Combination of vemurafenib and cobimetinib in patients with advanced *BRAF*^{V600}-mutated melanoma: a phase 1b study

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Summary

Background Addition of a MEK inhibitor to a BRAF inhibitor enhances tumour growth inhibition, delays acquired resistance, and abrogates paradoxical activation of the MAPK pathway in preclinical models of *BRAF*-mutated melanoma. We assessed the safety and efficacy of combined BRAF inhibition with vemurafenib and MEK inhibition with cobimetinib in patients with advanced *BRAF*-mutated melanoma.

Methods We undertook a phase 1b study in patients with advanced *BRAF*^{V600}-mutated melanoma. We included individuals who had either recently progressed on vemurafenib or never received a BRAF inhibitor. In the dose-escalation phase of our study, patients received vemurafenib 720 mg or 960 mg twice a day continuously and cobimetinib 60 mg, 80 mg, or 100 mg once a day for either 14 days on and 14 days off (14/14), 21 days on and 7 days off (21/7), or continuously (28/0). The primary endpoint was safety of the drug combination and to identify dose-limiting toxic effects and the maximum tolerated dose. Efficacy was a key secondary endpoint. All patients treated with vemurafenib and cobimetinib were included in safety and efficacy analyses (intention-to-treat). The study completed accrual and all analyses are final. This study is registered with ClinicalTrials.gov, number NCT01271803.

Findings 129 patients were treated at ten dosing regimens combining vemurafenib and cobimetinib: 66 had recently progressed on vemurafenib and 63 had never received a BRAF inhibitor. Dose-limiting toxic effects arose in four patients. One patient on a schedule of vemurafenib 960 mg twice a day and cobimetinib 80 mg once a day 14/14 had grade 3 fatigue for more than 7 days; one patient on a schedule of vemurafenib 960 mg twice a day and cobimetinib 60 mg once a day 21/7 had a grade 3 prolongation of QTc; and two patients on a schedule of vemurafenib 960 mg twice a day and cobimetinib 60 mg 28/0 had dose-limiting toxic effects—one developed grade 3 stomatitis and fatigue and one developed arthralgia and myalgia. The maximum tolerated dose was established as vemurafenib 960 mg twice a day in combination with cobimetinib 60 mg 21/7. Across all dosing regimens, the most common adverse events were diarrhoea (83 patients, 64%), non-acneiform rash (77 patients, 60%), liver enzyme abnormalities (64 patients, 50%), fatigue (62 patients, 48%), nausea (58 patients, 45%), and photosensitivity (52 patients, 40%). Most adverse events were mild-to-moderate in severity. The most common grade 3 or 4 adverse events were cutaneous squamous-cell carcinoma (12 patients, 9%; all grade 3), raised amounts of alkaline phosphatase (11 patients, 9%]), and anaemia (nine patients, 7%). Confirmed objective responses were recorded in ten (15%) of 66 patients who had recently progressed on vemurafenib, with a median progression-free survival of 2·8 months (95% CI 2·6–3·4). Confirmed objective responses were noted in 55 (87%) of 63 patients who had never received a BRAF inhibitor, including six (10%) who had a complete response; median progression-free survival was 13·7 months (95% CI 10·1–17·5).

Interpretation The combination of vemurafenib and cobimetinib was safe and tolerable when administered at the respective maximum tolerated doses. The combination has promising antitumour activity and further clinical development is warranted in patients with advanced *BRAF*^{V600}-mutated melanoma, particularly in those who have never received a BRAF inhibitor; confirmatory clinical testing is ongoing.

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Introduction

Monotherapy with a BRAF inhibitor such as vemurafenib or dabrafenib in patients with *BRAF*^{V600}-mutant metastatic melanoma results in a high rate of tumour response and improvement in progression-free survival[1,2] and overall survival[1] compared with chemotherapy. However, acquired resistance develops in most patients, which frequently reactivates the MAPK pathway through MEK.[3–5] Theoretically, treatment with a MEK inhibitor should benefit some patients with acquired resistance. However, sequential use of a MEK inhibitor after progression on a BRAF inhibitor does not result in clinically meaningful antitumour activity either in vitro[6,7] or in patients.[8] Concurrent administration of MEK and BRAF inhibitors is effective on cell lines with acquired resistance to BRAF inhibitors[9] and can achieve responses in patients with *BRAF*^{V600}-mutant melanoma

progressing on single-agent BRAF inhibitors.[10] For patients who have not previously received a BRAF inhibitor, treatment with a combination of a BRAF inhibitor and a MEK inhibitor could lead to greater initial antitumour activity and prevent or delay MAPK-driven acquired resistance mechanisms.[11]

BRAF inhibition is associated with increased occurrence of squamous-cell carcinomas and hyperkeratotic skin lesions. Findings from animal models suggest that combined treatment with BRAF and MEK inhibitors might attenuate development of such lesions by blocking paradoxical activation of the MAPK pathway induced by single-agent BRAF inhibitors.[12,13] Therefore, the combination of a BRAF inhibitor and a MEK inhibitor might have the additional benefit of lessening the frequency of these adverse events in patients.[12–15]

We designed a phase 1b study to test the safety of the MEK inhibitor cobimetinib in combination with the BRAF inhibitor vemurafenib. We also aimed to define a dosing regimen for continued clinical testing.

Methods

Study design

We designed an open-label, multicentre, phase 1b, dose-escalation study to assess the safety, tolerability, pharmacokinetics, and activity of cobimetinib in combination with vemurafenib. We undertook the study at ten cancer centres located in the USA and Australia (appendix p 18).

We included individuals with unresectable, locally advanced, or metastatic melanoma who had the *BRAF*^{V600} mutation. Eligibility criteria were: age 18 years or older; unresectable stage IIIC or stage IV melanoma; positive for the *BRAF*^{V600} mutation on real-time PCR assay (cobas 4800 *BRAF*^{V600} Mutation Test, Roche Molecular Systems, Branchburg, NJ, USA); measurable disease, according to Response Evaluation Criteria in Solid Tumors (RECIST) version 1.1; Eastern Cooperative Oncology Group (ECOG) performance status of 0 or 1; estimated life expectancy of at least 12 weeks; and adequate haematological, hepatic, and renal function (appendix p 19).

We allowed patients to enrol if they had received any treatment previously for brain metastases and had at least a 3-week history of stable disease. We initially enrolled only patients who had previously received and progressed on vemurafenib; the protocol was later amended (on July 13, 2011) to include patients who had never received a BRAF inhibitor. We assessed patients separately according to previous treatment received. We did baseline ophthalmological examination and excluded patients if we identified findings or risk factors of retinal vein thrombosis or retinal detachment.

We undertook the study according to the provisions of the Declaration of Helsinki and its amendments and relevant Good Clinical Practice guidelines. The local institutional review board, independent ethics committee, or research ethics board of all participating study sites approved the study. All study participants provided written informed consent. An independent data safety monitoring board monitored and evaluated safety data from the study.

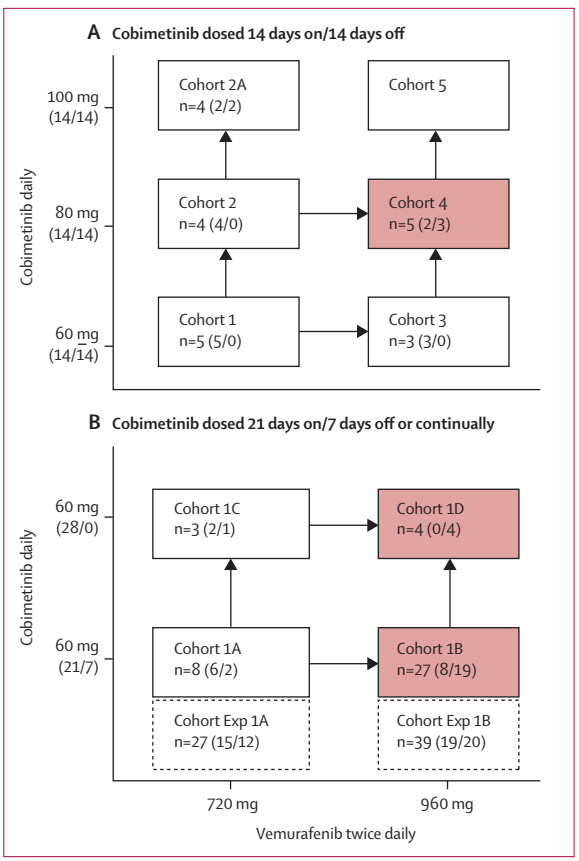

Figure 1: **Dose-escalation scheme and cohort assignment**
Dose-escalation scheme for cobimetinib 14 days on/14 days off (14/14; A) and 21 days on/7 days off (21/7) and continuous (28/0; B) Numbers (n) indicate patients assigned to every cohort, and numbers in parentheses are patients who progressed on vemurafenib/patients who had never received a BRAF inhibitor. Red boxes show cohorts in which dose-limiting toxic effects were identified. Dotted outlines indicate cohorts that were expanded.

See Online for appendix

Procedures

Our study consisted of two stages: dose escalation and cohort expansion. We administered vemurafenib in a regimen approved by the US Food and Drug Administration. We gave patients an oral dose of vemurafenib twice a day, in a continuous (28-day) cycle. For cobimetinib, we used intermittent schedules, at doses and in regimens tested in the single-agent phase 1 study of this agent.[16] We administered cobimetinib orally once a day, according to one of the following 28-day cycles: 14 consecutive days of cobimetinib followed by a 14-day drug holiday (14/14), starting two dose levels below the single-agent maximum tolerated dose; 21 consecutive days of cobimetinib followed by a 7-day drug holiday

	Vemurafenib dose	Cobimetinib dose	Enrolled (n)	Recently progressed on vemurafenib (n)	Never received a BRAF inhibitor (n)	Dose-limiting toxic effect (n)
1	720 mg twice a day	60 mg once a day, 14 days on/14 days off	5	5	0	0
2	720 mg twice a day	80 mg once a day, 14 days on/14 days off	4	4	0	0
2A	720 mg twice a day	100 mg once a day, 14 days on/14 days off	4	2	2	0
3	960 mg twice a day	60 mg once a day, 14 days on/14 days off	3	3	0	0
4	960 mg twice a day	80 mg once a day, 14 days on/14 days off	5	2	3	1 (grade 3 fatigue >7 days)
5	960 mg twice a day	100 mg once a day, 14 days on/14 days off	0	0	0	NA
1A	720 mg twice a day	60 mg once a day, 21 days on/7 days off	8	6	2	0
Exp 1A	720 mg twice a day	60 mg once a day, 21 days on/7 days off	27	15	12	NA
1B	960 mg twice a day	60 mg once a day, 21 days on/7 days off	27*	8	19	1 of 6* (grade 3 prolongation of QTc)
Exp 1B	960 mg twice a day	60 mg once a day, 21 days on/7 days off	39	19	20	NA
1C	720 mg twice a day	60 mg once a day, for 28 days	3	2	1	0
1D	960 mg twice a day	60 mg once a day, for 28 days	4	0	4	2 (grade 3 stomatitis and fatigue >7 days; grade 3 arthralgia and myalgia)

QTc=corrected QT interval. Exp=expansion cohort. NA=not assessed. *Assessment of dose-limiting toxic effects in cohort 1B was based on the first six patients enrolled into this cohort; further patients were enrolled while awaiting other cohorts to be declared safe and tolerable.

Table 1: **Cohort characteristics and dose-limiting toxic effects**

(21/7), starting at the single-agent maximum tolerated dose; or a continuous daily schedule (28/0).

We planned ten dose-escalation cohorts (figure 1); dose escalation proceeded in a standard 3+3 design. In brief, we started dose escalation in cohort 1 (vemurafenib 720 mg twice a day and cobimetinib 60 mg every day 14/14). After we declared cohort 1 safe and tolerable, as defined in the protocol (according to the absence of dose-limiting toxic effects), we continued dose escalation with simultaneous enrolment of cohorts 1A, 2, and 3. We enrolled cohort 2A only after we declared cohort 2 safe and tolerable, and we enrolled cohort 4 only after we declared cohorts 2 and 3 safe and tolerable. We enrolled cohorts 1B and 1C simultaneously only when we declared cohort 1A safe and tolerable, and we enrolled cohort 1D only after we declared both cohorts 1B and 1C safe and tolerable. We did not enrol patients into cohort 5. We gave priority for enrolment to advancing cohorts, but patients could be accrued into a previously cleared cohort if a slot did not exist in the currently enrolling dose cohort.

We expanded cohorts 1A and 1B (Exp 1A and Exp 1B) because both cohorts were declared safe and tolerable after dose escalation; furthermore, they delivered the single-agent maximum tolerated dose and schedule of cobimetinib and, in the case of cohort 1B, the approved dose and schedule of vemurafenib.

We defined the maximum tolerated dose as the highest dose of both vemurafenib and cobimetinib at which no more than one of six patients had dose-limiting toxic effects, including: grade 3 or higher non-haematological and non-hepatic organ toxic effects; and grade 4 or higher haematological toxic effects (except febrile neutropenia at grade 3 or higher). We set the assessment period for dose-limiting toxic effects at 28 days. We replaced

	Recently progressed on vemurafenib (n=66)	Never received a BRAF inhibitor (n=63)
Median age (years)	52·5 (range 19–88)	56·0 (range 21–74)
Men	42 (64%)	35 (56%)
White ethnic origin	63 (95%)	62 (98%)
ECOG performance status at baseline		
0	23 (35%)	41 (65%)
1	43 (65%)	22 (35%)
Stage at enrolment		
Unresectable stage IIIC	3 (5%)	7 (11%)
Stage IVA	4 (6%)	3 (5%)
Stage IVB	5 (8%)	9 (14%)
Stage IVC	54 (82%)	44 (70%)
Increased amount of lactate dehydrogenase at baseline	39 (62%)*	29 (46%)
Median follow-up (months)	6·3 (IQR 3·0–11·8)	12·7 (IQR 9·0–16·5)

Data are number of patients (%), unless otherwise stated. ECOG=Eastern Cooperative Oncology Group. *Data available for 63 patients.

Table 2: **Patients' characteristics and follow-up**

patients who discontinued study treatment or who had missed 3 days or more in total of either study drug during the assessment period for dose-limiting toxic effects, for reasons other than a dose-limiting toxic effect. However, we included these patients in safety and efficacy analyses.

We administered vemurafenib and cobimetinib until either patients withdrew consent, unacceptable adverse effects arose, or disease progression. We allowed patients to receive treatment beyond disease progression if the clinical investigator judged that clinical benefit from the combination would continue, with agreement from the medical monitor.

We assessed two populations: patients whose melanoma had progressed on vemurafenib immediately preceding enrolment in this trial (either progression defined using RECIST version 1.1 or unequivocal clinical progression in the opinion of the investigator); and individuals who either had never received a BRAF inhibitor, had not received previous treatment for advanced melanoma, or were previously treated but had not been exposed previously to BRAF or MEK inhibitor therapy. In patients whose melanoma had recently progressed on vemurafenib, either we continued with the drug after progression, until administration of the first study dose of vemurafenib and cobimetinib, or we restarted the drug during the 28-day screening period before cycle 1 day 1 of this study. We did not allow these patients to receive antimelanoma treatment in the intervening period, except with vemurafenib, and we excluded individuals if relevant toxic effects that needed treatment discontinuation had arisen while on vemurafenib previously. For patients who had never received a BRAF inhibitor but who had received an investigational agent previously, we required a 4-week washout period, and any adverse events must have recovered to at least grade 1 before enrolment.

Every treatment cycle was 28 days. We assessed patients every week during cycles 1 and 2; thereafter, visits took place once every cycle. Our assessments included a physical examination, electrocardiography, and laboratory studies. We graded adverse events according to the Common Terminology Criteria for Adverse Events version 4.0.

To manage adverse events, we allowed modifications to the dose of vemurafenib and cobimetinib—ie, reductions, interruptions, and permanent discontinuation. In general, for adverse events of grade 1 or 2, we did not modify the dose, according to the protocol. For most clinically significant adverse events of grade 3 or higher, dosing of either vemurafenib or cobimetinib, or both drugs, was interrupted depending on the nature of the event—eg, abnormal liver function test results for vemurafenib, or diarrhoea for cobimetinib. We resumed the dose at the same or a lower amount if the adverse event resolved to grade 1 or lower within 28 days. If a dose reduction was indicated according to the protocol, we decreased the dose of vemurafenib by 240 mg twice a day and reduced the

	Recently progressed on vemurafenib (n=66)			Never received a BRAF inhibitor (n=63)		
	All grades	Grade 3	Grade 4	All grades	Grade 3	Grade 4
Most common adverse events						
Non-acneiform rash*	22 (33%)	1 (2%)	0	55 (87%)	9 (14%)	0
Diarrhoea	31 (47%)	2 (3%)	0	52 (83%)	5 (8%)	0
Fatigue	18 (27%)	1 (2%)	0	44 (70%)	6 (10%)	0
Photosensitivity or sunburn*	10 (15%)	1 (2%)	0	42 (67%)	2 (3%)	0
Liver enzyme abnormality*	22 (33%)	2 (3%)	2 (3%)	42 (67%)	9 (14%)	3 (5%)
Nausea	22 (33%)	2 (3%)	0	36 (57%)	2 (3%)	0
Arthralgia	8 (12%)	1 (2%)	0	30 (48%)	7 (11%)	0
Increase in creatine phosphokinase	10 (15%)	0	1 (2%)	27 (43%)	2 (3%)	0
Pyrexia	11 (17%)	1 (2%)	0	27 (43%)	1 (2%)	0
Vomiting	13 (20%)	1 (2%)	0	27 (43%)	0	0
Blood and lymphatic-system disorders						
Anaemia	10 (15%)	4 (6%)	0	20 (32%)	5 (8%)	0
Lymphopenia	2 (%)	1 (2%)	0	1 (2%)	1 (2%)	0
Abdominal pain	10 (15%)	1 (2%)	0	12 (19%)	0	0
Cardiac disorders						
Congestive cardiomyopathy	0	0	0	1 (2%)	1 (2%)	0
Eye disorders						
Retinal detachment	1 (2%)	0	0	1 (2%)	0	0
Chorioretinopathy	0	0	0	3 (5%)	0	0
Retinopathy	0	0	0	2 (3%)	0	0
Macular oedema	0	0	0	1 (2%)	0	0
Vogt-Koyanagi-Harada syndrome	0	0	0	1 (2%)	1 (2%)	0
Gastrointestinal disorders						
Constipation	9 (14%)	1 (2%)	0	10 (16%)	0	0
Abdominal distension	6 (9%)	1 (2%)	0	2 (3%)	0	0
Gastro-oesophageal reflux disease	0	0	0	5 (8%)	1 (2%)	0
Gastrointestinal haemorrhage	2 (3%)	0	1 (2%)	0	0	0
Colitis	0	0	0	1 (2%)	1 (2%)	0
Haematemesis	1 (2%)	1 (2%)	0	0	0	0
Small intestinal obstruction	1 (2%)	1 (2%)	0	0	0	0
Administration-site and general disorders						
Mucosal inflammation	3 (5%)	0	0	4 (6%)	1 (2%)	0
Asthenia	5 (8%)	0	0	1 (2%)	1 (2%)	0
Non-cardiac chest pain	0	0	0	1 (2%)	1 (2%)	0
Hepatobiliary disorders						
Biloma	0	0	0	1 (2%)	1 (2%)	0
Infections and infestations						
Urinary-tract infection	3 (5%)	0	0	7 (11%)	2 (3%)	0
Skin infection	1 (2%)	0	0	3 (5%)	1 (2%)	0
Pneumonia	3 (5%)	2 (3%)	0	1 (2%)	0	0
Sepsis	0	0	0	3 (5%)	1 (2%)	1 (2%)
Ophthalmic herpes zoster	0	0	0	1 (2%)	1 (2%)	0
Pneumonia, chlamydial	0	0	0	1 (2%)	1 (2%)	0
Pyelonephritis	1 (2%)	1 (2%)	0	0	0	0
Wound infection	1 (2%)	1 (2%)	0	0	0	0
Injury, poisoning, and procedural complications						
Femur fracture	1 (2%)	1 (2%)	0	1 (2%)	0	0
Procedural pain	1 (2%)	1 (2%)	0	0	0	0

(Table 3 continues on next page)

	Recently progressed on vemurafenib (n=66)			Never received a BRAF inhibitor (n=63)		
	All grades	Grade 3	Grade 4	All grades	Grade 3	Grade 4
(Continued from previous page)						
Investigations						
Increase in alkaline phosphatase	10 (15%)	3 (5%)	1 (2%)	21 (33%)	7 (11%)	0
Increase in aspartate aminotransferase	6 (9%)	0	0	22 (35%)	5 (8%)	0
Increase in alanine aminotransferase	4 (6%)	0	0	23 (36%)	6 (10%)	0
Increase in creatinine	6 (9%)	0	0	20 (32%)	1 (2%)	0
Increase in bilirubin	6 (9%)	0	0	8 (13%)	1 (2%)	0
QT interval prolonged	5 (8%)	2 (3%)	0	4 (6%)	1 (2%)	0
Increase in γ-glutamyltransferase	1 (2%)	0	1 (2%)	7 (11%)	3 (5%)	3 (5%)
Increase in uric acid	0	0	0	4 (6%)	0	1 (2%)
Weight increased	0	0	0	4 (6%)	1 (2%)	0
Increase in aminotransferase	0	0	0	3 (5%)	1 (2%)	0
Metabolism and nutrition disorders						
Hypokalaemia	4 (6%)	0	0	10 (16%)	3 (5%)	0
Hypophosphataemia	3 (5%)	2 (3%)	0	9 (14%)	4 (6%)	0
Hyperglycaemia	2 (3%)	0	0	10 (16%)	2 (3%)	0
Hyponatraemia	3 (5%)	0	0	9 (14%)	3 (5%)	1 (2%)
Hypoalbuminaemia	4 (6%)	1 (2%)	0	7 (11%)	0	0
Hyperuricaemia	0	0	0	7 (11%)	2 (3%)	0
Dehydration	4 (6%)	1 (2%)	0	2 (3%)	0	0
Musculoskeletal and connective tissue disorders						
Myalgia	4 (6%)	0	0	15 (23%)	1 (2%)	0
Back pain	10 (15%)	0	0	7 (11%)	2 (3%)	0
Neck pain	4 (6%)	1 (2%)	0	2 (3%)	0	0
Joint range of motion decreased	1 (2%)	1 (2%)	0	1 (2%)	0	0
Neoplasms: benign, malignant, and unspecified (including cysts and polyps)						
Cutaneous squamous-cell carcinoma	1 (2%)	5 (8%)	0	7 (11%)	7 (11%)	0
Seborrhoeic keratosis	1 (2%)	0	0	11 (17%)	1 (2%)	0
Basal-cell carcinoma	1 (2%)	0	0	6 (10%)	2 (3%)	0
Keratoacanthoma	1 (2%)	1 (2%)	0	1 (2%)	1 (2%)	0
Adrenal neoplasm	0	0	0	1 (2%)	1 (2%)	0
Tonsil cancer	1 (2%)	0	1 (2%)	0	0	0
Nervous-system disorders						
Headache	13 (20%)	0	0	17 (27%)	2 (3%)	0
Paraesthesia	5 (8%)	0	0	4 (6%)	1 (2%)	0
Convulsion	0	0	0	5 (8%)	2 (3%)	0
Syncope	2 (3%)	0	0	3 (5%)	3 (5%)	0
Presyncope	0	0	0	1 (2%)	1 (2%)	0
Nerve VII paralysis	0	0	0	2 (3%)	2 (3%)	0
Psychiatric disorders						
Depression	4 (6%)	0	0	5 (8%)	1 (2%)	0
Mental status changes	1 (2%)	1 (2%)	0	1 (2%)	0	0
Suicidal ideation	0	0	0	1 (2%)	1 (2%)	0
Renal and urinary disorders						
Haematuria	3 (5%)	0	0	4 (6%)	1 (2%)	0
Renal failure	2 (3%)	0	0	2 (3%)	1 (2%)	0
Nephrolithiasis	0	0	0	1 (2%)	1 (2%)	0
Reproductive-system and breast disorders						
Pelvic pain	1 (2%)	1 (2%)	0	0	0	0

(Table 3 continues on next page)

dose of cobimetinib by 20 mg a day, independently, depending on causality (as assessed by the clinical investigator). If the study treatment was interrupted for more than 28 days for whatever reason, we judged the patient had been discontinued from the study.

We did tumour assessments at 6-week intervals during the treatment period for all patients. Investigators used RECIST version 1.1 to assess tumour responses and progression. We obtained plasma samples for pharmacokinetic characterisation of cobimetinib and vemurafenib on days 1 and 14 of cycle 1 and on day 8 of cycles 2 and 3. For patients who had recently progressed on vemurafenib, we did additional pharmacokinetic characterisation on day –1, before combination treatment started.

Outcomes

Primary objectives of the dose-escalation stage were to establish the safety, tolerability, and pharmacokinetics of the combination of vemurafenib and cobimetinib and to define the recommended phase 2 or 3 dose and schedule. In the cohort-expansion stage, we aimed to establish further the safety profile and pharmacokinetics of the treatment combination. A secondary objective was to assess efficacy of the combination of vemurafenib and cobimetinib, by measuring objective responses, progression-free survival, duration of response, and overall survival.

Statistical analysis

The cutoff date for safety and efficacy follow-up data was Oct 1, 2013. We included in the safety and efficacy analyses all patients who received any amount of study treatment (intention-to-treat analysis). We did not use any formal statistical model in the efficacy analysis and we planned no formal hypothesis testing. To summarise data for safety, clinical activity, and pharmacokinetics of vemurafenib and cobimetinib, we used descriptive statistics (SAS version 9.2). We recorded all adverse events arising on or after treatment on cycle 1 day 1 in frequency tables, and we defined events according to terms in the Medical Dictionary for Regulatory Affairs (MedRA).

The clinical investigator assessed objective response, which they defined as complete (CR) or partial response (PR). We confirmed tumour response at least 4 weeks after the initial documentation was received, using RECIST version 1.1. We defined progression-free survival as the interval between treatment start date and date of progression or death from any cause, whichever came first; we categorised progression using RECIST version 1.1. For patients who did not progress and who survived, we censored data at the last evaluable tumour assessment date. We calculated duration of response only for patients who had an objective response and defined it as the interval between the date of the earliest qualifying response and the date of progressive disease (PD) or death. Patients without progression after an objective response were censored at the last evaluable

tumour assessment date. More details on statistical analyses are provided in the appendix (pp 14–17).

This study is registered with ClinicalTrials.gov, number NCT01271803.

Role of the funding source

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Results

Between Feb 17, 2011, and July 23, 2013, 129 patients were enrolled and received vemurafenib plus cobimetinib (table 1). 66 patients had recently progressed on vemurafenib monotherapy and 63 had never received a BRAF inhibitor (table 2). Patients in whom melanoma progressed while they were taking vemurafenib were more likely to have an ECOG performance status of 1, stage IV M1c disease, and an increased amount of lactate dehydrogenase in serum than were the population who had never had a BRAF inhibitor (table 2). Cohorts 1, 2, and 3 included only patients who had recently progressed on vemurafenib. Within the population who had never received a BRAF inhibitor, 44 (70%) of 63 patients had stage IV M1c disease and 29 (46%) had raised lactate dehydrogenase activity. Expansion cohorts were opened for both populations at a cobimetinib dose of 60 mg daily 21/7 in combination with vemurafenib at 720 mg (Exp 1A) or 960 mg (Exp 1B) twice a day.

As doses and exposure increased, the number of toxic effects also grew, as expected. Four dose-limiting toxic effects were recorded, one each in cohorts 4 and 1B and two in cohort 1D (table 1); no dose-limiting toxic effects were noted in the other cohorts. In cohort 4 (vemurafenib 960 mg twice a day and cobimetinib 80 mg daily 14/14), one of five patients had grade 3 fatigue. Most patients in 14/14 cohorts entered the study after progression on single-agent vemurafenib. After an initial tumour response, investigators saw that the 14-day cobimetinib dosing holiday afforded an opportunity for tumour regrowth. Therefore, no additional patients were accrued into cohorts with cobimetinib on the 14/14 schedule. In cohort 1B (vemurafenib 960 mg twice a day and cobimetinib 60 mg daily 21/7), one of six patients initially enrolled into the cohort had grade 3 QT prolongation, but the regimen met protocol-specified criteria to be declared safe and tolerable. In cohort 1D (vemurafenib 960 mg twice a day and cobimetinib 60 mg, 28/0), two of four patients had a dose-limiting toxic effect: either grade 3 stomatitis and fatigue or grade 3 arthralgia and myalgia. Therefore, the dose administered to cohort 1B, which delivered both drugs at their respective single-agent maximum tolerated doses, was defined as the maximum tolerated dose of the combination, and the 21/7 dosing schedule was subsequently expanded (Exp 1B). No dose-limiting toxic effects were recorded in either expansion cohort (Exp 1A and Exp 1B).

Overall, adverse events were more common in patients who had never received a BRAF inhibitor than in those who enrolled after progression on vemurafenib (table 3). Total duration of exposure was longer for patients who had never received vemurafenib, which might have played a part in the different rate of toxic effects. Common adverse events in 63 patients who had never received a BRAF inhibitor and in 66 individuals recently progressing on vemurafenib were non-acneiform rash, diarrhoea, fatigue, photosensitivity or sunburn, and liver

	Recently progressed on vemurafenib (n=66)			Never received a BRAF inhibitor (n=63)		
	All grades	Grade 3	Grade 4	All grades	Grade 3	Grade 4
(Continued from previous page)						
Respiratory, thoracic, and mediastinal disorders						
Hypoxia	3 (5%)	1 (2%)	0	0	0	0
Pleural effusion	2 (3%)	1 (2%)	0	0	0	0
Pulmonary embolism	0	0	0	1 (2%)	1 (2%)	0
Skin and subcutaneous tissue disorders						
Rash	18 (27%)	0	0	32 (51%)	1 (2%)	0
Acneiform rash	5 (8%)	0	0	19 (31%)	1 (2%)	1 (2%)
Maculopapular rash	2 (3%)	1 (2%)	0	19 (31%)	4 (6%)	0
Hyperkeratosis	2 (3%)	1 (2%)	0	9 (14%)	0	0
Erythaematous rash	3 (5%)	0	0	7 (11%)	1 (2%)	0
Generalised rash	1 (2%)	0	0	3 (5%)	3 (5%)	0
Acne	2 (3%)	1 (2%)	0	2 (3%)	0	0
Morbilliform rash	0	0	0	1 (2%)	1 (2%)	0
Vascular disorders						
Hypertension	6 (9%)	1 (2%)	0	17 (27%)	4 (6%)	0
Hypotension	4 (6%)	1 (2%)	0	2 (3%)	1 (2%)	0

Data are number of patients (%). No grade 5 adverse events were recorded. *Medical Dictionary for Regulatory Activities (MedDRA) grouping.

Table 3: **Adverse events**

	Recently progressed on vemurafenib (n=66)	Never received a BRAF inhibitor (n=63)
Complete response	0	6 (10%)
Partial response	10 (15%)	49 (78%)
Stable disease	28 (42%)	6 (10%)
Progressive disease	24 (36%)	2 (3%)
Not assessable or not done	4 (6%)	0

Data are number of patients (%).

Table 4: **Best response to treatment**

enzyme abnormalities (table 3). No grade 5 events were recorded. Details of all adverse events, by grade, are provided in the appendix (pp 1–12). Cutaneous squamous-cell carcinoma or keratoacanthoma, previously shown to be associated with monotherapy with BRAF inhibitors, developed in eight (13%) of 63 patients who had never received a BRAF inhibitor and six (9%) of 66 patients who had recently progressed on vemurafenib (table 3).

Toxic effects typical of MEK inhibition[11] were also more common in patients who had never received a BRAF inhibitor than in those who progressed on vemurafenib, including diarrhoea, an increase in creatine phosphokinase activity, and acneiform rash (table 3). No rhabdomyolysis associated with augmented activity of creatinine phosphokinase was reported. In the 63 patients who had never received a BRAF inhibitor, seven (11%) had the underlying diagnosis of serous chorioretinopathy (five grade 1 and two grade 2), with reported adverse events of chorioretinopathy, retinopathy, macular oedema, and retinal detachment (table 3). Retinal vein occlusion was not reported in any patients. One individual had symptomatic grade 3 cardiomyopathy, which resulted in permanent discontinuation of cobimetinib; ejection function and heart failure symptoms subsequently improved on vemurafenib monotherapy.

Dose modifications because of adverse events are shown in the appendix (p 13). As expected in view of the difference in toxic effects between populations, the proportion of dose interruptions or reductions in patients who had never received a BRAF inhibitor was higher than in individuals recently progressing on vemurafenib. In the population who had never received a BRAF inhibitor, permanent withdrawal of both vemurafenib and cobimetinib because of adverse events happened in two patients, for grade 3 rash and grade 3 QT interval prolongation; vemurafenib alone was stopped

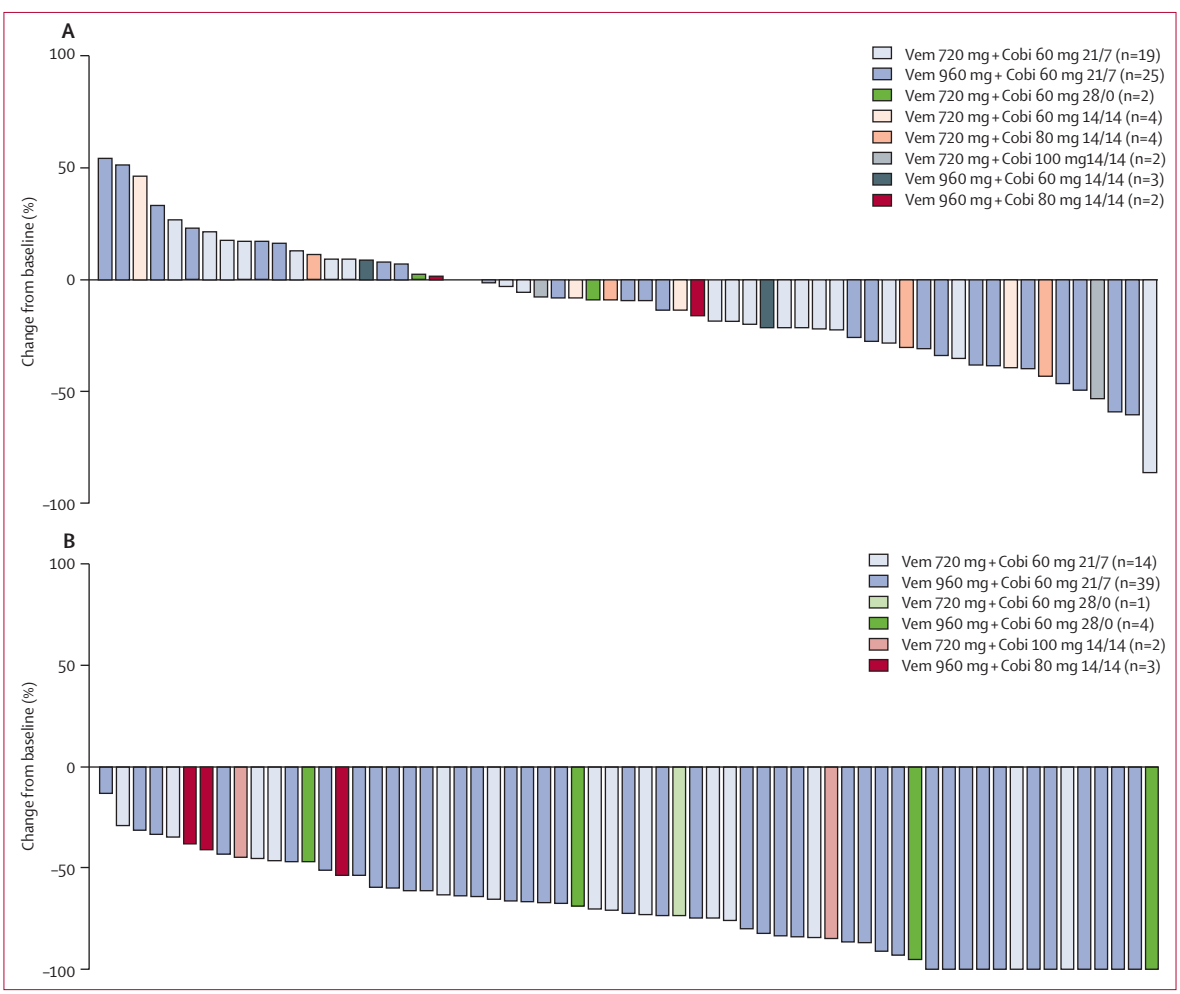

Figure 2: **Best change from baseline in target lesions**
Waterfall plots show tumour response, which was measured as the change from baseline in the sum of the longest diameters of target lesions, as defined by RECIST version 1.1. Coloured bars represent doses and schedules of vemurafenib (Vem) and cobimetinib (Cobi). Patients receiving the same regimen from both the expansion and escalation cohorts are represented by the same colour within each waterfall plot. 14/14=14 days on/14 days off. 21/7=21 days on/7 days off. 28/0=continuously. (A) Patients who had recently progressed on single-agent vemurafenib (n=66). (B) Individuals who had never received a BRAF inhibitor (n=63).

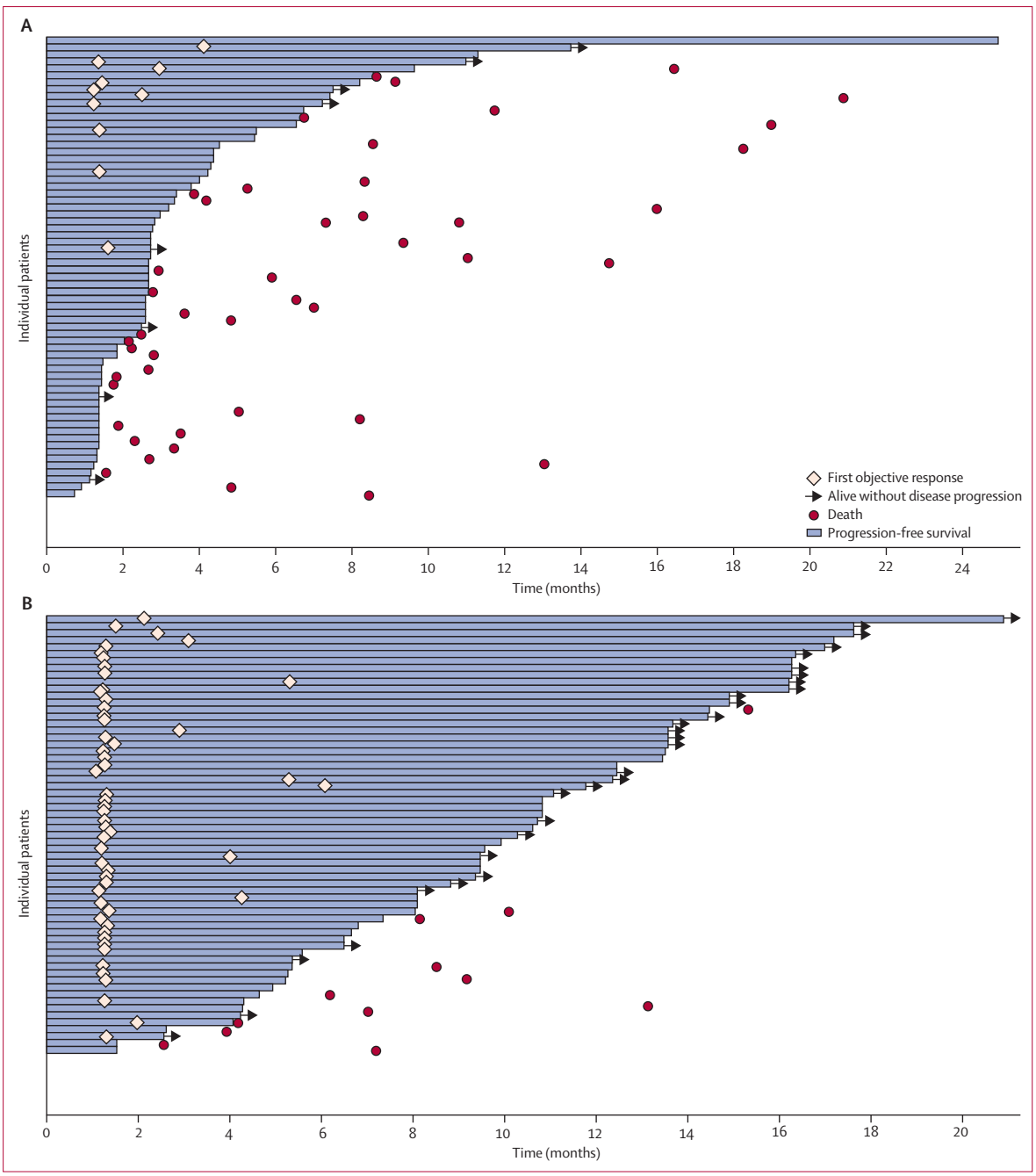

Figure 3: **Time to first response, progression, and death**
(A) Patients who recently progressed on single-agent vemurafenib. (B) Patients who had never received a BRAF inhibitor.

permanently in two patients for a grade 4 rise in γ-glutamyltransferase activity and a grade 2 increase in creatinine; and cobimetinib alone was discontinued permanently in one patient for grade 3 cardiomyopathy. Although dose interruptions or modifications were frequent, particularly in patients who had never received a BRAF inhibitor, the combination of vemurafenib and cobimetinib was sufficiently well-tolerated that permanent withdrawal of either agent only occurred in about 5% of patients. The proportion of patients who had never received a BRAF inhibitor and who needed modifications to the recommended phase 2/3 dose was similar to the overall proportion of dose modifications in the population who had never received a BRAF inhibitor (appendix p 13).

Analysis of pharmacokinetic data showed that vemurafenib exposure was not altered by coadministration

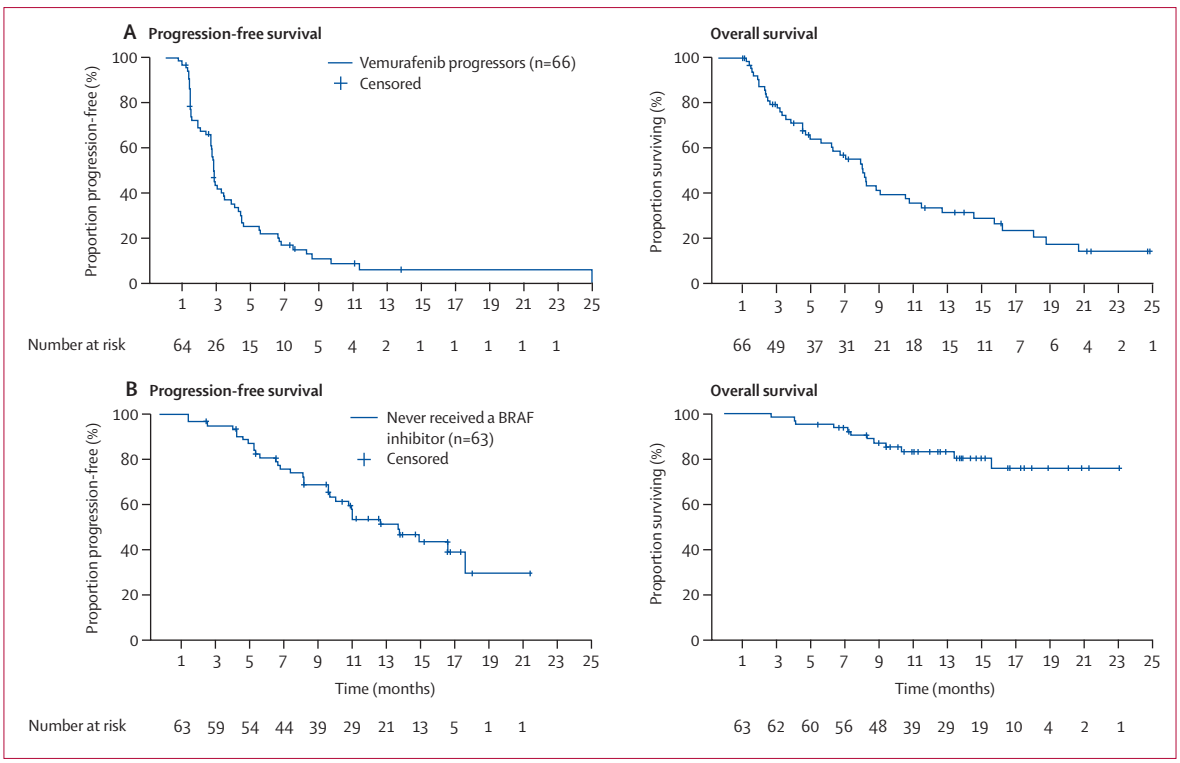

Figure 4: **Progression-free and overall survival**
(A) Patients who recently progressed on single-agent vemurafenib. (B) Patients who had never received a BRAF inhibitor.

with cobimetinib. In patients who recently progressed on vemurafenib, day –1 vemurafenib pharmacokinetic data (vemurafenib single-agent dosing) were comparable with those obtained for vemurafenib in cycle 1 day 14 (vemurafenib in combination with cobimetinib). Similarly, vemurafenib did not alter pharmacokinetic measurements for cobimetinib. Pharmacokinetic data for cobimetinib in combination with vemurafenib were comparable with those for cobimetinib alone (maximum drug plasma concentration and area under the concentration–time curve at steady state) recorded in the phase 1 dose-escalation and expansion study.[16]

Ten (15%) of 66 patients who had recently progressed on vemurafenib had a confirmed objective response, with no CRs (table 4). 28 (42%) had stable disease whereas 24 (36%) had PD as their best response. Figure 2A presents target lesion responses for patients who had recently progressed on vemurafenib. In the ten patients who had an objective response, median time to objective tumour response was 1·5 months (95% CI 1·3–4·2), with a response duration of 6·7 months (4·9–not evaluable). Six of 13 patients who had recently progressed on vemurafenib and with progression-free survival of longer than 6 months did not achieve a PR, including an individual with a best response of stable disease who was progression-free for more than 2 years (figure 3A).

All 63 patients who had never received a BRAF inhibitor had evidence of target lesion reduction after combination treatment (figure 2B). In this population, 55 (87%) patients had confirmed objective responses, including CRs in six (10%) people. Only two (3%) individuals had PD as their best response (figure 3B). Both of these patients had a reduction in their RECIST-defined target lesions, but new tumour lesions developed. Tumour response happened rapidly, typically by the time of the first tumour assessment (median time to objective response 1·4 months, 95% CI 1·2–6·2). The median duration of response was 12·5 months (95% CI 9·7–not evaluable).

During enrolment, patients were selected using the cobas assay, which detects all people with $BRAF^{V600E}$ mutations and most patients with $BRAF^{V600K}$ mutations. Post-hoc sequencing of 94 unique tumour samples obtained from this study showed that seven tumours had a mutation other than $BRAF^{V600E}$. Consistent with efficacy data reported previously with single-agent vemurafenib,[17] in three patients who had never received a BRAF inhibitor and who had mutations other than $BRAF^{V600E}$, one individual had a CR, one had a PR, and one had stable disease. By contrast, in four patients who had recently progressed on vemurafenib and who had mutations other than $BRAF^{V600E}$, two people had PD as their best overall response, one had stable disease, and one could not be assessed.

Figure 4 presents progression-free and overall survival curves for both populations. 58 (88%) of 66 patients

who had recently progressed on vemurafenib progressed or died during the study period. In this population, median progression-free survival was 2·8 months (95% CI 2·6–3·4). Of the 63 patients who had never received a BRAF inhibitor, 33 (52%) progressed or died during the study period, with a median progression-free survival of 13·7 months (95% CI 10·1–17·5). The remaining 38 patients who had not progressed at the time of data cutoff were allowed to continue with study treatment.

45 (68%) of 66 patients who had recently progressed on vemurafenib died. Median overall survival was 8·3 months (95% CI 5·95–10·87). Estimated 1-year overall survival in this population was 32% (19–45). Only 12 (19%) of 63 patients who had never received a BRAF inhibitor had died at the time of reporting, corresponding to an estimated 1-year overall survival of 83% (95% CI 73–93).

Discussion

The findings of our study show that the combination of vemurafenib and cobimetinib was safe and tolerable at the respective single-agent maximum tolerated doses (960 mg of vemurafenib twice a day and 60 mg cobimetinib daily 21/7). Permanent discontinuation because of toxic effects was low. Tumour responses and progression-free survival were encouraging in patients with *BRAF*^{V600}-mutated melanoma who had never received a BRAF inhibitor.

Because MEK inhibition diminishes BRAF-induced paradoxical activation of the MAPK pathway in normal tissue (ie, without a *BRAF* mutation), MEK inhibition might have positively affected tolerability of the treatment combination, despite both drugs targeting the same pathway.[13] Consistent with this idea, the proportion of cutaneous squamous-cell carcinomas we noted in this work (8–11%) seemed lower than that reported in studies of vemurafenib monotherapy (19–26%).[1,18,19]

The number of adverse events in patients who had recently progressed on vemurafenib was lower than that in individuals who had never received a BRAF inhibitor. This difference could be attributable to several factors, including a longer duration on study for patients who had never received a BRAF inhibitor, previous exposure to vemurafenib and ongoing management of vemurafenib-related adverse events in individuals who had recently progressed on vemurafenib, and assignment of patients who had recently progressed on vemurafenib to a cohort that delivered vemurafenib at or below the previously tolerated dose. With respect to cohort assignments, 39 (62%) of 63 patients who had never received a BRAF inhibitor were treated at the maximum tolerated dose of each drug, compared with 27 (41%) of 66 with previous exposure to vemurafenib. Photosensitivity was recorded less frequently in patients who had recently progressed on vemurafenib than in individuals who had never received a BRAF inhibitor (table 3), which might reflect effective avoidance of ultraviolet exposure in people who have been exposed previously to vemurafenib and have experience of photosensitivity or sunburn.

***Panel*: Research in context**

Systematic review

We searched PubMed and meeting abstracts from the American Society of Clinical Oncology and the European Society of Medical Oncology for 2012, 2013, and 2014 (if available), with the terms: "advanced melanoma", "clinical trial", "BRAF inhibitor", and "MEK inhibitor". The results showed that, in addition to the combination of vemurafenib and cobimetinib, BRAF and MEK inhibitor combinations of dabrafenib and trametinib[20,21] and LGX818 and MEK162[22] have been assessed in patients with advanced *BRAF*-mutated melanoma. Data for the dabrafenib and trametinib combination, which was investigated in a randomised phase 2 study, showed that the combination might result in increased response and extended progression-free survival over BRAF-inhibitor monotherapy.[21]

Interpretation

The results of our study are promising and support the idea that combined inhibition of BRAF and MEK might result in improvement of clinical outcome in patients with *BRAF*-mutated melanoma who have never received a BRAF inhibitor. The combination of vemurafenib and cobimetinib can be delivered at the respective single-agent maximum tolerated doses, with tolerable and manageable adverse events. Cutaneous squamous-cell carcinomas were reported less frequently compared with historical data for BRAF-inhibitor monotherapy, supporting the idea that combined inhibition reduces paradoxical activation of the MAPK pathway. The promising effectiveness of this combination must be confirmed in a randomised phase 3 study.

Although in our study we allowed patients with treated and stable brain metastases to be accrued, we did not specifically obtain data on characteristics of brain metastases nor did we detail previous treatment received for brain lesions. Therefore, subgroup analyses based on these characteristics are not possible. Serial surveillance of cardiac function by echocardiography was not done in this study, therefore, we cannot exclude asymptomatic impairment of cardiac function. We might have underestimated cardiac function toxic effects of the combination of vemurafenib and cobimetinib.

Pharmacokinetic analysis showed that exposure of vemurafenib or cobimetinib was not altered when administered in combination. Cobimetinib is metabolised by CYP3A4 and UGT2B7, whereas CYP3A plays a part in the metabolism of vemurafenib. Cobimetinib is not an inhibitor or inducer of CYP3A and vemurafenib is a mild inducer of CYP3A; therefore, no substantial interaction between drugs was noted. Detailed pharmacokinetic and biomarker analyses of vemurafenib in combination with cobimetinib are planned.

Although our study was mainly a phase 1b study, the expansion stage allowed for improved precision in defining the safety and efficacy of the combination in a large and homogenous population receiving a uniform dose and schedule. Confirmed responses to the cobimetinib and vemurafenib combination in patients who had never received a BRAF inhibitor were noted in 55 (87%) of 63 patients, and six (10%) patients achieved a CR. This proportion seems substantially higher than in previous studies of vemurafenib monotherapy (confirmed responses in 53–57% of patients)[1,18,19] or of

MEK inhibitor monotherapy (responses in 22% of patients).[11] Based on the encouraging results of our study, a randomised phase 3 trial comparing cobimetinib and vemurafenib with vemurafenib monotherapy is ongoing (NCT01689519). The maximum tolerated doses of cobimetinib and vemurafenib that we identified in our study are being administered to previously untreated patients with advanced *BRAF*^{V600}-mutated melanoma.

Ten (15%) of 66 patients who recently progressed on vemurafenib had a tumour response, which is notably lower than in patients who had never received a BRAF inhibitor. This finding suggests that reduction of disease burden is most effective with the drug combination when the majority of cells are sensitive to vemurafenib and before any selective pressure from genomic or epigenetic changes has taken place. Although the most common cause of acquired resistance to BRAF inhibitors is reactivation of the MAPK pathway through MEK,[3–5] by the time resistance to BRAF inhibitor monotherapy develops, evidence can be seen of branched tumour heterogeneity, leading to development of several mechanisms of resistance in parallel in different metastatic lesions.[3] During evaluation of the schedule of continuous vemurafenib combined with cobimetinib in a 14/14 schedule, tumour regrowth was noted in some patients during the 14-day period off cobimetinib. When a longer period of exposure to cobimetinib was investigated (21/7 schedule), no tumour regrowth was seen during the 7 days off cobimetinib.

Patients who had never received a BRAF inhibitor achieved a median progression-free survival of more than 13 months, which is notably longer than that recorded in previous studies of vemurafenib monotherapy, in which median progression-free survival was less than 7 months.[1,18,19] Although this observation will undergo definitive assessment in the randomised trial that is underway, the finding suggests that combined inhibition of BRAF and MEK with vemurafenib and cobimetinib might delay the emergence of resistant tumours, which can be predicted from data of preclinical studies showing combined activity of BRAF and MEK inhibitors in cells resistant to BRAF-inhibitor monotherapy. As discussed in the panel, emerging clinical evidence suggests that the combination of dabrafenib and trametinib might extend progression-free survival over dabrafenib alone.[20] By contrast, progression-free survival was less than 3 months in patients who had recently progressed on vemurafenib, suggesting that this approach does not enable durable control of *BRAF*^{V600}-mutated melanoma. Findings of ongoing clinical and correlative research studies will further define the role of the combination of vemurafenib and cobimetinib for treatment of patients with *BRAF*^{V600}-mutated melanoma.

Contributors

AR contributed to the design and implementation of the study, patients' treatment, data analysis and interpretation, and writing of the report. RG contributed to patients' recruitment, patients' treatment, and data collection, and reviewed the report. AP contributed to patients' accrual and reviewed the report. OH contributed to study design, data collection, data analysis, data interpretation, and writing of the report. TFG contributed to patients' enrolment, data collection, and data analysis, and proofread the report. AD contributed to data collection, data analysis, and data interpretation, and edited and approved the final report. LF contributed to patients' accrual and reviewed and approved the report. TL contributed to data collection, data interpretation, and writing of the report. BC provided study materials, contributed to patients' enrolment, collected and assembled data, contributed to data interpretation, provided substantive suggestions for revisions, critically reviewed the report, and approved the final version. KL contributed to data collection and writing and revision of the report. DK contributed to data collection, data interpretation, and writing of the report. PB contributed to interpretation of results, provided substantive suggestions for revisions and reviewed subsequent iterations, and reviewed and approved the final version of the report. MY was the study statistician and contributed to data collection, data analysis, data interpretation, and writing of the report. IC contributed to study design, data analysis, and data interpretation, and reviewed and edited the report. LM contributed to study design, data analysis, data interpretation, and writing and review of the report. NC contributed to study design, data collection, data analysis, data interpretation, and writing and final approval of the report. IP contributed to study design, data collection, data analysis, and data interpretation, and writing, review, and final approval of the report. GAM contributed to study design, data collection, and data interpretation, and writing of the report.

Declaration of interests

AR has received honoraria and grants from Genentech for participating in this clinical trial. RG has received personal fees from Roche/Genentech for participation in clinical trials, involvement with advisory boards, and speaking; he has also received a grant from GlaxoSmithKline for participation in the data-monitoring committee for the Metric trial. OH received a research grant from Genentech during implementation of this study and has previously been compensated as a consultant and speaker for Genentech. TFG has received personal fees from Roche/Genentech for services on an advisory board. LF received a grant from Roche/Genentech related to implementation of this study and has previously received an honorarium for services on a speaker's bureau from Roche/Genentech. TL received a grant from Roche/Genentech during implementation of this study; he has previously received grants from Abbott, Abraxis, Acceleron, Amgen, Argos, AstraZeneca, Aveo, Biovex, Bristol-Myers Squibb, Eisai, Eli Lilly, GlaxoSmithKline, Genentech, Celgene, Hoffmann-La Roche, Immatics, Merck, Novartis, Pfizer, Prometheus, Roche, Synta, Threshold, and Wyeth and personal fees from Argos, Aveo, Bristol-Myers Squibb, GlaxoSmithKline, Genentech, Celgene, Novartis, Pfizer, Prometheus, and Wyeth. BC has received honoraria for consultancy from Genentech, Bristol-Myers Squibb, CytRx, Prometheus, GlaxoSmithKline, and Merck; and has received payment for services on speakers' bureaus from Genentech, Bristol-Myers Squibb, and Prometheus and for educational lectures from Merck. KL has received grants from Roche. PB has received research support from Genentech during implementation of this trial; he has previously received grants from Bristol-Myers Squibb, Medimmune, and Merck and he is a member of the speakers' bureau for Bristol-Myers Squibb. MY was an employee of the trial sponsor Roche/Genentech with company stock options. IC is an employee and stockholder with Roche and has a combination therapy patent pending. LM and NC are employees of Genentech. IP received an institution grant from Genentech during implementation of this study. GAM has previously received research support grants from Millennium, Novartis, and Pfizer. AP, AD, and DK declare no competing interests.

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