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Trastuzumab

Prop INN

Antineoplastic MAb

Herceptin®

Immunoglobulin G1 (human-mouse monoclonal rhuMab HER2 γ1-chain anti-human p185$^{c\text{-}erbB2}$ receptor), disulfide with human-mouse monoclonal rhuMab HER2 light chain, dimer

CAS: 180288-69-1

EN: 198466

Introduction

Protooncogenes encoding growth factors and their receptors are known to play a crucial role in the pathogenesis of several human malignancies. The human epidermal growth factor receptor 2 (HER2; erbB2 or neu) was identified and the HER2 oncogene gene, also known as *neu* or *c-erbB-2*, was found to encode a 185 kDa transmembrane glycoprotein receptor (p185^{HER2}) which is partially homologous to the epidermal growth factor receptor and has endogenous tyrosine kinase activity (1-3). In 1986, researchers discovered that overexpression of the HER2 gene was common in ductal carcinomas such as ovarian and breast cancer and were correlated with survival times and potential for relapse. In addition, results from several studies strongly suggest that HER2 gene may be involved in the pathogenesis and poor prognosis of HER2-overexpressing tumors (4, 5). Studies demonstrated that transfection of HER2 into non-neoplastic cells resulted in malignant transformation and that transgenic mice expressing HER2 develop mammary tumors (6-8).

According to the American Cancer Society, each year 180,000 U.S. woman are diagnosed with breast cancer and approximately 164,000 of these new diagnoses are metastatic breast cancer with overexpression of HER2 in tumors in 25-30% of the cases. Interference with HER2 expression is clearly a strategy of major therapeutic implications for treatment of aggressive cancers associated with overexpression of the HER2 protein (ECD^{HER2}). Antibodies directed at p185^{HER2} have been shown to inhibit tumor growth in transformed cells providing further evidence that this receptor is an excellent target for the treatment of breast cancer (9-13). The murine monoclonal antibody MAb 4D5 is directed against the extracellular domain of p185^{HER2} and potently inhibits growth of cells *in vitro* and in xenograft models of human breast cancer (14-16).

However, due to the immunogenic nature of murine antibodies which limits their use therapeutically, MAb 4D5 was humanized by including the 4D5 complementarity determining regions onto a human IgG1 framework. Characterization of biological activity of the several resulting humanized versions led to development of only one recombinant humanized anti-p185^{HER2}, rhuMAb HER2 (Herceptin®; trastuzumab) intended for use as a therapy for woman with metastatic breast cancer. Trastuzumab is the first product engineered which targets an underlying genetic defect resulting in cancer.

Pharmacological Actions

Trastuzumab is a humanized recombinant monoclonal antibody directed against the extracellular domain of HER2. Trastuzumab has been reported to have antiproliferative and cytostatic effects on HER2-overexpressing breast cancer cells *in vitro* and in mouse xenograft models (17-20). The results of an assay using purified peripheral blood mononuclear cells (PBMCs) and SKBR-3 cells, an overexpressing breast cancer cell line, have shown that the tumor cell target must be opsonized by trastuzumab for *in vivo* antibody-dependent, cell-mediated cytotoxicity (ADCC) (18). In another assay, the activity of trastuzumab was characterized in an ADCC assay using PBMC and SKBR-3 cells. Results showed that trastuzumab mediated ADCC via FcγRIII-leukocytes (19).

An 18-h *in vitro* assay was developed using PBMCs from normal and trastuzumab-treated patients and chromium-labeled SKBR-3 target cells in order to evaluate the cytotoxic potential of trastuzumab through evaluation of ADCC *in vitro*. The assay, which uniquely used 7 effector:target ratios with or without trastuzumab, allows for analysis of multiple samples without the need for overnight storage or cyropreservation of PBMCs.

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Trastuzumab was found to be effective only in HER2-overexpressing cell line with no affect on cells expressing normal levels of HER2 protein (20).

The sequence and interval of drug delivery for combined trastuzumab and doxorubicin therapy for breast cancer was determined in a study in which 14 days following xenograft implants, mice were treated with doxorubicin, trastuzumab or a combination of the two agents administered simultaneously or 1-4 days apart and compared to untreated animals. A differential equation model involving log tumor volume *vs.* time was devised and tumor volumes were measured from days 14-42 after implant. Simultaneous administration of the two agents was determined to be the optimal mode of delivery since synergistic action was not observed with separate administration and treatment with doxorubicin prior to trastuzumab resulted in antagonistic action (21).

Pharmacokinetics

In a pharmacokinetic study, 45 metastatic breast cancer patients with HER2- overexpressing carcinomas were administered a loading dose of 250 mg trastuzumab i.v. followed by 100 mg/week. Out of 41 patients, 90% displayed plasma trastuzumab trough levels above the targeted level of 10 μg/ml. The mean serum half-life dependent on circulating ECD^{HER2} released from tumors was found to be 8.3 ± 5.0 days (22, 23).

Anti-p185HER2 immunoliposomes have been designed combining the targeting and antiproliferative activities of trastuzumab with advanced drug delivery and their pharmacokinetics and pharmacodynamics were reported in several studies. The anti-$p185^{HER2}$ immunoliposomes were constructed with trastuzumab-Fab' fragments conjugated to PEG (0-12 mol%) and small unilamellar long-circulating liposomes and loaded with doxorubicin for specificity of delivery of the chemotherapeutic agent to HER2-overexpressing cell, safely disregarding normal cells. Rapid intracellular uptake of the encapsulated agents occurred when target cells were treated with doxorubicin-loaded anti-$p185^{HER2}$ *in vitro* and circulation of intact constructs in normal rats injected i.v. was prolonged, illustrating the improved pharmacokinetics of this drug delivery system as compared to free doxorubicin treatment. The pharmacokinetics of doxorubicin and Fab' from the doxorubicin-loaded anti-$p185^{HER2}$ immunoliposomes were found to be biexponential with terminal a half-life and mean residence time of > 11 h and 16-26 h, respectively. Tumor localization of doxorubicin was observed when doxorubicin-loaded anti-$p185^{HER2}$ immunoliposomes were systemically administered to $p185^{HER2}$-overexpressing tumor xenograft-bearing nude mice. Moreover, doxorubicin-loaded anti-$p185^{HER2}$ immunoliposome treatment in SCID mice with BT-474 tumor xenografts resulted in significant regression of established tumors and partial and complete responses, in contrast to treatment with maximum tolerated doses of free doxorubicin which yielded minimal inhibition of growth with a higher observed systemic toxicity. Therefore, use of the anti-$p185^{HER2}$ immunoliposome delivery system markedly enhanced the doxorubicin therapeutic index as well as the agent's antiproliferative efficacy (24, 25).

Clinical Studies

In a phase II study, trastuzumab (250 mg i.v. over 90 min followed by 100 mg/week x 10) was administered to 46 patients with stage IV breast cancer and antitumor activity was evaluated. Out of 43 evaluable patients with HER2 antigen shedding of less than 0.5 μg/ml, 5 objective responses, 1 complete response, 4 partial responses and 2 minor responses were observed. Another patient had a > 50% regression in tumor volume with regrowth occurring at 11 weeks. The overall response rate was 11.6%, with responses noted in the liver, mediatinum, lymph nodes and chest wall lesions. Stable disease was observed in 14 patients lasting for a mean duration of 5.1 months. Fever and chills were observed in 5 patients following the initial infusion. Other adverse effects included transient pain at tumor sites, demonstrating that trastuzumab was well tolerated (22, 23) (Box 1).

Trastuzumab therapy (250 mg i.v. over 90 min followed by 100 mg/week x 10) was determined to be well tolerated with minimal side effects in a phase II study involving 44 metastatic breast cancer patients. Partial or complete responses were observed in 5 patients. During and following initial infusions, transient fever and chills were observed in 5 patients. Fever was not observed with subsequent infusions (26).

The safety and efficacy of trastuzumab monotherapy (4 mg/kg followed by 2 mg/kg/week i.v. for 1 year and 6 months) was examined in an open-label study involving 222 patients with HER2-overexpressing metastatic breast cancer. An overall response rate of 21% was observed with complete, partial and minor responses in 4%, 17% and 7% of the patients, respectively. Stable and progressive disease were observed in 30% and 42% of the patients, respectively. Duration of tumor responsiveness to treatment was estimated (Kaplan-Meier) to be 8.4 months with a mean survival of 13 months. Trastuzumab treatment was terminated in 2 patients due to adverse effects and a decrease in cardiac ejection fraction was observed in 9 patients with a prior history of cardiac events (27) (Box 2).

In a phase II open-label multicenter study, metastatic breast cancer patients with immunohistochemically defined 2+ or 3+ HER2-overexpression were administered trastuzumab (250 mg followed by 100 mg/week x 8) and cisplatin (75 mg/m^2 on days 1, 29 and 57) with 54% and 69% of the patients undergoing prior hormone therapy or radiotherapy, respectively. After 8 weeks, responsive patients and those exhibiting stable disease were placed on maintenance therapy. One complete response, 8 partial responses, 9 minor plus stable diseases and 18 progressive diseases were observed out of the 36 evalu-

Box 1: Efficacy and safety of trastuzumab in patients with metastatic breast cancer overexpressing the antigen (22, 23).

Study Design	Open clinical trial
Study Population	Patients with metastatic breast carcinoma overexpressing the 185-kd transmembrane glycoprotein receptor $p185^{HER2}$ (n = 46)
Intervention Groups	Trastuzumab, 250 mg i.v. → 100 mg i.v. 1x/week x 10 weeks
Withdrawals [causes]	3 patients [bacteremia in 1, death in 1, patient's request in 1]
Adverse Effects	Fever on 5 occasions, pain at tumor site in 3, diarrhea in 2, nausea/vomiting in 1
Significance of Results	Response rate, 5/43 (11.6%) patients [complete remission in 1/43 (2.3%); partial remission in 4/43 (9.3%)]
Conclusions	Trastuzumab is effective and well tolerated, which suggests that growth factors can induce regression of cancer

Source: Prous Science CTLine database.

Box 2: Efficacy and safety of trastuzumab in patients with relapsing metastatic breast cancer overexpressing HER2 (27).

Study Design	Open clinical trial
Study Population	Patients with relapsing metastatic breast cancer overexpressing HER2 (n = 222)
Intervention Groups	Trastuzumab, 4 mg/kg → 2 mg/kg/week x 11 months (median)
Withdrawals	2 due to adverse effects. 9 patients never received treatment
Adverse Effects	Ejection fraction decrease in 9 patients
Significance of Results	Complete remission rate, 8/222 (3.6%) patients Partial remission rate, 36/222 (16.2%) patients Response rate, 21% Response duration (median), 8.4 months Survival time (median), 13 months
Conclusions	Trastuzumab is active in HER2-overexpressing metastatic cancer and has a favorable toxicity profile

Source: Prous Science CTLine database.

able patients, suggesting that addition of trastuzumab to therapy improved the overall response rate well above those expected with cisplatin treatment alone. No serious adverse effects were observed with trastuzumab treatment except the side effects associated with cisplatin, including grade 3-4 toxicities in 54% of the patients (27, 28) (Box 3).

Preliminary results from a phase III study examining the cytotoxic potential of trastuzumab and the immune status of treated patients with metastatic breast cancer demonstrated in an in vitro ADCC assay with treated patient PBMCs, an enhancement of cytotoxic activity, suggesting that the antitumor activity of effector cells may also be increased *in vivo* (29).

Further studies have also demonstrated the additive effects of trastuzumab given in conjunction with chemotherapy in patients with HER2-overexpressing metastatic breast cancer. In a randomized phase III trial, 469 patients were administered initial chemotherapy with either doxorubicin-cyclophosphamide (60 mg/m^2 and 600 mg/m^2, respectively) or paclitaxel (175 mg/m^2 x 3 h) every 3 weeks for 6 cycles with half of the patients receiving trastuzumab (4 mg/kg followed by 2 mg/kg/week i.v.). Adverse side effects such as grade 3-4 myocardial dysfunction were observed in 18% of the patients receiving doxorubicin-cyclophosphamide plus trastuzumab as compared to only 3%, 0% and 2% in those treated with doxorubicin-cyclophosphamide alone, paclitaxel alone or paclitaxel plus trastuzumab, respectively. Addition of trastuzumab to chemotherapy did not increase the incidence of side effects. The overall response rate was significantly improved in patients receiving both chemotherapy and trastuzumab (62% *vs.* 36.2% with chemotherapy alone) as well as the time to disease progression (8.6 *vs.* 5.5 months with chemotherapy alone). These results suggest that the combination of paclitaxel and trastuzumab is the better regimen when risk and benefit factors are taken into account (30) (Box 4).

Box 3: Response rate in patients with HER2-overexpressing metastatic breast cancer to trastuzumab plus cisplatin (28).

Study Design	Open clinical trial
Study Population	Patients with HER2-overexpressing metastatic breast cancer (n = 36)
Intervention Groups	Trastuzumab, 250 mg → 100 mg/week x 8 weeks Cisplatin, 75 mg/m^2 on days 1, 29 and 57
Significance of Results	Complete remission rate, 1/36 (2.8%) patients Partial remission rate, 7/36 (19.4%) patients
Conclusions	Trastuzumab plus cisplatin improves response rates in patients with metastatic breast cancer expressing the HER2/neu growth factor receptor

Source: Prous Science CTLine database.

Box 4: Synergism between trastuzumab and chemotherapy in metastatic breast cancer patients (30).

Study Design	Randomized clinical trial
Study Population	Patients with metastatic breast cancer (n = 469)
Intervention Groups	(AC) Doxorubicin, 60 mg/m^2 + cyclophosphamide, 600 mg/m^2 1x/3 weeks x 6 cycles (AC+H) Doxorubicin, 60 mg/m^2 + cyclophosphamide, 600 mg/m^2 1x/3 weeks x 6 cycles + trastuzumab, 4 mg/kg 2 mg/kg i.v. 1x/week (T) Paclitaxel, 175 mg/m^2 1x/3 weeks x 6 cycles (T+H) Paclitaxel, 175 mg/m^2 1x/3 weeks x 6 cycles + trastuzumab, 4 mg/kg → 2 mg/kg i.v. 1x/week
Adverse Effects	AC, 71% AC+H, 68% T, 59% T+H, 70%
Significance of Results	Response rate, AC (42.1%) < AC+H (64.9%) Response rate, T (25%) < T+H (57.3%) Time to progression, AC (6.5 months) < AC+H (9 months) Time to progression, T (4.2 months) < T+H (7.1 months)
Conclusions	Trastuzumab markedly increases the clinical benefit of chemotherapy in metastatic breast cancer patients overexpressing HER2

Source: Prous Science CTLine database.

Genentech's trastuzumab has been available to the U.S. oncology medical community since October 5, 1998. Trastuzumab was approved by the FDA for the treatment of metastatic breast cancer in patients with tumors overexpressing the HER2 protein. Used as first-line therapy, it is given in combination with paclitaxel; as second- or third-line therapy, trastuzumab is used alone. The product is supplied as a lyophilized, sterile powder for i.v. infusion containing 440 mg trastuzumab/vial (31).

The FDA has also approved the immunohistochemical test DAKO's HercepTest for marketing, designed specifically to identify breast cancer patients whose tumors overexpress the HER2 protein and who may therefore benefit from treatment with trastuzumab (32).

Manufacturer

Genentech, Inc. (US); licensed to Roche outside the U.S.

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