Prognostic markers and tumour growth kinetics in melanoma patients progressing on vemurafenib

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Abstract

The BRAF inhibitor vemurafenib is an effective drug in patients with BRAF mutant metastatic melanoma, but resistance occurs after a median of 6 months. The anti-CTLA4-antibody, ipilimumab, is a standard first-line and second-line treatment option in Europe, with a median time to response of 2–3 months, but some patients show rapid clinical deterioration before that. The aim of this analysis was to identify prognostic markers for survival after failure of vemurafenib treatment to identify patients who have a sufficient life expectancy to respond to new immunotherapy treatments. We retrospectively analysed 101 consecutive unselected patients treated with vemurafenib for metastatic melanoma at a single institution. The association between clinical parameters and death within 3 months after cessation of vemurafenib (n=69) was assessed by binary logistic and Cox regression. Of the patients, 45% died within 3 months of progression on vemurafenib. Elevated baseline serum lactate dehydrogenase, absence of normalization of serum lactate dehydrogenase on vemurafenib therapy, performance status of at least 2 at progression and time from primary tumour to metastatic disease less than 5 years were identified as poor prognostic markers. In an exploratory tumour growth kinetics analysis (n=16), we found that following cessation of vemurafenib, approximately a third each showed a stable, decelerated or accelerated rate of tumour growth. Patients with these poor prognostic markers are unlikely to have sufficient life expectancy to complete ipilimumab treatment after failure with vemurafenib. Consideration needs to be given to the elective use of immunotherapy before patients become resistant to vemurafenib. This requires prospective randomized evaluation. Our tumour growth kinetics analysis requires confirmation; however, it may suggest that intermittent vemurafenib treatment should be investigated in clinical trials.

Introduction

Malignant melanoma is currently the ninth most common cancer in Europe, and incidence rates have been increasing worldwide (http://www.cancer.org; http://www.cancerresearchuk.org). Until recently, therapeutic options for patients with metastatic melanoma were limited, and 5-year survival rates of 10–20% largely reflected the natural history of the disease.

Major treatment advances have been made over the past 4 years, with the approval of five agents by the European Medical Association for the treatment of metastatic melanoma: in 2011, ipilimumab [1] – an anti-CTLA4 antibody – and vemurafenib [2] – a selective BRAF inhibitor – were approved. More recently, the BRAF inhibitor, dabrafenib, and the MEK inhibitor, trametinib, were also approved [3,4]. In addition, the new anti-PDL1-agent, pembrolizumab, and the anti-PD1-agent, nivolumab, have been approved.

Approximately 40% of melanomas harbour activating mutations in the protein kinase BRAF (Catalogue of Somatic Mutations in Cancer database). In metastatic melanoma patients harbouring a BRAF V600 mutation, treatment with vemurafenib results in improved progression-free survival (PFS) and overall survival (OS) compared with chemotherapy with dacarbazine, especially in patients with large tumour burden [5–7]. The most important limitation of BRAF-targeted therapy is the relatively short duration of the antitumour effect: the median PFS is 6–7 months and resistance is almost universal.

In contrast, immunotherapy has limited response rates of 10–20% with ipilimumab and 30% with pembrolizumab or nivolumab in metastatic melanoma; however, longterm control can be achieved in some patients independent of the BRAF mutation status, and better responses are often seen in patients with lower tumour burden [1, 8–10]. Until late 2013, ipilimumab was licensed only in Europe as a second or subsequent line of treatment.Therefore, until recently, in Europe, outside of the context of clinical trials, standard treatment for BRAF mutant melanoma consisted of vemurafenib followed by ipilimumab. Immunotherapy typically requires at least 8–12 weeks for a response to be seen, and consequently, patients embarking on this therapy need to have a life expectancy of at least 3 months. Clinical experience suggests that patients with rapidly growing disease after

progression on vemurafenib do not benefit from immunotherapy [9]. To date, apart from adequate performance status (PS), prognostic markers of potential benefit have not been defined. However, we have shown that a serum lactate dehydrogenase (LDH) level greater than two times the upper limit of normal is a negative prognostic marker for long-term benefit from ipilimumab [11].

We carried out a retrospective analysis to identify prognostic parameters and which patients have a sufficient life expectancy to respond to new immunotherapy treatments following progression on vemurafenib. We also carried out an exploratory comparative analysis of tumour growth kinetics during progression on vemurafenib and after the cessation of the drug but before the instigation of subsequent therapy.

Methods

Patients

In this retrospective case series, we analysed all patients who had been treated with and progressed on vemurafenib as a single-agent therapy for metastatic melanoma harbouring a BRAF V600 mutation at a single institution (Royal Marsden Hospital, London, UK) from March 2010 to May 2013. Patients were treated with vemurafenib within the BRAF inhibitor in Melanoma (BRIM-3) study (NCT01006980, enrolment period March 2010 until December 2010), the vemurafenib safety study (NCT01307397, enrolment period March 2011 until January 2013) as well as off trial after approval from the European Medicines Agency in December 2011. Exclusion criteria were treatment with vemurafenib for malignancies other than melanoma, cessation of vemurafenib for reasons other than progressive disease, such as toxicity, and lack of radiological confirmation of progressive disease, for example, death before radiological confirmation of progressive disease.

The primary aim of the analysis was the identification of the proportion of patients with early death (ED; defined as death ≤90 days after progression on vemurafenib) and their clinical characteristics compared with patients with late death (LD; defined as death >90 days after progression on vemurafenib). Any patient who was alive but had not yet reached at least 90 days of follow-up after cessation of treatment were

not included in the ED/LD analysis. We also described the PFS, OS and prognostic markers for OS for all patients analysed.

In an exploratory analysis, we measured tumour growth rates (TGRs) while patients were progressing on vemurafenib compared with those after stopping vemurafenib but before subsequent therapy or best supportive care was instigated. Only patients who had not/not yet started further treatment after stopping vemurafenib were included in the analysis. TGRs were measured as percentage per week and assessed using a modified form of Response Evaluation Criteria In Solid Tumours (RECIST 1.1) by two independent observers: TGRs were calculated as the change in the sum diameters of the target lesions and up to five new lesions between the most recent computed tomography (CT) scan before and at the time of stopping vemurafenib (TGR1) and between the CT scan at cessation of vemurafenib and the subsequent CT scan after stopping vemurafenib (TGR2). An increase of at least 5% per week between TGR1 and TGR2 (ΔTGR) was considered as accelerated, −5% >ΔTGR< 5% per week was considered stable and ΔTGR of up to −5% per week was considered as decelerated tumour growth. These cutoff values correspond to 20% tumour growth as per RECIST 1.1 – that is, progressive disease over a 4-week interval.

Statistical methods

All statistical analyses were carried out using the SPSS software, version 21.0 (SPSS Inc., Chicago, Illinois, USA). Univariate binary logistic regression analyses were used to test for any variable associated with ED. Univariate Cox regression analyses were used to test for any variable associated with OS. P-values less than 0.05 were considered significant. Variables with P less than 0.05 were entered into a multivariate model in a forward stepwise manner. PFS, postprogression survival (PPS) and OS analyses were carried out using the Kaplan–Meier method. PFS was defined as the time from the start of vemurafenib to progression. PPS was defined as the time from cessation of vemurafenib until death. OS was defined as the time from the start of vemurafenib until death. Patients alive at time of evaluation were censored at the last follow-up (cutoff 30 September 2013). Patients lost to follow-up were censored at the last follow-up.

Results

Clinical characteristics of patients with progression on vemurafenib

Between March 2010 and May 2013, 101 patients were treated with vemurafenib as a single agent for metastatic melanoma. At the time of evaluation, 69 (68%) had stopped vemurafenib for radiologically confirmed progressive disease (Table 1). This formed the case series population.

The patients were treated outside trials, within the vemurafenib safety study and within the BRIM-3 study (20, 70 and 10%, respectively). An overview of the clinical characteristics of all patients who progressed on vemurafenib is given in Table 2. The majority of the patients were treatment-naive before starting vemurafenib (70%): 21 (30%) patients had undergone at least one systemic treatment before vemurafenib, including two patients having being treated with ipilimumab. Before commencing vemurafenib, 70% of the patients had an elevated LDH, 16% had an Eastern Cooperative Oncology Group PS of at least 2 and 20% had central nervous system (CNS) disease; 26% of the patients had two of these three characteristics, and one patient had all three.

The median follow-up from starting vemurafenib was 20.4 months [95% confidence interval (CI): 15.4–25.4]. The median PFS was 4.1 months (95% CI: 2.7–5.6; Table 3). The median PPS was 3.2 months (95% CI 2.2–4.2). In nine of 69 patients the date of death was unknown because of loss to follow-up.

Post vemurafenib therapy

After progression on vemurafenib, 28 patients (41%) received further active therapy (Table 2); the same proportion of patients received best supportive care because of clinical deterioration, according to the his/her preference and/or as ipilimumab was not yet available at the time of progression. Thirteen patients (19%) were lost to follow-up. Out of the 56 patients in whom postvemurafenib management is known, 21 (38%) received subsequent treatment with ipilimumab and seven (12%) received other systemic therapies, but only eight of these patients were able to complete the full course of four doses of ipilimumab. This represents 14% of the total population in whom follow-up management is known and 38% of those who commenced ipilimumab. Seven died before completion of ipilimumab therapy, and three patients

discontinued ipilimumab for progression on treatment (Table 2). All patients who were treated with ipilimumab had a PS of up to 1 before starting this therapy. All patients who completed four doses of ipilimumab had a normal baseline LDH before starting vemurafenib or an elevated baseline LDH that normalized while on treatment with vemurafenib. One of those eight patients (12%) responded to ipilimumab, whereas the others showed progressive disease.

Prognostic impact of clinical features

We explored the prognostic significance of clinical parameters in all patients who progressed on vemurafenib: age, sex, CNS involvement at baseline, progression within the CNS, elevated LDH at baseline and at progression, absence of normalization of LDH on vemurafenib, PS at baseline and progression, time between diagnosis of the primary tumour and development of metastatic disease and stage of metastatic disease at the start of vemurafenib.

In multivariate analysis, PS of at least 2 at progression and time between diagnosis of the primary tumour and development of metastatic disease less than 5 years were associated with poor OS [hazard ratio (HR) 3.4 (95% CI: 1.7–7.0), P =0.001 and HR 6.4 (95% CI: 2.5–16.4), P <0.001, respectively; Table 4].

Patients with elevated baseline LDH levels showed a tendency towards poorer OS compared with those with normal LDH [HR 1.8 (95% CI: 1.0–3.3), P = 0.06]. However, the absence of normalization of LDH was an independent marker for poor OS [HR 8.6 (95% CI: 2.8–26.6), P <0.001; Table 4]. No difference in outcome was seen between patients with normal baseline LDH levels and those with elevated baseline LDH levels that normalized (Fig. 1). Approximately two-thirds of the patients (60%) with LDH normalization showed a second rise in LDH before subsequent progressive disease. In 87% of patients, progression occurred within the next 2 months, with a median time to progression of 29 days (range 0–168) from secondary abnormal LDH.

Prognostic markers for early death

Thirty-one of 69 patients (45%) died within 90 days (ED), whereas 30 patients (43%) survived longer than 90 days (LD; Table 2). Eight patients had not yet reached 90 days since stopping vemurafenib at the time of the analysis and were excluded from

the analysis.There were no differences between patients with ED/LD in relation to age, sex, stage of metastatic disease, prior treatment or PS at baseline. Elevated prevemurafenib baseline LDH [odds ratio (OR) 5.5 (95% CI: 1.3–23.0), P = 0.02], PS of at least 2 at the time of progression [OR 6.9 (95% CI: 1.6–29.4), P= 0.01] and time between diagnosis of the primary tumour and development of metastatic disease less than 5 years [OR 12.8 (95% CI: 1.9–83.3), P = 0.008] were correlated with ED (Table 4). All patients who had elevated baseline LDH before vemurafenib treatment and never showed normalization of their LDH (n =8) belonged to the ED group.
Influence of stopping of vemurafenib on tumour growth

We analysed the preprogression and postprogression tumour growth kinetics in 16 out of the 69 patients (three ED patients, 12 LD patients and one patient who had not yet reached 90 days of follow-up) in whom postprogression CT scanning was performed following the cessation of vemurafenib treatment and before the instigation of further active therapy or best supportive care. The reasons for the lack of evaluable CT were as follows: immediate initiation of a further treatment (n =15), palliative care without a further scan (n =27), external CT (n= 7), partial CT (n= 3) and no subsequent scan yet at the time of evaluation (n =1).

The median time between CT scans before and at the time of stopping vemurafenib was 8 weeks (range 4–12 weeks). The median time between CT scans at and after stopping vemurafenib was 5 weeks (range 4–14 weeks). The median TGRs before and after progression on vemurafenib were +6.1% per week (range − 3.5 to +156.5%) and + 14.7% per week (range +0.7 to +45.2%), respectively – that is, a 2.4-fold median increase in growth rate. Seven out of 16 patients (44%) showed an acceleration of tumour growth, with ΔTGR of at least 5% per week after stopping vemurafenib (Fig. 2a), whereas in five patients (31%), the tumour growth was stable (− 5%>ΔTGR<5% per week; Fig. 2b), and in four patients (25%), a deceleration in tumour growth was seen (ΔTGR≤−5% per week; Fig. 2c). The median OS for these three groups was 9.0 (range 2.0–13.0), 10.0 (range 3.0–20.0) and 11.0 (range 8.0–23.0) months, respectively. None of the patients with decelerated TGR had received immunotherapy before vemurafenib. Three out of 16 patients (two with decelerated TGR and one with accelerated TGR) received ipilimumab after vemurafenib. One each completed the full course of ipilimumab, but both showed subsequent progression.

Discussion

Our data demonstrate that a significant number of patients with progressive disease on vemurafenib do not have sufficient life expectancy to potentially complete subsequent treatment with ipilimumab or to even respond to new immunotherapy, and others have made a similar observation [12]. In this analysis, the median survival was 3.2 months, with 45% of patients dying within 3 months (ED) of stopping vemurafenib. An elevated baseline LDH, PS of at least 2 at progression and time between diagnosis of the primary tumour and development of metastatic disease less than 5 years were correlated with ED. All three parameters are well-known prognostic factors for metastatic melanoma.

Despite our limited sample size, these data suggest that if ipilimumab is to be administered after vemurafenib, then serious consideration needs to be given to a vemurafenib 'induction' strategy. For example, patients could receive vemurafenib until maximum response, normalization of poor prognostic factors or for an arbitrary period such as 8–16 weeks (two to four cycles), followed by immunotherapy utilizing immune checkpoint inhibition. In a recent small series, all 11 patients treated early with ipilimumab after two to four induction cycles of vemurafenib were alive at a median follow-up of 1 year, compared with seven out of eight patients who were reported to have died on switching to vemurafenib on progression [13]. This strategy of elective sequential hybrid therapy of targeted agent immunotherapy requires prospective evaluation. A current prospective trial investigates how BRAF/MEK inhibitor treatment and immunotherapy treatment should be sequenced in patients with advanced BRAF mutant melanoma (NCT02224781).

We have found that our patients with an elevated baseline LDH that normalized while on vemurafenib showed a significantly better OS than patients without normalization of this serum enzyme. The normalization of LDH may also be important when we try to predict who will benefit from postvemurafenib ipilimumab. Only eight patients (14%) out of the 56 patients in whom postvemurafenib treatment is known were able to receive all four doses of ipilimumab; normalization of LDH while on vemurafenib was a feature of those who were able to complete this immunotherapy. A previously reported case series of 28 patients treated with BRAF

inhibitors followed by ipilimumab suggested clinical baseline markers such as elevated baseline LDH, PS greater than 0 and CNS involvement to be indicators of failure of completion of second-line treatment [14]. The relationship between normalization of LDH and the ability of patients to receive or complete subsequent therapy is important as it impacts how we sequence new treatments and design elective sequential hybrid therapy.

Our exploratory study of TGR suggests that a majority of patients will show a steady or even an accelerated pace of disease after cessation of vemurafenib, but approximately one-third showed a deceleration of tumour growth. Although the accuracy of the method that we used is limited and the number of patients with accelerated tumour growth is likely to be underestimated in this selective analysis as a significant number had no subsequent CT scanning after stopping of vemurafenib, our findings imply that tumour evolutionary pressure is not always towards a more malignant phenotype. Preclinical data have demonstrated increased tumour proliferation of resistant tumours only in the presence of vemurafenib; however, following drug cessation, tumour regression was observed [15]. This finding is consistent with our tumour kinetic analysis, which shows that following cessation ofvemurafenib, 25% of patients had a deceleration of TGR. An obvious clinical consequence of this observation is that intermittent dosing of targeted agents may be a therapeutic strategy for some patients. Indeed, this approach has been shown to delay the onset of drug resistance in mice [15].

Rationally designed prospective clinical trials based on observations such as those presented here are urgently required to try and improve outcomes for patients by maximizing the relatively limited benefit from our existing therapies.

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TABLES AND FIGURES

Table 1: Overview of patients on vemurafenib

	n (% of total)
Total patients on vemurafenib	101
Cessation of vemurafenib at the time of evaluation	87 (86)
Cessation of vemurafenib for progressive disease	69 (68)
Cessation of vemurafenib for toxicities	6 (6)
Progressive disease not confirmed radiologically before cessation of vemurafenib	12 (12)
No cessation of vemurafenib at time of evaluation	14 (14)

Table 2: Clinical characteristics of all patients with progression on vemurafenib, those with early death (death ≤3 months after progression on vemurafenib) and those with late death (death >3 months after progression on vemurafenib)

	All	Early death	Late death
Total (n)	69	31	30
Baseline			
Age (range) (years)	52.7 (18–77)	52.4 (18–77)	52.3 (22–74)
Male [n (%)]	40 (58)	19 (61)	15 (50)
Female [n (%)]	29 (42)	12 (39)	15 (50)
Time from primary tumour to metastatic disease <5 years [n (%)]	53 (77)	29 (94)*	20 (67)
Systemic treatment before vemurafenib [n (%)]	21 (30)	9 (29)	10 (33)
Stage M1c [n (%)]	55 (80)	28 (90)	21 (70)
Normal LDH [n (%)]	21 (30)	4 (13)	14 (47)
Elevated LDH [n (%)]	48 (70)	27 (87)*	16 (53)
PS 0/1 [n (%)]	58 (84)	23 (74)	27 (90)
PS ≥2 [n (%)]	11 (16)	8 (26)	3 (10)
CNS involvement [n (%)])	14 (20	8 (26)	5 (17)
On treatment with vemurafenib			
LDH, normalized [n (%)]	38/48 (79)	19/27 (70)	16/16 (100)
LDH, not normalized [n (%)]	10/48 (21)	8/27 (30)	–
Elevated LDH at progression [n (%)]	34 (49)	19 (61)	12 (40)
PS ≥2 at progression [n (%)]	26 (38)	18 (58)*	5 (17)
Postprogression treatment [n (%)]	28 (41)	7 (23)**	20 (67)
Ipilimumab	21 (30)	5 (16)	14 (47)
Completed ipilimumab	8/21 (38)	–	8/14 (57)
Discontinued ipilimumab (PD/death)	10/21 (48)	5/5 (100)	5/14 (36)
Discontinued ipilimumab (toxicities)	2/21 (10)	–	1/14 (7)
Lost to follow-up	1/21 (5)	–	–

CNS, central nervous system; LDH, lactate dehydrogenase; n, number of patients; PD, progressive disease; PS, performance status.P-values indicate correlation between clinical variables and early death.(*P<0.05; **P<0.001.)

Table 3: Clinical and radiological outcome of all patients with progression on vemurafenib and of patients with early death (death ≤3 months after progression on vemurafenib) and late death (death >3 months after progression on vemurafenib)

	All	Early death	Late death
Total (n)	69	31	30
Clinical outcome			
1-year OS rate (95% CI) (%)	27 (15–39)	3 (0–9)	47 (29–66)
Median OS (95% CI) (months)	8.5 (6.7–10.2)	4.4 (3.7–5.2)	11.7 (10.2–13.1)
Median PFS (95% CI) (months)	4.1 (2.7–5.6)	3.7 (3.5–3.9)	6.4 (5.2–7.7)
Median PPS (95% CI) (months)	3.2 (2.2–4.2)	1.3 (0.6–2.0)	5.1 (3.9–6.3)
Radiological outcome			
Clinical benefit (CR, PR, SD) [n (%)]	60 (87)	26 (84)	29 (97)
Primary refractory disease [n (%)]	9 (13)	5 (16)	1 (3)
Best response after cycle 2 [n (%)]	54 (78)	29 (93)	20 (60)
Median time from BL to BR (95% CI) (months)	1.8 (1.2–7.1)	1.7 (1.2–7.1)	1.8 (1.2–5.5)
Median time from BR to PD (95% CI) (months)	2.0 (0.1–12.9)	1.9 (0.1–12.9)	3.7 (0.2–11.7)

BL, baseline; BR, best response; CI, confidence interval CR, complete remission; OS, overall survival; PD, progressive disease; PFS, progression-free survival; PPS,postprogression survival; PR, partial response; SD, stable disease.

Table 4: Prognostic factors for early death and overall survival:multivariate analysis

	OR	95% CI	*P*-value
Prognostic factors for early death			
Time from primary tumour to metastatic disease <5 years	12.82	1.93–83.33	0.008
PS at progression ≥2	6.86	1.60–29.42	0.010
Elevated LDH at baseline	5.48	1.31–23.01	0.020
	HR	95% CI	*P*-value
Prognostic factors for overall survival			
Time from primary tumour to metastaticdisease <5 years	6.41	2.49–16.39	<0.001
Normal LDH at baseline (reference)	1		
LDH, normalized	1.17	0.56–2.41	0.680
LDH, not normalized	8.62	2.80–26.61	<0.001
Overall			<0.001
PS at progression ≥2	3.45	1.70–7.02	0.001

Elevated LDH at baseline, LDH, normalized and not normalized, PS ≥2 at progression, time from primary tumour to metastatic disease ≥5 years were all entered into a multivariate model in a forward conditional selection manner. CI, confidence interval; HR, hazard ratio; LDH, lactate dehydrogenase; PS, performance status.

Figure 1: Overall survival curves of patients with normal baseline LDH and elevated baseline LDH with normalization and without normalization. Overall survival was defined as time from start of vemurafenib until death. Vertical lines indicate that the patients' data were censored. The median follow-up period on vemurafenib was 20.4 months. No significant difference in overall survival was observed between patients with normal baseline LDH and those with normalized LDH. CI, confidence interval; HR, hazard ratio; LDH, serum lactate dehydrogenase.

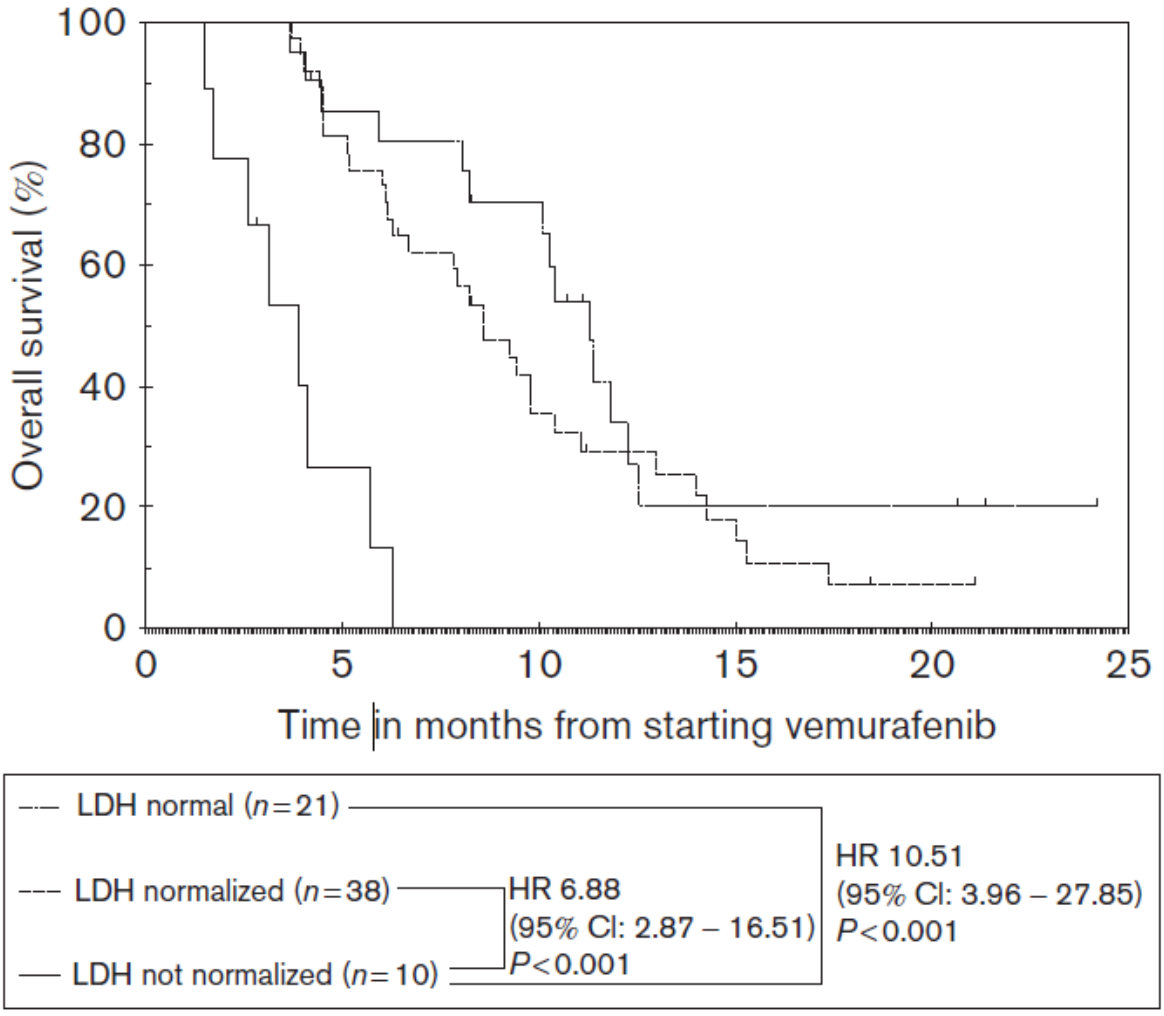

Figure 2: Tumour growth rates (TGR) before and after progression on vemurafenib. TGR1 [left side of the panels (a), (b) and (c)]: tumour growth rate before progression on vemurafenib; TGR2 [right side of panels (a), (b) and (c)]: tumour growth rate after progression on vemurafenib. Change in TGR (ΔTGR) was calculated as difference between TGR1 and TGR2. (a) Patients with accelerated TGR; (b) patients with stable TGR; (c) patients with decelerated TGR. Legends of panels (a), (b) and (c) include the sum of the target lesions in mm at the time of cessation of vemurafenib. ** Patients with early death.