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Dexverapamil to overcome epirubicin resistance in advanced breast cancer

Abstract Resistance to cytotoxic chemotherapy is a major problem in the management of patients with metastatic breast cancer. Various data suggest P-glycoprotein-associated multidrug resistance (MDR) to be a relevant resistance mechanism in this tumor. The purpose of this study was to evaluate feasibility and activity of combining oral dexverapamil, a second-generation chemosensitizer currently in clinical development for MDR reversal, with epirubicin in patients with epirubicin-refractory high-risk metastatic breast cancer. Patients first received epirubicin alone at 120 mg/m^2. In cases of clinical refractoriness, epirubicin was continued at the same dose and schedule but supplemented with oral dexverapamil. Dexverapamil was given at 300 mg every 6 h for a total of 13 doses and commenced 2 days prior to epirubicin administration. At the time of this interim analysis, 41 patients had received epirubicin alone and 20 proceeded to treatment with epirubicin plus dexverapamil. Of the 20 patients, 14 were considered evaluable for toxicity and activity. Addition of dexverapamil resulted in a significant decrease in mean heart rate and blood pressure as well as prolongation of PQ time as compared to epirubicin alone. However, these cardiovascular effects of dexverapamil were usually mild, and subjective tolerance of treatment was good. In 7/14 patients, dose escalation of dexverapamil was feasible. Dexverapamil had no effect on epirubicin toxicities and did not require reduction of the epirubicin dose. In 2/14 patients, the addition of dexverapamil to epirubicin was able to convert progressive disease and no changes respectively, into partial responses. In 3 patients with progressive disease, addition of dexverapamil temporarily prevented further tumor progression. Analyses of dexverapamil and nor-dexverapamil plasma levels, of in vitro reversal activity of patient sera containing dexverapamil, and of epirubicin pharmacokinetics without and with dexverapamil are currently in progress. Addition of oral dexverapamil to epirubicin 120 mg/m^2 proved to be feasible in a multiinstitutional setting. Patient accrual is continuing to determine whether dexverapamil is capable of overcoming epirubicin refractoriness in a significant number of patients with metastatic breast cancer.

Key words Dexverapamil · Epirubicin · Breast cancer

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Introduction

Resistance to cytotoxic chemotherapy is a common problem in the clinical management of cancer patients. Cancers frequently respond initially to cytotoxic treatment but eventually progress or relapse despite chemotherapy, and show resistance not only to the cytotoxic agents used but also to drugs patients had not received before. A similar experimental phenomenon has been termed multidrug resistance or MDR (Endicott and Ling 1989). Various molecular mechanisms have been identified that can render

cancer cells multidrug-resistant, including overexpression of the MDR gene *MDR1*/P-glycoprotein (Pgp), alterations in topoisomerase II or changes in the glutathione system (Moscow and Cowan 1988; Gottesman and Pastan 1993). The mechanism that has received most clinical attention in recent years has been MDR. In breast cancer, overexpression of Pgp/*MDR1* has been detected at various frequencies (Lehnert 1993). In locally advanced breast cancer, Pgp/*MDR1* positivity has been associated with a poorer response to doxorubicin-based pre-operative chemotherapy as well as a shorter time to treatment failure (Verrelle et al. 1991).

In 1981, the calcium-channel blocker verapamil was reported to be capable of overcoming MDR in preclinical models (Tsuruo et al. 1981). These observations have prompted a number of studies, which have shown a variety of compounds to share this ability with verapamil (Ford and Hait 1990; Lehnert 1994). Recently, high-dose infusional verapamil has been found capable of reversing clinical drug resistance in patients with multiple myeloma (Salmon et al. 1991) and of enhancing chemotherapy activity in heavily pretreated patients with drug-refractory malignant lymphomas (Miller et al. 1991). These studies have also made clear, however, that the plasma/tissue concentrations needed for effective MDR reversal cannot be achieved by racemic verapamil because of its cardiac effects.

Various studies have found the D enantiomer of verapamil to have similar molar potency in reversing MDR to the S enantiomer or the racemic mixture of verapamil (Gruber et al. 1988; Mickisch et al. 1990; Pirker et al. 1990). D-Verapamil had been previously demonstrated to have five- to tenfold lower cardiac activity as compared to the *S* enantiomer (Echizen et al. 1985). Hence, it was hoped that dexverapamil might be able to yield higher plasma/tissue concentrations as compared to the racemic mixture, which in turn might enhance clinical effectiveness and utility of MDR reversal. The goal of this multiinstitutional phase II trial was to evaluate whether treatment with 120 mg/m^2 epirubicin plus oral dexverapamil is feasible in a multiinstitutional setting and whether the addition of dexverapamil is able to overcome epirubicin resistance in patients with metastatic breast cancer.

Patients and methods

Major eligibility criteria for study entry were histologically or cytologically proven breast cancer; age between 18 and 65 years; measurable metastatic disease with documented tumor progression; adequate bone marrow, liver and renal function; no significant cardiac disease; resting systolic blood pressure of at least 13.3 kPa (100 mm Hg); heart rate of at least 50/min; no concomitant treatment with cardiac or antihypertensive agents or agents known to be capable of reversing MDR; cumulative doses of doxorubicin, epirubicin and mitoxantrone, respectively lower than 240 mg/m^2, 360 mg/m^2 and 56 mg/m^2, and written informed consent.

To be able to assess dexverapamil effects on epirubicin activity and toxicity, a two-stage design was used. Patients first received epirubicin alone at three-weekly intervals. In cases of progressive disease or no change after two cycles or progression after temporary response, patients continued to receive epirubicin at the same dose and schedule sponses to two epirubicin cycles continued to receive epirubicin alone until tumor progression. Patients were withdrawn from study if any of the following occurred: progressive disease after two cycles or no change after four cycles of epirubicin plus dexverapamil, progression after temporary response to epirubicin plus dexverapamil, severe toxicity, or a total cumulative dose of epirubicin above 1000 mg/m^2.

Epirubicin was given at a dose of 120 mg/m^2 as a 15-min infusion every 3 weeks. The epirubicin dose was reduced by 25% if any of the following occurred: a white blood cell nadir below 1.0 × 10^9/l, leukocytopenia of WHO grade 3 or 4 associated with infection, a platelet nadir below 50 × 10^9/l, mucositis of WHO grade 3 or 4, or mucositis of WHO grade 2 lasting more than 7 days. Dexverapamil was given at 300 mg every 6 h for a total of 13 doses, starting 2 days prior to epirubicin administration. The dexverapamil dose was reduced to 250 mg if treatment was associated with a drop in systolic blood pressure below 10.7 kPa (80 mm Hg) or any fall in blood pressure associated with symptoms or prolongation of PQ time of 0.28 s or more. The dexverapamil dose was escalated to 350 mg if no undue toxicities were observed to 300 mg. To allow close cardiovascular monitoring, patients were hospitalized for the first cycle of dexverapamil and for any further cycle of dose-modified dexverapamil.

As collateral studies, the determination of dexverapamil and nordexverapamil plasma levels and analysis of the ability of dexverapamil-containing patient sera to reverse MDR in vitro were mandatory, detection of *MDR1*/Pgp expression in tumor biopsies and pharmacokinetics of epirubicin without and with dexverapamil were optional.

The endpoint used for statistical evaluation of dexverapamil activity was response rate, and a response rate of 20% was selected as the activity level of interest. The design described by Gehan and Schneiderman was used to calculate the number of patients needed to determine the response rate with a standard error of below 0.10 and to ensure that the risk of missing a response rate of 20% was 0.05 or less (Gehan and Schneiderman 1982). Tumor responses were evaluated

Table 1 Patient and tumor characteristics (n = 14)

Characteristic	No. of patients
Age (years), median (range) 55 (35–64)	
Menopausal status	
Premenopausal	4
Postmenopausal	10
Disease-free interval	
<2 years	6
⩾2 years	4
None	4
Sites of metastases (mutually non-exclusive)	
Liver	8
Lung	4
Bone	8
Others	8
2 sites	7
⩾3 sites	7
Dominant site of disease	
Visceral	11
Bone	1
Soft tissue	2
Prior chemotherapy	11
Adjuvant only	4
Palliative only	5
Adjuvant + palliative	2
Prior anthracyclines	8
Best prior response to palliative chemotherapy	
Complete response	1
Partial response	0
No change	2
Progressive disease	4

Table 2 Cardiovascular toxicity. All values are means ± SEM (*PQ*, *EPI* epirubicin, DVPM dexverapamil

Parameter	EPI alone	EPI + DVPM	P^a
Heart rate (beats/min)	78±10	69±8	<0.01
Blood pressure (mm Hg)			
Systolic	132±15	111±15	<0.01
Diastolic	82±5	69±8	<0.01
PQ time (ms)	166±20	191±23	<0.01

[a] Calculated by Student's *t*-test

Table 3 Cardiovascular toxicity per patient (n = 14) (*EPI* epirubicin, *DVPM* dexverapamil)

Parameter	Number of patients	
	EPI alone	EPI + DVPM
Heart rate		
>60 beats/min	12	6
50–59 beats/min	2	6
40–49 beats/min	0	2
Systolic blood pressure		
>100 (mm Hg)	14	6
90–99 (mm Hg)	0	3
80–89 (mm Hg)	0	5
Atrioventricular block		
First degree	2	6
Higher degree	0	0
Atrioventricular rhythm	0	0

according to UICC (International Union Against Cancer) criteria (Hayward et al. 1977); toxicities were graded according to World Health Organization (WHO) criteria (Miller et al. 1991).

At the time of this interim analysis, 41 patients had received epirubicin alone and 20/41 proceeded to the second stage. Of the 20 patients, 14 were considered evaluable for toxicity and dexverapamil activity and are the subject of this report. Their main characteristics are shown in Table 1. Eleven patients presented with visceral metastases, 8 patients had liver metastases and 8 patients had at least two sites of disease. Eight patients had previously received anthracyclines, 7 patients had had prior palliative chemotherapy, and only 1/7 patients had experienced a response to such therapy. Accordingly, the vast majority of the study population had poor risk criteria.

Results

The cardiovascular toxicities observed in patients receiving epirubicin alone compared to those receiving epirubicin supplemented with dexverapamil are summarized in Tables 2, 3. Although the addition of dexverapamil resulted in a significant drop in heart rate and blood pressure as well as prolongation of PQ time, these effects were usually mild. In the 12 patients who received at least two cycles of dexverapamil, dose escalation of dexverapamil was possible in 7 and dose reduction was required in 2 patients.

Non-cardiovascular toxicities were similar in patients receiving epirubicin alone or with dexverapamil (Table 4). Six and 7 patients, on epirubicin alone and epirubicin plus dexverapamil respectively, experienced white blood cell

Table 4 Non-cardiovascular toxicity per patient (n = 14) (*EPI* epirubicin, *DVPM* dexverapamil)

Parameter	Number of patients									
	EPI alone					EPI + DVPM				
	0[a]	1	2	3	4	0[a]	1	2	3	4
White blood cells	1	3	4	4	2	1	2	4	6	1
Platelets	11	2	1	0	0	11	2	0	1	0
Hemoglobin	8	4	2	0	0	4	6	4	0	0
Mucositis	13	0	1	0	0	11	0	3	0	0
Nausea/vomiting	10	2	2	0	0	8	4	1	1	0

[a] Grading according to WHO criteria

Table 5 Response evaluation (n = 14) (*EPI* epirubicin, *DVPM* dexverapamil)

Response	Number of patients	
	EPI alone	EPI + DVPM
Complete	0	0
Partial	1	2
No change	7	9
Progressive disease	6	3

complications. All other toxicities were mild and subjective tolerance of treatments was usually good. In 1 patient, dexverapamil cycles were associated with a generalized flush, which rapidly dissolved after each treatment course.

Tumor responses to epirubicin alone and with dexverapamil are shown in Table 5. In 2/14 patients the addition of dexverapamil to epirubicin was able to convert progressive disease and no change, respectively, into partial responses. In 3 patients who progressed to epirubicin alone, the addition of dexverapamil temporarily prevented further tumor progression.

Analyses of dexverapamil and nor-dexverapamil plasma levels and of the ability of dexverapamil-containing patient sera to reverse MDR in vitro are currently in progress, as are analyses of epirubicin pharmacokinetics without/with dexverapamil.

Discussion

In the present study, oral dexverapamil given at 300 mg every 6 h for a total of 13 doses was well tolerated by the patients when added to epirubicin at 120 mg/m^2. In half of the evaluable patients, dose escalation of dexverapamil was feasible. Analyses of plasma levels of dexverapamil and nor-dexverapamil are currently in progress, as are analyses of the in vitro ability of patient sera containing dexverapamil to reverse MDR. These data will be critical for determining whether the dexverapamil regimen used in this study was able to yield plasma levels deemed sufficient for effective MDR reversal.

Dexverapamil had no effect on epirubicin toxicity.

oral racemic verapamil when added to the same dose of epirubicin as used in the present study (Mross et al. 1993a), and for high-dose racemic verapamil added to chemotherapy with vincristine, doxorubicin and dexamethasone (VAD) in patients with multiple myeloma (Salmon et al. 1991). Nonetheless, in 30% of the patients with VAD-refractory multiple myeloma, verapamil has been capable of re-inducing remission to VAD. Lack of enhanced epirubicin toxicity following addition of dexverapamil is suggestive of little pharmacokinetic interaction between the two agents. This might be due to not having achieved plasma/tissue concentrations of dexverapamil sufficient for inhibition of the physiological Pgp function. Addition of cyclosporin A to etoposide, for instance, has been reported to result in an approximately 50% increase in the area under the etoposide time curve (AUC) as a result of reduced hepatic and renal elimination (Lum et al. 1992). This increase in etoposide AUC has been associated with significantly increased bone marrow toxicity. It must be emphasized, though, that those observations have not proved pharmacokinetic interaction between cyclosporin and etoposide to be the result of cyclosporin-induced Pgp inhibition in liver and kidney cells. Thus it might be possible to achieve Pgp-blocking concentrations of MDR modulators without altering the pharmacokinetics of cytotoxic agents. In recently reported preliminary studies, dexverapamil has been observed to result in altered distribution of doxorubicin rather than in reduced elimination (Scheithauer et al. 1993). Addition of racemic verapamil to epirubicin has been found to alter the metabolism of epirubicin significantly without affecting parameters such as AUC, terminal half-life or volume of distribution (Mross et al. 1993b). Analyses of epirubicin pharmacokinetics without and with dexverapamil are in progress in a subset of this study population.

In 2 of the 14 patients, addition of dexverapamil was capable of inducing partial remissions when added to epirubicin. One of these patients had tumor progression to epirubicin alone. In the other patient, the tumor showed no change after two cycles of epirubicin and the possibility cannot be excluded that this patient would have gone into remission without receiving added dexverapamil. However, on the basis of experience with 120 mg/m^2 epirubicin combined with cyclosphosphamide in patients with metastatic breast cancer, remissions are usually achieved rapidly, i.e., following two treatment cycles, or not at all (Marschner et al. 1994).

On the basis of these preliminary data, adding oral dexverapamil to epirubicin at the doses and schedule used in this study appears to be feasible in a multiinstitutional setting. Subjective tolerance was good and objective toxicities were moderate. Further patient accrual is ongoing to determine whether dexverapamil is capable of overcoming epirubicin refractoriness in a significant number of patients with metastatic breast cancer.

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