Review

Anti-CD20 Monoclonal Antibody as a New Treatment Modality for B-Cell Lymphoma

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Patients with follicular non-Hodgkin's lymphoma (NHL) generally have an indolent clinical course and respond well to standard chemotherapeutic regimens. However, the response duration is rather short. There has been no therapeutic regimen superior to CHOP chemotherapy for aggressive NHL in past 20 years. Novel therapeutic strategies are necessary for these types of B-cell lymphomas which express CD20 antigen on the surface of tumor cells. Chimeric human/mouse anti-CD20 monoclonal antibody, rituximab, has a powerful therapeutic ac-

tivity with minimal adverse reaction in B-cell lymphoma through complement-dependent cytotoxicity (CDC) or antibody-dependent cell-mediated cytotoxicity (ADCC) and apoptotic mechanisms. The combination of rituximab and CHOP chemotherapy produced a high response rate (95%) in patients with indolent B-cell NHL and prolonged the progression-free and overall survivals in patients with diffuse large-B-cell lymphoma. Confirmation of CD20 expression of tumor cells is essential to predict the response to rituximab in B-cell lymphoma.

Key words: CD20 antigen, Monoclonal antibody, Rituximab, Malignant lymphoma

I. Introduction

The treatment of advanced-stage non-Hodgkin's lymphoma over 50 years was based primarily on chemotherapy and radiotherapy. However, The optimal treatment of follicular lymphoma has not yet been determined. In patients with a low tumor burden, a watchful waiting strategy until disease progression has no negative influence on survival [1, 11, 17]. Chemotherapies containing anthracyclines have not prolonged the survival of patients with follicular lymphoma. On the other hand, the CHOP regimen (cyclophosphamide, doxorubicin, vincristine, prednisolone) has been well established as the standard chemotherapy for aggressive lymphoma according to the results of the randomized trials comparing with 2nd and 3rd generation chemotherapies because of similar efficacy and less toxicity [7]. However, The event-free and overall survivals remain worse in elderly patients and patients having poor prognostic factors accord-

ing to International Prognostic Index (IPI) [15].

In this respect, immunotherapy with monoclonal antibodies might provide new promising strategies to prolong the survival without major toxicity when combined with chemotherapy. Attempts to treat B-cell malignancies with monoclonal antibodies reactive to B-cell antigen idiotypes began more than a decade ago. This type of murine monoclonal antibody therapy was, however, limited by the development of human anti-mouse antibody responses, although significant clinical activity was observed. The genetic engineering to reconstitute antibodies genetically by joining the variable region genes allowed the development of human/mouse chimeric antibody having advantages of reduced immunogenicity and an enhanced ability to interact with human effector cells. Among the various monoclonal antibodies, the chimeric human/mouse anti-CD20 monoclonal antibody, rituximab, has a powerful therapeutic activity with minimal adverse reaction in B-cell lymphomas [9].

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II. Expression of CD20 in B-Cell Lymphoma

The CD20 antigen is a non-glycosylated, 297-amino acid phosphoprotein that penetrates the plasma membrane four times (Fig. 1) [14]. Both the amino and carboxy termini of the protein are located within the cytoplasm, while only a short segment of the protein is located in extracellular parts. The cytoplasmic domains contain both serine and threonine residues but no tyrosine residues. The intracellular domains also contain consensus phosphorylation sequences for serine/threonine kinases. The structure of CD20 may form an ion channel. The function of CD20 is thought to be involved in B-cell activation, the regulation of cell growth, and transmembrane calcium flux. CD20 expression is restricted to the B-cell lineage. It does not modulate or shed in response to stimulation. CD20 is reliably present in almost all B-cell non-Hodgkin's lymphomas. The CD20 antigen is, therefore, an ideal target monoclonal antibody therapy. CD20 antigen is first expressed at the pre-B cell stage of B-cell development. Mature B-cells also express CD20, and B-cell activation results in an additional increase in CD20 expression. As B-cells differentiate and mature toward plasma cells, the CD20 antigen begins to disappear from the surface (Fig. 2) [6]. The CD20 antigen is not expressed in plasma cells.

Flow-cytometry and immuno-histochemical staining of B-cell lymphoma tissues in 150 patients showed that CD20 antigen was expressed in 95% of samples. There was a good correlation in the incidence of expression between CD20 and CD19 ($r=0.89$). Discrepancy in expression of CD20 and CD19 was observed in 6 cases of diffuse large B-cell lymphoma, in which CD20 did not express in spite of strong expression of CD19 (Fig. 3).

Fig. 1. The structure of CD20.

III. Chimeric Anti-CD20 Monoclonal Antibody

The chimeric human/mouse anti-CD20 monoclonal antibody, rituximab is a human IgG1k antibody with mouse variable regions isolated from a murine anti-CD20 antibody. *In vitro* studies demonstrated that rituximab was able to lyse CD20 positive cells through complement-dependent cytotoxicity (CDC) or antibody-dependent cell-mediated cytotoxicity (ADCC) [13] and apoptotic mechanisms [3]. It has also been shown to sensitize drug-resistant lymphoma cell lines to killing by cytotoxic drugs.

IV. Efficacy and Safety of Rituximab

The clinical response seems to correlate with the intensity of CD20 expression rather than the incidence. We experienced a case of follicular lymphoma successfully treated with rituximab as a single agent. A 40 year old male with follicular lymphoma in stage IV, who had been heavily treat-

Fig. 2. CD20 expression during B-cell differentiation.

ed with chemotherapy, was given with rituximab at a dose of 375 mg/m² by intravenous infusion every 1 week for a total of 8 doses. Rituximab was administered at an initial rate of 25 mg/hr to avoid infusion-related reactions. The bulky tumors began to decrease in size after 2 weeks and the patient achieved complete remission (CR) after completion of 8 doses. No relapse has been observed for more than 2 years until now.

The Japanese phase II trial was conducted to determine whether this dose is practically applicable to Japanese patients with relapsed indolent B-cell lymphoma with respect to safety and efficacy [12]. A total of 90 patients were enrolled and treated with rituximab at 375 mg/m² once weekly of 4 doses. Hematological toxicities at grade 3 or more occurred in 18 patients. Non-hematological toxicities were observed more frequent in patients with extranodal disease or bone marrow involvement. The overall response in low-grade NHL was around 60%. The median progression-free survival in low-grade B cell NHL was 245 days. Pharmaco-

Fig. 3. The relationship between CD20 and CD19 expression on the surface of B-cell lymphomas. There is a good correlation between these antigen expressions. Six samples from aggressive lymphoma were CD20 negative and CD19 positive as indicated as right-lower part of the figure.

Before treatment

8 weeks

16 weeks

After 1 year

Fig. 4. CT findings before and after treatment with rituximab in a patient with follicular lymphoma. The patient continues complete response for more than 1 year.

Fig. 5. Event-free survival among 399 patients assigned to CHOP or CHOP plus rituximab in GELA study.

kinetic analysis showed that elimination of half-life of the drug was around 18 days and the serum antibody levels were still measurable at three months.

Czuczman *et al.* [5] first reported the results of a phase II study on the combination of rituximab and CHOP chemotherapy for indolent B-cell lymphoma. The high clinical response (95%) suggests an additive therapeutic benefit of the combination without significant added toxicity [10]. As to aggressive non-Hodgkin's lymphoma, Vose *et al.* [16] reported the results of phase II study of rituximab in combination with CHOP regimen in previously untreated patients. The clinical responses were at least comparable to those achieved with CHOP alone. Recently, Coiffier *et al.* [4] reported the results of randomized phase III study in which CHOP chemotherapy plus rituximab compared with CHOP alone in elderly patients with diffuse large-B-cell lymphoma. They have demonstrated that the addition of rituximab to the CHOP regimen increases the complete-response rate and prolongs event-free survival and overall survival, without a clinically significant increase in toxicity. CHOP plus rituximab therapy seems to be a novel standard therapy for aggressive B-cell lymphoma.

V. Conclusions and Future Direction

In general, low-grade or follicular non-Hodgkin's lymphoma is assumed to have an indolent course when compared with intermediate- and high-grade non-Hodgkin's lymphoma. Although treatment of low-grade lymphomas with chemotherapeutic regimens is characteristically associated with a high initial response rate, the clinical course consists of a pattern of repeated relapse. Subsequent remissions occur at a lower rate with a shorter duration. Patients eventually succumb to the disease or its complications with a median survival of approximately 6~7 years. For these reasons, novel therapeutic agents and strategies have been needed to manage this group of patients. Many clinical trials

for the study of rituximab in combination with a variety of cytotoxic agents are now conducted all over the world. Rituximab has also been reported to be effective for other types of B-cell malignancies, such as mantle cell lymphoma, plasmacytoma, B-cell chronic lymphocytic leukemia [2, 8]. The usefulness of rituximab is also reported in non-malignant diseases such as autoimmune diseases, graft-versus-host disease after allogeneic marrow transplantation. Recent studies have been devoted to other types of antibody-mediated therapy including anti-CD19 and CD22 antibodies coupled with immunotoxins, and anti-CD20 monoclonal antibody radiolabeled with iodohippurate sodium I¹³¹ or yttrium Y⁹⁰.

VI. References

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