Amifostine Pretreatment for Protection Against Cyclophosphamide-Induced and Cisplatin-Induced Toxicities: Results of a Randomized Control Trial in Patients With Advanced Ovarian Cancer

By George Kemp, Peter Rose, John Lurain, Michael Berman, Alberto Manetta, Bernard Roullet, Howard Homesley, Dominique Belpomme, and John Glick

**Purpose:** Serious cumulative toxicity is a well-recognized consequence of chemotherapy. Amifostine, an organic thiophosphate, has demonstrated the ability to protect selectively a broad range of normal, but not neoplastic, tissues from the cytotoxic effects of chemotherapy and radiotherapy. This study was designed to determine if amifostine could reduce the serious toxicities associated with cyclophosphamide and cisplatin (CP), without reducing antitumor efficacy in patients with ovarian cancer.

**Patients and Methods:** Two hundred forty-two patients with advanced ovarian cancer were randomized to receive six cycles of cyclophosphamide (1,000 mg/m²) and cisplatin (100 mg/m²) with or without amifostine (910 mg/m²) every 3 weeks for six cycles. The occurrence of hematologic, renal, neurologic, and ototoxicity was evaluated. Antitumor efficacy was assessed by pathologic tumor response and survival.

**Results:** Pretreatment with amifostine before each cycle of chemotherapy resulted in a reduction of cumulative toxicities. Hematologic toxicity consisted of grade 4 neutropenia associated with fever and/or infection that required antibiotic therapy ($P = .005$), days in hospital ($P = .019$), and days on antibiotics ($P = .031$). Platinum-specific toxicities consisted of protracted serum creatinine elevations ($P = .004$), $40\%$ reduction from baseline in creatinine clearance ($P = .001$), and severity of neurologic toxicity ($P = .029$). Twenty-four percent of CP patients compared with $9\%$ of amifostine plus CP patients discontinued therapy because of protocol-specified toxicity ($P = .002$). Pathologic tumor response rates were $37\%$ with amifostine and $28\%$ in controls, with comparable median survival times of 31 months. Amifostine was generally well tolerated; the principal side effects were emesis and a transient decrease in blood pressure.

**Conclusion:** Pretreatment with amifostine reduces the cumulative hematologic, renal, and neurologic toxicities associated with the CP regimen, with no reduction in antitumor efficacy.


The narrow therapeutic index of chemotherapy is well recognized and is attributable to the relative inability of cytotoxic drugs to discriminate effectively between normal and target tissues. As a consequence, a broad range of toxicities are encountered in clinical practice that not only have an impact on patients' quality of life and their ability to accept potentially effective treatment, but may also have serious, life-threatening consequences. The pharmacoeconomic impacts of toxic chemotherapy, with the necessity to hospitalize patients and administer expensive supportive care measures, are of increasing concern in this time of cost containment and health care reform.

Amifostine, formerly known as WR-2721, represents a unique adjunct for the management of cancer patients receiving alkylating agent and cisplatin-based chemotherapy, or radiation therapy. This drug arose from a classified nuclear warfare project sponsored by the United States Army, and was ultimately selected from more than 4,400 chemicals screened because of its superior radioprotective properties and safety profile. With declassification of the project, amifostine was subsequently evaluated for its potential role in reducing the toxicity of therapeutic radiation, as well as drugs that alter the structure and function of DNA, such as alkylating agents and cisplatin. A profile emerged from preclinical studies that demonstrated the ability of amifostine to protect selectively a broad range of normal, but not neoplastic, tissues from the cytotoxic effects of chemotherapy and radiation therapy. Additionally, amifostine has shown the capacity to diminish substantially the potential genotoxic and carcinogenic properties of these same therapeutic modalities.
Phase II clinical trials in patients who received either cyclophosphamide or cisplatin confirmed the profile evolved from preclinical studies.18-21 Amifostine pretreatment was shown to decrease both the degree and duration of neutropenia associated with cyclophosphamide treatment, as well as the treatment-limiting nonhematologic toxicities of cisplatin therapy, namely, nephrotoxicity, neurotoxicity, and ototoxicity. In view of the spectrum of protective activity, it was logical to evaluate the potential use of amifostine with a combination of cisplatin and cyclophosphamide (CP). Advanced ovarian cancer was chosen as the model for evaluation, since the CP regimen was the accepted standard of treatment in 1988 when this study was initiated.

PATIENTS AND METHODS

Study Design

A controlled, multicenter trial in patients with Federation of International Gynecologic Oncologists (FIGO) stage III or IV epithelial ovarian cancer was designed to determine the ability of amifostine to protect against the hematologic, renal, neurologic, and auditory toxicities associated with CP therapy with preservation of antitumor effect. Patients were randomized to receive six cycles of cyclophosphamide 1,000 mg/m² and cisplatin 100 mg/m² with or without pretreatment with amifostine 910 mg/m² administered at 3-week intervals. Patients were stratified by cancer center and extent of residual disease (< 2 cm v > 2 cm). Chemotherapy was to be initiated within 6 weeks of surgery. Following chemotherapy, patients with nonmeasurable disease or those with measurable disease who achieved a clinical complete response were to undergo second-look laparotomy. The protocol-specified procedures to be followed for reassessed laparotomies including washings and biopsies to be taken to assess pathologic tumor response. Patients were monitored for toxicity and survival.

Study Population

Women with biopsy-proven FIGO stage III or IV epithelial ovarian cancer who met the following criteria were eligible: Gynecologic Oncology Group (GOG) performance status of 0, 1, or 2; no prior chemotherapy or radiation therapy; and adequate bone marrow function (WBC count ≥ 3,000 cells/µL, neutrophil count ≥ 2,000 cells/µL, and platelet count ≥ 100,000 cells/µL), renal function (serum creatinine concentration ≤ 1.5 mg/dL), and hepatic function (bilirubin and AST levels ≤ two times the upper limit of normal).

Treatment Plan

All patients received intravenous hydration with 5% dextrose or 0.45% normal saline at a rate of 200 mL/h for 6 hours before treatment. Patients were to receive drug therapy only when urine output was greater than 150 mL/h for 3 hours. Amifostine was reconstituted with 9.5 mL normal saline and the dose of 910 mg/m² was administered as a 15-minute intravenous infusion to patients in a supine position. Within 15 minutes of completion of the amifostine infusion, cyclophosphamide 1,000 mg/m² was administered in 250 mL of 5% dextrose in water or 500 mL normal saline over 20 minutes. Immediately following the infusion of cyclophosphamide, patients in both treatment arms were given mannitol (12.5 g as an intravenous bolus) followed by a 30-minute infusion of cisplatin 100 mg/m². After chemotherapy was completed, patients received a 6-hour infusion of mannitol at a rate of 10 g/h (250 mL/h). Intravenous hydration (> 150 mL/h) was continued until discharge.

Initially, all corticosteroids (eg, dexamethasone, a standard component of antiemetic regimens for intensive cisplatin therapy) were prohibited in this study, as there was concern that they could confound the assessment of the effect of amifostine on neutrophils. However, because initial experience demonstrated intolerable nausea and vomiting for patients in both arms of the trial, the protocol was amended to require that dexamethasone (10 mg intravenously) be given 30 minutes before initiation of therapy and 4, 8, and 12 hours after completion of each cycle of treatment to patients in both treatment groups to ensure that the effects of amifostine would not be confounded by those of dexamethasone. The administration of other antiemetics such as metoclopramide, diphenhydramine, and lorazepam were permitted in this study. Ondansetron was also permitted once it became commercially available.

Following the protocol-specified interim analysis, the results of which met the protocol defined early-stopping rules by demonstrating a reduction in hospitalizations for grade 4 neutropenia associated with fever and/or infection in the amifostine arm, accrual to the protocol was temporarily halted. To provide greater power to assess the effect of amifostine on survival and nonhematologic toxicities, accrual was reopened, but the protocol was amended to allow the use of colony-stimulating factors (CSFs) if grade 4 neutropenia and fever persisted for more than 12 hours. However, the actual use of granulocyte colony-stimulating factor (G-CSF) was minimal (5% of patients overall and < 3% of patients following cycles 2 to 6) and thus did not affect interpretation of the results.

Study Outcome Variables

The primary end point to assess amifostine’s protection against hematologic toxicity was the incidence of grade 4 neutropenia (< 500 neutrophils/µL) associated with fever and/or infection that required antibiotic therapy. The incidence of grade 4 neutropenia, as well as the need to delay or reduce the dose of chemotherapy due to hematologic toxicity, was also a protocol-defined end point for hematologic toxicity. Chemotherapy was to be delayed until the neutrophil count recovered to ≥ 1,500/µL and platelet count recovered to ≥ 100,000/µL. Patients who required a delay in chemotherapy for greater than 2 weeks beyond the next scheduled course of chemotherapy (day 35) were to be withdrawn from study for hematologic toxicity. The doses of cyclophosphamide and cisplatin were reduced for hematologic toxicity based on nadir counts as listed in Table 1.

The ability of amifostine to protect against cisplatin-associated nephrotoxicity was evaluated by the need to delay or discontinue

<table>
<thead>
<tr>
<th>Platelet Nadir/µL</th>
<th>Neutrophil Nadir/µL</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,500</td>
<td>1,499-1,000</td>
</tr>
<tr>
<td>50,000-74,999</td>
<td>75/100</td>
</tr>
<tr>
<td>&lt; 50,000</td>
<td>50/75</td>
</tr>
</tbody>
</table>

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the dose of cisplatin. Cisplatin was to be delayed if serum creatinine levels did not return to ≤ 1.5 mg/dL, and/or creatinine clearance to ≥ 65 mL/min. Patients whose serum creatinine levels remained elevated (> 1.5 mg/dL) for greater than 2 weeks after the next scheduled course of chemotherapy (day 35) were to be withdrawn from the study for nephrotoxicity. Cisplatin was to be reduced by 20% if serum creatinine levels increased to greater than 1.0 mg/dL above baseline. Renal toxicity was further assessed by changes from baseline in creatinine clearance and incidence and severity of hypomagnesemia.

Neurologic toxicity was evaluated by a clinical neurologic examination performed by a third party blinded to the patient's treatment or adverse events reported by the patient. Neurologic examinations were performed at baseline and before the fourth, fifth, and sixth cycles of cisplatin-based therapy, as well as monthly for the first 3 months following completion of protocol therapy. Using the National Cancer Institute (NCI) common toxicity criteria, the occurrence and severity of symptoms of peripheral neuropathy or decrease in neurologic function and the cumulative dose of cisplatin at the onset of neuropathy were compared between the two treatment arms. Baseline audiograms were to be performed on all patients. Before each course of chemotherapy, patients were asked whether they had sustained any hearing loss. An audiogram was to be performed on any patients who reported impairment, with a moderate hearing loss requiring a 25% reduction in dose of cisplatin and a severe clinical hearing loss requiring discontinuation of cisplatin.

Preservation of antitumor efficacy was assessed by tumor response at second-look surgery and survival. Patients with either a complete or partial clinical response were to have their tumor assessed by second-look laparotomy. Responses assessed at surgical restaging were categorized as follows: (1) complete pathologic response—no tumor detectable and washings and multiple biopsy specimens were negative for tumor; and (2) partial pathologic response—≥ 50% reduction in disease compared with initial surgery.

Responses were determined by the study investigator responsible for the care of the patient, and reviewed and verified by the principal investigator for this study. Survival was defined as the observed length of life from protocol entry to death or date of last follow-up evaluation.

Adverse events and laboratory abnormalities were assessed using NCI toxicity criteria. For patients randomized to receive amifostine, blood pressure was measured before, at least every 5 minutes during the infusion, and 5 minutes postinfusion. A hypotensive event was defined as a decrease from baseline in systolic blood pressure as listed in Table 2. Initially, hypotensive events required stopping the amifostine infusion regardless of the transient nature of the decrease in blood pressure. However, after clinical experience with drug administration was obtained in approximately 40 patients, the protocol was amended to allow the amifostine infusion to be restarted if blood pressure returned to baseline within 5 minutes and the patient was asymptomatic.

Statistical Methods

The study was planned to accrue 200 patients. This sample size would provide greater than 80% power to detect a 50% reduction in hematologic toxicity and renal toxicity. With 100 patients per arm, the 95% confidence interval would lie within ± 10% of the true tumor response rate.

The protocol specified an interim analysis halfway through with early-stopping rules using the methodology reported by Pocock. As described earlier, the data met the criteria for early stopping and accrual was temporarily halted after 121 patients had been accrued and approximately 100 patients had completed protocol therapy. When accrual was reopened, the sample size was revised to 242 patients.

Due to complications from extensive pelvic surgery, it was not possible to determine the cause of fever or infection in 13 patients. Therefore, they were excluded from the analysis of neutropenic fever and/or infection for the first course of therapy. All other analyses presented are based on intent-to-treat basis, ie, no data are excluded due to protocol violations. Categorical data were compared using a χ2 statistic. Survival and the cumulative dose of cisplatin at the onset of neurologic toxicity were analyzed using a log-rank statistic. All P values presented are based on two-sided tests of significance.

RESULTS

Demographics

Two hundred forty-two patients were accrued to this study from April 1988 through September 1993: 122 patients were randomized to receive amifostine plus CP and 120 patients were randomized to receive CP alone. The median follow-up time is greater than 41 months. As shown in Table 3, the two treatment arms were well matched with respect to age, race, FIGO stage, extent of residual disease, and performance status.

Myelosuppression

The primary end point to assess hematologic toxicity was the cumulative incidence of grade 4 neutropenia associated with fever and/or infection that required antibiotic therapy. The percentage of patients who experienced neutropenia-associated events was reduced by 53% in the amifostine arm (P = .019). Grade 4 neutropenia associated with fever and/or infection that required antibiotic therapy continued to be a problem in the CP arm. Despite dose reductions and treatment delays, multiple episodes occurred only in the control arm. Over six cycles of therapy, pretreatment with amifostine reduced the total incidence of neutropenia-associated events by 62% (P = .005) (Fig 1). The decreased incidence of neutropenia-associated events resulted in a significant reduction in days in hospital (P = .019), as well as days on antibiotics (P = .031) (Table 4).

<table>
<thead>
<tr>
<th>Variable</th>
<th>Mean Baseline Systolic Blood Pressure (mm/Hg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 100</td>
<td>100-119</td>
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<tr>
<td>120-139</td>
<td>140-179</td>
</tr>
<tr>
<td>≥ 180</td>
<td></td>
</tr>
</tbody>
</table>

Table 2. Protocol Definition of Hypotension Requiring Interruption of Amifostine Infusion

Decrease in systolic blood pressure during infusion of amifostine (mm/Hg)

20 25 30 40 50
Table 3. Baseline Patient Characteristics

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Amifostine + CP (N = 122)</th>
<th>CP (N = 120)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td></td>
<td></td>
<td>.475</td>
</tr>
<tr>
<td>Median</td>
<td>55</td>
<td>55</td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>21-75</td>
<td>25-78</td>
<td></td>
</tr>
<tr>
<td>Race</td>
<td></td>
<td></td>
<td>.544</td>
</tr>
<tr>
<td>Black</td>
<td>11</td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>103</td>
<td>104</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>7</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Not specified</td>
<td>1</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>FIGO stage</td>
<td></td>
<td></td>
<td>.945</td>
</tr>
<tr>
<td>III</td>
<td>103</td>
<td>100</td>
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</tr>
<tr>
<td>IV</td>
<td>19</td>
<td>18</td>
<td></td>
</tr>
<tr>
<td>Not specified</td>
<td>0</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Extent of residual disease (cm)</td>
<td></td>
<td></td>
<td>.371</td>
</tr>
<tr>
<td>&lt; 2</td>
<td>79</td>
<td>71</td>
<td></td>
</tr>
<tr>
<td>≥ 2</td>
<td>43</td>
<td>49</td>
<td></td>
</tr>
<tr>
<td>Measurable disease</td>
<td></td>
<td></td>
<td>.492</td>
</tr>
<tr>
<td>Yes</td>
<td>25</td>
<td>29</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>97</td>
<td>91</td>
<td></td>
</tr>
<tr>
<td>Histology‡</td>
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<td></td>
<td>.787</td>
</tr>
<tr>
<td>Serous adenocarcinoma</td>
<td>93</td>
<td>85</td>
<td></td>
</tr>
<tr>
<td>Endometrioid adenocarcinoma</td>
<td>14</td>
<td>16</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>15</td>
<td>19</td>
<td></td>
</tr>
<tr>
<td>GOG performance status</td>
<td></td>
<td></td>
<td>.433</td>
</tr>
<tr>
<td>0, fully active</td>
<td>46</td>
<td>53</td>
<td></td>
</tr>
<tr>
<td>1, ambulatory</td>
<td>63</td>
<td>52</td>
<td></td>
</tr>
<tr>
<td>2, self-care</td>
<td>13</td>
<td>15</td>
<td></td>
</tr>
</tbody>
</table>

*Pearson χ² test, 2-sided.
†Some patients had > 1 type of histologically confirmed ovarian malignancy.

To exclude the confounding effects of extensive pelvic surgery, the incidence of neutropenia-associated events that occurred during cycles 2 to 6 was compared. Consistent with protection against cumulative hematologic toxicity, there was a significant reduction in the incidence \( (P = .012) \) and consequent days on antibiotics \( (P = .005) \) and days in hospital \( (P = .009) \) in the amifostine arm (Fig 2).

Reductions in the doses of cyclophosphamide and cisplatin for hematologic toxicity were comparable between the two treatment arms. However, despite dose reductions for grade 4 hematologic toxicity in the earlier cycles, a

Table 4. Hematologic Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Amifostine + CP</th>
<th>CP</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Incidence (by patient) of neutropenia with fever and/or infection requiring antibiotic therapy</td>
<td>11/111</td>
<td>10</td>
<td>25/118</td>
</tr>
<tr>
<td>Days in hospital</td>
<td>89</td>
<td>226</td>
<td>.019</td>
</tr>
<tr>
<td>Days on antibiotics</td>
<td>111</td>
<td>284</td>
<td>.031</td>
</tr>
<tr>
<td>Incidence of withdrawals for hematologic toxicity</td>
<td>1/122</td>
<td>1</td>
<td>8/120</td>
</tr>
<tr>
<td>% of patients with grade 4 neutropenia whose neutrophil count failed to recover to ≥ 1,500/µL by day 22 ± 3 days</td>
<td>43/98</td>
<td>44</td>
<td>64/99</td>
</tr>
</tbody>
</table>

*Two-sided.
significantly greater proportion of patients in the control arm (43% v 22% in the amifostine plus CP arm) experienced grade 4 neutropenia following the last cycle of chemotherapy (P = .001). The duration of grade 4 neutropenia, assessed by the failure of granulocyte counts to recover to ≥ 1,500/µL by day 22, was also significantly greater in the control arm (65% in the control arm v 44% in the amifostine plus CP arm, P = .004). These differences became more pronounced with multiple cycles of chemotherapy. Over the six cycles, hematologic toxicity resulted in withdrawal from chemotherapy for eight patients treated with CP alone compared with one patient treated with amifostine plus CP (P = .016).

Although cyclophosphamide and cisplatin are less toxic to the megakaryocytes and RBCs than the myeloid series, the data showed that pretreatment with amifostine resulted in an 88% reduction (P = .169) in the number of platelet units transfused and a 29% reduction in the RBC units transfused (P = .230).

Nephrotoxicity

One of the most common early signs of cisplatin renal toxicity is the excessive loss of urinary magnesium. By the last cycle of therapy, the incidence and severity of hypomagnesemia was significantly reduced in the amifostine plus CP arm (P = .001). Grade 3 or 4 hypomagnesemia, the latter characterized by seizures, occurred only in the control arm.

As defined in the protocol, cisplatin therapy was to be delayed until serum creatinine levels were ≥ 1.5 mg/dL. Overall, there was a statistically significantly greater proportion of patients in the control arm compared with the amifostine plus CP arm whose serum creatinine levels were elevated to greater than 1.5 mg/dL at day 22 (± 3 days), when the next scheduled course of chemotherapy was to be administered: 15% in the CP arm versus 5% in the amifostine plus CP arm (P = .014).

Failure of serum creatinine levels to return to ≥ 1.5 mg/dL by day 35, ie, 2 weeks after the next scheduled course of chemotherapy, precluded further administration of cisplatin. Over the course of the study, 15 patients in the CP arm compared with two patients pretreated with amifostine had treatment-limiting nephrotoxicity. This difference is statistically significant (P = .001).

Consistent with the cumulative nature of cisplatin-induced nephrotoxicity, the differences become more pronounced in the latter cycles of chemotherapy (Fig 3). By cycle 6, 36% of patients in the CP arm compared with only 10% in the amifostine arm had protracted elevations in creatinine that required a delay or discontinuation of cisplatin (P = .003).

Serum creatinine levels vary based on patient’s age and body-surface area, and may overestimate the renal function of women with ovarian cancer. Therefore, creatinine clearance values, calculated using the formula of Cockcroft and Gault, 24 which accounts for these factors, were used to compare renal function. By the last cycle of chemotherapy, 30% of patients on the CP arm compared with 13% on the amifostine plus CP arm had ≥ 40% reductions from baseline in creatinine clearance (P = .001). The effect of concurrent doses of nephrotoxic antibiotics was also evaluated. Fifteen patients on the amifostine plus CP arm and 20 on the CP arm received these drugs. In this subset of patients, 60% of controls compared with 12% of those given amifostine plus CP had ≥ 40% reductions in creatinine clearance (P = .032). Overall, an analysis of the percent of patients with a ≥ 40% reduction in creatinine clearance, with use of
nephrotoxic antibiotics as a covariant, shows a significant reduction in loss of renal function in patients pretreated with amifostine ($P < .001$).

**Neurotoxicity**

The incidence of neurologic toxicity, defined as the occurrence of symptoms of peripheral neuropathy or a decrease in neurologic function (defined as ability to perform daily activities), is related to the cumulative dose of cisplatin. By cycle 5, the difference between the two treatment arms was statistically significant ($P = .015$). Over the six cycles of chemotherapy, there was a significant reduction in the severity of peripheral neuropathy ($P = .029$) (Table 5). Two patients in the control arm and none in the amifostine arm required discontinuation of cisplatin due to neurologic toxicity. Pretreatment with amifostine resulted in a 43% reduction in the incidence of protocol-specified ototoxicity ($P = .108$), ie, clinical hearing loss or tinnitus, that required dose reduction or discontinuation of cisplatin (16% in the control arm v 9% in the amifostine plus CP arm).

Over the six cycles of therapy, 26% of patients (31 of 120) on the control arm had treatment-limiting renal, neurologic, or ototoxicity from cisplatin, compared with 10% (12 of 122) pretreated with amifostine ($P = .001$) (Fig 4).

**Antitumor Effect**

Tumor response was determined by histologic examination of biopsy specimens taken at second-look laparotomy. Eighty-six patients randomized to receive amifostine plus CP and 79 patients randomized to receive CP were eligible for second-look laparotomy. Five additional patients in each treatment arm who did not meet the protocol-defined eligibility criteria for second-look surgery did undergo this procedure. Approximately one third of eligible patients in both treatment arms did not undergo a second-look operation; they were considered as nonresponders for the purpose of this intent-to-treat analysis. The data demonstrate consistently across cohorts that amifostine does not reduce the antitumor efficacy of the CP regimen as determined by histopathologic assessment following second-look surgery (Table 6).

**Table 5. Severity of Neurologic Toxicity Following the Last Cycle of Chemotherapy**

<table>
<thead>
<tr>
<th>Chemotherapy</th>
<th>Grade 0</th>
<th>Grade 1</th>
<th>Grade 2</th>
<th>Grade 3</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amifostine + CP</td>
<td>55</td>
<td>29</td>
<td>29</td>
<td>9</td>
<td>.029</td>
</tr>
<tr>
<td>CP</td>
<td>39</td>
<td>31</td>
<td>35</td>
<td>15</td>
<td></td>
</tr>
</tbody>
</table>

The median follow-up time for the 242 patients treated in this protocol is 41 months. Fifty-three percent of patients on each treatment arm have died; the median survival time is 31 months for both treatment arms. As shown in Fig 5, there is no difference in the survival curves between the two treatment arms; comparable survival between the two treatment arms is also demonstrated when survival curves are compared in accordance with extent of residual tumor mass following debulking surgery (< 2 cm v ≥ 2 cm). For patients with less than 2 cm residual disease, the median survival times were 37.6 months in the amifostine plus CP arm and 39.7 months in the CP arm. For patients with ≥ 2 cm residual disease, the median survival times were 27.6 months in the amifostine plus CP arm and 23.7 months in the CP arm.

**Safety**

The principal side effects observed with amifostine were nausea and/or vomiting and hypotension characterized by a transient decrease in blood pressure. The only other side effects that occurred more frequently in the amifostine plus CP arm relative to the CP arm were flushing, sneezing not associated with other signs of allergic reaction, dizziness, sleepiness, hiccups, and chills. These side effects were all transient in nature and did not interfere with the patient’s therapy.

The incidence of nausea and vomiting was high in both treatments arms, as would be expected since this CP regimen is highly emetogenic. Eighty-eight percent of patients in the control arm experienced nausea and vomiting; vomiting was characterized as severe in 23% of patients. The incidence of nausea and vomiting was increased to 96%, with 30% characterized as severe, in patients who received amifostine (Table 7). Seven patients (6%) in the amifostine plus CP arm and six (5%)
in the CP arm discontinued chemotherapy due to nausea and vomiting. None of the patients who discontinued therapy had received ondansetron, as it was not approved for clinical use until late in the course of this study.

Of 122 patients treated with amifostine, 75 (61.5%) had reductions in systolic blood pressure that met the protocol definition of hypotension requiring interruption of the amifostine infusion (Table 2): 17 patients required discontinuation of the amifostine infusion before they had received the full protocol dose (including five patients who had their infusion terminated before the protocol amendment that allowed the infusion to be restarted); 27 patients had their infusion temporarily interrupted, but their blood pressure recovered to the threshold value within 5 minutes and they went on to receive the full protocol dose; and 31 patients received the full protocol dose with no interruption. Early in the study, two patients (1.6%) in the amifostine plus CP arm discontinued the protocol due to hypotension. A transient reduction in blood pressure during one infusion did not imply that a patient would experience a similar reduction in subsequent cycles. Over the course of the study, 145 of 581 (25%) of the amifostine infusions were associated with reductions in blood pressure. Reductions were generally noted toward the end of the infusion. The mean time to onset was 14 minutes and the mean duration was 6 minutes. Hypotension was readily managed by placing the patient into a supine position, or if already in a supine position, into the Trendelenburg position and infusing additional normal saline. Overall, hypotension did not result in any lasting sequelae, including impairment in cardiac, renal, or CNS function or symptoms, in any of the 122 patients treated with amifostine on this study.

DISCUSSION

Cancer chemotherapy, while continuing to improve tumor response and prolong patient survival, can also produce serious and possible life-threatening toxicities. The management of serious treatment-related complications also has pharamacoeconomic consequences. Much of the focus of supportive care has been placed on acute hematologic toxicity, and a considerable effort has been expended on the development of CSFs as a means to accelerate the release of granulocytes from the bone marrow. While CSFs are effective for the management of acute neutropenia, repeated courses of chemotherapy required for the management of most tumors result in progressive cumulative damage to bone marrow stem cells, including thrombocytopenia, anemia, and a diminished neutrophil response to subsequent doses of the bone marrow stimulant.25,26 Indeed, cumulative toxicity to a wide range of organs is one of the major unmet needs that confronts the field of cancer chemotherapy. Each course of treatment causes progressive injury to normal tissues, which initially may be subclinical, but in time will result in serious loss of organ function, manifested by morbidity or reduced tolerance of further treatment.

Whereas, for alkylating agents, such as cyclophosphamide, it is the bone marrow stem cell that is placed at risk, in the case of cisplatin, it is the kidney, peripheral nervous system, and eighth cranial nerve that may sustain...

### Table 6. Pathologic Tumor Response Rates in Patients Who Underwent Second-Look Surgery

<table>
<thead>
<tr>
<th>Response Rate</th>
<th>Amifostine + CP</th>
<th>CP</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>Pathologic complete response rate</td>
<td>26/60</td>
<td>43.3</td>
</tr>
<tr>
<td>Pathologic complete response rate + clinical partial response rate</td>
<td>45/60</td>
<td>75.0</td>
</tr>
</tbody>
</table>

### Table 7. Percent of Patients Experiencing Nausea/Vomiting

<table>
<thead>
<tr>
<th>Grade</th>
<th>Amifostine + CP (N = 122)</th>
<th>CP (N = 120)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Any</td>
<td>96</td>
<td>88</td>
<td>.02</td>
</tr>
<tr>
<td>3/4</td>
<td>30</td>
<td>23</td>
<td>.22</td>
</tr>
</tbody>
</table>
serious and permanent damage. Aggressive regimens of intravenous hydration have reduced the potential for renal tubular damage by the heavy metal, but renal toxicity nevertheless remains a serious problem and deaths continue to be reported in the literature. Importantly, the loss of renal function from cisplatin is typically permanent and surviving patients are placed at risk for further complications, including overt renal failure, if further nephrotoxic factors are superimposed on the baseline injury caused by cisplatin. In patients who relapse, treatment options are limited, since further cisplatin may be precluded, and serious toxicity may be produced if renally excreted chemotherapeutic agents, such as carboplatin, methotrexate, bleomycin, and ifosfamide, are administered. In addition, drugs and diagnostic aids, such as intravenous contrast materials, required for the primary medical or supportive care are impacted by cisplatin-related cumulative renal damage, either because of an interference of their urinary elimination or the potential for these agents to contribute further to preexisting renal damage. Currently, there is no effective measure to manage cumulative toxicity, other than to reduce dose and/or lengthen treatment schedules with the risk of losing control of the tumor. Amifostine, administered before each cycle of chemotherapy, represents the first systemically administered selective protective agent with the potential to protect against the cumulative toxicities associated with specific forms of chemotherapy.

Interest in amifostine as a cytoprotective agent arose from its history as a nuclear warfare project and preclinical studies that demonstrated protection of a broad range of normal tissues from the cytotoxic effects of chemotherapy and radiation. The mechanism for this selective cytoprotection has been carefully defined and is based on systemic and cellular pharmacokinetics. Both animal and human data demonstrated that amifostine is rapidly cleared from plasma and taken up in normal tissues. With 90% of the drug cleared from the plasma within 6 minutes, there is virtually no plasma in the systemic circulation that could potentially interact with the chemotherapeutic agents when they are administered 15 minutes after completing the infusion.

Amifostine is a prodrug that is dephosphorylated to the active metabolite, the free thiol (WR-1065), at the tissue site by cell membrane-bound and capillary alkaline phosphatase. Recent studies have shown that the specific activity of membrane-bound alkaline phosphatase is higher in normal human cells compared with the enzyme activity in the cell membranes of cancer. This biotransformation at the tissue site accounts for the rapid transport into the normal cell with negligible transport into the tumor cell. Transport into normal cells has been characterized as carrier-mediated facilitated diffusion process, whereas entry into tumor tissue has been reported to rely on the slower passive diffusion process. Complementary mechanisms based on differences in normal versus neoplastic capillary alkaline phosphatase activity, as well as the pH differences between normal and tumor tissues, also favor the conversion of the prodrug to the active uptake form (WR-1065) in normal tissues. The end result is as much as a 100-fold greater steady-state concentration of the free thiol into normal organs such as the bone marrow, kidney, and heart versus tumor tissue. Conceptually, this may be viewed as a selective temporary state of acquired resistance to the effects of chemotherapy and radiation therapy for normal tissues, analogous to a heightened intracellular concentration of the endogenous protector, glutathione.

Once the free thiol, WR-1065, has entered a normal cell, it is available to bind directly to, and thus detoxify, the active species of alkylating agents or cisplatin. As a corollary, in vitro, as well as in vivo, rodent studies have demonstrated the capacity of amifostine to reduce the genotoxic and carcinogenic properties of the therapeutic modalities in question. The second mechanism of protection involves the ability of the free thiol to act as a scavenger of oxygen free radicals, such as those derived from radiation therapy or from specific drugs, such as doxorubicin-derived superoxide anions, which have been implicated in the production of drug-induced cardiac toxicity. A broad range of normal tissues have been protected by amifostine in preclinical studies, and the consistent demonstration of reduced toxicity for the bone marrow and kidney by alkylating agents and/or cisplatin contributed to the rationale for the design of this clinical trial. This is in contrast to the results of an extensive data base for amifostine, which demonstrates no reduction on the cytotoxicity of chemotherapeutic agents—including anthracyclines, taxanes, vinca alkaloids, and other compounds—in a number of human cancer cell lines; in some instances, apparent tumor sensitization and antitumor synergy was observed. Similarly, in vivo studies that used human tumor xenografts of ovarian, lung, and breast cancer and of melanoma have demonstrated either no reduction or increased antitumor efficacy, while normal organs evidenced decreased toxicity.

The present phase III trial of CP with or without amifostine in women with stage III/IV ovarian cancer was designed based on an extensive preclinical data base and phase II clinical trials, which demonstrated that amifostine could selectively protect normal tissues from...
AMIFOSTINE REDUCTION OF CUMULATIVE TOXICITY

The toxicities of cyclophosphamide and cisplatin without reducing their antitumor effect with two principal objectives: the reduction of serious and life-threatening toxicities, with emphasis on hematologic and renal toxicities, and preservation of antitumor activity. The results of this multicenter, randomized trial confirm the ability of amifostine, administered before each cycle of chemotherapy, to protect these normal organs from the cumulative toxicities associated with six cycles of intensive CP. Amifostine pretreatment resulted in a significant decrease in the number of patients who had protracted elevations in serum creatinine levels that precluded further cisplatin therapy (30% in the amifostine arm vs. 61% in the control arm; P = 0.005). Associated with this hematoprotective effect was a 61% reduction in the number of days in the hospital for neutropenia-associated complications (P = 0.019), as well as a 61% reduction in days on antibiotic therapy (P = 0.031). The significant reduction in days in the hospital and days on antibiotic therapy required to treat fever and infections associated with grade 4 neutropenia in the amifostine plus CP arm should have significant pharmacoeconomic benefits. The cytoprotective effect of amifostine became increasingly evident during the latter cycles of therapy, which demonstrates the ability to reduce cumulative toxicity.

Similarly, amifostine protected the kidney from the toxic effects of cisplatin treatment. This was evident by significant decreases in treatment delays and in the number of patients who had protracted elevations in serum creatinine levels that precluded further cisplatin therapy in the amifostine arm. By the last cycle of therapy, 30% of CP patients sustained a ≥ 40% reduction in creatinine clearance. In contrast, 13% of patients treated with amifostine evidenced this degree of renal injury (P = 0.001). The loss of renal function in the control arm is consistent with published reports and the results of the Southwest Oncology Group trial using the same cisplatin dose. A 61% reduction in days on antibiotic therapy (P = 0.031). The significant reduction in days in the hospital and days on antibiotic therapy associated with grade 4 neutropenia in the amifostine plus CP arm should have significant pharmacoeconomic benefits. The cytoprotective effect of amifostine became increasingly evident during the latter cycles of therapy, which demonstrates the ability to reduce cumulative toxicity.

Moreover, in the amifostine plus CP arm, the concurrent use of antibiotics with recognized nephrotoxic potential, or preexisting diabetes or hypertension, did not result in an increase in the incidence of serious reductions in creatinine clearance as did the control arm. Available follow-up data from this patient population suggest that this loss in renal function persists for years, which is consistent with literature that indicate cumulative injury is typically permanent. A decrease of ≥ 40% in the creatinine clearance level represents significant renal impairment comparable to the loss of one kidney, equivalent, in essence, to a chemical nephrectomy. It makes continued therapy with cisplatin or other nephrotoxic drugs hazardous: dose reductions, typically imprecise, are generally required for drugs such as carboplatin, methotrexate, or bleomycin, for which main route of elimination is the kidneys.

Peripheral neuropathy has increasingly become a serious clinical problem with prolonged cisplatin treatment. Over the six cycles of therapy in this study, the severity of cisplatin neurologic toxicity was reduced by pretreatment with amifostine (P = 0.029); clinical hearing loss was also reduced by 40%, although the difference was not statistically significant (P = 0.108). Treatment with cisplatin is typically limited to six courses in recognition that further cumulative doses of cisplatin will likely produce a treatment-limiting toxicity (predominantly renal, neurologic, or ototoxicity), although one cannot predict in an individual patient which clinical manifestation will occur first or predominate. The effect of amifostine on global cisplatin toxicities shows that 26% of the patients (31 of 120) on the control arm had treatment-limiting renal, neurologic, or ototoxicity of a magnitude to require discontinuation of cisplatin. This was reduced to 10% (12 of 122) with amifostine pretreatment (P = 0.001).

In contrast to the cytoprotection of normal organs, the coadministration of amifostine did not protect tumor cells against the antineoplastic effects of the CP regimen. Tumor response data as assessed by second-look laparotomy were comparable in the two groups and, with a median follow-up duration of 41 months, there is no difference in survival between control and amifostine-treated patients; approximately 53% of patients in both treatment arms have died, and the median survival time of patients in both arms is 31 months. These survival data compare favorably with those reported in other multicenter studies that have attempted to use comparable doses of cyclophosphamide and cisplatin in patients with advanced ovarian cancer.

Amifostine at the recommended dose and schedule is generally well tolerated. The principal side effects are nausea and vomiting and hypotension characterized as a transient reduction in systolic blood pressure. It is recommended that drugs that could potentiate hypotension not be administered in conjunction with amifostine and that patients with preexisting cardiovascular or cerebrovascular conditions be carefully monitored. In this trial, antihypertensive medication was temporarily stopped for 24 hours before amifostine administration. It is important that patients be adequately hydrated before the amifostine infusion and that appropriate antiemetic therapy be administered. An improved antiemetic regimen for amifostine given in conjunction with emetogenic chemotherapy arose from recent studies with paclitaxel incorporating a...
dose of 20 mg of intravenous dexamethasone and a sero-
totonin antagonist (ie, ondansetron) before therapy has
greatly reduced these side effects. This regimen has now
been evaluated in more than 300 amifostine plus chem-
otherapy infusions. Only 1% of infusions were associated
with grade 3/4 vomiting and less than 1% of patients had
to have infusions discontinued for hypotension. 83

The ability of amifostine to provide selective protection
of normal tissues, while not interfering with antitumor
activity, suggests that this agent serves as an important
new adjunct in cancer management. Amifostine’s protec-
tive effects should apply to all cisplatin- and alkylating-
based regimens and thus should prove beneficial for ovar-
ian cancer patients treated with cisplatin and paclitaxel.
Consistent with the protection of cumulative toxicities,
such as cisplatin-induced nephrotoxicity, the effect of
amifostine is most apparent in the latter cycles of chem-
otherapy. However, to realize the full benefit of this selec-
tive protective agent, amifostine must be administered
before each cycle of therapy, rather than when toxicity
occurs. New trials have been initiated to explore further
the potential role of amifostine with agents such as pacli-
taxel, carboplatin, and doxorubicin, as well as radiation
therapy.

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