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Construction and Immunological Evaluation of Immunostimulatory Nanocomposites with Vaccine Potential

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Abstract

For a long time, the most important means of controlling infectious diseases is prevention, and vaccination is considered the most effective measures, so the preparation and improvement of the vaccine is not only supported by the medical profession, it is the concern of the broad masses of the people. A number of biomolecules have been used as antigens to construct novel vaccine preparations, including Virus-like particles (VLPs) or bacteria, dendritic cells, nucleic acids and peptides. Immunostimulatory nanocomposites are based on the above molecules as vectors, encapsulating immune adjuvants, and nanoparticles for the construction of novel vaccines. Compared to traditional vaccines, adjuvants, nanoparticles alone, the efficacy of immunostimulatory nanocomposites as potential vaccines shows increased $CD4^+$, CD8+ T cell count, secretion of cytokine interferon-gamma and IL-4 functions also significantly enhanced, in addition, the use of nanoparticles enhances the Immune irritation of adjuvants, VLPs and other molecules on humoral and cellular immune responses. Thus, immunostimulatory nanocomposites may be potential vaccines for future prevention and treatment applications.

Keywords: Immunostimulatory nanocomposites; Virus-like particles; Vaccine; Adjuvant; Nanoparticles; Humoral and cellular immunity

Background

The concept of vaccine in the early development has been simply set for the disease to produce immunity inactivated or attenuated pathogens, that is, vaccines are made from pathogens [1]. Traditional vaccines include vaccines for current national immunization programs and live attenuated hepatitis A vaccine, rubella attenuated live vaccines, mumps attenuated live vaccines and rabies vaccines [2]. Traditional vaccines are limited by the production process, and their storage, use and transportation requirements are high and side effects are prone to occur after inoculation, and the immune effect is affected by the condition of the body [3]. Further studies will be necessary to address these questions.

In recent years, with the progress of modern biotechnology, and in-depth understanding of pathogen causative factors, the definition of the vaccine has also changed. It is now believed that the vaccine is a pathogenic protein (polypeptide, peptide), polysaccharide or nucleic acid, in the form of a single component or complex particles containing active ingredients, or through live attenuated pathogens or carrier after entering the body can produce inactivated, destroys or inhibits pathogenic specificity immune response [4]. Vaccines aim at generating the body's protective and therapeutic immune responses against many diseases, such as infectious diseases and cancers [5]. A variety of new vaccines (hepatitis B subunit vaccine, HIV vaccine and genetic engineering vaccine [6-8]) research has become a hot issue.

With advances in immunology and molecular biology, in order to compensate for the shortcomings of vaccine formulations, this requires the systematic design of vaccines [9]. The rapid development of nanotechnology provides new hope for the generation of new adjuvant systems based on nanoparticles. At present, there are two kinds of nano-adjuvant: inorganic nano-adjuvant and organic nano-adjuvant, according to the two adjuvants made of different Immunostimulatory Nanocomposites. The construction and immunization of this new vaccine remains to be further studied.

Construction and Immunological Evaluation of Inorganic Nanocomposites

Currently, the most studied inorganic nano-adjuvant complexes include nano-aluminum compounds, calcium nanoparticles, nano-gold particles and so on. The preparation methods of nanoparticle composite carrier include the physical and chemical methods [10], the following paragraphs describe the construction and related immune function of several inorganic nanocomposites.

The nano-aluminum compounds are the first nano-vaccine to be studied [11], in which the aluminum hydroxide adjuvant is also approved by the FDA as a human vaccine adjuvant. Tang Cheng et al. [12] used Newcastle disease virus as antigen, first produced a sterile aluminum hydroxide nanoparticle suspension and conventional aluminum hydroxide suspension, and then mixing the two with the inactivated ND antigen. The results show that Nano-aluminum hydroxide adjuvants can induce higher levels of antibodies to stimulate the expression of CD4 and CD8 molecules in chicken peripheral blood lymphocytes. Therefore, Nano-aluminum hydroxide adjuvant can help chicken body produce more humoral immunity and cellular immunity than conventional aluminum hydroxide adjuvant. There is evidence that humoral and cellular immunity are important for preventing viral pathogens such as HIV. Andreas Frey et al. [13] co-link the HIV-1gp 120 C4 region to the peptide co-conjugate to the nano-Al2O3 adjuvant. Compared with the adjuvant MDP (hydrophilic muramyl dipeptide), nano-Al2 O3 Can induce the strongest antibody titers and have the strongest reactivity to HIV-1gp 120-infected cells. The results

show that this novel vaccine method can be used to induce an immune response against conformation sensitive viral antigens without the need for additional adjuvants.

In the inorganic nano-adjuvant, people have more research on calcium nanoparticles; the results found that calcium nanoparticles as immune adjuvant have a unique advantage [14]. The nanoparticle CA can be used as an anti-idiotypic antibody NP30 for schistosomiasis vaccines. Feng Zhen-qing et al. [15] immunized BALB/c mice with CA-NP30 activity to prepare CA-NP30 conjugate (CA-NP30). The results showed that CA could significantly enhance the protective immunity of NP30 to *Schistosoma japonicum* infection. Compared with NP30 alone immunized group, the specific IgG, IgG1, IgG2a and anti-NP30 antibody levels of CA-NP30 immunized group were significantly increased. The concentration of IFN-γ in supernatant of spleen cells was greatly increased. CA-NP30 may enhance the humoral and cellular immune response in mice. Sanganagouda Koppad et al. [16] coupled calcium phosphate (CaP) particles with inactivated Newcastle disease virus (NDV) vaccines. Compared to commercial live vaccines (RDV'F '), CaP-conjugated NDV vaccines can also induce long-term hematocrit inhibition (HI) and enzyme-linked immunosorbent assay (ELISA) titers, (CMI) from 2 weeks to 4 weeks after inoculation. In general, CaP-conjugated NDV vaccines resulted in a stronger and longer immune response than commercial live vaccines. So the nano-calcium phosphate can induce the body to produce a strong immune response, activation of the immune system to achieve the effect of immune protection, and nano-phosphate can reduce the injection of local inflammatory response, and induced Th1 cell response, better than traditional hydrogen aluminum adjuvant. Calcium nanoparticles can induce innate immunity and adaptive immunity through DC activation. Viktoriya Sokolova et al. [17] prepared a vaccine made of CpG, poly (I: C) functionalized calcium phosphate nanoparticles, and combinations thereof. In addition, the viral peptide of influenza A virus hemagglutinin (HA) is encapsulated in the granules. Their goal is to develop calcium phosphate nanoparticles that can act as carriers of immunologically active oligonucleotides into dendritic cells for activation. It was observed that functionalized CaP nanoparticles induced congenital inflammatory cytokines IL-12p70 and TNF-α and strongly stimulated antigen-specific $CD4^+$ T cells. Depending on the antigen encapsulated in the nanoparticles, they can become effective antiviral vaccines by their ability to induce effective antigen-specific cellular immunity.

Nano-gold particles can be used as a new generation of vaccine research. Wang Yarun et al. [18] synthesized a novel immunostimulating nanocomposite (CpG-Au@HBc VLP) by encapsulating CpG gold nanoparticles with self-assembling engineered virus-like particles using the principle of electrical interaction. Monodisperse and uniform size of CpG-Au@HBc VLP showed an increased number of $CD4^+$, $CD8^+$ T cells and a greater secretion of cytokine interferon-gamma than HBc VLP with conventional Freund's adjuvant adjuvant. Besides, using Au nanoparticles also enhanced immunogenicity of CpG and VLP in humoral and cellular immune pathways. Listeria is caused by *Listeria monocytogenes (Listeria monocytogenes)*, which affects pregnant women and their newborns, and also affects the presence of tumors or chronic ones that are immunologically impaired Patients with immune disease. There is currently a need to produce an effective vaccine that produces protective T cell immunity. Ricardo Calderon-Gonzalez et al. [19] have now developed an effective T cell vaccine with the gold nanoparticle antigen delivery method in conjunction with the novel polysaccharide nanoparticle adjuvant (GNP-LLO91-99 nano-vaccines formulated with AdvaxTM), Electrophoresis results show that the vaccine is cytotoxic and is effective in delivering Listeria epitopes to DCs *in vitro* and *in vivo* and induces effective CD4 and $CD8^+$ T cell immunity, providing a powerful animal model for listeriosis Protective effects.

Construction and Immunological Evaluation of Organic Nanocomposites

With the global research on nano-vaccines continue to achieve results, organic nano-adjuvant also gradually into the research hot spots. The organic nanocomposites are mainly Polylactic-coglycolicacid (PLGA), chitosan and liposomes [20-22]. The following paragraphs describe the construction and related immune function of several organic nanocomposites.

PLGA (Polylactic acid-glycolic acid copolymer) is a copolymer of lactic acid (Lacticacid, LA) and Glycolicacid (GA), is a novel organic nano-adjuvant, which has good stability and safety. Zhang Weifeng et al. [23] used PLGA nanoparticles to formulate antigens by encapsulating the antigen within the nanoparticles or by simply mixing the soluble antigen with the nanoparticles. The results show that the combined formulation (composed of antigen in the nanoparticles, antigens mixed with the nanoparticles) induced a more potent antigen-specific immune response than each one-component preparation. Mice immunized with a combined vaccine formulation showed enhanced antigen-specific IgG antibody induction with high affinity, increased secretion of cytokines by spleen cells, and improved production of memory T cells.PLGA nanoparticles are a powerful carrier for vaccines. Corbin Clawson et al. [24] synthesized PLGA-NP loaded with Hp91 peptide, or by conjugation to its carrier on the surface. It was subsequently found that nano-PLGA and short peptide carrier-coated post-stimulated DC (Dendritic cells) cells were equivalent to 5 times the immune effect. In addition, Hp91 conjugated to NPs may potentially cross-link the receptor, leading to stronger DC activation. In conclusion, ISP (immunostimulatory peptides) Hp91 delivered by PLGA-NPs not only retains its ability to activate DCs, whereas PLGA-NPs carrying Hp91 are stronger than free peptides in activating DCs. Wang Fang et al. [25] encapsulated pcDNA-SG encoding T and B cell epitopes of foot-and-mouth disease virus (FMDV) into PLGA microparticles and then immunized mice. Confocal laser scanning microscopy showed that the expression of SG immunogen in lymphocytes of mice incubated with PLGA-pcDNASG microparticles was prolonged compared to mice immunized with naked pcDNA-SG. PLGA-pcDNA-SG particles showed virus-specific antibodies with higher titers, exhibiting stronger immunogenicity, IFN-production and lymphocyte proliferation. PLGA-DNA particles can enhance humoral and cellular immunity.

Chitosan is a polysaccharide consisting of a copolymer of N-acetylglucosamine and glucosamine, derived from chitin, which is found in crustaceans. M. Iqbal et al. [26] constructed a nano-vaccine with a respiratory syncytial virus (RSV) anti-cancer gene plasmid with chitosan, which induces peptide and viral specificity in BALB/c mice compared to intradermal immunization induction CTL response to intranasal immunity. A significant reduction in viral load was observed in the lungs of immunized mice compared to the control group after RSV challenge in chitosan/DNA immunized mice. These results indicate that the vaccine significantly inhibits the inflammatory response of lung tissue and exhibits a good immune enhancement effect. Zhai Yongzhen et al. [27] encoded plasmids of the pre-membrane (prM) and envelope (E) proteins of Japanese encephalitis virus and mouse GM-CSF (pJME/GM-CSF), then injected into the

mouse muscle to raise large and multifocal large aggregates of macrophages and granulocytes. The chitosan-PJME/GM-CSF nanoparticles prepared by intramuscular injection can prolong the duration of infiltration of the injection site, increase the content of APC in the spleen, improve the function of DC, induce CTL activity was significantly higher than that of pJME. These results indicate that the vaccine is superior to standard pJME/GM-CSF administration in DC recruitment, antigen processing and expression, induction of cellular immunity, and enhancement of vaccines. The above results show that chitosan-βJME/GM-CSF nanoparticles enhance the immunological adjuvant performance of GM-CSF by intramuscular injection, which provides experimental basis for the extensive application of chitosan.The development of a new and effective vaccine against *Mycobacterium tuberculosis* (M.tb) is a challenge to prevent tuberculosis infection. Feng Ganzhu et al. [28] developed a novel nanoparticle-based recombinant DNA vaccine comprising Esat-6 T-cell epitopes (Esat-6/3e) and fms-like tyrosine kinase 3 ligand (FL) genes (Called Esat-6/3e-FL) and encapsulated with chitosan (CS) nanoparticles. The plasmid DNA vaccine was then intramuscularly injected into C57BL/6 mice and enhanced with Esat-6/3e peptide. The results showed that immunized mice significantly enhanced T cell responses and protected against M.tb H37Rv, and nano-chitosan significantly increased the immunization and protection of DNA vaccines, whereas nano-Esat-6/3e-FL was a vaccine against M.tb in mice.

Liposomes are used as effective vectors for gene transfer to have a greater advantage than viral vectors. Hamouda et al. [29,30] developed an influenza vaccine adjuvant oil-in-water nanoemulsion-based adjuvant W805EC, which immunized ferrets or mice by nasal cavity to produce safe and specific cellular and humoral immune responses. The hemagglutination inhibitory antibody produced by the nanoemulsion-based adjuvant vaccine was 19 to 90 times that of without the adjuvant vaccine. The amount of antigen used in the immunized ferret was 1/50 of the adjuvant vaccine, indicating that the use of NE adjuvant intranasally administered antigen to effectively produce mucosal and serum antibody responses as well as the strong cellular Th1 immune response, Immunized nanoemulsion-based adjuvant can effectively enhance the immunogenicity of influenza vaccine hemagglutinin antigen, reduce the amount of antigen, help reduce the cost of vaccines and accelerate the mass production of influenza vaccine. James J. Moon et al. [31] used a lipid double layer cross-linked multilayer liposome carrier, allows the encapsulated protein to remain at a high concentration over a long period of time and slowly release in the blood. They immunized the mice with the vector-encapsulated antigen OVA and the Immune irritation monophosphoryl lipid A (MPLA) and found that these vesicles carrying the antigen/adjuvant formed a very potent whole protein vaccine, causing endogenous T cells and antibody responses comparable to those of the strongest vaccine vector, which can significantly enhance humoral and cellular immune responses.Hu Yun et al. [32] assembled liposomal protein-based nanoparticles as a nicotine hapten delivery system. They constructed nanoparticles by conjugating the model hapten carrier protein (bovine serum albumin (BSA)) to cationic liposomes. As a result, the NicAb titers that could be caused by the lipid complex vaccine with Alum were significantly higher than that induced by either the vaccine without Alum or Nic-BSA with Alum. It was confirmed that the nano-lipid complex had significant immunostimulatory effect.

With the advancement of medicine and the continuous development of nanotechnology, there are some other organic nano-carriers as a new adjuvant. Nano-bead conjugated peptides can provide effective and universal immunization regimens to induce protective immunity and are potentially used for immunotherapy. Theodora Fifis et al. [33] conjugated peptides containing ovalbumin (OVA)-dependent CD4 and CD8T epitopes to 0.05 μm nano-beads and found that these nano capsules produced strong immune responses and inhibit the growth of tumor cells expressing CD8T epitopes, experiments show that the inert nano-sphere (particle size 40~50 nm) adjuvant mainly induced $CD8^+$ T cell immune response. It allows dendritic cells to concentrate on lymph nodes, stimulate APC recognition for antigens, induce high levels of IFN-γ, and to some extent enhance humoral immune responses. In order to verify that carbon nanotubes can be used as new vaccines or drug delivery devices, it has now explored their complementary interactions with a portion of the human immune system. Carolina et al. [34] reported that carbon nanotubes can activate the complement system through classical and bypass pathways, and they demonstrate that the blood complement system is activated by classical pathways and bypass pathways, while observing the effects of carbon nanotubes on complement and plasma proteins , Indicating that carbon nanotubes can act as immune adjuvants. Anthracycline plays an indisputable key role in the treatment of many cancer diseases. Simeonova et al. [35] found that poly-butylcyanoacrylate, which combines Epirubicin, inhibits NK cell activity, without binding or simple mixing of the reference sample will stimulate the activity of NK cells.

In summary, the above-mentioned immunostimulatory nanocomposites are capable of keeping the antigen effective to stimulate the body to produce antibodies as compared to the use of conventional vaccines, common adjuvants, nanoparticles alone [36], induce the strongest antibody titers, and thereby stimulating humoral immune responses. Moreover, most of them can activate and ingest higher DCs, activate $CD4^+$ T cell subsets, induce Th1 response and promote cellular immunity. In addition, the nanocomposites can stimulate the secretion of IFN-γ, TNF-α, IL-2, IL-4, and further enhance the humoral and cellular immune function of the vaccine [18].

Conclusion

Through the process of dissociation and recombination, we developed immunostimulatory nanocomposites with potential for vaccine, which showed strong humoral and cellular immune stimulating ability. With advances in medicine and the continuous development of nanotechnology, immunostimulatory nanocomposites will serve as a new class of vaccines that will play an increasingly important role in medical research as they are superior to conventional vaccines and adjuvants. Thus, it is reasonable to believe that a combination of a new expression vector and more immunogenic antigens can stimulate a stronger immune response and provide better protection. The next step in our research direction will be more on the mechanism of the composite material in order to better apply to people.

References

1. Cunningham AL, Garçon N, Leo O, Friedland LR, Strugnell R, et al. (2016) Vaccine development: From concept to early clinical testing. Vaccine 34: 6655-6664.
2. Usonis V, Bakasenas V, Kaufhold A (1999) Reactogenicity and immunogenicity of a new live attenuated combined measles, mumps and rubella vaccine in healthy children. Pediatr Infect Dis J 18: 42-48.

3. Rathore AS, Bhalghat M, Patra AK (2012) Vaccines -key considerations for development and production of vaccine products. Biopharm International 25: S28-S34.
4. Jing Y, Wei Y, Feng Z (2003) Bacillus Calmette-Guerin Vaccine polysaccharide nucleic acid in treatment of congestive heart failure. Chin J New Drugs and Clin Remed 22: 33-35.
5. Larson HJ, Jarrett C, Eckersberger E, Smith DM, Paterson P (2014) Understanding vaccine hesitancy around vaccines and vaccination from a global perspective: A systematic review of published literature. Vaccine 32: 2150-2159.
6. Bertoletti A, Ferrari C (2003) Kinetics of the Immune Response during HBV and HCV Infection. Hepatology 38: 4-13.
7. Liu J, Ostrowski M (2017) Development of targeted adjuvants for HIV-1 vaccines. AIDS Res Ther 14: 43.
8. Dertzbaugh MT (1998) Genetically Engineered Vaccines: An Overview. Plasmid 39: 100-113.
9. Enrqiuez N, Delden CV, Siegrist CA (2005) Vaccine Prevention in Solid Organ Transplantation (SOT) Candidates and Recipients: A Systematic Approach is needed. Florida Entomologist 88: 247-252.
10. Yang L, Li W, Kirberger M, Liao W, Ren J (2016) Design of nanomaterial based systems for novel vaccine development. Biomater Sci 4: 785-802.
11. Rostami H, Ebtekar M, Ardestani MS, Yazdi MH, Mahdavi M (2017) Co-utilization of a TLR5 agonist and nano-formulation of HIV-1 vaccine candidate leads to increased vaccine immunogenicity and decreased immunogenic dose: A preliminary study. Immunol Lett 187: 19-26.
12. Cheng Tang, Xing Huang, Fa Long Yang, et al. (2008) Aluminum hydroxide nanoparticles against chicken Newcastle disease antigen Immune adjuvant effect. Chin J Vet Scien 38: 1060-1064.
13. Frey A1, Mantis N, Kozlowski PA, Quayle AJ, Bajardi A, et al. (1999) Immunization of mice with peptomers covalently coupled to aluminum oxide nanoparticles. Vaccine 17: 3007-3019.
14. He Q, Mitchell AR, Johnson SL, Wagner-Bartak C, Morcol T, et al. (2000) Calcium Phosphate Nanoparticle Adjuvant. Clin Diagn Lab Immunol 7: 899-903.
15. Feng ZQ, Zhong SG, Li YH, Li YQ, Qiu ZN, et al. (2004) Nanoparticles as a vaccine adjuvant of anti-idiotypic antibody against schistosomiasis. Chin Med J (Engl)117: 83-87.
16. Koppad S, Raj GD, Gopinath VP, Kirubaharan JJ, Thangavelu A, et al. (2011) Calcium phosphate coupled Newcastle disease vaccine elicits humoral and cell mediated immune responses in chickens. Res Vet Sci 91: 384-390.
17. Sokolova V, Knuschke T, Kovtun A, Buer J, Epple M , et al. (2010) The use of calcium phosphate nanoparticles encapsulating Toll-like receptor ligands and the antigen hemagglutinin to induce dendritic cell maturation and T cell activation. Biomaterials 31: 5627-5633.
18. Wang Y, Kang N, Liu Y, Shan W, Zhuang G, et al. (2016) Construction and Immunological Evaluation of CpG-Au@HBc Virus-Like Nanoparticles as a Potential Vaccine. Nanoscale Res Lett 11: 338.
19. Calderon-Gonzalez R, Marradi M, Garcia I, Petrovsky N, Alvarez-Dominguez C (2015) Novel nanoparticle vaccines for Listeriosis. Hum Vaccin Immunother 11: 2501-2503.
20. Sah H, Thoma LA, Desu HR, Sah E, Wood GC (2013) Concepts and practices used to develop functional PLGA-based nanoparticulate systems. Int J Nanomedicine 8: 747-765.
21. Vasiliev YM (2015) Chitosan-based vaccine adjuvants: incomplete characterization complicates preclinical and clinical evaluation. Expert Rev Vaccines 14: 37-53.
22. Hadinoto K, Sundaresan A, Cheow WS (2013) Lipid–polymer hybrid nanoparticles as a new generation therapeutic delivery platform: A review. Eur J Pharm Biopharm 85: 427-443.
23. Zhang W, Wang L, Liu Y, Chen X, Liu Q, et al. (2014) Immune responses to vaccines involving a combined antigene nanoparticle mixture and nanoparticle-encapsulated antigen formulation. Biomaterials 35: 6086-6097.
24. Clawson C, Huang CT, Futalan D, Seible DM, Saenz R, et al. (2010) Delivery of a peptide via poly(d, l-lactic-co-glycolic)acid nanoparticles enhances its dendritic cell-stimulatory capacity. Nanomedicine 6: 651–661.
25. Wang F, He XW, Jiang L, Ren D, He Y, et al. (2006) Enhanced immunogenicity of microencapsulated multiepitope DNA vaccine encoding T and B cell epitopes of foot-and-mouth disease virus in mice. Vaccine 24: 2017–2026.
26. Iqbal M, Lin W, Jabbal-Gill I, Davis SS, Steward MW, et al. (2003) Nasal delivery of chitosan–DNA plasmid expressing epitopes of respiratory syncytial virus (RSV) induces protective CTL responses in BALB/c mice. Vaccine 21: 1478-1485.
27. Zhai Y, Zhou Y, Li X, Feng G (2015) Immuneenhancing effect of nanoDNA vaccine encoding a gene of the prME protein of Japanese encephalitis virus and BALB/c mouse granulocytemacrophage colonystimulating factor. Mol Med Rep 12: 199-209.
28. Feng G, Jiang Q, Xia M, Lu Y, Qiu W, et al. (2013) Enhanced Immune Response and Protective Effects of Nano-chitosan-based DNA Vaccine Encoding T Cell Epitopes of Esat-6 and FL against Mycobacterium Tuberculosis Infection. Plos One 8: e61135.
29. Hamouda T, Sutcliffe JA, Ciotti S, Baker JR Jr (2011) Intranasal immunization of ferrets with commercial trivalent influenza vaccines formulated in a nanoemulsion-based adjuvant. Clin Vaccine Immunol 18: 1167-1175.
30. Hamouda T, Chepurnov A, Mank N (2010) Efficacy, immunogenicity and stability of a novel intranasal nanoemulsion -adjuvanted influenza vaccine in a murine model. Human Vaccine 6: 585-594.
31. Moon JJ, Suh H, Bershteyn A (2011) Interbilayer-Crosslinked Multilamellar Vesicles as Synthetic Vaccines for Potent Humoral and Cellular Immune Responses. Nat Mat 10: 243-251.
32. Hu Y, Zheng H, Huang W, Zhang C (2014) A novel and efficient nicotine vaccine using nano-lipoplex as a delivery vehicle. Hum Vaccin Immunother 10: 64-72.
33. Fifis T, Mottram P, Bogdanoska V (2004) Short peptide sequences containing MHC class I and/or class II epitopes linked to nano-beads induce strong immunity and inhibition of growth of antigen-specific tumour challenge in mice. Vaccine 23: 258-266.
34. Salvador-Morales C, Flahaut E, Sim E, Sloan J, Green ML, et al. (2006) Complement activation and protein adsorption by carbon nanotubes. Mol Immunol 43: 193-201.
35. Simeonova MY, Antcheva MN (2007) Effect of farmorubicin both free and associated with poly (butylcyanoacrylate) nanoparticles on phagocytic and NK activity of peritoneal exudate cells from tumor-bearing mice. J Drug Target 15: 302-310.
36. Ikoba U, Peng H, Li H, Miller C, Yu C, et al. (2015) Nanocarriers in therapy of infectious and inflammatory diseases. Nanoscale 7: 4291-4305.