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Original Research

# Phase I clinical trial of a novel procaspase-3 activator SM-1 with temozolomide in recurrent high-grade gliomas

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ABSTRACT

*Objective:* Despite a standard of care, the mortality of recurrent high-grade gliomas (HGGs) remains high. SM-1 is a novel molecular activator that has shown to target procaspase-3, which is overexpressed in HGGs. A phase I clinical trial was conducted to evaluate the safety, pharmacokinetics, and primary clinical efficacy of SM-1 plus TMZ. Participants received escalating doses of daily oral SM-1 (450, 600, and 800 mg) plus standard TMZ therapy.

*Methods:* In the preclinical study, the synergistic effects of SM-1 and temozolomide (TMZ) in rodent models were evaluated. In the clinical study, adult patients received SM-1 therapy in various doses in combination with a standard TMZ dosing. The tolerability and pharmacokinetics data of the combination therapy were tested. The primary efficacy was measured by tumor response in accordance with the RANO criteria.

*Results:* A total of 13 patients with recurrent HGG were enrolled, with 11 patients completed $\geq$ two cycles of therapy and received tumor assessment. Among them, one patient had complete response, whereas two patients had partial response for the best change from baseline. No dose-limited toxicities were observed, and no maximum tolerated dose was reached.

*Conclusion:* SM-1 has the potential to enhance antitumor activity while alleviating the side effects of TMZ. SM-1 exhibited mild toxicity in patients with recurrent HGG. The combination of SM-1 and TMZ warrants further investigation, with promising clinical outcomes. The monotherapy phase and expansion phase of SM-1 are still ongoing. (ClinicalTrials.gov number, CTR20221641).

## Introduction

Gliomas are the most common subset of primary brain tumor in adults [1]. WHO Grade 3 and 4 gliomas are categorized as high-grade gliomas (HGGs), accounting for more than 50% of all malignant brain tumors [2]. For recurrent HGG patients, surgery is a viable option, but outcomes hinge on complete tumor resection and tumor invasiveness, with anatomical complexities affecting the success of re-operation. [3,4]. In cases of repeated radiotherapy, evidence to support its efficacy in extending survival time is insufficient [3]. Drug therapy appears to present fewer limitations and provide more considerable benefits in treating patients with recurrent HGG. Given that no therapeutic strategy is curative for recurrent HGG, enrollment in clinical trials is the preferred option if possible.

Although advances in genomic profiling have been made, HGGs remain lethal and inevitably progressive. Effective and less toxic new therapies for HGGs are still elusive. Temozolomide (TMZ), a common alkylating agent, is a crucial part of both newly diagnosed and recurrent HGGs [5,6], it provides more clinical benefits to patients with methylated O-6-methylguanine-DNA methyltransferase (MGMT) genes than to those with unmethylated MGMT [7]. However, a major concern in patients with HGGs is TMZ resistance, which is mainly related to hyperactivation of MGMT [8]. Combination regimens of multiple drugs often exhibit better inhibition of tumor cells compared to single-drug chemotherapy and is a growing research area [9,10]. In light of this, pro-apoptotic drugs may result in cell cycle arrest as cells attempt to

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repair DNA, thereby potentially enhancing tumor sensitivity to TMZ.

Procaspase-3 (PC-3) is the zymogen precursor to caspase-3, which is an executioner caspase responsible for the cleavage of numerous proteins in apoptosis [11]. Due to the overexpression of PC-3 in HGGs and its crucial role in apoptotic-inducing agents, PC-3 activation can be a novel anticancer target [12–16]. A team screened a small molecular compound known as procaspase-activating compound 1 (PAC-1) in 2006 [17], and subsequent experiments explored its potential in the treatment of gliomas [18,19]. Furthermore, a phase I study for advanced malignancy treatment identified the safety profile of PAC-1 [20]. A clinical trial continued to explore the tolerability and pharmacokinetics (PK) of PAC-1 plus TMZ, and although the dose escalation was stopped of PAC-1 at 625 mg, it provided preliminary evidence of the safety profile and clinical benefit of the combination [21].

Shenzhen Zhenxing Medical Technology Co., Ltd. synthesized a series of new compounds and selected SM-1 with significant anticancer activity to develop more potent PC-3 activators [22]. SM-1 is a small-molecule PC-3 activator that has the potential to be a proapoptotic agent for the treatment of cancer [22]. Previous studies indicated that SM-1 induced apoptosis in human gastric carcinoma cells [23], colorectal cancer cells [24], and other human carcinoma cell lines [22]. The phase I clinical trial of SM-1 with concomitant TMZ chemotherapy was initiated in patients with recurrent HGGs.

## Materials and methods

### *Study designs and participants*

This study was a prospective, open-label, dose-escalation phase I study of SM-1 with TMZ in patients with recurrent HGGs. The clinical trial enrolled patients at Beijing Tiantan Hospital, and it was sponsored by Shenzhen Zhenxing Medical Technology Co., Ltd., with drug supply from CTR20221641 program.

Patients aged 18 years or older with a KPS score $\geq$ 60 and advanced HGGs (except for brain stem gliomas) that recurred or progressed following standard of care. Patients were required to have histologically confirmed (including resection or biopsy) or radiographically measurable disease in accordance with response assessment in neuro-oncology (RANO) criteria [25]. Additional eligibility criteria were adequate hepatic function (total bilirubin $\leq 1.5 \times$ the upper limit of normal (ULN), aspartate aminotransferase, alanine aminotransferase $\leq 2.5 \times$ ULN, and $\leq 5 \times$ ULN if liver metastases are present), adequate renal function (creatinine clearance $\geq$ 60 mL/min according to Cockcroft Gault Formula), adequate bone marrow function (absolute neutrophil count $\geq 1.5 \times 10^3$ μL, hemoglobin $\geq$ 9 g/dL, and platelets $\geq 100 \times 10^3$ μL), and adequate cardiac function (LVEF $\geq$ 50%). Patients were excluded if they underwent surgery or radiotherapy or received an investigational agent or any other form of anticancer therapy within 4 weeks from the start of the study. The exclusion criteria also included active severe infection, medical history of immunodeficiency disorders, cardiovascular diseases, active viral infections (including hepatitis B, hepatitis C, syphilis, and HIV), history of drug or alcohol abuse, bleeding/thrombotic disorders, problematic wound healing, factors affecting oral drug absorption (such as inability to swallow, chronic diarrhea, and intestinal obstruction), and known hypersensitivity to any component of SM-1 or TMZ. Patients who were pregnant or lactating, received hematopoietic cytokine therapy within 7 days prior to enrollment, or had other uncontrolled diseases were excluded. Patients with other malignancies were eligible only if they had been progression-free for at least 5 years and were deemed at low risk of recurrence by the investigator or if the tumor was *in situ*.

The study was conducted following the principles of the Declaration of Helsinki and Good Clinical Practice guidelines. The protocol was approved by the Ethics Committees of the Beijing Tiantan Hospital of Capital Medical University (Approval No. YW2022-025-01). A written informed consent was obtained from each participant or their legal representative before any study-specific procedure.

### *Treatment plan*

During dose escalation, the patients received single-dose administration of SM-1 on cycle 0 day 1, and completed PK sampling and relevant safety assessment. Cycle 0 lasted for 7 days, followed by multiple-dose administration (cycles 1, 2, and 3+). SM-1 was administered orally at 450, 600, or 800 mg (dose level 1, 2, or 3) daily in 28-day cycles in a standard 3 + 3 design, with concomitant TMZ chemotherapy. During the combination therapy, TMZ was taken once a day at 150 or 200 mg/m$^2$ for 5 days in a row, followed by a 23-day break before repeating the next dosage cycle, for up to 6 cycles in total. After the combination therapy, the participants entered the phase of SM-1 monotherapy in the opinion of the investigator (the schematic diagram is shown in Fig. 1). No placebo was administered, with no intra-patient dose escalation.

Dose-limiting toxicities (DLTs) were evaluated during cycles 0 and 1 of the dose-escalation phase and defined as any grade 3 or higher event reported to be related or possibly related to SM-1. If the patient did not experience any DLTs or grade 2 adverse events (AEs) that were deemed related to the study treatment, a protocol amendment may be considered to include the evaluation of higher dose levels.

Treatment was intended to continue until progressive disease (PD) per RANO criteria by brain enhanced MRI every two cycles (or MRI at any time when PD was suspected), or until unacceptable toxicity, which was evaluated using the NCI Common Terminology Criteria for Adverse Events (CTCAE, version 5.0), or initiation of a new therapy, or consent withdrawal. Patients could withdraw from the study treatment at any time at the request of their own or their parents, or at the discretion of the investigator for safety, behavioral, or administrative reasons.

### *Safety*

The safety population consisted of all patients who received any amount of SM-1 and was used for all safety analyses. The primary objective of this study was an assessment of safety and tolerability. Safety was continuously assessed through physical examination, vital signs, clinical laboratory, electrocardiogram, and AEs. The AEs were coded by system organ class and preferred term by using the current version of the Medical Dictionary for Regulatory Activities (MedDRA). They were documented and graded according to CTCAE (version 5.0).

### *PK assessments*

The PK analysis population was defined as those patients who received at least one SM-1 dose and provided sufficient data for a concentration–time profile. Serial blood samples were drawn before and after dosing at multiple timepoints (1, 2, 3, 4, 6, 8, 12, 24, 48, and 96 h after the first dose in C0D1) in the escalation phase to determine the circulating plasma concentrations of SM-1. Blood sampling was conducted before and after dosing on C1D5 to obtain the PK data for multiple-dose administration.

The exploratory assessments included confirmation of baseline MGMT promoter methylation status determined by hospitals or laboratories that accord with national standards (not limited to detection method).

### *Exploratory evaluation of clinical efficacy*

Radiographic evaluations were conducted every two cycles to evaluate the clinical efficacy of the treatment, and the responses were classified as complete response (CR), partial response (PR), stable disease (SD), or PD. During the study treatment, brain MRIs could be conducted at any time when PD was suspected according to the protocol and opinion of investigators. All patients were followed up until 28 days after their last dose or the initiation of a new antitumor therapy.

To explore the potential efficacy of SM-1/TMZ combined therapy,


**Fig. 1.** Schematic diagram of phase I clinical trial.
The primary objective of this study was to determine the safety and tolerability of SM-1/TMZ combination therapy on patients with recurrent HGG.

the Rat C6 glioma cell line was cultured and subsequently injected beneath the rat's skull, at a location slightly to the right rear of the top of the brain of the Wistar mouse (female, 120-150g) to establish a tumor model. The rats were divided into groups for drug administration based on their respective weights, as outlined below:

(1) Control group (CON);
(2) SM-1 group including (a) 40mg/kg, (b) 80mg/kg and (c) 160mg/kg;
(3) TMZ group including (a) 6.25mg/kg, (b) 12.5mg/kg and (c) 25mg/kg;
(4) Combination group including (a) SM-1(40mg/kg)/TMZ(6.25mg/kg) group, (b) SM-1(80mg/kg)/TMZ(6.25mg/kg) group, (c) SM-1(160mg/kg)/TMZ(25mg/kg) group.

The mice received daily oral doses according to their respective groupings, with SM-1 dissolved in a 40% hydroxypropyl-β-cyclodextrin solution and TMZ dissolved in a 0.5% sodium carboxymethyl cellulose solution. The experiment ended when reaching the point at which half of the rats in the CON had expired. The data were presented as means ± standard deviation (SD), with tumor growth inhibition (%) calculated as follows: (1 - [total weight of tumor-bearing brain in the administration group / total weight of tumor-bearing brain in the CON]) × 100%. Group-level statistical analysis was conducted using one- or two-way analysis of variance tests (ANOVA) via GraphPad Prism5. Survival analysis was performed using the Kaplan-Meier method, also with GraphPad Prism5.

### *Statistical analysis*

The data cutoff date for the statistical analysis was March 27, 2024. All data were summarized or listed on the basis of the relevant analysis population. Descriptive statistics were applied for clinical and PK parameters.

## Results

### *Patients*

From September 2022 to March 2024, a total of 13 participants were screened for enrollment, of whom 11 received the study treatment [median age, 53 years, (range of 31–62 years)]. Four and seven were male and female patients, respectively, and all were Han Chinese. According to the 2021 WHO Classification, most patients (n = 8) had been diagnosed with GBM at screening, and the rest had anaplastic astrocytoma, gliosarcoma mixed with anaplastic astrocytoma, and anaplastic oligodendrogliomas. Except for two cases (18.2%) of grade 3 glioma, the remaining nine patients had grade 4 glioma. All tumors were supratentorial. The patient characteristics are detailed in Table 1.

One patient was not included in the DLT set, and another patient was not included in the effectiveness analysis set. Subject S01005 (dose level 2) withdrew from the study due to AE, with the last dosing on C1D4. Subject S01012 (dose level 3) withdrew from the study due to PD, with the last dosing on C1D10.

### *Safety*

All patients had at least one AE. The profile is shown in Table 2. and the details are shown in Supplementary Tables 1 and 2. Three patients (27.3%) experienced a grade 3 TEAE, including decreased lymphocyte count and cerebral edema at dose level 3 and cerebral edema at dose level 2. All other reported TEAEs were grade 1 or 2.

All patients experienced AEs that were possibly or unlikely related or unrelated to SM-1, with the common ones being metabolism and nutrition disorders (especially hypertriglyceridemia), eye disorders, and decreased lymphocyte count. No AEs resulted in dose reduction. One patient (S01005, dose level 2) discontinued the study treatment due to AE (intracranial hemorrhage, grade 1, possibly related to SM-1/TMZ; cerebral edema, grade 3, unlikely related to SM-1/TMZ), which was consistent with prior medical history. The AE (cerebral edema) was initially rated as grade 2 during the screening period and upgraded to grade 3 by a follow-up CT scan on C1D4. Given the patient's insufficient duration of drug exposure, this AE was deemed to be related to the tumor.

No DLTs were observed. No patient experienced an SAE in this phase I trial.

The results demonstrated that SM-1 could be combined safely with TMZ at the recommended dose, with acceptable toxicity and side effects.

### *Pharmacokinetics*

PKs are summarized in Table 3. In patients with HGG, SM-1 could be absorbed and eliminated rapidly across the dose levels, with a median $T_{max}$ of 4.05 h (range of 1.95–8.00 h). The $t_{1/2}$ was similar, with a median of 16.58 h after a single dose. After multiple oral administrations of different doses of SM-1, the overall PK characteristics were similar to those observed after a single administration. The analysis supported a daily dose regimen. The PK data indicated a dose-dependent increase in AUC, $C_{max}$, $AUC_{0\text{-}24h}$, and $AUC_{(0\text{-}\infty)}$ in all patients, and wide interpatient variabilities were observed across dosage levels due to the relatively small number of patients.

### *Exploratory evaluation of clinical efficacy*

At the time of data cutoff (March 27, 2024), the median duration was 66 days (range of 18–272 days). One patient was still on treatment and remained suggestive of clinical benefit. Nine out of the eleven patients stopped because of progression. The remaining one patient withdrew because of AE and died during the follow up period, which was possibly related to his previous medical history (S01005, April 14, 2023). A

**Table 1**
Patient demographics, baseline characteristics by patient identification number.

| # | Dose level | Age (years) | Sex | BSA ($m^2$) | KPS | Diagnosis | Tumor grade (per WHO 2021) | MGMT | IDH | Status | Weeks of SM-1 treatment | Number of surgeries | Recurrence before SM-1 (times) | Prior TMZ therapy duration (cycles) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| S01001 | 1 | 31 | Female | 1.48 | 80 | anaplastic astrocytoma | 3 | methylated | Mutant | Discontinued due to PD | 42.14 | 1 | 2 | 7 |
| S01003 | 1 | 53 | Male | 1.74 | 60 | gliosarcoma mixed with anaplastic astrocytoma | 4 | methylated | Mutant | Discontinued due to PD | 24.14 | 2 | 3 | 7 |
| S01004 | 1 | 55 | Female | 1.55 | 70 | anaplastic oligodendrogliomas | 3 | methylated | Mutant | Treatment ongoing | 72.29 | 2 | 2 | 12 |
| S01005 | 2 | 46 | Male | 1.93 | 60 | Glioblastoma | 4 | unmethylated | Wild type | Died* | 0.57 | 2 | 3 | 7 |
| S01006 | 2 | 56 | Female | 1.64 | 70 | Glioblastoma | 4 | unmethylated | Wild type | Discontinued due to PD | 8.14 | 1 | 1 | 8 |
| S01008 | 2 | 60 | Female | 1.72 | 70 | Glioblastoma | 4 | methylated | Wild type | Discontinued due to PD | 56.57 | 1 | 1 | 6 |
| S01009 | 2 | 59 | Female | 1.32 | 70 | Glioblastoma | 4 | unmethylated | Wild type | Discontinued due to PD | 8.00 | 2 | 1 | 11 |
| S01010 | 3 | 48 | Female | 1.7 | 70 | Glioblastoma | 4 | N/A | Wild type | Discontinued due to PD | 8.00 | 3 | 5 | 18 |
| S01011 | 3 | 62 | Female | 1.8 | 60 | Glioblastoma | 4 | methylated | Wild type | Discontinued due to PD | 32.29 | 2 | 2 | 6 |
| S01012 | 3 | 46 | Male | 1.68 | 60 | Glioblastoma | 4 | unmethylated | Wild type | Discontinued due to PD | 1.57 | 2 | 2 | 6 |
| S01013 | 3 | 50 | Male | 1.97 | 80 | Glioblastoma | 4 | unmethylated | Wild type | Discontinued due to PD | 8.14 | 1 | 1 | 6 |

N/A, not available; Prior TMZ therapy duration, the duration of TMZ administration subsequent to concurrent chemoradiotherapy.
*The patient exited the clinical trial due to disease progression and passed away four months later.

**Table 2**
AE summary.

| AE summary | 450mg n=3(%) | 600mg n=4(%) | 800mg n=4(%) | Total n=11 (%) |
|---|---|---|---|---|
| Total patients | 3 | 4 | 4 | 11 |
| ≥Grade 3 TRAEs reported | 0 | 0 | 1(25%) | 1(9.1%) |
| AEs per patient, median (range) | 11.33(9-13) | 8.25(5-16) | 4.5(0-10) | 7.73(0-16) |
| TEAE related to SM-1 | 3(100%) | 4(100%) | 3 (75.0%) | 10 (90.9%) |
| TEAE related to TMZ | 3(100%) | 4(100%) | 3 (75.0%) | 10 (90.9%) |
| SAE | 0 | 0 | 0 | 0 |
| TEAE leading to discontinuation of the study drug | 0 | 1 (25.0%) | 1 (25.0%) | 2 (18.2%) |
| TRAE leading to discontinuation of the study drug (SM-1 related) | 0 | 1 (25.0%) | 0 | 1(9.1%) |
| TRAE leading to discontinuation of the study drug (TMZ related) | 0 | 1 (25.0%) | 0 | 1(9.1%) |
| TEAEs leading to discontinuation | 0 | 0 | 1 (25.0%) | 1(9.1%) |
| TEAE leading to discontinuation of the study drug (TMZ related) | 0 | 0 | 0 | 0 |
| TEAE leading to discontinuation of the study drug (TMZ related) | 0 | 0 | 0 | 0 |

(1) The severity of AEs is assessed on the basis of CTCAE5.0 criteria.
(2) AEs per patient are defined as the average of AEs of patients till the cut-off date. If the same patient experiences the same AE multiple times, these AEs are aggregated and counted as one event to avoid double counting.
(3) Treatment-related adverse events (TRAEs) are defined as all AEs that are categorized as definitely related, probably related, or possibly related to the study drug.
(4) Severe TRAEs are defined as SAEs that are categorized as definitely related, probably related, or possibly related to the study drug.
(5) Treatment-emergent adverse events (TEAEs) are defined as all AEs that occur from the first dose of the study drug until the safety follow-up visit.
(6) In the case of the same subject experiencing the same AE multiple times, the severity analysis considers the most severe occurrence of that AE in which the subject participated.

**Table 3**
Pharmacokinetic results.

| Parameters, units (median, range) | Cycle (C), Day (D) | 450mg/d N=3 | 600mg/d N=4(C0D1)* | 800mg/d N=4 |
|---|---|---|---|---|
| Cmax, ng/mL | C0D1 | 733.00 (310.50-1015.00) | 1208.45 (941.20-1604.00) | 1469.50 (1169.00-1597.00) |
| | C1D5 | 1306.00 (1026.00-1706.00) | 1622.00 (1508.00-2039.00) | 2360.50 (1553.00-3133.00) |
| Tmax, hours | C0D1 | 5.95 (2.97-5.98) | 5.02 (4.00-8.00) | 3.53 (1.95-6.02) |
| | C1D5 | 4.00 (4.00-4.02) | 3.98 (3.00-6.02) | 2.53 (2.02-4.05) |
| $t_{1/2}$, hours | C0D1 | 20.26 (11.81-27.45) | 19.00 (15.93-31.42) | 16.04 (15.07-23.05) |
| | C1D5 | 17.91 (12.16-18.78) | 22.94 (19.28-24.96) | 13.05 (7.70-24.92) |
| $AUC_{0-24h}$, ng·h/mL | C0D1 | 9160.61 (5389.07-10005.78) | 15531.14 (12520.43-23013.21) | 18164.66 (9853.89-21692.09) |
| | C1D5 | 16789.55 (13334.91-21597.01) | 23591.62 (21681.73-31695.42) | 29938.04 (19397.69-40672.34) |
| $AUC_{(0-\infty)}$, ng·h/mL | C0D1 | 12091.11 (11592.63-17381.05) | 25878.37 (20820.78-45921.77) | 29141.11 (13176.22-42606.24) |
| | C1D5 | 16751.48 (13301.29-21551.63) | 23473.99 (21681.92-31609.38) | 40511.91 (24442.33-84652.10) |

*One patient at dose level 2(S01005, 600mg) withdrew from the study early and PK samples for C1D5 were not collected, resulting in the inability to calculate PK parameters.

notable detail that the best overall therapeutic efficacy was shown by one patient (9.1%) having CR while on treatment with SM-1/TMZ and two patients (18.2%) having PR. In addition, two patients had SD with a reduction in tumor size. The new therapy had an overall response rate (ORR) of 27.3% and a disease control rate (DCR) of 45.5%, indicating the treatment's effectiveness in managing HGGs. The duration of treatment is shown in Fig. 2.

Exploratory assessments including MGMT status are shown in Table 1. Although assessment by genetic subtype was not a prespecified analysis, the responses of patients with methylated MGMT promoter were better than those of others (Fig. 3).

The antitumor activity of SM-1 was an exploratory objective in this dose-escalation study. Considering that it is an early phase I trial, the results are encouraging.

As for the preclinical study, the primary index of the anti-tumor activity was the survival rate. Rats were administered a total of 11 doses of SM-1 at 40mg, 80mg, and 160mg, and the survival rates for the three groups were 60%, 80%, and 90% respectively, while the survival rates for the CON was 40% (Supplementary Table 2). Moreover, the brain wet weight/body weight index of surviving animals in SM-1 group was lower than that of the CON (Supplementary Table 3), indicating that SM-1 had anti-cancer activity as a single agent. The survival rates of the combination therapy groups were higher than the respective corresponding monotherapy groups, with a survival rate of 90% in the SM-1 (40mg/kg)/TMZ(6.25mg/kg) group and 100% in the other group. According to the one belt one line model, the survival rates of the rat C6 glioma model were analyzed by separately simulating the dose-response curves of TMZ and SM-1. Fig. 4 demonstrates the synergy between SM-1 and TMZ in the orthotopic C6 glioma rat model.

*Clinical effects of special interest*

A 60-year-old female patient (Patient S01008) presented with bilateral lower limb weakness, increased headache, nausea, and vomiting. After the intracranial mass was confirmed by MRI, the patient underwent gross tumor resection of the corpus callosum and left thalamus (2021-11-18). The pathological diagnosis was GBM. The molecular test results suggested IDH wild type, TERT+, and positive methylation at the MGMT promoter (20%). The fluorescence in-situ hybridization indicated no loss of chromosome 1p/19q and a Ki-67 labeling index of 30%. After concurrent chemoradiotherapy (60Gy/30f, PTV; 75 mg/m$^2$ of TMZ daily) and six cycles of TMZ chemotherapy (200 mg/m$^2$, days 1–5, every 28 days), the symptoms of fatigue and weakness worsened, and the MRI of the patient confirmed a progression on December 2022.

After screening for eligibility, treatment with 600 mg of SM-1 (D1-28) combined with 260 mg of TMZ (D1-5) was provided in a 28-day cycle from December 27, 2022. After a single cycle, the clinical symptoms revealed slight improvement. A reduction in neoplastic tissue in the corpus callosum was described at the first follow-up (administration for two cycles), and the lesion was determined as stable disease in accordance to the RANO criteria. The patient subsequently followed the medication regimen on the basis of the clinical trial protocol. A new MRI after another two cycles of therapy demonstrated that the patient achieved PR with a 56.93% reduction in the cross-sectional area of the target lesion compared with the baseline at the third tumor assessment. The serial MRI evaluation of Patient S01008 is shown in Fig. 5. The PFS was 13.2 months.

## Discussion

This study is the first reported phase I clinical trial of a combination

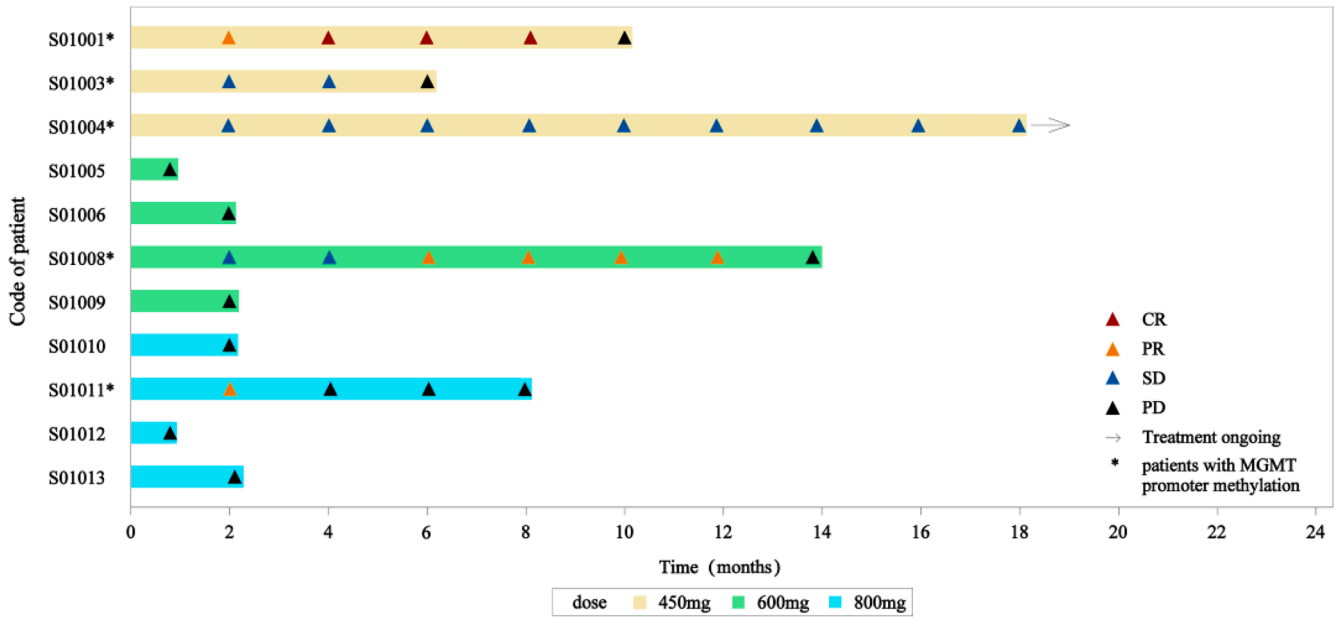


**Fig. 2.** Duration of treatment and response to treatment at different dose levels.
Responses were assessed according to RANO criteria, CR complete response, PR partial response, SD stable disease, PD progressive disease.

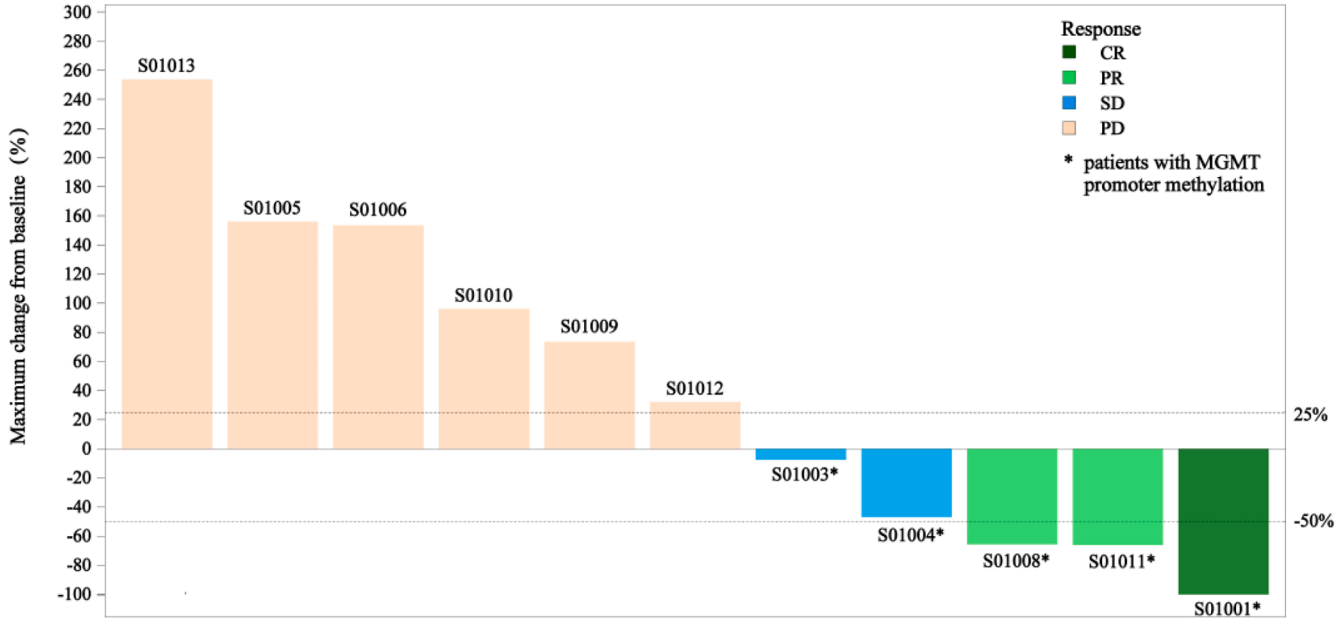


**Fig. 3.** Best change from baseline in the product of perpendicular diameters of the largest tumor cross-section of all target lesions in the subject (FAS)

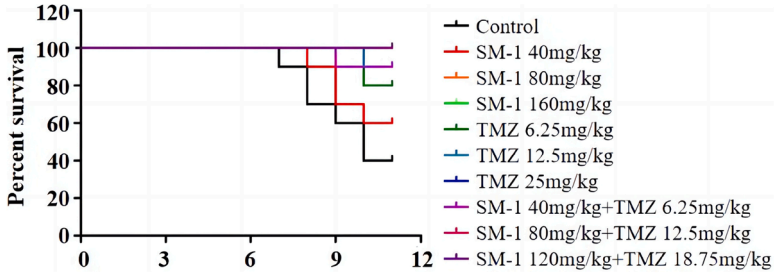


**Fig. 4.** The impact of SM-1/TMZ combination on orthotopic C6 glioma rats' survival
A significant statistical difference was found between the group treated with the drugs and the control group (C6 glioma cells implanted in Wistar rats, Log-Rank test, $P<0.01$).

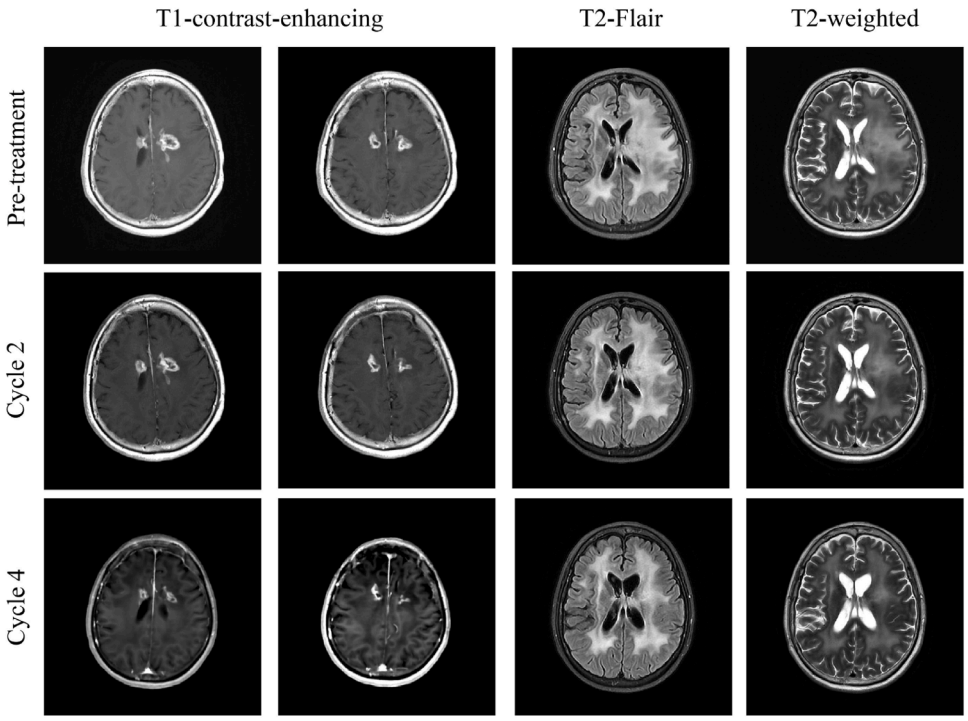


**Fig. 5.** Tumor responses by consecutive T1-weighted MRI scans with intravenous contrast, and MRI images of T2-weighted and T2 flair in Patient S01008. Pre-treatment: baseline images of the first recurrence after Stupp regimen. Cycle 2: images obtained at 8 weeks of SM-1/TMZ administration, Cycle 4: images obtained at 16 weeks of SM-1/TMZ administration, Baseline images demonstrate the bilateral frontal lobe and periventricular nodular lesions. Cycle2 images demonstrated a slight decrease in lesion size compared to the baseline images. A continued decrease in lesion size is seen in Cycle 4 images. In addition, T2-weighted images showed a widespread abnormal signal in the brain with a reduced extent compared to before.

of SM-1 and TMZ therapy, using a standard 3 + 3 dose-escalation design.

The study tested three dose levels, assessing the safety and PK in patients with recurrent HGG. The majority of AEs were hyperlipidemia, decreased lymphocyte count, and optic nerve disorder. Occasional mild-to-moderate toxicities possibly related to SM-1 were observed, with the majority of AEs being grades 1 and 2, suggesting that SM-1 was well tolerated in patients even at the highest dose. In this study, SM-1 at 800 mg dose level demonstrated relative safety and partial therapeutic efficacy when combined with the standard TMZ therapy. Although the results have not been disclosed, during the SM-1 monotherapy dose-escalation phase for solid tumors, hallucinations were observed in a participant at the 1050 mg dose level. Therefore, prioritizing the safety of the patients, we did not further escalate the dose to explore the maximum tolerated dose (MTD) in order to avoid potential serious adverse events. PKs were examined on all patients, and the PK data supported good absorption of orally administered SM-1, and once-daily oral dosing is very convenient for patients.

Although this study was not powered for survival analysis, SM-1 demonstrated its potential clinical activity. Our preclinical results showed that SM-1 had inhibitory effects in the glioma model and might synergize with TMZ in rodent models. And in the clinical trial, patients enrolled had all been treated with TMZ for at least 6 months, which indicates a reduced likelihood of response to TMZ monotherapy, yet some still benefit from SM-1/TMZ therapy. The variability may be influenced by various factors like the gene status. Therefore, we are considering a comprehensive assessment of the cycles of TMZ treatments received, and the interval since the last administration. Considering that the level of PC-3 was observed to be increased in glioma cells, future directions may consider the collection of related data in patients to explore the clinical mechanism of SM-1 in subsequent trials, particularly the mechanism of its synergistic effects with TMZ [13]. In addition, one patient finished six cycles of SM-1/TMZ therapy and was still in the period of SM-1 monotherapy (S01004, PFS = 16.8 months), showing the sustained efficacy of SM-1.

Although the results are promising for patients with recurrent HGG, this clinical trial focused primarily on safety and PK, and it has limitations shared by other early phase trials, including low sample size, lack of comparative data and proper control group, and absence of *in-vivo* PC-3 detection. Moreover, the SM-1 concentrations in the cerebrospinal fluid were not obtained because lumbar puncture was not conducted on the patients. Further exploration will be conducted in the next phase.

In conclusion, the preliminary results support the combined administration of SM-1 with TMZ in patients with HGG. This trial will proceed to its phase Ib/II clinical trial and further investigate different cohorts to offer a well-tolerated option to enhance the efficacy of conventional chemotherapy.

## Author contribution

M.H searched the literature and drafted the manuscript. Z.K and S.L drafted and proofread the manuscript. B.Z guided the writing of the paper. Y.X and L.L assisted in writing the paper. W.L was responsible for selecting the topic and critically revising important intellectual content. All authors contributed to the article and approved the submitted version.

## Novelty and impact

In this phase I trial, we present SM-1, a novel procaspase-3 activator, in combination with TMZ for recurrent high-grade gliomas. This single-center, open-label study establishes safety profile and primary efficacy of SM-1, highlighting a synergistic potential with TMZ. Our findings lay the foundation for personalized, genetically informed glioma treatments, marking a significant step forward in glioma therapy.

## Ethics statement

Approval of the research protocol by an Institutional Reviewer Board: The protocol was approved by the Ethics Committees of the Beijing Tiantan Hospital of Capital Medical University (Approval No. YW2022-025-01).

Informed Consent: Written informed consent was obtained from each participant or their legal representative before any study-specific procedure.

Registry and the Registration No. of the study/trial: CTR20221641

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## CRediT authorship contribution statement

**Mengqian Huang:** Writing – review & editing, Writing – original draft. **Zhuang Kang:** Writing – review & editing, Project administration. **Shenglan Li:** Writing – review & editing, Formal analysis. **Botao Zhang:** Methodology, Data curation. **Yantao Xiao:** Visualization, Project administration. **Shangwei Li:** Data curation. **Wenbin Li:** Writing – review & editing, Project administration, Funding acquisition, Conceptualization.

## Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

Yantao Xiao and Shangwei Li serve as employees to Shenzhen Zhenxing Pharmaceutical Technology Co., Ltd.; The study was designed under the responsibility of Shenzhen Zhenxing Pharmaceutical Technology Co., Ltd., in conjunction with the steering committee of Beijing Tiantan Hospital of Capital Medical University; Study drug SM-1 was donated/provided by Shenzhen Zhenxing Pharmaceutical Technology Co., Ltd.; All authors had full access to all of the data in the study and had final responsibility for the decision to submit for publication; other authors have no conflict of interest.

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## Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.neo.2025.101141.

## Data availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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