Clinical Cancer Research

Phase Ib Study of Buparlisib plus Trastuzumab in Patients with HER2-Positive Advanced or Metastatic Breast Cancer That Has Progressed on Trastuzumab-Based Therapy

Cristina Saura[1], Johanna Bendell[3], Guy Jerusalem[6], Shaun Su[4], Qinhua Ru[4], Stefan De Buck[8], David Mills[8], Sophie Ruquet[9], Ana Bosch[10], Ander Urruticoechea[2], Joseph T. Beck[11], Emmanuelle Di Tomaso[12], David W. Sternberg[4], Cristian Massacesi[9], Samit Hirawat[5], Luc Dirix[7], and Jose Baselga[10]

Abstract

Purpose: Phosphoinositide 3-kinase (PI3K)/AKT/mTOR pathway activation in patients with HER2-positive ($HER2^+$) breast cancer has been implicated in *de novo* and acquired trastuzumab resistance. The purpose of this study was to determine the clinical activity of the PI3K inhibitor buparlisib (BKM120) in patients with $HER2^+$ advanced/metastatic breast cancer resistant to trastuzumab-based therapy.

Experimental Design: In the dose-escalation portion of this phase I/II study, patients with trastuzumab-resistant locally advanced or metastatic $HER2^+$ breast cancer were treated with daily oral doses of buparlisib and weekly intravenous trastuzumab (2 mg/kg). Dose escalation was guided by a Bayesian logistic regression model with overdose control.

Results: Of 18 enrolled patients, 17 received buparlisib. One dose-limiting toxicity of grade 3 general weakness was reported at the 100-mg/day dose level (the single-agent maximum tolerated dose) and this dose level was declared the recommended phase II dose (RP2D) of buparlisib in combination with trastuzumab. Common (>25%) adverse events included rash (39%), hyperglycemia (33%), and diarrhea (28%). The pharmacokinetic profile of buparlisib was not affected by its combination with trastuzumab. At the RP2D, there were two (17%) partial responses, 7 (58%) patients had stable disease ($\geq$6 weeks), and the disease control rate was 75%. Pharmacodynamic studies showed inhibition of the PI3K/AKT/mTOR and RAS/MEK/ERK pathways.

Conclusions: In this patient population, the combination of buparlisib and trastuzumab was well tolerated, and preliminary signs of clinical activity were observed. The phase II portion of this study will further explore the safety and efficacy of this combination at the RP2D. *Clin Cancer Res; 20(7); 1935–45.* ©*2014 AACR.*

Authors' Affiliations: [1]Medical Oncology Department, Vall d'Hebron University Hospital, Vall d'Hebron Institute of Oncology (VHIO), Barcelona, Spain; [2]Catalan Institute of Oncology, Barcelona, Spain; [3]Sarah Cannon Research Institute, Nashville, Tennessee; [4]Novartis Pharmaceuticals, East Hanover, NJ; [5]Novartis Pharmaceuticals, Florham Park, New Jersy; [6]C.H.U. Sart-Tilman, Liege; [7]Oncologisch Centrum AZ-St. Augustinus Oncology, Antwerp, Belgium; [8]Novartis Pharma AG, Basel, Switzerland; [9]Novartis Oncology, Paris, France; [10]Memorial Sloan–Kettering Cancer Centre, New York, New York; [11]Highlands Oncology Group, Fayetteville, Arkansas; and [12]Novartis Institutes for BioMedical Research, Cambridge, Massachusetts

Note: Supplementary data for this article are available at Clinical Cancer Research Online (http://clincancerres.aacrjournals.org/).

A. Bosch and J. Baselga were affiliated with Hospital Clínic Universitario, Valencia, Spain, and Vall d'Hebron, Barcelona, Spain, respectively, when this research was conducted.

Corresponding Author: Cristina Saura, Vall d'Hebron University Hospital, Passeig Vall d'Hebron 119, Edifici Maternoinfantil Planta 14, 08035 Barcelona, Spain. Phone: 0034 932746085; Fax: 0034 932746059; E-mail: csaura@vhebron.net

doi: 10.1158/1078-0432.CCR-13-1070

Introduction

HER2 is overexpressed in 15% to 30% of breast cancers, and is associated with aggressive disease and poor prognosis (1–3). Although use of the HER2 antibody trastuzumab has led to important clinical benefits for patients with HER2-positive ($HER2^+$) breast cancer, 50% to 74% of patients with metastatic disease do not respond to treatment (4, 5), and approximately 75% progress within a year (4). For patients who do not respond, or who progress on single-agent trastuzumab, treatment options include the HER2 small-molecule inhibitor lapatinib (6), combination treatment with trastuzumab and another HER2 antibody pertuzumab (7), or the use of trastuzumab emtansine (T-DM1; ref. 8), an antibody–drug conjugate. These treatment options have been shown to offer some additional benefit, with objective response rates of 22%, 24%, and 44%, respectively, and median progression-free survival times of 8.4, 5.5, and, 9.6 months, respectively (6–8). Despite these new therapeutic options, disease progression on

Translational Relevance

Although trastuzumab has provided important clinical benefits to patients with HER2-positive breast cancer, *de novo* or acquired resistance to HER2-directed therapy remains a major obstacle. Activation of the phosphoinositide 3-kinase (PI3K)/AKT/mTOR pathway is observed in approximately 75% of HER2$^+$ breast cancers. Inhibition of this pathway has been shown to restore sensitivity to trastuzumab in resistant breast cancer models. Importantly, preclinical models show that continued HER2 blockade is required for tumor regression in response to PI3K inhibition even after the development of trastuzumab resistance. Here, we show that the combination of the PI3K inhibitor buparlisib and trastuzumab was well tolerated, and displayed preliminary clinical activity in patients with advanced or metastatic trastuzumab-resistant HER2$^+$ breast cancer. Clinical responses to buparlisib and trastuzumab were reported in patients with PI3K pathway–activated tumors, and evidence of inhibition of both the PI3K/AKT/mTOR and RAS/MEK/ERK signaling pathways in patients treated with this combination was observed.

HER2-directed therapy is experienced by most patients, and new strategies are needed to delay or overcome the onset of tumor progression. In addition, the improved extracranial disease control and extended survival of patients treated with HER2-directed therapies, such as trastuzumab, are associated with an increased incidence of relapse within the brain (9), and thus there is a particular need for targeted therapies that can be safely and successfully combined with trastuzumab; can penetrate the blood–brain barrier; and can delay or control relapse at this site.

The phosphoinositide 3-kinase (PI3K)/AKT/mTOR pathway is one of the most frequently dysregulated signaling pathways in cancer (10) and is important for the oncogenic function of HER2 (11). Activation of the PI3K/AKT/mTOR pathway—defined as mutation or amplification of the *PIK3CA* gene, which encodes the p110α catalytic subunit of PI3K; loss of PTEN protein expression; or overexpression of AKT—was identified in 75% of HER2$^+$ breast cancers in one study (12). Taking several studies into consideration, a recent review reported *PIK3CA* alterations occur in 20% to 25% of HER2$^+$ breast cancers, PTEN alterations in 30% to 40%, and AKT alterations in 0% (13). Tumors with PTEN loss, which have been shown to be reliant on the p110β subunit of PI3K (14), have been associated with poor clinical outcome in patients with breast cancer (15). In addition, evidence suggests a direct link between trastuzumab resistance and PI3K/AKT/mTOR pathway activation through either PTEN loss or activating *PIK3CA* mutations (16–18). Furthermore, inhibition of the PI3K/AKT/mTOR pathway can restore sensitivity to trastuzumab in trastuzumab-resistant HER2$^+$ breast cancer xenograft models (19–21).

Buparlisib (BKM120) is an oral pan-PI3K inhibitor that inhibits all four isoforms of class I PI3K (α, β, δ, and γ) with at least 50-fold selectivity compared with its activity against other proteins or lipid kinases (22). Single-agent buparlisib has demonstrated antitumor activity in a variety of cell lines, including HER2$^+$ MDA453 and BT474 cells, and in xenograft models from cancers with or without alteration in *PIK3CA* and/or PTEN (20, 22). In a murine model of trastuzumab-resistant HER2$^+$ breast cancer generated through long-term treatment of trastuzumab-sensitive BT474 cells, single-agent buparlisib inhibited tumor proliferation, but only the combination of trastuzumab and buparlisib was enough to induce tumor regression (20). In a phase I, dose-finding study, the maximum tolerated dose (MTD) of single-agent buparlisib was established at 100 mg/day in patients with advanced solid tumors, including breast cancer (CBKM120X2101/NCT01068483; ref. 23, 24). In this study, buparlisib was generally well tolerated and showed encouraging single-agent activity (23, 24).

The high frequency of PI3K/AKT/mTOR pathway alterations in HER2$^+$ breast cancer, combined with the role of the pathway in resistance to trastuzumab, and preclinical evidence that combined PI3K inhibition restores sensitivity to trastuzumab and has greater antitumor activity than single-agent PI3K inhibition, supports the rationale for clinical evaluation of combined targeting of PI3K and HER2 in patients with HER2$^+$ breast cancer. Here, we report on the phase I dose-escalation portion of this phase I/II study, the primary aim of which was to determine the MTD and/or recommended phase II dose (RP2D) of continuous daily buparlisib in combination with weekly trastuzumab in patients with HER2$^+$ locally advanced or metastatic breast cancer whose disease had progressed on trastuzumab-based therapy.

Patients and Methods

Patient population

Patients aged ≥18 years with HER2$^+$ metastatic breast cancer and evidence of disease progression as per Response Evaluation Criteria In Solid Tumors version 1.0 (RECIST v1.0) and resistance to trastuzumab were included. Resistance to trastuzumab was defined as recurrence while on or within 4 weeks since the most recent infusion for patients who received trastuzumab for metastatic disease (or within 12 months for patients who received trastuzumab as adjuvant therapy). Resistance to T-DM1 was considered as equivalent to trastuzumab for the purpose of defining study eligibility. Other key inclusion criteria included World Health Organization (WHO) performance status ≤2; provision of a representative tumor sample for the determination of tumor molecular status; ≥1 but ≤4 prior lines of HER2-directed therapy (trastuzumab, lapatinib, and/or T-DM1); ≤4 lines of prior cytotoxic chemotherapy.

Patients were excluded if they had received previous treatment with a PI3K inhibitor or had a contraindication to trastuzumab treatment. Patients with untreated brain metastases were also excluded; however, patients were

eligible if the brain metastases were previously treated, they had completed therapy (including radiation and/or surgery) >4 weeks earlier, and were clinically stable (as determined by the investigator) with respect to the tumor at the time of study entry. Other exclusion criteria included a medically documented history of, or active, major mood or psychiatric disorder; Common Terminology Criteria for Adverse Events (CTCAE) ≥grade 3 anxiety; poorly controlled diabetes mellitus (HbA_{1c} >8%); and fasting plasma glucose >140 mg/dL or >7.8 mmol/L.

Approval was obtained from the ethics committees of participating institutions and regulatory authorities. All participating patients provided written informed consent and agreed to comply with the protocol. The study was conducted in accordance with the Declaration of Helsinki and guidelines for Good Clinical Practice as defined by the International Conference on Harmonization.

Study design and buparlisib dose escalation

This was a phase Ib/II, multicenter, open-label, dose-escalation study of buparlisib in combination with trastuzumab. Here, we report on the phase Ib portion only, the primary objective of which was to determine the MTD and/or RP2D of buparlisib in combination with trastuzumab in this patient population. Patients received oral buparlisib as a once-daily hard gelatin capsule starting on day 1 of a continuous 28-day cycle. The initial starting dose of buparlisib for this dose-escalation study was 50 mg/day.

The starting dose of 50 mg/day was selected based on the results of the Bayesian logistic regression model with overdose control (BLRM EWOC), in which the highest possible starting dose derived from the prior distribution of dose-limiting toxicity (DLT) rates—taking into account the potential higher toxicity when buparlisib and trastuzumab are combined compared with the single agent—was 50 mg/day (to achieve a conservative safety margin, a median 50% increase in odds of DLT was assumed). This starting dose is also supported by the results of the first-in-man single-agent study of buparlisib (CBKM120X2101/NCT01068483), in which no DLTs were observed at doses ≤50 mg/day (23).

A 4-mg/kg loading dose of i.v. trastuzumab was administered on day-7 at the discretion of the investigator if clinically indicated. This was followed by fixed weekly i.v. doses of 2 mg/kg trastuzumab starting on cycle 1 day 1. Treatment continued until disease progression, unacceptable toxicity, investigator decision, or patient withdrawal of consent.

Buparlisib dose escalation was guided by an adaptive BLRM EWOC (25), and each dose cohort consisted of newly enrolled patients, among whom three to six must be evaluable for dose determination. Patients were considered evaluable as part of the dose-determining set if they experienced a DLT in the first cycle or they received buparlisib for ≥21 days, received all four scheduled doses of trastuzumab, and completed all safety evaluations required for dose-determining decisions in the first cycle. Dose-escalation beyond the single-agent MTD of 100 mg/day buparlisib (23) was not permitted, and intrapatient dose escalation was not permitted during the first four cycles of treatment. Before a dose could be declared to be the MTD, at least 15 evaluable patients had to be included in the dose-escalation part of the study, with at least 6 evaluable patients treated at the estimated RP2D (MTD or lower dose) level for one treatment cycle. The MTD was defined as the highest drug dose that does not cause medically unacceptable DLTs during the first cycle of treatment in more than 33% of treated patients. DLTs were defined as an adverse event or laboratory abnormality that were considered to be related to buparlisib treatment; met any of the CTCAE criteria outlined in Supplementary Table S1; occurred <28 days following the first dose of buparlisib; and were considered unrelated to the disease, disease progression, intercurrent illness, or concomitant medications.

Statistical analysis

A two-parameter BLRM was fitted on the cycle 1 DLT data (i.e., absence or presence of DLT) accumulated throughout the dose escalation to model the dose–toxicity relationship of buparlisib in combination with trastuzumab. After each cohort of patients was treated and evaluated for DLTs, the next recommended dose of buparlisib to be administered in combination with trastuzumab was the one with the highest posterior probability of DLT in the target toxicity interval (16%–33%, among the doses fulfilling the overdose criterion; <25% chance of excessive toxicity).

Safety assessments

Routine clinical and laboratory assessments, including hematology, coagulation, and biochemistry assessments, were conducted at baseline and at regular intervals throughout the study. Other safety assessments included glucose monitoring by urine dipstick test, blood markers of glucose homeostasis, and patient self-rating depression and anxiety questionnaires [PHQ-9 (26) and GAD-7 (27), respectively]. Adverse events were recorded continuously from the start of study treatment until 28 days after treatment discontinuation, and were graded using the National Cancer Institute's CTCAE v3.0. To be evaluable as part of the safety set, patients must have received at least one dose of buparlisib or trastuzumab and had at least one valid postbaseline assessment.

Efficacy assessments

Tumor radiologic response was assessed by computed tomography scan (preferred) or magnetic resonance imaging according to RECIST v1.0 at baseline and every 8 weeks thereafter until disease progression or end of treatment. Assessments at the end of treatment were only performed if the previous evaluation was >21 days earlier. Complete and partial responses (CR and PR) were defined as at least two determinations of CR or PR ≥4 weeks apart before progression; stable disease (SD) and prolonged SD were defined as at least one SD assessment or better ≥6 weeks and ≥24 weeks, respectively, after the start of treatment and not qualifying for CR or PR; progressive disease was defined as progression and not qualifying for CR, PR, or SD; all other cases were considered unknown.

Pharmacokinetic profiling

Blood samples for full pharmacokinetic profiling of buparlisib were collected on days 1 and 8 of cycle 1 and day 1 of cycle 2 at the following time points: predose, 0.5 hours, 1 hour, 1.5 hours, 2 hours, 3 hours, 4 hours, 6 hours, 8 hours, and 24 hours postdose. Serum levels of buparlisib were analyzed by liquid chromatography-tandem mass spectrometry.

PI3K pathway status determination

Archival biopsies of primary tumors were collected from all patients for the analysis of PI3K pathway activation status. Tumor tissue was assessed for the presence of *PIK3CA* mutations and *PTEN* mutations as determined by SNaPshot genotyping assay or Sanger sequencing, and loss of PTEN protein expression, as defined by an immunohistochemistry H-score <50.

Pharmacodynamic assessments

Changes from baseline in levels of phosphorylated (Ser240)-S6 ribosomal protein (pS6) and phosphorylated eukaryotic translation initiation factor 4E-binding protein 1 (4E-BP1) were determined using 3-mm diameter, full-thickness skin biopsies obtained during screening and between 3 and 6 hours postdose on day 1 of cycle 2. Paired (pre- and post-treatment) tumor biopsies collected at baseline and on day 28 of cycle 1 were analyzed for changes from baseline in pAKT, p4E-BP1, Ki-67, pERK, and pMEK by immunohistochemistry, using the following rabbit monoclonal antibodies: pAKT473 (Cell Signaling Technology; cat. #3787), p4E-BP1 (Cell Signaling Technology; cat. #2855), Ki-67 (Ventana; cat. #790-4288), Phospho-p44/42 MAPK (Erk1/2; Thr202/Tyr204; Cell Signaling Technology; cat. #4376), pMEK (Cell Signaling Technology; cat. #2338).

Results

Patient characteristics

Eighteen female patients from seven centers located in three countries (Belgium, Spain, and USA) were enrolled into the dose-escalation portion of this study between May 27, 2010, and April 28, 2011. One patient received only a loading dose of trastuzumab, but did not receive buparlisib; this patient was evaluable for safety but not for dose determination or tumor response. Of the 17 remaining patients who received at least one dose of buparlisib, 5 received buparlisib 50 mg/day and 12 received buparlisib 100 mg/day. All 17 patients who received buparlisib had received prior antineoplastic therapy (chemotherapy, hormonal therapy, or targeted therapy), which included HER2-directed therapy. The median number of prior antineoplastic regimens, HER2-directed therapies, and cytotoxic chemotherapies for all patients were 4 (range, 1–8), 3 (range, 1–5), and 3 (range, 1–4), respectively (Table 1).

Activation of the PI3K pathway was detected in archival primary tumor samples from 7 (41%) patients, of which 5 (29%) had a *PIK3CA*-activating mutation (one E545G, one E545K, one K1063E, and two H1047R) and 3 (18%) had a *PTEN* mutation (one P285S, one D375N, and one Q97X). One tumor sample had both an activating *PIK3CA* and a *PTEN* mutation. Loss of PTEN was not identified in any of the tumor samples.

Dose escalation and RP2D

Of the 18 patients enrolled on the study, 1 patient did not receive buparlisib and 2 patients discontinued without experiencing DLTs in the first cycle and before meeting the minimum drug exposure, as defined in the protocol, for dose determination (1 patient discontinued due to disease progression after receiving 14 doses of buparlisib and four doses of trastuzumab; and another patient discontinued due to disease progression after receiving 21 doses of buparlisib and three doses of trastuzumab). Therefore, 15 patients (4 patients in the buparlisib 50-mg/day cohort and 11 patients in the buparlisib 100-mg/day cohort) were evaluable for dose determination. Only one DLT of grade 3 general weakness for >7 consecutive days was observed in a patient treated with buparlisib 100 mg/day. As dose escalation beyond the single-agent MTD of 100 mg/day was not permitted, the MTD of buparlisib in combination with trastuzumab as per the BLRM EWOC was not reached (Supplementary Fig. S1). The RP2D of buparlisib in combination with i.v. trastuzumab 2 mg/kg weekly was declared as 100 mg/day.

Patient disposition

As of June 22, 2012, all patients had discontinued treatment: 13 (77%) patients treated with buparlisib discontinued due to disease progression, including 1 who developed a new lesion in the central nervous system (CNS); 1 (6%) patient had a suspected study drug–related grade 3 allergic reaction; and 3 (18%) patients withdrew consent (Supplementary Table S2). The median exposure to study treatment was 10.9 weeks (range, 1.0–41.0); 8.0 weeks (range, 4.0–15.9) in the 50-mg/day cohort; and 13.9 weeks (range, 4.0–41.0) in the 100-mg/day cohort (Supplementary Table S3).

Safety and tolerability

The most common study drug–related all-grade adverse events in the full cohort were rash in 7 (39%) patients, hyperglycemia in 6 (33%) patients, and diarrhea in 5 (28%) patients (Table 2; Supplementary Table S4). No grade 3/4 study drug–related adverse events were experienced by patients treated with buparlisib 50 mg/day. The most common grade 3/4 study drug–related adverse events in patients treated with buparlisib 100 mg/day were alanine aminotransferase increase in 3 (25%) patients, and hyperglycemia, aspartate aminotransferase increase, and asthenia in 2 (17%) patients each. Grade 3 study drug–related psychiatric adverse events were experienced by 2 (17%) patients receiving buparlisib 100 mg/day: 1 patient experienced both grade 3 anxiety and grade 3 altered mood; the other patient experienced grade 3 affective disorder.

Serious adverse events suspected to be study drug related occurred in 2 patients treated at 100 mg/day (Supplementary Table S5): 1 patient experienced grade 3 asthenia and grade 3 altered mood; and 1 patient experienced grade 3 asthenia, grade 2 and grade 3 affective disorder, and grade 2

Table 1. Patient baseline characteristics and tumor status (full analysis set)

	All ($N = 17$)	50 mg/day ($n = 5$)	100 mg/day ($n = 12$)
Median age, y (range)	47 (28–70)	47 (41–55)	47 (28–70)
≥65 years, *n* (%)	1 (6)	0	1 (8)
Female, *n* (%)	17 (100)	5 (100)	12 (100)
Postmenopausal, *n* (%)	10 (59)	3 (60)	7 (58)
WHO performance status, *n* (%)			
0	8 (47)	2 (40)	6 (50)
1	9 (53)	3 (60)	6 (50)
Median number of prior antineoplastic regimens (range)	4 (1–8)	4 (3–5)	4 (1–8)
Median number of prior cytotoxic chemotherapy (range)	3 (1–4)	3 (2–4)	3 (1–4)
Median number of prior HER2-directed therapies (range)[a]	3 (1–5)	3 (1–4)	3 (1–5)
Setting of last trastuzumab treatment, *n* (%)			
Adjuvant	5 (29)	2 (40)	3 (25)
Metastatic	12 (71)	3 (60)	9 (75)
Hormonal status, *n* (%)			
ER and/or PgR positive	8 (47)	2 (40)	6 (50)
ER and PgR negative	9 (53)	3 (60)	6 (50)
Most common site of metastases, *n* (%)			
Nodes	10 (59)	4 (80)	6 (50)
Bone	9 (53)	2 (40)	7 (58)
Liver	9 (53)	2 (40)	7 (58)
Lung	8 (47)	1 (20)	7 (58)
Brain	1 (6)	0	1 (8)
Skin	1(6)	1 (20)	0
Others	4 (24)	2 (40)	2 (17)
Mutational status of tumor			
Activating mutation of *PIK3CA*	5 (29)	1 (20)	4 (33)
PTEN mutation	3 (18)	1 (20)	2 (17)
Total patients with activated PI3K pathway[b]	7 (41)	2 (40)	5 (42)

Abbreviations: ER, estrogen receptor; PgR, progesterone receptor.

[a]Includes HER2-directed agents defined for eligibility determination (trastuzumab, T-DM1, and lapatinib), as well as experimental HER2-directed agents that were not considered for eligibility purposes.

[b]PI3K pathway activation is defined as *PIK3CA* mutation, and/or *PTEN* mutation, and/or PTEN null or low expression by immunohistochemistry (H-score <50).

depression. There were two on-treatment deaths during the study, both of which occurred after discontinuation of buparlisib: 1 patient who received buparlisib 50 mg/day died of disease progression 12 days after receiving the last dose of buparlisib; the other patient had received buparlisib 100 mg/day and died of respiratory failure 14 days after receiving the last dose of buparlisib. The latter patient was initiated on carboplatin and gemcitabine in combination with trastuzumab immediately after study treatment discontinuation, and the respiratory failure that occurred 2 weeks thereafter was suspected to be related to the study indication and the subsequent chemotherapy. Neither death was suspected to be buparlisib related.

Pharmacokinetic analysis

All 17 patients who received buparlisib were evaluable for pharmacokinetics analysis. The buparlisib pharmacokinetic profile from day 8 of cycle 1 shows that it is rapidly absorbed following administration with a median time to reach peak plasma concentration of 1.3 hours (range, 0.5–2.0) and 1.5 hours (range, 1.0–4.0) in the 50-mg/day and 100-mg/day cohorts, respectively (Fig. 1; Supplementary Table S6). After reaching peak drug concentration (C_{max}), the level of buparlisib in both the 50-mg/day and 100-mg/day cohorts decreased in a biexponential manner with a mean effective half-life ($T_{1/2,acc}$) at day 8 of 38.4 and 43.3 hours, respectively. The pharmacokinetics of buparlisib when administered in combination with trastuzumab was, therefore, similar to that reported previously with single-agent buparlisib (23).

Clinical activity

Among the 17 patients evaluable for response, PRs were observed in 2 patients treated with buparlisib 100 mg/

Table 2. Summary of adverse events suspected to be study-drug related (safety set)

	All ($N = 18$[a])		50 mg/day ($n = 5$)		100 mg/day ($n = 12$)	
Adverse event	**All *n* (%)**	**G3/4 *n* (%)**	**All *n* (%)**	**G3/4 *n* (%)**	**All *n* (%)**	**G3/4 *n* (%)**
Any	16 (89)	8 (44)	4 (80)	0	12 (100)	8 (67)
Rash	7 (39)	1 (6)	1 (20)	0	6 (50)	1 (8)
Hyperglycemia	6 (33)	2 (11)	1 (20)	0	5 (42)	2 (17)
Diarrhea	5 (28)	0	1 (20)	0	4 (33)	0
Asthenia	4 (22)	2 (11)	0	0	4 (33)	2 (17)
Mood altered	4 (22)	1 (6)	2 (40)	0	2 (17)	1 (8)
Nausea	4 (22)	0	2 (40)	0	2 (17)	0
Pruritus	4 (22)	0	1 (20)	0	3 (25)	0
Alanine aminotransferase increased	3 (17)	3 (17)	0	0	3 (25)	3 (25)
Aspartate aminotransferase increased	3 (17)	2 (11)	0	0	3 (25)	2 (17)
Stomatitis	3 (17)	0	0	0	3 (25)	0
Vomiting	3 (17)	0	2 (40)	0	1 (8)	0
Affective disorder	2 (11)	1 (6)	0	0	2 (17)	1 (8)
Anxiety	1 (6)	1 (6)	0	0	1 (8)	1 (8)
Blood glucose increased	1 (6)	1 (6)	0	0	1 (8)	1 (8)
Hypersensitivity	1 (6)	1 (6)	0	0	1 (8)	1 (8)
Photosensitivity reaction	1 (6)	1 (6)	0	0	1 (8)	1 (8)

NOTE: The table includes all study drug–related adverse events that occurred in >2 patients and all adverse events that occurred at G3 or G4.

Abbreviation: G, grade.

[a]One patient received loading dose trastuzumab, but no buparlisib. No study drug–related adverse events were observed in this patient.

day. One of the patients who achieved a PR had received one prior cytotoxic therapy and one prior HER2-directed therapy in the adjuvant setting; the other had received four prior lines of cytotoxic therapy, and five prior lines of HER2-directed therapy, and the last line of trastuzumab-based treatment was in the metastatic setting. Both patients had PI3K pathway–activated tumors (Table 3, Fig. 2). SD (≥6 weeks) as best overall response was achieved

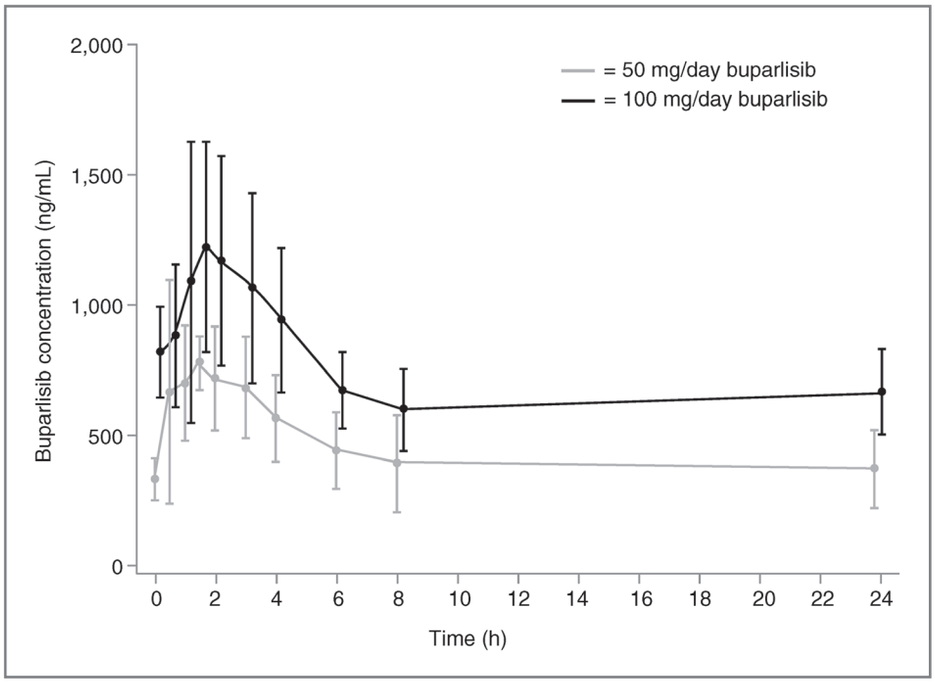

Figure 1. Plasma pharmacokinetic profile of buparlisib in blood serum on cycle 1 day 8. Dots, mean plasma concentrations; error bars, standard deviation of the mean.

Table 3. Best overall response (full analysis set)

Best overall response (RECIST)	All ($N = 17$)	50 mg/day ($n = 5$)	100 mg/day ($n = 12$)
CR, *n* (%)	0	0	0
PR, *n* (%)	2 (12)	0	2 (17)
SD, *n* (%)	8 (47)	1 (20)	7 (58)
SD ≥24 weeks, *n* (%)	1 (6)	0	1 (8)
PD, *n* (%)	5 (29)	2 (40)	3 (25)
Unknown, *n* (%)	2 (12)	2 (40)	0
DCR (CR or PR or SD), *n* (%)	10 (59)	1 (20)	9 (75)
90% CI for DCR[a]	(36.4–78.8)	(1.0–65.7)	(47.3–92.8)
CBR (CR or PR or SD ≥24 weeks), *n* (%)	3 (18)	0	3 (25)
90% CI for CBR	(5.0–39.6)	(0.0–45.1)	(7.2–52.7)

CI, confidence interval; PD, progressive disease.
[a]90% CIs for DCR and CBR were obtained using the exact binomial 90% CI test.

by 7 (58%) patients in the 100-mg/day cohort, 3 of whom had PI3K pathway–activated tumors, and by one (20%) patient without a PI3K pathway–activated tumor in the 50-mg/day cohort. Prolonged SD (≥24 weeks) was experienced by 1 patient with HER2$^+$, hormone receptor–negative breast cancer with an activating *PIK3CA* mutation (H1047R) receiving 100 mg/day of buparlisib. Two patients in the 50-mg/day cohort had an unknown response as they discontinued treatment before the first RECIST evaluation due to withdrawal of consent and symptomatic progression of disease. The disease control rate (DCR; CR, PR, or SD ≥6 weeks) was 59% (75% in the 100-mg/day cohort and 20% in the 50-mg/day cohort); and the clinical benefit rate (CBR; CR, PR, or SD ≥24 weeks) was 18% (25% in the 100-mg/day cohort and 0% in the 50-mg/day cohort).

Pharmacodynamic assessments

Pre- and posttreatment skin biopsies were available for 3 patients treated at buparlisib 50 mg/day and 3 patients treated at buparlisib 100 mg/day, and were evaluated for levels of pS6 and p4E-BP1. Reductions in pS6 were noted in four of the six evaluated paired biopsies, and the largest reduction was observed in a patient in the 100-mg/day cohort and who obtained a PR. In contrast, with the exception of 1 patient, treatment with buparlisib was not

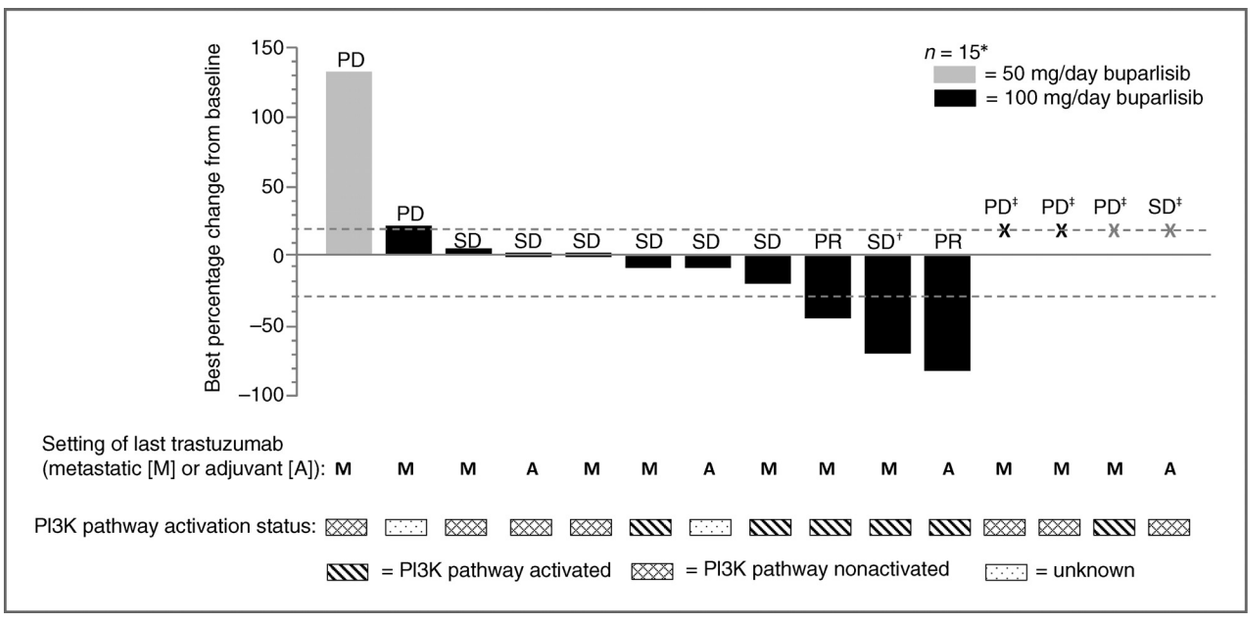

Figure 2. Waterfall plot of best percentage change from baseline in sum of longest diameters and best overall response by PI3K pathway activation status. PD, progressive disease. *, Patients with missing best percentage change from baseline and unknown overall response are not included; †, unconfirmed PR; ‡, patients with missing best percentage change from baseline.

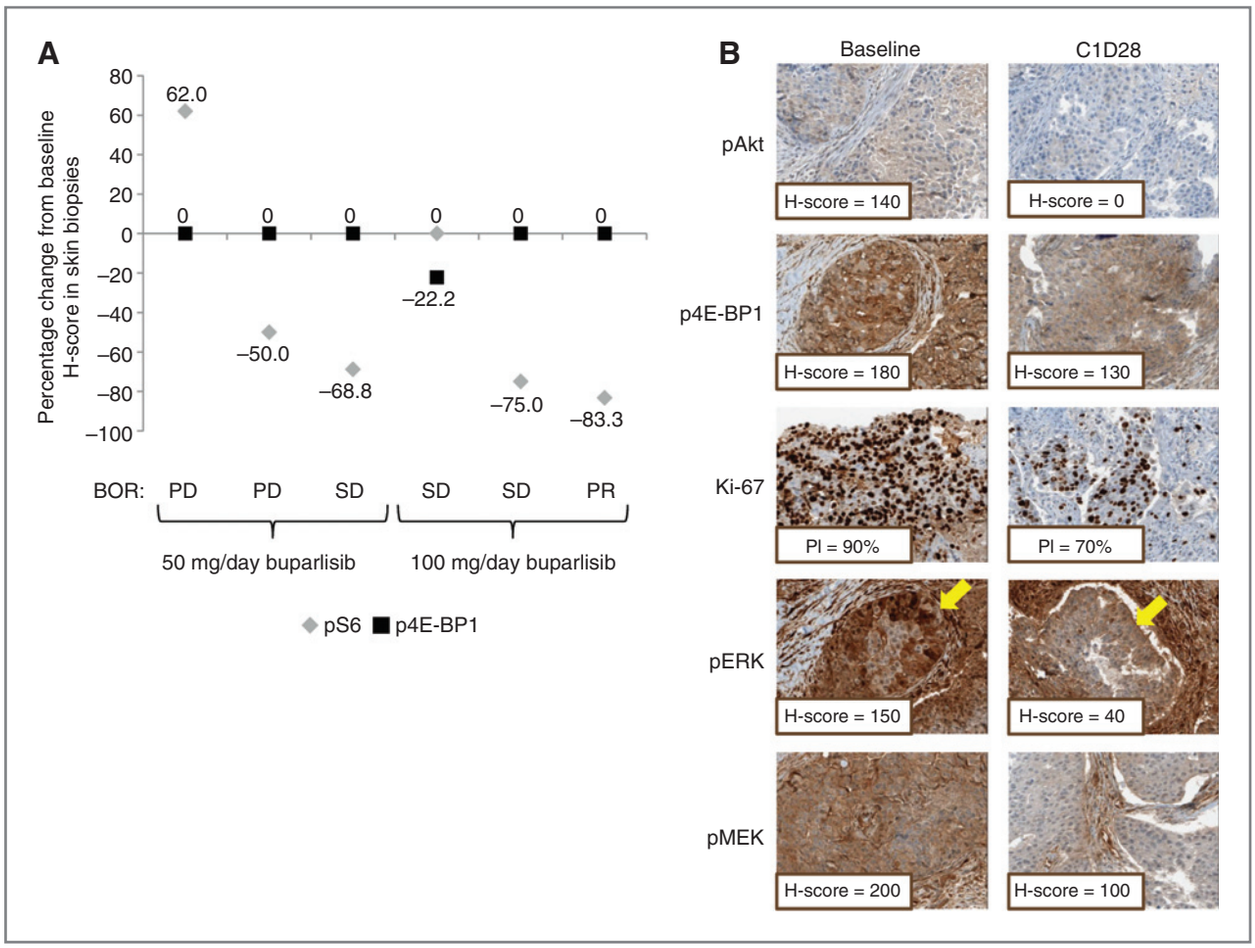

Figure 3. Changes from baseline in biomarkers in skin and tumor. A, percentage change from baseline H-score in skin biopsy biomarkers: pS6 and p4E-BP1. B, inhibition of the PI3K/AKT/mTOR and RAS/RAF/MEK pathway and reduction in cellular proliferation in tumor tissue following 1 cycle of treatment. 4E-BP1, eukaryotic translation initiation factor 4E-binding protein 1; BOR, best overall response; C, cycle; D, day; p, phosphorylated; PD, progressive disease; PI, proliferation index. Arrows point to tumor cells with changes in expression.

associated with a reduction in levels of p4E-BP1 in the skin (Fig. 3).

Analysis of a paired tumor biopsy taken at baseline and at cycle 1 day 28 from a patient with a non–PI3K pathway-activated tumor who received 100 mg/day of buparlisib and had a 75% reduction in pS6 in the skin, revealed a 100% reduction in pAKT, a 28% reduction in p4E-BP1, a 20% reduction in tumor cell proliferation (Ki-67), a 73% reduction in pERK, and a 50% reduction in pMEK (Fig. 3) following buparlisib/trastuzumab treatment. The best overall response in this patient was SD, and the patient remained on treatment for 3 months.

Discussion

This dose-escalation study provides evidence for the feasibility of combining buparlisib with a fixed weekly dose of trastuzumab in patients with locally advanced or metastatic HER2$^+$ breast cancer that has progressed following trastuzumab-based therapy. As only one DLT was observed at the 100-mg/day dose of buparlisib, and dose escalation beyond 100 mg/day (the single-agent MTD) was not permitted, the MTD of buparlisib in combination with trastuzumab was not reached. The RP2D of buparlisib in combination with trastuzumab was declared as buparlisib 100 mg/day and trastuzumab 2 mg/kg weekly.

The combination of buparlisib and trastuzumab was generally well tolerated, and the safety profile was similar to that reported previously with single-agent buparlisib (23) and with other PI3K inhibitors (28–32). The pharmacokinetic profiles of buparlisib at both the 50-mg/day and 100-mg/day dose levels when administered in combination with trastuzumab were similar to those reported previously by Bendell and colleagues (23) for single-agent buparlisib. This observation indicates that the drug disposition of buparlisib is not affected by its combination with trastuzumab.

Hyperglycemia, which has been reported here and in some other trials of PI3K/AKT/mTOR pathway inhibitors (23, 33–35), is a likely on-target effect of PI3K inhibition, as

this signaling axis mediates the actions of insulin, including glucose transport and glycogen synthesis (36–39); thus, PI3K inhibition might be expected to cause an increase in blood glucose and the compensatory release of insulin and C-peptide from pancreatic β cells (39). The hyperglycemia observed in this trial was generally controlled through the use of concomitant glucose-lowering medications, such as metformin and insulin and/or buparlisib dose interruption and reduction.

A recurrent side effect associated with buparlisib treatment was the occurrence of psychiatric adverse events, and this observation is likely associated with penetration of buparlisib across the blood–brain barrier and inhibition of PI3K/AKT/mTOR signaling in the CNS (40, 41). In separate studies, dysregulation of the PI3K pathway has been associated with changes in serotonin levels and in psychiatric disturbances, such as anxiety and depression (42–44). In the current study, the median time-to-first occurrence of psychiatric adverse events (regardless of study drug relationship) was 7.1 weeks (7.1 weeks in the 50-mg/day cohort and 6.1 weeks in the 100-mg/day cohort). Psychiatric adverse events were generally well managed through buparlisib dose modification and the use of concomitant treatments, such as benzodiazepine derivatives and selective serotonin reuptake inhibitors. Two patients (11%) developed grade 3 suspected study drug–related psychiatric adverse events and no patients permanently discontinued treatment due to such events.

The ability of buparlisib to cross the blood–brain barrier and inhibit the PI3K/AKT/mTOR pathway, combined with evidence of antitumor activity in the brain of a mouse model of HER2$^+$ metastatic breast cancer (40) as well as preliminary clinical activity in patients with brain metastases treated with single-agent buparlisib (23, 41), support the evaluation of buparlisib in combination with trastuzumab in patients with HER2$^+$ breast cancer and brain metastases. In the current trial, 1 patient (two prior lines of cytotoxic chemotherapy and two prior lines of HER2-directed therapy) had a baseline brain lesion (nontarget). This patient remained on treatment for 49 days, and achieved an unconfirmed PR in target lesions (liver and lymph nodes). An additional arm of this trial will evaluate buparlisib in combination with trastuzumab and capecitabine in patients with HER2$^+$ breast cancer brain metastases.

Biomarker analysis of paired pre- and posttreatment skin biopsies and tumor biopsies, as well as the occurrence of hyperglycemia, demonstrates that buparlisib in combination with trastuzumab successfully inhibits the PI3K/AKT/mTOR pathway in patients with advanced trastuzumab-resistant HER2$^+$ breast cancer. Similar to previous reports of dose-dependent inhibition of pS6 in the skin with single-agent buparlisib (45), the most prominent inhibition of pS6 was seen at the highest dose of buparlisib. Furthermore, of the skin biopsies analyzed, the greatest reduction in pS6 was associated with the best clinical response, and thus further investigation into the use of pS6 levels in skin as a surrogate biomarker for response is warranted. In contrast, changes in p4E-BP1 in skin biopsies were generally not observed. In the paired tumor biopsies, in addition to evidence of successful PI3K/AKT/mTOR pathway inhibition (reduced pAKT and p4E-BP1), reductions in pERK and pMEK were also observed, and most likely reflect inhibition of the RAS/MEK/ERK pathway through successful inhibition of HER2 by trastuzumab.

Preliminary signs of clinical activity were observed in this study (two PRs; 75% DCR; and 25% CBR at the RP2D), and although the sample size in this present trial is small, the activity was similar to that reported previously with the mTOR inhibitor everolimus in combination with trastuzumab (35% CBR; ref. 46). Preclinical data have demonstrated that continued HER2 blockade is required for potent tumor regression in response to PI3K inhibition even after the development of trastuzumab resistance (19, 20). Furthermore, in the present study, inhibition of the PI3K/AKT/mTOR and RAS/MEK/ERK pathways has been observed in paired tumor biopsies. Taken together, these data suggest that the clinical activity observed in this trial is likely the result of the combined activities of both buparlisib and trastuzumab and that PI3K inhibition with buparlisib can restore sensitivity to trastuzumab. However, single-agent buparlisib has also been shown to have activity in patients with advanced solid tumors (23, 47) and specifically in patients with metastatic breast cancer (24), and so the possibility of the observed activity in this trial being the result of single-agent buparlisib cannot be ruled out.

Tumor activation of the PI3K pathway through *PIK3CA* and/or *PTEN* alteration was present in both of the 17 (12%) patients who responded to treatment and in almost half (3 of 7) of those who achieved SD. The ongoing phase II portion of this trial, as well as other studies of buparlisib in breast cancer and other tumor types, will serve to delineate any predictive value of PI3K pathway activation for buparlisib clinical activity. In addition to alterations in *PIK3CA* and *PTEN*, it is likely that there are other genes whose expression may be associated with response to combined treatment with buparlisib and trastuzumab, such as overexpression of other receptor tyrosine kinases, or expression of oncogenic RAS. Comprehensive gene expression profiling of tumor samples to identify a gene expression signature associated with response to combined PI3K and HER2 inhibition may therefore be informative in defining predictive biomarkers of sensitivity and resistance. Similar studies have recently identified a gene expression signature associated with *in vitro* response to the PI3K inhibitor GDC-0941 (48).

In conclusion, the combination of buparlisib plus trastuzumab had a tolerable safety profile, and the study determined the RP2D to be buparlisib 100 mg/day and trastuzumab 2 mg/kg weekly. Buparlisib and trastuzumab produced a pharmacodynamic response in both the PI3K/AKT/mTOR and RAS/MEK/ERK signaling relay and preliminary evidence of clinical activity has been observed. The phase II portion of this trial is further evaluating the safety and efficacy of combined buparlisib and trastuzumab in this patient population.

Disclosure of Potential Conflicts of Interest

G. Jerusalem has received speakers bureau honoraria from and is a consultant/advisory board member for Novartis. D. Mills is an employee of Novartis Pharma AG. E. Di Tomaso is an employee of Novartis Pharmaceutical. D.W. Sternberg is an employee of Novartis Oncology. S. Hirawat is an employee of Novartis Pharmaceuticals Corporation. J. Baselga is a consultant/advisory board member for Roche. No potential conflicts of interest were disclosed by the other authors.

Authors' Contributions

Conception and design: C. Saura, S. Su, Q. Ru, S. De Buck, J.T. Beck, E. DiTomaso, C. Massacesi, S. Hirawat, J. Baselga

Development of methodology: Q. Ru, D. Mills, C. Massacesi, S. Hirawat, L. Dirix, J. Baselga

Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.): C. Saura, J. Bendell, G. Jerusalem, Q. Ru, A. Bosch, A. Urruticoechea, J.T. Beck, E. Di Tomaso, C. Massacesi, S. Hirawat, L. Dirix, J. Baselga

Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis): C. Saura, G. Jerusalem, Q. Ru, S. De Buck, D. Mills, S. Ruquet, E. Di Tomaso, C. Massacesi, S. Hirawat, L. Dirix, J. Baselga

Writing, review, and/or revision of the manuscript: C. Saura, J. Bendell, G. Jerusalem, S. Su, Q. Ru, S. De Buck, D. Mills, S. Ruquet, A. Bosch, A. Urruticoechea, J.T. Beck, E. Di Tomaso, D.W. Sternberg, C. Massacesi, S. Hirawat, L. Dirix, J. Baselga

Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): S. Su, Q. Ru, D. Mills, J.T. Beck, C. Massacesi, L. Dirix

Study supervision: C. Saura, S. Su, Q. Ru, J.T. Beck, D.W. Sternberg, C. Massacesi, S. Hirawat, L. Dirix

Acknowledgments

The authors thank Xumei Zhao, of Novartis Pharmaceutical Corporation, and Amanda Quinn, PhD, for medical editorial assistance. The authors would also like to thank the patients and their families.

Grant Support

G. Jerusalem receives research funding from Novartis Pharmaceuticals Corporation (grant number BEL00034/2011). Financial support for medical editorial assistance was provided by Novartis Pharmaceuticals.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked *advertisement* in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

Received April 18, 2013; revised November 29, 2013; accepted December 7, 2013; published OnlineFirst January 27, 2014.

References

1. Slamon DJ, Clark GM, Wong SG, Levin WJ, Ullrich A, McGuire WL. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. Science 1987;235:177–82.
2. Slamon DJ, Godolphin W, Jones LA, Holt JA, Wong SG, Keith DE, et al. Studies of the HER-2/neu proto-oncogene in human breast and ovarian cancer. Science 1989;244:707–12.
3. Shah S, Chen B. Testing for HER2 in breast cancer: a continuing evolution. Patholog Res Int 2010;2011:903202.
4. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. N Engl J Med 2001;344:783–92.
5. Vogel CL, Cobleigh MA, Tripathy D, Gutheil JC, Harris LN, Fehrenbacher L, et al. Efficacy and safety of trastuzumab as a single agent in first-line treatment of HER2-overexpressing metastatic breast cancer. J Clin Oncol 2002;20:719–26.
6. Geyer CE, Forster J, Lindquist D, Chan S, Romieu CG, Pienkowski T, et al. Lapatinib plus capecitabine for HER2-positive advanced breast cancer. N Engl J Med 2006;355:2733–43.
7. Baselga J, Gelmon KA, Verma S, Wardley A, Conte P, Miles D, et al. Phase II trial of pertuzumab and trastuzumab in patients with human epidermal growth factor receptor 2-positive metastatic breast cancer that progressed during prior trastuzumab therapy. J Clin Oncol 2010;28:1138–44.
8. Verma S, Miles D, Gianni L, Krop IE, Welslau M, Baselga J, et al. Trastuzumab emtansine for HER2-positive advanced breast cancer. N Engl J Med 2012;367:1783–91.
9. Lin NU, Winer EP. Brain metastases: the HER2 paradigm. Clin Cancer Res 2007;13:1648–55.
10. Liu P, Cheng H, Roberts TM, Zhao JJ. Targeting the phosphoinositide 3-kinase pathway in cancer. Nat Rev Drug Discov 2009;8:627–44.
11. Yakes FM, Chinratanalab W, Ritter CA, King W, Seelig S, Arteaga CL. Herceptin-induced inhibition of phosphatidylinositol-3 kinase and AKT is required for antibody-mediated effects on p27, cyclin D1, and antitumor action. Cancer Res 2002;62:4132–41.
12. Lopez-Knowles E, O'Toole SA, McNeil CM, Millar EK, Qiu MR, Crea P, et al. PI3K pathway activation in breast cancer is associated with the basal-like phenotype and cancer-specific mortality. Int J Cancer 2010;126:1121–31.
13. Hernandez-Aya LF, Gonzalez-Angulo AM. Targeting the phosphatidylinositol 3-kinase signaling pathway in breast cancer. Oncologist 2011;16:404–14.
14. Wee S, Wiederschain D, Maira SM, Loo A, Miller C, DeBeaumont R, et al. PTEN-deficient cancers depend on PIK3CB. Proc Natl Acad Sci U S A 2008;105:13057–62.
15. Depowski PL, Rosenthal SI, Ross JS. Loss of expression of the PTEN gene protein product is associated with poor outcome in breast cancer. Mod Pathol 2001;14:672–76.
16. Nagata Y, Lan KH, Zhou X, Tan M, Esteva FJ, Sahin AA, et al. PTEN activation contributes to tumor inhibition by trastuzumab, and loss of PTEN predicts trastuzumab resistance in patients. Cancer Cell 2004;6:117–27.
17. Berns K, Horlings HM, Hennessy BT, Madiredjo M, Hijmans EM, Beelen K, et al. A functional genetic approach identifies the PI3K pathway as a major determinant of trastuzumab resistance in breast cancer. Cancer Cell 2007;12:395–402.
18. O'Brien NA, Browne BC, Chow L, Wang Y, Ginther C, Arboleda J, et al. Activated phosphoinositide 3-kinase/AKT signaling confers resistance to trastuzumab but not lapatinib. Mol Cancer Ther 2010;9:1489–502.
19. Chakrabarty A, Bhola NE, Sutton CR, Ghosh R, Kuba MG, Dave B, et al. Trastuzumab-resistant cells rely on a HER2-PI3K-FoxO-survivin axis and are sensitive to PI3K inhibitors. Cancer Res 2012;73:1190–200.
20. O'Brien NA, McDonald K, Tong L, Von Euw E, Conklin D, Kalous O, et al. PI3K/mTOR inhibition overcomes *in vitro* and *in vivo* trastuzumab resistance independent of feedback activation of pAKT. Cancer Res 2012;72(24 Suppl.):Abstr P4-08-01.
21. Serra V, Markman B, Scaltriti M, Eichhorn PJ, Valero V, Guzman M, et al. NVP-BEZ235, a dual PI3K/mTOR inhibitor, prevents PI3K signaling and inhibits the growth of cancer cells with activating PI3K mutations. Cancer Res 2008;68:8022–30.
22. Maira SM, Pecchi S, Huang A, Burger M, Knapp M, Sterker D, et al. Identification and characterization of NVP-BKM120, an orally available pan-class I PI3-kinase inhibitor. Mol Cancer Ther 2012;11:317–28.
23. Bendell JC, Rodon J, Burris HA, de Jonge M, Verweij J, Birle D, et al. Phase I, dose-escalation study of BKM120, an oral pan-class I PI3K inhibitor, in patients with advanced solid tumors. J Clin Oncol 2012;30:282–90.
24. Rodon J, Bendell JC, Razak A, Homji N, Trandafir N, Quadt C, et al. Safety profile and clinical activity of single-agent BKM120, a pan-class I PI3K inhibitor, for the treatment of patients with metastatic breast carcinoma. Cancer Res 2011;71(24 Suppl.):Abstr P3-16-01.
25. Neuenschwander B, Branson M, Gsponer T. Critical aspects of the Bayesian approach to phase I cancer trials. Stat Med 2008;27:2420–39.

26. Kroenke K, Spitzer RL, Williams JB. The PHQ-9: validity of a brief depression severity measure. J Gen Intern Med 2001;16:606–13.
27. Spitzer RL, Kroenke K, Williams JB, Lowe B. A brief measure for assessing generalized anxiety disorder: the GAD-7. Arch Intern Med 2006;166:1092–7.
28. Von Hoff DD, LoRusso P, Demetri GD, Weiss GJ, Shapiro G, Ramanathan RK, et al. A phase I dose-escalation study to evaluate GDC-0941, a pan-PI3K inhibitor, administered QD or BID in patients with advanced or metastatic solid tumors. J Clin Oncol 2011;29(15 Suppl.): Abstr 3052.
29. Peyton JD, Rodon Ahnert J, Burris H, Britten C, Chen LC, Tabernero J, et al. A dose-escalation study with the novel formulation of the oral pan-class I PI3K inhibitor BEZ235, solid dispersion system (SDS) sachet, in patients with advanced solid tumors. J Clin Oncol 2011:29(15 Suppl.):Abstr 3066.
30. Wagner AJ, Bendell JC, Dolly S, Morgan JA, Ware JA, Fredrickson J, et al. A first-in-human phase I study to evaluate GDC-0980, an oral PI3K/mTOR inhibitor, administered QD in patients with advanced solid tumors. J Clin Oncol 2011;29(15 Suppl.):Abstr 3020.
31. Edelman G, Bedell C, Shapiro G, Pandya SS, Kwak EL, Scheffold C, et al. A phase I dose-escalation study of XL147 (SAR245408), a PI3K inhibitor administered orally to patients (pts) with advanced malignancies. J Clin Oncol 2010;28(15 Suppl.):Abstr 3004.
32. Markman B, Dienstmann R, Tabernero J. Targeting the PI3K/AKT/mTOR pathway—beyond rapalogs. Oncotarget 2010;1:530–43.
33. Tabernero J, Rojo F, Calvo E, Burris H, Judson I, Hazell K, et al. Dose- and schedule-dependent inhibition of the mammalian target of rapamycin pathway with everolimus: a phase I tumor pharmacodynamic study in patients with advanced solid tumors. J Clin Oncol 2008;26:1603–10.
34. Yap TA, Yan L, Patnaik A, Fearen I, Olmos D, Papadopoulos K, et al. First-in-man clinical trial of the oral pan-AKT inhibitor MK-2206 in patients with advanced solid tumors. J Clin Oncol 2011;29: 4688–95.
35. Busaidy NL, Farooki A, Dowlati A, Perentesis JP, Dancey JE, Doyle LA, et al. Management of metabolic effects associated with anticancer agents targeting the PI3K-AKT-mTOR pathway. J Clin Oncol 2012; 30:2919–28.
36. Foukas LC, Claret M, Pearce W, Okkenhaug K, Meek S, Peskett E, et al. Critical role for the p110alpha phosphoinositide-3-OH kinase in growth and metabolic regulation. Nature 2006;441:366–70.
37. Taniguchi CM, Emanuelli B, Kahn CR. Critical nodes in signalling pathways: insights into insulin action. Nat Rev Mol Cell Biol 2006;7:85–96.
38. Knight ZA, Gonzalez B, Feldman ME, Zunder ER, Goldenberg DD, Williams O, et al. A pharmacological map of the PI3-K family defines a role for p110alpha in insulin signaling. Cell 2006;125:733–47.
39. Engelman JA, Luo J, Cantley LC. The evolution of phosphatidylinositol 3-kinases as regulators of growth and metabolism. Nat Rev Genet 2006;7:606–19.
40. Nanni P, Nicoletti G, Palladini A, Croci S, Murgo A, Ianzano ML, et al. Multiorgan metastasis of human HER-2(+) breast cancer in Rag2 (−/−);Il2rg(−/−) mice and treatment with PI3K inhibitor. PLoS ONE 2012;7:e39626.
41. Maira M, Schnell C, Lollini P, Chouaid C, Schmid P, Nanni P, et al. Preclinical and preliminary clinical activity of NVP-BKM120, an oral pan-class I PI3K inhibitor, in the brain. Ann Oncol 2012;23(Suppl. 9): Abstr 1675P.
42. Bandaru SS, Lin K, Roming SL, Vellipuram R, Harney JP. Effects of PI3K inhibition and low docosahexaenoic acid on cognition and behavior. Physiol Behav 2010;100:239–44.
43. Tohda C, Nakanishi R, Kadowaki M. Hyperactivity, memory deficit and anxiety-related behaviors in mice lacking the p85alpha subunit of phosphoinositide-3 kinase. Brain Dev 2009;31:69–74.
44. Ackermann TF, Hortnagl H, Wolfer DP, Colacicco G, Sohr R, Lang F, et al. Phosphatidylinositide dependent kinase deficiency increases anxiety and decreases GABA and serotonin abundance in the amygdala. Cell Physiol Biochem 2008;22:735–44.
45. Rodon J, Bendell J, Razak ARA, De Jonge MJA, Eskens F, Di Tomaso E, et al. A phase I dose-escalation and expansion trial of BKM120, an oral pan-PI3K inhibitor, in patients with advanced solid tumors: analysis of pharmacodynamic biomarker data. Ann Oncol 2012;23(Suppl. 9):Abstr 457P.
46. Morrow PK, Wulf GM, Ensor J, Booser DJ, Moore JA, Flores PR, et al. Phase I/II study of trastuzumab in combination with everolimus (RAD001) in patients with HER2-overexpressing metastatic breast cancer who progressed on trastuzumab-based therapy. J Clin Oncol 2011;29:3126–32.
47. Doi T, Ando Y, Bando H, Yoshino T, Inada M, Mitsuma A, et al. Phase I dose-escalation study of BKM120, an oral pan-class I PI3K inhibitor, in Japanese patients with advanced solid tumors. Mol Cancer Ther 2011;10(11 Suppl.):Abstr B159.
48. O'Brien C, Wallin JJ, Sampath D, GuhaThakurta D, Savage H, Punnoose EA, et al. Predictive biomarkers of sensitivity to the phosphatidylinositol 3′ kinase inhibitor GDC-0941 in breast cancer preclinical models. Clin Cancer Res 2010;16:3670–83.