16. Kleinerman RA, Liebermann JV, Li FP. Second cancer following cancer of the male genital system in Connecticut, 1935–82. In: Multiple Primary Cancers in Connecticut and Denmark. *Natl Cancer Inst Monogr* 1985, **68**, 139–147.
17. Hay JH, Duncan W, Kerr GR. Subsequent malignancies in patients irradiated for testicular tumours. *Br J Radiology* 1984, **57**, 397–602.
18. Fosså SD, Langmark F, Aass N, Andersen A, Lothe R, Børresen AL. Second non-germ cell malignancies after radiotherapy of testicular cancer with or without chemotherapy. *Br J Cancer* 1990, **61**, 639–643.
19. Kaldor JM, Day NE, Band P, *et al.* Second malignancies following testicular cancer, ovarian cancer and Hodgkin's disease: an international colaborative study among cancer registries. *Int J Cancer* 1987, **39**, 571–585.
20. Hellbardt A, Rene-Olivier M, Obradovic M, Mermillod B, Paunier J-P. The risk of second cancer (sc) in patients treated for testicular seminoma. *Int J Rad Oncol Biol Phys* 1990, **18**, 1327–1331.
21. Kim JH, Chu FC, Woodard HQ, Melamed MR, Huvos A, Cantin J. Radiation-induced soft-tissue and bone sarcoma. *Radiology* 1978, **129**, 501–508.
22. Glanzmann C, Schultz G, Lütolf UM. Lond-term morbidity of adjuvant infradiaphragmatic irradiation in patients with testicular cancer and implications for the treatment of stage I seminoma. *Radiother Oncol* 1991, **22**, 12–8.
23. Duchesne GM, Horwich A, Dearnaley DP, Nicholls J, *et al.* Orchidectomy alone for stage 1 seminoma of the testis. *Cancer* 1990, **65**, 1115–1118.
24. von der Maase H, Engelholm SA, Rørth M, *et al.* The DATECA study group. Non-seminomatous testicular germ cell tumours in Denmark 1976–80. *Acta Radiol Oncol* 1984, **23**, 255.

Acknowledgements—We are grateful to all the departments of oncology, radiotherapy and pathology because they made it possible to investigate hospital records and review histological specimens. We thank Gitte Grandt Pedersen for typing the manuscript.

Eur J Cancer, Vol. 29A, No. 5, pp. 668–671, 1993.
Printed in Great Britain

0964–1947/93 $6.00 + 0.00

Induction Chemotherapy and Intensification with Autologous Bone Marrow Reinfusion in Patients with Locally Advanced and Disseminated Breast Cancer

Nanno H. Mulder, Paula O.M. Mulder, Dirk Th. Sleijfer, Pax H.B. Willemse, Els van der Ploeg, Wil V. Dolsma and Elisabeth G.E. de Vries

In 56 patients with disseminated or locally advanced breast cancer it was attempted to reach a state of no evidence of disease by a remission induction regime containing prednisone, 5-fluorouracil, methotrexate, doxorubicin and vincristine. If successful, patients received an intensification regimen consisting of cyclophosphamide (7 g/m²) and etoposide (1.5 g/m²) with autologous bone marrow reinfusion. The complete remission rate of the induction regimen was 52% and the partial remission rate 42%. 32 patients received the intensification regimen. Two toxic deaths occurred. The median time to disease progression in the group with disseminated disease was 15 months. After a median observation of 4 years, 11 out of 19 patients with locally advanced breast cancer were free of disease. It is concluded that this approach may lead to prolonged disease-free survival in patients with locally advanced breast cancer, but does not influence the survival in disseminated disease.
Eur J Cancer, Vol. 29A, No. 5, pp. 668–671, 1993.

INTRODUCTION

PATIENTS WITH disseminated breast cancer are not curable by present day chemo-, hormonal or immunotherapy or combinations of treatment modalities. Even in cases of a complete remission, characteristically reached in approximately 20% of patients by chemotherapy, remission duration is limited and all patients relapse [1]. Partial remissions are even shorter and median survival is less than 2 years [2]. Patients with locally advanced disease, inflammatory breast cancer, thoracic wall infiltration or gross axillary involvement have a prognosis comparable to those with systemic disease [3–5].

Breast cancer is, however, sensitive to chemotherapy, and a case can be made for better results of more intensive regimens [6]. Therefore, the use of ablative doses of chemotherapy with bone marrow rescue seems to be a valid option to study. The results of such treatment in patients who have measurable tumours indicate high remission rates but usually short remission duration and survival [7–16].

These results parallel those with acute leukaemia, where cure is uncommonly achieved by ablative treatment in relapse, but the same therapy in remission can cure [17]. Clearly, the optimal

Correspondence to N.H. Mulder.
N.H. Mulder, D. Th. Sleijfer, P.H.B. Willemse and E.G.E. de Vries are at the Department of Medical Oncology; P.O.M. Mulder is at the Department of Intensive Care Medicine; E. van der Ploeg is at the Department of Surgical Oncology; and W.V. Dolsma is at the Department of Radiotherapy, University Hospital Groningen, Oostersingel 59, 9713 EZ Groningen, The Netherlands.
Received 5 Oct. 1992; accepted 25 Nov. 1992.

situation for intensive treatment is when minimal residual cancer is present.

Our treatment strategy for patients with advanced breast cancer, therefore, aimed at the induction of a complete remission by six courses of chemotherapy, consisting of methotrexate, fluorouracil (5-FU), doxorubicin, vincristine and prednisone. If a situation of no evidence of disease was achieved, this treatment was followed by intensive chemotherapy and autologous bone marrow reinfusion.

A substantial number of patients so treated are at risk of relapse at the site of the former bulky primary tumours [4] or metastases and, therefore, these sites were treated with surgery and/or radiotherapy. Finally, we added to the treatment described above 2 years of treatment with tamoxifen. The preliminary results of this approach were reported earlier [18].

PATIENTS AND METHODS

Pre- or perimenopausal women (up till 2 years postmenopausal) with histologically proven breast cancer were eligible for this study. The stages IIIb and IV (1983 AJCC TNM-classification) [19] were included. Patients with disseminated disease who were refractory to hormonal manipulation, or were considered to be non-eligible for such therapy on clinical grounds or because of lack of oestrogen receptor or unknown receptor status were entered in this study.

Staging investigations included radionuclide bone scan, liver ultra-sound, chest X-ray and bilateral iliac crest biopsies.

A WHO performance score of 0, 1 or 2 was required. Serum creatinine levels had to be below 120 μmol/l and bilirubin below 30 μmol/l, in order to permit full dosing of induction chemotherapy.

Informed consent was given by all patients before induction treatment and before intensification. The study was approved by the local medical ethical committee.

Treatment schedule

1.5 g/m^2 of methotrexate (MTX) was rapidly infused when alkalinisation of urine was reached following a 6-h infusion of 1 l of sodium bicarbonate 1.4%. 1 h after MTX, 5-FU was given intravenously also in a dose of 1.5 g/m^2, followed by 2 l of sodium bicarbonate (1.4%) over 4 h. Starting 24 h after MTX, leucovorin was given orally in a dose of 15 mg/m^2, every 6 h for 2 days. Prednisone, 40 mg/m^2 orally, was started 24 h after MTX and given for 14 consecutive days. On day 14 doxorubicin 50 mg/m^2 and vincristine 1.0 mg/m^2 for a maximum dose of 2 mg were given. Courses were repeated after 4 weeks, for a total number of 6.

Dose adjustment was done by postponing treatment until leucocytes were above 3.0 × 10^9/l and thrombocytes above 120 × 10^9/l. Dose reduction, 25% of responsible drugs, was given in case toxicity of grade 3 or 4 (WHO criteria) [19] of vital extramedullary organs in the previous course. After completion of remission induction chemotherapy, patients were restaged.

Patients with marrow invasion after induction chemotherapy were excluded for intensification.

In patients who obtained a complete remission and in those patients who had no evidence of disease after six courses of induction chemotherapy, bone marrow was harvested in a minimal amount of 1 × 10^8 cells/kg body weight. These patients received 7 g/m^2 of cyclophosphamide divided over 3 consecutive days. Mesna was given in a dose of 50% of the cyclophosphamide dose. Etoposide, 1.5 g/m^2, was divided over six equal doses and given at 12-h intervals during the same 3 days.

Bone marrow was reinfused on the 7th day after the start of ablative chemotherapy. Patients were treated in a single room, without isolation procedures.

After complete recovery of bone marrow, tamoxifen was given in a dose of 20 mg/day for a period of 2 years.

In case of voluminous locations of disease at the start of treatment, as is the case in patients with T4 breast cancer who have not undergone surgery, irradiation followed the intensification regimen. In T4 tumours a minimal tumour dose of 60 Gy, during 6–7 weeks, was applied. In case of bulky metastatic disease prior to treatment an appropriate irradiation field to osseous metastases and/or positive lymph nodes was given and here the minimal tumour dose was 45 Gy in 5 weeks.

Those patients who still had evidence of disease after six induction courses were treated with the best treatment option available tailored to the individual need of the patient.

Response criteria

The WHO criteria were used. By these criteria patients with pleural effusion only or bone metastases only were not evaluable for response [20].

Survival was determined from the start of treatment till death, time to disease progression from start of treatment till progression. No evidence of disease required the absence of clinical signs and symptoms of cancer, negative double sided iliac crest biopsy, no evidence of tumour on bone scan or negative biopsy or fine needle aspiration of remaining hot spots. Negative liver echography and chest X ray were required and negative fine needle aspiration or biopsy of T4 tumours.

RESULTS

From 1986 to 1989 56 women have been entered into this study. Their mean age was 40 years, range 27–52. Their survival is shown in Fig. 1.

14 patients had T4 breast cancer, all of them with skin infiltration. 7 of them were classified as having inflammatory breast cancer.

14 patients had N3 lymph node metastases and 28 had metastatic cancer. 1 of those 28 also had a T4 tumour. Major locations of tumour in these patients with metastatic disease were lung in 6 patients, liver in 7, bone in 14, soft tissue 1, lymph nodes in 4 patients. 9 patients were found to have bone marrow involvement on bilateral iliac crest biopsy.

From the 56 patients treated with remission induction, 20 are not evaluable for response, because of initial surgical treatment of stage IIIb tumours (12 patients) or not evaluable metastatic disease, 36 are evaluable for response. The overall response

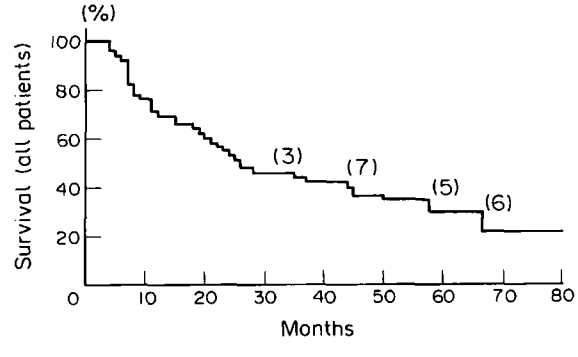

Fig. 1. Survival of all patients entered into the study.

Table 1. Treatment results of induction chemotherapy prior to intensification and ABMT

Stage	Number	Evaluable	CR	PR	PD
N3	14	6	6		
T4	14	10	6	4	
IV	28	20	7	11	2

Complete response (CR) 52%; partial response (PR) 42%; PD, progressive disease.

rate of the remission induction regimen is 94%, the complete response rate is 52%. Results per stage are given in Table 1.

The 9 patients who started with bone marrow involvement had a repeat biopsy after six courses; in 5 patients no tumour invasion could be detected any more. Complete responses of T4 tumours were confirmed histologically.

In 32 patients who had no evidence of disease after induction chemotherapy bone marrow was harvested. These patients received the intensification regimen with reinfusion of autologous marrow. All patients with locally advanced disease and 2 with stage IV received additional irradiation subsequently. Of these 32 patients, 11 had N3 tumours, 8 had T4 tumours and 13 had metastatic disease.

In the 19 patients with locally advanced disease who received the intensification programme, the median observation time is 48 months; 8 patients had relapsed, 4 of 8 with T4 tumours. 11 (58%) patients are disease-free. Relapse occurred after a median 36 months, range 16–53 months. 6 of these 8 patients have died; median survival after relapse was 7 months.

Of the 13 patients with metastatic disease who received the intensification programme, 12 have relapsed, and of these 11 have died. Median disease-free survival was 15 months, median survival 27 months. 1 patient survives with no evidence of disease after 41 months. Median survival after relapse is 8 months. The time to disease progression is shown in Fig. 2.

Of the patients who did not receive intensification, 5 patients with locally advanced disease had no evidence of disease after induction chemotherapy but refused intensification (2 patients) or had concomitant medical or psychological problems (3 patients). 3 of these 5 patients have relapsed.

Of 15 patients with metastatic disease who still had evidence of disease after induction, 12 have died, with a median survival of 15 months (6–54+).

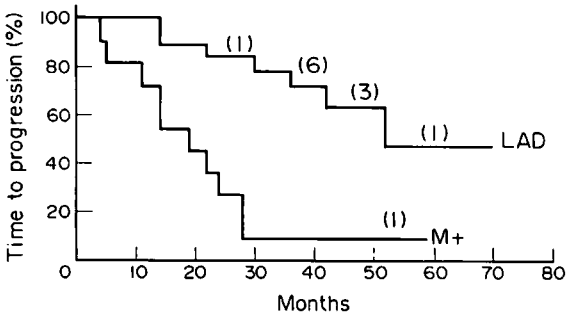

Fig. 2. Time to progression of 19 patients with locally advanced disease (LAD) and of 13 patients with disseminated disease (M+) receiving the intensification programme.

Toxicity

Induction regimen. In 2 patients treatment was stopped during induction therapy: 1 patient discontinued therapy because of subjective toxicity, in the other patient treatment was stopped because of severe cerebellar ataxia, which slowly resolved after stopping chemotherapy. In 4 other patients mild symptoms of cerebellar toxicity developed, in 1 patient this required dose adjustment of 5-FU.

In addition, 1 patient developed symptomatic cytomegalovirus infection, 1 patient had pulmonary embolism, and 2 had peripheral thrombosis. Common toxicities occurring in all patients were conjunctivitis, oral mucositis, grade 2–3, and leukopenia grade 2–3. No clinical symptoms of cardiac toxicity or peripheral neuropathy were evident.

Intensification regimen. All patients experienced a period with lowered numbers of leucocytes or granulocytes. The median duration of the period with leucocytes below 1.0×10^9/l was 16 days (range 9–22) and with thrombocytopenia below 40×10^9/l was 13 days (range 8–18). Thrombocytes were kept above a level of 20×10^9/l by platelet transfusion. One or more episodes of fever occurred in all patients. Median number of days with axillary temperature > 38°C was 9. Blood cultures were positive in 8 patients, 2 patients died from pulmonary infection; in addition, infection was considered life threatening in 2 patients. Moderate to severe nausea and vomiting occurred in all patients on the days of chemotherapy.

The main subjective toxicity was oral mucositis grade 3, in all patients. No bleeding incidents occurred and none of the patients experienced haemorrhagic cystitis or symptoms of cardiac toxicity.

DISCUSSION

The choice of drugs in the remission induction treatment in this study was dictated by the wish to avoid development of resistance to the agents used in intensification. Clearly the regimen leads to a large number of complete remissions, at the cost of considerable but not life-threatening toxicity.

The use of etoposide in the intensification was promoted by our experience in a dose escalation study of etoposide that suggested activity of that agent at high doses, as two clinical and one partial response occurred in breast cancer patients with end-stage disease [21].

The optimal moment of administering ablative intensification therapy is probably in the situation of no evidence of disease, and the best candidates to receive such treatment are patients who have demonstrated sensitivity to chemotherapy. All patients with metastatic disease who went into our intensification programme had shown disappearance of tumour, but some patients with locally advanced disease had prior surgery of all tumour, so tumour sensitivity was not demonstrable in these patients.

The main problem with applying intensive chemotherapy, especially in a situation of no evidence of disease, is the substantial toxicity inherent to such regimens, especially the risk of toxic death [22, 23], as evidenced in 2/32 patients in this study. Kennedy recently accumulated data from four studies aimed at intensification after induction treatment; from the 160 patients in these studies 10% died from treatment [24]. These events are usually due to granulopenic infection, a problem that can now be coped with better using granulocyte growth factors alone or in combination with blood stem cell harvesting and reinfusion.

The results of the regimen described here are, as far as metastatic disease is concerned, conclusive. Clearly, a median

time to relapse of 15 months is insufficient for such intensive treatment. Vincent *et al.* [25] also found no prolonged survival from intensification with one alkylator, high-dose melphalan, in patients responding to conventional induction. However, Williams *et al.* [26], Kennedy *et al.* [24] and Dunphy *et al.* [27] using intensification based on two alkylators also report median time to progression of 10–13 months.

In studies of the type reported here approximately 10% of patients remain disease free for a prolonged period of time. Although 5-year survival was found in between 2 and 18% [28], in a recent overview of conventional chemotherapy prolonged disease-free survival is rare outside an intensification study.

The perspective for the regimen in locally advanced disease is somewhat brighter. After 4 years median observation, 58% of patients remain free of disease (95% confidence level 33–80%). However, it is difficult to attribute this result to any one part of the regimen, as 2 out of 5 patients who were in complete remission but who did not receive intensification also survive disease free for 44+ and 36+ months. The long-term (5-year) disease-free survival of this locally advanced group of patients is reported to be around 30% after treatment with a combination of surgery, chemotherapy and radiotherapy [29–32]. Therefore, proof of superiority of intensification has to await comparative studies.

At present the best intensification regimen for patients with surgically incurable breast cancer has not been determined, it seems unlikely that any regimen so far available will substantially improve the chances of disease-free survival for the majority of patients. A goal for the future will be to determine subgroups of patients that might benefit from intensification, and to limit toxicity further to an acceptable level.

1. Fischer J, Rose CJ, Rubens RD. Duration of complete responses to chemotherapy in advanced breast cancer. *Eur J Cancer Clin Oncol* 1982, **18**, 747.
2. Henderson IC. Chemotherapy for advanced disease. In Bonadonna G, ed. *Breast Cancer, Diagnosis and Management*. New York, Wiley, 1984, 247.
3. DeLena M, Zucali R, Viganotti G, Valagussa P, Bonadonna G. Combined chemotherapy-radiotherapy approach in locally advanced (T3b-T4) breast cancer. *Cancer Chemother Pharmacol* 1978, **1**, 53.
4. Rubens RD, Sexton S, Tong D, Winter PJ, Knight RK, Hayward JL. Combined chemotherapy and radiotherapy for locally advanced breast cancer. *Eur J Cancer Clin Oncol* 1980, **16**, 351.
5. DeLena M, Varini M, Zucali R, *et al.* Multimodal treatment for locally advanced breast cancer. *Cancer Clin Trials* 1981, **4**, 229.
6. Hryniuk W, Bush H. The importance of dose intensity in chemotherapy of metastatic breast cancer. *J Clin Oncol* 1984, **2**, 1281.
7. Douer D, Champlin RE, Ho WG, *et al.* High-dose combined-modality therapy and autologous bone marrow transplantation in resistant cancer. *Am J Med* 1981, **71**, 973.
8. Stewart PS. Autologous bone marrow transplantation in metastatic breast cancer. *Breast Cancer Res Treat* 1982, **2**, 85.
9. Lazarus HM, Herzig RH, Graham-Pole J, *et al.* Intensive melphalan chemotherapy and cryopreserved autologous bone marrow transplantation for the treatment of refractory cancer. *J Clin Oncol* 1983, **1**, 359.
10. Corringham R, Gilmore M, Prentice HG. High-dose melphalan with autologous bone marrow transplant. *Cancer* 1983, **52**, 1783.
11. Tannir N, Spitzer G, Schell F, Legha S, Zander A, Blumenschein G. Phase II study of high-dose amsacrine (AMSA) and autologous bone marrow transplantation in patients with refractory metastatic breast cancer. *Cancer Treat Rep* 1983, **67**, 599.
12. Tannir N, Spitzer G, Dicke K, Schell F, Distefano A, Blumenschein G. Phase I-II study of high dose mitomycin with autologous bone marrow transplantation in refractory metastatic breast cancer. *Cancer Treat Rep* 1984, **68**, 805.
13. Maraninchi D, Abecasis M, Gastaut JA, *et al.* High-dose melphalan with autologous bone marrow rescue for the treatment of advanced adult solid tumors. *Cancer Treat Rep* 1984, **68**, 471.
14. Knight III WA, Page CP, Kuhn JG. High dose L-pam with autologous bone marrow infusion for advanced, steroid hormone receptor negative, breast cancer. *Breast Cancer Res Treat* 1984, **4**, 336.
15. Eder JP, Antman KH, Peters W, *et al.* High-dose combination alkylating agents with bone marrow support as initial treatment for metastatic breast cancer. *J Clin Oncol* 1986, **4**, 1592.
16. Peters WP, Shpall EJ, Jones RB, Bast R, Affronti M, Peters WP. High-dose combination alkylating agent therapy with autologous bone marrow support as initial therapy for metastatic breast cancer. *J Clin Oncol* 1988, **6**, 1368.
17. Thomas ED, Buckner CD, Clift RA. Marrow transplantation for acute non-lymphoblastic leukemia in first remission. *N Engl J Med* 1979, **300**, 597.
18. Mulder NH, Sleijfer DTh, De Vries EGE, Willemse PHB. Intensive induction chemotherapy and intensification with autologous bone marrow reinfusion with stage IIIb and IV breast cancer. *Proc Am Soc Clin Oncol* 1988, **7**, abstract 26.
19. Beahrs OH, Myers MH, eds. *Manual for Staging of Cancer*. American Joint Committee on Cancer, 2nd ed. Philadelphia, Lippincott, 1983, 127.
20. Miller AB, Hoogstraten B, Staquet M, Winkler A. Reporting results of cancer treatment. *Cancer* 1981, **47**, 207.
21. Mulder NH, Meinesz AF, Sleijfer DTh, *et al.* High dose etoposide with or without cyclophosphamide and autologous bone marrow transplantation in solid tumors. In McVie JG, Dalesion O, Smith IE, eds. *Autologous Bone Marrow Transplantation and Solid Tumours*. New York, Raven Press, 1984, 125.
22. Henderson IC, Hayes DF, Gelman R. Dose–response in the treatment of breast cancer: a critical review. *J Clin Oncol* 1988, **6**, 1501.
23. Jones RB, Shpall EJ, Ross M, Bast R, Affronti M, Peters WP. AFM induction chemotherapy, followed by intensive alkylating agent consolidation with autologous bone marrow support (ABMS) for advanced breast cancer. Current results. *Proc Am Soc Clin Oncol* 1990, **9**, abstract 30.
24. Kennedy MJ, Beveridge RA, Rowley SD, Gordon GB, Abeloff MD, Davidson NE. High dose chemotherapy with reinfusion of purged autologous bone marrow following dose intense induction as initial therapy for metastatic breast cancer. *J Natl Cancer Inst* 1991, **83**, 920.
25. Vincent MD, Powles TJ, Coombes RC, McElwain TJ. Late intensification with high dose melphalan and autologous bone marrow support in breast cancer patients responding to conventional chemotherapy. *Cancer Chemother Pharmacol* 1988, **21**, 255.
26. Williams SF, Mick RM, Desser R, Golick J, Beschorner J, Bitzan JD. High dose consolidation with a autologous stem cell rescue in stage IV breast cancer. *J Clin Oncol* 1989, **7**, 1824.
27. Dunphy FR, Spitzer G, Buzdar AU, *et al.* Treatment of estrogen receptor-negative or hormonally refractory breast cancer with double high-dose chemotherapy intensification and bone marrow support. *J Clin Oncol* 1990, **8**, 1207.
28. Falkson G, Tormey DC, Carey P, Witze R, Falkson HL. Long term survival of patients treated with chemotherapy for metastatic breast cancer. *Eur J Cancer* 1991, **27**, 973.
29. Hortobagyi GN, Ames FC, Buzdar AU, *et al.* Management of stage III primary breast cancer with primary chemotherapy, surgery and radiation therapy. *Cancer* 1988, **62**, 2507.
30. Derman DP, Browde S, Kessel IL, *et al.* Adjuvant chemotherapy (CMF) for stage III breast cancer: a randomized trial. *Int J Radiat Oncol Biol Phys* 1989, **17**, 257.
31. Henderson MA, McBride CM. Secondary inflammatory breast cancer: treatment options. *Southern Med J* 1989, **81**, 1512.
32. Fields JN, Kuske RR, Perez CA, Fineberg BB, Bartlett N. Prognostic factors in inflammatory breast cancer–univariant and multivariant analysis. *Cancer* 1989, **63**, 1225.