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Invited review

# Vaccination strategies in lymphoproliferative disorders: Failures and successes

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ABSTRACT

Anti-tumor vaccines in lymphoproliferative disorders hold out the prospect of effective tumor therapies with minimal side effects.

The addition of immunotherapy to old and new chemotherapy regimens has improved both response rates and disease-free survival, leading in many cases to an extended overall survival.

Ideally, an antigen that is used for vaccination would be specifically expressed in the tumor; it must have an important, causal part in the multifactorial process that leads to cancer, and it must be expressed stably even after it is attacked by the immune system.

Immunotherapies, which aim to activate the immune system to kill cancer cells, include strategies to increase the frequency or potency of antitumor T cells, to overcome suppressive factors in the tumor microenvironment, and to reduce T-cell suppression systemically.

In this review, we focus on the results of clinical trials of vaccination in lymphoma, and discuss potential strategies to enhance the efficacy of immunotherapy in the future.



**Contents**



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## 1. Introduction

It is well known that spontaneous tumor regression is not rarely observed in patients with lymphoma. Unfortunately, it is incontrovertible that immune surveillance of tumors in their early stages is an imperfect deficient mechanism against cancer. Any significant anti-cancer immunity is offset by the induction of resistance mechanisms, such as the appearance of a subset of T cells or myeloid cells with suppressor function, or the release of tumor-produced immunosuppressive cytokines [1].

Moreover, generally, lymphomas are weakly immunogenic due to a decreased expression of MHC class I molecules on the surface of malignant cells, which prevent the recognition of tumor-associated antigens (TAAs) by cytotoxic T lymphocytes (CTLs) [2,3].

Nevertheless, anti-tumor vaccines hold out the prospect of effective tumor therapies with minimal side effects. The addition of immunotherapy to old and new chemotherapy regimens has improved both response rates and disease-free survival, leading in many cases to an extended overall survival.

A successful example is the anti-CD20 antibody rituximab acting as passive vaccination against B cell lymphoma. Addition of the anti-CD20 mAb rituximab to conventional and high-dose chemotherapy protocols has drastically ameliorated the prognosis of B-Non Hodgkin Lymphomas (NHLs). In spite of this clinical success, resistance occurs in almost half of the treated patients, resulting in nonresponse to treatment or early relapse [4]. Moreover, rituximab targets CD20 in general, thus depleting not only B cell lymphoma cells but also normal B cells.

It is envisioned that a personalized active vaccination strategy targeting tumor-specific antigens may evoke an even better and more sustained therapeutic response. Hence, different NHL biotargets and more potent mAbs are continuously being sought.

In this review, we focus on the results of clinical trials of vaccination in lymphoma, and discuss potential strategies to enhance the efficacy of immunotherapy in the future.

### 1.1. Literature search

The research was performed on PubMed, covering the period starting from 1976 through 2014. Research in PubMed was performed using medical subjects headings (MeSH®) to report the most common methodologies employed to study vaccination in lymphoproliferative disorders. The keywords used to search were based on the following logical linguistic pattern: ("Lymphoma" [Mesh]) OR ("Lymphoproliferative disease" [Mesh]) OR ("Cancer" [Mesh]) AND ("Vaccination" [Mesh]).

### 1.2. Inclusion and exclusion criteria. Study selection

The research was limited to clinical cross-sectional studies, case-control studies, and genetic association studies published in peer-reviewed journals.

The research was conducted independently by two authors, who evaluated whether the information of each reference was relevant or not. Each disagreement between the two reviewers was resolved by discussion until a consensus was reached. If the abstract did not include enough information to evaluate inclusion or exclusion, the full text of publication was reviewed if available. Otherwise, the paper was excluded.

### 1.3. Immunoglobulins and antigenic determinants

Immunoglobulins (Ig) are glycoproteins formed by two identical heavy and two identical light polypeptide chains. The *N*-terminal ends of each pair of heavy-light chains consist of two variable (V) Ig domains ($V_L$ and $V_H$) that form a unique surface for antigen binding. V regions are generated during B cell ontogeny by the so-called VDJ rearrangement of the germ-line Ig genes. This genetic rearrangement allows for the tremendous initial diversity of human Igs in naïve B cells, a critical feature of the immune system, which is further increased and reshaped by somatic hyper-mutation of V regions in antigen-stimulated mature B cells.

The association of the two V domains generates the idiotype (Id), a distinctive structure and a unique collection of antigenic determinants called idiotopes [5–8].

A number of different methods are presently employed to reproduce the clonal, patient- and tumor-specific idiotype in the laboratory: large scale culture of hybridomas, recombinant technology and DNA vaccines (Table 1).

**Table 1**
Active vaccination strategies.

| | |
|---|---|
| Idiotype vaccination | Hybridomas |
| | Recombinant technology |
| | DNA vaccines |
| Vaccine with tumor antigens | MAGE |
| | NY-ESO 1 |
| | PASD-1 |
| | CD20 |
| | c-MYC |
| | Stress proteins |
| | IGKV3-20 |
| | IGKV3-15 |
| | T-cell leukemia/lymphoma 1 oncoprotein |
| Antigen presenting immune cells | Id-pulsed Dendritic cells |
| | Fusion of DCs with tumor cells |
| | Tumor-derived RNA- pulsed DCs |
| | Fusion of DC with chemokines (Cd40, MIP-1 alpha, RANTES) |
| Enhanced T-cell activation | Anti-CTLA-4mAb |
| | Anti-PD-1 |
| Immunostimulatory adjuvants | Keyhole Lympet Hemocyanin |
| | GM-CSF |
| | alphaCD19-Id |
| | Bacteriophages |
| | Supernatant of necrotic tumor cells |
| | Anti CD40, antiOX40, anti-41BB, anti-CD27, anti-GITR |
| | Syntetic immune primic center |
| | Rhamnogalacturonan II |
| | FR-derived nona peptide |

### 1.4. Hybridomas and DNA vaccines

A major difficulty in production of Id vaccines originates from its patient-specific nature that requires the generation of a custom-made product. However, the manufacturing issues were overcome by advances in hybridoma and recombinant DNA technology. The Id may be used as either protein or DNA in therapeutic vaccines. In the traditional rescue hybridization technique, the Id protein is produced by fusing the lymphoma cells with mouse myeloma cells to generate Id-secreting hybridomas. For recombinant Id protein production, genes encoding the tumor-specific Ig variable regions are cloned by polymerase chain reaction, ligated into an expression vector and transfected into bacterial, plant, insect, or mammalian cells that then produce the Id protein. For Id DNA vaccination, the immunoglobulin heavy and light chains are cloned and inserted into a plasmid vector for naked DNA injection [9,10].

DNA vaccines offer several benefits such as specific targeting, use of multiple genes to augment immunity and reduced