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SYSTEMATIC REVIEW

From anthelmintic to neuro-oncology: A systematic review of mebendazole repurposing for brain tumour therapy

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Abstract

Aim: Mebendazole (MBZ), a benzimidazole anthelmintic with established clinical use, has emerged as a repurposing candidate for primary brain tumours due to its multimodal anticancer actions and central nervous system penetrance. This systematic review synthesizes preclinical and clinical evidence evaluating MBZ's efficacy, mechanisms of action and translational relevance.

Methods: This systematic review was conducted in accordance with the Joanna Briggs Institute (JBI) methodology. Systematic searches were performed in PubMed, EMBASE, SCOPUS and Web of Science using predefined eligibility criteria. A total of 22 studies were included (17 preclinical and five clinical/population).

Results: Preclinical work across glioblastoma, diffuse midline glioma, medulloblastoma and meningioma demonstrates consistent tumour growth suppression and survival extension via microtubule depolymerization, kinase inhibition, angiogenesis blockade, Hedgehog pathway interference, apoptosis/pyroptosis induction and impairment of DNA repair. MBZ also potentiates standard therapies, enhancing the effects of alkylators, radiotherapy and autophagy inhibitors. Efficacy was influenced by formulation, with polymorph C demonstrating superior brain penetration and tolerability. Additional delivery strategies, including efflux inhibition, intranasal microemulsions and nanosuspensions, further improved exposure. Clinically, MBZ was generally tolerable at high oral doses in early-phase studies, but evidence of efficacy remained modest, inconsistent and inconclusive.

Conclusion: MBZ shows broad preclinical anticancer activity and acceptable tolerability in early human studies, but current clinical evidence does not demonstrate meaningful efficacy in brain tumour patients. Further well-designed comparative trials with clear formulation reporting and integrated pharmacokinetic and biomarker analyses are needed.

KEYWORDS
anticancer, brain tumours, drug repurposing, mebendazole, multimodal therapy

1 | INTRODUCTION

Primary malignant brain tumours remain among the most lethal of human cancers despite decades of research.[1] Standard-of-care treatment, which includes maximal safe surgical resection followed by radiotherapy and chemotherapy,[2] yields only modest improvements in survival, with a 5-year overall survival of around 22%.[3] The World Health Organization (WHO) formally classified primary brain tumours into three malignancy grades: II, III and IV.[3] The fifth edition of the WHO Classification of Tumours of the Central Nervous System, published in 2021, integrates molecular profiling with histopathology to create more biologically and prognostically coherent categories. In adults, diffuse gliomas were consolidated into three groups: astrocytoma with isocitrate dehydrogenase (IDH) mutation, oligodendroglioma with IDH mutation and 1p/19q codeletion and glioblastoma that is IDH wild-type. Central to these revisions is IDH status, which reclassified IDH-mutant glioblastomas as grade 4 astrocytomas, and restricted the diagnosis of glioblastoma to IDH wild-type tumours.[4]

Formally known as glioblastoma multiforme (GBM), IDH wild-type glioblastoma is the most common and aggressive primary brain malignancy in adults, with a life expectancy of around 12–16 months.[5] Following standard treatment, disease recurrence is almost inevitable, driven largely by intrinsic tumour heterogeneity, an infiltrative growth pattern that precludes complete resection and acquired resistance to alkylating chemotherapy.[5,6] Despite this dismal outlook, the Food and Drug Administration (FDA) has not approved any new systemic therapies for GBM since the introduction of **temozolomide** (TMZ) in 2005,[7] underscoring the urgent need for innovative treatment strategies. One of the principal barriers to therapeutic progress in neuro-oncology is the blood–brain barrier (BBB). This tightly regulated interface restricts the penetration of many anticancer agents into brain parenchyma, limiting their efficacy even when they demonstrate potent cytotoxicity *in vitro*.[8] Drug efflux transporters such as **P-glycoprotein** and **BCRP** further reduce brain accumulation of chemotherapy,[9] underscoring the need for agents with both intrinsic anticancer activity and favourable pharmacokinetic profiles for central nervous system (CNS) delivery.

In this context, drug repurposing has emerged as a potential strategy to accelerate the identification of novel treatments for brain tumours. Repurposing leverages the existing safety and pharmacology data of clinically approved drugs, thereby reducing the cost and time required for translation into oncology.[10] Several non-oncology drugs, including antiepileptics,[11] antimalarials,[12] antidepressants[13] and antipsychotics[14] have been investigated in this capacity. Notably, benzimidazole anthelmintics such as **mebendazole** (MBZ) have gained attention in the treatment of brain tumours, owing to their well-documented safety profiles, pharmacokinetics and accumulating preclinical evidence of anticancer efficacy.[15] MBZ, first established as a treatment for helminthic infections in 1972,[16] has demonstrated pleiotropic anticancer activity across multiple tumour types.[17] Importantly, MBZ exhibits the ability to penetrate the BBB, further strengthening its translational relevance in the context of brain cancer.[15]

The aim of this systematic review is to synthesize the available evidence on the anticancer actions of MBZ in primary brain tumours, with particular emphasis on its mechanisms of action, preclinical efficacy, delivery strategies and translational potential. By critically appraising the breadth of existing research, this review seeks to clarify MBZ's potential role within the evolving therapeutic landscape of neuro-oncology, guided by the following research questions:

1. What biological mechanisms underlie MBZ's proposed anticancer activity in preclinical models?
2. Which brain tumour types have been investigated with MBZ treatment, and what patterns of sensitivity, tumour progression and survival outcomes have been reported?
3. How does MBZ interact with standard brain cancer therapies, including chemotherapy and radiotherapy?
4. What clinical evidence exists regarding the use of MBZ as an adjunctive or standalone therapy in patients with brain cancer?

2 | METHODOLOGY

2.1 | Protocol and registration

This systematic review adheres to the reporting standards outlined by the Joanna Briggs Institute (JBI) and follows the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 guidelines.[18] An a priori protocol was developed and registered with the Open Science Framework (OSF) (DOI: 10.17605/OSF.IO/U8WDY). The review was initially registered as a scoping review protocol, but the final approach was refined and conducted as a systematic review. This deviation from the original protocol is documented on the OSF.

2.2 | Search strategy

A structured search was developed using the JBI three-step framework. An initial exploratory search (30 July 2025, PubMed and Google Scholar) identified core terms ('mebendazole', 'glioma', 'brain tumour'), which were expanded iteratively (e.g. 'glioblastoma', 'astrocytoma', 'medulloblastoma') and applied to title and abstract fields. Final strategies were translated across databases using the TERA Polyglot Search Translator[19] (Supplementary 1). Grey literature was searched using Research Rabbit,[20] Litmaps,[21] and TERA Farmer.[22]

2.3 | Information sources

The database search was conducted across four databases (PubMed, EMBASE, SCOPUS and Web of Science) on 31 July 2025, with all retrieved records exported into EndNote X9[23] for screening and organization. The search was subsequently re-run on 12 January 2026 to identify newly published studies, and any additional eligible records were incorporated into the final analysis.

2.4 | Study selection

Duplicates were removed using TERA Deduplicator[19] with manual verification. Title and abstract screening was performed independently by two reviewers (LO and CB) in TERA Screenatron. Studies were included if they investigated MBZ's anticancer effects in brain tumours (*in vitro*, *in vivo* or clinical) and reported original data on cancer-related outcomes (e.g. viability, apoptosis, tumour regression). Exclusion criteria were non-English publications, non-oncological studies, articles without original data and conference abstracts. Full-text screening was conducted in Covidence,[24] with disagreements resolved through discussion.

2.5 | Charting, collating and summarizing the data

Two reviewers (LO and CB) independently extracted data from included studies using a predefined Excel template, following established methodological guidelines. Extracted variables comprised study metadata (author, year, country), sample characteristics (cancer type, cell lines, animal models), experimental methods, assays performed, MBZ exposure parameters (e.g. IC_{50} values, dosages) and key outcomes. Effect measures were defined a priori for each evidence stream. For *in vitro* studies, IC_{50} values and relative cell viability were collected; for *in vivo* studies, tumour volume, histopathological endpoints and survival metrics (e.g. median overall survival) were extracted; and for clinical studies, tumour response, progression-free survival (PFS) and overall survival (OS) were recorded where available. Due to significant heterogeneity in study design, outcome measures and reporting, quantitative synthesis was not feasible. Data were therefore synthesized descriptively and structured according to evidence stream (*in vitro*, *in vivo*, clinical) and outcome domain. Methodological quality was assessed using design-specific risk of bias tools.

2.6 | Risk of bias and certainty of evidence assessment

Methodological quality was independently appraised by two reviewers (LO and CB) using validated, design-specific tools appropriate to each evidence stream: QUIN[25] for *in vitro* studies, SYRCLE[26] for animal studies, ROBINS-I v2[27] for non-randomized clinical studies and

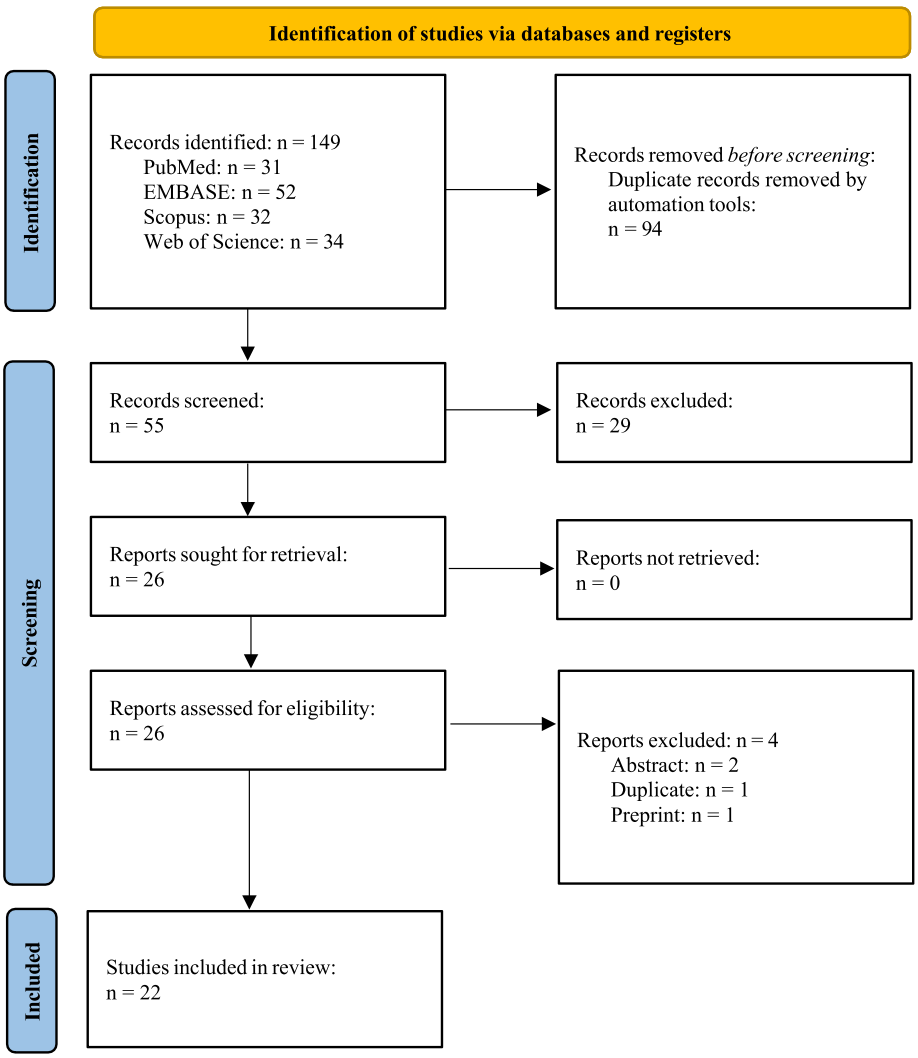

FIGURE 1 PRISMA flow diagram summarizing study identification, screening, eligibility assessment and inclusion. Duplicate records were removed using TERA Deduplicator with manual verification, and screening workflows were supported by TERA Screenatron and Covidence. Data extraction was performed in Microsoft Excel.

RoB 2[28] for randomized trials. Outputs were visualized using *robvis*[29] programme. Certainty of evidence was not graded formally due to the predominantly preclinical evidence base and limited clinical data.

3 | RESULTS

3.1 | Search results

Database searching yielded 149 citations, of which 94 were automatically removed as duplicates. Title and abstract screening of the remaining 55 citations led to the exclusion of 29, leaving 26 articles for full-text screening. After retrieving and screening these 26 articles, four articles were excluded, leaving a final number of 22 articles from the primary database search (Figure 1).

3.2 | Synthesis of results

Twenty-two studies met the inclusion criteria, comprising 17 preclinical (*in vitro*, Table 3; *in vivo*, Table 4) and five clinical or population-based studies (Table 5). Tumour types included GBM, diffuse midline glioma (DMG), medulloblastoma (MB), malignant meningioma and other high-grade gliomas, studied using human and animal models. Research originated from North America (n = 15), Europe (n = 2), Asia (n = 4) and South America (n = 1).

Overall, the evidence base was heavily weighted towards preclinical research. Across experimental models, MBZ demonstrated multimodal anticancer activity, including microtubule disruption, kinase inhibition, angiogenesis blockade, interference with oncogenic signalling, apoptosis induction and enhancement of chemo- and radiotherapy responses. In contrast, the clinical literature was limited to a small number of early-phase or non-randomized studies, together with one randomized phase II trial. These studies generally supported the safety and feasibility of high-dose oral MBZ, but provided only modest and context-dependent signals of efficacy.

3.3 | Risk of bias assessment

In vitro studies frequently lacked sample size justification, randomization, blinding, cell line authentication and adequate reporting, leading to many 'Not reported/Inadequate' judgements (Supplementary 2 Figure 1). *In vivo* studies showed similar deficiencies, with incomplete outcome data often unaddressed (Supplementary 2 Figure 2). In the single randomized trial,[49] risk of bias was 'Low' for OS but 'Some concerns' for PFS due to the open-label design and non-uniform imaging (Supplementary 2 Figure 3). Non-randomized studies[47,48,50,51] were commonly at 'Serious' or 'Critical' risk of bias from confounding and participant selection, though outcome measurement and missing data was generally 'Low' (Supplementary 2 Figure 4). Overall, reporting limitations were pervasive in preclinical work, while in clinical studies the main vulnerabilities related to progression outcomes rather than OS.

3.4 | Mechanisms of Mebendazole's anticancer activity

MBZ acts through multiple cellular mechanisms. Its best-established effect is microtubule depolymerization, causing G2/M arrest and apoptosis via **caspase** activation, **PARP** cleavage and altered **Bcl-2** signalling.[35,38,46] It also inhibits several kinases, most potently **MAPK14** (IC_{50} 0.10 ± 0.05 μM), as well as **ABL1**, **ERK2**, **VEGFR-2** and **PDGFRα**, disrupting survival signalling and angiogenesis.[33] Anti-angiogenic effects are supported by reduced microvessel density and selective VEGFR-2 suppression in tumour vasculature.[40] MBZ modulates transcriptional pathways, notably inhibiting Hedgehog signalling through GLI1/PTCH1 downregulation and suppression of primary cilium formation, with activity against **vismodegib**-resistant **SMO** mutants.[40,42] In GBM and DMG, it activates p53/p21/p27 regulators and induces apoptosis and pyroptosis through mitochondrial dysfunction, **NF-κB** activation and **NLRP3** inflammasome signalling.[35] Finally, MBZ interferes with DNA damage repair by blocking nuclear import of **Chk2** and Nbs1 at nanomolar concentrations, prolonging γH2AX signalling and enhancing radiosensitivity.[31]

3.5 | Cancer types and patterns of sensitivity to Mebendazole

MBZ exhibits broad anticancer activity across GBM, MB, meningioma and other brain tumour types (Table 1), with IC_{50} values generally in the low nanomolar to sub-micromolar range. In GBM models (U87, U251, T98G, GL261), MBZ reduced proliferation and migration with IC_{50} values of 0.2–0.6 μM,[33–35,39] often more selective for tumour over normal astrocytes. MB lines (D425Med, UW228) were similarly sensitive (0.22–0.5 μM).[40,41] DMG responded at <1 μM in most lines tested, including ONC201-resistant variants.[38,46] Canine gliomas mirrored this sensitivity, with IC_{50} values as low as 30 nM, and MBZ demonstrated selectivity over primary fibroblasts, supporting cross-species therapeutic relevance.[30,36] Meningioma cell lines were uniformly sensitive (0.26–0.42 μM).[44] Taken together, MBZ displays consistent cytotoxicity across multiple CNS cancers, with variations in sensitivity mapped partly to tumour subtype and resistance background.

3.6 | Effects of Mebendazole on tumour progression and survival in animal models

Across preclinical models, MBZ reproducibly suppressed tumour growth and extended survival. In syngeneic GL261 gliomas, oral MBZ (50 mg/kg/day) extended mean survival by ~63% and prolonged survival in MB xenografts by over 100%.[39,40] The most potent polymorph, MBZ-C, achieved therapeutic brain levels exceeding tumour IC_{50} by >20-fold and nearly doubled survival. Combination with the efflux inhibitor **elacridar** extended lifespan four-fold in mice.[41] In the orthotopic GL261 glioma model, MBZ polymorph C significantly prolonged survival, whereas **vincristine** did not improve survival

TABLE 1 Summary of included studies on mebendazole treatment across cancer types categorized by country and year of publication.

		Animal		Human					Population/clinical trial				
Author (year)	Country	GBM	Astrocytoma	GBM	DMG	MB	Meningioma	ODG	GBM	DMG	ODG	Astrocytoma	MB
Kipper et al. (2018)[32]	Brazil	×	×	✓	×	×	×	×	×	×	×	×	×
Ren et al. (2022)[35]	China	×	×	✓	×	×	×	×	×	×	×	×	×
Ariey-Bonnet et al. (2020)[33]	France	×	×	✓	×	×	×	×	×	×	×	×	×
Gentile et al. (2025)[38]	Italy	×	×	×	✓	×	×	×	×	×	×	×	×
Patil et al. (2020)[47]	India	×	×	×	×	×	×	×	✓	×	✓	✓	×
Patil et al. (2022)[49]	India	×	×	×	×	×	×	×	✓	×	×	×	×
Mena-Hernandez et al. (2020)[45]	Mexico	✓	×	×	×	×	×	×	×	×	×	×	×
Jo et al. (2022)[34]	Republic of Korea	×	×	✓	×	×	×	×	×	×	×	×	×
Bai et al. (2011)[39]	USA	✓	×	✓	×	×	×	×	×	×	×	×	×
Bai et al. (2015)[40]	USA	×	×	×	×	✓	×	×	×	×	×	×	×
Bai et al. (2015)[41]	USA	✓	×	×	×	✓	×	×	×	×	×	×	×
Larsen et al. (2015)[42]	USA	✓	×	×	×	✓	×	×	×	×	×	×	×
De Witt et al. (2017)[43]	USA	✓	×	×	×	×	×	×	×	×	×	×	×
Lai et al. (2017)[30]	USA	✓	✓	×	×	×	×	×	×	×	×	×	×
Markowitz et al. (2017)[31]	USA	✓	×	✓	×	×	×	×	×	×	×	×	×
Skibinski et al. (2018)[44]	USA	×	×	×	×	×	✓	×	×	×	×	×	×
Gallia et al. (2021)[48]	USA	×	×	×	×	×	×	×	✓	×	×	✓	×
Wright et al. (2022)[36]	USA	✓	×	×	×	×	×	×	×	×	×	×	×
Krystal et al. (2024)[50]	USA	×	×	×	×	×	×	×	×	✓	×	✓	×
Patel et al. (2024)[37]	USA	×	×	✓	×	×	×	×	×	×	×	×	×
Yamashita et al. (2025)[46]	USA	×	×	✓	✓	×	×	✓	×	×	×	×	×
Phan et al. (2025)[51]	USA	×	×	×	×	×	×	×	✓	✓	×	✓	✓

Abbreviations: GBM, Glioblastoma; DMG, Diffuse Midline Glioma; MB, Medulloblastoma; ODG, Oligodendroglioma; USA, United States of America.

despite greater *in vitro* potency. Vincristine was also associated with significant peripheral neuropathy, which was not observed with MBZ.[43] In malignant meningioma xenografts, MBZ extended survival by 58%, with further gains when combined with local radiation (+105%) when compared to control.[44] Recent IDH1-mutant glioma studies showed MBZ alone increased median survival by 87%, while MBZ plus radiotherapy produced long-term tumour-free survivors (>50%), a benefit not seen with radiation alone.[46] When treated with intranasal MBZ microemulsions, all rats survived to study endpoint compared to rapid decline in controls, suggesting alternative formulations may further enhance delivery and tolerability.[45]

3.7 | Interactions between Mebendazole and standard cancer therapies

MBZ frequently acts as a chemosensitizer and radiosensitizer. In GBM, MBZ enhanced the efficacy of TMZ, reducing cell viability in resistant and sensitive models, and synergized with **vinblastine** to suppress long-term tumour proliferation.[32,39] Triple-therapy with TMZ + MBZ + **chloroquine** produced the greatest cytotoxic effect *in vitro*, overcoming TMZ resistance.[34] In vismodegib-resistant MB, MBZ restored Hedgehog pathway suppression and extended survival by 150%.[40,42] MBZ sensitized glioma cells to ionizing radiation by impairing DNA repair protein trafficking, matching its radiosensitization EC_{50} (~25–35 nM),[31] with additive survival benefits when combined with radiotherapy in both meningioma and IDH1-mutant glioma.[44,46] In veterinary models, MBZ combined with mistletoe extract enhanced cytotoxicity beyond either agent alone.[36] These findings highlight MBZ's synergistic profile, enhancing chemotherapy, autophagy inhibitors and radiotherapy.

3.8 | Mebendazole and the hallmarks of cancer

To integrate mechanistic findings across diverse experimental models, MBZ-associated effects were mapped onto the Hallmarks of Cancer framework (Table 2).[52] Across 17 mechanistic studies, MBZ most

TABLE 2 Mapping of *in vitro* and *in vivo* studies onto the hallmarks of cancer framework, indicating the number and proportion of studies associated with each hallmark (studies may overlap across multiple hallmarks).

Hallmarks of cancer	Main molecular target/representative mechanistic link	References	*N* (%)
Sustaining Proliferative Signalling	Tubulin/microtubule depolymerization; MAPK14 inhibition; Hedgehog pathway suppression (SMO-GLI1/PTCH1)	Kipper,[32] Ren,[35] Ariey-Bonnet,[33] Gentile,[38] Jo,[34] Bai,[39] Bai,[40] Bai,[41] Larsen,[42] De Witt,[43] Lai,[30] Skibinski,[44] Patel,[37] Yamashita[46]	**14 (82.4)**
Resisting Cell Death	Caspase-3/PARP cleavage; Bcl-2 family modulation; NF-κB/NLRP3/GSDMD-mediated pyroptotic signalling	Ren,[35] Gentile,[38] Jo,[34] Skibinski,[44] Wright,[36] Patel,[37] Yamashita[46]	7 (41.2)
Activating Invasion and Metastasis	EMT-related markers: E-cadherin, N-cadherin, β-catenin, MMP2, Slug	Ren,[35] Jo,[34] Patel[37]	3 (17.6)
Enabling Replicative Immortality	Reduced long-term proliferation, clonogenic growth and neurosphere/spheroid formation	Kipper,[32] Patel,[37] Yamashita[46]	3 (17.6)
Evading Growth Suppressors	p53/p21/p27 axis	Ren,[35] Gentile[38]	2 (11.8)
Deregulating Cellular Metabolism	Mitochondrial membrane potential disruption	Ren,[35] Jo[34]	2 (11.8)
Genome Instability and Mutation	Impaired DNA damage response via Chk2 and Nbs1 nuclear trafficking; sustained γH2AX signalling	Markowitz,[31] Yamashita[46]	2 (11.8)
Inducing or Accessing Vasculature	VEGFR2 blockade; reduced CD31-positive microvessel density; PDGFRα modulation	Bai,[40] Skibinski[44]	2 (11.8)
Unlocking Phenotypic Plasticity	Hedgehog pathway suppression via SMO/primary cilium inhibition with downstream GLI1/PTCH1 repression	Larsen[42]	1 (5.9)
Avoiding Immune Destruction	–	–	0 (0)
Non-mutational Epigenetic Reprogramming	–	–	0 (0)

Bcl-2, B-cell lymphoma 2; **CD31**, cluster of differentiation 31; **Chk2**, checkpoint kinase 2; **EMT**, epithelial-mesenchymal transition; **GSDMD**, gasdermin D; **MAPK14**, mitogen-activated protein kinase 14; **MMP2**, matrix metalloproteinase 2; **Nbs1**, Nijmegen breakage syndrome 1; **NF-κB**, nuclear factor kappa B; **NLRP3**, NLR family pyrin domain containing 3; **PARP**, poly (ADP-ribose) polymerase; **PDGFRα**, platelet-derived growth factor receptor alpha; **PTCH1**, patched 1; **SMO**, smoothened; **VEGFR2**, vascular endothelial growth factor receptor 2; **γH2AX**, phosphorylated histone H2AX.

consistently targeted pathways related to sustaining proliferative signalling (82.4%), primarily through microtubule depolymerization, MAPK14 inhibition and Hedgehog pathway suppression. Evidence for resisting cell death was also substantial (41.2%), with studies reporting caspase-3/PARP cleavage, modulation of Bcl-2 family proteins and pyroptotic signalling. Smaller proportions of studies linked MBZ to activating invasion and metastasis (17.6%) and enabling replicative immortality (17.6%), including effects on EMT-related markers, clonogenicity and neurosphere or spheroid formation. More limited evidence implicated MBZ in evading growth suppressors, deregulating cellular metabolism, genome instability and mutation and inducing or accessing vasculature (each 11.8%), while only one study supported a role in unlocking phenotypic plasticity (5.9%). No studies addressed avoiding immune destruction or non-mutational epigenetic reprogramming. Overall, this mapping highlights that the preclinical evidence for MBZ is concentrated on proliferation and cell death pathways, while several hallmark domains remain comparatively underexplored.

3.9 | Clinical evidence for Mebendazole in cancer treatment

Clinical evidence for MBZ in brain tumours remains limited and is currently more informative regarding safety and feasibility than efficacy. Early-phase studies in adults and paediatric patients indicate that high-dose oral MBZ is generally tolerable, whether administered alone or in combination with standard therapies, with the main reported toxicities including reversible liver enzyme elevation and expected haematological adverse events associated with partner regimens.[47,48,50] However, evidence of antitumour efficacy in humans is modest and inconsistent. In adult gliomas, MBZ combined with TMZ was feasible and associated with a median OS of 21 months in a single-arm phase I study, although the absence of a comparator limits interpretation.[48] In recurrent high-grade glioma, phase I dose-escalation work established recommended doses for MBZ when combined with TMZ or **lomustine**, but survival outcomes remained limited.[47] Most notably, in the randomized phase II trial, the addition of MBZ to lomustine or TMZ did not meet the predefined efficacy target overall and did not improve median survival in the full study population.[49] A possible signal of benefit was observed only in a post hoc subgroup of patients with good performance status receiving lomustine plus MBZ.[49]

Paediatric data similarly support tolerability more strongly than efficacy. In children and young adults with HGG or DMG, MBZ combined with bevacizumab and irinotecan was well tolerated, and occasional radiographic responses were observed. However, PFS remained short, and all evaluable patients ultimately progressed.[50] Likewise, in paediatric patients receiving MBZ monotherapy for refractory or recurrent brain tumours, the drug was well tolerated, but no objective responses were observed, and PFS was limited.[51] Taken together, current clinical studies suggest that MBZ is safe and feasible at high doses, but do not establish meaningful single-agent efficacy and provide only tentative support for a role in selected combination settings.

4 | DISCUSSION

This systematic review identified a substantial preclinical literature supporting MBZ as a biologically plausible repurposing candidate in neuro-oncology, but only limited clinical evidence regarding its effectiveness in patients. Across *in vitro* and *in vivo* models, MBZ demonstrated multimodal anticancer actions, including cytoskeletal disruption,[34,35,38,39,43,46] angiogenesis inhibition,[40,44] kinase modulation,[33] Hedgehog pathway blockade[42] and interference with DNA damage repair (Tables 3 and 4).[31] These mechanisms map onto several Hallmarks of Cancer and help to explain the breadth of activity observed across tumour types and resistant disease contexts (Table 2). However, although early-phase clinical studies generally support the safety and feasibility of high-dose oral MBZ, evidence of meaningful efficacy in humans remains modest, inconsistent and not yet established. Accordingly, MBZ should presently be viewed as a promising but unproven translational candidate rather than a clinically validated therapy for brain tumours.

4.1 | Mechanistic targets and pathway modulation

MBZ exerts multifaceted anticancer effects through both canonical and non-canonical pathways. Its best-established activity is microtubule disruption, where binding to tubulin leads to depolymerization, G2/M cell cycle arrest and caspase-dependent apoptosis in multiple brain tumour models.[34,35,38,39,43,46] This mechanism is reminiscent of vinca alkaloids such as vincristine and vinblastine,[53] as well as **colchicine**,[54] which similarly destabilize microtubules and block the cell cycle at the G2/M phase to trigger apoptosis. However, MBZ appears to achieve these effects at sub- to low-micromolar concentrations while showing greater selectivity for tumour cells over normal counterparts.[39] Unlike classical microtubule inhibitors, MBZ has demonstrated more effective BBB penetration and a favourable neurotoxicity profile in preclinical models, which may provide an advantage in the treatment of brain tumours.[39,43] This distinction was highlighted by De Witt et al.[43] who directly compared MBZ polymorph C with vincristine in an orthotopic GL261 glioma model. Although vincristine was more potent *in vitro*, MBZ produced a significant survival benefit in vivo, whereas vincristine, even at doses approaching its maximum tolerated dose, did not improve survival. Importantly, vincristine caused significant peripheral neuropathy, while MBZ showed only a non-significant trend towards increased footpad sensitivity.[43] These findings suggest that, in neuro-oncology, therapeutic usefulness may depend less on nominal *in vitro* potency alone than on the capacity to achieve effective intracranial exposure without dose-limiting off-target toxicity. More broadly, because the BBB restricts therapeutic delivery and the blood-tumour barrier (BTB) is highly heterogeneous,

TABLE 3 Summary of *in vitro* studies investigating the anticancer properties of mebendazole organized by year of publication.

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
Lai et al. (2017),[30] United States of America	• To evaluate the *in vitro* chemosensitivity of three canine glioma cell lines to MBZ and fenbendazole. • To determine the effects of MBZ and fenbendazole on cell viability and tubulin structure in canine glioma cells. • To assess whether these drugs have selective cytotoxicity for tumour cells compared to normal fibroblasts.	**Cell lines used:** • Canine GBM: • G06-A • SDT-3G • Canine anaplastic astrocytoma: • J3T • Primary canine fibroblasts	• MTT cell viability assay • Immunofluorescence microscopy • Western blot	**IC_{50}:** • MBZ (after 72 hrs): • J3T = 0.03 ± 0.003 μM • G06-A = 0.08 ± 0.015 μM • SDT-3G = 0.03 ± 0.006 μM • Fenbendazole (after 72 hrs): • J3T: 0.550 ± 0.015 μM • G06-A: 1.530 ± 0.159 μM • SDT-3G: 0.690 ± 0.095 μM	• MBZ and fenbendazole are highly cytotoxic to canine glioma cell lines at low micromolar/submicromolar concentrations; MBZ demonstrated greater potency than fenbendazole. • Primary canine fibroblasts exposed to similar IC_{50} concentrations did not show significant cell death, suggesting selectivity towards tumour cells. • IF showed significant morphological changes in tubulin architecture in all three glioma cell lines following treatment with MBZ and fenbendazole (clumping, peripheralization, cell rounding), indicating anti-tubulin activity. • Western blot confirmed that the anti-tubulin antibody used was specific to canine α-tubulin. • MBZ and fenbendazole have high safety margins and could potentially be repurposed for veterinary neuro-oncology.
Markowitz et al. (2017),[31] United States of America	• To test whether the microtubule-targeting agent MBZ can radiosensitize glioma cells by acting during interphase (not just mitosis). • To elucidate the mechanism of MBZ-induced radiosensitization, specifically its effect on DNA damage response protein trafficking.	**Cell lines used:** • Patient derived GBM: • GBM14 • Mouse GBM: • GL261	• WST-1 cell viability assay • Immunofluorescence microscopy (MPM2) • Western blot • Cytoplasmic and nuclear fractionation	• **Potency (EC_{50}/DEF_{50}):** • Radiosensitization (DEF_{50}) = 35 nM (95% CI: 9–50 nM) • Inhibition of nuclear trafficking of Chk2 = 31 nM (95% CI: 17–45 nM) • Inhibition of nuclear trafficking of Nbs1 = 25 nM (95% CI: 18–32 nM) • Induction of mitotic arrest = 192 nM (95% CI: 127–257 nM)	• MBZ sensitizes glioma cells to ionizing radiation even when only interphase cells are present. • Radiosensitization is maximal when MBZ is applied post-IR, during interphase. • MBZ prolongs the DNA damage response (increases/sustains γH2AX post-IR) by delaying DNA repair. • MBZ disrupts nuclear import of key DDR proteins (Chk2, Nbs1), causing their cytoplasmic retention at low nanomolar concentrations. • The EC_{50} for radiosensitization matches the EC_{50} for DDR protein mis-localization, both much lower than the concentration needed to induce mitotic arrest. • Supports a mechanism in which MBZ radiosensitises by inhibiting interphase DNA repair, independently of mitotic arrest.
Kipper et al. (2018),[32] Brazil	• To identify molecular predictors of TMZ resistance in GBM and test whether combination therapies using vinblastine and MBZ can potentiate the effects of TMZ, particularly in resistant glioma models.	**Cell lines used:** • Human GBM: • U87 • A172 • U251 • U138 • U343 • Patient derived: • GBM, Grade IV	• TCGA bioinformatic analyses • Acute drug sensitivity assay • Long-term proliferation assay (cumulative population doubling) • Flow cytometry (PI staining)	**IC_{50}:** • Not described	• MBZ, when combined with TMZ (with or without vinblastine), significantly enhanced the reduction of cell numbers in most GBM and glioma cell cultures, including some that were resistant to TMZ alone. • The combination of TMZ, vinblastine and MBZ provided the strongest suppression of cell growth and long-term proliferation in both established GBM cell lines and patient-derived glioma cultures. • MBZ was especially effective in cases where low expression of FGFR3 and AKT2 genes predicted poor

TABLE 3 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
		• Anaplastic astrocytoma, Grade III • Fibrillar astrocytoma, Grade II • Gemistocytic astrocytoma, Grade II • Murine GBM: • C6	• Nuclear morphometric analysis • qPCR		response to TMZ. The triple combination induced cell cycle arrest, polyploidy, and features of senescence in patient-derived glioma cultures.
Ariey-Bonnet et al. (2020),[33] France	• To investigate the mechanism of action of MBZ repurposed for treatment of brain tumours. • Use in silico molecular target prediction followed by experimental (*in vitro*) validation, with a focus on GBM.	**Cell lines used:** • Human GBM: • U87 • U87vIII • T98G • U251	• Alamar Blue cell viability assay • *In vitro* kinase assays • RNA-seq (TCGA/ GTEx) and qRT-PCR • Thermal shift assay • Isothermal titration calorimetry • Differential scanning fluorimetry • NanoBRET target engagement assay • Molecular modelling/ docking • siRNA knockdown	**IC_{50}:** • U251 = 288 ± 3 nM • T98G = 2.1 ± 0.6 μM • Kinase inhibition: • MAPK14: 0.10 ± 0.05 μM • ABL1: 0.35 ± 0.12 μM • ERK2: 24.7 ± 10.4 μM • NanoBRET in-cell assay (MAPK14 inhibition in U87 cells): IC_{50} = 4.1 ± 1.1 μM • ITC (MBZ-MAPK14 binding): Kd = 1.27 ± 0.02 μM	• MBZ strongly inhibits viability of all tested GBM cell lines *in vitro*. • In silico prediction identified 21 putative molecular targets for MBZ, including 12 significantly upregulated in GBM. • Experimental validation showed direct kinase inhibition by MBZ, with strongest effect against MAPK14 (p38α MAPK), and also on ABL1 and ERK2, though less potently. • MBZ binds directly to MAPK14, confirmed by several biophysical assays. • Gene silencing of MAPK14 in GBM spheroids decreased tumour growth and reduced sensitivity to MBZ, confirming MAPK14 as an important mediator of MBZ's anticancer effect.
Jo et al. (2022),[34] Republic of Korea	• To investigate the potential of MBZ, a microtubule depolymerizing drug, as a repositioned agent for GBM treatment. • To explore strategies to improve MBZ efficacy using drug combinations, specifically with autophagy inhibition (via chloroquine) and with the standard agent TMZ.	**Cell lines used:** • Human GBM: • U87-MG • U373 • T98G • LN18 • TMZ-resistant U87-MG and U373 cells were generated by escalating TMZ from 25 μM to 800 μM.	• MTS cell viability assay • Boyden chamber migration assay • Flow cytometry (PI staining) • DAPI staining • Western blot	**IC_{50}:** • Proliferation = 0.22–0.65 μM • Migration = 0.88–2.25 μM)	• MBZ significantly inhibits proliferation and migration of GBM cell lines. • MBZ induces G2-M cell cycle arrest and apoptosis in GBM cells. • MBZ triggers autophagy in these cells, without upregulating Beclin 1, ATG5 or ATG7. • Combining MBZ with chloroquine enhances MBZ's anti-proliferative effects. • Triple therapy (TMZ + MBZ + chloroquine) is most effective at suppressing GBM cell growth. • MBZ (alone or with chloroquine) also effectively inhibits the proliferation of TMZ-resistant GBM cells, suggesting potential to overcome resistance.
Ren et al. (2022),[35] China	• To identify GBM hub genes and repurpose existing drugs with potential anti GBM activity using weighted gene co expression network analysis (WGCNA) and the Connectivity Map (CMAP) platform.	**Cell lines used:** • Human GBM • U-87 MG • U-251 MG	• TCGA/WGCNA bioinformatic analyses • Cell proliferation assay (CCK-8) • DNA synthesis assay	**IC_{50}:** • U 87 MG (24 hrs): • MBZ = 0.207 μM • Flubendazole = 0.170 μM • Fenbendazole = 0.182 μM • U 251 MG (24 hrs):	• Potently and dose-dependently inhibited proliferation of U-87 and U-251 cells • Suppressed DNA synthesis, migration and invasion *in vitro*. • Altered EMT marker expression • upregulated E-cadherin

(Continues)

TABLE 3 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
	• To test the anti-GBM activity of azole compounds and determine their mechanisms of action.		• Transwell migration and invasion assays • Flow cytometry (PI staining) • Mitochondrial membrane potential assay (JC-1) • Annexin V/PI staining • LDH release assay • Western blot • Caspase inhibition assay (Z-VAD-FMK)	• MBZ = 0.253 μM • Flubendazole = 0.224 μM • Fenbendazole = 0.226 μM	• downregulated N-cadherin, β-catenin, MMP2, Slug. • Arrested cell cycle at G2/M via P53/P21/Cyclin B1 signalling. • Reduced mitochondrial membrane potential, altered Bcl-2 family protein expression, induced caspase and PARP cleavage-indicating mitochondrial-dependent apoptosis. • Induced pyroptosis via NF-κB/NLRP3/GSDMD pathway (increased cell swelling/LDH release/ nuclear translocation of NF-κB). • Cytotoxic/apoptotic/pyroptotic effects prevented by pan-caspase inhibitor Z-VAD-FMK, showing caspase dependence.
Wright et al. (2022),[36] United States of America	• To evaluate the cytotoxicity of European mistletoe (*Viscum album*) extract against canine high-grade astrocytoma cells *in vitro*. • To assess the additive effect of mistletoe extract in combination with MBZ at its IC_{50} concentration.	**Cell lines used:** • Canine astrocytoma: • SDT 3G (grade IV astrocytoma	• MTT cell viability assay • IC_{50} determination (non-linear regression)	**IC_{50}:** • MBZ = 0.03 μM • Mistletoe extract = 5.644 ± 0.09 μg/mL	• Mistletoe extract alone caused dose-dependent cytotoxicity in SDT 3G cells with low experimental variability, repeatable across different passages. • MBZ alone at IC_{50} reduced cell viability to ∼55.5% of control (≈50% cell death). • MBZ + high-dose mistletoe (μg/mL): • Further 39.3% greater cell death *vs*. MBZ alone ($p = 0.0034$) • 40% greater cell death *vs*. mistletoe alone at same dose ($p = 0.0008$) • MBZ + low-dose mistletoe (2.5 ng/mL) • no additional effect beyond MBZ alone ($p = 0.98$) • Findings support further investigation of mistletoe, alone or as an adjunct to MBZ
Patel et al. (2024),[37] United States of America	• To develop, optimize and characterize sterile, injectable nanosuspensions of MBZ coated with albumin and polysorbate-80 for potential use in GBM therapy. • To assess *in vitro* cytotoxicity, colony-forming efficiency, cell migration, 3D spheroid activity and sterility of the MBZ nanosuspension.	**Cell lines used:** • Human GBM: • U-87 MG • LN-229	• Particle size, polydispersity index, and zeta potential measurements • Differential scanning calorimetry • X-ray diffraction • Transmission electron microscopy • MTT cell viability assay • Clonogenic assay • Cell migration assay • 3D tumour spheroid assay	**IC_{50}:** • U-87 MG • MBZ = 4.7 ± 0.09 μM • MBZ nanosuspension = 0.49 ± 0.02 μM • LN-229 • MBZ = 0.66 ± 0.15 μM • MBZ nanosuspension = 0.48 ± 0.05 μM	• Optimized MBZ nanosuspension with albumin and polysorbate-80 had a mean particle size of ∼208 nm, PDI 0.21 and zeta potential −20.4 mV. • MBZ nanosuspension showed much greater *in vitro* cytotoxicity against GBM cell lines than plain MBZ. • MBZ nanosuspension achieved a 2.65-fold greater inhibition of colony formation and 1.16-fold higher inhibition of cell migration (compared to MBZ) *in vitro*. • In 3D GBM spheroid models, MBZ NS reduced tumour spheroid growth by 50% and increased apoptosis *vs*. control. • Results support MBZ NS as a promising, sterile, injectable formulation for potential GBM therapy.

TABLE 3 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
			• Live/dead or apoptosis staining in 3D spheroids • Sterility testing		
Gentile et al. (2025),[38] Italy	• To evaluate the anticancer effects and mechanisms of repurposed MBZ, alone and in combination with the investigational anticancer compound ONC201, in DMG cell lines.	**Cell lines used:** • Human DMG: • SU-DIPG-XIII-FL • SU-DIPG-XIII-P* • SU-DIPG-VI • SU-DIPG-XLVIII • SU-DIPG-XXXVIII • SF8628 • SF7761 • ONC201-resistant human DMG cell line: • SU-DIPG-XIII-R	• CellTiter-Glo cell viability assay • Flow cytometry (cell cycle analysis) • Flow cytometry (Annexin V/PI staining) • Western blot • Checkerboard drug combination assay • Synergy analysis (CompuSyn/ CalcuSyn; SynergyFinder)	**IC_{50}:** • SF7761 = 102 nM (95% CI: 72–149 nM) • SF8628 = 258 nM (95% CI: 202–344 nM) • SU-DIPG-VI = not reached • SU-DIPG-XIII = 330 nM (95% CI: 236–495 nM) • SU-DIPG-XIII-P* = 162 nM (95% CI: 93–302 nM) • SU-DIPG-XXXVIII = 321 nM (95% CI: 221–516 nM) • SU-DIPG-XLVIII = 958 nM (95% CI: 637–2519 nM)	• MBZ reduced viability in 6/7 DMG cell lines, with IC_{50} within clinically achievable levels. • Induced G2/M arrest (via p21, p27, ±p53) and triggered apoptosis (cleaved caspase-3, PARP, Annexin V/PI). • MBZ + ONC201 showed additive/synergistic effects, enhancing apoptosis beyond either agent alone. • MBZ remained effective in ONC201-resistant lines. • Activity and synergy were independent of H3K27 and largely independent of TP53 status. • MBZ is a promising low-toxicity candidate for front-line or salvage therapy in DMG, including in combination with ONC201.

ABL1, ABL proto-oncogene 1, non-receptor tyrosine kinase; **A172**, human glioblastoma cell line; **AKT2**, AKT serine/threonine kinase 2; **Annexin V/PI**, annexin V/propidium iodide apoptosis assay; **ATG5/7**, autophagy-related proteins 5/7; **BBB**, blood–brain barrier; **BrdU**, 5-bromo-2′-deoxyuridine; **C6**, rat glioma cell line; **CCK-8**, Cell Counting Kit-8; **CI**, confidence interval; **CMAP**, Connectivity Map; **DAPI**, 4′,6-diamidino-2-phenylindole; **DEF50**, dose enhancement factor at 50% effect; **DMG**, diffuse midline glioma; **DSC**, differential scanning calorimetry; **EC50**, half maximal effective concentration; **EMT**, epithelial–mesenchymal transition; **ERK2**, extracellular signal-regulated kinase 2 (MAPK1); **FGFR3**, fibroblast growth factor receptor 3; **GBM**, glioblastoma; **GBM14**, patient-derived glioblastoma cell line; **GL261**, murine glioma cell line; **GTEx**, Genotype-Tissue Expression; **H3K27**, histone H3 lysine 27; **IC_{50}**, half maximal inhibitory concentration; **IF**, immunofluorescence; **IR**, ionizing radiation; **ITC**, isothermal titration calorimetry; **JC-1**, mitochondrial membrane potential dye; **Kd**, dissociation constant; **LDH**, lactate dehydrogenase; **LN-229**, human glioblastoma cell line; **LN18**, human glioblastoma cell line; **MAPK14**, mitogen-activated protein kinase 14 (p38α); **MBZ**, mebendazole; **MMP2**, matrix metalloproteinase 2; **MPM2**, mitosis-specific phospho-epitope antibody; **MTS**, tetrazolium-based cell viability assay; **MTT**, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide; **NanoBRET**, bioluminescence resonance energy transfer target engagement assay; **Nbs1**, nibrin (NBN); **NF-κB**, nuclear factor kappa B; **NLRP3**, NLR family pyrin domain containing 3; **ONC201**, dordaviprone; **PARP**, poly (ADP-ribose) polymerase; **PDI**, polydispersity index; **PI**, propidium iodide; **qPCR**, quantitative polymerase chain reaction; **qRT-PCR**, quantitative reverse transcription PCR; **RNA-seq**, RNA sequencing; **SDT-3G**, canine glioma cell line; **SF7761/SF8628**, human DMG cell lines; **siRNA**, small interfering RNA; **SU-DIPG**, diffuse intrinsic pontine glioma cell line series; **T98G**, human glioblastoma cell line; **TCGA**, The Cancer Genome Atlas; **TEM**, transmission electron microscopy; **TMZ**, temozolomide; **TP53**, tumour protein p53; **U-87 MG/U251/U373/U87-MG/U87vIII/U138/U343**, human glioma cell lines; **WGCNA**, weighted gene co-expression network analysis; **WST-1**, tetrazolium-based cell viability assay; **Z-VAD-FMK**, pan-caspase inhibitor (carbobenzoxy-valyl-alanyl-aspartyl-[O-methyl]-fluoromethylketone).

TABLE 4 Summary of *in vivo* tumour model studies of mebendazole (including associated *in vitro* experiments reported within the same studies), organized by year of publication.

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
Bai et al. (2011),[39] United States of America	• To investigate the anticancer potential of benzimidazole antihelminthics, particularly MBZ, in GBM using *in vitro* GBM models and *in vivo* mouse models, as well as to explore potential mechanisms of action.	**Cell lines used:** • Human GBM: U87-MG, D54, H80, H247, H392, H397, H502, H566 • Human GBM neurosphere line: 060919 • Mouse GBM: GL261 • Mouse nontumor astrocytes (from C57BL/6 brain) **Animal model:** • Female C57BL/6 mice implanted intracranially with 40 000 GL261 cells • Human GBM xenograft: Female athymic nude mice implanted intracranially with 150 000 060919 cells	• Cell viability assay (Cell Counting Kit-8; WST-8) • Western blot (tubulin polymerization) • Immunofluorescence microscopy • In vivo survival analysis (Kaplan–Meier) • In vivo bioluminescence imaging (tumour growth) • Combination efficacy testing (MBZ + TMZ)	**IC_{50}:** • GL261 = 0.24 μM • 060919 = 0.1 μM • Mouse astrocytes = 0.4 μM **Animal model:** • MBZ 100 mg/kg/day showed toxicity; 50 mg/kg/day (oral) was well tolerated and used for efficacy studies. **Animal Survival (mean):** • GL261: MBZ increased mean survival from 30 (control) to 49 days (∼63% increase) • 060919 xenograft: MBZ increased mean survival from 48 (control) to 65 days • Combination (GL261 model): MBZ + TMZ 50 days; TMZ alone: 41 days; control: 29–30 days.	• MBZ showed low micromolar cytotoxicity in human and mouse GBM cell lines, with MBZ more selective for tumour cells over normal astrocytes. • MBZ disrupted microtubule structure, similar to colchicine. • Oral MBZ (50 mg/kg) significantly prolonged survival in both syngeneic (GL261) and xenograft (060919) GBM models. • Effective in both TMZ-sensitive and -resistant tumours. • MBZ + TMZ provided additional survival benefit, though not significantly better than MBZ alone. • Primary action via microtubule depolymerization; other effects (e.g. Bcl-2 inhibition, anti-angiogenesis) may contribute.
Bai et al. (2015)[40] United States of America	• To evaluate the efficacy of MBZ against various subtypes of MB, including tumours with acquired resistance to the SMO inhibitor vismodegib. To characterize MBZ's impact on VEGFR2 kinase and tumour angiogenesis.	**Cell lines used:** • Human MB: D425Med (group 3; c-MYC and OTX2 amplified), UW228 • Human umbilical vein endothelial cells (HUVECs) for angiogenesis studies **Animal model:** • PTCH+/−, p53−/− mouse-derived spontaneous MB allograft (SHH group) • Vismodegib-resistant SMO-D477G mutant allograft (acquired resistance model) • Group 3 human MB D425Med orthotopic xenograft (nude mice)	• Cell-free kinase inhibition assay (VEGFR2; ADP-Glo) • Western blot (VEGFR2 phosphorylation) • Immunohistochemistry and immunofluorescence microscopy (VEGFR2, CD31, VEGF) • Microvessel density quantification (CD31) • In vivo bioluminescence imaging (tumour growth)	**IC_{50}:** • D425Med = 0.22 μM • UW228 = 0.50 μM • VEGFR2 kinase inhibition = 4.3 μM **Animal model:** • MBZ 50 mg/kg/day (oral gavage, maximum tolerated, based on prior studies) • Median Survival: ◦ PTCH+/−, p53−/− allograft (SHH group) MBZ improved median survival by 150% vs control	• MBZ potently inhibited VEGFR2 activity *in vitro* by competing with ATP binding. • MBZ inhibited VEGFR2 autophosphorylation in HUVECs and blocked VEGFR2 phosphorylation in tumour tissue in vivo. • MBZ selectively impaired tumour angiogenesis (reduced microvessel density in tumours), with no discernible effect on normal brain vasculature. • MBZ significantly prolonged survival in orthotopic MB models, as well as in a vismodegib-resistant (SMO mutant) sonic hedgehog model.

TABLE 4 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
			• In vivo survival analysis (Kaplan–Meier) • Western blot (tubulin polymerization) • Phosphorylation assay (PDGFRα; NIH3T3)	◦ SMO-D477G vismodegib-resistant allograft median survival extended from 12 days (control) to 30 days (MBZ; +150%) • D425Med xenograft (group 3) median survival from 21 (control) to 48 days (+129%)	• MBZ reduced tumour burden in vivo as measured by bioluminescence imaging. • Tubulin disruption by MBZ was cell-line dependent: evident in UW228, but not D425 at 1 μM. • MBZ also inhibited PDGFRα phosphorylation in NIH3T3 cells, suggesting broader tyrosine kinase inhibitory potential.
Bai et al. (2015)[41] United States of America	• To compare the brain penetration, pharmacokinetics, efficacy and toxicity of the three MBZ polymorphs (A, B, C), especially in the context of brain cancer therapy. • To determine whether specific MBZ polymorphs, alone or combined with the P-glycoprotein/BCRP inhibitor elacridar, can improve antitumor efficacy and brain tissue bioavailability. • To measure MBZ and its main metabolites in plasma, brain, and brain tumour tissue.	**Cell lines used:** • Mouse GBM: GL261 (with luciferase, 'GL261-luc') • Human MB: D425 **Animal model:** • C57BL6 mice: Used for orthotopic glioma (GL261-luc) and D425 MB xenograft models • Mice were treated with oral MBZ (different polymorphs or tablets) at 50 mg/kg daily for the first 20 days, then 5 days a week thereafter • Combination studies: • Elacridar 50 mg/kg orally, given 2 h before MBZ, for either 7 or 14 days	• Cell viability assay (Cell Counting Kit-8) • Infrared spectroscopy (polymorph/tablet characterization) • LC–MS/MS quantification (MBZ and metabolites in plasma/brain/tumour) • Pharmacokinetic analysis (Cmax, Tmax, AUC; brain/plasma and tumour/plasma ratios) • In vivo survival analysis (Kaplan–Meier) • Combination efficacy testing (MBZ ± elacridar)	• MBZ polymorphs A, B and C were equally cytotoxic to GL261 glioma cells • MBZ IC_{50} for D425 and GL261: approximately 0.11–1 μM **Pharmacokinetics MBZ-C:** • Brain Cmax: 2016 ng/g (7.1 μM) • Brain levels remained >2.7 μM for 1–8 h • Brain-to-plasma AUC ratio: 0.82 (0–24 h) • Plasma AUC: 16039 h·ng/mL ◦ Brain AUC: 13134 h·ng/g	• Only polymorphs B and C prolonged survival in GL261 glioma-bearing mice: ◦ MBZ-C: 48.5 days vs control 29 ◦ MBZ-B: 45 days, but associated with more weight loss ◦ MBZ-A: No benefit • MBZ-C reached brain concentrations >29-fold GL261 IC_{50} • MBZ + elacridar further improved survival: • In GL261 MBZ-C alone = 53 days ◦ Elacridar (7 d) = 92.5 days ◦ Elacridar (14 d) = 110.5 days • In D425 MB: ◦ MBZ alone = 52 days ◦ Elacridar: 77 days • MBZ-C is the most effective and best tolerated polymorph for brain tumours • Combination with elacridar enhances efficacy, but longer treatment may increase toxicity • Supports prioritization of MBZ-C for future brain cancer trials
Larsen et al. (2015),[42] United States of America	• To determine whether MBZ can inhibit Hedgehog signalling at clinically relevant concentrations. • To assess the effect of MBZ on the formation of the primary cilium, Hedgehog target gene expression, and growth of Hedgehog-driven cancer cells and tumours.	**Cell lines used:** • Human: ◦ hTERT-RPE1 ◦ DAOY (MB, Hh-subtype) ◦ 293 T • Mouse: ◦ NIH3T3 ◦ C3H10T1/2	• Reporter assay (GLI-luciferase) • qRT-PCR (Hedgehog target genes) • Cell proliferation assay (BrdU incorporation ELISA)	**IC_{50}:** • DAOY MB ◦ Suppression of GLI1 expression (Hedgehog pathway transcription factor) by MBZ = 516 ± 81 nM **Animal model:**	• MBZ suppressed GLI1 and PTCH1 expression in Hedgehog-activated cell lines at low micromolar–nanomolar concentrations. • MBZ strongly inhibited Hedgehog-dependent DAOY cells but had modest impact on Hedgehog-

(Continues)

TABLE 4 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
	• To evaluate the capacity of MBZ to inhibit Hedgehog pathway activation, including against mutations that confer resistance to standard Hedgehog inhibitors.	◦ Shh-Light2 ◦ SMO−/− MEFs **Animal model:** • Syngeneic GL261 gliomas in 4–6-week-old female nude (nu/nu) mice (GL261: PTEN-mutant mouse glioma line) • DAOY orthotopic medulloblastoma xenografts in 5–6-week-old female athymic mice (cerebellar injection of luciferase-expressing DAOY cells)	• Cell viability assay (CellTiter-Blue) • Clonogenic assay • Flow cytometry (Annexin V) • Western blot • Immunofluorescence microscopy • In vivo bioluminescence imaging • In vivo survival analysis (Kaplan–Meier)	• DAOY xenograft (MBZ 50 mg/kg, oral, daily) ◦ Increased median survival +38 days; decreased GLI1/ PTCH2; decreased tumour growth • DAOY xenograft (25 mg/kg, oral, daily) ◦ Increased median survival +19 days; decreased GLI1/ PTCH1 • GL261 glioma (oral MBZ) • Decreased GLI1 mRNA/ protein	responsive, non-tumour hTERT-RPE1 cells. • In GL261 glioma model, decreased GLI1 mRNA/protein in tumours. • Inhibited primary cilium formation, preventing SMO localization and Hedgehog activation; activity unaffected by vismodegib-resistant SMO mutations. • Combination with vismodegib showed additive Hedgehog pathway inhibition.
De Witt et al. (2017),[43] United States of America	• To investigate whether MBZ can serve as a safer and more effective replacement for vincristine (the standard microtubule inhibitor) for the treatment of brain tumours • Compare their mechanisms of action and efficacy in a mouse GBM model	**Cell lines used:** • Mouse GBM: GL261 **Animal model:** • Female C57BL/6 mice, age 3 months • Orthotopic brain tumour model using GL261/GLuc (GL261 cells transduced with Gaussia luciferase for blood-based measurement).	• Cell viability assay (MTT) • Western blot (tubulin polymerization) • Immunofluorescence microscopy (MPM2) • In vivo survival analysis (Kaplan–Meier) • Von Frey filament testing (peripheral neuropathy) • Body weight monitoring	EC_{50}: • MBZ (Polymorph C): ◦ Microtubule depolymerization = 132 nM ◦ Cell viability = 160 nM ◦ Mitotic arrest = 192 nM • Vincristine: ◦ Microtubule depolymerization = 1.36 nM ◦ Cell viability = 2 nM ◦ Mitotic arrest = 2.49 nM **Animal model:** • MBZ-C 50 mg/kg or 100 mg/kg (oral gavage, daily) • Vincristine 0.5 mg/kg or 1.0 mg/kg (weekly, intraperitoneal injection) • Combination: Rapid weight loss <1 week triggering euthanasia.	• MBZ and vincristine share the same mechanism of action (microtubule inhibition) • MBZ (both 50 mg/kg and 100 mg/kg daily oral doses) significantly extended survival in glioma-bearing mice, while vincristine (0.5 or 1.0 mg/kg weekly i.p.) did not improve survival. • Vincristine caused significant peripheral neuropathy; MBZ did not show statistically significant neurotoxicity at tested doses. • Combination of MBZ and vincristine resulted in rapid severe toxicity and weight loss. • Authors conclude that MBZ is more effective and safer than vincristine for brain tumour treatment in this model, supporting its use as a replacement for vincristine.

TABLE 4 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
Skibinski et al. (2018),[44] United States of America	• To assess the efficacy and mechanisms of MBZ alone and combined with radiation in malignant meningioma, *in vitro* and in an intracranial mouse model.	**Cell lines used:** • Human meningioma: ◦ KT21MG1 ◦ IOMM LEE ◦ AC-1 ◦ SF4068 ◦ SF6717 ◦ SF1335 ◦ SF1335 + YAP **Animal model:** • Female athymic nude mice (6 weeks old) • Intracranial implantation of KT21MG1-luciferase cells (50 000 cells in 0.5 μL Matrigel)	• Cell viability assay (CCK-8) • Clonogenic assay • Cell proliferation assay (BrdU ELISA) • Caspase-3/7 activity assay • Western blot • Immunohistochemistry (cleaved caspase-3, Ki67, CD31) • H&E staining • In vivo bioluminescence imaging (tumour growth) • In vivo survival analysis (Kaplan–Meier)	**IC_{50}:** • KT21MG1 (72 hrs): 0.391 ± 0.03 μM • IOMM LEE (72 hrs): 0.390 ± 0.01 μM • AC 1 (72 hrs) = 0.342 ± 0.05 μM • SF4068 (72 hrs) = 0.421 ± 0.04 μM • SF6717 (72 hrs) = 0.412 ± 0.02 μM • SF1335 (72 hrs) = 0.372 ± 0.04 μM • SF1335 + YAP (72 hrs) = 0.262 ± 0.03 μM **Animal model:** • KT21MG1 intracranial xenograft (oral MBZ 50 mg/kg/day in high-fat diet, ad libitum) ◦ Increased median survival by +58% *vs.* control (19 to 30 days) • KT21MG1 intracranial xenograft + single-dose local radiation (12 Gy, day 5) ◦ Median survival 33.5 days • KT21MG1 intracranial xenograft + combination of MBZ 50 mg/kg/day + radiation (12 Gy, day 5) ◦ Median survival increased by +105% *vs.* control (19 to 39 days)	• MBZ alone is cytotoxic to malignant meningioma cells at sub micromolar concentrations, suppressing proliferation and colony formation, and inducing caspase dependent apoptosis. • Combination with radiation further reduced colony formation in vitro and extended survival in vivo. • In tumours, MBZ and combination treatment decreased proliferation (Ki67), increased apoptosis (cleaved caspase 3), and lowered angiogenesis (CD31). • Supports MBZ, especially combined with radiation, as a preclinical candidate for malignant meningioma therapy.
Mena-Hernandez et al. (2020),[45] Mexico	• To develop and characterize MBZ microemulsions suitable for intranasal administration.	**Cell lines used:** • None **Animal model:** • Male Wistar rats (200–230 g) for orthotopic GBM model	• Fourier-transform infrared spectroscopy (FTIR) • Differential scanning calorimetry (DSC)	**IC_{50}:** • None **Animal model:** • MBZ microemulsion (formulation B): 250–	• Developed a stable, mucoadhesive MBZ microemulsion suitable for intranasal use (particle size ~145 nm; pH 4.5–6.5).

(Continues)

TABLE 4 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
	• To evaluate the efficacy and safety of intranasal MBZ microemulsion in a rat orthotopic GBM model.	• C6 rat glioma cell line (implanted intracranially; 1×10^6 cells/3 μL) • Sprague–Dawley rats (200–250 g) for nasal toxicity testing	• X-ray powder diffraction • Solubility screening (oils/surfactants) • Microemulsion characterization (particle size, zeta potential, PDI, pH, conductivity, TEM, rheology) • In vivo survival analysis (Kaplan–Meier) • Body weight monitoring • In vivo near-infrared fluorescence imaging (tumour growth) • Histology (H&E)	260 μg/mL (intranasal dose; 40 μL daily per rat; equivalent to 10.4 μg MBZ/day)	• The selected formulation (B: OA/Labrafil 1:1, sodium hyaluronate) had the best MBZ solubility, rheology, and stability. • 14 days of dosing caused no detectable damage or inflammation in nasal mucosa or effect on animal health. • Intranasal MBZ microemulsion significantly prolonged survival and reduced tumour growth in the orthotopic rat GBM model *vs*. control and vehicle. ◦ All MBZ-treated rats survived to day 50; controls/vehicle survived 20–30 days. ◦ Treated animals showed less body weight loss and lower fluorescence signal (smaller tumour burden) ◦ Tumours in MBZ-treated rats had lower cellularity, mitoses, and necrosis on histology. • Intranasal MBZ microemulsion is safe and shows therapeutic potential for GBM in preclinical models.
Yamashita et al. (2025),[46] United States of America	• To evaluate the effect of MBZ alone and combined with ionizing radiation therapy in preclinical models of IDH1 mutant glioma. • To investigate underlying mechanisms including effects on proliferation, apoptosis, DNA damage, and cell cycle arrest in vitro and in vivo.	**Cell lines used:** • Primary focus: ◦ BT142 (human, patient-derived, IDH1-R132H mutant, grade III oligodendroglioma) • Other patient-derived glioma lines (IDH- mutant and wild-type): ◦ JHH 136 ◦ JHU 0879 ◦ Br23c ◦ JHH 520 ◦ JHH 227A ◦ TS603 ◦ CCF STTG1 • Human:	• Cell proliferation assays (Alamar Blue; CellTiter-Glo) • Neurosphere formation assay • Caspase-3/7 activity assay • Western blot • ELISA • Immunohistochemistry • Flow cytometry • In vitro radiation treatment schedule • In vivo survival analysis (Kaplan–Meier) • Histology (H&E)	**IC_{50}:** • Cancer cell lines (broad panel) (72 hrs): 130–884 nM • Patient derived IDH mutant glioma lines (72 hrs): 215–474 nM **Animal model:** • MBZ oral gavage 50 mg/kg (sesame oil vehicle), 5 days/week × 4 weeks then 2 days/week until endpoint: ◦ median survival 187 days vs control 100 days (+87%)	• MBZ induced nanomolar-level growth inhibition *in vitro*, reduced neurosphere formation, increased caspase-dependent apoptosis, and elevated DNA damage marker pγH2AX. • Caspase inhibition (Z-VAD-FMK) blunted MBZ-induced apoptosis, DNA damage, and growth inhibition. • MBZ disrupted microtubule polymerization and arrested cells in G2/M; effects independent of IDH1-mutant oncometabolite (2-HG) inhibition and MAPK pathway changes. • MBZ + RT further reduced proliferation, enhanced G2/M arrest,

TABLE 4 (Continued)

Author/ year/ country	Research aims	Sample details	Assays performed	Potency measures	Key findings
		◦ U251 ◦ U87 ◦ DIPG1 (diffuse intrinsic pontine glioma) ◦ DIPG16A (brainstem glioma) ◦ H4 (neuroglioma) • Other human cancer/ comparator lines: ◦ HT1080 (fibrosarcoma) ◦ SW1080 (colorectal cancer) ◦ SW1353 (chondrosarcoma, IDH2 R172S mutant) ◦ C8 D1A (astrocyte derived comparator line) **Animal model:** • Female athymic nude mice (6–8 weeks old) • BT142 cells (3×10^5) were orthotopically implanted in a stereotaxic frame		• RT alone 5 Gy single fraction on day 10 post-implantation: ◦ median survival 130 days (NS vs control) • Oral MBZ as above + RT day 10: ◦ Median survival not reached at end of study (146–295 days range) ◦ Significantly longer than MBZ alone ($p = 0.039$) and RT alone ($p = 0.0007$) ◦ 6/11 mice (54.4%) long-term survivors (>290 days) with no histologically detectable tumour	modulated DNA damage and cell cycle proteins, and reduced neurosphere formation more than either agent alone. • In vivo, MBZ + RT produced the greatest survival benefit, with over half of mice achieving long-term, tumour-free survival. • MBZ, especially in combination with radiation, is a promising, well-tolerated candidate for IDH-mutant glioma therapy.

AUC, area under the concentration–time curve; **BBB**, blood–brain barrier; **BCRP**, breast cancer resistance protein; **BrdU**, 5-bromo-2′-deoxyuridine; **CCK-8**, Cell Counting Kit-8; **CD31**, cluster of differentiation 31 (platelet endothelial cell adhesion molecule-1); **CI**, confidence interval; **Cmax**, maximum (peak) concentration; **CMYC**, MYC proto-oncogene; **CNS**, central nervous system; **DAPI**, 4′,6-diamidino-2-phenylindole; **DEF50**, dose enhancement factor at 50% effect; **DMG**, diffuse midline glioma; **DSC**, differential scanning calorimetry; **EC50**, half-maximal effective concentration; **ELISA**, enzyme-linked immunosorbent assay; **EMT**, epithelial–mesenchymal transition; **FTIR**, Fourier-transform infrared spectroscopy; **GBM**, glioblastoma multiforme; **GLI1**, glioma-associated oncogene homologue 1; **GTEx**, Genotype-Tissue Expression project; **H&E**, haematoxylin and eosin; **Hh**, Hedgehog; **HUVEC**, human umbilical vein endothelial cell; **IDH**, isocitrate dehydrogenase; **IC_{50}**, half-maximal inhibitory concentration; **IF**, immunofluorescence; **IHC**, immunohistochemistry; **IR**, ionizing radiation; **LC–MS/MS**, liquid chromatography–tandem mass spectrometry; **MB**, medulloblastoma; **MBZ**, mebendazole; **MPM2**, mitotic phosphoprotein monoclonal antibody 2; **MTT**, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide; **NIH3T3**, NIH Swiss mouse embryo fibroblast cell line (3 T3); **NS**, not significant; **OTX2**, orthodenticle homeobox 2; **PBS**, phosphate-buffered saline; **PDGFRα**, platelet-derived growth factor receptor alpha; **PDI**, polydispersity index; **PFS**, progression-free survival; **P-gp**, P-glycoprotein (ABCB1); **PTCH**, patched 1; **qRT-PCR**, quantitative reverse-transcription polymerase chain reaction; **RNA-seq**, RNA sequencing; **RT**, radiotherapy; **SEM**, standard error of the mean; **SHH**, Sonic hedgehog; **siRNA**, small interfering RNA; **SMO**, smoothened; **TCGA**, The Cancer Genome Atlas; **TEM**, transmission electron microscopy; **Tmax**, time to maximum concentration; **TMZ**, temozolomide; **VEGF**, vascular endothelial growth factor; **VEGFR2**, vascular endothelial growth factor receptor 2; **WST-8**, water-soluble tetrazolium salt-8.

strategies to enhance drug penetrance across these interfaces remain a major research focus.[55]

Beyond its role in the cytoskeleton, MBZ has been found to directly inhibit kinases, including MAPK14, with additional activity against ABL1 and ERK2.[33] Mechanistically, this positions it alongside other kinase inhibitors such as **sorafenib**, **ralimetinib** and **imatinib**, but with the advantage of multi-target engagement. However, as highlighted in clinical experience with other broad-spectrum kinase inhibitors, this breadth of activity can also raise safety concerns, since off-target effects may contribute to increased toxicity and limit tolerability.[56] Importantly, these kinase-directed effects are complemented by additional pathway-specific actions, underscoring the diverse mechanisms through which MBZ may exert anticancer activity. Aberrant Hedgehog pathway signalling is known to drive tumourigenesis by promoting uncontrolled proliferation, survival and stem-like behaviour in cancer cells.[57] MBZ has been shown to prevent primary ciliary formation in this pathway, thereby blocking SMO localization and down-regulating GLI1/PTCH transcriptional activity (Figure 2).[42] This mirrors the action of clinically approved SMO inhibitors such as vismodegib and **sonidegib**, but with the added advantage that MBZ retains activity in SMO-mutant, vismodegib-resistant settings by acting upstream of SMO.[40]

Angiogenesis, the process of new blood vessel formation from pre-existing vessels, is normally governed by a tightly regulated angiogenic switch. However, this control is lost in pathological states like cancer, driving uncontrolled cell growth and excessive vascularization.[58] MBZ has been shown to interfere with tumour vascularization by inhibiting VEGFR2,[40] one of the key regulators of this process and a major therapeutic target.[59] In addition to VEGFR2 inhibition, MBZ alters PDGFRα phosphorylation, culminating in the selective reduction of microvessel density while largely sparing normal brain vasculature.[40] This profile resembles the actions of established anti-angiogenic agents such as **sunitinib**,[60] but is distinct in that it unites these effects with cytoskeletal disruption and additional signalling modulation, thereby offering a multi-faceted therapeutic advantage. Additionally, MBZ impairs the DNA damage response by preventing nuclear import of key repair proteins such as Chk2 and Nbs1, sustaining γH2AX signalling and delaying repair following radiation exposure.[31] These actions align conceptually with PARP inhibitors such as **olaparib**, which are currently being evaluated in clinical trials for brain

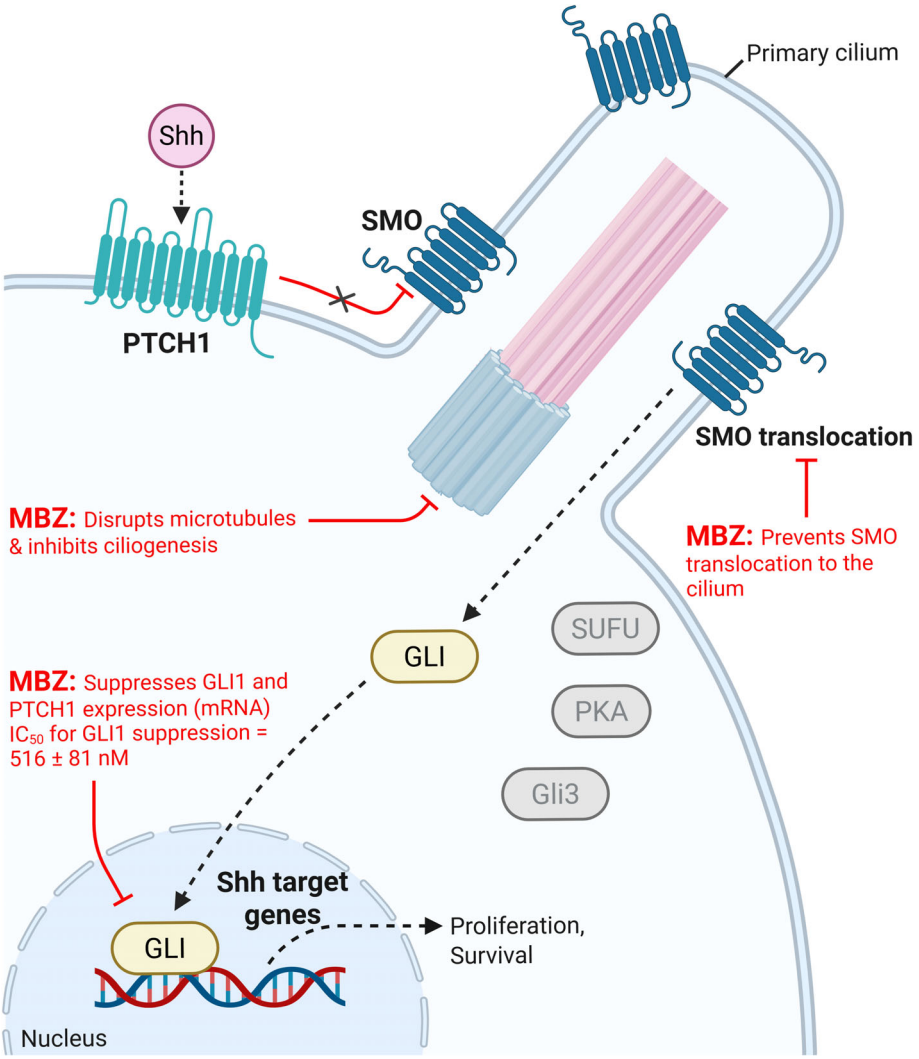

FIGURE 2 Mebendazole blocks primary-cilium-dependent Hedgehog signalling by preventing SMO translocation and downstream GLI activation, including in vismodegib-resistant SMO-mutant settings. In the canonical pathway, sonic hedgehog (Shh) binds the receptor PTCH1 at the ciliary membrane. Ligand binding causes PTCH1 to exit the primary cilium, which relieves repression of SMO and allows SMO to translocate from the plasma membrane/endosomal pool into the cilium. Ciliary SMO then initiates signalling that converts GLI transcription factors from SUFU-bound, inactive forms to active forms that accumulate at the ciliary tip, translocate to the nucleus and drive transcription of target genes, including GLI1 (an amplifier of pathway output) and PTCH1 (negative-feedback receptor). MBZ suppresses hedgehog pathway activity, with reported GLI1 expression suppression at an IC_{50} of 516 ± 81 nM (in DAOY cells) and retains activity against vismodegib-resistant SMO mutants. Figure created in BioRender. O'Callaghan, L. (2026) https://BioRender.com/ottz6hu.

cancer,[7] as well as checkpoint kinase inhibitors like **prexasertib**,[61] thereby positioning MBZ as a potential radiosensitizer in gliomas.

Further anticancer actions attributed to MBZ include suppression of migration, invasion and EMT, evidenced by down-regulation of mesenchymal markers (N-cadherin, **β-catenin**, **MMP2**, Slug) and up-regulation of the epithelial marker E-cadherin.[35] While these effects mirror those of Wnt/β-catenin and MMP inhibitors such as **marimastat**,[62] it remains unclear whether MBZ engages these pathways directly or whether these changes arise secondary to broader cytotoxic stress. Similarly, the reported ability of MBZ to modulate programmed cell death appears highly context-dependent. Autophagy induction has been observed in some tumour models,[34] whereas pyroptosis via NF-κB/NLRP3/GSDMD signalling has been described in others.[35] These observations invite comparison with established modulators of autophagy (e.g. chloroquine derivatives)[63] and pro-pyroptotic agents (e.g. anthracyclines),[64] but also raise questions about the reproducibility and clinical relevance of these mechanisms in brain tumours. These findings illustrate that MBZ exerts anticancer effects through many mechanisms, including cytoskeletal disruption, kinase inhibition, pathway-specific signalling interference, angiogenesis blockade and impairment of the DNA damage response. This breadth positions MBZ as distinct from single-target agents, effectively combining the therapeutic actions of multiple existing drug classes within a single compound, thereby underscoring its potential as a versatile candidate for brain tumour therapy.

4.2 | Therapeutic applications, resistance and combination strategies

MBZ exhibits broad and pleiotropic activity in neuro-oncology, demonstrating efficacy across diverse tumour histologies, including GBM, DMG/diffuse intrinsic pontine glioma (DIPG), MB (both sonic hedgehog [SHH] and Group-3 subtypes) and malignant canine glioma, as well as in treatment-resistant disease states that remain highly relevant in current clinical practice. Across preclinical models, MBZ demonstrates efficacy consistent with a multi-target mechanism, encompassing microtubule disruption, VEGFR2-mediated anti-angiogenesis, Hedgehog pathway inhibition and therapy-enhancing effects through modulation of DNA damage and autophagy.[39,40,42] This breadth of activity helps to explain why MBZ shows efficacy across tumours as distinct as H3K27-altered DMG and SHH MB, and why its effects are often preserved regardless of TP53 status.[46] Notably, MBZ has retained potency in ONC201-resistant DMG and SMO-mutant, vismodegib-resistant MB, reinforcing its pathway-agnostic utility in tumours with constrained mutational targetability.[40] This cross-resistance activity suggests that MBZ can act as a 'fail-safe' across diverse resistance pathways, which is an appealing attribute in diseases like GBM, where adaptive resistance is near-universal,[65] thereby supporting continued investigation in rational combination strategies.

From a therapeutic standpoint, the most tangible translational opportunity lies in rational combination strategies. MBZ augments alkylators (e.g. TMZ and lomustine) and ionizing radiation in multiple GBM systems.[48] MBZ has also been shown to enhance the effects of radiation in malignant meningioma,[44] and demonstrates synergy with autophagy inhibitors such as chloroquine, where triple-agent regimens achieve more pronounced tumour suppression than doublet combinations.[34] These findings are biologically plausible: microtubule inhibition induces mitotic stress by delaying cell-cycle progression and promoting mitotic slippage,[66] alkylating agents impose additional genotoxic damage,[67] and autophagy blockade removes a critical survival mechanism.[68] Together, these cellular insults drive cells more decisively towards apoptosis. The observation that certain triple-agent regimens outperform doublets across models strengthens the rationale for prospectively evaluating MBZ-based combinations, provided that pharmacokinetic and dose-density considerations are carefully addressed.[48] Emerging evidence also suggests that MBZ may complement immunotherapy by modulating immune checkpoints and promoting immunogenic cell death,[69] although this remains to be fully tested in brain tumour settings. Realizing these opportunities, however, will depend on overcoming pharmacokinetic variability and ensuring consistent exposure, which remain major barriers to clinical reproducibility.

4.3 | Formulation, delivery and brain exposure

A central challenge in the translational development of MBZ is achieving consistent and therapeutically relevant brain exposure.[70] Comparative studies of MBZ polymorphs show that polymorph C achieved superior brain penetration, tolerability and survival benefit relative to polymorphs A and B in preclinical models.[41] These findings indicate that formulation is biologically important and may influence reproducibility across studies. However, current human data are insufficient to conclude that inconsistent clinical efficacy is primarily a consequence of polymorph variability or inadequate CNS delivery alone. Rather, formulation likely represents one of several interacting translational barriers, alongside limited trial size, heterogeneous populations and the possibility that preclinical activity may not fully translate into clinically meaningful benefit. Brain and tumour exposure may also be affected by efflux transporters at the BBB. Bai et al.[41] showed that co-administration of MBZ polymorph C with elacridar improved survival in a mouse glioma model, suggesting that transporter-mediated efflux may restrict CNS exposure (Figure 3). More broadly, poor aqueous solubility, variable oral absorption and substantial pharmacokinetic heterogeneity remain recognized challenges for MBZ repurposing.[41,71,72] Alternative delivery approaches, including nanosuspensions and intranasal microemulsions, have shown encouraging preclinical results by improving solubility, tissue exposure, or local tolerability.[37,45]

These findings support continued investigation of formulation and delivery optimization, but such strategies should not be interpreted as a substitute for demonstrating clinical efficacy. Improved exposure may strengthen the translational case for MBZ, yet it remains necessary to determine, in adequately designed human

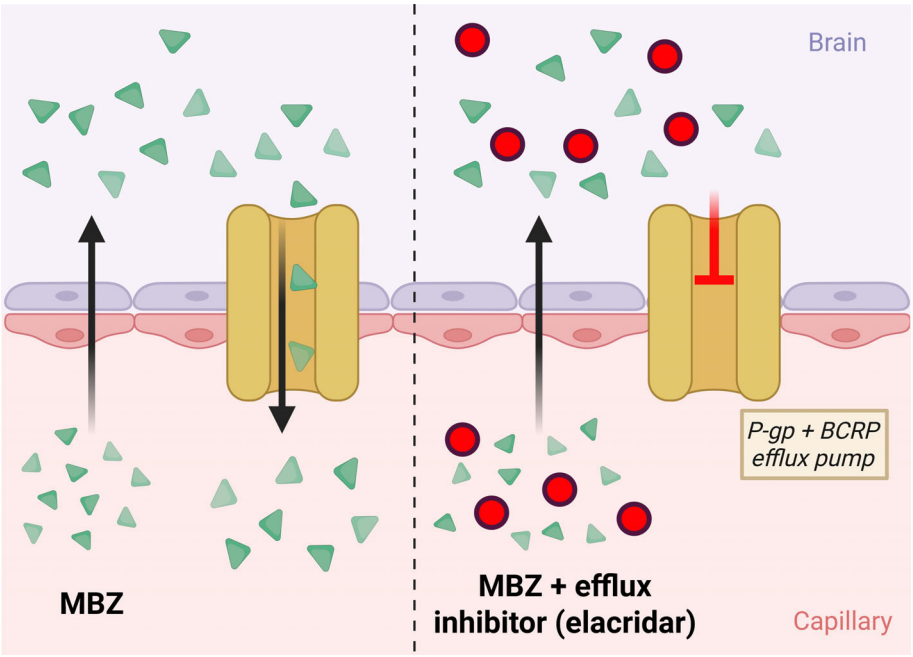

FIGURE 3 Blood–brain barrier efflux limits brain penetration of mebendazole polymorph C, whereas elacridar increases central nervous system exposure. Left panel: schematic of a brain microvascular endothelial cell with tight junctions sealing the paracellular route. MBZ enters the cell by passive transcellular diffusion but is actively transported back to the luminal (blood) side by P-glycoprotein (P-gp; ABCB1/MDR1) on the apical membrane, resulting in low net flux to the abluminal (brain) side and limited brain accumulation. Right panel: co-administration of elacridar (GF120918), an inhibitor of P-glycoprotein and breast cancer resistance protein (BCRP/ABCG2), blocks apical efflux. With efflux activity suppressed, a greater fraction of MBZ crosses the endothelium and reaches the brain interstitial space, increasing intraparenchymal exposure. In the referenced preclinical study, the reported brain-plasma ratio of 0.82 was observed for MBZ polymorph C. Figure created in BioRender. O'Callaghan, L. (2026) https://BioRender.com/9e2532n.

studies, whether enhanced delivery translates into better patient outcomes. Future trials should, therefore, report the polymorph and formulation used explicitly, incorporate pharmacokinetic measurements and relate exposure to clinically meaningful endpoints.

4.4 | Clinical evidence, safety profile and translational trial design

The current clinical evidence does not establish MBZ as an effective treatment for brain tumours in humans. Although several early-phase studies demonstrate that high-dose oral MBZ is feasible to administer, the available efficacy signals are modest, inconsistent and largely derived from small, heterogeneous cohorts (Table 5).[47–50] The strongest comparative evidence comes from the randomized phase II trial in recurrent GBM, in which MBZ added to lomustine or TMZ did not meet the predefined efficacy benchmark in the overall study population and did not produce a clear survival advantage overall.[49] A possible benefit was observed only in a post hoc subgroup of fitter patients receiving lomustine plus MBZ, which should be regarded as hypothesis-generating rather than definitive.

Single-arm adult studies likewise provide limited evidence of efficacy. In newly diagnosed high-grade glioma, MBZ combined with TMZ was associated with median OS of 21 months,[48] but the absence of a control group prevents attribution of this outcome to MBZ itself. Similarly, phase I dose-escalation work in recurrent high-grade glioma established feasible dose levels for combination regimens, yet survival outcomes remained limited, and the studies were not designed to demonstrate efficacy.[47] Paediatric data follow a similar pattern: combination therapy with bevacizumab and irinotecan yielded occasional radiographic responses, but PFS remained short,[50] while MBZ monotherapy showed no objective responses and limited PFS in recurrent or refractory disease.[51] Taken together, the human literature presently supports tolerability more strongly than effectiveness. This distinction is important. The presence of multiple biologically plausible anticancer mechanisms and consistent preclinical activity does not, on its own, justify a strong claim for clinical usefulness. At present, the most defensible interpretation is that MBZ remains an investigational repurposing candidate whose efficacy in humans is uncertain. Future trials are therefore needed not because efficacy has already been demonstrated and requires refinement, but because clinically meaningful benefit has not yet been established. Such studies should be adequately powered, include appropriate comparators, and clearly define whether MBZ is being evaluated as a monotherapy strategy or as a component of rational combination regimens.

Safety and feasibility data remain encouraging. Across adult and paediatric studies, MBZ has generally been well-tolerated at high oral doses, with adverse events including gastrointestinal symptoms,

TABLE 5 Population-based studies on the anticancer effects of mebendazole.

Author/ year/ country	Research aims	Study design/cohort	Data analysis	Key findings
Patil et al. (2020),[47] India	• To identify the maximum tolerated dose of MBZ in adults with recurrent high-grade glioma when combined with lomustine, TMZ, or TMZ plus re-irradiation. • To assess the acute toxicity, safety, and tolerability of high-dose oral MBZ with these regimens for future clinical translation.	**Design:** • Multi-arm, open-label, single-centre, phase 1 dose-escalation trial (Accelerated titrated design 4, with modified Fibonacci escalation schema). **Cohort:** • 11 adult patients with recurrent high-grade glioma (GBM n = 6, grade 2–3 astro−/oligodendroglia n = 5) • Median age: 46 years (range 25–68) • Gender: 8 males, 3 females • All had prior surgery, radiotherapy, and TMZ. **Intervention:** • Arm A1: MBZ + TMZ + re-irradiation (1 patient) • Arm B1: MBZ + lomustine (9 patients) • Arm C1: MBZ + TMZ (1 patient) • MBZ was administered orally as 100 mg chewable tablets at 5 dose levels (100–1600 mg three times daily [TDS]). • Polymorph was not reported. • Dose-limiting toxicity (DLT) was assessed during the first cycle/first 1–6 weeks, depending on treatment arm.	• Descriptive statistics for patient characteristics, adverse events, and dose escalation. • Toxicity graded by CTCAE v4.03. • Kaplan–Meier method for PFS and OS; Brookmeyer & Crowley for CI. • Primary outcome: maximum tolerated dose defined as the dose at which ≥2 dose limiting toxicities observed per arm.	• Maximum tolerated dose (and recommended phase 2 dose) of MBZ: ◦ 1600 mg TDS (4800 mg/day) with TMZ (± re-irradiation) ◦ 800 mg TDS (2400 mg/day) with lomustine • No dose limiting toxicities observed at any dose in TMZ-containing arms; maximum tolerated dose reached at 1600 mg TDS in CCNU arm (due to neutropenia/thrombocytopenia). • Commonest adverse events: anaemia (82%), nausea (64%), fatigue (55%); grade ≥3 toxicity infrequent. • Median PFS: 6.3 months; median OS: 7.7 months. • High-dose MBZ was generally well tolerated and feasible for oral administration in this setting.
Gallia et al. (2021),[48] United States of America	• To evaluate the maximum tolerated dose, safety, toxicity, and plasma levels of high-dose MBZ in combination with TMZ in patients with newly diagnosed high-grade gliomas. • To provide preliminary estimates of OS and PFS in this patient group.	**Design:** • Single-centre, open-label, nonrandomized, phase 1 dose-escalation trial using a standard 3 + 3 design. **Cohort:** • 24 adult patients (≥18 years) with newly diagnosed malignant glioma (WHO grade III or IV) who had completed surgery and postoperative chemoradiation per the Stupp protocol. ◦ Diagnosis: 18 GBM, 6 anaplastic astrocytoma ◦ Median age: 49.8 years (range 27.8–67.5) ◦ All patients white; KPS: range 60–100 **Intervention:** • Oral MBZ-C was administered at 4 dose levels (25, 50, 100, and 200 mg/kg/day in	• Adverse events graded by CTCAE v4.0, dose-limiting toxicity defined as any nonhematologic grade ≥3 or hematologic grade ≥4 toxicity. • Plasma trough concentrations (Cmin) of MBZ and metabolites measured at weeks 4, 8, and 16 via LC–MS. • Descriptive statistics for demographics and toxicity, Kruskal-Wallis ANOVA for dose comparisons, Mann–Whitney U-tests for correlation, Kaplan–Meier for OS/PFS estimates.	• High-dose oral MBZ (up to 200 mg/kg/day) combined with TMZ was safe and well tolerated in adults with newly diagnosed high-grade gliomas. • The most common side effect was reversible elevation of liver enzymes, observed only at the highest dose. • No severe adverse events attributable to MBZ occurred. • Median OS was 21 months; median PFS was 13 months. • Findings support further clinical trials of MBZ in this patient population.

(Continues)

TABLE 5 (Continued)

Author/ year/ country	Research aims	Study design/cohort	Data analysis	Key findings
		divided doses) in combination with standard adjuvant TMZ (75 mg/m^2). • MBZ was provided as a custom non-commercial IND formulation of polymorph C in 500 mg chewable tablets. • An expansion cohort included 15 patients treated at the highest dose level (200 mg/kg/day)		
Patil et al. (2022),[49] India	• To evaluate whether the addition of MBZ to lomustine or TMZ improves 9-month OS in adults with recurrent GBM ineligible for re-irradiation. • To determine progression free survival, safety/ tolerability, and factors impacting OS. • To explore post hoc whether ECOG performance status (PS) influences treatment benefit.	**Design:** • Single-centre, randomized, open-label phase II (1:1), historical-control benchmark OS ≥55%. Sample size: 44 pts/arm; India; CTRI/2018/01/011542. **Cohort:** • 88 adults, median age ∼41, 73–75% male, ECOG PS 0–1 (71%), ECOG PS 2–3 (29%). • IDH-mutated: 36%; <50% had MGMT status. • All had prior surgery & RT; most had prior TMZ. **Intervention:** • Patients were randomized 1:1 to TMZ-MBZ or CCNU-MBZ. • TMZ-MBZ: TMZ 200 mg/m^2 on days 1–5 every 28 days (up to 12 cycles) plus MBZ 1600 mg TID (4.8 g/day). • CCNU-MBZ: CCNU 110 mg/m^2 on day 1 every 42 days (up to 6 cycles) plus MBZ 800 mg TID (2.4 g/day). • MBZ was administered as polymorph B (Mebex 100 mg chewable tablets; Cipla Limited). • Treatment continued until disease progression, unacceptable toxicity, or withdrawal, and the MBZ dose remained unchanged throughout treatment.	• A'Hern one stage design • Kaplan–Meier for OS and PFS; reverse Kaplan–Meier for median follow up. • Brookmeyer & Crowley method for medians and 9-month OS estimates. • Cox proportional hazards regression; hazard ratios via Efron's method for ties. • OS restricted to ECOG PS 0–1 subgroup. • Toxicity graded per CTCAE v4.03	• Nine-month OS 36.6% for TMZ + MBZ; 45.0% for lomustine + MBZ; both below the 55% target. • Median OS 6.70 months vs 6.53 months; no significant difference. • Median progression-free survival 4.13 months vs 4.27 months; no significant difference. • In patients with good performance status (score 0–1), lomustine + MBZ met the 55% target (57.9%); TMZ + MBZ was 39.6%. • Good performance status was the only factor linked to longer survival. • Severe side effects occurred in 18.6% for TMZ + MBZ and 9.5% for lomustine + MBZ; most common were low platelets, low neutrophils, liver enzyme increases, and low blood sodium. • Both combinations were safe but did not improve survival overall; possible benefit for lomustine + MBZ in fitter patients.
Krystal et al. (2024),[50] United	• To determine the maximally tolerated dose of MBZ when given in combination with bevacizumab and irinotecan in children and young adults with HGG and DMG.	**Design:** • Multicentre, open-label, nonrandomized, phase 1 dose-escalation trial using a standard 3 + 3 design (NCT01837862). **Cohort:**	• Monitored for dose-limiting toxicities using Common Terminology Criteria for Adverse Events (CTCAE v4.0). • Objective response was evaluated via MRI using Response Assessment in	• No dose-limiting toxicities observed at doses up to 200 mg/kg/day mebendazole (MTD not reached).

TABLE 5 (Continued)

Author/ year/ country	Research aims	Study design/cohort	Data analysis	Key findings
States of America	• To describe progression-free survival and OS in this cohort. • To evaluate clinical and MRI responses after adding mebendazole to multimodal therapy.	• 10 paediatric and young adult patients (ages 1–21 years) with HGG or DMG. ◦ Diagnosis: 7 DMG, 1 anaplastic astrocytoma, 1 spinal HGG. ◦ Median age at diagnosis: 9 years (range 4–18). ◦ Tumour location: brainstem, thalamus, septum pellucidum/corpus callosum, spine. ◦ All patients received prior radiation; 7 treated at initial diagnosis, 2 at first relapse. **Intervention:** • MBZ, supplied as Janssen 100 mg or 500 mg chewable tablets (polymorph not reported), was administered orally twice daily across three sequential dose cohorts: ◦ 50 mg/kg/day (n = 3) ◦ 100 mg/kg/day (n = 4; one unevaluable) ◦ 200 mg/kg/day (n = 3) • MBZ was combined with bevacizumab 10 mg/kg IV and irinotecan 125–150 mg/m^2 IV on days 1 and 15 of each 28-day cycle, with treatment continued for up to 12 cycles or until progression, toxicity, or withdrawal.	Neuro-Oncology (RANO) criteria every three cycles. • Progression-free survival and OS were calculated as means (months) from the start of study treatment. • Descriptive statistics were used for demographics, AEs, and outcomes due to small sample size (no formal power analysis).	• Most common grade 3/4 adverse events: neutropenia (33%), lymphopenia (22%). • No unexpected toxicities attributable to MBZ. • Overall response rate: 33% (2 subjects had partial responses, 1 had a complete response sustained for 10 months). • Mean progression-free survival: 4.7 months. • Mean OS: 11.4 months. • All evaluable patients were eventually removed due to disease progression. • MBZ is safe and well-tolerated at high doses in combination with bevacizumab and irinotecan in paediatric HGG/DMG. • Provides foundation for further studies, though efficacy remains to be established in larger cohorts.
Phan et al. (2025),[51] United States of America	• To determine the safety, tolerability and maximum tolerated dose of oral MBZ administered as single-agent therapy in paediatric patients with refractory, progressive, or recurrent brain tumours. • To explore PFS as a secondary outcome to inform future combination or translational studies.	**Design:** • Two-centre, open-label, non-randomized, phase 1 dose-escalation trial using a standard 3 + 3 design. **Cohort:** • 17 paediatric patients (aged 1–21 years) with refractory, progressive, or recurrent brain tumours. • Tumour types included: ◦ diffuse intrinsic pontine glioma (n = 3), diffuse midline glioma (n = 5), glioblastoma multiforme (n = 2), medulloblastoma (n = 2), anaplastic astrocytoma (n = 2), and single cases of anaplastic ependymoma and glioneuronal tumour.	• Descriptive statistics for patient characteristics and adverse events. • Toxicities graded using CTCAE v4.0. • Maximum tolerated dose defined per standard 3 + 3 criteria. • Kaplan–Meier method used to estimate PFS from initiation of MBZ to progression or death. • Radiographic response assessed using RANO criteria, with confirmatory imaging required for CR/PR.	• No dose-limiting toxicities were observed at any dose level; MBZ was well tolerated up to 2500 mg/m2/day, establishing this dose as the MTD. • 121 adverse events were reported, with 69 possibly/probably MBZ-related; most were grade 1–2. • Seven grade 3–4 MBZ-related AEs occurred, with no treatment-related deaths. • Five patients did not complete the DLT period due to early progression or death and were replaced for MTD determination. • No objective radiographic responses were observed.

(Continues)

TABLE 5 (Continued)

Author/ year/ country	Research aims	Study design/cohort	Data analysis	Key findings
		• Median age: 12 years (range 3–17) • Prior treatment burden: ◦ Prior therapy: 0–4 previous treatment lines; 14 treated at progression and 3 for anticipated progression. • Eligibility required adequate organ/marrow function and KPS/Lansky ≥50. **Intervention:** • Oral MBZ monotherapy, using a non-commercial 500 mg polymorph C tablet formulation manufactured by Aurochem Laboratories for Johns Hopkins clinical trials, was administered three times daily with meals at dose levels of 1250, 1875, and 2500 mg/m^2/day. • Treatment continued until toxicity or progression, with DLT assessed over a 1-month observation period.		• Mean PFS was 7.6 weeks (range 2–24 weeks); a single prolonged case was confounded by recent surgery and re-irradiation. • Overall, MBZ demonstrated acceptable safety but limited single-agent efficacy in this population.

AEs, adverse events; **ANOVA**, analysis of variance; **CCNU**, lomustine (1-[2-chloroethyl]-3-cyclohexyl-1-nitrosourea); **CI**, confidence interval; **CMIN/Cmin**, minimum (trough) concentration; **CR**, complete response; **CTCAE**, Common Terminology Criteria for Adverse Events; **CTRI**, Clinical Trials Registry–India; **D1–5**, days 1 to 5; **DLT**, dose-limiting toxicity; **DMG**, diffuse midline glioma; **ECOG**, Eastern Cooperative Oncology Group; **GBM**, glioblastoma multiforme; **HGG**, high-grade glioma; **IDH**, isocitrate dehydrogenase; **IND**, Investigational New Drug; **IV**, intravenous; **KPS**, Karnofsky Performance Status; **LC–MS**, liquid chromatography–mass spectrometry; **MRI**, magnetic resonance imaging; **MTD**, maximum tolerated dose; **NCT**, National Clinical Trial (ClinicalTrials.gov identifier); **NS**, not significant; **O6-methylguanine-DNA methyltransferase** (**MGMT**), O6-methylguanine-DNA methyltransferase; **OS**, overall survival; **PFS**, progression-free survival; **PR**, partial response; **PS**, performance status; **q28d/q42d**, every 28/42 days; **RANO**, Response Assessment in Neuro-Oncology; **RT**, radiotherapy; **Stupp protocol**, standard concomitant chemoradiotherapy with temozolomide followed by adjuvant temozolomide; **TDS/TID**, three times daily; **TMZ**, temozolomide; **WHO**, World Health Organization.

reversible transaminase elevation, anaemia and regimen-related haematological toxicity.[47–51] Importantly, this safety profile supports continued clinical investigation. However, tolerability should not be conflated with efficacy, and the current evidence base is not yet sufficient to support routine use in neuro-oncology outside of research settings. Future translational trial design should therefore prioritize rigorous efficacy assessment, while also improving reporting consistency. In particular, studies should specify the MBZ polymorph and formulation used, given the known pharmacokinetic differences between polymorphs and the inconsistent reporting across the current clinical literature. Although Gallia et al.[48] and Phan et al.[51] explicitly reported polymorph C and Patil et al.[49] reported polymorph B, other studies did not clearly specify the polymorph used. Pharmacokinetic and pharmacodynamic endpoints should be incorporated prospectively, and patient-centred outcomes such as cognition, corticosteroid dependence and health-related quality of life may be valuable adjuncts to conventional survival measures in this setting.[73–75]

4.5 | Limitations and future directions

Despite compelling preclinical evidence, the translational trajectory of MBZ in neuro-oncology faces important limitations. To date, studies have focused on primary brain tumours such as GBM, DMG and MB, yet metastatic brain cancer represents a far more common clinical challenge,[76] and remains largely unexamined in MBZ research. This restricts the external validity of current findings and leaves open questions about whether MBZ's mechanisms extend to secondary brain malignancies. Furthermore, there is marked heterogeneity in experimental design, with some studies relying on human-derived cell lines and others on animal-derived systems or a combination of both. While each model provides unique insights, animal systems remain imperfect surrogates for human disease, with differences in biology, pharmacokinetics and treatment response that complicate cross-comparison and raise concerns about the predictive validity of preclinical findings.[77]

Beyond these biological considerations, the quality of evidence across different streams is uneven. *In vitro* studies frequently lacked reporting of sample-size justification, randomization and blinding, raising concerns about reproducibility. This reflects a well-recognized, discipline-wide issue, with surveys and multicentre experiments revealing widespread irreproducibility in preclinical biomedical research.[78–80] *In vivo* models consistently demonstrated survival benefit, but deficiencies in allocation concealment and blinding reduced confidence in internal validity. Clinical evidence to date has come from small, early-phase trials, the majority of which were single-centre and geographically restricted. Non-randomized studies[47,48,50] contributed valuable safety and feasibility data but were consistently subject to serious or critical risks of bias, primarily due to confounding, selection effects and the absence of comparators. In contrast, the single randomized trial[49] offered comparatively stronger internal validity, with lower risk of bias overall, although the efficacy signals were modest and the limited sample size constrained statistical power and generalizability. Collectively, these methodological limitations constrain the confidence with which efficacy findings can be interpreted and reinforce the need for more rigorous and transparent trial design in future work.

Variability in MBZ polymorph usage and the frequent omission of formulation reporting introduce an additional biological confounder, as polymorph C achieved superior bioavailability and tolerability in preclinical work compared with other forms.[41,81] Without consistent reporting and standardization, interpretation across studies remains difficult. Future case reports and clinical trials should therefore explicitly declare the polymorph and formulation used. This is especially important in light of the current evidence base, where inadequate reporting may obscure whether inconsistent outcomes reflect pharmacokinetic variability, study design differences or limited intrinsic clinical activity.

Looking forward, the immediate priority is not simply to optimize formulation, but to determine more clearly whether MBZ improves clinically meaningful outcomes in humans. Formulation and delivery innovations such as nanosuspensions, lipid-based carriers and intranasal systems may enhance exposure and deserve further evaluation,[37,45,70] but they should be developed alongside, rather than instead of, rigorous clinical testing. Future studies should prioritize biomarker-informed and pharmacokinetically integrated trial designs, include appropriate comparators, and focus on settings where MBZ has the strongest rationale, such as rational combination regimens or biologically selected subgroups. Only through this combination of critical efficacy testing, transparent formulation reporting, and improved translational design can MBZ's true role in neuro-oncology be defined.

Future research should also prioritize biomarker-enriched trial designs that integrate pharmacokinetic and pharmacodynamic assessments to establish clear exposure-response relationships.[82,83] Incorporating biomarkers that reflect MBZ's mechanistic targets, such as angiogenesis or Hedgehog pathway activity, could help to align patient selection and trial endpoints with the drug's underlying biology. In parallel, more radical solutions such as the rational design of molecules with reduced efflux affinity, chemical modification of the BBB or the development of targeted and bypass delivery systems could be pursued.[84] Finally, MBZ's potential as part of multimodal regimens requires further investigation, particularly through combination strategies that exploit radiosensitization, autophagy inhibition and pathway-specific vulnerabilities such as Hedgehog or mitochondrial stress responses.[31,34,42] Collectively, these efforts highlight that MBZ's successful translation will depend not only on its diverse intrinsic pharmacology, but also on resolving the pharmacokinetic and delivery challenges that currently limit reproducibility. Only through standardization, innovation and biomarker-guided design can MBZ progress from a promising repurposed agent to a clinically reliable component of brain tumour therapy.

4.6 | Summary

MBZ demonstrates broad preclinical anticancer activity in brain tumour models, including effects on microtubules, angiogenesis,

Hedgehog signalling and DNA damage-response pathways, supporting its biological plausibility as a repurposed neuro-oncology agent. However, the available clinical literature is limited and does not yet establish clear efficacy in humans. Early-phase studies indicate that high-dose oral MBZ is feasible and generally well tolerated, but observed antitumour effects have been modest, inconsistent and difficult to interpret owing to small sample sizes, heterogeneous populations, non-randomized designs and inconsistent formulation reporting. MBZ should, therefore, be regarded as a promising but unproven candidate. Future research should prioritize rigorous comparative clinical trials, transparent reporting of polymorph and formulation and integration of pharmacokinetic and biomarker data to determine whether the preclinical promise of MBZ can translate into meaningful patient benefit.

4.7 | Nomenclature of targets and ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in http://www.guidetopharmacology.org, and are permanently archived in the Concise Guide to PHARMACOLOGY 2025/26.[85–88]

AUTHOR CONTRIBUTIONS

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The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

DATA AVAILABILITY STATEMENT

Data are available upon reasonable request to the corresponding author.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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