acid) [PLA], polyethylene glycol [PEG], polylactide, poly-caprolactone [PCL], poly(alkyl cyanoacrylates), poly(methyl methacrylates), poly (phosphazenes), polylactide (PLA), poly (fumaric-co-se bacic), poly(lactide-co-glycolides) [PLGs], polylactide-co-poly(ethylene glycol) [PELA] and carboxymethylethylcellulose, poly(oxazoline) have been utilized [**32,35,38,70-72**]. On the other hand, natural polymers such as polysaccharides like chitosan, p-glucan, acetylated dextran and poly ((R)-3-hydroxybutyrate) (PHB) synthesized by some bacteria were also used [**35,73**].

4. Polymeric hydrogels for vaccine delivery systems

In the recent time, different polymeric hydrogel based systems have been developed for vaccine delivery [**4,71,79-86**]. The hydrogel based system encapsulates a vaccine molecule which is being released upon entering the body of the animal and can possibly undergo degradation.

The delivery of various hydrogel based devices containing the vac-cine/antigen molecules has been performed for oral, intramuscular and transcutaneous immunization [**67,71**]. In the mid-90s, poly (meth-acrylic acid) hydrogels have been developed for the model antigen delivery to the ruminants. This hydrogel based model antigen delivery system administered orally to the gastrointestinal tract of sheep [**87**]. Additionally, the DNA vaccines was successfully delivered with agarose hydrogel based device. This was a slow release system which successfully elicited antigen specific immune response in cattle [**88**].

On the other hand, a hydrogel based vaccination system have been reported by Ishii et al., 2008 [**89**], where it was focused the antigen delivery at the animal's transcutaneous region. The efficiency of this

system was also enhanced by an application of silver immersed patch to a hairless rat' s back skin. This transcutaneous hydrogel based vaccine delivery systems demonstrated notable antigen specific humoral immune response for adenoviral infection [**89**].

Different hydrogel based delivery systems were developed for efficient delivery of vaccine and thereby elicit the antigen mediated immune response (Fig. 2.).

a. Hydrogel Patch based system

Hydrogel based systems were used to develop "hydrogel patch" for successful immunization process. Matsuo et al., 2011 [**90**] have demonstrated a hydrogel patch containing tetanus and diphtheria toxoid, that could efficiently introduce antigenic proteins to the outer layer of the skin and could induce toxoid specific IgG development near the patch area. Furthermore, Hirobea et al., 2012 [**91**] developed a hydrogel patch for the tetanus and diphtheria vaccination for human individuals. This hydrogel patch was also harboring diphtheria and tetanus toxoids, that was applied to human study volunteers. There was no adverse effect observed in the study individuals. However, the vaccination system was found to elicit efficient immune response.

b. Thermo-sensitive hydrogel system

Thermoresponsive property of hydrogel was also utilized for the development of hydrogel based vaccine delivery system. In this regard, Wu et al., 2012a [**92**] have developed a thermal-sensitive hydrogel vaccine delivery which was intended to utilize H5N1 influenza vaccination. The major constituent of this system was a solution of N-[(2-hydroxy-3-trimethylammonium) propyl] chitosan chloride (HTCC) and a, p-glycerophosphate (a, P-GP), which formed a hydrogel in the physiological temperature. This further efficiently deliver H5N1 antigen through intranasal route and stimulate antigen specific CD8+ immune response [**92**]. It has also shown that quarternized chitosan/a, P-glycerophosphatethermosensitive hydrogel based vaccination system exhibited notable humoral and cell-mediated immune response for Ebolavirus glycoprotein antigen. It has been thought that the thermal sensitivity of the hydrogel based system might have attributed the significant antigen mediated immune response [**93,94**]. Another study had also indicated the efficiency of chitosan based gel system for vaccine delivery specific to the prevention ovine brucellosis [**95**]. On other hand, a thermosensitive hydrogel based vaccine delivery system was developed with the polymers, PLGA and PEG for the model antigen, ovalbumin delivery. The sol-gel transition temperature of this hydrogel based system was shown around 32°C [**96**]. This hydrogel can also found efficient. Furthermore, for the immunization specific to Echinococcus granulosus infection in grazing animals, a thermosensitive chitosan hydrogel based delivery system was developed and found efficient [**97**]. Intranasal delivery of thermosensitive polymeric hydrogel based system with Gantrez® AN119 and the surfactant Pluronic® F127 (PF127) was also developed. This hydrogel based system shown a notable "immu-noadjuvant" ability [**98**].

c. Bio bullets

Hydrogel based biobullets have been developed for ballistic vaccine delivery. A poly(ethylene glycol) based hydrogel was prepared through photopolymerization as a vaccine delivery system to deliver bacterial vaccine. This hydrogel based system was especially developed in the form of 'thermoplastic degradable bio bullet's which was intended to use balistically to the bison. Brucellaabortus strain RB51 (RB51) live vaccines have been encapsulated and demonstrated notable viability with this hydrogel based biobullets [**99**]. In general, the PEG based hydrogel system have been considered efficient for vaccine delivery [**99-102**].

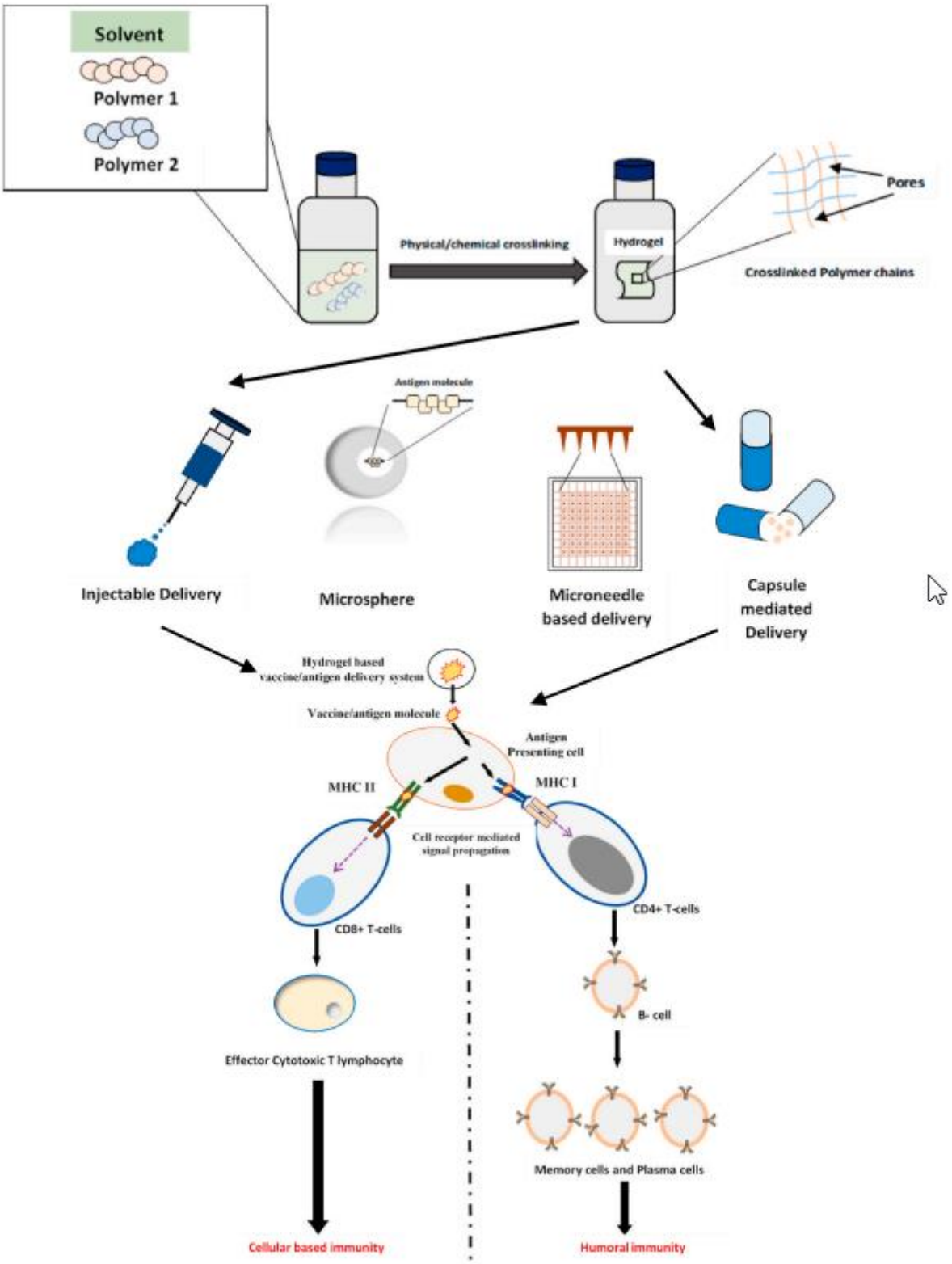

Fig. 2. Diagrammatic representation of different hydrogel based vaccine delivery systems. Polymers can be used to develop hydrogels by physical/chemical crosslinking; then they can be used to prepare different vaccine delivery system which will develop antigen mediated immune response.

In general, the PEG based hydrogel system have been considered efficient for vaccine delivery [**99-102**].

d. Capsule based systém

The hydrogel capsules were considered as exciting devices for vaccine delivery. Chong et al., 2009 [**31**] developed degradable polymeric hydrogel capsules for oligopeptide antigen delivery. This system involves oligopeptide antigens which were covalently linked to negatively charged polymeric component and then the conjugate adsorbed on amine functionalized silica.

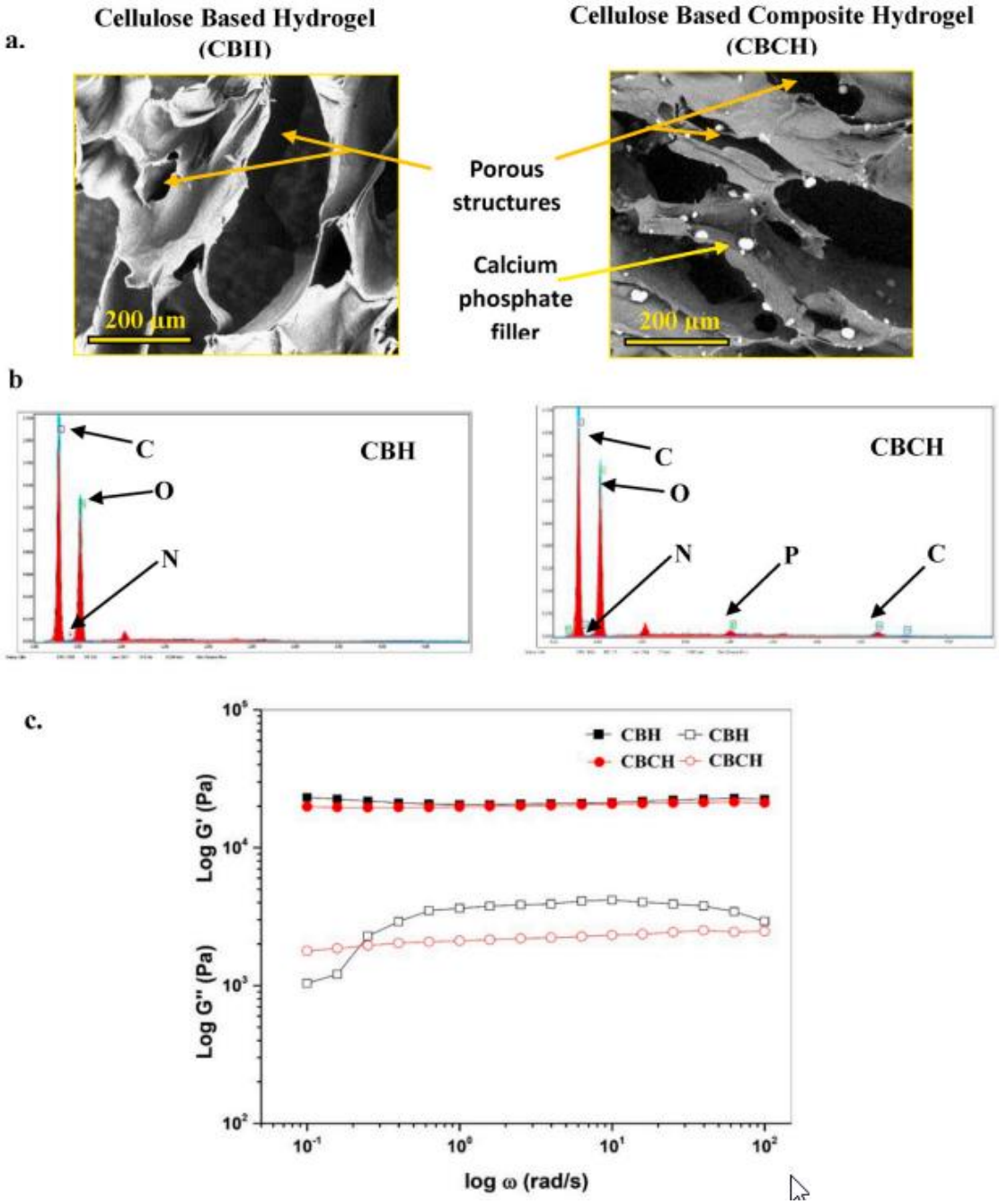

Fig. 3. Cellulose based hydrogels (CBH) and calcium filled cellulose based composite hydrogels (CBCH) and their properties. (a) SEM cross-sectional images showing porous structures; (b) SEM-EDX confirms the presence of the ingredients like cellulose and calcium within the hydrogel; (c) rheological properties (storage modulus [filled boxes] and loss modulus [open boxes]) of the hydrogels (Basu et al., unpublished data)

This vaccine delivery system has been found to deliver the antigen efficiently to the antigen presenting cells (APC) and thus could able to develop immune response. Furthermore, Sexton et al., 2009 [**103**] reported a modified poly (methacrylic acid) hydrogel capsule for the delivery of model antigen in the transgenic mouse model system [**16,103**]. In this system, poly (methacrylic acid) was modified by thiol groups, which covered themselves into disulfide linkage to make the delivery system stable with the external environment. This hydrogel capsule system showed degradation when it comes in contact with the cellular environment and released the vaccine to the target and finally the capsule is phagocytosed by immune system's APC and dendritic cells (DC) [**16**].

e. Peptide gels and nanogels

Peptide gels and Nanogels are considered as efficient vaccine delivery system. A novel peptide nanofiber hydrogel was developed by Li et al., 2013 [**104**]. This hydrogel based system has considered to act as an adjuvant for the vaccine delivery related to respiratory syndrome virus. A supramolecular peptide hydrogel was developed which acted as adjuvant for the vaccine for West Nile virus and significantly elicit immune response [**82**]. The peptide-hydrogel vaccine delivery system was also exhibited promising results as vaccine adjuvant which notably raised antigen mediated immune response [**105,106**] (Wang et al., 2017, Wang et al., 2018). On the other hand, Azegami et al., 2017 [**107**] developed a nanogel formulation for nasal vaccine delivery. Furthermore, a novel nanogel based vaccine deliver system was also developed for the delivery of immunogenic proteins. This nanogel was composed of ethylenediamine groups which notably interact with the antigenic protein molecule. The system has high efficiency of antigenic protein delivery [108]. The nanogel based devices generally entrapped and interacted with the antigenic molecules through the hydrophobic interactions with the polymeric network of the gel [**109,110**].

f. Injectable hydrogel systems

Different injectable hydrogel based systems have also been developed for efficient vaccine delivery. Huang et al., 2011 [**111**] reported the potential self-assembling peptide hydrogel which can work as an adjuvant for swine H1N1 influenza killed vaccine. Moreover, Wu et al., 2011 [**112**], developed an injectable hydrogel based system containing biodegradable nanoparticle. This system involved nanoparticle loaded with human basic fibroblast growth factor (bFGF) which was introduced into a hydrogel. This system convert into a gel at physiological temperature and release the cargo. This delivery system demonstrated the production of strong immune response in presence of the antigen molecule. Furthermore, Wu et al., 2012 [**113**] developed PCL and PEG based injectable hydrogel based delivery system which demonstrated notable antigen mediated immunogenicity. Additionally, an injectable hydrogel has also been developed by utilizing pentablock co-polymer made up of PEG, PCL, PLA for sustain release of vaccine. This vaccine has demonstrated a significant ability in the context of antigen mediated immune response [**114**]. A novel injectable self-assembled poly (L-valine) hydrogel delivery system was also developed for the dual delivery of antigen molecule and TLR agonist. This system exhibited high antigen persistence at the delivery site. Additionally, the hydrogel based vaccine formulation showed antitumour effect in melanoma bearing mice [**115**]. Another injectable hydrogel formulation was developed that encapsulates model antigen, ovalbumin and granulocyte-macrophage colony stimulating factor for efficient delivery of antigen and enhanced immunogenic reaction [**116**]. Furthermore, a vaccine system was developed with PEG-b-poly (L-alanine) for the codelivery of immune check point inhibitor and a tumour vaccine. This vaccine system induced tumor-infiltrating CD8+ T-cells and has found efficient for immunotherapy related to B16F10 melanoma [**86**]. Asai et al., 2019 [**117**] also developed an injectable polypeptide hydrogel system which can able sustain delivery of cargo antigen. On the other hand, an injectable polymer-nanoparticle hydrogels were also developed which can deliver the subunit vaccine. In this type of hydrogel vaccine delivery system, the

polymer and the nanoparticles were interacted with dynamic and multivalent noncovalent interactions [**4**].

g. Micro-particle, liposome based system

Different polymeric microparticle, liposome based systems were also prepared [**118**]. Polymer such as Poly (e-caprolactone) was used to prepare microparticle loaded with model antigen bovine serum protein (BSA) for oral vaccine delivery [**119**]. Jianga et al., 2005 [**30**] have been prepared a PLGA microparticle based system for the controlled delivery of protein antigens. A gel core liposome was also developed by using the synthetic polymer, poly acrylic acid (PAA) for intramuscular delivery of 'transmission blocking malaria vaccine'. In this system, the liposome core was gel based and prepared with biocompatible polymer PAA within the phospholipid bilayer **[120**]. Gordon et al., 2010 [**121**] have developed chitosan hydrogel with silica nanoparticle containing model antigen molecule, ovalbumin (OVA). This hydrogel based nanoparticle containing antigen delivery system have been found to efficiently induce CD4+ T cell proliferation which demonstrates its functionality. Furthermore, Hariyadi et al., 2014 [**122**] developed an efficient vaccination formulation strategy for the preparation of freeze dried oral vaccines using alginate hydrogel microspheres. On ther hand, quater-nized chitosan hydrogel microparticles were developed for porcine reproductive and respiratory syndrome virus (PRRSV) inactivated vaccine [**123**] and H5N1 split vaccine [**124**].

h. Microneedle based systems

Hydrogel based microneedle system has become a notable approach for vaccine delivery [**125**]. The hydrogel based microneedle vaccine delivery system could swell after in contact with interstitial skin fluid [**126,127**]. The model protein antigen (like ovalbumin) can then release through the hydrogel structure into the skin. The hydrogel based

microneedles were developed by using different polymers and polymeric mixtures like PEG and poly (methylvinylethercomaleic acid) with sodium carbonate [**126**]. The hydrogel based microneedle will then can be removed intact from the skin region after the delivery of the vaccine. This device has found efficient for the delivery of cargo vaccine [**127**].

5. Cellulose based vaccine delivery system

Cellulose has been considered as significant biopolymer which is abundant in nature [**128**]. This polymer is majorly synthesized by the plants. However, certain bacterial strains also synthesize bacterial cellulose [**129**]. This natural polymer has a significant hydrophilicity and biocompatibility [**130**].

Cellulose based hydrogel formulation has a significant material property. Research indicated that cellulose based hydrogel exhibit significant cross-sectional porous structures and viscoelastic property (Fig. 3.), which are important characteristics of this hydrogels to become an efficient delivery system for vaccines [**94,131**].

Study reported that various cellulose derivatives like ethyl cellulose (EC), methyl cellulose (MC), hydroxypropylmethyl cellulose (HPMC), hydroxypropyl cellulose (HPC), ethylhydroxyethylcellulose (EHEC) have been utilized in different biomedical application and for the development of vaccine delivery systems like vaccine bullets [**99,132**]. Bacterial nanocellulose and polyacrylic acid based hydrogel microparticles were utilized for the delivery of model antigen, ovalbumin. The study showed the high entrapment efficiency and release of ovalbumin by hydrogel microparticles and notable antigen mediated immune response through the production of significant anti-Ova-IgG in the intestinal region of mice [**133**]. Furthermore, similar study was performed with antigen containing nanocellulose

hydrogel based delivery system. This system was also found efficient to elicit antigen specific immune response through stimulating interferon-y-producing cells. This nanocellulose based system exhibited notable cellular infiltration and vaccine delivery. Interestingly, after the delivery the hydrogel matrix was found to be degraded by the cell itself [**33**]. Recently, Kim et al., 2020 [**134**] has developed Microneedle based vaccine delivery system with car-boxymethyl cellulose at room temperature for SARS-CoV2 antigenic protein which could become an interesting vaccine delivery vehicle for SARS-COV2 immunization in future.

6. Conclusion and future perspective

A suitable vaccine delivery system not only ensures the proper delivery of the vaccine, but also plays a crucial role to generate strong immune response within the animal immune system. Thus, the development of a vaccine delivery system is crucial for the efficiency of the vaccine molecule by enhancing its effect and thereby facilitating immunogenic response. The application of hydrogel based systems for vaccine delivery have been considered as an exciting approach. Different formulations like hydrogel based bio-bullets, microspheres, microneedles, injectable delivery systems have been developed for efficient vaccine delivery. These delivery systems have found significant in antigen mediated immune response.

Various synthetic and natural polymers have been utilized to develop hydrogel based delivery systems for vaccination. It has been reported that these systems have been successfully deliver the cargo vaccine/ antigen molecule through nasal, injectable route and can elicit a significant immune response. Natural polymers like chitosan, cellulose have been utilized for the development of hydrogel based vaccine delivery system. On the other hand, PEG, PLA have also been used. These polymers are biocompatible and thus showed promising results in vaccine delivery. Hydrogels contain significant porous structures and rheological properties (as shown in Fig. 3). These hydrogel-based devices can efficiently deliver the vaccine cargo to the immune system [**94**].

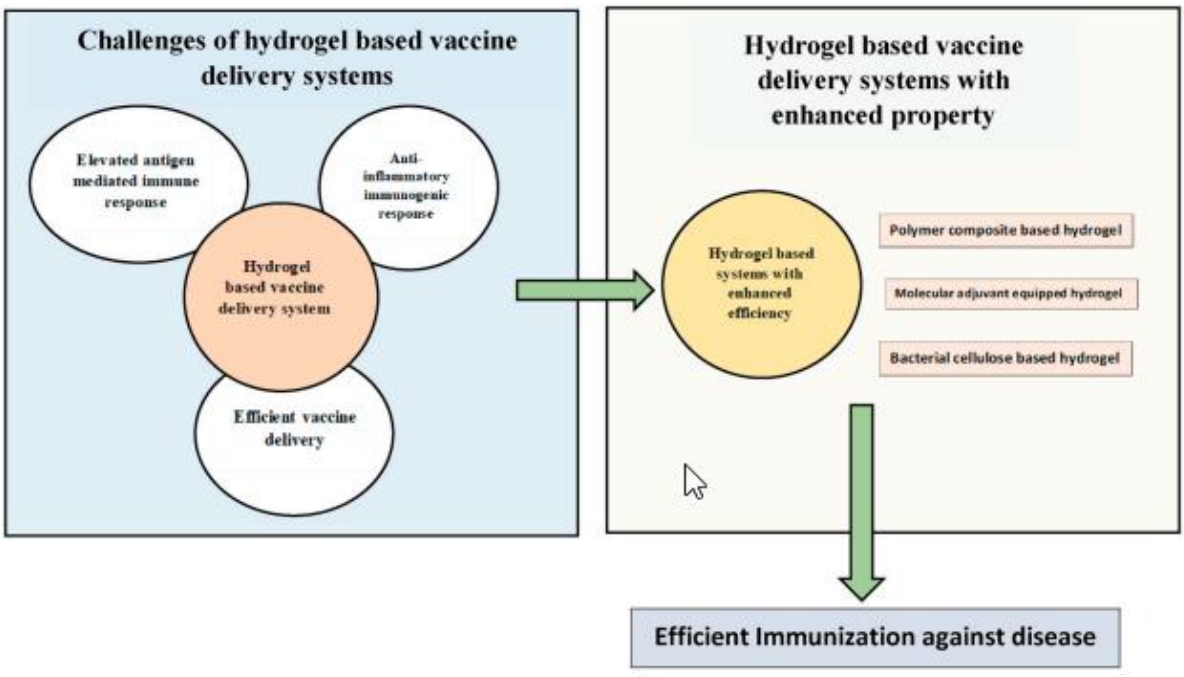

Fig. 4. Challenges and future prospects of hydrogel based vaccine delivery systems.

The key challenges related to the application of hydrogel based delivery systems are: (i) it must play a key role in facilitating the development of an elevated antigen mediated immune response; (ii) the hydrogel based system must face and bypass the inflammatory immunogenic response; (iii) it must exhibit efficient vaccine delivery. Thus, considering the need a holistic approach is needed, where the structural characteristics of the system like porosity must comprise the necessary properties related to the efficient vaccine delivery. In this context, utilizing a hydrogel based composite could be a promising approach (Fig. 4.). Hydrogel composites can be prepared with biocompatible polymers, reinforcement/particulate filler materials etc., which can develop an efficient hydrogel based system that can strongly recruit immunological cells [**135**]. There is a meagre amount of researches has been performed with hydrogel based composite system for its efficiency for vaccine delivery [**4,110,135**]. The system could be reinforced with various reinforcement agents like nanoparticles, filler particles and reinforcement fibers like cellulose fibers. Thus can become an effective vaccine delivery system. However, the interaction between the components of the composite hydrogels and the interaction with the vaccine must also be analyzed before its application.

Previous research indicated that application of molecular adjuvants can enhance the immunogenicity of a vaccine delivery system [**48**]. The efficiency of vaccine delivery systems can thus be enhanced by the simultaneous utilization of various other adjuvant molecules with the hydrogel based delivery device which will further strengthen the immunogenic response within the animal.

Bacterial cellulose based composite hydrogel system can also be utilized. The significant difference between cellulose and bacterial cellulose is that the bacterial cellulose is free from hemicellulose and lignin impurities. On the other hand, the bacterial cellulose nanofibers are also considered as reinforcement agents of a polymeric hydrogel system [**130**]. Composite hydrogels prepared with bacterial cellulose might increases the efficiency of the vaccine delivery system. Additionally, the composite can be prepared by utilizing bacterial cellulose and particulate filler materials like calcium phosphates, carbonates. This composite hydrogel also contains significant characteristics like porosity and rheological properties. Thus this hydrogel could also become a plausible system for vaccine delivery due to its notable properties. However, further studies are required to understand the role of bacterial cellulose based composite hydrogel system for its efficiency in regards to its immunogenicity and for the delivery of a vaccine.

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