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ORIGINAL ARTICLE

A phase I study of vinflunine in combination with capecitabine in patients with metastatic breast cancer previously treated with anthracyclines and taxanes

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Abstract
Purpose A phase I study was performed to determine the maximal tolerated dose (MTD), recommended dose (RD), safety and efficacy of vinflunine when combined with capecitabine in patients with metastatic breast cancer (MBC) previously treated with anthracyclines and taxanes, with pharmacokinetic blood sampling to test potential drug–drug interactions.
Patients and methods Sixteen patients with MBC who had received anthracyclines and taxanes in the neo/adjuvant setting, if progression occurred during or within 12 months of chemotherapy completion, and/or as first-line chemotherapy of MBC were enrolled. Vinflunine (VFL) was given on day 1 with capecitabine (CAPE) twice daily from days 1 to 14, every 3 weeks. Three dose levels (DL) were investigated (DL1: VFL 280 mg/m² and CAPE 1,650 mg/m²/day, DL2: VFL 320 mg/m² and CAPE 1,650 mg/m²/day and DL3: VFL 280 mg/m² and CAPE 2,000 mg/m²/day).
Results The RD was established as vinflunine 280 mg/m² on day 1 plus capecitabine 1,650 mg/m²/day on days 1 to 14 given every 3 weeks. Dose-limiting toxicities were grade 4 neutropenia lasting at least 7 days for 2 patients, anorexia with fatigue for 1 patient and diarrhoea with fatigue, anorexia and febrile neutropenia for 1 patient. Neutropenia was the main toxicity of the combination, it was reported in 15 patients (93.8%) with grade 3 in 7 patients (43.8%) and 22.6% of cycles and grade 4 in 7 patients (43.8%) and 19.8% of cycles. Complications were rare with only one patient experiencing febrile neutropenia at DL exceeding the RD. The most frequent non-haematological toxicities were fatigue and gastrointestinal disorders; however, no grade 3 or 4 episode was observed at the RD. Hand–foot syndrome was reported in 5 patients (31.3%) and 22.6% of cycles, no episode of grade 3 was seen. Concerning pharmacokinetics, no modifications were detected for VFL, while slight accumulation between days 1 and 14 was observed for 5-FU formed from CAPE. The risk of clinical significant drug–drug interaction was considered weak. Objective partial responses were reported in 7 patients, yielding a response rate of 43.8% in the all-treated population according to the investigator assessment.
Conclusions The combination of vinflunine and capecitabine is safe and showed promising antitumour activity in MBC patients who have failed prior anthracyclines and taxanes. Further clinical development of this combination is warranted.

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Keywords Vinflunine · Capecitabine · Anthracycline/taxane-pretreated · MBC · Phase 1 · Second line

Introduction

Anthracyclines and taxanes constitute the most active cytotoxic agents in breast cancer and are increasingly used in the adjuvant setting and as first-line chemotherapy of metastatic breast cancer (MBC). For patients with MBC who have failed anthracyclines and taxanes, treatment options are limited and there is a need for new effective agents. Capecitabine has demonstrated significant antitumour activity [6] in this patient's population and has been approved in this setting.

Vinflunine is a novel antitubulin agent [17] obtained by using superacidic chemistry. It suppresses microtubule dynamics, causes cell cycle arrest in the G_2/M phase and ultimately leads to apoptotic cell death [16, 18, 19]. The low affinity of VFL for tubulin and its readily reversible effect on it may reduce clinical neurotoxicity [12, 20]. It is noteworthy that no grade 3 or 4 neuropathy has been observed in clinical trials of vinflunine.

Vinflunine has shown clinical activity against multiple tumour types including metastatic breast cancer, advanced bladder cancer, advanced non-small cell lung cancer and mesothelioma. In metastatic breast cancer, vinflunine 320 mg/m² given every 3 weeks was assessed in two multicenter phase II studies as second- and third-line chemotherapy after anthracycline and taxane failure. Among 60 patients treated in second-line, 18 responses were documented, yielding a response rate of 30% (95% CI: 19–43).

In the 21 patients who relapsed during or within 6 months of completing a taxane-based regimen, the response rate was 34.4%. Overall median progression-free survival and median survival were 3.7 and 14.3 months, respectively [8].

Among 56 patients treated in third-line, a response rate of 13% (95% CI: 5–24), a median progression-free survival of 2.6 months and a median survival of 11.4 months were reported [14].

Based on a potent synergism of the combination of vinflunine and 5-fluorouracil in preclinical models [2], a phase I study was designed to establish the optimal doses of vinflunine and capecitabine to be used in MBC patients and to investigate potential drug–drug interactions when co-administered.

Patients and methods

The study protocol and its amendments were submitted and approved by the Ethics Committee and the Health Authorities according to local requirements. The trial was designed according to the current Declaration of Helsinki (Somerset West 1996) and conducted in accordance with Good Clinical Practice Guidelines. Written informed consent was obtained from each participating patient before study entry. The cut-off date for analysis was 2 November 2005.

Patients selection

Patients were recruited from 4 centres between September 2003 and November 2005. Patients were required to have histologically proven metastatic breast carcinoma with documented progressive disease, to have received prior anthracycline and taxane-based therapy in the neo/adjuvant setting and/or in the first-line treatment of MBC. Patients who progressed during or within 12 months of completing neo/adjuvant chemotherapy that contained an anthracycline and a taxane were also eligible. Until RD, patients with non-measurable disease according to RECIST were allowed; however, at the RD, only patients with at least one measurable lesion were to enter the study. Other requirements included age $\geq$18 years and $\leq$75 years; predicted life expectancy $\geq$3 months; a WHO performance status (PS) of 0, 1 or 2; no severe or uncontrolled medical conditions; no symptomatic peripheral neuropathy grade $\geq$2 according to the National Cancer Institute Common Toxicity Criteria (NCI CTC) (version 2.0); an ECG without clinically significant abnormality; adequate haematologic (absolute neutrophil count $\geq 2.0 \times 10^9$/L, platelets $\geq 100 \times 10^9$/L), liver (bilirubin $\leq 1.5 \times$ upper normal limit UNL, transaminases $\leq$2.5, UNL unless due to liver involvement) and renal functions within 7 days before study drug administration.

Study design and dose escalation scheme

A standard phase I design was used with 3 to 6 patients per dose level (DL). The maximal tolerated dose (MTD) was defined as the DL at which two or more out of three or two or more out of 6 patients developed a dose-limiting toxicity (DLT) during the first cycle.

The recommended dose (RD) for further clinical testing was defined as the DL below the MTD and had to be assessed in at least 6 patients. DLTs were one of the following: grade 4 neutropenia lasting 7 days or more, febrile neutropenia as defined by the NCI CTC; neutropenic infection (grade 3 or 4 infection concomitant with grade $\geq$3 neutropenia); grade 4 thrombocytopenia or thrombocytopenia with bleeding or requiring platelet transfusion; any grade $\geq$3 non-haematological toxicity except inadequately treated nausea or vomiting; and delay of more than 3 weeks in starting the second cycle because of toxicity.

Study drug administration

To be evaluable for MTD determination, each patient had to receive one cycle of treatment except in case of discontinuation due to the occurrence of DLT. Each patient had to receive at least two cycles of treatment, to be evaluable for efficacy except in the case of early progression. Vinflunine was given intravenously as a 20-min infusion once every 3 weeks.

Capecitabine was supplied as 150 and 500 mg tablets and administered orally twice daily from days 1 to 14 every 3 weeks.

Individual doses of both drugs were determined as the product of patient's body surface area times the dosage. It was rounded to the nearest 100 mg of the total daily dose for capecitabine.

Three DLs were investigated: DL1 (VFL 280 mg/m² and CAPE 1,650 mg/m²/day), DL2 (VFL 320 mg/m² and CAPE 1,650 mg/m²/day) and DL3 (VFL 280 mg/m² and CAPE 2,000 mg/m²/day).

On the day of cycle start (cycle defined as a three-week period), neutrophils had to be $\geq 1.5 \times 10^9$/L and platelets $\geq 75 \times 10^9$/L. If a patient required a cycle delay, both drugs were delayed for a maximum of 2 weeks.

No concomitant prophylactic use of GCSF was allowed, and it could be given only in case of febrile neutropenia according to local practice.

The dose of vinflunine had to be reduced from 320 to 280 mg/m² and from 280 to 250 mg/m² for subsequent cycles in the event of grade 4 neutropenia lasting for 7 days or more, febrile neutropenia or grade 4 thrombocytopenia.

If grade ≥ 2 constipation lasting 5 days or more occurred, the dose of vinflunine had to be reduced to the next lower dose for subsequent cycles.

If grade ≥ 2 mucositis lasting 5 days or more occurred, both the dose of vinflunine and capecitabine had to be reduced to the next lower dose for subsequent cycles.

In case of grade ≥ 2 hand–foot syndrome, capecitabine was withheld until resolution to grade ≤ 1 and the dose reduced to the next lower dose for subsequent cycles.

Pretreatment evaluation and follow-up

At baseline, physical examination and routine laboratory tests were performed and WHO performance status was determined. Complete blood cell count and differential white blood cell count were obtained on a weekly basis. Biochemical profile was performed pretreatment and then before each cycle. All toxicities were graded using the National Cancer Institute Common Toxicity Criteria (version 2.0). Tumour measurements were performed at baseline and every 6 weeks. Responses were assessed using RECIST [30].

Pharmacokinetic assessment

PK were assessed on day 1 (VFL + CAPE) and day 14 (CAPE alone) of the first cycle. Blood samples were collected at pre-dose, 20 min (end of VFL infusion), 45 min, 4, 11, 48, 96 and 168 h for VFL and at pre-dose, 30 min, 1, 2, 3, 4, 5 and 7 h for CAPE. Bioanalysis methods were HPLC/UV with a lower limit of quantification (LLOQ) of 2 ng/mL for VFL and for its metabolite DVFL (4-0 deacetylvinflunine) in blood [35] and 50 ng/mL for CAPE and its metabolites (5′-DFCR, 5′-DFUR and 5-FU) in plasma [25].

For VFL, the area under the curve of blood concentrations versus time from zero to infinity (AUC_{inf}) and total clearance (Cl_{tot}) were obtained by Bayesian estimation (in-house population PK model, NONMEM®, GloboMax-ICON, USA). Since DVFL was not included in this model its blood concentrations were graphically compared with those of a REFERENCE group (79 patients from phase I studies, VFL single agent). For CAPE and its active metabolite 5-FU, AUC_{last} (AUC from zero to last quantifiable concentration) was calculated by non-compartmental methods (Kinetica®, Thermo Electron Corp., USA). Interaction impact on VFL was controlled according to international guidelines [CPMP/EWP/560/95, Dec. 1997] by the 90% confidence interval (CI) for log-transformed ratios of Cl_{tot} of TEST group (patients from present study, VFL + CAPE) *versus* the REFERENCE group. Residual error was obtained from ANOVA. It was concluded to a non drug–drug interaction if the calculated 90%CI was within the acceptance range [0.80–1.25].

Similar approach was used for CAPE and 5-FU by comparing AUC_{last} between day 1 (CAPE + VFL) and day 14 (CAPE alone). Statistics were performed with SAS® software (SAS Institute Inc., USA) with a 5%-α risk.

Statistical analysis

The primary objective of the study was to analyse the DLTs during the first cycle of treatment in each DL and to determine the MTD. The secondary objectives were to determine the recommended DL for further clinical testing, the safety profile and the antitumour activity of the combination. For response rate, exact 95% confidence intervals (CLs) were provided.

Patients having received at least one cycle of study treatment were evaluable for MTD. For safety analysis, patient had to receive at least one study drug administration to be

Table 1 Patient characteristics

Characteristics	No of patient (%)
No of registered and treated patients	16
Age (years)	
Median	52.8
Range	32.3–74.6
WHO performance status	
0	10 (62.5)
1	5 (31.3)
2	1 (6.3)
Prior chemotherapy	
Neo/adjuvant only	4 (25.0)
Neo/adjuvant and advanced disease	11 (68.8)
Advanced disease only	1 (6.3)
Number of involved organs	
1	3 (18.8)
2	4 (25.0)
≥3	9 (56.3)
Visceral involvement	
Yes	10 (62.5)
No	6 (37.5)

Table 2 Number of patients and cycles per dose level

Dose level	Vinflunine (mg/m²)/Capecitabine (mg/m²/day)	No of patients	No of cycles
1	280/1,650	7*	53
2	320/1,650	3	20
3	280/2,000	6	33

* At least 6 patients to be enrolled at the RD as per protocol

evaluable, and for tumour response, patient assessed at least once after the second cycle were evaluable.

Results

Patient characteristics

Characteristics of the 16 patients are listed in Table 1. All the patients had received prior chemotherapy containing an anthracycline and a taxane: the majority (68.8%) underwent neo/adjuvant chemotherapy and first-line chemotherapy for advanced disease.

Drug delivery

A total of 106 cycles were administered with a median number of 8 cycles (range: 1–10). DLs are depicted in Table 2.

Table 3 DLTs during the first cycle per dose level

Dose level	No of pts with DLT/Nb of pts	Type of DLTs
1	0/7	–
2	2/3	G4 neutropenia ≥7 days G4 anorexia and G3 fatigue
3	2/6	G4 neutropenia ≥7 days and G3 constipation
		G3 diarrhoea, G3 fatigue,
		G3 anorexia and febrile neutropenia

Cycle delay occurred in 8 patients (50%) and 11% of cycles. It was observed at DL1 (57.1% of patients) and DL3 (66.7%). The main reason for cycle delay was neutropenia. The dose of vinflunine was reduced in 4 patients (25%) at DL2 and DL3. Reduction was due to the occurrence of grade 4 neutropenia lasting at least 7 days in 2 patients, grade 2 constipation lasting at least 5 days and grade 4 anorexia in one patient each.

Capecitabine was reduced in 3 patients, all of them were treated at the DL3 where capecitabine was given at 2,000 mg/m²/day. The main reason for capecitabine dose reduction was hand–foot skin reaction. Capecitabine was cancelled or missed in 181 of the 1,549 daily administrations given (11.7%).

Determination of the MTD

All the 16 patients enrolled were evaluable for the determination of the MTD. Table 3 describes the DLTs. Neutropenia was the main limiting toxicity of the combination at DL2 (VFL 320 mg/m² and CAPE 1,650 mg/m²/day) and at DL3 (VFL 280 mg/m² and CAPE 2,000 mg/m²/day).

Among the first 3 patients enrolled at DL1 (VFL 280 mg/m² and CAPE 1,650 mg/m²/day), no DLT was observed. As a consequence, dose escalation was continued to DL2 where the dose of vinflunine was increased from 280 to 320 mg/m² and the dose of capecitabine was similar to DL1 and then to DL3 where the dose of vinflunine was similar to DL1 and the dose of capecitabine was increased from 1,650 to 2,000 mg/m²/day. At DL2, two out of the 3 patients enrolled experienced a DLT, and at DL3, two out of 6 patients also had a DLT. Therefore, criteria for the MTD were met both at DL2 and DL3.

As per protocol, additional patients were enrolled at DL1 up to a total of 7 patients to confirm that DL1 is the RD. Since no DLT was observed in any of the 7 patients, DL1 combining vinflunine 280 mg/m² on day 1 and capecitabine 1,650 mg/m²/day from days 1 to 14 was considered to be the RD.

Table 4 Incidence of worst grade for haematological disorders per cycle and per dose level

Haematological toxicity	NCI CTC grading								
	DL1 (n = 53)			DL2 (n = 20)			DL3 (n = 33)		
	All grades N (%)	G3 N (%)	G4 N (%)	All grades N (%)	G3 N (%)	G4 N (%)	All grades N (%)	G3 N (%)	G4 N (%)
Anaemia	49 (92.5)	2 (3.8)	–	20 (100)	–	–	30 (90.9)	1 (3.0)	–
Neutropenia	40 (75.5)	11 (20.8)	2 (3.8)	18 (90.0)	4 (20.0)	6 (30.0)	30 (90.9)	9 (27.3)	13 (39.4)
Febrile neutropenia	–	–	–	–	–	–	1 (3.0)	1 (3.0)	–
Thrombocytopenia	14 (26.4)	–	–	12 (60.0)	–	–	14 (42.4)	–	–

Table 5 Incidence of worst grade for most common drug-related non-haematological adverse events per cycle and per dose level

Adverse Event	NCI CTC grading								
	DL1 (n = 53)			DL2 (n = 20)			DL3 (n = 33)		
	All grades N (%)	G3 N (%)	G4 N (%)	All grades N (%)	G3 N (%)	G4 N (%)	All grades N (%)	G3 N (%)	G4 N (%)
Abdominal pain	2 (3.8)	–	–	7 (35.0)	–	–	3 (9.1)	1 (3.0)	–
Constipation	9 (17)	–	–	10 (50.0)	–	–	7 (21.2)	1 (3.0)	–
Diarrhoea	16 (30.2)	–	–	6 (30.0)	–	–	2 (6.1)	1 (3.0)	–
Nausea	8 (15.1)	–	–	7 (35.0)	–	–	10 (30.3)	–	–
Stomatitis	6 (11.3)	–	–	1 (5.0)	–	–	7 (21.2)	–	–
Vomiting	7 (13.2)	–	–	2 (10.0)	–	–	5 (15.2)	–	–
Fatigue	16 (30.2)	–	–	17 (85.0)	3 (15.0)	–	13 (39.4)	1 (3.0)	1 (3.0)
Anorexia	5 (9.4)	–	–	5 (25.0)	–	1 (5.0)	5 (15.2)	1 (3.0)	–
Sensory neuropathy	1 (1.9)	–	–	3 (15.0)	–	–	4 (12.1)	–	–
Alopecia	16 (30.2)	NA	NA	3 (15.0)	NA	NA	8 (24.2)	NA	NA
Hand–foot syndrome	6 (11.3)	–	NA	–	–	NA	17 (51.5)	–	NA
Weight loss	13 (24.5)	–	–	8 (40.0)	–	–	14 (42.4)	–	–

Safety results

All patients were evaluable for safety analysis. The haematological disorders based on the analysis of blood tests are displayed by cycles and by DL in Table 4.

Neutropenia occurred in 15 patients (93.8%) with grade 3 and 4 in 7 patients (43.8%) each. At the RD (DL1), grade 3 and 4 events were reported in 20.8% and 3.8% of the cycles, respectively. Complications were rare: only one patient presented with febrile neutropenia at DL3.

The non-haematological adverse events are shown by cycle and by DL in Table 5. The most frequent events were gastrointestinal disorders and fatigue.

However, the incidences of grade 3 and 4 were low. It is noteworthy that no grade 3 or 4 events were reported at the RD (DL1). Drug-related constipation was seen in 11 patients (68.8%) and 24.5% of cycles with one episode of grade 3 reported at DL3. Drug-related diarrhoea was found in 8 patients (50%) and 22.6% of cycles with also one episode of grade 3 seen at DL3. Drug-related fatigue occurred in 14 patients (87.5%) and 43.4% of cycles; grade 3 episodes were reported in 3 patients (two at DL2 and one at DL3) and a single episode of grade 4 was seen at DL3. Grade 1 or 2 hand–foot syndrome was observed in 5 patients (31.3%) and 23% of cycles with a higher incidence reported at DL3 where the dose of capecitabine was 2,000 mg/m²/day. No toxic death occurred during study treatment.

Pharmacokinetics

Among the 16 patients included in the study, 14 were evaluable for PK at D1. Two patients were not evaluable: one due to thawed blood samples during shipment and the other because of missing administrations of CAPE in the days before PK assessment. On D14, eight patients were evaluable. Blood samples were missing in 4 patients. In two other patients, few CAPE administrations were skipped on the days before PK assessments.

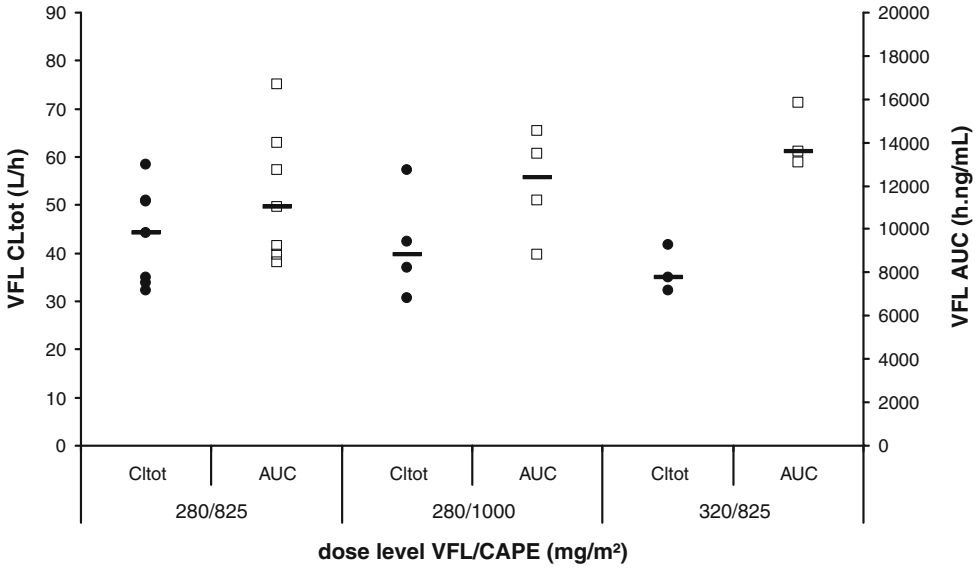

Fig. 1 Vinflunine (VFL) individual and median blood PK parameters (Cltot, *black circles* and AUCinf, open *squares*) according to each dose level

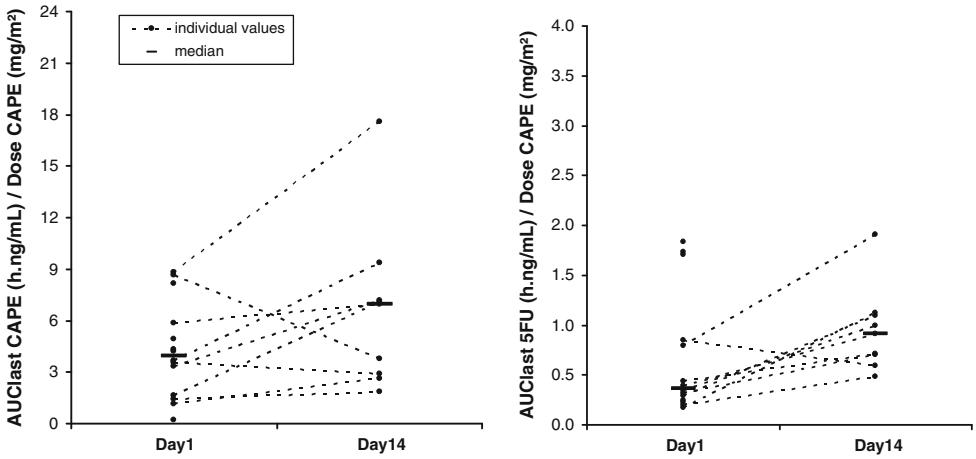

Fig. 2 Capecitabine (CAPE) and 5-FU AUClast values (h.ng/mL) normalised by CAPE dose levels (mg/m²) at day 1 (vinflunine + CAPE) and day 14 (CAPE alone)

Blood PK profiles of VFL were comparable between the three dose levels with a multi-exponential decay as previously described [3]. The median VFL AUC_{inf} was 11,043 and 12,406 h.ng/mL at 280 mg/m^2 (DL1 and DL3), and 13,599 at 320 mg/m^2 (DL2). Despite the limited number of patients, PK parameters indicated a dose-proportional exposure increase between 280 and 320 mg/m² with moderate inter-patient variability (coefficients of variation, CV in %, at 13–27%) (Fig. 1).

Therefore, all the dose levels were pooled in a same TEST group to be compared with the REFERENCE group. Concerning VFL, the ratio of Cl_{tot} geometric means (TEST/REFERENCE) was 0.99 and the 90%CI [0.88–1.11] confirmed the absence of difference between the two groups, demonstrating the non influence of CAPE on VFL clearance when both drugs were combined. This was further confirmed by the PK profile of DVFL, a metabolite formed through esterase hydrolysis. All the individual concentration values fell within the 95%CI of values of the REFERENCE group (data not shown) indicating the absence of interaction or impact of CAPE on this metabolism pathway. Concerning CAPE, the mean AUCs at D14 of both CAPE and 5-FU were higher than those at D1.

At DL1 and DL2 (825 mg/m^2), the mean CAPE AUC_{last} was 3,399 and 3,748 h.ng/mL at D1 ($n = 7$), as compared to 4,602 and 8,141 h.ng/mL at D14 ($n = 5$). For 5-FU, corresponding values were 322 and 362 h.ng/mL at D1 ($n = 3$), and 626 and 1,107 h.ng/mL at D14 ($n = 3$).

However, large inter-individual variability (CV up to 109%) with ranges that overlapped between D1 and D14, and large intra-individual variability (CV = 55%) were observed (Fig. 2). The 90%CI of the AUC last/Dose geometric mean ratio (D1/D14), for both CAPE [0.40–0.96] and 5-FU [0.28–0.58], was not within the regulatory range, indicating a non-equivalence of exposures between D1 and D14.

Antitumour activity

Responses were assessed by the investigator in all patients. One patient was not available for tumour response because she was prematurely discontinued for toxicity before the first tumour assessment scheduled at cycle 2. Partial responses were documented in 7 patients (43.8%, 95% CI [19.8–70.1]) and stable disease in 7 patients (43.8%). Responses were seen at the 3 DLs with 4 responses at the RD (DL1).

Discussion

Vinflunine single agent has shown significant antitumour activity as second-line or third-line treatment of anthracycline–taxane-pretreated patients with MBC [8, 14]. Vinflunine in association with other cytotoxics is currently investigated in MBC and in other indications: doxorubicin [34], gemcitabine [5] carboplatin [32] and cisplatin [28]. The present study tested VFL in association with capecitabine in MBC.

Capecitabine is an effective agent that is well tolerated in combination regimens. Clinical trials are now studying the potential of capecitabine-based combinations to provide new effective options for patients after failure of anthracyclines and taxanes. Additionally, capecitabine causes minimal myelo-suppression, making it particularly suited to combination with myelotoxic agents such as vinflunine.

The present phase 1 study investigated vinflunine in combination with capecitabine as second-line treatment in patients with MBC after failure of anthracycline- and taxane-based therapy. Vinflunine was initially tested at the dose of 280 mg/m² and then increased at 320 mg/m² which is the dose generally used in monotherapy. The starting dose of capecitabine was 1,650 mg/m²/day given from days 1 to 14.

This dose was then increased to 2,000 mg/m²/day which was shown to be manageable in combination with docetaxel [24] and ixabepilone [31].

The criteria for MTD were reached when vinflunine 320 mg/m² was associated with the starting dose of capecitabine 1,650 mg/m²/day (dose level 2) and again when vinflunine 280 mg/m² was combined with capecitabine 2,000 mg/m²/day (dose level 3). Therefore, vinflunine 280 mg/m² on day 1 plus capecitabine 1,650 mg/m²/day given from days 1 to 14 every 3 weeks (dose level 1) was considered to be the recommended dose.

Results of the present study have shown that the combination is safe and manageable. Neutropenia was the main limiting toxicity but the incidence of complications was low: a single patient experienced febrile neutropenia at dose level 3. The incidences of grade 3–4 non-haematological adverse events were also low with no grade 3 or 4 episode reported at the RD (dose level 1). This safety profile compares favourably with other capecitabine doublets. The combination of capecitabine with docetaxel was associated with a higher incidence of grade 3–4 gastrointestinal disorders (grade 3 stomatitis, diarrhoea and nausea were 17, 14 and 6%, respectively and grade 4 stomatitis, diarrhoea and nausea were 0,4, 0,4 and 0%, respectively) and grade 3 hand–foot syndrome (24%); however, the dose of capecitabine used was 2,500 mg/m²/day. Association of capecitabine 2,000 mg/m²/day and ixabepilone led to a high incidence of grade 3–4 sensory neuropathy (grade 3: 20% and grade 4: 1%).

In terms of antitumour activity, 7 objective responses were reported in the 16 treated patients, yielding a response rate of 43.8%. Despite the small number of patients enrolled in this phase I study, the activity of the combination looks promising. Similarly, encouraging results have been reported when capecitabine was combined with vinorelbine with response rates ranging from 49 to 50% in phase II studies conducted in patients pretreated with anthracyclines and taxanes [1, 11].

Concerning the search for drug–drug interaction, the suspected mechanism was competition/inhibition on esterases. Capecitabine is an orally administered fluoropyrimidine derivative, and the active moiety 5-FU is formed through 3 metabolism steps, the first one being controlled by liver carboxylesterases [22, 23, 27, 29]. Vinflunine circulates in blood mostly as parent compound and DVFL, its only active metabolite, is formed through various esterases [9].

Therefore, the mechanism most likely involved in the drug–drug interaction is a substrate competition on esterases.

The metabolism process should be affected, with a decrease in DVFL and/or 5-FU production(s), whereas parent(s) compound(s) is (are) increased. Current data indicate no impact on VFL metabolism. Regarding CAPE, exposures were higher at D14 (CAPE alone) than at D1 (CAPE + VFL). Of note, from the identified mechanism of drug–drug interaction, blood exposure of CAPE metabolites should have been increased and that of CAPE hydroxylated metabolite should have been decreased if a substrate competition on esterase activity had occurred between VFL and CAPE. However, the opposite was observed with an apparent lower exposure of parent compound at D1 (VFL + CAPE) *versus* D14 (CAPE alone). Consequently, current data are more likely would suggest an accumulation of CAPE and 5-FU over repeated administrations.

A time-dependant increase in 5-FU plasma concentrations is well established, as several independent studies reported mean increases in 5-FU plasma AUC of up to 60% [27] or even more [25], after 14 days of treatment, with no

expected clinical consequences [10, 15, 27, 33]. The literature data on CAPE accumulation are more controversial: some studies with CAPE given as a monotherapy or in combination with leucovorin, paclitaxel or docetaxel report no modification of CAPE AUC [21, 27], while a combination trial with cisplatin reported a slight CAPE AUC increase similar to our study, although the statistical significance was not reached [25]. The apparent increase in mean CAPE AUC reported here should thus be more likely attributed to the well-described PK variability of CAPE [27] and to the relatively low number of patients in our study.

Concerning the other theoretical mechanisms of drug–drug interaction between VFL and CAPE, they are very unlikely:

1. Absorption process is not involved because only CAPE is orally administered [26].
2. Binding of both compounds in plasma (including metabolites) is moderate (54% for CAPE [27] and 78% for VFL [4, 7]). Albumin is mostly involved for CAPE instead of lipoproteins for VFL.
3. Metabolism of CAPE goes through carboxyl esterases, cytidine deaminase, thymidine deaminase and then dihydropyrimidine dehydrogenase [22], whereas CYP3A4 is mostly involved for VFL except various esterases for one metabolite [9, 13].
4. Finally, elimination of CAPE is essentially in urines [27] whereas that of VFL is mainly in faeces [13].

In conclusion, the combination of vinflunine 280 mg/m² on day 1 and capecitabine 1,650 mg/m²/day from days 1 to 14 appears safe and effective in patients with metastatic breast cancer previously treated with anthracyclines and taxanes. Given the limited number of therapeutic options in this patient population, this combination warrants further clinical testing.

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