



Cover Story

Core-shell polymer particles as flexible platform for vaccination



Vaccination is a facile and powerful way to protect against infectious diseases and other devastating illnesses including certain cancers. Traditional vaccines have relied on live-attenuated or inactivated organisms, attenuated bacteria, outer membrane vesicles, or inactivated toxins as the source for antigens [1]. Recently, recombinant protein subunit vaccines have been extensively investigated along with various adjuvants for the treatment of cancers or persistent viral infections, such as HIV. Therapeutic vaccination against such diseases needs not only long-term antibody responses, but also strong and effective specific cytotoxic T cell responses. Recent studies suggest that the use of nanoparticles for the co-delivery of a protein antigen and an adjuvant is effective in fortifying vaccine potency [2]. This approach guarantees that the antigen and adjuvant are delivered to the same antigen presenting cells, dendritic cells (DCs) in particular, to ensure that those DCs that take up the antigen are the ones being activated. While traditional adjuvants, such as aluminum and emulsions (e.g. AS03), can significantly enhance the antibody-mediated humoral responses, most of them failed to induce favorable cell-mediated immune responses.

Professor Wim Hennink and his colleagues, as well as others, have demonstrated that poly-uridine (PolyU), a ligand for toll like receptors (TLRs) 7/8, encapsulated into synthetic polymeric nanocomplexes can serve as a strong adjuvant [3,4]. This RNA adjuvant can trigger the innate immune system in such a way that it supports long-lasting antibody responses and strong CD8+ cytolytic T cell responses when co-administered with an antigen. Capitalizing on the promising adjuvant properties of RNA nanocomplexes, professor Hennink and his team have explored ways to co-deliver RNA (PolyU) and an antigen (OVA) in the same polymeric nanoparticle. Unlike traditional delivery systems such as PLGA particles that require co-encapsulation of both antigen and adjuvant within the polymeric core, the study by the Hennink team shows an innovative co-delivery approach in which an antigen is clicked onto a preformed polyU adjuvant nanoparticle [5]. To achieve covalent linkage of antigens to the core (RNA adjuvant) nanoparticles, two clickable cationic polymers that contain azide or bicyclo[6.1.0]nonyne (BCN) groups were introduced into the core nanoparticle. BCN-modified OVA and glycosylated polymers (containing mannose or galactose units) were sequentially clicked onto the RNA core. In addition, disulfide groups conveyed nanoparticle stability outside cells, but, once taken up by antigen presenting cells, they enabled release of the loaded antigen and RNA. The generated nanoparticles resemble the structural organization of viruses in terms of size and supramolecular organization,

and are therefore named virus mimicking particles (VMPs) despite being fully synthetic. They showed that these negatively charged reduction-sensitive VMPs (~200 nm, -14 mV) were stable in a physiological environment. The immunogenicity of these VMP vaccines have been tested both *in vitro* and *in vivo*. The surface mannosylated VMP (VMP-Man) showed 5 times higher cellular uptake by bone marrow-derived DCs compared to the galactosylated VMP (VMP-Gal) counterpart. Moreover, VMP-Man efficiently activated DCs and greatly facilitated the MHC I antigen presentation *in vitro*. Vaccination of mice with VMP-Man elicited robust OVA-specific cytotoxic T cell responses, as well as stronger humoral immune responses compared with adjuvanted soluble OVA.

The strength of the Hennink team's vaccine delivery system lies in the flexibility at which antigen shell layers can be "clicked" onto the universal RNA core nanoparticle, enabling versatile adaptation of the vaccine. This avoids, unlike the traditional PLGA-based nanoparticulate vaccines, the requirement for encapsulation of the antigen, which is, from a pharmaceutical point of view, cumbersome as different antigens will have different requirements for optimal encapsulation. Moreover, since the antigen is physically linked to the adjuvant nanoparticle, it assures co-delivery of adjuvant and antigen into antigen presenting cells, necessary for robust immune responses. This elegant core-shell approach may have clinical utility, especially for personalized cancer vaccines based on tumor neoantigens, which often need a fast and flexible manufacturing platform. Moreover, important from a pharmaceutical perspective is that the vaccine platform developed in this study can be stored in a dry-powder form after freeze-drying favoring vaccine shelf-life. Although this study demonstrates the colloidal stability and targeting ability of these VMPs, further work is required to assess their drainage properties to sentinel lymph nodes, where immune responses are orchestrated. In addition, since only a model antigen OVA was studied in this work, the efficacy of this vaccine platform should be tested in more relevant animal models for tumor growth, such as spontaneous occurring tumors in mice or dogs. Overall, the work by the Hennink team is a new, original approach to antigen delivery using a core-shell platform that affords flexibility necessary for personalized vaccines.

References

- [1] G.J. Nabel, Designing Tomorrow's Vaccines, *N. Engl. J. Med.* 368 (2013) 551–560.
- [2] M.-L. De Temmerman, J. Rejman, J. Demeester, D.J. Irvine, B. Gander, S.C. De Smedt, Particulate vaccines: on the quest for optimal delivery and immune response, *Drug Discov. Today* 16 (2011) 569–582.

- [3] B. Lou, A. De Beuckelaer, G.R. Dakwar, K. Remaut, J. Grooten, K. Braeckmans, B.G. De Geest, E. Mastrobattista, S. De Koker, W.E. Hennink, Post-PEGylated and crosslinked polymeric ssRNA nanocomplexes as adjuvants targeting lymph nodes with increased cytolytic T cell inducing properties, *J. Control. Release* 284 (2018) 73–83.
- [4] R. Heidenreich, E. Jasny, A. Kowalczyk, J. Lutz, J. Probst, P. Baumhof, B. Scheel, S. Voss, K.J. Kallen, M. Fotin-Mleczek, A novel RNA-based adjuvant combines strong immunostimulatory capacities with a favorable safety profile, *Int. J. Cancer* 137 (2015) 372–384.
- [5] B. Lou, A. De Beuckelaer, E. Boonstra, D. Li, B.G. De Geest, S. De Koker, E. Mastrobattista, W.E. Hennink, Modular core-shell polymeric nanoparticles mimicking viral structures for vaccination, *J. Control. Release* 293 (2019) 48–62.

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