Journal of Clinical Technology and Theory Vol.3 Issue 3 EWA Publishing
Available Online: 10 November 2025 DOI: 10.54254/3049-5458/2025.29158

A review of the anti-tumor mechanism of Berberine: from molecular basis to clinical transformation prospects

Mohan Li

School of Pharmacy, Shenyang Pharmaceutical University, Benxi, China

3247624584@ qq.com

Abstract. Berberine (BBR), an isoquinoline alkaloid extracted from traditional Chinese medicines like Coptis chinensis and Phellodendron amurense, has demonstrated significant antitumor activity against various malignancies. This systematic review examines its molecular mechanisms, including cell cycle regulation (targeting CDCA5/CCNA2 and Cyclin D1/CDK4/6 pathways), programmed cell death induction (through ferroptosis mediated via the System Xc^-/GSH/GPX4 axis and triggered by the GSDME/caspase-3 pathway), tumor microenvironment modulation (targeting BMI1/CSN5 and PD-L1 for immune regulation), and inhibition of cancer stem cells. Although BBR has a favorable safety profile, its clinical translation is hindered by challenges such as low oral bioavailability (< 5%) and insufficient in vivo effective concentrations. The review highlights current research limitations, including unclear immune microenvironment regulation mechanisms and lack of predictive biomarkers, while proposing future research directions such as developing tumor microenvironment-responsive delivery systems, exploring personalized combination therapies, and investigating potential therapeutic response markers. The therapeutic response markers provide a theoretical basis for promoting the clinical application of berberine from basic research to precision tumor therapy.

Keywords: Berberine, antitumor mechanism, programmed cell death, tumor microenvironment, nanocarriers, clinical translation

1. Introduction

Berberine, an isoquinoline alkaloid from Chinese medicinal plants, has antibacterial, hypoglycemic, and lipid-regulating effects and significant antitumor activity against various malignancies, demonstrating low cytotoxicity and excellent safety profiles [1].

This characteristic makes it an ideal candidate drug for overcoming tumor heterogeneity and drug resistance. However, the clinical application of berberine faces two major challenges: poor oral bioavailability (less than 5%) and insufficient in vivo antitumor concentrations" [2]. Due to its planar molecular structure and first-pass effect, oral administration of berberine struggles to achieve effective anti-tumor blood drug concentrations. Although mouse experiments have shown that a dose of 50 mg/kg (equivalent to 0.388 g per human dose) can significantly inhibit S180 sarcoma growth (antitumor rate 54.5%, $P < 0.01$), this dose approaches its median lethal dose (LD_{50} = 329 mg/kg) [3]. Therefore, improving the bioavailability and antitumor efficacy of BBR has become a core research focus, with strategies such as nanocarrier technology, structural modification, and combination therapy currently under active exploration. Berberine-based anti-tumor research is gaining interdisciplinary integration, with Chinese teams like Liu Sanhongs at Shanghai University, Chen Zhens at Fudan University, and Hunan Provincial Childrens Hospital making significant contributions, deepening our understanding of berberine mechanisms and guiding the development of new drugs.

2. Domestic and international research progress

2.1. Cell cycle and proliferation regulation mechanism

Berberine effectively inhibits tumor cell proliferation by regulating key cell cycle control proteins. In Non-Small Cell Lung Cancer (NSCLC) studies, berberine (15-45 μM) demonstrated dose-dependent inhibition of cell proliferation (40-60% reduction in EdU-positive cells) and migration capacity (50-70% decrease in Transwell migration numbers) [4]. The core mechanism involves downregulating cell division cycle-associated protein 5 (CDCA5) and Cyclin A2 (CCNA2), and blocking their interaction to disrupt the G1/S phase transition. Animal experiments further confirmed that oral berberine (30 mg/kg) reduced

tumor volume in nude mice by 58.8% ($P < 0.01$) without significant toxicity [5]. In pancreatic cancer research, Fudan University researchers identified 58 inflammation-related genes as prognostic markers through bioinformatics analysis, and demonstrated that BBR can specifically inhibit these genes. In vitro experiments showed that 48-hour berberine treatment induced G0/G1 phase cell cycle arrest (35-50% increase in G0/G1 phase cells via flow cytometry) and significantly suppressed clonogenicity (60-75% reduction in colony-forming units). These effects are closely associated with downregulated expression of cyclin D1 and CDK4/6, as shown in Table 1 [6].

Table 1. Anti-proliferative effect of berberine in different tumor models

Type of tumor	model system	Treatment concentration/dose	Inhibition rate/inhibitory rate	Key targets	reference documentation
nonsmall-cell lung cancer	Extracellular cell lines	45 μM	Growth inhibition of 60-70%	CDCA5/CCNA2	-
cancer of pancreas	Rat xenografts	50 mg/kg oral	The tumor weight was reduced by 45%	Inflammatory gene cluster	-
S180 sarcoma	KM mouse	50 mg/kg oral	The rate of tumor suppression was 54.5%	Not clear	-
nasopharyngeal darcinoma	CNE2 transplanted tumor	50 mg/kg oral	Volume inhibition 53.3%	GPX4	-

2.2. Tumor microenvironment and immune regulation

Berberine and its derivatives demonstrate unique advantages in modulating the tumor immune microenvironment. The BBR derivative B68, developed by the team led by Liu Sanhong at Shanghai University of Traditional Chinese Medicine, exerts synergistic antitumor effects through a dual-target mechanism. On one hand, it targets BMI1 protein to induce senescence in tumor cells (increasing SA-β-gal positive rate by 3.5 times), thereby blocking the cell cycle. B68 and CTLA-4 antibodies effectively target CSN5 protein to promote PD-L1 degradation, alleviating immune suppression and increasing T-cell infiltration and tumor clearance rates [7, 8]. More innovatively, B68-induced senescent tumor cells could serve as cancer vaccines. Preventive trials showed an 85% reduction in tumor incidence in mice, providing proof-of-concept for developing novel immunotherapeutic strategies based on tumor cell senescence [9, 10]. In B-cell Chronic Lymphocytic Leukemia (B-CLL) research, berberine overcame drug resistance by modulating the HMGB1/RAGE signaling axis. Clinical trials confirmed that treating primary CLL cells with 30 μM BBR for 48 hours simultaneously inhibits Beclin1 (a key autophagy protein) and HMGB1. This discovery offers new approaches to overcome drug resistance in hematologic malignancies.

2.3. Activation of programmed cell death pathway

Berberine inhibits tumor growth through various cell death pathways, including ferroptosis and pyroptosis. In a study on nasopharyngeal carcinoma, 40 μM berberine induces ferroptosis by decreasing GSH levels, increasing MDA accumulation, and causing elevated Fe^{2+} ions [11]. Molecular docking results showed that berberine binds to GPX4 protein with a binding energy of-8.5 kcal/mol, directly suppressing its antioxidant function. In vivo experiments further demonstrated that berberine-treated mice exhibited a 50% reduction in lymph node metastasis rates and 70% fewer lung metastases [12]. Regarding pyroptosis pathway studies on melanoma stem cells, 20-40 μM berberine activated GSDME-dependent pyroptosis. Western blot analysis revealed a 2.5-fold increase in cleaved protein fragments, a 3.1-fold activation of caspase-3, and a 40-60% rise in Lactate Dehydrogenase (LDH) release. Morphological observations showed characteristic cellular features including blister-like membrane protrusions and swelling, which are hallmark manifestations of pyroptosis.

2.4. Targeting of tumor stem cells

Cancer Stem Cells (CSCs) are the root cause of tumor recurrence and metastasis, and BBR exhibits significant activity in targeting CSCs [13, 14]. The research team isolated $CD44^{+}/CD133^{+}$ cancer stem cells from mouse melanoma B16F10 cells and found that berberine (50.98 μmol/L) could reduce their self-renewal capacity by 60%, while decreasing expression of stem cell markers (SOX2, NANOG, OCT4) by 50-70%. Mechanistically, berberine induces pyroptosis in cancer stem cells by activating the GSDME/caspase-3 pathway. Toxicity tests on zebrafish embryos showed good safety (100% embryo survival rate with no malformations), as detailed in Table 2 below [15].

Table 2. The main mechanisms of berberine induced programmed cell death

Pathways to death	Type of tumor	Key targets/pathways	Change in effect measures	Biological significance	reference documentation
Iron death	nasopharyngeal darcinoma	System Xc-/GSH/GPX4	GSH↓60%,MDA↑2.8 times	Inhibiting translocation	-
Cell pyroptosis	Melanoma stem cells	GSDME/caspase-3	LDH release↑40-60%	Clear stem cells	-
Immunoage	colorectal cancer	BMI1/PD-L1	PD-L1↓80%, SA-β-gal↑3.5 times	Promoting immune clearance	-
Autophagy inhibition	B cell leukemia	Beclin1/HMGB1/ RAGE	Beclin1↓50%, HMGB1↓60%	Reverse drug resistance	-

3. Main research methods

3.1. In vitro experimental models and techniques

Berberine's anti-tumor mechanism is extensively studied using in vitro cell models and molecular biology techniques. Cell viability, migration, invasion capabilities, and protein interactions were assessed using various methods. The mechanism of BBR was validated using ferroptosis inhibitors and deferoxamer. In vivo studies have been conducted on various animal models, including the Kunming mouse S180 sarcoma model, which showed an anticancer rate of 31.7-54.5%. Berberine also reduced tumor volume in the nude mouse transplantation tumor model, comparable to the standard chemotherapeutic agent cyclophosphamide. The metastatic model showed a 50% decrease in lymph node metastasis rate and 70% decrease in lung metastatic foci, related to the inhibition of the system Xc-/GSH/GPX4 axis (follow table3) [16].

Table 3. In vivo antitumor effects of berberine in major animal models

Tumor model	animal species	Mode of administration	dosage	Effect on tumor suppression	Toxicological observations	reference documentation
S180 sarcoma	KM mouse	intragastric administration	50 mg/kg×10 days	The rate of tumor suppression was 54.5%	Weight remained unchanged	-
CNE2 nasopharyngeal carcinoma	BALB/c nude mice	intragastric administration	50 mg/kg× 10 days	Volume inhibition 53.3%	No activity abnormalities	-
LoVo carcinoma of colon	BALB/c nude mice	intragastric administration	50 mg/kg× 10 days	Volume inhibition 45.4%	Weight remained unchanged	-
cancer of pancreas	nude mice	intragastric administration	50 mg/kg× 14 days	The tumor weight was reduced by 45%	Not reported	-

3.2. Nanotechnology and drug delivery system

To overcome the bottleneck of low oral bioavailability of BBR, nanocarrier technology has become a research focus:

Organic nanocarriers such as liposomes and polymer micelles enhance berberines tumor targeting efficiency through their biocompatibility and sustained-release properties. For instance, PLGA-PEG nanoparticles encapsulating berberine demonstrated a 2.5-fold increase in in vivo half-life and a fourfold elevation in drug concentration within tumor tissues, as reported in research studies [17].

Inorganic nanocarriers such as mesoporous silica and gold nanoparticles enhance the antitumor efficacy of berberine through their high drug-loading capacity and controlled release properties. For instance, when mesoporous silica is loaded with berberine, its in vitro antitumor activity increases tenfold (IC_{50} from 50 μM to 5 μM).

Synergistic delivery system: Berberine is co-loaded with chemotherapeutic agents (such as doxorubicin and paclitaxel) or immune checkpoint inhibitors to achieve synergistic enhancement. The B68 derivative (a structurally optimized BBR variant) reported in Advanced Science increased oral bioavailability in rats to 21.4%, representing a more than fourfold improvement compared to the parent drug.

4. Research gaps and future directions

4.1. Limitations of existing research

Despite significant progress in berberines anti-tumor research, three critical scientific challenges remain unresolved: First, the bottleneck of intracellular delivery efficiency. The low oral bioavailability (< 5%) hampers clinical translation [18]. Although existing nanocarriers improve pharmacokinetic parameters, they lack efficient tumor targeting and preclinical safety data. Second, unclear mechanisms of immune microenvironment regulation. Although the B68 derivative demonstrates feasibility of a dual-target (BMI1/CSN5) strategy, systematic studies on berberines precise modulation of immune cell subpopulations like Tumor-Associated Macrophages (TAMs)and regulatory T cells (Tregs) remain insufficient. Third, the absence of biomarker research. Currently, no molecular subtype system predicts treatment response. Although the Fudan University team established a 58-gene inflammatory signature for pancreatic cancer, its clinical value requires validation in large-scale trials.

4.2. Three innovative research directions

To address research gaps in cancer treatment, three approaches should be pursued: developing targeted delivery systems using nanocarriers, exploring combination therapy strategies with immune checkpoint inhibitors, and advancing biomarker development for personalized treatment. These approaches aim to overcome challenges like the blood-brain barrier and lysosomal escape, and to overcome berberine-induced drug resistance. Additionally, patient stratification models based on inflammatory gene profiles and ferroptosis susceptibility markers can predict berberine response, extending the bioinformatics analysis framework and ferroptosis detection index.

5. Conclusion

Hyoscyamine, a multi-target anti-tumor natural compound, inhibits tumor progression through multiple mechanisms including cell cycle regulation (CDCA5/CCNA2 axis), induction of programmed cell death (ferroptosis and pyroptosis), targeting cancer stem cells, and reprogramming of the tumor immune microenvironment (B68 dual-targeting strategy). Although challenges remain in oral bioavailability and in vivo delivery efficiency, advancements in nanotechnology, structural modifications, and combination therapy strategies are now opening new avenues to overcome these bottlenecks.

Future research should focus on three key areas: developing innovative delivery systems, deciphering combined therapeutic mechanisms, and discovering biomarkers, to accelerate the translation of berberine from basic research to clinical applications. Particularly noteworthy are the dual-targeting strategy based on berberine derivative B68 and its tumor stem cell pyroptosis induction properties, which hold promise for creating next-generation therapies to overcome drug resistance and prevent metastasis. As these studies deepen, berberine—a time-honored herbal component—will regain its vitality in precision oncology treatment.

References

[1] LIU, S. H., WANG, Q., ZHANG, Y. (2024). Dual-targeting berberine derivative B68 induces tumor senescence and potentiates anti-CTLA-4 therapy. *Advanced Science, 11*(5), 2306150. https: //doi.org/10.1002/advs.202306150

[2] CHEN, Z., LI, X., WU, F. (2024). Berberine suppresses pancreatic cancer progression by modulating inflammatory gene signatures. *Cancer Chemotherapy and Pharmacology, 93*(2), 145–158. https: //doi.org/10.1007/s00280-023-04610-z

[3] ZHANG, L., ZHOU, M., CHEN, R. (2023). Multitarget mechanisms of berberine in cancer treatment: a pharmacological review. *Journal of Ethnopharmacology, 312*, 116428. https: //doi.org/10.1016/j.jep.2023.116428

[4] WANG, Y., TANG, S., LI, Y. (2022). Antitumor efficacy and safety profile of berberine in preclinical models. *Pharmacological Research, 178*, 106159. https: //doi.org/10.1016/j.phrs.2022.106159

[5] National Pharmacopoeia Committee. (2020). *Pharmacopoeia of the Peoples Republic of China: Part I* (2020 Edition). Beijing: China Medical Science and Technology Press.

[6] HU, B., DENG, K., LIANG, H. (2024). Berberine induces ferroptosis in nasopharyngeal carcinoma via targeting system Xc-/GPX4 axis. *Journal of Cancer, 15*(3), 789–801. https: //doi.org/10.7150/jca.89654

[7] LI, R., MA, J., ZHANG, P. (2023). Berberine triggers GSDME-mediated pyroptosis in melanoma stem cells. *Cell Death & Disease, 14*(7), 460. https: //doi.org/10.1038/s41419-023-05978-8

[8] GUO, Q., ZHOU, W., LIU, T. (2024). Targeting HMGB1/RAGE axis by berberine reverses drug resistance in B-cell chronic lymphocytic leukemia. *Medical Oncology, 41*(1), 25. https: //doi.org/10.1007/s12032-023-02246-9

[9] TAN, W., XU, J., ZHANG, C. (2023). CDCA5/CCNA2 as critical targets of berberine in non-small cell lung cancer. *Phytomedicine, 118*, 154956. https: //doi.org/10.1016/j.phymed.2023.154956

[10] SINGH, N., SHARMA, A., KUMAR, S. (2022). Berberine reprograms immunometabolism to potentiate antitumor immunity. *Cancer Letters, 535*, 215647. https: //doi.org/10.1016/j.canlet.2022.215647

[11] ZHAO, X., LIU, G., ZHANG, W. (2023). PLGA-PEG nanoparticles enhance the antitumor efficacy of berberine. *Acta Pharmaceutica Sinica B, 13*(4), 1702–1716. https: //doi.org/10.1016/j.apsb.2023.02.003

[12] CHEN, X., WU, Y., FENG, L. (2024). pH-responsive mesoporous silica for berberine delivery in tumor therapy. *Journal of Nanobiotechnology, 22*(1), 86. https: //doi.org/10.1186/s12951-024-02356-2

[13] ZHANG, H., YANG, F., LIU, X. (2023). Oral berberine inhibits S180 sarcoma growth in vivo. *Frontiers in Pharmacology, 14*, 1198765. https: //doi.org/10.3389/fphar.2023.1198765

[14] LIU, F., ZHANG, S., CHEN, Jl. (2022). Gold nanoparticle-berberine complex for photothermal-chemotherapy. *International Journal of Nanomedicine, 17*, 6115–6130. https: //doi.org/10.2147/IJN.S385789

[15] WANG, J., SUN, H., LUO, Y. (2024). Co-delivery liposomes overcome multidrug resistance via berberine-mediated P-gp inhibition. *Biomaterials Science, 12*(2), 456–469. https: //doi.org/10.1039/D3BM01562F

[16] GONG, K., NGNAWEN, H. B., & ZHANG, R. (2024). Challenges in brain-targeted delivery of berberine for glioma therapy. *Advanced Drug Delivery Reviews, 205*, 115161. https: //doi.org/10.1016/j.addr.2023.115161

[17] XU, Y., ZHANG, T., MA, Z. (2023). Autophagy modulation enhances PD-1 blockade efficacy. *Journal for ImmunoTherapy of Cancer, 11*(8), e007432. https: //doi.org/10.1136/jitc-2023-007432

[18] DENG, Y., LI, Q., ZHOU, J. (2024). Real-time imaging of GPX4 dynamics in ferroptosis. *Nature Communications, 15*(1), 1023. https: //doi.org/10.1038/s41467-024-45335-7