

OTHERS

Phase I study of combined therapy with vorinostat and gefitinib to treat *BIM* deletion polymorphism-associated resistance in *EGFR*-mutant lung cancer (VICTROY-J) : a study protocol

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Abstract : The *BIM* deletion polymorphism is reported to be associated with poor outcomes of epidermal growth factor receptor (*EGFR*)-mutant non-small cell lung cancer (NSCLC) treated with *EGFR*-TKIs, including gefitinib. We have shown that a histone deacetylase inhibitor, vorinostat, can epigenetically restore *BIM* function and apoptosis sensitivity to *EGFR*-TKIs in *EGFR*-mutant NSCLC cells with *BIM* deletion polymorphisms. The purpose of this study is to determine the feasibility of combined treatment of vorinostat with gefitinib in *BIM* deletion polymorphism positive *EGFR*-mutant NSCLC patients. *BIM* deletion polymorphism positive *EGFR*-mutant NSCLC patients treated with at least one *EGFR*-TKI and one regimen of chemotherapy are being recruited to this study. Vorinostat (200-400 mg) will be administered orally once daily on days 1-7, and gefitinib 250 mg orally once daily on days 1-14. With a fixed dose of gefitinib, the dose of vorinostat will be escalated following a conventional 3+3 design. The primary endpoint is to define the maximum tolerated dose (MTD) of vorinostat combined with 250 mg of gefitinib. This is the first phase I study of combined therapy with vorinostat and gefitinib for NSCLC patients double selected for an *EGFR* mutation and *BIM* deletion polymorphism. *J. Med. Invest.* 64 : 321-325, August, 2017

Keywords : *EGFR* mutation, *BIM* polymorphism, gefitinib, vorinostat, non-small cell lung cancer

INTRODUCTION

The majority of patients with non-small cell lung cancer (NSCLC) with epidermal growth factor receptor (*EGFR*) activating mutations, such as exon 19 deletion and L858R point mutation, show marked responses to *EGFR* tyrosine kinase inhibitors (*EGFR*-TKIs), such as gefitinib, erlotinib, and afatinib (1-4). However, 20-30% of patients with *EGFR*-activating mutations show intrinsic resistance to *EGFR*-TKIs. Molecular mechanisms of the intrinsic resistance are not fully understood (5).

BIM, also called Bcl-2-like protein 11, is a pro-apoptotic molecule that belongs to the Bcl-2 family. *BIM* upregulation is essential for the induction of apoptosis in lung cancer cells with *EGFR* mutations treated with first-generation *EGFR*-TKIs, and a low *BIM* protein level is associated with resistance to *EGFR*-TKIs (6, 7). Recently, an East Asian-specific 2,903 bp deletion polymorphism in the *BIM* gene was discovered, whose incidence was around 13% and 0.5% for heterozygous and homozygous carriers, respectively (8). Importantly, the *BIM* deletion polymorphism results in the preferential splicing of exon 3 over the BH3-encoding exon 4 in the *BIM* pre-mRNA, and leads to the production of inactive *BIM* isoforms lacking the BH3 domain. This in turn reduces expression of

pro-apoptotic *BIM* protein isoforms in *EGFR*-mutant lung cancer cell lines following TKI exposure and is sufficient to confer TKI resistance (8). Since its initial discovery, several meta-analyses have reported an association between *BIM* deletion polymorphism and shorter progression-free survival (PFS) of patients with NSCLC harboring *EGFR* mutations, who received gefitinib or erlotinib treatment (9-13).

Vorinostat (suberoylanilide hydroxamic acid [SAHA]), has been approved in 20 countries to date including Japan for cutaneous T-cell lymphoma as monotherapy, is a small-molecule inhibitor of histone deacetylase (HDAC) that induces cell differentiation, cell cycle arrest, and apoptosis in several types of tumor cell lines (14). We previously reported that the combined use of vorinostat and gefitinib was able to preferentially upregulate the expression of pro-apoptotic *BIM* isoforms in *EGFR*-mutant NSCLC cell lines with the *BIM* deletion polymorphism, and overcome *EGFR*-TKI resistance *in vitro* and *in vivo* (15). Two clinical trials, a phase I/II study combining gefitinib and vorinostat in patients with advanced NSCLC regardless of presence/absence of *EGFR* mutation in Korea (16) and a phase I/II study combining erlotinib and vorinostat with advanced *EGFR*-mutant NSCLC patients after *EGFR*-TKI progression in Spain (17) have been performed. However, the combination treatment did not show significant efficacy in these patient population and novel biomarker is warranted. Therefore, based on our preclinical findings, we designed the present phase I study named VICTROY-J “Vorinostat-Iressa Combined Therapy on Resistance by *BIM* Polymorphism in *EGFR* Mutant Lung Cancer” to evaluate the safety of combined therapy with vorinostat and gefitinib, and to

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determine the maximum tolerated dose (MTD) of vorinostat combined with a fixed dose of gefitinib for Japanese patients with *EGFR*-mutant NSCLC with a *BIM* deletion polymorphism.

METHODS AND DESIGN

Purpose

The primary objective is to determine the MTD of vorinostat combined with a fixed dose of gefitinib for patients with *EGFR*-mutant NSCLC with a *BIM* deletion polymorphism. The secondary objective is to evaluate the safety and efficacy of the combined therapy with vorinostat and gefitinib in the early-phase trial setting.

Study design

This study is an open-label, multi-institutional phase I dose-escalation study of participating institutions, include 5 specialized centers in Japan as of November 2016. Participating institutions are listed in Appendix 1.

Three to six patients will be enrolled at each dose level of vorinostat. With a fixed dose of gefitinib, dose escalation of vorinostat following a conventional 3+3 design using an escalation scheme will be used (Figure 1). Initially, 3 patients are enrolled to level 1. If one or two patients experience DLT, 3 additional patients are enrolled to the level. If 3 of 6 patients experience DLT, the previous level is declared the MTD. If 2 or less of 6 patients experience DLT, dose escalation is permitted to continue. After the termination of protocol treatment, any treatment is allowed.

Ethical considerations and registration

This study was conducted in accordance with the International Committee for Harmonization Good Clinical Practice (ICH-GCP) guideline and the Declaration of Helsinki. The study protocol was approved by the institutional review boards of all participating institutions. Informed consent will be provided for all patients

before registration. This study was registered with ClinicalTrials.gov (NCT02151721) and UMIN Clinical Trials Registry (UMIN 000015193).

Endpoint

The primary endpoint is MTD, which is defined as the highest dose level at which 2 or less of 6 patients experience a dose-limiting toxicity (DLT). Toxicities will be graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE) version 4.0. DLT is defined as follows ; grade ≥ 1 intestinal lung disease ; grade ≥ 4 neutropenia lasting 5 days or more ; febrile neutropenia ; grade ≥ 3 thrombopenia requiring platelet transfusion ; grade ≥ 4 thrombopenia ; any grade uncontrollable skin toxicity ; grade ≥ 3 nonhematological toxicity. DLT will be evaluated during the first two cycles (14 days per cycle) of therapy.

The secondary endpoints are pharmacokinetics and pharmacodynamics of vorinostat and gefitinib, progression-free survival (PFS), overall survival (OS), response rate (RR), duration of response and complete response, disease control rate (DCR), and incidence of adverse events defined by Common Terminology Criteria for Adverse Events (CTCAE) version 4.0.

Eligibility criteria

Inclusion criteria

Prior to enrollment in the study, patients must fulfill all of the following criteria : histologically or cytologically diagnosed NSCLC (excluding squamous cell carcinoma) ; NSCLC of clinicopathologic stage IIIB or IV for which radical radiation therapy is impractical or there is a recurrence after surgery ; *EGFR* mutations (deletion of exon 19 and L858R mutation of exon 21) for which the clinical benefits of an *EGFR*-TKI (gefitinib or erlotinib) are recognized by testing methods that are listed by the national health insurance ; having a history of treatment with an *EGFR*-TKI (gefitinib or erlotinib) and a history of pathologic deterioration during treatment ; having a history of treatment with cytotoxic anticancer agents (not including pre- or postoperative chemotherapy in the previous 1 or

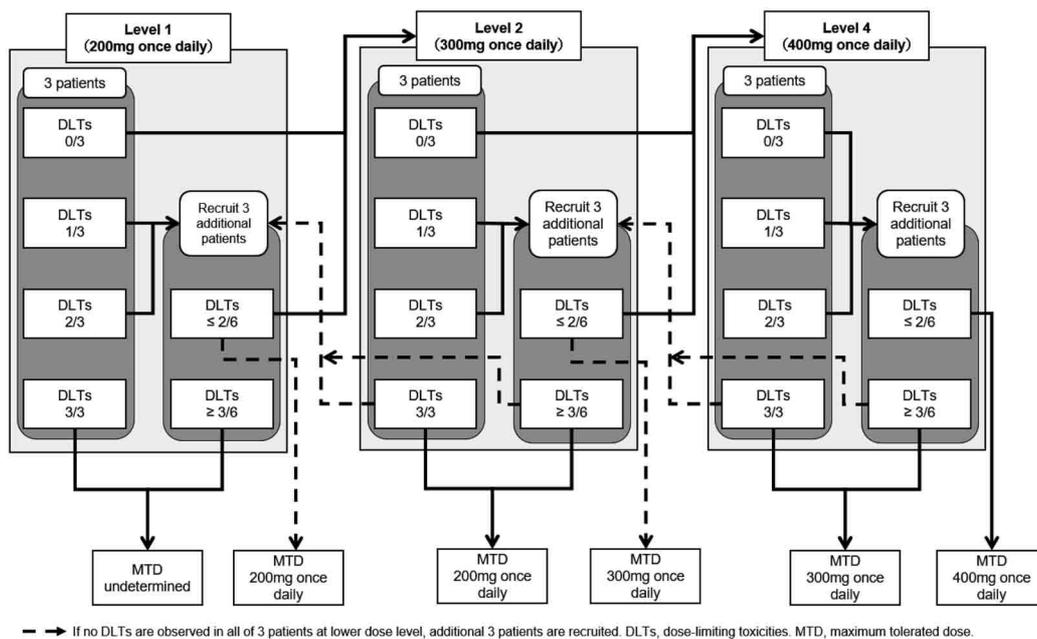


Figure 1. Study design.

Dashed arrow ; if no DLTs are observed in all of 3 patients at lower dose level, additional 3 patients are recruited. DLTs, dose-limiting toxicities. MTD, maximum tolerated dose.

with gefitinib in Japanese *EGFR*-mutant NSCLC patients with *BIM* deletion polymorphism in this investigator-initiated trial, we would like to conduct phase II study in cooperation with pharmaceutical companies. If successful, this combined treatment with vorinostat and gefitinib may lead to substantial and important changes in the management of patients with *EGFR* mutant NSCLC with a *BIM* deletion polymorphism.

CONFLICT OF INTERESTS-DISCLOSURE

Yoshinori Hasegawa obtained speakers fees and research grant from AstraZeneca, Taiho, and MSD. Toshiaki Takahashi obtained speakers fees from AstraZeneca and Taiho and research grant from AstraZeneca, MSD, and Taiho. Nobuyuki Katakami obtained speakers fees and research grant from AstraZeneca and Taiho. Akira Inoue obtained speakers fees from AstraZeneca, Taiho and advisory fees from AstraZeneca and MSD. Seiji Yano obtained speakers fees and research grants from AstraZeneca and Taiho. The other authors have nothing to disclose.

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APPENDIX 1. Participating institutions

Participating institution	Principal investigator
1. Kanazawa University Hospital	Shinji Takeuchi
2. Nagoya University Hospital	Yoshinori Hasegawa
3. Institute of Biomedical Research and Innovati on Hospital	Nobuyuki Katakami
4. Tohoku University Hospital	Akira Inoue
5. Shizuoka Cancer Center	Toshiaki Takahashi

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