RAPID PUBLICATION

Further Evaluation of Intensified and Increased Total Dose of Cyclophosphamide for the Treatment of Primary Breast Cancer: Findings From National Surgical Adjuvant Breast and Bowel Project B-25

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Purpose: **In 1989, the National Surgical Adjuvant Breast and Bowel Project initiated the B-22 trial to determine whether intensifying or intensifying and increasing the total dose of cyclophosphamide in a doxorubicin-cyclophosphamide combination would benefit women with primary breast cancer and positive axillary nodes. B-25 was initiated to determine whether further intensifying and increasing the cyclophosphamide dose would yield more favorable results.**

Patients and Methods: **Patients (n = 2,548) were randomly assigned to three groups. The dose and intensity of doxorubicin were similar in all groups. Group 1 received four courses, ie, double the dose and intensity of cyclophosphamide given in the B-22 standard therapy group; group 2 received the same dose of cyclophosphamide as in group 1, administered in two courses (intensified); group 3 received double the dose of cyclophosphamide (intensified and increased) given in group 1. All patients received recombinant human granulocyte colony-stimulating factor. Life-table estimates were used to determine disease-free survival (DFS) and overall survival.**

Results: **No significant difference was observed in DFS (*P* = .20), distant DFS (*P* = .31), or survival (*P* = .76) among the three groups. At 5 years, the DFS in groups 1 and 2 (61% *v* 64%, respectively; *P* = .29) was similar to but slightly lower than that in group 3 (61% *v* 66%, respectively; *P* = 08). Survival in group 1 was concordant with that in groups 2 (78% *v* 77%, respectively; *P* = .71) and 3 (78% *v* 79%, respectively; *P* = .86). Grade 4 toxicity was 20%, 34%, and 49% in groups 1, 2, and 3, respectively. Severe infection and septic episodes increased in group 3. The decrease in the amount and intensity of cyclophosphamide and delays in therapy were greatest in courses 3 and 4 in group 3. The incidence of acute myeloid leukemia increased in all groups.**

Conclusion: **Because intensifying and increasing cyclophosphamide two or four times that given in standard clinical practice did not substantively improve outcome, such therapy should be reserved for the clinical trial setting.**

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DURING THE MID-1980s, findings from studies conducted to evaluate postoperative systemic adjuvant chemotherapy in patients with primary operable breast cancer revealed a modest benefit in disease-free survival (DFS) and overall survival. It was thought, however, that the results would have been better had more drug been administered.[1] That thesis was proposed as a result of (1) a greater remission rate among patients with advanced disease and (2) a retrospective assessment of relapse-free survival in women with early-stage disease, which revealed that higher dose-intensity, ie, amount of drug administered per unit of time, and greater cumulative dose improved patient outcome.[2-6]

In order to test the hypothesis that increased dose and intensity of chemotherapy would result in improved DFS and survival, the National Surgical Adjuvant Breast and Bowel Project (NSABP) formulated, in the late 1980s, a strategy for evaluating the worth of administering, in the adjuvant setting, different intensities and cumulative doses of a single agent in a multiple-agent regimen. It was recognized that in order to appropriately evaluate this hypothesis, it would be necessary for drug to be given at

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increasing levels of dose-intensity and total dose. Consequently, the NSABP initiated two trials, B-22 and B-25, to be conducted sequentially. The aim of these studies was to determine whether higher but fewer doses (dose intensification) of cyclophosphamide in a doxorubicin-cyclophosphamide combination would more effectively prolong DFS and overall survival than would the same cumulative dose of cyclophosphamide given over a more prolonged period of time and whether increasing both the dose-intensity and the cumulative dose of cyclophosphamide, ie, administering a higher dose, would result in greater prolongation of DFS and overall survival than would giving cyclophosphamide at a lower dose and intensity over the same period of time, as is done with standard doxorubicin-cyclophosphamide therapy for breast cancer.

These trials were designed to ensure (1) strict control over treatment delays and dose reductions, (2) enough properly selected patients who would be randomly assigned to the treatment groups that would receive the same therapy but at different dose intensities and in different cumulative doses, (3) a dose-intensity difference that would be sufficient to provide a reasonable chance of detecting differences in treatment failure and survival rates, (4) therapy that demonstrated a beneficial effect when administered at a standard dose and intensity, and (5) acceptable patient morbidity and mortality.

A recent report of findings from the B-22 study through 5 years of follow-up failed to demonstrate a significant improvement in either DFS or overall survival when the same total dose of cyclophosphamide administered in four courses of standard doxorubicin-cyclophosphamide therapy was intensified, ie, given in two courses, or was intensified and increased, ie, doubled in each of four courses. Although these findings failed to support the thesis that intensifying or increasing the total dose of cyclophosphamide would result in a benefit, it was thought that the increased doses of cyclophosphamide administered in B-22 might not be high enough to demonstrate an advantage that might only be evident from increased and/or more intensified therapy. For that reason, the B-25 trial was designed as a sequel to B-22. This report presents the B-25 findings through 5 years of follow-up and relates them to the results obtained from B-22.

PATIENTS AND METHODS

Study Design

Women with primary, operable breast cancer, one or more histologically proven positive axillary nodes, and no evidence of metastatic disease were eligible for the study if they fulfilled specific criteria common to all NSABP trials that evaluated systemic therapy[7,8] and if they gave written consent to participate. The design and conduct of the B-25 trial were, in general, similar to those of B-22.[9] The eligibility criteria and all other aspects of the B-25 study were similar to those of B-22, and for the most part, the same investigators participated in both studies. The B-25 study was designed to evaluate the outcome of therapy administered at levels of intensity and total dose that were greater than those in the B-22 trial. In order to maintain a link with B-22, group 1 (the ''standard'' or control group) of the B-25 trial was identical with the third group in B-22, ie, the group in which the cyclophosphamide was not only intensified but increased (doubled). In the second group of patients in B-25, the intensity of the cyclophosphamide was increased by 100% over that in the control group, while the total dose was left unchanged; in the third group, both the dose-intensity and total dose were increased by 100%. Unlike patients in the B-22 study, to whom recombinant human granulocyte colony-stimulating factor (G-CSF; Amgen, Thousand Oaks, CA) was administered at the discretion of the investigator, women in all three treatment groups of B-25 received G-CSF. Patients were stratified according to number of positive nodes (one to three, four to nine, and ≥ 10), age (≤ 49 years or ≥ 50 years), and type of surgery (lumpectomy or total mastectomy). Randomization was performed within strata, and a biased-coin approach was used to ensure treatment balance within an institution.

Patient accrual in the B-25 study began on April 1, 1992, and was terminated on February 28, 1994. During that time, 2,548 patients were randomized among three treatment groups. The relationship of those three groups to the three groups in the B-22 study is depicted in Table 1. Treatment in all three of the groups in B-25 began within 2 to 5 weeks after surgery. Women enrolled in group 1 received doxorubicin (Adriamycin; Pharmacia-Upjohn, Kalamazoo, MI) 60 mg/m^2 intravenously and cyclophosphamide (Neostar; Pharmacia-Upjohn) 1,200 mg/m^2 intravenously every 21 days for four cycles (courses). Women in group 2 received doxorubicin therapy identical to that in group 1 and two courses of cyclophosphamide, ie, one dose of 2,400 mg/m^2 on day 1 of course 1 and another dose of 2,400 mg/m^2 on day 1 of course 2. In group 2, no cyclophosphamide was given during cycles 3 and 4. The total dose of cyclophosphamide administered to women in group 2 was identical to that in group 1, but the dose of drug was given in two instead of in four courses. In group 3, doxorubicin was administered in the same manner as in groups 1 and 2. However, the increased dose of cyclophosphamide (2,400 mg/m^2) given for two cycles in group 2 was given for four cycles in group 3. All patients who were ≥ 50 years of age received tamoxifen 20 mg/d beginning on day 1 of cycle 1 and continuing for 5 years. The administration of tamoxifen began concurrently with the first cycle of chemotherapy. G-CSF, which was given to all patients to shorten the nadir of the granulocyte counts with the intent of decreasing episodes of infection, was administered at 5 μg/kg subcutaneously beginning on day 2 of each cycle of therapy, starting at least 24 hours after the last dose of chemotherapy, and continuing daily

Table 1. Treatment Groups After Randomization

Protocol	Group* (cyclosphosphamide dose)	No. of Courses†	Dose (mg/m^2/wk)	Total Dose (mg/m^2)
B-22	1. Standard	4	600	2,400
	2. Intensified	2	1,200	2,400
	3. Intensified and increased	4	1,200	4,800
B-25‡	1. Standard or control (as group 3 of B-22)	4	1,200	4,800
	2. Intensified	2	2,400	4,800
	3. Intensified and increased	4	2,400	9,600

*Each group also received four courses of doxorubicin 60 mg/m^2 every 21 days (total dose, 240 mg/m^2).

†Every 21 days.

‡G-CSF was administered to each patient on day 2 of each cycle of therapy.

until a granulocyte count of greater than 10,000 was reached after day 7, at which time it was discontinued for the rest of that cycle. The dose of G-CSF was doubled, and ciprofloxacin was subsequently added for patients who had recurrent septic episodes; these were defined as a temperature of greater than 38.5°C and/or evidence of systemic infection in the presence of lowered granulocyte count requiring hospitalization. The finding of a positive blood culture was not used as the sole criterion for diagnosing sepsis. Specific instructions for the management of patients whose counts failed to return to stated levels were outlined in the protocol.

Study Information

Only three (0.1%) of the 2,548 patients enrolled onto the study had no follow-up evaluation as of December 31, 1998, the cutoff date used for analysis of end points, ie, DFS, distant disease-free survival (DDFS), and overall survival. No data with regard to DDFS appear in the tables or figures in this article. The average time on study for the 2,545 patients with follow-up (the intention-to-treat [ITT] cohort) was 69.2 months. Thirty-four women with follow-up (1%) were declared ineligible: 12 (1%) in group 1, 13 (2%) in group 2, and nine (1%) in group 3 (Table 2). Four patients were ineligible because the time from their initial diagnosis to definitive operation was greater than 28 days; 26 patients were ineligible for a variety of other reasons. Removal of the 34 ineligible patients with follow-up left a total of 2,511 eligible patients with follow-up information. There was either missing information about the number of doses of cyclophosphamide received for another 27 patients or evidence that they received no protocol therapy. Removal of those patients left a cohort of 2,484 patients, ie, the per-protocol cohort. As of December 31, 1998, no follow-up information had been received for 3.9% of the patients for more than 6 months.

Table 2. B-25 Study Information

Patient Status and Reasons for Ineligibility	Group 1	Group 2	Group 3	Total
Randomized, no. of patients	850	848	850	2548
Randomized with follow-up	849	847	849	2545
Ineligible, no. of patients	11	11	9	31
With follow-up	12	13	9	34
Reason ineligible				
Time from diagnosis to operation (> 28 days)	2	0	2	4
Time from operation to randomization (> 35 days)	1	0	1	2
Advanced disease	1	0	0	1
Prior cancer	1	0	1	2
Tumor at line of resection	1	1	1	3
Abnormal hematology or chemistry	1	0	0	1
Cardiac disease	0	1	0	1
Other	5	11	4	20
Eligible with follow-up and treatment information,* no. of patients	837 827	834 824	840 833	2511 2484
Mean time on study,† months				
Patients with follow-up	69.2	69.2	69.2	69.2
Per-protocol cohort	69.2	69.2	69.2	69.2
Mean follow-up time, months				
Patients with follow-up	55.3	55.4	54.9	55.2
Per-protocol cohort	55.4	55.4	54.3	55.2

*Cyclophosphamide information for at least one course of therapy, ie, the "per-protocol" cohort.

†All patients with follow-up.

Table 3. B-25 Patient Entry and Characteristics

Characteristic	Group 1 (n = 850)	Group 2 (n = 848)	Group 3 (n = 850)
Mean time on study,* months	69	69	69
Age			
≤ 49 years	59	59	59
50-59 years	28	26	28
≥ 60 years	13	15	12
Race			
White	86	86	87
Black	7	9	7
Other	6	5	5
Unknown	1	1	1
Clinical tumor size			
≤ 2.0 cm	51	53	57
≥ 2.1 cm	48	46	42
Unknown	1	1	1
Mean ± SD	2.41 ± 1.67	2.33 ± 1.58	2.27 ± 1.73
No. of positive nodes			
1-3	56	56	57
4-9	31	31	31
10+	12	12	11
Unknown	1	1	1
Type of surgery			
Lumpectomy + axillary dissection	36	36	36
Modified radical mastectomy	64	64	64
Estrogen receptor			
0-9 fmol	37	35	36
10-49 fmol	21	24	22
50-99 fmol	10	11	11
100+ fmol	15	15	15
Unknown	18	15	16
Progesterone receptor			
0-9 fmol	37	38	39
10+ fmol	45	48	44
Unknown	18	15	16

NOTE. Values are percent of patients entered on study.

*As of December 31, 1998.

Patient Characteristics

The distribution of patient characteristics was similar among the three treatment groups (Table 3). It is worth noting that 59% of the patients were ≤ 49 years of age, a little over 50% had tumors of ≤ 2.0 cm in size, approximately 55% had one to three positive nodes, and 12% had ≥ 10 positive nodes. Lumpectomy had been performed in 36% of the patients, and between 35% and 39% of tumors were either estrogen receptor (ER)–negative or progesterone receptor (PgR)–negative, ie, had receptor values of 0 to 9 femtomoles/mg cytosol protein (fmol). Tumors from 2,533 patients were analyzed; in 57%, ER and PgR assays were performed by means of conventional cytosol analyses; in 37%, histochemical analyses were carried out; and in 6%, the method of ER determination was not specified. For each method of analysis, the distribution was similar across the three groups.

Toxicity Information

The percentage of patients who experienced selected acute toxicities as of July 7, 1998, is shown for all patients for whom toxicity information was available. Toxicity information was received for 2,534

Table 4. Toxicity Information

	Group 1 (n = 850)*	Group 2 (n = 848)	Group 3 (n = 850)
No. of patients with toxicity information	842	845	847
No. of courses of chemotherapy reported	3,336	3,307	3,235
Average no. of courses per patients with data	3.9	3.9	3.8

*Number of patients on study.

patients, who received a total of 9,878 courses of chemotherapy (Table 4). Patients discontinued protocol therapy for a variety of reasons, which are listed in Table 5 according to age so as to indicate the effect of age on patient withdrawal from the study. In all groups, more patients ≥ 50 years of age either began only one agent or discontinued one or more agents. The frequency with which protocol therapy was discontinued in women aged ≥ 50 years was fairly similar in groups 1 and 2 but considerably greater in group 3. The greatest toxicity experienced per patient is shown in Table 6; ''overall'' grade refers to the percent of patients who experienced maximum toxicity of that grade. Patients who had more than one grade of a particular toxicity were classified according to the greatest toxicity they experienced. Information about patients who experienced less toxicity than the grades selected for presentation is not shown.

No dose reductions of either doxorubicin or cyclophosphamide were permitted in B-25 as a result of toxicity, except when a septic episode occurred. If a patient had one septic episode, the dose of G-CSF was doubled, but full doses of doxorubicin and cyclophosphamide were maintained. If she had a second septic episode, the G-CSF dosage was maintained at double the original dose, doses of doxorubicin-cyclophosphamide were maintained at the full dose, and ciprofloxacin was administered. If a patient experienced a third septic episode despite having received G-CSF and an antibiotic, the initially stipulated dose of doxorubicin-cyclophosphamide was reduced by 25%. When patients experienced hematologic, gastrointestinal, hepatic, or renal toxicities, administration of chemotherapy was delayed until the full dose could be tolerated. The protocol specifically mandated the course of action to be taken in the event that specific side effects, such as hepatic dysfunction, renal and cardiac toxicities, sepsis, or hemorrhagic cystitis, occurred. The protocol stipulated that cyclophosphamide be discontinued in the presence of grade 2 gross hemorrhagic cystitis.

For each group, the ratios of drug doses received to assigned doses were calculated and expressed as percentages. This was performed for each course as well as for the total amount of therapy received across all courses. Findings are presented in Table 7 for the group of patients who had the poorest tolerance for cyclophosphamide. Patients in the 10th percentile demonstrated the least ability to tolerate the dosage as stipulated at randomization. Delays in drug administration are presented as days between courses of therapy for the 90th, 75th, and 50th percentiles of patients. Women in the 90th percentile experienced the longest delays.

Statistical Considerations

The percentage of patients who were disease-free or who survived through 5 years after surgery was estimated by the actuarial life-table method.[10] In the analysis of DFS, an event was defined as follows: the first documented evidence of local, regional, or distant recurrence; recurrence of tumor in the ipsilateral breast after lumpectomy; second primary cancer (other than basal or squamous cell cancer of the skin or in-situ cervical cancer); or death without recurrence of cancer. Death from any cause was the end point for overall survival. All analyses obtained using the 2,545 randomized patients with follow-up are referred to as ITT analyses; those obtained using the 2,484 eligible patients with follow-up and with treatment information are referred to as analyses per protocol.[11] No difference in findings was observed when either of the two cohorts was used for the life-table analyses. The data presented in this article are those that were obtained using the ITT analysis. The adjusted analyses accounted for number of positive nodes, age, and type of surgery.[12,13] Adjusted and unadjusted analyses yielded similar results. Findings using adjusted analyses are presented. The statistical significance of the difference between the life-table distribution by treatment was determined by a summary χ^2 test (log-rank statistic).[14] All *P* values in the figures presented are related to the entire period of observation and were not truncated at 5 years. When 5-year results are specifically referred to, the associated *P* values relate to only 5 years of follow-up. Two-sided *P* values of less than .05 were considered statistically significant. At the alpha = .05 level (unadjusted for multiple comparisons), there was a greater than 80% power to detect a 6% difference in DFS and a 5% difference in overall survival between treatment groups at 5 years. Adjusted curves for DFS and overall survival were computed using the summary relative odds method, with the standard group designated as the reference group for the adjusted curves.[13] Cumulative odds ratios of greater than 1.0 indicate that patients in group 1 had, on average, worse outcomes than patients in the other groups, whereas cumulative odds ratios of less than 1.0 indicate that patients in group 1 had, on average, better outcomes than patients in the other groups. Treatment by covariate interactions[15] were tested via Cox proportional hazards models[16] to determine whether treatment effects on survival and DFS were different according to nodal status

Table 5. Reasons for Discontinuing Protocol Therapy, According to Patient Age

	Group 1		Group 2		Group 3	
Reason	≤ 49 Years (n = 498)*	≥ 50 Years† (n = 344)	≤ 49 Years (n = 499)	≥ 50 Years† (n = 346)	≤ 49 Years (n = 500)	≥ 50 Years† (n = 347)
Refused to begin therapy	2	2	2	3	3	1
Began only one agent or discontinued one or more agents	13	35	6	42	53	91
Toxicity, resulting in						
Patient withdrawal	4	4	2	10	25	24
Physician discontinuing therapy	3	10	3	12	20	31
Other reasons, resulting in						
Patient withdrawal	6	11	0	7	6	19
Physician discontinuing therapy	0	5	1	10	1	9
Unknown	0	5	0	3	1	8
Total	15	37	8	45	56	92

*Based on all patients for whom information was available.

†All patients ≥ 50 years of age received tamoxifen.

Table 6. Greatest Toxicity Per Patient

Toxicity (grade)	Group 1 (n = 842) (%)	Group 2 (n = 845) (%)	Group 3 (n = 847) (%)
Overall			
0	1	1	1
1	9	7	3
2	51	42	31
3	18	16	16
4	20	34	49
Death within 30 days of chemotherapy	< 1 (1)*	< 1 (2)	< 1 (2)
Granulocytes (day 1)			
3 500-999	3	5	4
4 < 500	2	2	2
Infection			
3 Severe	6	10	19
4 Life-threatening	< 1	3	5
5 Death	0	< 1	< 1
Septic episode			
4	15	28	44
Nausea			
3 No intake	11	12	15
Vomiting			
3 6-10 episodes every 24 hours	11	11	14
4 Parenteral support required	4	6	6
Diarrhea			
3 7-9 episodes every 24 hours	1	1	2
4 ≥ 10 episodes every 24 hours (hemorrhagic, support required)	1	< 1	1
Stomatitis			
3 Inability to eat	2	2	2
4 Support required	0	0	1
Hemorrhagic cystitis			
3 Gross (+ clots)	0	0	0
4 Transfusion required	0	0	0
Phlebitis/thromboembolism			
Deep-vein, embolism	0	< 1	< 1
Cardiac (dysrhythmia, functional, ischemia)			
3	< 1	< 1	< 1
4	< 1	< 1	< 1
Leukemia			
Acute myeloid	(4)	(5)	(6)
Myelodysplastic syndrome (MDS)	(0)	(5)	(2)

NOTE. Overall grade 4 toxicities exclude alopecia, irregular menses, hot flashes, fluid retention, vaginal discharge, nadir grades, and weight gain or loss. Septic episodes are classified as grade 4.

*Numbers in parentheses indicate number of patients.

(one to three nodes, four to nine nodes, or ≥10 nodes) and age (< 50 years, ≥ 50 years) at randomization. Subset analyses for age and nodal categories were performed using life-table methodology.

RESULTS

DFS and Overall Survival

Simultaneous comparisons of the three treatment groups using life-table analyses over all follow-up time indicated no significant difference in DFS ($P = .20$), DDFS ($P = .31$), or overall survival ($P = .76$). When the DFS of women in group 1 was compared with that of women in group 2, the outcome was virtually identical (Fig 1). At 5 years, the DFS was 61% in the former and 64% in the latter. For the entire period of observation, the relative odds estimate was 1.10 ($P = .23$; 95% confidence interval [CI], 0.95 to 1.28). At 5 years, the DFS was 61% in group 1 and 66% in group 3; the relative odds estimate over the entire period of observation was 1.15 ($P = .08$; 95% CI, 0.98 to 1.36).

Findings among the three groups relative to survival were concordant. Analyses revealed no significant difference in survival across all groups or across any of the pairwise comparisons. The 5-year survival for women in group 1 was 78%; for those in group 2, 77%; and for those in group 3, 79% (Fig 1). A comparison between group 1 and group 2 demonstrated relative odds of 0.98 ($P = .81$; 95% CI, 0.80 to 1.19) over the entire period of observation. A comparison between group 1 and group 3 showed relative odds of 1.04 ($P = .72$; 95% CI, 0.85 to 1.26).

When patients within a treatment group were evaluated according to their nodal status, there was little evidence, in any group with positive nodes (one to three, four to nine, or ≥ 10) that either intensifying or intensifying and increasing the total dose of cyclophosphamide resulted in a DFS

Table 7. Comparison of Dose and Intensity of Cyclophosphamide Received by Treatment Group and by Course Within Treatment Group Compared With Dose and Intensity Specified at Randomization

Treatment Group	No. of Patients*	Percent of Dose and Intensity at Randomization: 50th (median)†	25th	10th‡
1. Standard	818	99.8	99.0	94.0
Total drug§	827	99.9	99.5	97.9
Course 1	826	99.9	99.2	97.7
Course 2	827	99.8	99.0	97.2
Course 3	818	99.9	98.8	96.2
Course 4				
2. Intensified	816	100.0	99.5	97.1
Total drug§	824	99.9	99.6	98.3
Course 1	819	100.0	99.2	96.8
Course 2		—	—	—
Course 3		—	—	—
Course 4				
3. Intensified and increased total dose	815	99.8	94.5	73.1
Total drug§	830	99.9	99.6	98.5
Course 1	826	100.0	99.3	96.5
Course 2	829	99.9	98.8	76.7
Course 3	826	99.8	93.9	0.0
Course 4				

*Numbers may vary due either to patient withdrawal or to lack of patient information.

†Percentile of patients.

‡Ten percent of the patients received equal to or less than the dose and intensity of the displayed value, whereas 90% received a greater dose and intensity than in the displayed value.

§Total drug is the amount of drug administered across the four courses of chemotherapy.

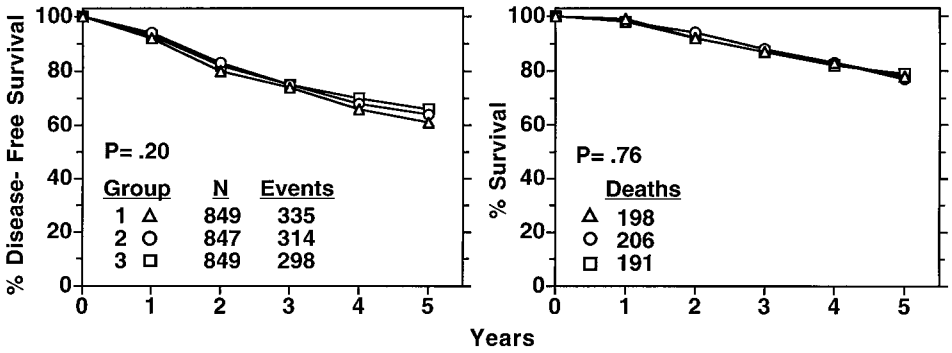
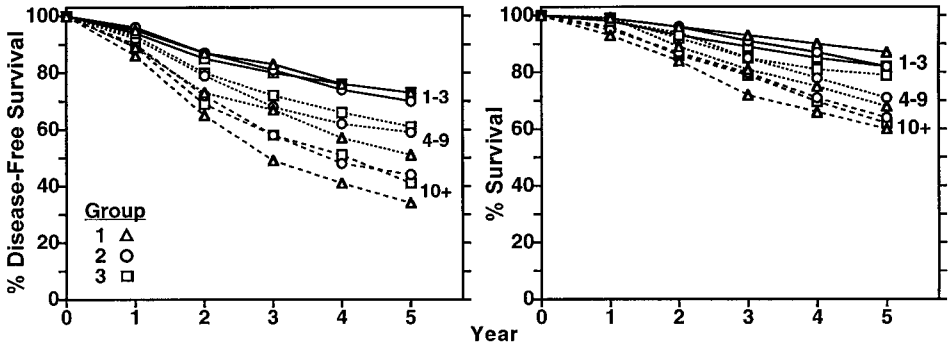


Fig 1. Life-table analysis through 5 years: DFS and overall survival of patients according to treatment group, after adjustment for number of nodes, age, and type of surgery. *P* values relate to tests for heterogeneity across all groups over the entire observation period.

outcome that was superior to that observed in the group that received standard therapy (Fig 2). The DFS at 5 years in patients with one to three positive nodes was 73%, 71%, and 73% in groups 1, 2, and 3, respectively. Neither the test for homogeneity nor the pairwise comparisons between standard cyclophosphamide therapy versus intensified cyclophosphamide or intensified and increased cyclophosphamide therapy were significant ($P = .72$, .45, and .83 for groups 1, 2, and 3, respectively). Although the test for homogeneity was barely significant in patients with four to nine positive nodes ($P = .05$), the comparison between group 1 and group 3 was significantly better in the latter group (51% *v* 61%, respectively; $P = .02$), whereas the comparison between group 1 and group 2 (59%) was not ($P = .15$). In patients with ≥ 10 positive nodes, the 5-year DFS of the three groups was similar, ie, 34%, 44%, and 41% in groups 1, 2, and 3, respectively. Neither the test for heterogeneity nor the two pairwise comparisons were significant ($P = .14$, .05, and .18 for groups 1, 2, and 3, respectively), although the comparison between groups 1 and 2 yielded a nearly significant result in favor of group 2.

When survival was examined according to treatment and nodal status, there was a significant interaction ($P = .04$). Women in group 1 who had one to three positive nodes had a nearly significantly better 5-year survival (87%) than women in group 2 (82%; $P = .08$) and those in group 3 (82%; $P = .06$). A test for heterogeneity among all three groups in patients with one to three positive nodes was not significant ($P = .11$). However, women with four to nine positive nodes in group 3 had a significantly better 5-year survival (79%) than did women with the same number of nodes in group 1 (68%; $P = .01$), and the test for heterogeneity among all groups was significant ($P = .03$) (Fig 2). In patients with ≥ 10 positive nodes, the three groups were not significantly different ($P = .78$) with regard to survival (60% in group 1, 64% in group 2, and 62% in group 3).



	#Nodes	N	Events	P
△		482	134	
○	1-3	481	148	.72
□		485	135	
△		268	131	
○	4-9	267	113	.05
□		266	103	
△		99	70	
○	10+	99	55	.14
□		98	60	

	Deaths	P
△	67	
○	90	.11
□	86	
△	90	
○	77	.03
□	61	
△	41	
○	39	.78
□	44	

Fig 2. Life-table analysis through 5 years: DFS and overall survival of patients according to number of positive nodes and treatment group. *P* values relate to tests for heterogeneity across all groups over entire observation period.

When women in each treatment group were evaluated according to their age and nodal status, the findings relating to their outcome were less consistent than those observed when they were examined relative to treatment and nodal status regardless of age. Whereas no significant qualitative interactions among age, nodal status, and the therapy in group 1 versus the therapy in group 2 were observed for DFS ($P = .27$), DDFS ($P = .13$), and overall survival ($P = .14$), there were highly significant interactions between age, nodal status, and the therapy in group 1 versus that in group 3 DFS ($P = .002$), DDFS ($P = .002$), and overall survival ($P = .0003$). These three-way interactions between group 1 and group 3 were not as significant when age and nodal status were analyzed as continuous variables (DFS, $P = .01$; DDFS, $P = .04$; and survival, $P = .06$). The observed interactions were due mainly to the apparent benefit in DFS and overall survival from intensified and increased cyclophosphamide in women whose age at entry onto the study was less than 50 years and who had four to nine positive nodes (Figs 3 and 4). Similar benefits were not observed for women younger than 50 years of age with one to three or ≥ 10 positive nodes or for those ≥ 50 years old in any treatment group within any nodal group.

When annual event rates were compared among the three groups, either overall or according to patient age (≤ 49 or ≥ 50 years), only among women aged ≤ 49 years was there a statistically significant difference ($P = .02$) in favor of group 3 (Table 8). When annual mortality rates were determined, there was no significant difference among the patients in the three groups when they were evaluated overall or according to age. The annual event rates and mortality rates for women with ER-negative tumors (0 to 9 fmol) or with tumors of any degree of ER positivity were similar among the three treatment groups, with the exception of annual event rates for women with tumors having an ER value of 10 to 49 fmol.

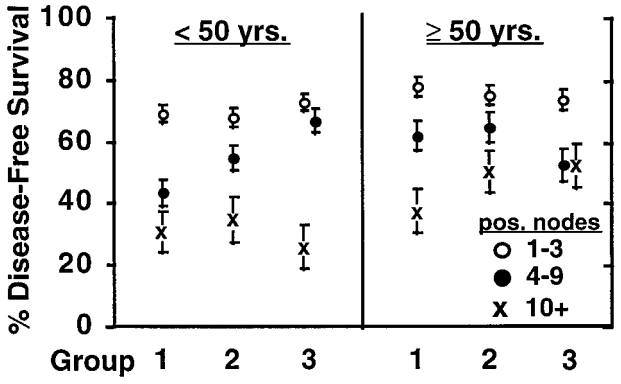


Fig 3. DFS at 5 years according to treatment group, age, and nodal status (Φ = 5-year DFS ± 1 SE).

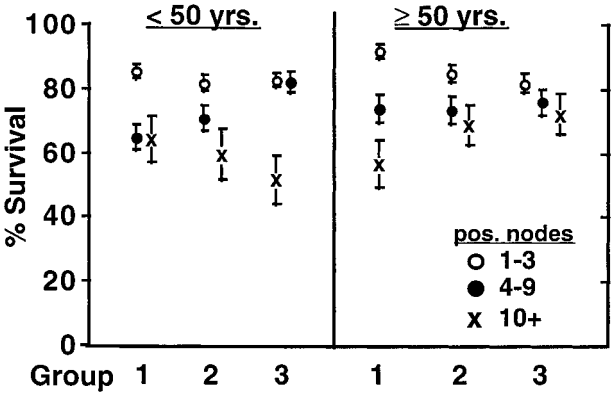


Fig 4. Survival at 5 years according to treatment group, age, and nodal status (Φ = 5-year survival ± 1 SE).

No significant differences in annual event rates or in mortality rates were evident when these rates were determined according to type of surgery.

Sites of Treatment Failure

Examination of the distribution of first sites of treatment failure in the various groups failed to demonstrate that one regimen was more effective than another in reducing the incidence of a first treatment failure at a particular site (Table 9). Of interest is the relatively low overall incidence of local and regional treatment failures observed in all three groups.

Toxicity

Information regarding the severity of toxicity, ie, the greatest toxicity per patient, is presented in Table 6. The findings indicate that the higher the dose of cyclophosphamide per course and the greater the number of courses of the higher dose of drug, the greater the evidence of severe toxicity. Whereas 20% of the women in group 1 demonstrated grade 4 toxicity, this figure increased to 34% in women in group 2 and to 49% in those in group 3 (Table 6). Most of the grade 4 toxicities were septic episodes. A total of 726 patients had at least one septic episode: 124 in group 1, 232 in group 2, and 370 in group 3. There were 76 life-threatening infections—four in group 1, 26 in group 2, and 46 in group 3. Also reported were 126 cases of vomiting that required parenteral support (31 in group 1, 49 in group 2, and 46 in group 3) On the other hand, there was little difference in the incidence of other toxicities in the three groups. A total of 21 cases of either acute myeloid leukemia or other myeloproliferative disorders have been reported (Table 6). Four cases occurred in women randomly assigned to group 1, 10 in women in group 2, and eight in women in

Table 8. Annual Event and Mortality Rates by Age, Tumor ER Status, and Type of Surgery

No. of Patients	Characteristic	Annual Event Rate (%)			Overall *P*	Annual Mortality Rate (%)			Overall *P*
		Group 1	Group 2	Group 3		Group 1	Group 2	Group 3	
	Age*								
1,506	≤ 49 years	11.1	10.0	8.4	.02	5.5	5.4	4.6	.33
1,039	≥ 50 years	8.1	7.9	8.8	.67	4.4	5.1	5.4	.46
	Tumor ER								
910	0-9 fmol	11.2	10.1	11.1	.66	7.1	6.7	7.2	.84
561	10-49 fmol	10.3	11.8	6.8	.004	4.8	5.7	3.7	.15
272	50-99 fmol	10.1	7.2	7.2	.29	3.7	4.5	3.0	.50
385	≥ 100 fmol	8.9	7.0	8.1	.55	2.5	3.6	4.6	.17
417	Unknown	7.5	6.7	7.6	.84	4.3	4.9	3.7	.87
	Surgery*								
917	Lumpectomy + axillary dissection	7.3	7.3	6.4	.61	3.4	4.1	3.5	.88
1,628	Modified radical mastectomy	11.5	10.2	9.9	.24	6.0	6.0	5.8	.92
2,545	All patients	9.9	9.1	8.6	.22	5.1	5.3	4.9	.79

*As reported at time of randomization.

group 3. Five deaths occurred in patients within 30 days after they received chemotherapy; one of these occurred in group 1, two in group 2, and two in group 3.

An increase in the frequency of toxicities greater than or equal to grade 4 was observed across age groups (younger than 50, 50 to 59, or ≥ 60 years) (Table 10). Similar increases were observed across treatment arms for the three age groups. When the frequency of the occurrence of one or more highest levels of a toxicity was evaluated according to the course of therapy, the lowest frequency occurred in group 1. The frequency was similar (approximately 4% to 9%) for each of the four courses of doxorubicin-cyclophosphamide (Table 10). In group 2, the frequency of toxicity was greatest in the first and second courses, ie, when intensified cyclophosphamide was administered. That incidence then decreased in courses three and four where no cyclophosphamide was given. Thus, the incidence of toxicity was lower in both of these courses than in group 1. The incidence of severe toxicities in group 3 was almost identical in the first two courses to that observed in group 2, but that level of toxicity persisted in the third and fourth courses as well.

Table 9. Sites of First Reported Treatment Failure

Site of Recurrence	Group 1 (n = 849)*		Group 2 (n = 847)		Group 3 (n = 849)	
	No. of Patients	%	No. of Patients	%	No. of Patients	%
Local (chest wall and/or scar)	48	5.7	40	4.7	42	4.9
Regional (axillary supraclavicular, parasternal nodes)	33	3.9	31	3.7	34	4.0
Distant	181	21.3	178	21.0	165	10.4
Skeletal	63	7.4	64	7.6	57	6.7
Respiratory	48	5.7	29	3.4	34	4.0
Digestive	31	3.7	39	4.6	32	3.8
Other	39	4.6	46	5.4	42	4.9
Combination of local, regional, distant	10	1.2	7	0.8	3	0.4
Ipsilateral breast†	26	8.5	16	5.2	13	4.2
Contralateral breast‡	18	2.1	14	1.7	12	1.4
Second primary tumors§	15	1.8	19	2.2	24	2.8
Dead, NED	4	0.5	9	1.1	5	0.6
Total first events	335	39.5	314	37.1	298	35.1
Alive, event-free	514	60.5	533	62.9	551	64.9

*Number of patients with follow-up.

†Among lumpectomy patients only: standard therapy, 307 patients; intensified, 305 patients; intensified and increased dose, 306 patients.

‡Includes second cancers.

§Except cancer of the opposite breast.

Relatively few patients in group 1 required blood transfusions, the greatest number of which (9%) were administered after the fourth course of therapy (Table 11). In group 2, the most transfusions were given after course 2 (13%); the numbers after courses 3 and 4 were 7% and 4%, respectively. In contrast, in group 3, nearly one half of the patients required transfusions—41% after course 3 and 45% after course 4.

Compliance With Therapy

Except in the third and fourth courses of group 3, more than 90% of the dose and intensity of cyclophosphamide specified at randomization (Table 7) was administered to patients in the three groups. Only in the 10th percentile of patients was the total amount of drug and dose-intensity found to be appreciably less than the protocol-designated dose (73.1%). Most of the reduction in amount of drug and intensity in the 10th percentile of women was observed in the later courses of therapy, ie, they received 77% and 0% in courses 3 and 4, respectively. Most of the dose reductions were made in response to septic episodes. When compared

Table 10. Percent of Patients With One or More Toxicities ≥ Grade 4* According to Age and Course of Therapy

Treatment Group	Age at Entry			Course of Therapy			
	< 50 Years (n = 1,497)† (%)	50-59 Years (n = 697)† (%)	≥ 60 Years (n = 340)† (%)	1 (n = 2,529)† (%)	2 (n = 2,491)† (%)	3 (n = 2,454)† (%)	4 (n = 2,404)† (%)
1. Standard	16.3	21.4	36.4	8.9	4.2	6.0	7.0
2. Intensified	31.5	37.8	37.1	25.4	14.4	1.3	1.2
3. Intensified and increased total dose	47.2	51.0	54.7	23.8	17.4	19.9	20.2

*Excluding alopecia, irregular menses, hot flashes, fluid retention, vaginal discharge, and weight gain or loss.
†Number of patients for whom toxicity information was available as of July 7, 1998.

with the protocol-stipulated dose and intensity for cyclophosphamide, the dose and intensity of doxorubicin received by treatment group and by course within treatment group were highly concordant with that reported for cyclophosphamide (data not shown), except that the doxorubicin was administered in courses 3 and 4 in group 2 whereas cyclophosphamide was not.

When evaluated according to course of therapy, the rate of delay in administration of therapy was less than 10% among all courses in the three groups, except for the increased incidence of delay in course 4 (16%) in group 3 (Table 12). The duration of the interval between courses of therapy was similar among the three groups. The greatest delays (28 days) occurred between the second and third and the third and fourth courses. As stipulated in the protocol, the median interval between all courses was 21 days.

DISCUSSION

The results of the B-25 trial confirm and augment the findings from the B-22 study, which demonstrated that intensifying and either maintaining or increasing the total dose of cyclophosphamide in a doxorubicin-cyclophosphamide regimen failed to improve patient outcome. The only manifestation of an improved outcome was observed in the women in group 3 of B-25, who received twice the intensity and total dose of the drug as women in group 1 of that study. The small but questionably meaningful increase (5%) in DFS at 5 years of follow-up was of borderline significance ($P = .08$); no benefit in overall survival was evident. Furthermore, when examined overall or according to age or ER status, annual event and mortality rates, for the most part, failed to show a significant advantage from dose manipulation, except in women ≤ 49 years of age, in whom a slight but significant benefit in the annual event rate was observed when both dose-intensity and total dose were doubled.

Another indication of the failure to achieve a meaningful clinical benefit from the administration of an intensified and increased total dose of cyclophosphamide was the observation that, at 5 years of follow-up, the DFS and overall survival of women who received that regimen (66% and 79%, respectively) were highly concordant with those of women in the standard therapy group of B-22, who received the dose and intensity of drug generally administered in clinical practice (62% and 78%, respectively). Those and other cross-protocol comparisons were carried out because, in B-22, the outcome of patients who received the intensified and increased dose of cyclophosphamide was virtually identical to that of women in the standard therapy group of B-25. The DFS at 5 years was 63% and 61%, respectively, and the overall survival was 78% and 78%, respectively (Fig 5). The similarity of the findings among all treatment groups is further demonstrated by the fact that the individual DFS and overall survival outcomes of patients in the six groups comprising B-22 and B-25 were concordant, despite the manipulation of dose-intensity and total dose of therapy (Fig 6). Thus, the findings from the two studies are consistent, in that doubling or quadrupling the intensity and total dose of cyclophosphamide failed to demonstrate that manipulating the drug dosage resulted in a clearly significant clinical benefit. Additional follow-up time is necessary to assess the significance of the slight advantages that have been observed. Characteristics of patients in B-25 were similar to those of patients in B-22, with the exception that patients in

Table 11. Percent of Patients Requiring RBC or Platelet ± RBC Transfusions According to Course of Therapy and Treatment Received

Transfusion	Course 1			Course 2			Course 3			Course 4		
	Standard (n = 836)*	Int (n = 834)	Int. & Inc (n = 840)	Standard (n = 832)	Int (n = 817)	Int & Inc (n = 820)	Standard (n = 831)	Int (n = 809)	Int & Inc (n = 787)	Standard (n = 815)	Int (n = 800)	Int & Inc (n = 736)
RBCs	< 1	1	2	2	4	6	4	6	14	7	3	17
Platelets ± RBCs	0	1	2	< 1	7	8	1	1	25	2	1	26
Total	< 1	2	4	2	13	14	5	7	41	9	4	45

*Number of patients for whom information was available.
Abbreviations: Int, intensified; Int & Inc, intensified and increased dose.

Table 12. Dose Delays and Duration of Interval Between Courses

Treatment Group	Course 1		Course 2		Course 3		Course 4		Days Between Courses of Therapy†								
									Courses 1 and 2			Courses 2 and 3			Courses 3 and 4		
	No. of Patients Delaying Therapy*	%	No. of Patients Delaying Therapy*	%	No. of Patients Delaying Therapy*	%	No. of Patients Delaying Therapy*	%	90th‡	75th	50th	90th	75th	50th	90th	75th	50th
1. Standard	836	< 1	832	3	831	4	815	6	23	21	21	23	21	21	24	21	21
2. Intensified	834	< 1	817	4	809	8	808	7	23	21	21	25	21	21	24	21	21
3. Intensified and increased total dose	840	< 1	820	5	787	9	744	16	24	21	21	28	21	21	28	23	21

*Each patient is included in the total only once per course. Information on delays is based on dates that blood samples were taken.

†The protocol stipulated 21-day intervals between courses.

‡Percentile of patients. Ten percent of the patients experienced a delay equal to or greater than that of the displayed value, and 90% experienced a delay of less than the displayed value.

B-25 were somewhat younger; 59% and 52% of the women were ≤ 50 years of age in B-25 and B-22, respectively, and in B-25, tumors were slightly smaller.

One intriguing aspect of the B-22 study relates to the biologic significance of the findings, which demonstrated that the outcome in any category of node-positive patients did not improve when either the intensity or the total dose of cyclophosphamide was increased. Certainly, in light of the findings from both B-22 and B-25, which show a lack of benefit, those that relate to the effect of treatment on patients within various node-positive categories are not surprising. They do, however, seem antithetical to the view that nodal status correlates with the extent of residual tumor burden after surgery and, consequently, with patient outcome. That thesis contends that less residual tumor should be present in patients with negative nodes than in patients with one to three positive nodes and that the extent of occult tumor becomes progressively greater as the number of positive nodes increases. Consequently, it has been proposed that high-dose chemotherapy would more likely improve the outcome of patients with fewer positive nodes, which should be associated with a smaller residual tumor burden, and that those patients should, thus, demonstrate a better response to treatment.[17-19] However, the findings from B-25, which showed that intensifying and increasing the total dose of cyclophosphamide even further did not improve the outcome of patients with fewer positive nodes, served to confirm the results from B-22, which also failed to support that thesis. The findings from B-25 also demonstrated that the outcome of women with 10 or more positive nodes was little altered by either the degree of intensification or amount of total dose administered.

In both studies, despite the various manipulations of drug dose and intensity, a similarity in DFS was observed in women in each of the three nodal categories evaluated. This phenomenon is illustrated in Fig 7 , in which the outcomes of patients in each of the six treatment groups comprising both trials have been plotted. When survival was taken into consideration, the outcomes of women with one to three or 10 or more positive nodes in the six treatment groups were highly concordant, despite the variable intensity and total dose of drug administered. However, for women with four to nine positive nodes, the findings with regard to DFS and survival were somewhat discordant, in that fewer events and deaths occurred among those who received the greatest intensified and total dose of drug in B-25. Because of a lack of improvement in DFS and in overall survival in women with either one to three or 10 or more positive nodes, it is difficult to assess the clinical significance of the survival findings in the subgroup of patients with four to nine positive

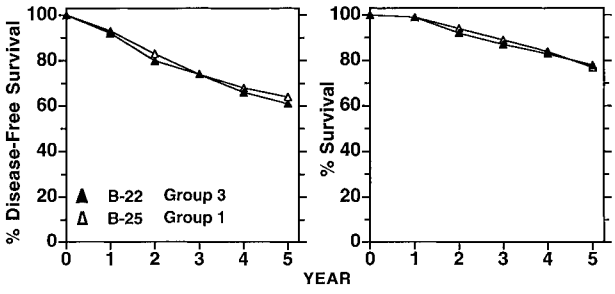


Fig 5. Life-table analysis, through 5 years, comparing DFS and overall survival of B-22 patients in group 3 with that of B-25 patients in group 1. Both groups received the standard dose of doxorubicin and the same total dose and increased intensity of cyclophosphamide.

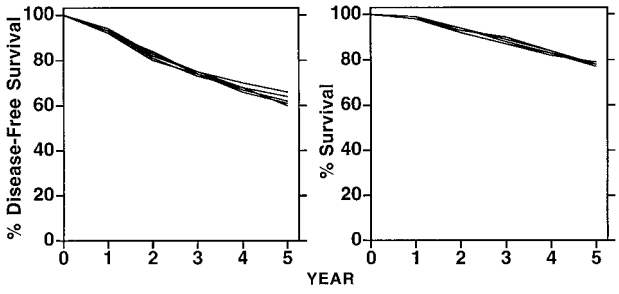


Fig 6. Life-table plots, through 5 years, of DFS and overall survival of patients in each of the groups comprising the B-22 study and each of the groups comprising the B-25 study. (The overlapping lines prevent the use of symbols.)

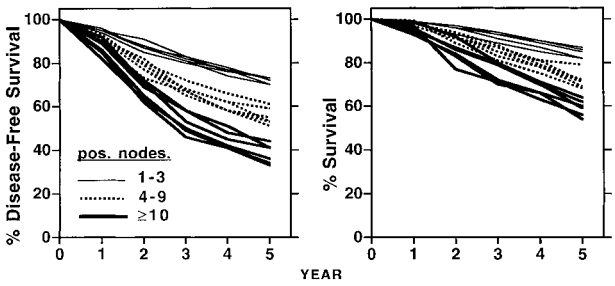


Fig 7. Life-table plots, through 5 years, of DFS and overall survival of patients in each of the groups comprising the B-22 study and each of the groups comprising the B-25 study, according to number of positive nodes.

nodes. To attach meaningful significance to this discrepancy is difficult in that we are unable to evoke a biologic explanation for it. Thus, the findings from both the B-22 and B-25 studies provide little evidence to support the concept that increasing the intensity and total dose of cyclophosphamide in a doxorubicin-cyclophosphamide combination results in a better outcome, at least in part, by more effectively eradicating the smaller number of residual tumor cells putatively associated with fewer positive nodes.

The findings that relate to the incidence of local and regional treatment failure in both the B-22 and B-25 studies are of particular interest for several reasons. First, the distributions of such events among the treatment groups in both studies at similar follow-up times were almost identical, despite the variation in intensity and total dose of cyclophosphamide administered. Thus, it would seem that the standard dose and intensity of doxorubicin and cyclophosphamide generally used in clinical practice yields a degree of local and regional control that is not enhanced by higher doses of the same therapy. Second, in both studies, the incidence of both local and regional treatment failures was surprisingly low. In the B-22 trial, only 6% of first treatment failures occurred at local sites such as the chest wall and/or scar, and 4% occurred at regional nodal sites. In B-25, this incidence was similar (5% and 4%, respectively). These findings are significant in view of a recent revival of interest in the use of postoperative radiation therapy to diminish the frequency of treatment failures at those sites and, ultimately, to improve survival outcome. Two recent studies support that claim.[20,21] In both, the incidence of local-regional treatment failures in women who received chemotherapy but no radiation therapy after surgery was much greater than that observed in our studies. As expected, the high rates of local-regional recurrence were markedly decreased by radiation therapy. Although the follow-up time in both studies was longer than that in our trials, because the majority of local-regional treatment failures are observed within the first 5 or 6 years of follow-up, it is highly unlikely that the lower incidence of such failures observed in our studies will approximate those noted by the other investigators. The basis for the recommendation that radiation therapy be used to reduce local-regional treatment failures is related to the incidence of such recurrences in those studies and to the conclusion that chemotherapy was ineffective in reducing that incidence. Our findings, however, demonstrate not only a lower incidence of such treatment failures but also show that effective chemotherapy can prevent local-regional disease. Consequently, unlike other investigators,[22] we do not subscribe to the universal use of postoperative local-regional radiation therapy to treat node-positive and node-negative patients who receive postoperative chemotherapy.

Although toxicity data from both B-22 and B-25 have been meticulously gathered and documented, the significance of such information might be considered irrelevant in view of the limited benefit achieved from the dose manipulations of cyclophosphamide that gave rise to the toxicities observed. However, knowledge of the extent of the toxicities that occurred might be important for several reasons. First, such information might serve as an indication of whether enough of the protocol-stipulated dose of drug was administered, despite the toxicity encountered, to provide an adequate test of the concepts being evaluated; second, it might be of value in determining whether the severity of the toxicities that occurred was within boundaries that would allow for further escalation of the intensity and total dose, should that be considered worth doing; and third, it might indicate whether the severity of the toxicities observed would preclude the use of therapy that has demonstrated a minimal benefit.

No added benefit was observed from intensifying cyclophosphamide without increasing the total dose of the drug in either the B-22 or B-25 trial. This was not, however, because patients received an amount of drug that was less than that mandated by the protocol, nor was it due to the fact that a greater proportion of women experienced either a delay in receiving therapy or an increase in the number of days between therapy. Thus, it would seem that an appropriate test of cyclophosphamide intensification was carried out in both studies. Consequently, because there is little or no justification for using intensified doxorubicin-cyclophosphamide therapy in the clinical setting, the toxicities observed, while greater than those that occur after the administration of standard therapy, are not of consequence within the context of this report. Some might view the relatively small benefit observed as providing justification for the use of an intensified and increased total dose of cyclophosphamide in clinical practice. However, before such a recommendation can be made, the toxicities that occur as a result of the regimen must be taken into consideration.

Particularly noteworthy in that regard is the observation that patients 50 years of age and older were not as likely to tolerate high-dose chemotherapy; in B-25, 26% of them discontinued protocol therapy whereas only 11% of those

who received standard therapy, as in B-22, did so. Moreover, when examined according to age group, toxicities of grade 4 or higher occurred in 47% to 55% of women who received high-dose chemotherapy, as compared with 6% to 8% of women on standard therapy in B-22. That increase was apparent after each of the four courses of therapy and was approximately 20% versus less than 10% after standard therapy. There was a markedly greater increase in the number of septic episodes after high-dose chemotherapy (44% *v* 3%). Those events were largely responsible for the finding that for women in the lowest 10th percentile of the B-25 study, only 77% of the protocol-required dose of drug was received in course 3 and 0% in course 4. This increase occurred despite the administration of G-CSF in all patients in the B-25 study. Moreover, the fact that almost 50% of women who received high-dose chemotherapy required blood transfusions after courses 3 and 4 further demonstrates the severity of toxicity associated with that regimen. Of major significance was the finding that four deaths occurred because of toxicity in patients who received the increased drug dose, whereas only one death was reported in the group of women on standard therapy. Finally, there is evidence that high-dose chemotherapy may be mutagenic.[23] Six cases of acute myeloid leukemia and two of myelodysplastic syndrome occurred in patients receiving therapy in which the dose was intensified and increased; after standard therapy, three cases of acute myeloid leukemia and one case of myelodysplastic syndrome were reported. In B-25, five cases of each type of blood dyscrasia occurred when the dose was intensified but the total dose was not increased, whereas in B-22, at a similar follow-up time, only one of each was observed when the dose was intensified but not increased. In view of the extent of the toxicities that occurred and the failure to observe a meaningful benefit when the dose and intensity of cyclophosphamide were increased, we have concluded that attempting to improve patient outcome by administering the drug at an increased dose, as used in B-25, is not advisable.

A few comments about how our findings compare with those of others who have evaluated drug intensification and increased total dose of therapy are appropriate. During the last decade, despite the extensive use of high-dose chemotherapy in conjunction with autologous bone marrow or stem-cell rescue, the paucity of data from appropriate randomized clinical trials has prevented definition of the role of such therapy in breast cancer patients who have a large number of positive nodes. Similarly, there is little or no information from trials that have been specifically conducted to evaluate the worth of intensifying and increasing the total dose of the same drug or regimen to a level short of that requiring the need for autologous stem-cell administration. Results that have recently been reported from two randomized phase II trials[24,25] have failed to demonstrate a benefit from the use of ''high-dose'' adjuvant chemotherapy in women with positive nodes. In both studies, patients received several courses of standard ''multimodality treatment'' before being randomized to receive either more of the same therapy at the same dose or a regimen that contained other chemotherapeutic agents as well as autologous stem-cell transplantation. Thus, each of the studies compared two different regimens of therapy, not increased dose(s) of a similar drug or regimen, an aim that was different from that of our studies. The trial most often considered supportive of the hypothesis that increased dose and dose-intensity of a particular drug or combination of drugs could lead to improvement of DFS and overall survival was the one conducted by the Cancer and Leukemia Group B (CALGB), which compared the outcome of patients who received three different dose levels of cyclophosphamide, doxorubicin, and fluorouracil. Recently updated findings from that study[26] show that in no group was the dose intensification and total dose greater than that which might be considered to be the standard dose for the chemotherapy used.[27] Consequently, the major finding from that trial demonstrated that dose reduction resulted in a poorer outcome than that achieved from using doses of a therapy known to be efficacious. One recent study in patients with metastatic breast cancer seems to have some relevance to our findings.[28] Results from that trial demonstrated an increase in response rate and time to disease progression when the dose of epirubicin administered was increased from one that was most likely ineffective (40 mg/m^2) to one that was more effective (90 mg/m^2); no further increase was found, however, by increasing the dose from 90 to 135 mg/m^2. The conclusion from that study may be viewed in part as being similar to the one reached in the CALGB trial, in that when a suboptimal dose of a therapy was increased to a level of clinical effectiveness, a benefit was observed. It demonstrates, however, that unlike what was observed in the CALGB study but similar to what was demonstrated in our trials, no further benefit may occur when an additional increase in drug dose is administered. Thus, to our knowledge, the B-22 and B-25 studies are the only prospective randomized trials that provide information obtained in the adjuvant setting about the consequences of increasing the total dose and intensity of a drug beyond that demonstrated to be clinically effective.

Our observation of an equivocally limited clinical benefit from intensifying and increasing the total dose four-fold beyond that demonstrated to be clinically effective strongly supports the concept that an effective dose of drug exists beyond which no further benefit is likely to be achieved. In that regard, our findings indicate the propriety of continuing to administer cyclophosphamide at a dose of 600 mg/m^2 per course in the doxorubicin-cyclophosphamide regimen used in clinical practice. Our findings also fail to support several concepts that relate to the use of high-dose

chemotherapy and seem to contradict the findings obtained from in vitro studies that demonstrate a steep dose-response curve for all alkylating agents (including cyclophosphamide), which maintain that small increases in drug dose lead to a disproportionate increase in tumor cell kill.[27,29,30] In addition, our findings also fail to provide support for the thesis that higher doses of chemotherapy overcome drug resistance and, thus, confer a benefit.[27,29,30] It remains possible, however, that higher doses of multiple alkylators, possibly with stem-cell support, may overcome drug resistance."[31]

When the B-22 trial was formulated, we realized that the findings, especially if they proved to be negative, would provoke criticism and that our decision to manipulate the dose of only one of the two agents in the regimen, ie, cyclophosphamide and not doxorubicin, was likely to be challenged. The rationale for making those choices has already been presented.[9] Most important, when we designed the study, we recognized that there needed to be sufficient points on the dose-escalation curve to test the hypothesis that increased dose and intensity of a drug is beneficial. It was for that reason that we decided to conduct the B-25 study sequentially, thus ensuring that the number of points on the curve be adequate to test the hypothesis while at the same time ensuring that the eligibility criteria and all other aspects of the study would be similar to those of the B-22 trial. We believe that sufficient levels of dose and intensity have been evaluated in the B-22 and B-25 trials to warrant the conclusion that further increasing the dose of cyclophosphamide above that used in standard therapy without stem-cell support is unlikely to be feasible.

When our findings are considered together with the results reported from studies that have been conducted by other investigators, including those from randomized trials evaluating the worth of high-dose chemotherapy with stem-cell replacement in the adjuvant setting, the case for the use of such therapy seems weak.[32-34] Until evidence is obtained from additional studies testing a variety of new concepts[35,36] for more effectively administering therapeutic agents, we recommend that the use of such therapy be limited to the clinical trial setting.

ACKNOWLEDGMENT

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APPENDIX
Clinical Centers That Contributed 10 or More Patients to NSABP B-25

Name	Principal Investigator	Program Coordinator
Albert Einstein Medical Center, Philadelphia, PA	A.M. Desai	E. Barksdale
Alliant Hospitals, Inc, Louisville, KY	J.T. Hamm	B. MacCracken
Baptist Hospital East, Louisville, KY	M. Grimaldi	J. Sisk
Baptist Medical Centers, Birmingham, AL	T.A. Gaskin III	K.T. Hawkins
Baptist Regional Cancer Institute, Jacksonville, FL	N. Abramson	P. Stokes
Baystate Medical Center, Springfield, MA	W. Reed and G. Makari-Judson	T. Barron
Berkshire Medical Center, Pittsfield, MA	H. Zimbler	G.F. Gero
Billings Interhospital Oncology Project, Billings, MT	N. Hammond	S. Hall
Boston Medical Center, Boston, MA	M.T. Kavanah	D. McDonald
CCOP, Allegheny, Pittsburgh, PA	R. Pugh	
CCOP, Alton Ochsner Medical Foundation, New Orleans, LA	C.G. Kardinal	M. Bateman
CCOP, Atlanta Regional, Atlanta, GA	E.W. Franklin III	P. Remke
CCOP, Brooklyn, NY	S. Rafla-Demetrious	A. Angelone
CCOP, Central Illinois, Decatur, IL	J.L. Wade III	J. Scott
CCOP, Central Los Angeles, Los Angeles, CA	C.A. Presant	S. Soto
CCOP, Christiana Care Health System, Inc, Newark, DE	T. Wozniak	P. Eppes
CCOP, Columbia River Oncology Program, Portland, OR	K.S. Lanier	L. Birenbaum
CCOP, Columbus, OH	L.R. Laufman	M. Bacher
CCOP, Dayton, OH	H.M. Gross	M. Biery
CCOP, Evanston Hospital/Kellogg Cancer Center, Evanston, IL	J.D. Khandekar	B. Marks
CCOP, Kansas City, MO	R.J. Belt	P. Brinkman
CCOP, Marshfield Clinic, Marshfield, WI	J.L. Hoehn	L. Weigel
CCOP, Metro-Minnesota, Minneapolis, MN	S. Rousey	B. Michel
CCOP, Mt Sinai Medical Center, Miami Beach, FL	E. Davila	J.W. Grimes
CCOP, Natalie Warren Bryant at Saint Francis Hospital, Tulsa, OK	K.S. Weibel	S. Segler
CCOP, North Shore, Manhasset, NY	L. Weiselberg	C. Yerys
CCOP, Northwest, Tacoma, WA	H.I. Pierce	K. Hart
CCOP, Sacramento, CA	V. Caggiano	R. Beale
CCOP, Santa Rosa Memorial Hospital, CA	L.W. Keiser	S. Silkworth

APPENDIX
Clinical Centers That Contributed 10 or More Patients to NSABP B-25 (Cont'd)

Name	Principal Investigator	Program Coordinator
CCOP, Scott and White Clinic, Temple, TX	R.R. Young	S. Madsen
CCOP, SE Cancer Control Consortium, Winston-Salem, NC	J.N. Atkins	R.R. Burgess
CCOP, St Louis, MO	A.O. Greco	C. Licavoli
CCOP, Syracuse Hematology and Oncology Association, Syracuse, NY	J.J. Kirshner	P. Barden
CCOP, The Duluth Clinic, Duluth, MN	S.A. Kuross	M. Shene
CCOP, Upstate Carolina, Spartanburg, SC	J.D. Bearden III	P. Williams
CCOP, Virginia Mason, Seattle, WA	P.L. Weiden	B. Edelheit
CCOP, Wichita, KS	S.R. Dakhil	M. Good
Centre Hospitalier de l'Université de Montreal, Quebec, Canada	A. Robidoux	L. Robitaille
CHA-Pavillon Saint-Sacrement, Quebec City, Quebec, Canada	L. Deschenes	E.P. Marcoux
Colorado Cancer Research Program, Denver, CO	R.F. Berris	N.J. Morton
Comprehensive Cancer Research Group, Miami, FL	A.J. Koletsky	C. Byrne
Cross Cancer Institute Edmonton, Alberta, Canada	A.W. Lees	C. Danbrook
Glens Falls Hospital, Glens Falls, NY	R.W. Sponzo	B.A. Sponzo
Good Samaritan Hospital, Cincinnati, OH	R.E. Welling	P.A. Tudor
Greenville Memorial Hospital, Greenville, SC (CGOP)	M.A. O'Rourke	J. Vergnolle
Harrington Cancer Center, Amarillo, TX (CGOP)	B. Pruitt	K. Thomas
Hartford Hospital, Hartford, CT	P.A. DeFusco	J.M. Kulko
Indiana Regional Cancer Center, Indianapolis, IN	S.R. Gunale	P. Hess
Jewish General Hospital, Montreal, Quebec, Canada	F. Patenaude	D. Poulin
John Wayne Cancer Institute, Santa Monica, CA	A.E. Giuliano	M. Brennan
Kaiser Permanente, Northern California Region, CA	L. Fehrenbacher	A. Rodoni
Kaiser Permanente, San Diego, CA	J. Polikoff	S. Cory
Lehigh Valley Hospital, Allentown, PA	H.C. Hoover, Jr	A. Geshan
MBCCOP, VA Commonwealth University, Richmond, VA	H.D. Bear	L. Keener
Memorial Cancer Institute, Long Beach, CA	J. Link	F. Magy
Michael Reese Hospital, Chicago, IL	B.L. Fuller	S.J. Love
Michigan State University, East Lansing, MI	N.V. Dimitrov	C. Robins
Mount Sinai Medical Center, Cleveland, OH	R.S. Bornstein	S. Reynolds
New England Medical Center Hospitals, Inc, Boston, MA	R. Graham	M.J. Scannell
Providence Hospital, Mobile, AL	M. Meshad and T.A. Beeker	P. Shaw
Puget Sound Oncology Consortium, Seattle, WA	R.B. Clarfeld	N.L. Knudsen
Riverside Cancer Institute, Columbus, OH	J.P. Kuebler	K. Stydnicki
Rockford Clinic, Rockford, IL	W.R. Edwards	S.M. Richter
Royal Victoria Hospital, Montreal, Quebec, Canada	H. Shibata	J.P. Aylward
Rush Presbyterian–St Luke's Medical Center, Chicago, IL	J. Wolter	D.B. Madrid
St Barnabas Hospital, Livingston, NJ	R.A. Michaelson	S. Gargiulo
St Luke's Hospital, Bethlehem, PA	A.N. Morrison	
St Mary's Hospital Center, Montreal, Quebec, Canada	P.D. Ahlgren	C. Desrosiers
Sutter Health Western Division Cancer Research Group, Marin County, CA	P.D. Eisenberg	L. Friedman
Thompson Cancer Survival Center, Knoxville, TN	T.J. Panella	J. Bruijn
Tom Baker Cancer Centre, Calgary, Alberta, Canada	A.H.G. Paterson	L. Dobni
Tulane University, New Orleans, LA	D.J. Beech	M. Ramsey
University of California, Davis, CA	S.A. Scudder	J. Houston
University of Cincinnati, OH	D. Hyams	M.A. Schilds
University of Hawaii, Honolulu, HI	R. Oishi	D. Coleman
University of Kentucky, Lexington, KY	E.H. Romond	M. Ashki
University of Medicine/Dentistry, New Brunswick, NJ	I.L. Wapnir	A.G. Owens
University of Michigan, Ann Arbor, MI	L. Baker	B. Golden
University of New Mexico, Albuquerque, NM	A. Mangalik	A. Parsons
University of N. Carolina, Chapel Hill, NC (CGOP)	M. Graham	G.C. Huitt
University of Pittsburgh Cancer Institute, Pittsburgh, PA	V.G. Vogel III	L. Robertson
University of Texas, San Antonio, TX	A.B. Cruz, Jr	I. Presas
Virginia Oncology Association, Newport News, VA	J.J. Schulz	O.M. Jackson
Walt Disney Memorial Cancer Institute, Orlando, FL	R.L. Moroose	J. Crofton

Abbreviations: CCOP, Community Clinical Oncology Program; CGOP, Cooperative Group Outreach Program; MBCCOP, Minority-Based Community Clinical Oncology Program.

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