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Multicentric parallel phase II trial of the polo-like kinase 1 inhibitor BI 2536 in patients with advanced head and neck cancer, breast cancer, ovarian cancer, soft tissue sarcoma and melanoma. The first protocol of the European Organization for Research and Treatment of Cancer (EORTC) Network Of Core Institutes (NOCI)

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ABSTRACT

Aims: BI 2536 is a selective and potent small-molecule inhibitor of polo-like kinase 1. We performed a multi-centre, multi-tumour phase II trial to investigate the efficacy, safety and pharmacokinetics of BI 2536 in five solid tumour types.

Patients and methods: Patients with advanced head and neck, breast and ovarian cancer, soft tissue sarcoma and melanoma were selected according to protocol-defined general and tumour-specific criteria. They were ⩾18 years old, had a good performance status, adequate bone marrow, renal and liver function, measurable progressive disease and had completed other relevant systemic treatments >4 weeks ago. BI 2536 200–250 mg was given intravenously on day 1 every 3 weeks until intolerance, progression or refusal. The study was based on a Simon two-stage design, with 12 patients entering in stage 1 and additional 25 patients to be entered in case of at least one response in the first stage. The rate of objective responses (RECIST criteria) was chosen as primary end-point.

Results: Seventy six patients were included, 71 started treatment and received a median

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Head and neck cancer
Breast cancer
Ovarian cancer
Soft tissue sarcoma
Melanoma

number of two cycles (four in ovarian cancer). Frequent grade 3–4 adverse events were neutropaenia (81.6%), thrombocytopaenia (19.7%), febrile neutropaenia (19.7%), anaemia (15.5%) and pain (9.9%). We did not observe confirmed objective responses. All cohorts were closed after the entry of 14–15 eligible non-responding patients. Pharmacokinetic analyses revealed multi-compartmental behaviour and a rapid distribution of BI 2536.

Conclusions: BI 2536 showed limited antitumour activity according to the design of this trial in five different tumour types. Derivatives of BI 2536 with a more favourable pharmacological profile are currently explored further in prospective studies.

1. Introduction

In 1991, the *polo* gene, encoding for an enzyme present in Drosophila melanogaster, was discovered and cloned. In Drosophila, mutations in polo were found to induce abnormal mitoses. In 1993, Clay and colleagues identified and cloned gene sequences in murine haematopoietic progenitor cells encoding for a protein kinase that shares extensive homology with the enzyme encoded by Drosophila's *polo* gene. The mouse gene was called *Plk*. The protein encoded by *Plk*, polo-like kinase, belongs to a family of serine/threonine kinases that are involved in cell growth and differentiation in various species. In 1994, both human and murine cDNAs that were homologous to Drosophila's *polo* gene and the related kinase were cloned. The human counterpart was named *PLK*. Polo-like kinases (PLKs) play a key role in processes such as cell division and checkpoint regulation of mitosis. About 80% of human tumours express high levels of PLK transcripts, while PLK mRNA is mostly absent in surrounding healthy tissues (reviewed in [1]). Overexpression of PLK is associated with poor prognosis in several tumour types and a lower overall survival rate. The overexpression of PLKs in human tumours, but not in healthy non-dividing cells, makes them an attractive, selective target for anticancer drug development.[2,3]

PLK inhibitors interfere with different stages of mitosis such as centrosome maturation, spindle formation, chromosome separation and cytokinesis. They induce mitotic chaos and severely perturb cell cycle progression ('Polo arrest'), eventually leading to cancer cell death. PLK1, the most extensively characterised member of the polo family, controls critical steps in the passage of cells through the M phase of the cell cycle, including initiation of entry into mitosis, centrosome separation necessary for the formation of a bipolar mitotic spindle, metaphase to anaphase transition and mitotic exit and onset of cell division (Fig. 1A).[1] PLK inhibitors act on the mitotic spindle in a completely different manner than the established anticancer agents such as vinca alkaloids or taxanes, which directly bind to structural components of the spindle, but do not interfere with the regulation of the mitotic process (Fig. 1B).

BI 2536 is a highly selective and potent small molecule PLK1 inhibitor.[4] The free base (BI 2536 BS) is used as an intravenous formulation. The molecular potency of BI 2536 is in the low nanomolar range. More than 45 kinases tested in parallel with PLK1 were not inhibited. In a panel of tumour cell lines, activity was not dependent on cellular origin or molecular phenotype. Three clinical phase I dose-ranging studies have been performed with BI 2536, testing four different 3-weekly schedules.[5–7] BI 2536 was administered either on day 1, days 1 and 8, days 1, 2 and 3 or as a 24-h infusion on day 1. BI 2536 was generally well tolerated, with a MTD defined in two independent phase I studies as once-weekly 200 mg/cycle or twice-weekly 100 mg/cycle. The dose-limiting toxicities (DLTs) were reversible and non-cumulative neutropaenia and thrombocytopaenia, as expected from preclinical testing of this compound. Other related adverse events were of mild to moderate intensity. BI 2536 had a favourable, dose-dependent safety profile. A pharmacokinetic (PK) analysis showed dose-proportional maximum plasma concentrations and total exposure, with a terminal elimination half-life of about 17–34 h. BI 2536 had a high total clearance from plasma and a high volume of distribution. Hints of antitumour activity were observed in the heavily pre-treated patient population involved in the phase I programme, providing the rationale for further clinical development of BI 2536 in various solid tumour indications.

The aim of the current multi-tumour phase II protocol was to screen for activity and evaluate the safety of BI 2536 in patients with various well-defined advanced cancers. The day 1 every 3 week schedule was chosen as a safe, feasible and tolerable regimen. The primary end-point was to assess the confirmed objective response (OR) rate (complete and partial responses) as defined by RECIST.[8] The study also assessed the duration of response in responding patients, progression-free survival and overall survival. The safety was documented by CTCAE version 3.0. The objective of the PK analysis was to describe the plasma concentration time-course following administration of a single administration of BI 2536.

2. Patients and methods

2.1. Trial design

This trial was performed as a set of parallel phase II investigations in five different solid tumours combined in one protocol. Each of the phase II cohorts had a similar design with OR as the primary end-point. The trial was a multi-centre, open-label, non-randomised study. Patients were registered at the EORTC Headquarters prior to the start of treatment after signing of informed consent and verification of eligibility. The study included a screening visit, consecutive study assessments and a post-treatment follow-up visit. During the

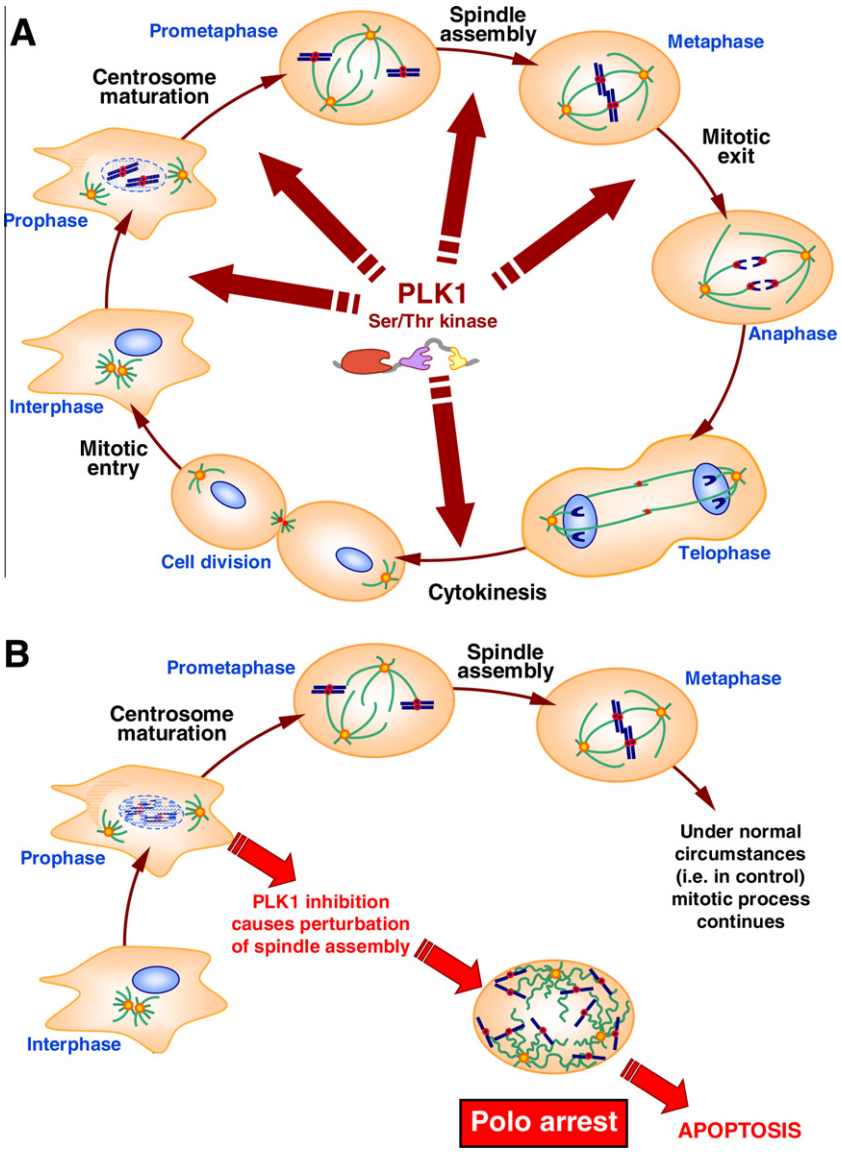

Fig. 1 – Physiological function of PLK1 (A) and cellular effects of PLK1 inhibition (B).

treatment phase, the patients underwent regular assessments for safety, clinical response and PK. Antitumour activity was assessed every other course until progression, and adverse events were assessed separately for each course of therapy. The Simon two-stage design was applied to each stratum: 12 or 37 eligible patients were to be registered for each tumour type, according to the number of responses observed among the first 12 eligible and evaluable patients.

2.2. *Patient selection*

The study protocol-defined general and tumour-specific eligibility criteria. All patients had measurable disease. Administration of any prior systemic treatment for the malignancy (including chemotherapy, radiotherapy, immunotherapy, hormonal therapy, treatment with monoclonal antibodies, tyrosine kinase inhibitors and other agents) had been completed for at least 4 weeks before study entry. Patients were at least 18 years old, had an ECOG performance status of 0-2 and adequate haematological function (ANC $\geqslant 1.5 \times 10^9$/l, platelets $\geqslant 100 \times 10^9$/l and haemoglobin $\geqslant 9$ mg/dl). Serum creatinine was $\leqslant 175$ μmol/l, bilirubin $\leqslant 1.5$ times ULN, AST/ALT $\leqslant 2.5 \times$ ULN in the absence of liver metastases and $\leqslant 5 \times$ UNL in case of liver metastases. Patients had no persistent toxicities from prior therapy and had progressive disease within the past 6 months prior to study entry according to RECIST. Written informed consent was obtained according to ICH/GCP and national/local regulations. The trial was approved by the responsible committees in all involved institutions.

Patients with head and neck cancer were entered if they had histologically or cytologically proven squamous cell carcinoma (excluding nasopharyngeal primaries), had distant metastasis or had disease recurrence with new non-irradiated lesions in pre-irradiated field as target lesions. They had no prior chemotherapy for recurrent or metastatic

disease. Breast cancer patients enrolled in this trial had histologically proven recurrent or metastatic adenocarcinoma and had failed prior taxane and anthracycline therapy, with one or two lines of chemotherapy given either as adjuvant treatment or for recurrent/metastatic disease. They did not qualify for Her-2 based therapy. In the ovarian cancer cohort, patients had histologically proven epithelial ovarian cancer, metastatic or inoperable locally advanced disease and were either progressing under or relapsing within 6 months of completing any line of platinum and taxane-based therapeutic regimen for advanced disease. Sarcoma patients qualified for this study if they had histologically proven advanced and/or metastatic malignant soft tissue sarcoma of high or intermediate grade. Patients were excluded if they had embryonal rhabdomyosarcoma, chondrosarcoma, osteosarcoma, Ewing sarcoma, gastrointestinal stromal tumours, dermatofibrosarcoma protuberans, inflammatory myofibroblastic sarcoma, neuroblastoma, malignant mesothelioma or mixed mesodermal tumours of the uterus. Patients had received no more than one combination or two single agents of chemotherapy for advanced disease and treatment had included an anthracycline if not medically contra-indicated. Patients with histologically proven metastatic malignant melanoma qualified if the primary site was non-ocular, if they had recurrent/metastatic disease and had received no more than one line of chemotherapy pending LDH $\leqslant 2 \times$ ULN. One prior line of previous immunotherapy was allowed.

The patient population was not selected on the basis of biological markers. PLK expression in tumour tissue was not assessed systematically.

2.3. Treatment

BI 2536 was given as a peripheral or central intravenous (i.v.) 1 h infusion at a flat dose of 200 mg on day 1 every 21 days, which was defined as one treatment cycle. One dose escalation to 250 mg after course 1 was allowed, in case they did not encounter drug-related CTCAE grade 3 or greater haematological toxicity or grade 2 or greater non-haematological toxicity (excluding fatigue, adequately controlled nausea/vomiting and alopecia). The drug was provided as 2 mg/ml ready to use solution for infusion in glass containers. The medication was filled in plastic bags pre-filled with 500 ml sodium chloride. The treatment was given until disease progression, intercurrent illness that prevented further drug administration, drug-related adverse event considered by the investigator to warrant permanent discontinuation of study drug, drug-related CTCAE grade 3 or greater non-haematological toxicity (excluding untreated nausea, vomiting or diarrhoea), a second episode of any drug-related adverse reaction necessitating another dose reduction or withdrawal or non-compliance of the patient.

The dose of BI 2536 could be reduced by 50 mg once during any treatment period according to protocol-defined criteria. Once the dose was reduced, it was not increased again during the further course of treatment. Treatment for subsequent cycles was given when the absolute neutrophil count (ANC) was $\geqslant 1.0 \times 10^9$/l, platelets were $\geqslant 75 \times 10^9$/l and all adverse drug reactions of the previous course had recovered to $\leqslant$grade 2. If treatment had to be postponed, the first day of treatment was considered day 1 of the next course. If treatment was delayed for more than 1 week due to drug-related adverse events the patient was withdrawn from the protocol treatment.

Concomitant medication was given at the discretion of the investigator. Prophylactic anti-emetics were not prescribed during course 1, but could be used as of cycle 2. Growth factors with short half-life could be used according to local practice for the treatment of haematotoxicity after the first treatment with BI 2536. Treatment with corticosteroids and bisphosphonates was allowed as long as the treatment started before entry into the study and the used dose was stable. Concomitant treatment with agents metabolised by CYP2C9 was avoided whenever possible. Palliative radiotherapy could be given during the study for bone pain or for other reasons for symptom relief. The irradiated area was not used for response assessment. Other anticancer agents or investigational drugs were not permitted during the conduct of the trial.

2.4. Investigations

The initial work-up of trial candidates included informed consent, medical history, concomitant therapy, physical examinations, vital signs and demographics (age, gender, ECOG performance status, vital signs, body weight, height), cancer signs and symptoms, 12-lead ECG, complete blood counts (red blood cells, haemoglobin, haematocrit, total white blood cells with ANC and platelets), serum chemistry (sodium, potassium, calcium, creatinine, urea, total bilirubin, AST, ALT, ALP, LDH, albumin, total protein, creatin phosphokinase, tumour markers and pregnancy test if appropriate) and tumour evaluation according to RECIST (X-rays, CT scans, MRI scans, measurement of clinical lesions). Blood counts were repeated at least weekly. Each new treatment cycle was preceded by assessment of the performance status, physical examination, blood pressure, vital signs, cancer symptoms, assessment of adverse events that had occurred since the previous visit, complete blood counts and serum chemistry including tumour markers. PK sampling was performed in all patients at least during course 1 and 3 according to protocol-specific instructions. The end of treatment evaluation included performance status, physical examination, vital signs, body weight, assessment of cancer symptoms and residual adverse events, ECG, complete blood counts, serum chemistry and disease assessment. Objective response, measured according to RECIST, was used as the principal end-point in this trial. Imaging was done at pre-baseline (for documentation of progressive disease), at baseline and after every other treatment course. For assessment of adverse events the Common Terminology criteria for Adverse Events (CTCAE) version 3.0 were applied (http://ctep.info.nih.gov/CTC3/default.htm).

2.5. Statistical design

This non-randomised multicentric parallel phase II trial of BI 2536 was performed in five cohorts of solid tumours. A separate phase II design was applied to each of the solid tumour strata. In each tumour type a Simon optimal two-stage design for confirmed objective response was used.[9] The type I error was 10%. For the null hypothesis of a 5% response rate and an alternative hypothesis of a 20% response rate, in stage I,

12 eligible patients were to be treated in each cohort. An additional 25 eligible patients were to be enrolled per tumour type where one or more responses were confirmed in stage I. If no responses were seen in the first 12 eligible patients, the stratum was closed for accrual. If the true response rate was 5%, the probability of early termination was 54%. On completion of stage II, the observation of at least four objective responses was considered sufficient to reject the null hypothesis. This design yields 90% power. Patients who dropped out due to early clinical progression at the end of course 1 were replaced. Secondary end-points included progression-free survival, clinical benefit rate defined as the fraction of eligible treated patients responding or documented as stable disease according to RECIST, overall survival, duration of response, adverse events and pharmacokinetics. Progression-free survival and overall survival were estimated by the Kaplan-Meier method in each stratum. Greenwood's variance estimate (after log–log transformation) was used to calculate confidence intervals. We did not perform formal comparisons between the different independent strata of the trial. In this report the efficacy data are summarised on the dataset of eligible patients. Baseline characteristics, dosing and safety data are summarised in the dataset of treated patients.

2.6. Pharmacokinetics

The objective of the PK analysis was to describe the plasma concentration time-course following administration of BI 2536. Plasma concentrations were assessed at courses 1 and 3 and optional in one additional course. The actual blood sampling times and the duration of the BI 2536 infusion (start and end time of infusion) were documented. For quantification of analyte plasma concentrations, 2.5–3.0 ml of blood was taken from a forearm vein in an EDTA anticoagulant blood drawing tube. The detection limit for BI 2536 in plasma was 0.5 ng/ml. Plasma samples were stored at −20 °C and shipped in bulk to the Department of Drug Metabolism and Pharmacokinetics/Bioanalytics, Boehringer Ingelheim Pharma GmbH & Co KG, Biberach, Germany. Analyte concentrations were determined by a validated high performance liquid chromatography tandem mass spectrometry assay at Boehringer Ingelheim[data on file].

3. Results

3.1. Patient characteristics

Of the 71 patients treated in this trial, 27 (38.0%) were male and 44 (62.0%) were female. The median age was 58 years and their median weight was 67 kg. Demographic data are summarised in Table 1. Sixty-one patients (85.9%) had previously undergone surgery for their tumour. The most common therapy combinations comprised surgery with chemotherapy (25 patients; 35.2%); surgery with chemotherapy, radiotherapy and other therapies (13 patients; 18.3%); and surgery with chemotherapy and radiotherapy (11 patients; 15.5%); each of the other combination regimens had been used in <10% of patients. Upon review, 17 of 76 (22.4%) patients included in this trial had some violation of eligibility criteria, of which five never started treatment with the study drug. These included six cases of insufficient documentation of prior progression and three cases of insufficient delay after prior treatment. The other reasons were need for local radiotherapy, previously irradiated indicator lesions, laboratory data violation (two cases), regression of metastases at screening, prior treatment that was not allowed (two cases) and start of treatment prior to registration through EORTC.

3.2. Treatment compliance

A total of 71 patients received at least one dose of study medication. The median actual dose intensity was 66.7 mg BI 2536/week, consistent with the planned dose intensity (66.7 mg/week). Most patients (46 patients; 64.8%) received the study treatment via the central venous route, for 17 patients (23.9%) peripheral venous access was used and for the remaining 8 patients (11.3%) both routes were used. Of the 65 patients who received more than one treatment course 54 (83.1%) underwent treatment without a delay in administration, 6 patients (9.2%) required a 4–7 d delay, 3 patients (4.6%) required an 8–14 d delay and 2 patients (3.1%) required a delay of more than 14 days. Of the 266 treatment courses received by patients, 13 courses (4.9%) required dose delay. Only one course was delayed due to a drug-related event. A total of 195 courses were administered after cycle 1

Table 1 – Patient characteristics.

	Tumour type					
	Head and neck cancer (N = 14) N (%)	Breast cancer (N = 14) N (%)	Ovarian cancer (N = 15) N (%)	Soft tissue sarcoma (N = 14) N (%)	Melanoma (N = 14) N (%)	Total (N = 71) N (%)
Age (years): median	59.4	57	64	57.4	56.7	57.7
Age (years): <50/50–59/60–69/⩾70	2/5/7/0	2/6/3/3	0/7/4/4	3/5/4/2	5/4/4/1	12/27/22/10
Sex: male/female	13/1	0/14	0/15	8/6	6/8	27/44
Performance status 0/1/2	2/11/1	8/6/0	6/5/4	7/7/0	8/5/1	31/34/6
Prior treatment						
Surgery	7 (50.0)	13 (92.9)	15 (100.0)	14 (100.0)	12 (85.7)	61 (85.9)
Chemotherapy	7 (50.0)	14 (100.0)	15 (100.0)	14 (100.0)	12 (85.7)	62 (87.3)
Radiotherapy	14 (100.0)	14 (100.0)	3 (20.0)	5 (35.7)	2 (14.3)	38 (53.5)
Hormonotherapy	0 (0.0)	11 (78.6)	2 (13.3)	2 (14.3)	0 (0.0)	15 (21.1)

(courses 2–23). Of these 13 cycles (6.7%) required a treatment delay, eight courses (4.1%) were administered after a 4–7 d delay, three courses (1.5%) were administered after an 8–14 d delay and two courses (1.1%) were administered after a delay of more than 14 days. A total of 16 patients (22.5%) underwent dose escalation at treatment course 2, as specified by the protocol.

3.3. Objective response and clinical benefit

The primary end-point of the trial was to document the OR as defined by RECIST. No patient experienced a complete or partial response. Accordingly duration of response was not assessed. The best response was stable disease, which was experienced by 18 patients (30.5%) based on the investigators assessment and by 25 patients (42.4%) based on the coordinating investigator's review of patient data. Overall we observed differences in the treatment outcome evaluations when comparing the investigator's assessment with the coordinating investigator's assessment in 8 patients. The highest rate of stable disease was seen in ovarian cancer. The response to treatment is summarised in Table 2. We did not observe clinically meaningful tumour marker responses. Clinical benefit was defined as the number of patients experiencing at least stable disease according to RECIST. Clinical benefit was documented in 25 patients (42.4%) (95% confidence interval (CI) 29.6%, 55.9%): head and neck cancer 3 (30.0%), breast cancer 5 (45.5%), ovarian cancer 10 (76.9%), soft tissue sarcoma 2 (15.4%) and melanoma 5 (41.7%).

3.4. Progression-free and overall survival

The median progression-free survival was 1.4 (95% CI 1.4, 2.5) months, with 3.5% of patients (95% CI 0.64%, 10.6%) estimated as being progression-free at 6 months after the start of treatment. The median overall survival was 9.5 (95% CI 6.2, 11.8) months, with 63.8% of patients (95% CI 50.1%, 74.7%) estimated as surviving at 6 months and 34.8% (95% CI 21.9%, 48.1%) as surviving 1 year after the start of treatment.

3.5. Adverse events, serious adverse events and deaths

Irrespective of causality, the most frequently reported clinical AEs during treatment were pain (88.7%) and fatigue (70.4%). The most common grade $\geqslant 3$ clinical AEs was febrile neutropaenia (19.7%). Grade 4 clinical AEs seen during the trial comprised febrile neutropaenia (4.2%) and dyspnoea (1.4%). The most frequently reported drug-related AEs observed during the trial were fatigue (39.4%), alopecia (26.8%) and febrile neutropaenia (19.7%; Tables 3a and 3b). A total of 41 patients died during or after the study, with 39 deaths being due to progressive disease and one death being due to drug-related shock. In addition, for one patient death was attributed to bleeding from brain metastases. In total, 30 patients (42.3%) experienced an SAE during or after treatment. Study drug-related SAEs comprised febrile neutropaenia/neutropenic fever (9.9%), fever (four reports in 3 patients; 5.6%), thrombocytopaenia and anaemia (each 4.2%), shock (2.8%) and abdominal abscess (1.4%). Overall, the most frequently observed grade $\geqslant 3$ laboratory abnormalities seen during treatment were neutropaenia (58 patients; 81.7%), leucopaenia (48 patients; 67.6%), thrombocytopaenia (14 patients; 19.7%) and anaemia (11 patients; 15.5%). CTC grade 3 clinical laboratory abnormalities were furthermore observed sporadically for hyponatraemia, -calcaemia, -kaliaemia, increased alkaline phosphatase and decreased albumin. A total of 7 patients (9.9%) had ECG findings classified as abnormal but not clinically relevant and one patient (1.4%) had findings classified as abnormal and clinically relevant. The latter patient experienced sinus bradycardia. Patients with non-clinically relevant ECG abnormalities included two patients with sinus arrhythmia and one patient with ventricular fibrillation, sinus tachycardia and left bundle branch block.

Table 2 – Efficacy of treatment with BI 2536 and survival outcomes.

	Tumour type					
	Head and neck cancer (N = 10) N (%)	Breast cancer (N = 11) N (%)	Ovarian cancer (N = 13) N (%)	Soft tissue sarcoma (N = 13) N (%)	Melanoma (N = 12) N (%)	Total (N = 59) N (%)
Response to treatment evaluated by the study coordinator[a]						
Stable disease	3 (30.0)	5 (45.5)	10 (76.9)	2 (15.4)	5 (41.7)	25 (42.4)
Progressive disease	5 (50.0)	6 (54.5)	2 (15.4)	11 (84.6)	6 (50.0)	30 (50.8)
Early death malignant disease					1 (8.3)	1 (1.7)
Early death toxicity	1 (10.0)					1 (1.7)
Not assessable	1 (10.0)		1 (7.7)			2 (3.4)
Time related end-points						
Median progression-free survival (month, 95% confidence interval)	1.15 (1.12, 4.14)	1.38 (1.28, 5.39)	2.76 (2.07, 4.11)	1.35 (1.28, 1.64)	1.30 (1.15, 2.53)	1.41 (1.35, 2.53)
Median overall survival (month, 95% confidence interval)	6.41 (4.90, 10.81)	13.27 (5.29, NA)	8.18 (5.72, 14.26)	11.83 (10.05, 15.28)	5.36 (4.24, 9.59)	9.53 (6.18, 11.83)

[a] This table is performed on the dataset of eligible patients.

Table 3a – Incidence of adverse events occurring in >10% of the treated patients.

Event	Incidence of adverse events occurring in >10% of treated population					
	Head and neck cancer N (%)	Breast cancer N (%)	Ovarian cancer N (%)	Soft tissue sarcoma N (%)	Melanoma N (%)	Total N (%)
Number of patients, N (%)	14 (100.0)	14 (100.0)	15 (100.0)	14 (100.0)	14 (100.0)	71 (100.0)
Patients with an AE	14 (100.0)	14 (100.0)	15 (100.0)	14 (100.0)	14 (100.0)	71 (100.0)
Pain	11 (78.6)	11 (78.6)	14 (93.3)	13 (92.9)	14 (100.0)	63 (88.7)
Fatigue	8 (57.1)	9 (64.3)	13 (86.7)	11 (78.6)	9 (64.3)	50 (70.4)
Other toxicity	5 (35.7)	4 (28.6)	12 (80.0)	3 (21.4)	5 (35.7)	29 (40.8)
Anorexia	4 (28.6)	4 (28.6)	7 (46.7)	3 (21.4)	5 (35.7)	23 (32.4)
Hair loss/alopecia	1 (7.1)	5 (35.7)	6 (40.0)	4 (28.6)	6 (42.9)	22 (31.0)
Other gastrointestinal	5 (35.7)	3 (21.4)	7 (46.7)	5 (35.7)	2 (14.3)	22 (31.0)
Constipation	2 (14.3)	2 (14.3)	8 (53.3)	5 (35.7)	3 (21.4)	20 (28.2)
Diarrhoea	2 (14.3)	2 (14.3)	7 (46.7)	6 (42.9)	3 (21.4)	20 (28.2)
Dyspnoea	2 (14.3)	3 (21.4)	5 (33.3)	1 (7.1)	7 (50.0)	18 (25.4)
Fever	3 (21.4)	4 (28.6)	5 (33.3)	4 (28.6)	2 (14.3)	18 (25.4)
Oedema	4 (28.6)	5 (35.7)	4 (26.7)	1 (7.1)	4 (28.6)	18 (25.4)
Nausea	4 (28.6)	3 (21.4)	3 (20.0)	3 (21.4)	4 (28.6)	17 (23.9)
Other pulmonary/upper respiratory	4 (28.6)	1 (7.1)	4 (26.7)	6 (42.9)	2 (14.3)	17 (23.9)
Cough	4 (28.6)	2 (14.3)	2 (13.3)	2 (14.3)	5 (35.7)	15 (21.1)
Other dermatology/skin	3 (21.4)	1 (7.1)	7 (46.7)		4 (28.6)	15 (21.1)
Other neurology	3 (21.4)	5 (35.7)	2 (13.3)	2 (14.3)	3 (21.4)	15 (21.1)
Febrile neutropaenia	2 (14.3)	1 (7.1)	5 (33.3)	3 (21.4)	3 (21.4)	14 (19.7)
Infection	4 (28.6)	3 (21.4)	4 (26.7)	2 (14.3)	1 (7.1)	14 (19.7)
Other constitutional symptoms	5 (35.7)	2 (14.3)	2 (13.3)	2 (14.3)	3 (21.4)	14 (19.7)
Sensory neuropathy	1 (7.1)	3 (21.4)	2 (13.3)	1 (7.1)	6 (42.9)	13 (18.3)
Vomiting	1 (7.1)		6 (40.0)	3 (21.4)	2 (14.3)	12 (16.9)
Weight loss	3 (21.4)	3 (21.4)	2 (13.3)	1 (7.1)	2 (14.3)	11 (15.5)
Mucositis	1 (7.1)	3 (21.4)	1 (6.7)	3 (21.4)	2 (14.3)	10 (14.1)
Other haemorrhage/bleeding	4 (28.6)	2 (14.3)	2 (13.3)		2 (14.3)	10 (14.1)
Other pain		2 (14.3)	4 (26.7)	1 (7.1)	2 (14.3)	9 (12.7)
Dizziness		1 (7.1)	4 (26.7)	1 (7.1)	2 (14.3)	8 (11.3)
Pruritis/itching	1 (7.1)	2 (14.3)	2 (13.3)	1 (7.1)	2 (14.3)	8 (11.3)

Table 3b – Frequency of patients with drug-related adverse events occurring in two or more patients, for all treated patients.

Event	Grade 3/4 (N = 71) N (%)	Any related event (N = 71) N (%)
Number of patients with any type of event	17 (23.9)	51 (71.8)
Fatigue	2 (2.8)	28 (39.4)
Hair loss/alopecia	–	19 (26.8)
Febrile neutropaenia	2 (2.8)	14 (19.7)
Anorexia	–	10 (14.1)
Other toxicity	6 (8.5)	10 (14.1)
Diarrhoea	–	9 12.7)
Nausea	–	9 (12.7)
Mucositis	–	8 (11.3)
Pain	–	5 (7.0)
Vomiting	–	5 (7.0)
Pruritis/itching	–	4 (5.6)
Other dermatology/skin	–	3 (4.2)
Other gastrointestinal	–	3 (4.2)
Rash/desquamation	–	3 (4.2)
Constipation	–	2 (2.8)
Dyspnoea	–	2 (2.8)
Fever	–	2 (2.8)
Other Haemorrhage/bleeding	1 (1.4)	2 (2.8)
Other neurology	–	2 (2.8)
Weight loss	–	2 (2.8)
Injection site reaction/extravasation changes	–	2 (2.8)

3.6. *Pharmacokinetics*

BI 2536 exhibited multi-compartmental PK behaviour. Plasma concentrations increased up to 0.5 h or 1 h after the start of infusion. After the end of the infusion concentrations decreased rapidly indicating rapid distribution. By 24 h after drug administration, the average BI 2536 plasma concentration was 24 ng/ml, which is less than 3.8% of the average maximum plasma concentration of 636 ng/ml. By 120 h after dose administration (last sampling time), the plasma concentration had decreased further to about 1.85 ng/ml, which is less than 0.5% of the maximum plasma concentration. The inter-patient variability of plasma concentrations was moderate (gCV: 54.0–75.3%). Low BI 2536 plasma concentrations were detected in some patients before dose administration in each treatment cycle indicating that BI 2536 did not accumulate. The plasma concentration time profiles of BI 2536 after infusion of 200 mg BI 2536 are illustrated in Fig. 2.

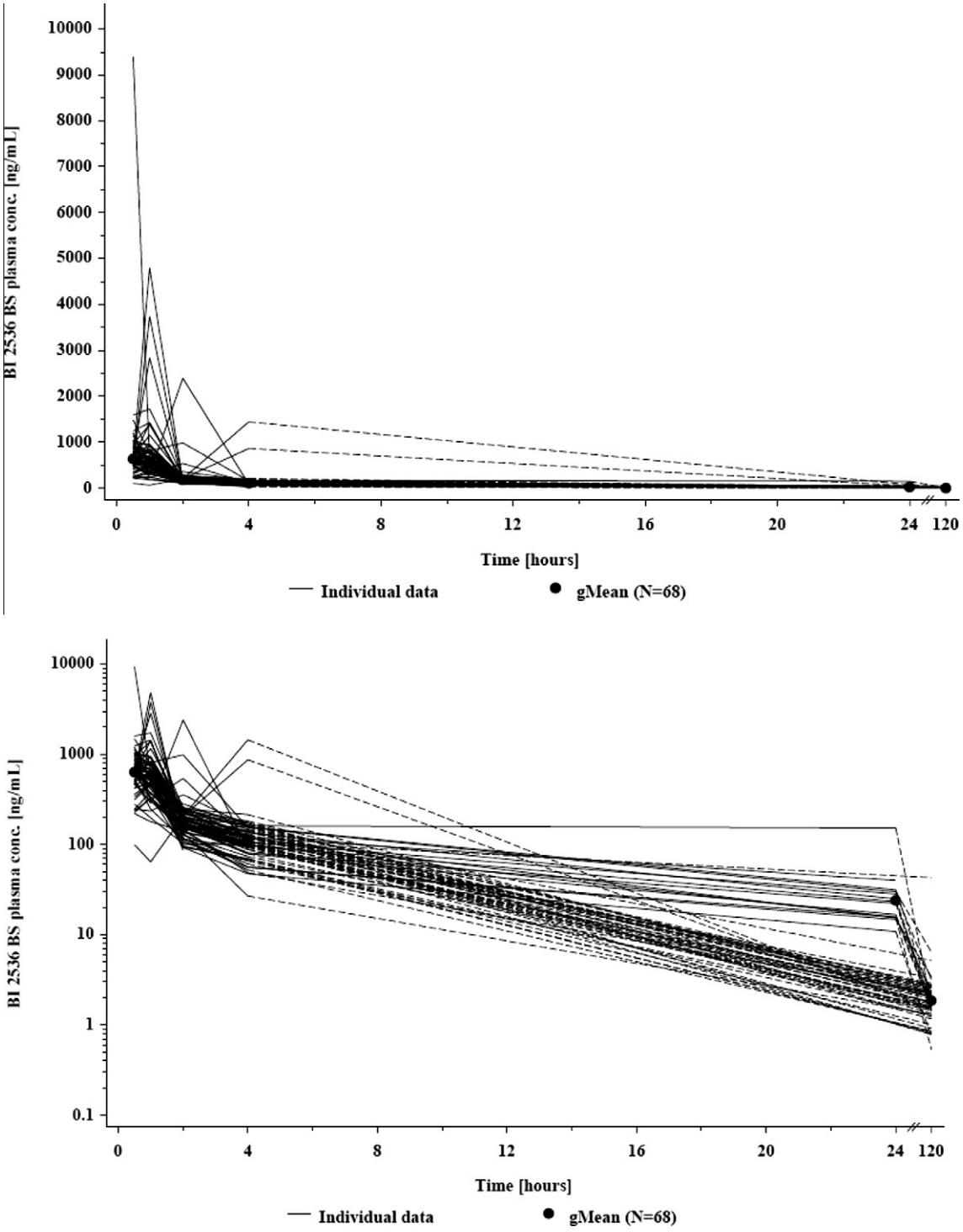

Fig. 2 – Drug plasma concentration time profiles of BI 2536 BS after 1 h i.v. infusion of 200 mg BI 2536 on day 1 of cycle 1.

4. Discussion

This study aimed to assess the efficacy of BI 2536 in five parallel patient cohorts, to characterise antitumour activity of the novel compound in head and neck cancer, breast cancer and ovarian cancer, soft tissue sarcoma and melanoma. The end-point was the determination of the objective response rate, but none of the first 14–15 evaluable patients per tumour type experienced a complete or partial response during the conduct of the trial. Consequently, all strata were closed after completion of stage 1. Of note is that individual patients experienced disease stabilisation for up to 23 treatment cycles, and all patients were progressing according to RECIST criteria at baseline. When assessed according to stringent criteria, 42.4% of patients experienced clinical benefit (95% CI 29.6–55.9%), but the median progression-free survival was only 1.4 months (95% CI 1.4, 2.5) and the median overall survival 9.5 months (95% CI 6.2, 11.8).

BI 2536 exhibited multi-compartmental PK behaviour. The plasma concentrations of BI 2536 increased during the infusion period. After the end of infusion BI 2536 showed a rapid disposition phase. The inter-patient variability was moderate, and the drug did not accumulate following repeated infusions. The PK characteristics identified were consistent with those reported in phase I trials.[5–7]

BI 2536 was easy to administer, and the schedule used was very patient friendly. The safety profile was consistent with the phase I experience, as the most frequently observed drug-related AEs were fatigue (39.4%), alopecia (26.8%) and febrile neutropaenia (19.7%). Our study confirms that BI 2536 has an acceptable safety profile for the treatment of patients with advanced solid tumours, as shown in the previous phase I and a limited number of phase II studies with this compound.

So far only three phase II studies have been reported using BI 2536 in patients with solid tumours, including patients with advanced NSCLC, pancreatic cancer and hormone-refractory prostate cancer (HRPC). These studies were using different 3-week schedules. In the NSCLC study, the efficacy, safety and PK of two dosing schedules of BI 2536 were investigated.[10] Patients were randomised to receive BI 2536 either on day 1 (200 mg) or on days 1–3 ($3 \times 50\ mg^{-3} \times 60$ mg) of 21-day treatment courses. Partial response and stable disease were observed in 54% of the patients. The progression-free survival and overall survival were 58 days (95% CI, 48–85) and 189 days (95% CI, 176–304; 47 patients censored). Grade 4 neutropaenia occurred in 36% of patients and two patients died due to sepsis and pulmonary haemorrhage. Other common adverse events, which were generally mild, included fatigue and nausea.

In the HRPC study, patients were treated with 200 mg BI 2536 (given as a 1-h i.v. infusion on day 1 every 3 weeks) and the dose was escalated to 250 mg from cycle 2 onward in patients with non-haematological toxicities of grade <2 and haematological toxicities of grade <3 in severity.[11] Six patients had stable disease, with one patient achieving a 25% prostate-specific antigen reduction. BI 2536 was generally well tolerated, with neutropaenia being the main AE. Grade 3 or 4 neutropaenia was observed in 20% of patients at a dose of 200 mg, and in 73% of patients after dose escalation to 250 mg.

In a study chemotherapy-naïve patients with advanced pancreatic cancer, patients randomly received either BI 2536 at a dose of 200 mg i.v. every 21 days or BI 2536 at a dose of 60 mg i.v. for 3 days every 21 days.[12] The incidences of AEs were similar in the two arms, with the most frequently reported events being fatigue (49%), nausea (42%) and neutropaenia (36%). Grade 3 or 4 adverse events were mainly haematological. The 1-year survival of 18% was similar to that often reported for gemcitabine in earlier trials.

Other phase I/II studies with BI 2536 include a completed trial in patients with non-Hodgkin's lymphoma, an ongoing study in patients with relapsed or refractory acute myeloid leukaemia and a small-cell lung cancer study that is currently under analysis[Boehringer Ingelheim, data on file].

The phase I/II evaluation of BI 2536 in solid tumours and haematological malignancies was accompanied by a dose escalation trial with a related back-up compound called BI 6727.[13,14] This drug is also a highly potent and selective inhibitor of PLK1, a second generation dihydropteridinone derivative with distinct PK properties as compared to BI 2536. A total of 65 patients have been treated with BI 6727 at doses from 12 to 450 mg. Reversible haematological toxicity was the main side-effect, neutropaenia and febrile neutropaenia were dose limiting. The dose of 300 mg was chosen for further development considering the overall safety of the compound. PK results indicated linearity in the therapeutic dose range, a large volume of distribution, moderate clearance and a long half-life around 110 h. Confirmed partial responses were observed in pre-treated patients with advanced urothelial cancer, ovarian cancer and metastatic melanoma. Stable disease as best response was reported in another 48% of patients.

Due to the favourable results with BI 6727 this drug was chosen for further clinical development, and is currently investigated in a number of combination phase I protocols and phase II studies. In addition, PLK inhibitors from other drug classes are explored in a number of early clinical trials.[15–22]

Conflict of interest statement

Patrick Schöffski: Advisory Board Member Boehringer Ingelheim Pharma; Jacques De Grève: unrestricted research grant 2009 Boehringer Ingelheim; Gerd Munzert, Holger Fritsch, Gertraud Hanft: Employee of Boehringer Ingelheim Pharma GmbH and Co. KG; and No conflict of interest for other authors.

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REFERENCES

1. Schöffski P. Polo-like kinase (PLK) inhibitors in preclinical and early clinical development in oncology. *Oncologist* 2009;**14**(6):559–70.
2. Schmit TL, Ahmad N. Regulation of mitosis via mitotic kinases: new opportunities for cancer management. *Mol Cancer Therapy* 2007;**6**:1920–31.
3. Strebhardt K, Ullrich A. Targeting polo-like kinase 1 for cancer therapy. *Nat Rev Cancer* 2006;**6**:321–30.
4. Steegmaier M, Hoffmann M, Baum A, et al. BI 2536, a potent and selective inhibitor of polo-like kinase 1, inhibits tumor growth in vivo. *Curr Biol* 2007;**17**(Suppl S2):316–22.
5. Ellis PM, Chu QS, Leighl NB, et al. A phase I dose escalation trial of BI 2536, a novel Plk1 inhibitor, with standard dose pemetrexed in previously treated advanced or metastatic non-small cell lung cancer (NSCLC). *J Clin Oncol* 2008;**26**:8115.
6. Hofheinz R, Hochhaus A, Al-Batran S, et al. A phase I repeated dose escalation study of the Polo-like kinase 1 inhibitor BI 2536 in patients with advanced solid tumors. *J Clin Oncol* 2006;**24**(Suppl 18):88s [abstract].
7. Mross K, Frost A, Steinbild S, et al. Phase I dose escalation and pharmacokinetic study of BI 2536, a novel polo-like kinase 1 inhibitor, in patients with advanced solid tumors. *J Clin Oncol* 2008;**26**:5511–7.
8. Therasse P, Arbuck SG, Eisenhauer EA, et al. New guidelines to evaluate the response to treatment in solid tumors. *J Natl Cancer Inst* 2000;**92**(3):205–16.
9. Simon R. Optimal two-stage designs for phase II clinical trials. *Controlled Clin Trials* 1989;**10**:1–10.
10. Von Pawel J, Reck M, Digel W, et al. Randomized phase II trial of two dosing schedules of BI 2536, a novel Plk-1 inhibitor, in patients with relapsed advanced or metastatic non-small-cell lung cancer (NSCLC). *J Clin Oncol* 2008;**26**:8030.
11. Pandha HS, Protheroe A, Wylie J, et al. An open label phase II trial of BI 2536, a novel Plk1 inhibitor, in patients with metastatic hormone refractory prostate cancer (HRPC). *J Clin Oncol* 2008;**26**:14547.
12. Mross K, Dittrich C, Aulitzky WE, et al. A randomized phase II trial of the novel polo-like kinase 1 inhibitor BI 2536 in chemonaïve patients with unresectable advanced pancreatic cancer—a study in cooperation with the CESAR network of investigators. In: *Presented at the 33rd annual congress of the European Society for Medical Oncology, September 12–16, 2008, Stockholm, Sweden*.
13. Schöffski P, Awada A, Dumez H, et al. A phase I single dose escalation study of the novel polo-like kinase I inhibitor BI 6727 in patients with advanced solid tumors. In: *Presented at the 20th European Organization for Research and Treatment of Cancer – National Cancer Institute – American Association for Cancer Research symposium on molecular targets and cancer therapeutics, October 21–24, 2008, Geneva, Switzerland*.
14. Rudolph D, Steegmaier M, Hoffmann M, et al. BI 6727, a polo-like kinase inhibitor with improved pharmacokinetic profile and broad antitumor activity. *Clin Can Res* 2009;**15**(9):3094–102.
15. Weinstein JM, Ghalib MH, Kumar M, et al. Phase I study of ON 01910.Na, a novel polo-like kinase 1 pathway modulator, administered as a weekly 24-h continuous infusion in patients with advanced cancer. *J Clin Oncol* 2008;**26**:2515.
16. Garland LL, Taylor C, Pilkington DL, et al. A phase I pharmacokinetic study of HMN-214, a novel oral stilbene derivative with Polo-like kinase-1-interacting properties, in patients with advanced solid tumors. *Clin Can Res* 2006;**12**:5182–9.
17. Von Hoff DD, Taylor C, Rubin S, et al. A phase I and pharmacokinetic study of HMN-214, a novel oral Polo-like kinase inhibitor, in patients with advanced solid tumors. *J Clin Oncol* 2004;**22**(Suppl 14):3034.
18. Blagden S, Olmos D, Sharma R, et al. A phase I first-in-human study of the polo-like kinase 1-selective inhibitor, GSK461364, in patients with advanced solid tumors. In: *Presented at the 20th European Organization for Research and Treatment of Cancer – National Cancer Institute – American Association for Cancer Research symposium on molecular targets and cancer therapeutics, October 21–24, 2008, Geneva, Switzerland*.
19. Chapman CM, Perez-Galan P, Wiestner A. ON 01910.Na, a novel clinical grade PLK-1 inhibitor, selectively induces apoptosis in human B-cell chronic lymphocytic leukemia (B-CLL). In: *Presented at the American Association of Cancer Research 2009 annual meeting, April 18–22, 2009, Denver, CO*.
20. Patnaik A, Forero L, Goetz A, et al. HMN-214, a novel oral antimicrotubular agent, inhibitor of Polo-like-and cyclin-dependent kinases: clinical, pharmacokinetic (PK), pharmacodynamic (PD) relationships observed in a phase I trial of a daily × 5 schedule every 28 days. *Proc Am Soc Clin Oncol* 2003;**22**:514 [abstract].
21. Ohnuma T, Cho SY, Roboz J, et al. Phase I study of ON 01910.Na by 3-day continuous infusion (CI) in patients (pts) with advanced cancer. *J Clin Oncol* 2006;**24**(Suppl 18):13137.
22. Donehower RC, Jimeno A, Li J, et al. Phase I study of ON 01910.Na, a novel cell cycle inhibitor in adult patients with solid tumors. *J Clin Oncol* 2006;**24**(Suppl 18):13026.