European Review for Medical and Pharmacological Sciences 2016; 20: 3465-3468

Treatment of advanced breast cancer with a combination of highly agglutinative staphylococcin and vinorelbine-based chemotherapy

Q.-T. YU[1], Z.-B. MENG[2]

[1]Department of Chemotherapy, and [2]Department of Interventional Therapy; Tumor Hospital Affiliated to Guangxi Medical University, Nanning, China

Abstract. – OBJECTIVE: Breast cancer is the most frequently diagnosed cancer among women in over 75% of countries worldwide accounting for one in four of all cancers in women. Highly agglutinative staphylococcin (HAS), a mixture of *Staphylococcus aureus* culture filtrates, has been used clinically as an immunomodifier in the treatment of a number of tumors for many years. The aim of present study is to evaluate the therapeutic effects and safety of treating advanced breast cancer patients with a combined therapy of HAS and vinorelbine-based chemotherapy compared to patients who receive standard chemotherapy alone.

PATIENTS AND METHODS: A total of 62 patients with advanced breast cancer were divided into 2 study groups. One group received intravenous injections of HAS and vinorelbine-based chemotherapy (n=31) compared to a control group assigned to receive vinorelbine-based standard chemotherapy (n=31).

RESULTS: Patients with advanced breast cancer who received HAS combined therapy showed a significantly higher overall response rate (Complete Response + Partial Response) of 67.7% compared with patients who received systemic chemotherapy alone (51.6%; $p < 0.05$). Overall, the occurrence of adverse effects was not significantly different between study groups. HAS was able to remedy the immunosuppressing effects of standard chemotherapy in these patients.

CONCLUSIONS: Treatment of advanced breast cancer with of HAS – in combination with vinorelbine-based chemotherapy – was generally more effective and just as safe compared to treatment with vinorelbine-based standard chemotherapy alone.

Key Words:
Highly agglutinative staphylococcin, HAS, Chemotherapy, Immunotherapy, Breast cancer.

Introduction

Breast cancer is the most frequently diagnosed cancer among women in 140 of 184 countries worldwide and accounts one in four of all cancers in women[1]. Since 2008, worldwide breast cancer incidence has increased by more than 20 percent with nearly 1.7 million new breast cancer cases diagnosed in 2012. In the US, breast cancer ranks second as a cause of cancer death in women, after lung cancer and is expected to account for 29% of new diagnoses in 2015[2]. According to the World Cancer Research Fund International, breast cancer mortality has increased by 14 percent in recent years.

Despite advances in detection, 5% of women diagnosed with breast cancer have metastasis at the time of the first presentation and an additional ~30% develop advanced or stage 4 breast cancer over time[3]. The stage of breast cancer is one of the most important factors in evaluating treatment options. For patients in whom chemotherapy is recommended, the choice of regimen (i.e. single-agent or a combination) and selection of a specific therapy depend on multiple factors, including the tumor burden (both in tumor volume and the presence of disease-related symptoms), general health status, prior treatments and toxicities, and patient preferences. Vinorelbine – from the vinca alkaloids class of compounds that interferes with microtubule assembly – has a high efficacy with a good tolerability at therapeutically effective doses[4]. Vinorelbine is recommended for the treatment of patients with advanced metastatic breast cancer who have failed standard first-line chemotherapy for metastatic disease, as well as for those who have relapsed within six months of anthracycline-based adjuvant therapy[4].

In recent years, novel immuno-oncology based therapies – which take advantage of the antitumor activity of the immune system – have become a highly promising method of cancer treatment[5,6]. Different immunological approaches are showing promise in development, and pre-clini-

Corresponding Author: Qitao Yu, MD; e-mail: qitao_yu1@163.com

cal and clinical evidence provides the rationale for investigating these newer immunotherapies in advanced cancers. Although breast cancer has been considered non-immunogenic, several recent clinical studies suggest that immunotherapy has the potential to improve clinical outcomes for patients. Emerging results from clinical trials evaluating immunotherapeutic agents, including vaccines and immune checkpoint agents, in breast cancer have shown promise, leading to increased interest in immunotherapy approaches. Furthermore, immunotherapy has fewer side effects and tumors are also less likely to develop resistance to immunotherapy because of the immune system's ability to target multiple cancer antigens simultaneously[7]. Ongoing and future studies will continue to evaluate novel immunotherapeutic strategies to include combination therapy regimens that will define the role of immunotherapy in the management of breast cancer. This new treatment approach, however, is still early in development, and more data from clinical studies are required.

Superantigens (SAgs) are toxins produced by many pathogenic bacteria and viruses that promote massive activation of the immune system through non-specific activation of T cells. Staphylococcal enterotoxins (SEs) are the most potent known SAgs and, therefore, have broad potential applications as immuno-therapeutic agents. Highly agglutinative staphylococcin (HAS) – a mixture of *Staphylococcus aureus* culture filtrates containing SEs – has been used clinically as a supplementary cancer therapeutic agent for almost two decades in China[8]. Treatment of malignant tumors with a combination of bacterial superantigens and other anti-cancer therapies have shown that HAS is capable of attenuating the leukopenia caused by chemotherapy or radiotherapy while strengthening of the immune system. The aim of the current study is to assess the efficacy and safety of treating advanced stage III and IV breast cancer patients with a combination therapy of HAS and vinorelbine-based chemotherapy compared to patients who receive vinorelbine-based standard chemotherapy alone.

Patients and Methods

Patients

In total, 62 women between the ages of 22 and 70 (median age of 43 years) were included in the study from February 1998 to July 2002. Among the patients, 23 did not undergo tumor excision, and presented with ipsilateral axillary lymph node metastases and pleural metastases. Ten patients had pulmonary metastases, 13 – hepatic metastases, and 17 patients presented with post-excision recurrences, chest-wall implantation metastatic carcinomas, and distant metastases. The tumor in 28 patients was diagnosed pathologically as stage III cancer, while the remaining 34 patients presented with the stage IV disease. Before the treatment, the average general condition score (Karnofsky score) was > 60. The patients demonstrated no abnormalities of their liver and kidney functions, and their blood analyses and ECG were normal.

Thirty-one patients were randomly enrolled into a study group to receive the combined therapy with highly agglutinative staphylococcin and vinorelbine-based chemotherapy, while the remaining 31 patients served as control group and were treated only with the vinorelbine-based chemotherapy. Informed consent was obtained from all patients, and the study was conducted in accordance with the Declaration of Helsinki and approved by Ethic Committee of our hospital.

Methods

Treatment

Highly agglutinative staphylococcin was provided by Shenyang Xiehe Biopharmacy Corporation (China). Chemotherapeutic drugs were obtained from.

Patients in control group were treated with epirubicin or adriamycin (50 mg on day 1), vinorelbine (40 mg on days 1 and 8), and cyclophosphamide (0.8 g on days 1 and 8), given by intravenous injections, 5-fluorouracil (4 g) in 1500 ml of normal saline, given via an infusion, and infusions of calcium folinate (200 mg) in 1500 ml of 5% glucose. The drugs were administered via a three-way infusion tube and given by continuous homogeneous drip phleboclysis at a moderate speed using an infusion pump for 48 hours (days 2 and 3), with 21 days being a cycle.

Patients in the combined therapy group were given the above chemotherapy and HAS, given for 10 consecutive days starting 1 week prior to chemotherapy (1000U-2000U). HAS was administered by intravenous injections once daily.

Evaluation of Treatment Effects

Peripheral blood cells were counted. In addition, NK cell activity and numbers of T cell sub-

sets were quantified before and 1 week after chemotherapy.

Evaluation of therapeutic effects was done according to the WHO 1997 evaluation criteria for solid tumors[12]. Therapeutic effects were classified into a complete response, a partial response, no change, and a progressive disease; the sum of complete and partial responses was considered as the overall response rate. The effects were classified, according to WHO criteria into 4 grades: complete response (CR), partial response (PR), no change (NC), or progressive disease (PD)[9]. A sum of CR+PR was calculated and defined the overall response rate (OR). In addition, adverse effects were evaluated, again, according to WHO criteria on the toxicity grading of anti-cancer drugs[10].

Adverse Effects

Adverse effects were registered, and liver and kidney functions were examined. According to the WHO grading criteria on acute and sub-acute toxicity of anti-cancer drugs, adverse effects were ranked as 0 (none), I (mild), II (moderate), III (severe), and IV (life-threatening).

Statistical Analysis

The *t*-test was used to compare quantitative data, while the chi-square test was used to compare frequencies. The *p*-value was considered significant at $p < 0.05$.

Results

The complete response and overall response rates in patients treated with HAS combined therapy were, respectively, 41.9% and 67.8% (Table I). This was significantly better then in the patients of the control group (respectively, 28.9% and 51.6%; $p < 0.05$).

Patients in the HAS combined therapy group showed a significantly ($p < 0.05$) lower decrease of white blood cells and platelets than patients in control group. The occurrence rates of nausea, vomiting, myocardial toxicity, hepatorenal damage, and decrease in hemoglobin were comparable between both patient groups. Overall, however, the prevalence of adverse effects was not significantly different between study groups.

In the control group, NK cell activity decreased significantly ($p < 0.05$) at the end of the first week after the chemotherapy. In contrast, NK cell activity in patients undergoing HAS combined therapy was increased compared with the levels before the chemotherapy ($p < 0.05$). In addition, we evaluated changes in T cell subsets in patients of both groups. After chemotherapy, the decrease in CD3 and CD4 counts was less pronounced in patients on combined therapy (Table II).

Discussion

Breast cancer has the highest incidence of cancers in women and about 250 thousand women die of breast cancer every year[12]. While substantial progress has been made in treating breast cancer with chemotherapy, the clinical efficacy of systemic chemotherapy is still unsatisfactory; thus, newer combined treatments are actively studied[13]. Specifically, adjuvant immune modulating therapies are of high interest, as T and NK cells play an important role in anti-tumor immunity[14]. Therefore, assessment of T cell status is one of the important methods to evaluate immune cells[15]. Here, we used HAS and vinorelbine-based chemotherapy to treat patients with advanced breast cancers. Our results indicate that treatment response was markedly higher combination HAS therapy versus vinorelbine-based chemotherapy alone. Importantly, this was not accompanied by increased adverse effects.

The main active ingredient of the combination therapy – highly agglutinative staphylococcin – is a staphylococcal enterotoxin C that does not exert toxic or other adverse effects, has high LD50 (> 500 mg/kg), can revert the leukopenia caused by chemo- and radiotherapy, inhibit and kill tumor cells, and significantly strengthen immune system. HAS is a new immunotherapeutic drug with direct and indirect antitumor effects[16,17]. Animal testing has shown that HAS inhibits tumor growth in mice. In addition, it also enhances the activity of NK cells and phagocytic function of macrophages, and increases lymphocyte transformation rate. In clinical studies, HAS, when used in combination with chemotherapy or radiotherapy, had more potent curative effect and diminished leukopenia rates compared with chemotherapy or radiotherapy alone[17,18]. As a superantigen, HAS can significantly improve CD3 and CD4 counts and CD4/CD8 ratio in patients with breast cancer, and reduce the levels of soluble Interleukin-2 (IL-2R) receptor[11]. Soluble IL-2R levels are significantly elevated in the blood serum of patients with leukemia, lymphoma, liver, breast and lung cancers[19].

Conclusions

The results from this study demonstrate that treatment of advanced breast cancer with a combination HAS + vinorelbine-based chemotherapy therapy is generally more effective and as safe compared to treatment with vinorelbine-based chemotherapy alone. Also, HAS was able to remedy the immunosuppressing effects of standard chemotherapy in these patients. A well-designed, randomized controlled trial with larger number of patients is necessary to validate our conclusions.

Conflict of Interest

The Authors declare that they have no conflict of interests.

References

1) Ferlay J, Soerjomataram I, Dikshit R, Eser S, Mathers C, Rebelo M, Parkin DM, Forman D, Bray F. Cancer incidence and mortality worldwide: Sources, methods and major patterns in GLOBOCAN 2012. Int J Cancer 2015; 136: E359-E386.
2) DeSantis CE, Fedewa SA, Goding SA, Kramer JL, Smith RA, Jemal A. Breast cancer statistics, 2015: Convergence of incidence rates between black and white women. CA Cancer J Clin 2016; 66: 31-42.
3) Early Breast Cancer Trialists' Collaborative Group (EBCTCG). Effects of chemotherapy and hormonal therapy for early breast cancer on recurrence and 15-year survival: An overview of the randomised trials. Lancet 2005; 365: 1687-1717.
4) Galano G, Caputo M, Tecce MF, Capasso A. Efficacy and tolerability of vinorelbine in the cancer therapy. Curr Drug Saf 2011; 6: 185-193.
5) Dempke WC, Sellmann L, Fenchel K, Edvardsen K. Immunotherapies for NSCLC: Are we cutting the gordian helix? Anticancer Res 2015; 35: 5745-5757.
6) Tian XL, Yan Z, Chen J, Zhao WH, Guo W. Clinical application of highly agglutinative staphylococcin in cancer treatment updates of the literature. Eur Rev Med Pharmacol Sci 2016; 20: 2718-2725.
7) Svoboda M, Navratil J, Slaby O. Immunotherapy for the prevention and treatment of breast cancer. Klin Onkol 2015; 28: 416-425.
8) Gu L, Yue J, Zheng Y, Zheng X, Wang J, Wang Y, Li J, Jiang Y, Jiang H. Evaluation of a recombinant double mutant of staphylococcal enterotoxin B (SEB-H32Q/K173E) with enhanced antitumor activity effects and decreased pyrexia. PLoS One 2013; 8: e55892.
9) Duffaud F, Therasse P. New guidelines to evaluate the response to treatment in solid tumors. Bull Cancer 2000; 87: 881-886.
10) Chanat C, Delbaldo C, Denis J, Bocaccio F, Cojean-Zelek I, Le Guyader N. Dose intensity and toxicity associated with Taxotere formulation: A retrospective study in a population of breast cancer patients treated with docetaxel as an adjuvant or neoadjuvant chemotherapy. Anticancer Drugs 2015; 26: 984-989.
11) Chen T. Academic review on highly agglutinative staphylococcin. Chinese Journal of Clinical Oncology 1998; 25: 236.
12) Kurosumi M. Significance and problems in evaluations of pathological responses to neoadjuvant therapy for breast cancer. Breast Cancer 2006; 13: 254-259.
13) Kasler M. Protocols for comprehensive diagnosis and treatment of breast cancer. Magy Onkol 2000; 44: 11-35.
14) Rivoltini L, Carrabba M, Huber V, Castelli C, Novellino L, Dalerba P, Mortarini R, Arancia G, Anichini A, Fais S, Parmiani G. Immunity to cancer: Attack and escape in T lymphocyte-tumor cell interaction. Immunol Rev 2002; 188: 97-113.
15) Hoshinaga K, Mohanakumar T, Goldman MH, Wolfgang TC, Szentpetery S, Lee HM, Lower RR. Clinical significance of in situ detection of T lymphocyte subsets and monocyte/macrophage lineages in heart allografts. Transplantation 1984; 38: 634-637.
16) Zhu ZZ. Immunological enhancement and antitumor effects of highly agglutinative staphylococcin. Chinese Journal of Oncology 1995; 17: 477.
17) Wen T. Minutes of 1997 highly agglutinative staphylococcin seminar. Chinese Journal of Oncology 1997; 19: 474.
18) Zhang J, Sun S, Chen J. Clinical observation on the treatment of middle and advanced malignant tumors with highly agglutinative staphylococcin and chemotherapy. Cancer Res Prev Treatm 1996; 23: 118-120.
19) Lippitz BE. Cytokine patterns in patients with cancer: A systematic review. Lancet Oncol 2013; 14: e218-e228.