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MONOGRAPH

VISMODEGIB

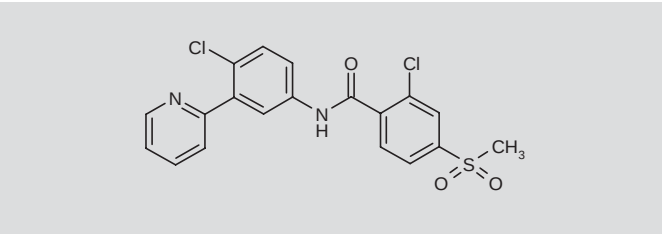
USAN

SMO Receptor Antagonist
Hedgehog Signaling Inhibitor
Oncolytic

CUR-691
GDC-0449
Hh-Antag691
HhAntag
R-3616
RG-3616

2-Chloro-*N*-[4-chloro-3-(pyridin-2-yl)phenyl]-4-(methylsulfonyl)benzamide

InChI: 1S/C19H14Cl2N2O3S/c1-27(25,26)13-6-7-14(17(21)11-13)19(24)23-12-5-8-16(20)15(10-12)18-4-2-3-9-22-18/h2-11H,1H3,(H,23,24)



$C_{19}H_{14}Cl_2N_2O_3S$
Mol wt: 421.297
CAS: 879085-55-9
EN: 473491

SUMMARY

When it became apparent that aberrant hedgehog (HH) pathway signaling was a likely mediator in the development of tumors, especially basal cell carcinoma, many companies began to investigate the use of small-molecule inhibitors of key steps in the HH pathway as potential anticancer agents. Under normal physiological conditions the pathway controls embryonic development via HH ligand binding to the patched (PTC) receptor, which inhibits a second receptor, smoothened (SMO), which in turn regulates glioma-associated oncogene (GLI)-mediated transcription of pro- and antiapoptotic genes. Genentech and Curis developed vismodegib (GDC-0449), a small-molecule inhibitor of SMO, which was proven to be the first such compound to successfully target tumorigenesis in humans. In preclinical pharmacokinetic and pharmacodynamic studies, vismodegib demonstrated promising characteristics for use in humans, more so than its predecessor Hh-Antag691. Vismodegib successfully entered phase I clinical trials in patients with basal cell carcinoma and medulloblastoma, and is currently under investigation in a number of phase II trials for various cancers, including basal cell carcinoma, medulloblastoma, pancreatic, ovarian, stomach and breast cancers, in addition to a number of phase II trials investigating its use in combination with other chemotherapeutic agents such as bevacizumab.

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*Synthesis prepared by R. Pandian, J. Bolós, R. Castañer. Thomson Reuters, Provença 388, 08025 Barcelona, Spain.

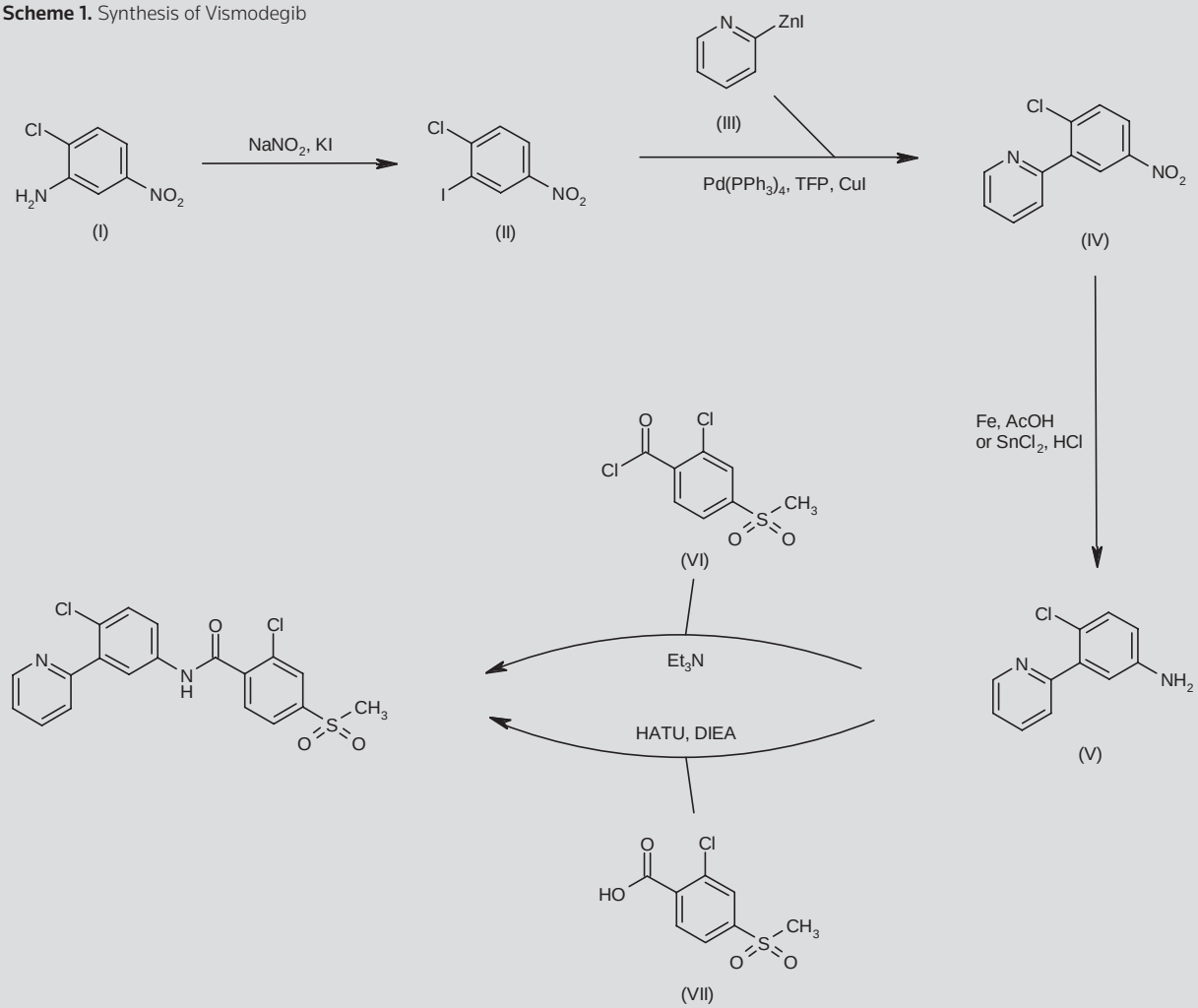
SYNTHESIS*

Sandmeyer reaction of 2-chloro-5-nitroaniline (I) with $NaNO_2$ in the presence of KI gives 1-chloro-2-iodo-4-nitrobenzene (II), which is then coupled with (2-pyridyl)zinc iodide (III) by means of $Pd(PPh_3)_4$, tris(2-furyl)phosphine and CuI in THF/DMAc to provide the phenylpyridine derivative (IV). After nitro group reduction in compound (IV) using either Fe and AcOH at 80 °C or $SnCl_2$ and HCl in EtOH (1), the resulting pyridylaniline (V) is finally acylated with either 2-chloro-4-(methylsulfonyl)benzoyl chloride (VI) in the presence of Et_3N in CH_2Cl_2 or 2-chloro-4-(methylsulfonyl)benzoic acid (VII) by means of HATU and DIEA in DMF (1, 2). Scheme 1.

BACKGROUND

The hedgehog (HH) pathway is a key signaling pathway in embryological development that is normally inactive in differentiated cells. The HH signaling pathway comprises three ligands (Sonic, SHH; Indian, IHH; and Desert, DHH) and two interacting transmembrane receptors, patched (PTC) and smoothened (SMO). In the absence of HH ligand binding, PTC and SMO form a repressive complex that inhibits cellular proliferation. The activation of SMO by the dissocia-

Scheme 1. Synthesis of Vismodegib



tion of the PTC receptor following HH ligand binding initiates G protein-mediated signaling cascades that result in upregulation of glioma-associated oncogenes GLI1-3. These proteins act as transcription factors that can alter gene expression in apoptotic and cell cycle-regulated pathways and in the HH signaling pathway itself, e.g., PTC1 (reviewed in 3). A characteristic of this aberrant activation is uncontrolled proliferation and subsequent tumor formation.

Constitutive reactivation of the HH pathway via mutations and aberrant autocrine/paracrine HH ligand stimulation has been linked to the promotion of a number of malignancies, including basal cell carcinoma (4), prostate (5), stomach (6), pancreatic (7) and colon cancers (8), medulloblastoma (9), glioma (10), lymphoma, leukemia and melanoma (11; for an extensive review, see 12, 13). Such mutations include those in PTC, in which deletion of the second allele results in constitutive upregulation of the PTC, SMO and GLI transcripts and missense activating mutations. In SMO, both types of mutation have been found to be largely associated with medulloblastoma, neuroectodermal tumors and basal cell carcinoma (14-16).

Many naturally occurring and synthetic small-molecule inhibitors of the HH signaling pathway that antagonize HH ligands or target SMO or events downstream of SMO activity, such as GLI activation, are being investigated as potential therapeutic anticancer drugs, and have been shown to halt the progression of various tumors (for an extensive review, see 13). Most, if not all, demonstrate antitumor activity, at least in vitro, with varying levels of potency, stability and toxicity reported. Hh-Antag691 is an SMO receptor antagonist that has demonstrated promising antiproliferative and proapoptotic activity both in cancer cell lines and in vivo in mammalian models, although its effectiveness as a human therapy is limited (17).

Curis and Genentech have developed an optimized version of Hh-Antag691 with improved potency, solubility, metabolic stability and absorption spectra, named vismodegib (GDC-0449) (1). Vismodegib is a small-molecule systemic HH inhibitor that acts as an SMO

receptor antagonist and is currently in phase II clinical trials for the treatment of ovarian cancer, basal cell carcinoma, refractory medulloblastoma, Gorlin syndrome, small cell lung cancer and advanced gastric, pancreatic and breast cancers. It is also being investigated in combination with chemotherapeutic agents such as bevacizumab (Avastin®; Genentech) as first-line therapy for the treatment of metastatic colorectal cancer.

PRECLINICAL PHARMACOLOGY

Preclinical assessment indicated that vismodegib could act as a potent inhibitor of HH-GLI signaling via SMO receptor antagonism both in vitro and in vivo. Nude mice implanted with human lung carcinoma Calu-6 cells, a clonal cell line that exhibits tumorigenesis dependent on HH signaling, were dosed orally with vismodegib (75 mg/kg b.i.d.) for 3 days, resulting in a 10-fold knock down in GLI expression, a measure of HH pathway (de)activation, in the stroma. In addition, vismodegib (12.5 mg/kg b.i.d. p.o.) elicited complete tumor regression in an HH pathway-dependent medulloblastoma allograft model generated from *Ptc*$^{+/-}$ mice (1). Furthermore, in a similar murine model in which *Ptc*$^{+/-}$ mice were bred with *p53*$^{-/-}$ mice to increase medulloblastoma formation, vismodegib (20 or 100 mg/kg b.i.d. for 4 days) decreased GLI mRNA in a dose-dependent manner, as evidenced by both quantitative polymerase chain reaction (qPCR) from whole cerebellum extract and in situ hybridization in brain sections. Decreased proliferation, increased macrophage infiltration, increased gliosis and increased cell death, indicative of growth inhibition, were also observed in these mice. The same study concluded that vismodegib exposure in *Ptc*$^{+/-}$ *p53*$^{-/-}$ mice (20 or 100 mg/kg for 14 days) dose-dependently decreased tumor size, with ablation of tumor seen at the higher dose (18). Hh-Antag691 (100 mg/kg b.i.d.), the vismodegib predecessor, also transiently reduced tumor size in a distinct murine model that exhibits HH signaling, the Cxcr6 mouse, in which one or both copies of the *Cxcr6* gene are knocked out, producing HH-dependent medulloblastoma in the animal, although Cxcr6 is itself not associated with the pathway (19).

In addition to targeting SMO, vismodegib has also been shown to target ATP-binding cassette (ABC) proteins that are naturally present in the blood–brain barrier and, as a result of HH signaling, are overexpressed in many cancers, and which are known to confer multidrug resistance. Human HEK-293 cells were manipulated to overexpress the ATP-binding cassette sub-family G member 2 (CD338) and were incubated with a florescent transporter substrate. Treatment of these cells with vismodegib resulted in inhibition of the transporter (IC_{50} = 1.4 μM), as demonstrated by increased retention of the fluorescent dye. Moreover, vismodegib at 10 μM reversed drug resistance to the antineoplastic agent mitoxantrone (5 μM) conferred by overexpression of the transporter. Most pertinent is that vismodegib was able to reverse drug resistance to toxic ABC transporter substrates in the human non-small cell lung cancer cell line NCI-H460 (17). It is worth noting that a subsequent study confirmed that Zhang et al. used the preoptimized version of vismodegib, Hh-Antag691, and as such, the results reported may not directly correlate with vismodegib (20).

PHARMACOKINETICS AND METABOLISM

The pharmacokinetic (PK) and pharmacodynamic (PD) profile of vismodegib has been extensively assessed in rodents, dogs, nonhuman primates and humans. Equilibrium dialysis of vismodegib at 1, 10 and 100 μM was performed in mice, rats, rabbits, dogs, cynomolgus monkeys and humans to assess plasma protein binding, which was found to be substantial (> 94%) and concentration-independent across all species tested. Blood-to-plasma partition values at 1, 10 and 100 μM vismodegib were obtained using the ratio of [^{14}C]-radiolabeled vismodegib to unlabeled drug in mice, rats, dogs, monkeys and humans (0.608-0.881 across all species irrespective of dose) (20).

Vismodegib (1 mg/kg i.v.) plasma clearance was low in mice (N = 27), rats (N = 3) and dogs (N = 3) at 23.0, 4.65 and 0.338 mL/min/kg, respectively, and moderate in monkeys (N = 3) at 19.3 mL/min/kg. The clearance in humans was predicted to be low at 0.649 mL/min/kg or 0.096 mL/min/kg, respectively, depending on whether monkey data were included in the prediction or not. The terminal half-life ($t_{1/2}$) of the drug ranged from 0.976 h (mice) to 41.8 h (dogs) and was predicted to be 13.6 h (using the higher predicted clearance) or 92.1 h in humans. The volume of distribution at steady state (V_{ss}) was low to moderate in all species (1.68, 0.490 ± 0.0653, 1.03 ± 0.119, 0.984 ± 0.342 and 0.766 L/kg, respectively, in mice, rats, dogs, monkeys and humans [predicted]) (20).

Bioavailability upon oral dosing (2 mg/kg for dogs and monkeys, and 5 mg/kg for rats and mice) was found to range between 13% and 53%, and renal clearance was negligible in all species.

The metabolic stability of vismodegib was assessed in vitro in hepatocytes. Following a 3-h incubation period, the amount of initial drug was 88% or greater in mice, rats and dogs, displaying high metabolic stability, and moderate in monkeys at approximately 44%. In addition, a total of six separate metabolites of vismodegib (three oxidative [M1-3] and three glucuronide [M4-6]) were identified in rats, dogs and humans, which is indicative of high drug stability. Recombinant human isoforms of cytochrome P450 where incubated with 50 μM vismodegib to determine those responsible for metabolite oxidation, namely P4503A4 and 3A5 for M1 and 2C9 for M3, for subsequent IC_{50} determination. The IC_{50} for vismodegib was assessed by P450 inhibition in human liver microsomes using standard probes, and inhibition constant values (K_i) indicated that it may be a moderate inhibitor of P4502C8 (6.0 μM) and P4502C9 (5.4 μM). Vismodegib at 15 μM was found not to inhibit P-glycoproteins (20).

The PK prolife of vismodegib has also been assessed in humans with refractory local or advanced metastatic solid tumors. Following a single dose, the peak plasma concentration (C_{max}) showed proportional progression at doses between 150 and 270 mg, but not at higher doses up to 540 mg. In healthy volunteers the $t_{1/2}$ was 10-14 days and in cancer patients plasma drug concentrations were maintained for up to 1 week. Following a multiple-dose regimen, steady state was reached at between 1 and 3 weeks. In agreement with previous animal studies, vismodegib exhibited high plasma protein binding. In particular, the compound showed strong, saturable and reversible binding to the alpha-1-acid glycoprotein, which the authors of the study suggest may explain the non-linear PK profile of vismodegib (21).

SAFETY

In all studies to date no serious adverse events (AEs) or dose-limiting toxicities (DLTs) have been reported. Grade 1 AEs such as stom-

ach cramps and nausea have been reported, and grade 3 AEs including fatigue, hyponatremia, muscle spasm and atrial fibrillation were noted in clinical studies.

CLINICAL STUDIES

Vismodegib has advanced to the clinical trial stage, with both phase I and phase II studies under way or proposed. An interventional, open-label, multicenter phase I investigation to evaluate the safety, tolerability, PK/PD and best dose of vismodegib for treating patients with locally advanced or metastatic solid tumors is being conducted by Genentech, the National Cancer Institute (NCI) and the Sidney Kimmel Comprehensive Cancer Center (22). The study began in April 2007 and data for primary outcome measures to assess DLT, AEs and single- and multiple-dose PK were collated in April 2009. Secondary outcome measures will include qPCR analysis of GLI mRNA levels from hair follicle and skin samples, tumor response and progression-free survival (PFS). In total, 33 adult patients (median age of 38-84 years) with metastatic or locally advanced basal cell carcinoma and an Eastern Cooperative Oncology Group (ECOG) performance rating < 2, were enrolled and received daily oral vismodegib at 150 mg (n = 17), 270 mg (n = 15) or 540 mg (n = 1). The initial dose was given on day 1 and then continued daily beginning on day 8 for a median period of 9.8 months. Tumor response was assessed by Response Evaluation Criteria in Solid Tumors (RECIST) or physical examination. No DLTs were recorded and overall 2 patients demonstrated a complete response, 16 showed a partial response (> 50% reduction in tumor diameter), 11 displayed stable disease and 2 disease progression. The overall response rate from patients with locally advanced or metastatic tumors was 60% and 50%, respectively. AEs related to the treatment included grade 3 fatigue (4 patients), hyponatremia (2 patients), muscle spasm (1 patient) and atrial fibrillation (1 patient). A single grade 4 AE was deemed not to be related to the treatment and a single patient withdrew from the study due to AEs. A second stage of this study enrolled 12 patients (non-basal cell carcinoma) to assess PK/PD of the recommended daily dose. This stage itself was further extended to include 20 patients with basal cell carcinoma who received the recommended daily dose or a higher dose of 250 mg/day, and 16 patients with solid tumors (10 with basal cell carcinoma); the latter group was included to assess the PK/PD profile of a new formulation of vismodegib (150 mg/day). Doses higher than 150 mg/day did not result in increased plasma concentrations (median C_{max} = 23 µM; steady-state concentration [C_{ss}] = 17 µM; median time to C_{ss} = 14 days). The majority of patients had elevated concentrations of GLI mRNA in tumor samples taken prior to vismodegib treatment, supporting a role for HH signaling in locally advanced and metastatic tumors. In 10 of 13 patients, a twofold downregulation of GLI mRNA expression was detected by qPCR on nontumor skin samples following vismodegib treatment compared to surrogate nontumor skin samples. The primary outcome data suggested that vismodegib displays a good PK/PD profile, and is effective in decreasing tumor size or halting disease progression at a dose of 150 mg/day; this was suggested to be the maximum effective dose for phase II investigations (21, 23-27).

A unique case study emerged from the above study; a single 26-year-old male with refractory medulloblastoma as a result of an *SMO* point mutation received 540 mg vismodegib on day 1 and day 8, and then daily for the remainder of the study. Within 2 months of the beginning of treatment, the patient showed marked improvement; he had no pain, no palpable nodules, had gained 7 kg and had returned to normal levels of physical activity. By the 3-month stage, however, scans revealed the emergence of new tumor masses and regrowth at some former tumor sites; although the response to vismodegib was rapid and remarkable and indicated that the compound could be useful for treating medulloblastoma, it was transient because the patient appeared to develop multidrug resistance (MDR) (28). Later molecular analysis showed that the patient's point mutation in *SMO* prevented the binding of vismodegib to SMO, which could in part explain the transient effect of the drug (29).

Currently, three phase II clinical trials of vismodegib are in the active stage. Genentech is sponsoring an interventional, double-blind, randomized, placebo-controlled study of vismodegib in combination with chemotherapy or bevacizumab as first-line therapy for metastatic colorectal cancer. The study began in March 2008 and the primary outcome measure of PFS will be assessed in March 2011. Secondary outcomes will include the analysis of HH ligand concentration in archived tissues and an assessment of grade 3/4 AEs. A total of 198 patients aged 18 and above with an ECOG performance status of 0 or 1 and adequate hepatic and renal function have been enrolled (30).

A second multicenter, randomized, double-blind, placebo-controlled phase II study is investigating the potential use of vismodegib as maintenance therapy in patients with ovarian cancer in the second or third remission. This study, sponsored by Genentech, began in September 2008 and the primary outcome measure of PFS will be assessed in January 2012. Secondary outcomes will include overall survival rates, the analysis of HH ligand concentration in archived tissues and an assessment of grade 3/4 AEs. A total of 104 female patients age 18 and above with epithelial ovarian, primary peritoneal or fallopian tube carcinoma and who are in second- or third-stage remission have been enrolled (31).

The final active multicenter, single-arm, two-cohort phase II study will evaluate the efficacy and safety of vismodegib in patients with advanced basal cell carcinoma. This study sponsored by Genentech began in January 2009 and the primary outcome measure of overall response rate will be determined in July 2012. Secondary outcomes will include response duration, PFS, overall survival and assessment of AEs. There are 100 patients enrolled aged over 18 years with inoperable advanced basal cell carcinoma (32).

A multicenter, nonrandomized, uncontrolled, open-label extension study of vismodegib in patients previously treated with the agent in phase I or II Genentech-sponsored studies in patients with basal cell carcinoma, ovarian carcinoma and metastatic colorectal carcinoma is currently enrolling. Enrollment in this study began in August 2009 and it is due for primary completion to measure AEs and AEs leading to discontinuation of treatment in August 2017 (33).

A number of studies sponsored by Genentech in collaboration with others are currently recruiting patients to investigate the safety and efficacy of vismodegib treatment in various cancers, including Gorlin syndrome (34) and refractory medulloblastoma in young patients (35) and in adults (36). A study is also proposed to investigate vismodegib in healthy female subjects of non-childbearing potential (37).

There are also a number of studies proposed but not yet recruiting to examine vismodegib safety and efficacy in metastatic adenocarcinoma of the pancreas (38), operable recurrent glioblastoma multiforme (39) and pancreatic ductal adenocarcinoma in the preoperative setting (40).

Recruitment is under way to investigate the efficacy of vismodegib in conjunction with other drugs and chemotherapy, i.e., in advanced stomach or gastroesophageal junction cancer (41), metastatic pancreatic cancer or inoperable solid tumors in conjunction with erlotinib or gemcitabine hydrochloride (42, 43), and in advanced breast cancer with RO-4929097 (44).

SOURCES

Curis, Inc. (US); Genentech Inc. (wholly owned subsidiary of Roche) (US); licensed to Chugai Pharmaceutical Co., Ltd. (JP).

DISCLOSURES

The author states no conflicts of interest.

REFERENCES

1. Robarge, K.D., Brunton, S.A., Castanedo, G.M. et al. *GDC-0449 - A potent inhibitor of the hedgehog pathway*. Bioorg Med Chem Lett 2009, 19(19): 5576-81.
2. Gunzner, J., Sutherlin, D., Stanley, M. et al. (Genentech, Inc.; Curis, Inc.). *Pyridyl inhibitors of hedgehog signalling*. EP 1789390, JP 2008511675, WO 2006028958.
3. Rohatgi, R., Scott, M.P. *Patching the gaps in hedgehog signaling*. Nat Cell Biol 2007, 9(9): 1005-9.
4. Williams, J.A., Guicherit, O.M., Zaharian, B.I. et al. *Identification of a small molecule inhibitor of the hedgehog signaling pathway: Effects on basal cell carcinoma-like lesions*. Proc Natl Acad Sci U S A 2003, 100(8): 4616-21.
5. Sanchez, P., Hernandez, A.M., Stecca, B. et al. *Inhibition of prostate cancer proliferation by interference with SONIC HEDGEHOG-GLI1 signaling.* Proc Natl Acad Sci U S A 2004, 101(34): 12561-6.
6. Yauch, R.L., Gould, S.E., Scales, S.J. et al (2008) *A paracrine requirement for hedgehog signaling in cancer.* Nature 2008, 455(7211): 406-10.
7. Tian, H., Callahan, C.A., DuPree, K.J., Darbonne, W.C., Ahn, C.P., Scales, S.J., de Sauvage, F.J. *Hedgehog signaling is restricted to the stromal compartment during pancreatic carcinogenesis*. Proc Natl Acad Sci U S A 2009, 106(11): 4254-9.
8. Varnat, F., Duquet, A., Malerba, M. et al. *Human colon cancer epithelial cells harbor active HEDGEHOG-GLI signaling that is essential for tumor growth, recurrence, metastasis and stem cell survival and expansion.* EMBO Mol Med 2009, 1(6-7): 338-51.
9. Sanchez, P., Ruiz i Altaba, A. *In vivo inhibition of endogenous brain tumors through systemic interference of hedgehog signaling in mice*. Mech Dev 2005, 122(2): 223-30.
10. Clement, V., Sanchez, P., de Tribolet, N., Radovanovic, I., Ruiz i Altaba, A. *HEDGEHOG-GLI1 signaling regulates human glioma growth, cancer stem cell self-renewal, and tumorigenicity*. Curr Biol 2007, 17(2): 165-72.
11. Stecca, B., Mas, C., Clement, V. et al. *Melanomas require HEDGEHOG-GLI signaling regulated by interactions between GLI1 and the RAS-MEK/AKT pathways*. Proc Natl Acad Sci U S A 2007, 104(14): 5895-900.
12. Teglund, S., Toftgard, R. *Hedgehog beyond medulloblastoma and basal cell carcinoma*. Biochim Biophys Acta 2010, 1805(2): 181-208.
13. Mas, C., Ruiz i Altaba, A. *Small molecule modulation of HH-GLI signaling: Current leads, trials and tribulations*. Biochem Pharmacol 2010, Epub ahead of print.
14. Reifenberger, J., Wolter, M., Knobbe, C.B. et al. *Somatic mutations in the PTCH, SMOH, SUFUH and TP53 genes in sporadic basal cell carcinomas*. Br J Dermatol 2005, 152(1): 43-51.
15. Wolter, M., Reifenberger, J., Sommer, C., Ruzicka, T., Reifenberger, G. *Mutations in the human homologue of the Drosophila segment polarity gene patched (PTCH) in sporadic basal cell carcinomas of the skin and primitive neuroectodermal tumors of the central nervous system.* Cancer Res 1997, 57(13): 2581-5.
16. Romer, J., Curran, T. *Targeting medulloblastoma: Small-molecule inhibitors of the Sonic hedgehog pathway as potential cancer therapeutics*. Cancer Res 2005, 65(12): 4975-8.
17. Zhang, Y., Laterra, J., Pomper, M.G. *Hedgehog pathway inhibitor HhAntag691 is a potent inhibitor of ABCG2/BCRP and ABCB1/Pgp*. Neoplasia 2009, 11(1): 96-101.
18. Romer, J.T., Kimura, H., Magdaleno, S. et al. *Suppression of the Shh pathway using a small molecule inhibitor eliminates medulloblastoma in Ptc1(+/-)p53(-/-) mice*. Cancer Cell 2004, 6(3): 229-40.
19. Sasai, K., Romer, J.T., Kimura, H., Eberhart, D.E., Rice, D.S., Curran, T. *Medulloblastomas derived from Cxcr6 mutant mice respond to treatment with a smoothened inhibitor*. Cancer Res 2007, 67(8): 3871-7.
20. Wong, H., Chen, J.Z., Chou, B. et al. *Preclinical assessment of the absorption, distribution, metabolism and excretion of GDC-0449 (2-chloro-N-(4-chloro-3-(pyridin-2-yl)phenyl)-4-(methylsulfonyl)benzamide), an orally bioavailable systemic hedgehog signaling pathway inhibitor.* Xenobiotica 2009, 39(11): 850-61.
21. Graham, R.A. *Pharmacokinetics of hedgehog pathway inhibitor GDC-0449 in patients with refractory or untreatable locally advanced or metastatic solid tumors: The role of alpha 1-acid glycoprotein protein binding*. 7th Annu Cancer Drugs Res and Dev (Jan 28-29, San Diego) 2010, Abst.
22. *GDC-0449 in treating patients with locally advanced or metastatic solid tumors. (NCT00607724).* ClinicalTrials.gov Web site, April 15, 2010.
23. Von Hoff, D.D., Lorusso, P.M., Rudin, C.M. et al. *Inhibition of the hedgehog pathway in advanced basal-cell carcinoma*. N Engl J Med 2009, 361(12): 1164-72.
24. Von Hoff, D.D., Rudin, M.C., LoRusso, M.P. et al. *Efficacy data of GDC-0449, a systemic Hedgehog pathway antagonist, in a first-in-human, first-in-class phase I study with locally advanced, multifocal or metastatic basal cell carcinoma patients.* Proc Am Assoc Cancer Res (AACR) 2008, 49: Abst LB-138.
25. LoRusso, P.M., Rudin, C.M., Borad, M.J. et al. *A first-in-human, first-in-class, phase (ph) I study of systemic hedgehog (Hh) pathway antagonist, GDC-0449, in patients (pts) with advanced solid tumors.* J Clin Oncol [44th Annu Meet Am Soc Clin Oncol (ASCO) (May 30-June 3, Chicago) 2008] 2008, 26(15, Suppl.): Abst 3516.
26. Rudin, C.M., Von Hoff, D.D., LoRusso, P.M. et al. *Updated safety and efficacy data from a first-in-human, first-in-class, phase I study of hedgehog pathway antagonist, GDC-0449.* Eur J Cancer Suppl [20th EORTC-NCI-AACR Symp Mol Targets Cancer Ther (Oct 21-24, Geneva) 2008] 2008, 6(12): Abst 355.
27. Doggrell, S.A. *The hedgehog pathway inhibitor GDC-0449 shows potential in skin and other cancers.* Expert Opin Investig Drugs 2010, 19(3): 451-4.
28. Rudin, C.M., Hann, C.L., Laterra, J. et al. *Treatment of medulloblastoma with hedgehog pathway inhibitor GDC-0449.* N Engl J Med 2009, 361(12): 1173-8.

29. Yauch, R.L., Dijkgraaf, G.J., Alicke, B. et al. *Smoothened mutation confers resistance to a Hedgehog pathway inhibitor in medulloblastoma*. Science 2009, 326(5952): 572-4.

30. *A study of GDC-0449 (hedgehog pathway inhibitor) with concurrent chemotherapy and bevacizumab as first-line therapy for metastatic colorectal cancer (NCT00636610).* ClinicalTrials.gov Web site, April 15, 2010.

31. *A study of GDC-0449 (hedgehog pathway inhibitor) as maintenance therapy in patients with ovarian cancer in a second or third complete remission (NCT00739661).* ClinicalTrials.gov Web site, April 15, 2010.

32. *A study evaluating the efficacy and safety of GDC-0449 (hedgehog pathway inhibitor) in patients with advanced basal cell carcinoma (NCT00833417).* ClinicalTrials.gov Web site, April 15, 2010.

33. *A study of GDC-0449 (hedgehog pathway inhibitor) in patients treated with GDC-0449 in a previous Genentech-sponsored phase I or II cancer study (NCT00959647).* ClinicalTrials.gov Web site, April 15, 2010.

34. *The purpose of this study is to determine the efficacy and safety of a systemic hedgehog pathway antagonist (GDC-0449) in patients with basal cell nevus syndrome (BCNS) (NCT00957229).* ClinicalTrials.gov Web site, April 15, 2010.

35. *GDC-0449 in treating young patients with medulloblastoma that is recurrent or did not respond to previous treatment (NCT00822458).* ClinicalTrials.gov Web site, April 15, 2010.

36. *GDC-0449 in treating adult patients with recurrent or refractory medulloblastoma (NCT00939484).* ClinicalTrials.gov Web site, April 15, 2010.

37. *A study of the hedgehog pathway inhibitor GDC-0449 in healthy female subjects of non-childbearing potential (NCT0091718).* ClinicalTrials.gov Web site, April 15, 1010.

38. *Hedgehog inhibitors for metastatic adenocarcinoma of the pancreas (NCT01088815).* ClinicalTrials.gov Web site, April 15, 2010.

39. *GDC-0449 in treating patients with recurrent glioblastoma multiforme that can be removed by surgery (NCT00980343).* ClinicalTrials.gov Web site, April 15, 2010.

40. *Hedgehog inhibition for pancreatic ductal adenocarcinoma (PDAC) in the preoperative setting (HIPPoS) (NCT01096732).* ClinicalTrials.gov Web site, April 15, 2010.

41. *Combination chemotherapy with or without GDC-0449 in treating patients with advanced stomach cancer or gastroesophageal junction cancer (NCT00982592).* ClinicalTrials.gov Web site, April 15, 2010.

42. *GDC-0449 and erlotinib hydrochloride with or without gemcitabine hydrochloride in treating patients with metastatic pancreatic cancer or solid tumors that cannot be removed by surgery (NCT0087816).* ClinicalTrials.gov Web site, April 15, 2010.

43. *Gemcitabine hydrochloride with or without GDC-0449 in treating patients with recurrent or metastatic pancreatic cancer (NCT01064622).* ClinicalTrials.gov Web site, April 15, 2010.

44. *GDC-0449 and RO4929097 in treating women with advanced breast cancer (NCT01071564).* ClinicalTrials.gov Web site, April 15, 2010.