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The preparation of recombinant chicken interleukin-7 and its antiviral activity against chicken IBDV infection

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Interleukin-7 (IL-7), produced mainly by thymic stromal cells, is an important cytokine playing a critical role in host immune system, especially in B cell growth and pre-B cell survival, differentiation and proliferation. IL-7 also stimulates pre-bone marrow cell differentiation into dendritic cells (DCs) and promotes DC maturation. Meanwhile, IL-7 triggers CD4⁺/CD8⁺T cell proliferation and maintains normal immune function. As IL-7 possesses the character of promoting the immune function it was extensively used as biological adjuvant to enhance the antigenicity of many vaccines including DNA vaccines. IL-7 has also been used to stimulate CD4⁺/CD8⁺T cell proliferation for the cancer patients after chemotherapy and the AIDS patients. Chicken infectious bursal disease (IBD), caused by IBD virus (IBDV), is an immunosuppressive viral disease, causing huge economic loss in poultry industry. Since IBDV attacked chicken bursal, which is a central immune organ for chicken B cell development and maturation, the B cells and antibodies against IBDV in the peripheral blood of IBDV-infected chicken were severely depleted during the late period of infection, leading to B cell-associated humoral immune suppression. As mentioned above, IL-7 has the ability to stimulate B cell differentiation and proliferation and restore humoral immune activity, whether IL-7 can revive the suppressed immune system of the IBDV-infected chicken, especially B cell-associated humoral immune system is still unknown. In this study, therefore, we prepared recombinant chicken IL-7 and investigated its therapeutic effects on IBD prevention and treatment. First, we amplify IL-7 cDNA by RT-PCR from chicken spleen, constructed His-tagged IL-7 eukaryotic expression vector using pcDNA3.1A plasmid. The recombinant chicken IL-7 was expressed in HEK293T cells, isolated by Ni-NTA Agarose affinity chromatography and identified for its biological activity by cell proliferation assay using 2E8 mouse B cell line. For therapeutic evaluation on IL-7 in chicken IBD, 4-week old chickens were treated with the recombinant IL-7 before or after IBDV challenge, the B cell number and IBDV antibody titers in the blood were analyzed by flow cytometry and ELISA, respectively. The histopathologic changes of IBDV-infected chicken tissues were observed as well. The results showed that the amplified chicken IL-7 gene sequence and its expression vector structure were correct based on the sequencing result, the IL-7 could stimulate 2E8 mouse B cell proliferation and maturation, suggesting the prepared IL-7 possesses the biological activity. After purification with Ni-NTA Agarose affinity chromatography, its purity reached more than 95%. The chicken experiment showed that the B cell numbers in the bursal significantly increased in IL-7-treated IBDV-infected chickens compared with control group (IBDV-infected chickens untreated with IL-7). The titers of anti-IBDV antibodies in the IL-7-treated IBDV-infected chicken were also significantly higher than that of IL-7-untreated chicken, indicating that chicken IL-7 can significantly stimulate humoral immune response to IBDV in IBDV-infected chicken, therefore the recombinant chicken IL-7 might have the potential therapeutic value to be further developed for IBD prevention and treatment.

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Antigen conjugated PMIDA coated cobalt oxide nanoparticles highly accelerate anticancer immune response

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The viability of using N-phosphonomethyliminodiacetic acid (PMIDA) modified cobalt oxide nanoparticles (CoO NPs) as an antigen carrier was studied. The whole Dalton's lymphoma (DL) cell lysate conjugated PMIDA-CoO NPs (CPCNs) were well characterized by DLS, TEM and SEM study. The CPCNs successfully activated antigen presenting cells (APCs) which was evident by the increasing levels of serum IFN- γ and TNF- α . Immunization of mice with the CPCNs induced IgG responses against the peptide, and increased the antibody dependent cellular cytotoxicity (ADCC). CPCNs also enhance the anti tumor CD4⁺T cell response in mouse. This result demonstrates that CPCNs are able to serve as antigen carriers to induce humoral immune responses against tumor antigens.

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