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N-(Phosphonacetyl)-L-aspartate (PALA) in Advanced Breast Cancer: a Phase II Trial of the EORTC Breast Cancer Cooperative Group*

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Abstract—*Twenty-nine evaluable patients with extensively pretreated breast cancer received PALA, a new pyrimidine antimetabolite. The drug was given by intravenous infusion over* 60 min, *at a daily dose of* 2.5 g/m^2 *for* 2 *consecutive days. Courses were repeated at* 2*-week intervals and doses were escalated to toxicity. Two objective partial remissions were observed, lasting for* 3 *and* 4.5 *months respectively. Toxic effects were dose-related and consisted mainly of mucocutaneous manifestations, i.e., skin rashes, stomatitis, diarrhea, conjunctivitis and corneal ulcerations. Evidence of antitumor potential in far-advanced disease and lack of myelosuppression point to the need for additional trials of PALA in a more favorable selection of patients with breast cancer.*

INTRODUCTION

N-(PHOSPHONACETYL)-L-ASPARTATE (PALA) is a new anticancer agent which was synthetized by Collins and Stark [1] as a transition-state inhibitor of aspartate transcarbamylase, a key enzyme in the *de novo* biosynthesis of pyrimidine nucleotides. This inhibition results in cytotoxic effects which may be reversed *in vivo* by uridine and carbamyl aspartate [2].

The spectrum of activity of PALA in experimental tumors appears to be unique [3–5]. PALA is curative without clear schedule dependency in the Lewis lung carcinoma; the drug is also effective against a variety of other murine solid tumors including CD8 F1 mammary carcinoma, glioma 26, B16 melanoma, colon carcinoma 26, C3H mammary carcinoma and mammary xenograft MX-1 [6, 7]. In contrast, L1210 and P388 murine leukemias are relatively or completely resistant to PALA. This unusual pattern of antitumor activity suggested that this compound would be effective against slowly growing tumors without affecting rapidly proliferating tissues and the bone marrow in particular [8]. Accordingly, toxicologic studies in animals confirmed the absence of drug-induced bone marrow impairment [6, 9]. Dose-limiting toxic effects in the dog were seen in the gastrointestinal tract and the central nervous system [9].

In humans, only phase I data were available when the present trial was initiated [10–15]. With any schedule, mucocutaneous toxicity was consistently found to be dose-related and dose-limiting, but no significant hematologic changes were observed. This report describes our phase II trial with this agent in patients with advanced breast cancer.

PATIENTS AND METHODS

Patients

All patients entered into the study had progressive, histologically proven recurrent or metastatic breast carcinoma, with measurable or evaluable lesions. Lymphoedema, hilar enlargement, pleural effusion, ascites, bone

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marrow invasion and/or osteoblastic skeletal infiltration were not considered as evaluable lesions. Patients who had previously received all conventional treatments, including surgery, radiotherapy, endocrine treatments and chemotherapy, were eligible provided that all drugs had been withdrawn for at least 4 weeks and that all related toxic manifestations had resolved. Concomitant radiotherapy for control of bone pain could be given, provided that all evaluable lesions were not included in the irradiated field. Patients with poor general condition (Karnofsky index below 50%), short life expectancy (< 3 months), central nervous system metastases or patients with a second cancer were not eligible. Other eligibility criteria included WBC $\geq 3000/mm^3$, platelets $\geq 75{,}000/mm^3$, creatinine ≤ 1.2 mg/dl and bilirubine ≤ 1.5 mg/dl.

Treatment

PALA was supplied by the National Cancer Institute (Bethesda, MD) in 10 ml ampuls containing 1 g of PALA in water for injection USP, with sodium hydroxide to adjust to pH 6.5–7.5. The intact ampuls were stored at 4°C and were diluted in 450 ml saline immediately prior to drug administration.

Treatment consisted of an intravenous infusion over 60 min at a daily dose of 2.5 g/m^2 for two consecutive days. Courses were repeated at 2-week intervals with dosage adjustments according to patient's individual tolerance. The protocol called for dose escalation by 20% when no toxicity was encountered during the previous course. Dosage was decreased by 20–50% when significant side-effects had occurred during the previous course.

All patients had to receive at least 3 courses of treatment unless there was evidence of rapid progression of the disease or unacceptable toxicity despite adequate dosage adjustments. Responders were treated until progression.

Follow-up and criteria of response

During the first two courses of treatment, physical examination, measurements of tumor lesions and recording of side-effects were repeated at weekly intervals. If complications did not occur, the patients were subsequently assessed at 2-week intervals, on the first day of treatment. Complete blood counts and automated chemical (SMA-12) profiles were also repeated before each course. Baseline investigations, including chest roentgenogram, skeletal survey and liver scan, were repeated after 6 courses of therapy in patients who did not present earlier evidence of progression of their disease.

U.I.C.C. criteria were used to evaluate response [16]. Briefly, objective remission required a decrease of 50% or greater in the sum of the products of the two largest perpendicular diameters of all measurable lesions and/or recalcification of osteolytic lesions; patients were classified as having stable disease when a decrease of less than 50% or an increase of less than 25% over original measurements was recorded; an increase of more than 25% over original measurements and/or the occurrence of new lesions, and/or progression of osteolytic lesions indicated progressive disease.

RESULTS

Forty-one eligible patients were included in the trial. In twelve of these, however, the therapeutic response could not be evaluated because of death within one month from initiation of treatment (7 patients), treatment withdrawal after one course for severe toxicity (2 patients) or treatment refusal (1 patient) and insufficient data (2 patients). Pretreatment characteristics in the 29 evaluable patients (Table 1) were similar to those in patients excluded from this analysis. All patients had far-advanced disease and had been heavily pretreated with surgery, radiotherapy, chemotherapy and endocrine treatments. Only two patients experienced partial remission (Table 2). The first one had extensive cutaneous lesions which markedly regressed for 4.5 months from initiation of the therapy. The other patient had a more than 50% reduction in the size of skin lesions and pulmonary nodules during a 3-month period. Worthy of note, both responders were considered resistant to conventional systemic treatment with hormones (2 and 3 drugs respectively) and chemotherapy (5 drugs each). Five patients, who had received 3–9 courses of PALA, had stabilization of their disease for 2–6 months. In 22 patients the dis-

Table 1. Pretreatment characteristics of evaluable patients

Total no.	29
Median age in years (range)	56 (32–72)
Median Karnofsky index (range)	80% (50–100)
Prior surgery	28
Prior chemotherapy	29
median no. drugs/patient (range)	5 (1–8)
Prior endocrine therapy	29
median no. drugs/patient (range)	2 (1–5)
Prior radiotherapy	29
Dominant site of disease	
soft tissue	6 (21%)
bone	7 (24%)
viscera	16 (55%)

Table 2. Response to PALA according to dominant site of disease

Dominant site	All patients	No. of patients Remission	No change	Progression
Soft tissue	6	1	2	3
Bone	7	0	0	7
Viscera	16	1	3	12
Total	29	2	5	22

ease clearly failed to respond to treatment and most of these showed evidence of malignant progression after 2 or 3 courses. At present, 19 of the 29 evaluable patients have died from their disease. The median survival time from the commencement of PALA was 106 days (range 33–480 days), reflecting a selection of poor patients in this trial.

A total number of 112 courses of therapy were given to the 29 patients evaluable for response, with a median number of 3 courses per patient (range 2–10). Therapy was generally administered on an out-patient basis. The toxic effects encountered during treatment are listed in Table 3. Although frequently reported, nausea and vomiting were generally mild. Mucocutaneous toxicity was frequently encountered and consisted mainly of diarrhoea, skin rashes and stomatitis, which often occurred simultaneously. These manifestations were rapidly reversible and had generally subsided at the time of scheduled retreatment. Severe side-effects were reported in 11 cases and could be alleviated by a 20% reduction in dosage of PALA in 9 patients. A 20% increase in the dosage of PALA was performed in 9 patients who had no side-effects after the first course and increased dosages could be maintained subsequently in 6 of these. In 3 cases, ophthalmic complications were observed consisting of conjunctivitis, which was associated with corneal ulcerations in 2 cases. These ulcerations healed rapidly after cessation of treatment. During the infusion of PALA, a few patients complained of drowsiness or mild headache and one had mild hypotension. No treatment-related hematologic changes nor toxic-related deaths were observed.

Table 3. Incidence of toxic effects in 33 patients

Toxic effect	Percentage of toxic patients
Nausea–vomiting	48 (3)*
Diarrhoea	48 (9)
Dermatitis	45 (24)
Stomatitis	42 (9)
Drowsiness–headache	12
Conjunctivitis	9
Corneal ulcerations	6
Hypotension	3

*Numbers in parentheses indicate % of patients who had dose reductions due to toxicity.

DISCUSSION

This trial is the first to present evidence of antineoplastic activity of PALA in advanced breast cancer. The response rate observed here was low (2/29) and remissions were short-lived. These results are, however, of interest, considering that they were obtained in patients with heavily pretreated end-stage disease. Short overall survival and a high proportion of early deaths clearly indicates how unfavourable the selection of patients was. Of note, one responder whose disease had failed to respond to prior chemotherapy with a conventional CMF regimen received phase I therapy when the disease became clearly refractory to PALA. She was subsequently treated with a combination of PALA (500 mg/m^2 on days 1–5) + 5FU (275 mg/m^2 on days 1–5), given at 3-week intervals. A second 50% regression of soft tissue lesions was obtained for 2 months with this 2-drug combination.

As described with other schedules [11–15], mucocutaneous toxicity was dose-related and represented the dose-limiting side-effect in most of our patients. We also observed ocular manifestations with corneal ulcerations in several cases. These complications should justify routine ophthalmologic controls in patients treated with PALA. The present schedule, consisting of two consecutive daily injections repeated at two-week intervals, is convenient for ambulatory patients. At the starting dose selected for this trial the drug is generally very well tolerated and subsequent dose escalations are often possible. The lack of hematologic toxicity of PALA deserves special consideration for combination chemotherapy.

The spectrum of antitumor activity of PALA in humans remains to be defined. Used as a single agent, the drug seemed to be inactive in

colorectal carcinoma [17, 18] and in non-small cell carcinoma of the lung [19]; hints of activity were reported by two groups in malignant melanoma [20, 21]. Our present results, obtained in far advanced cases, suggest that PALA might be an effective drug against breast cancer. Its actual antineoplastic activity should be preferably assessed in more favorable cases.

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