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EDITORIAL

Zhang SK, Jiang L, Jiang CL, Cao Q, Chen YQ, Chi H. Unveiling genetic susceptibility in esophageal squamous cell carcinoma and revolutionizing pancreatic cancer diagnosis through imaging. *World J Gastrointest Oncol* 2025; 17(6): 102544 [DOI: [10.4251/wjgo.v17.i6.102544](https://doi.org/10.4251/wjgo.v17.i6.102544)]

Li DH, Qiao C, Han YT, Ge JL. Kinesin family member 14 in digestive tract malignancies: Oncogenic mechanisms, clinical implications, and therapeutic prospects. *World J Gastrointest Oncol* 2025; 17(6): 105062 [DOI: [10.4251/wjgo.v17.i6.105062](https://doi.org/10.4251/wjgo.v17.i6.105062)]

Quan MJ, Lin Q. Prognostic value of post-neoadjuvant immunochemotherapy hypercoagulation in gastric cancer patients undergoing surgery. *World J Gastrointest Oncol* 2025; 17(6): 105085 [DOI: [10.4251/wjgo.v17.i6.105085](https://doi.org/10.4251/wjgo.v17.i6.105085)]

REVIEW

Jia C, Lang QF, Yin ZJ, Sun J, Meng QH, Pei TM. Role, mechanism, and application of N6-methyladenosine in hepatobiliary carcinoma. *World J Gastrointest Oncol* 2025; 17(6): 105140 [DOI: [10.4251/wjgo.v17.i6.105140](https://doi.org/10.4251/wjgo.v17.i6.105140)]

Mo L, Wang Y, Liang XY, Zou T, Chen Y, Tan JY, Wen J, Jian XH. Progress of traditional Chinese medicine in the prevention and treatment of colorectal cancer. *World J Gastrointest Oncol* 2025; 17(6): 105690 [DOI: [10.4251/wjgo.v17.i6.105690](https://doi.org/10.4251/wjgo.v17.i6.105690)]

Zhang Z, Wang HM, Xu ZX, Luan WY, Lin SX, Miao YD. Application of single-cell sequencing in the study of immune cell infiltration in inflammatory bowel disease and colorectal cancer. *World J Gastrointest Oncol* 2025; 17(6): 107382 [DOI: [10.4251/wjgo.v17.i6.107382](https://doi.org/10.4251/wjgo.v17.i6.107382)]

MINIREVIEWS

Marrelli D, Carbone L, Poto GE, Fusario D, Gjoka M, Andreucci E, Piccioni SA, Calomino N, Sandini M, Roviello F. Minimally invasive lymphadenectomy for gastric cancer: Could the robotic approach provide any benefits than laparoscopy? *World J Gastrointest Oncol* 2025; 17(6): 104015 [DOI: [10.4251/wjgo.v17.i6.104015](https://doi.org/10.4251/wjgo.v17.i6.104015)]

Zeng XF, Wang YW, Ou Y, Liu L. Role of myosin heavy chain 9 in gastrointestinal tumorigenesis: A comprehensive review. *World J Gastrointest Oncol* 2025; 17(6): 106617 [DOI: [10.4251/wjgo.v17.i6.106617](https://doi.org/10.4251/wjgo.v17.i6.106617)]

Massaro G, Paulet A, Lavacchi D, Brugia M, Rossini D, Giommoni E, Catalano M, Pillozzi S, Antonuzzo L, Roviello G. Immune checkpoint inhibitors in the first-line treatment of esophageal squamous cell carcinoma: Minireview for a big shift. *World J Gastrointest Oncol* 2025; 17(6): 106707 [DOI: [10.4251/wjgo.v17.i6.106707](https://doi.org/10.4251/wjgo.v17.i6.106707)]

Akkari I, Akkari H, Harbi R. Artificial intelligence to predict hepatocellular carcinoma risk in cirrhosis. *World J Gastrointest Oncol* 2025; 17(6): 107414 [DOI: [10.4251/wjgo.v17.i6.107414](https://doi.org/10.4251/wjgo.v17.i6.107414)]

Tang R, Qi SQ, Zhang T, Pan ZB, Xu JH. Progress in the study of therapeutic strategies for hepatoblastoma in children. *World J Gastrointest Oncol* 2025; 17(6): 107700 [DOI: [10.4251/wjgo.v17.i6.107700](https://doi.org/10.4251/wjgo.v17.i6.107700)]

ORIGINAL ARTICLE

Retrospective Cohort Study

Li YH, Qian GX, Yao L, Lei XD, Zhu Y, Tang L, Xu ZL, Bu XY, Wei MT, Lu JL, Jia WD. Preoperative model for predicting early recurrence in hepatocellular carcinoma patients using radiomics and deep learning: A multicenter study. *World J Gastrointest Oncol* 2025; 17(6): 106608 [DOI: [10.4251/wjgo.v17.i6.106608](https://doi.org/10.4251/wjgo.v17.i6.106608)]

Alpsoy A, Yavuz A, Simsek K, Altunay B, Karaca M, Unal B, Bassorgun CI, Tatli AM, Elpek GO. Evaluation of tumor budding, desmoplastic reaction, and lymphocytic infiltration in predicting survival for pancreatic ductal adenocarcinoma. *World J Gastrointest Oncol* 2025; 17(6): 107021 [DOI: [10.4251/wjgo.v17.i6.107021](https://doi.org/10.4251/wjgo.v17.i6.107021)]

Jung HN, Heo JH, Roh E, Kim BJ, Lee M, Kim JK, Kim JH, Han B, Han KD, Kang JG, Lee SJ, Ihm SH. Risk of hepatocellular carcinoma according to body mass index and waist circumference in nonalcoholic fatty liver disease. *World J Gastrointest Oncol* 2025; 17(6): 107364 [DOI: [10.4251/wjgo.v17.i6.107364](https://doi.org/10.4251/wjgo.v17.i6.107364)]

Retrospective Study

Liu QQ, Li YD, Chen JX, Zhang LL, Guan RC, Zhao W, Meng LY. Prognostic value of preoperative fibrinogen, neutrophil-to-lymphocyte ratio, serum alpha-fetoprotein, and prealbumin for patients with primary liver cancer undergoing transarterial chemoembolization. *World J Gastrointest Oncol* 2025; 17(6): 103198 [DOI: [10.4251/wjgo.v17.i6.103198](https://doi.org/10.4251/wjgo.v17.i6.103198)]

Gong X, Chen C, Shen JF. Gastric cancer in children infected with *Helicobacter pylori*. *World J Gastrointest Oncol* 2025; 17(6): 103632 [DOI: [10.4251/wjgo.v17.i6.103632](https://doi.org/10.4251/wjgo.v17.i6.103632)]

Qiu TH, Wen HY, Huang YL. Significance of hemoglobin and hematocrit changes in predicting patient survival and efficacy of neoadjuvant chemotherapy for advanced gastric cancer. *World J Gastrointest Oncol* 2025; 17(6): 104592 [DOI: [10.4251/wjgo.v17.i6.104592](https://doi.org/10.4251/wjgo.v17.i6.104592)]

Yang ZH, Liu SN, Chu FY, Yang C, Chen X. Efficacy and safety of donafenib plus transarterial chemoembolization and immunotherapy for hepatocellular carcinoma. *World J Gastrointest Oncol* 2025; 17(6): 105170 [DOI: [10.4251/wjgo.v17.i6.105170](https://doi.org/10.4251/wjgo.v17.i6.105170)]

Chen CR, Jin HL, Xu QJ, Yuan YL, Hu ZH, Liu Y, Lei HK. Development and validation of a nomogram for predicting postoperative venous thromboembolism risk in patients with hepatocellular carcinoma. *World J Gastrointest Oncol* 2025; 17(6): 105790 [DOI: [10.4251/wjgo.v17.i6.105790](https://doi.org/10.4251/wjgo.v17.i6.105790)]

Xu J, Li SX, Liu D, Chen LX, Chen X. Diagnostic value of serum pepsinogen, gastrin, and carbohydrate antigens in gastric ulcer and gastric cancer. *World J Gastrointest Oncol* 2025; 17(6): 105931 [DOI: [10.4251/wjgo.v17.i6.105931](https://doi.org/10.4251/wjgo.v17.i6.105931)]

Qiao SB, Niu ML, Liang WT, Zhang LJ, Chen X, Zhu YK. Analysis of serum S100A12, soluble advanced glycation end products receptor, and gut microbiome in elderly patients with colorectal cancer. *World J Gastrointest Oncol* 2025; 17(6): 106393 [DOI: [10.4251/wjgo.v17.i6.106393](https://doi.org/10.4251/wjgo.v17.i6.106393)]

Zeynelgil E, Duzkopru Y, Kocanoglu A, Karakaya S. Prognostic value of hemoglobin-to-red cell distribution width ratio and inflammation markers in colorectal cancer. *World J Gastrointest Oncol* 2025; 17(6): 106603 [DOI: [10.4251/wjgo.v17.i6.106603](https://doi.org/10.4251/wjgo.v17.i6.106603)]

Yang ZY, Wan WN, Zhao L, Li SN, Liu Z, Sang L. Noninvasive prediction of Ki-67 expression in pancreatic cancer via contrast-enhanced ultrasound quantitative parameters: A diagnostic model study. *World J Gastrointest Oncol* 2025; 17(6): 107919 [DOI: [10.4251/wjgo.v17.i6.107919](https://doi.org/10.4251/wjgo.v17.i6.107919)]

Yao ZY, Liu J, Ma X, Li WT, Shen Y, Cui YZ, Fang Y, Han ZX, Yang CH. Impact of fibrinogen-to-albumin ratio on the long-term prognosis of patients with advanced HER2-negative gastric cancer receiving immunochemotherapy. *World J Gastrointest Oncol* 2025; 17(6): 107980 [DOI: [10.4251/wjgo.v17.i6.107980](https://doi.org/10.4251/wjgo.v17.i6.107980)]

Randomized Controlled Trial

Dai YH, Zhuo-Ma SL, Luo YH. Effect of gastrointestinal endoscopy center care on the psychological state and pain level of colorectal cancer patients. *World J Gastrointest Oncol* 2025; 17(6): 106154 [DOI: [10.4251/wjgo.v17.i6.106154](https://doi.org/10.4251/wjgo.v17.i6.106154)]

Basic Study

Gao Z, Zhang X, He H. Role and mechanism of sarcosine dehydrogenase in the progression of gallbladder cancer through chemokine pathways. *World J Gastrointest Oncol* 2025; 17(6): 105016 [DOI: [10.4251/wjgo.v17.i6.105016](https://doi.org/10.4251/wjgo.v17.i6.105016)]

Wang L, Wang MH, Yuan YH, Xu RZ, Bai L, Wang MZ. Identification and validation of extracellular matrix-related genes in the progression of gastric cancer with intestinal metaplasia. *World J Gastrointest Oncol* 2025; 17(6): 105160 [DOI: [10.4251/wjgo.v17.i6.105160](https://doi.org/10.4251/wjgo.v17.i6.105160)]

Xu W, Li W, Ru J. Endothelial Per-Arnt-Sim domain-containing protein 1 expression is correlated with poor prognosis and promotes invasion and metastasis in gastric cancer. *World J Gastrointest Oncol* 2025; 17(6): 105213 [DOI: [10.4251/wjgo.v17.i6.105213](https://doi.org/10.4251/wjgo.v17.i6.105213)]

Wang ZW, Pan CY, Wei CL, Liao H, Zhang XP, Zhang CY, Yu L. Oxocresbanine inhibits the proliferation of hepatocellular carcinoma cells by promoting apoptosis and autophagy. *World J Gastrointest Oncol* 2025; 17(6): 105570 [DOI: [10.4251/wjgo.v17.i6.105570](https://doi.org/10.4251/wjgo.v17.i6.105570)]

Hua YQ, Guo KX, Ni P, Wang D, An TY, Gao YY, Zhang RG. RPF2 regulates the protein kinase B/mammalian target of rapamycin pathway in the pathogenesis of *Helicobacter pylori*. *World J Gastrointest Oncol* 2025; 17(6): 105664 [DOI: [10.4251/wjgo.v17.i6.105664](https://doi.org/10.4251/wjgo.v17.i6.105664)]

Dong YD, Wu XM, Liu WQ, Hu YW, Zhang H, Fang WD, Luo Q. Potential mechanism of *Camellia luteoflora* against colon adenocarcinoma: An integration of network pharmacology and molecular docking. *World J Gastrointest Oncol* 2025; 17(6): 105782 [DOI: [10.4251/wjgo.v17.i6.105782](https://doi.org/10.4251/wjgo.v17.i6.105782)]

Wei JR, Ouyang YN, Tang MT, Yuan JZ, Wang PL, Jiang LH, Wu LC. Charged multivesicular body protein 7 was identified as a prognostic biomarker correlated with metastasis in colorectal cancer. *World J Gastrointest Oncol* 2025; 17(6): 105967 [DOI: [10.4251/wjgo.v17.i6.105967](https://doi.org/10.4251/wjgo.v17.i6.105967)]

Wang YT, Yong YL, Liu ZK, Shen YX, Yang XM, Chen ZN. Regulator of chromosome condensation 1 promotes hepatocellular carcinoma proliferation via cell-division-cycle-associated-8 dependent phosphoinositide 3-kinase/protein kinase B signaling. *World J Gastrointest Oncol* 2025; 17(6): 106080 [DOI: [10.4251/wjgo.v17.i6.106080](https://doi.org/10.4251/wjgo.v17.i6.106080)]

Feng CZ, Zhong SQ, Ye SW, Zheng Z, Sun H, Zhou SH. Tumor-derived exosomal miR-425-5p and miR-135b-3p enhance colorectal cancer progression through immune suppression and vascular permeability promotion. *World J Gastrointest Oncol* 2025; 17(6): 106161 [DOI: [10.4251/wjgo.v17.i6.106161](https://doi.org/10.4251/wjgo.v17.i6.106161)]

META-ANALYSIS

Zhang W, Fu H, Liu ZR, Xu L, Che X, Ning YT, Zhan ZY, Zhou GC. Transarterial chemoembolization combined with lenvatinib vs transarterial chemoembolization combined with sorafenib for unresectable hepatocellular carcinoma: A systematic review and meta-analysis. *World J Gastrointest Oncol* 2025; 17(6): 105887 [DOI: [10.4251/wjgo.v17.i6.105887](https://doi.org/10.4251/wjgo.v17.i6.105887)]

SCIENTOMETRICS

Shang LQ, Guo HX, Wang P, Sun XH, You JQ, Ma JT, Wang LK, Liu JX, Wang ZQ, Shao HB. Global scientific trends on hepatocellular carcinoma research from 2004 to 2023: A bibliometric and visualized analysis. *World J Gastrointest Oncol* 2025; 17(6): 105781 [DOI: [10.4251/wjgo.v17.i6.105781](https://doi.org/10.4251/wjgo.v17.i6.105781)]

CASE REPORT

Xu XX, Gao YH, Du CZ, Gao XX, Chen P, Fan RF, Li HT, Qiao Z. Molecular surveillance-informed personalized multidisciplinary therapy achieves prolonged survival in a patient with Lynch syndrome-associated colorectal cancer: A case report. *World J Gastrointest Oncol* 2025; 17(6): 106316 [DOI: [10.4251/wjgo.v17.i6.106316](https://doi.org/10.4251/wjgo.v17.i6.106316)]

Suh JW, Jo JW, Park DG. Long-term survival of a patient with colorectal cancer with peritoneal carcinomatosis and low completeness of cytoreduction score: A case report. *World J Gastrointest Oncol* 2025; 17(6): 106846 [DOI: [10.4251/wjgo.v17.i6.106846](https://doi.org/10.4251/wjgo.v17.i6.106846)]

Jin CB, Li YS, Zhang J, Wu J, Tao WJ. Extensive bone metastases from an occult gastric primary: A case report. *World J Gastrointest Oncol* 2025; 17(6): 106848 [DOI: [10.4251/wjgo.v17.i6.106848](https://doi.org/10.4251/wjgo.v17.i6.106848)]

Hu RR, Liu M, Li HY. Primary squamous cell carcinoma of the descending colon with pancreatic metastasis: A case report. *World J Gastrointest Oncol* 2025; 17(6): 107272 [DOI: [10.4251/wjgo.v17.i6.107272](https://doi.org/10.4251/wjgo.v17.i6.107272)]

Kong XL, Lu XW, Dong SQ, Liu J, Zheng L, Chen LL, An ZH, Gao LM, Cao JL. Descending duodenal adenocarcinoma treated with pembrolizumab resulting in complete clinical response: A case report and literature review. *World J Gastrointest Oncol* 2025; 17(6): 107568 [DOI: [10.4251/wjgo.v17.i6.107568](https://doi.org/10.4251/wjgo.v17.i6.107568)]

LETTER TO THE EDITOR

Cai Z, Dai WD, Cai XW. Prognostic impact of inflammatory and nutritional markers in colorectal cancer patients with peritoneal metastasis. *World J Gastrointest Oncol* 2025; 17(6): 100010 [DOI: [10.4251/wjgo.v17.i6.100010](https://doi.org/10.4251/wjgo.v17.i6.100010)]

Kumar S. Esophageal squamous cell carcinoma susceptibility of activin A receptor type 1C variants in Chinese population. *World J Gastrointest Oncol* 2025; 17(6): 102687 [DOI: [10.4251/wjgo.v17.i6.102687](https://doi.org/10.4251/wjgo.v17.i6.102687)]

Arora J, Nassar M, Baraka B. Unravelling the potential of plasma DNA methylation in the detection and surveillance of esophageal cancer. *World J Gastrointest Oncol* 2025; 17(6): 103333 [DOI: [10.4251/wjgo.v17.i6.103333](https://doi.org/10.4251/wjgo.v17.i6.103333)]

Bae SU. Elevating surgical standards: The role of intraperitoneal isoperistaltic side-to-side anastomosis in colon cancer surgery. *World J Gastrointest Oncol* 2025; 17(6): 106645 [DOI: [10.4251/wjgo.v17.i6.106645](https://doi.org/10.4251/wjgo.v17.i6.106645)]

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AIMS AND SCOPE

The primary aim of *World Journal of Gastrointestinal Oncology* (WJGO, *World J Gastrointest Oncol*) is to provide scholars and readers from various fields of gastrointestinal oncology with a platform to publish high-quality basic and clinical research articles and communicate their research findings online.

WJGO mainly publishes articles reporting research results and findings obtained in the field of gastrointestinal oncology and covering a wide range of topics including liver cell adenoma, gastric neoplasms, appendiceal neoplasms, biliary tract neoplasms, hepatocellular carcinoma, pancreatic carcinoma, cecal neoplasms, colonic neoplasms, colorectal neoplasms, duodenal neoplasms, esophageal neoplasms, gallbladder neoplasms, *etc.*

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Basic Study

Oxocrebaine inhibits the proliferation of hepatocellular carcinoma cells by promoting apoptosis and autophagy

Zheng-Wen Wang, Cai-Yan Pan, Cang-Long Wei, Hui Liao, Xiao-Po Zhang, Cai-Yun Zhang, Lei Yu

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Abstract

BACKGROUND

Finding active lead anti-hepatocellular carcinoma compounds from traditional Chinese medicine has important research value.

AIM

To assess the detailed mechanism of oxocrebaine, a compound separated from the traditional Chinese medicinal plant *Stephania hainanensis* H.S. Lo et Y. Tsoong, and to evaluate its inhibition of the proliferation of human hepatocellular carcinoma cells *via* apoptosis and autophagy.

METHODS

MTT, BrdU labeling, and colony formation assays were used to assess the inhibitory effect of oxocrebaine on the growth and proliferation of human hepatocellular carcinoma Hep3B2.1-7 cells. Flow cytometry was used to detect the effect of oxocrebaine on the apoptosis of Hep3B2.1-7 cells. Western blotting was used to assess the expression of apoptosis-related proteins in Hep3B2.1-7 cells. The aforementioned methods were also used to evaluate the effects of oxocrebaine on cell proliferation, autophagy markers, and autophagy-related protein expression levels after adding autophagy inhibitor 3-mA. Furthermore, to verify

the anti-hepatocellular carcinoma effect of oxocrebanine *in vivo*, a nude mouse model was used to investigate the inhibitory effect of oxocrebanine treatment and its mechanism. Apoptosis was detected using a TUNEL assay and the expression of microtubule-associated protein 1 LC3 in tumor specimens was assessed using immunohistochemistry.

RESULTS

Oxocrebanine effectively inhibited the growth of Hep3B2.1-7 cells, whilst upregulating the protein expression of cleaved caspase-3, downregulating poly(ADP-ribose) polymerase 1 protein expression, increasing the levels of Bax and Bcl-2 antagonist/killer 1 protein expression, and decreasing the levels of Bcl-2 and myeloid cell leukemia 1 protein expression, which could promote apoptosis in Hep3B2.1-7 cells. Oxocrebanine promoted the transformation of LC3-I to LC3-II in Hep3B2.1-7 cells, suggesting the occurrence of autophagy, whilst the autophagy inhibitor 3-MA could reverse this process. Oxocrebanine was also shown to reduce the phosphorylation levels of the eukaryotic translation initiation factor 4EBP1 and ribosomal protein S6 kinase B1 (P70S6K), two downstream effector molecules in the PI3K/Akt/mTOR pathway, inducing autophagy in Hep3B2.1-7 cells. Moreover, the tumor-bearing nude mouse experiment indicated that oxocrebanine effectively inhibited the growth of Hep3B2.1-7 cells *in vivo*. The results of the TUNEL assay and immunohistochemistry also revealed that oxocrebanine induced apoptosis *in vivo* and increased the expression level of LC3, an autophagy marker.

CONCLUSION

Oxocrebanine can inhibit the proliferation of human hepatocellular carcinoma cells by promoting apoptosis and inducing autophagy *in vitro* and *in vivo*.

Key Words: *Stephania hainanensis* H.S. Lo et Y. Tsoong; Oxocrebanine; Hep3B2.1-7 cells; Apoptosis; Autophagy

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Core Tip: The present study revealed that oxocrebanine can inhibit the proliferation of hepatocellular carcinoma cells by promoting apoptosis and inducing autophagy both *in vitro* and *in vivo*. Oxocrebanine can induce apoptosis in Hep3b2.1-7 cells by upregulating cleaved caspase-3, Bax, and Bak protein expression levels and downregulating PARP1, Bcl-2, and Mcl-1 protein expression levels. The inhibition of Hep3b2.1-7 cell proliferation by oxocrebanine may be related to the induction of protective autophagy. The results of the TUNEL assay and immunohistochemistry also revealed that oxocrebanine induced apoptosis *in vivo* and increased the expression level of LC3, an autophagy marker.

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INTRODUCTION

Hepatocellular carcinoma (HCC) is a primary hepatoma with a high mortality rate, accounting for 75%-85% of primary hepatomas. Globally, China ranks first in hepatoma incidence[1-3]. Studies have reported that the 5-year average survival rate for patients with hepatoma in China is only 12.1%[4,5]. To achieve the goal set in the "Healthy China 2030" plan of increasing the overall 5-year cancer survival rate by 15%[6], it is of significant social importance to find highly effective and low-toxicity anti-HCC drugs. At present, the drugs used to treat HCC in clinical practice mainly include targeted drugs, chemotherapy drugs, and antiviral drugs. Although the above-mentioned drugs have demonstrated some therapeutic effects in clinical practice, challenges such as the low success rate of gene typing for targeted drugs, drug resistance, and toxic side effects remain significant concerns, especially in combination therapy, which can cause severe liver toxicity in patients with HCC[7-9]. Therefore, it is urgent to develop low toxicity and high efficiency anti-HCC drugs, especially by seeking lead compounds or structural optimization from traditional Chinese medicine as a breakthrough point. Exploring anti-HCC active lead compounds from traditional Chinese medicines is one effective path to treat HCC. The Li Chinese medicinal plant, *Stephania hainanensis* H.S. Lo et Y. Tsoong, commonly used in the Hainan Li ethnic group, is known for its analgesic, anti-swelling, and detoxifying effects, with high efficacy and low side effects. Furthermore, our previous research demonstrated that the Hainan *Dichroa febrifuga* compound oxocrebanine, which has a planar molecular structure, exerts dual inhibitory effects on topoisomerase and tubulin[10,11]. The present study aimed to assess the growth inhibitory effect of oxocrebanine on HCC cells and evaluate whether the inhibition is related to apoptosis and autophagy. Additionally, the present study aimed to assess the changes in apoptosis and autophagy-related pathway proteins following oxocrebanine treatment to elucidate the mechanism by which oxocrebanine induces apoptosis and autophagy *in vitro* and *in vivo*, in order to provide direction and evidence for the development of new

drugs for the treatment of HCC.

MATERIALS AND METHODS

MTT assay

Hep3B2.1-7 cells (Purchased from Zhejiang Nobo Biological Products Co., Ltd) were cultured with RPMI-1640 (Meilun Bio) for 24 hours. Treatment groups were administered oxocrebanine solutions at concentrations of 5, 10, 20, 40, and 80 $\mu\text{mol/L}$, while the control group received complete medium alone (Meilun Bio). Drug concentrations were selected based on preliminary experiments to ensure a dose-response range from slight to significant inhibition. Each concentration of oxocrebanine was assessed in six replicates. After 72 hours of cell culture, 10 μL of MTT solution (5 mg/mL) was added to each well. After 4 hours of incubation, the MTT solution was discarded and 150 μL of DMSO was added to each well. The plate was shaken for 10 minutes, and the optical density (OD) value was read using a microplate reader at a wavelength of 490 nm to calculate the cell inhibition rate. The experiment was repeated three times. The cell inhibition rate (%) was calculated as [(mean OD value of control group - mean OD value of treatment group)/mean OD value of control group] \times 100.

BrdU assay

Using the same experimental groups and drug concentrations as those in the MTT assay, cell proliferation was assessed per the BrdU Proliferation Assay Kit (Frdbio Bio) protocol. The cell proliferation rate (%) was calculated as (mean OD value of BrdU positive group/mean OD value of BrdU negative group) \times 100.

Plate colony formation assay

Hep3B2.1-7 cells were treated for 24 hours. Treatment groups were administered oxocrebanine solutions at concentrations of 25.45, 50.89, and 101.79 $\mu\text{mol/L}$, while the control group was administered 32.54 $\mu\text{mol/L}$ VP16 solution (Shanghai Tongtian Biotechnology Co., Ltd) and cultured in 1 mL complete culture medium (Meilun Bio). After 72 hours of drug treatment, the solutions were discarded, the culture medium was replaced with fresh medium, and the cells were further cultured for 14 days in an incubator. Thereafter, the cells were washed with PBS, fixed with 4% paraformaldehyde for 1 hour, stained with crystal violet for 25 minutes, washed again with PBS, air-dried, and imaged.

PI/Annexin V double staining

Hep3B2.1-7 cells were treated for 24 hours. Treatment groups were administered oxocrebanine solutions at concentrations of 25.45, 50.89, and 101.79 $\mu\text{mol/L}$, while the control group was administered 32.54 $\mu\text{mol/L}$ VP16 solution. After 48 hours of incubation, the cells were collected, resuspended in 200 μL of Binding Buffer, and stained with 10 μL of FITC-labeled Annexin V and 5 μL of PI staining solution. The cells were incubated in the dark for 15 minutes and then analyzed using flow cytometry.

Western blot analysis

After 24 hours of Hep3B2.1-7 cell culture, the treatment groups and drug concentrations were the same as in PI/Annexin V double staining. After 72 hours, proteins were extracted and quantified. Protein samples were subjected to 10% SDS-PAGE, transferred to PVDF membranes, blocked with 5% skim milk for 2 hours, and incubated overnight at 4 $^{\circ}\text{C}$ with primary antibodies against GAPDH or β -actin, c-caspase3, PARP1, Bcl-2, Bax, Bak and Mcl-1 (Beijing Boaoson Biotechnology Co., Ltd, bs-0755R, bs-2076R, bs-0032R, bs-0081R, bs-20763R, bs-0127R, bs-23315R). The membranes were washed three times with TBST, incubated with HRP secondary antibodies (Beijing Boaoson Biotechnology Co., Ltd, bs-40295G) for 2 hours, washed three times with TBST, and visualized using an ECL luminescence imaging system (General Electric Company of the United States, ImageQuant LAS500). Protein bands were semi-quantitatively analyzed using Image J software (National Institutes of Health).

MTT and BrdU labeling assays in the presence of an autophagy inhibitor

After 24 hours of Hep3B2.1-7 cell culture, the oxocrebanine + 3-MA group was treated simultaneously with the autophagy inhibitor 3-MA (Selleck) at 4 mmol/L and 50.89 $\mu\text{mol/L}$ oxocrebanine. The control group was cultured in RPMI 1640 medium (Meilun Bio) with 10% fetal bovine serum (Hangzhou Sijiqing Bioengineering Materials Co., Ltd). MTT and BrdU labeling assays were performed as previously described.

Western blot analysis after addition of an autophagy inhibitor and oxocrebanine

The same experimental doses and group assignments used in the aforementioned MTT and BrdU labeling assays in the presence of an autophagy inhibitor were conducted, and Western blot was performed as previously described. The antibodies used included those against GAPDH, microtubule-associated protein 1 LC3-I, and LC3-II (ab192890; Abcam), as well as the eukaryotic translation initiation factor 4EBP1, p-4EBP1, P70S6K, and p-P70S6K (bs-2559R, bs-3018R, bs-5671R, and bs-1426R, respectively; Beijing Boaoson Biotechnology Co., Ltd).

In vivo experiments

A total of 5×10^6 Hep3B2.1-7 cells in the log growth phase in 200 μL suspension were subcutaneously injected into the dorsal flank of male immunodeficient mice (BALB/cNu/Nu), aged 8-10 weeks. The nude mice ($n = 10$) were randomly

divided into either a control group or a treatment group, with five mice in each group. The mice were fed in specific-pathogen-free animal rooms, and their feed and drinking water were strictly sterilized using high-pressure and high-temperature sterilization methods. During the breeding process, direct contact between human skin and nude mice was avoided, and all items and utensils that came into contact with the nude mice were strictly disinfected. If any animals developed a serious illness or other health problems, such as infectious diarrhea or mousepox, and were unsuitable for the experiments, they were euthanized. Furthermore, the experimental animals were euthanized if the tumor size reached 2 cm. The experiment on the nude mice and related experimental protocols adhered to the 3R principle and were approved by the Medical Ethics Committee of Hainan Medical University (approval No. HYLL-2024-146). When the tumor volume increased to approximately 50 mm³, the nude mice were divided into control and treatment groups (10 mg/kg oxocrebanine). Each group contained five nude mice according to statistical requirements. All drugs were administered intraperitoneally every 3 days for five times. The mice were then observed for 3 days, during which tumor volume was measured every 3 days to plot tumor growth curves for each group. At the end of the observation, each group of nude mice were anesthetized by an intraperitoneal injection of 300 µL of 2% pentobarbital sodium (60 mg/kg), according to pharmacological experimental methodology[12] and literature report[13]. Subsequently, the tumors were imaged using a small animal *in vivo* live imaging system. The nude mice were then euthanized using 30% vol/minute CO₂ (40 cm × 30 cm × 25 cm box), followed by cervical dislocation to confirm death, ensuring that the nude mice quickly lost vital signs and avoiding other animals from seeing the process. Subsequently, the tumor tissues were removed intact, fixed using 4% paraformaldehyde, paraffin embedded, sectioned, and assessed for apoptosis using a TUNEL assay. Expression of the autophagy signature protein LC3 was also evaluated using immunohistochemical staining. In the same way of modeling and administration, another 10 nude mice were divided into control and treatment groups to record the survival time of the tumor-bearing nude mice and generate Kaplan-Meier survival curves. At the end of the experiment, all surviving nude mice were euthanized.

Statistical analysis

The data are presented as the mean ± SD from three independent experiments. Statistical analyses were performed using SPSS 21.0 for Windows (IBM Corp.). One-sided analysis of variance followed by the Fisher's least significant difference test was used to analyze the data from different groups. $P < 0.05$ was considered to indicate a statistically significant difference.

RESULTS

Inhibitory effect of oxocrebanine on proliferation of Hep3B2.1-7 cells

MTT, BrdU labeling, and colony formation assays were used to assess the effect of oxocrebanine on the growth and proliferation of Hep3B2.1-7 cells. The results of the MTT assay are presented in Table 1. After treatment of Hep3B2.1-7 cells with oxocrebanine, the inhibition rate increased significantly in a dose-dependent manner ($P < 0.01$) compared with the control group, and the IC₅₀ value was calculated to be 50.89 µmol/L. The BrdU labeling assay results indicated that the cell proliferation level decreased significantly ($P < 0.01$), which also showed a dose-dependent relationship (Table 2). Furthermore, plate colony formation in Hep3B2.1-7 cells decreased significantly after treatment with oxocrebanine ($P < 0.01$; Table 3 and Figure 1). The results of the aforementioned experiments suggest that oxocrebanine can inhibit the growth and proliferation of Hep3B2.1-7 cells.

Oxocrebanine induces apoptosis in Hep3B2.1-7 cells

The results of PI/Annexin V double staining combined with flow cytometry indicated that, compared with the control group, the apoptosis level of Hep3B2.1-7 cells in the VP16 group was significantly increased ($P < 0.01$; Table 4 and Figure 2). The apoptosis level in the oxocrebanine group was also significantly increased ($P < 0.05$ or $P < 0.01$) in a dose-dependent manner. Moreover, the cells treated with 50.89 µmol/L oxocrebanine exhibited early apoptosis (Figure 2D), whilst late apoptosis was observed at 101.78 µmol/L (Figure 2E).

Effect of oxocrebanine on expression of apoptosis effector proteins cleaved caspase3 and PARP1 in Hep3B2.1-7 cells

Western blot analysis was performed to measure the expression levels of cleaved caspase3 and PARP1 proteins in Hep3B2.1-7 cells after treatment with oxocrebanine (Figure 3). Compared with the control group, VP16 treatment significantly upregulated the expression of cleaved caspase3 ($P < 0.01$) but significantly downregulated the expression of PARP1 ($P < 0.01$). Similarly, oxocrebanine treatment significantly increased the expression of cleaved caspase3 ($P < 0.01$) but significantly decreased the expression of PARP1 ($P < 0.01$).

Effect of oxocrebanine on expression of apoptosis-related proteins Bcl-2, Bax, Bak, and Mcl-1 in Hep3B2.1-7 cells

Western blot analysis was performed to measure the expression levels of the apoptosis-related proteins Bcl-2, Bax, Bak, and Mcl-1 in Hep3B2.1-7 cells after treatment with oxocrebanine. Compared with the control group, VP16 or oxocrebanine significantly reduced the expression levels of Bcl-2 and Bax proteins ($P < 0.01$; Figure 4A-C). Furthermore, after treatment with VP16 or oxocrebanine, the expression level of Bak protein was significantly increased ($P < 0.01$), and the expression level of Mcl-1 protein was significantly decreased ($P < 0.01$; Figure 4D-G).

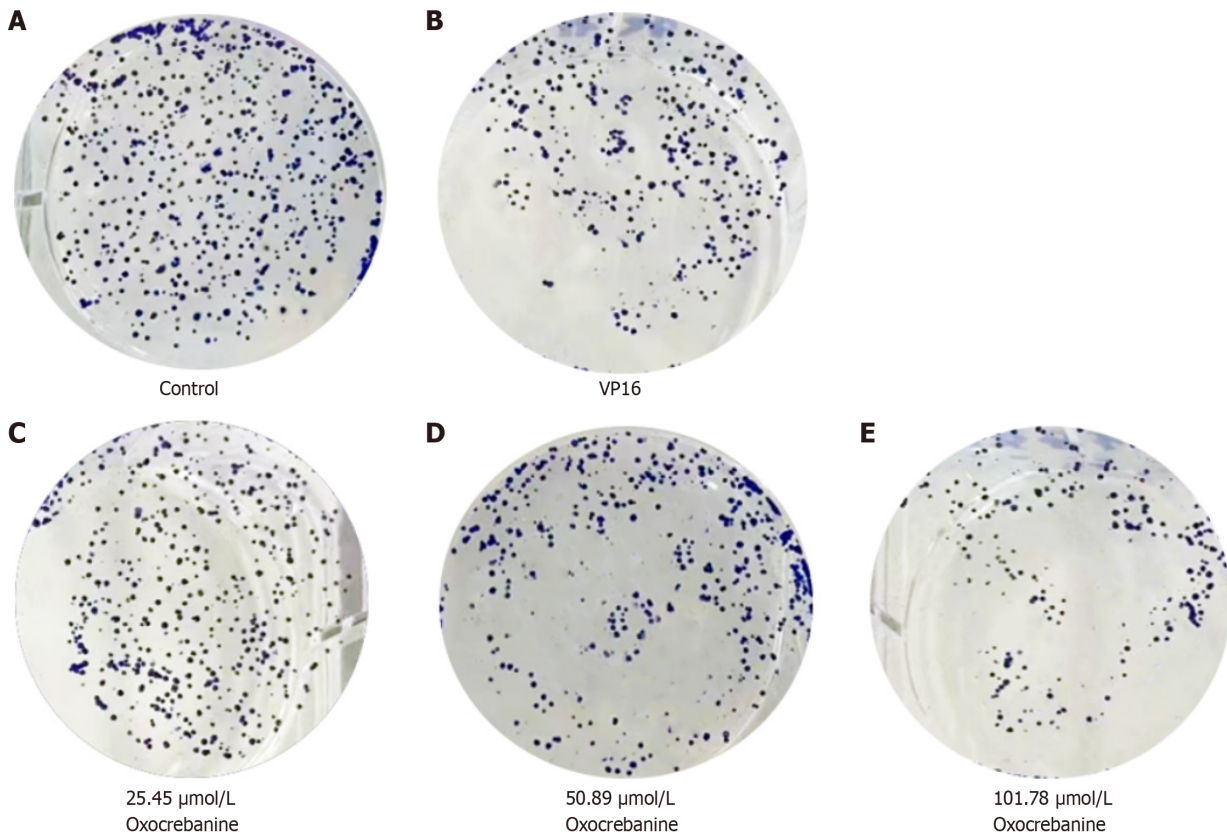


Figure 1 Effect of oxocorebanine on proliferation of Hep3B2.1-7 cells ($\times 100$). Hep3B2.1-7 cells were treated respectively with 1 mL of complete culture medium, VP16 at a concentration of 32.54 $\mu\text{mol/L}$, and oxocorebanine at concentrations of 25.45 $\mu\text{mol/L}$, 50.89 $\mu\text{mol/L}$, and 101.78 $\mu\text{mol/L}$. After 72 hours of drug treatment, the cells of each group were stained with crystal violet for 25 minutes and photographed. A: Control; B: VP16; C: 25.45 $\mu\text{mol/L}$ oxocorebanine; D: 50.89 $\mu\text{mol/L}$ oxocorebanine; E: 101.78 $\mu\text{mol/L}$ oxocorebanine.

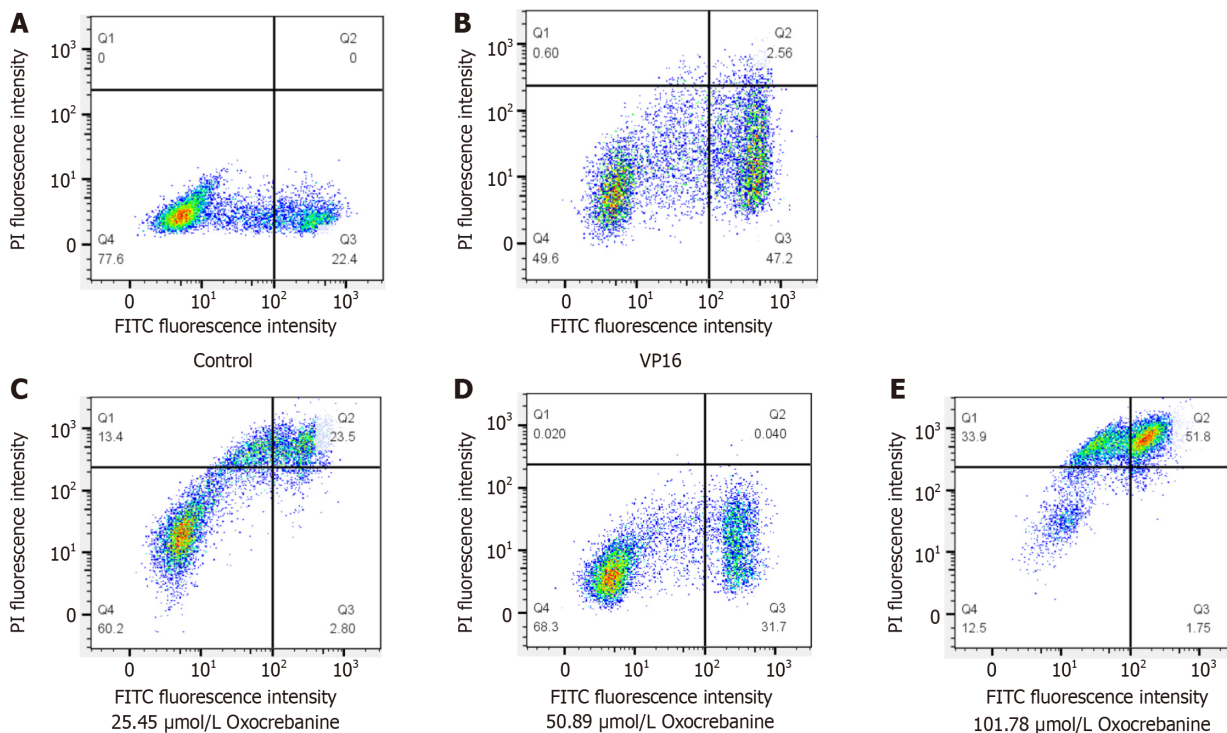


Figure 2 Effect of oxocorebanine on apoptosis of Hep3B2.1-7 cells. After 48 hours of incubation with complete culture medium, 32.54 $\mu\text{mol/L}$ VP16, or 25.45 $\mu\text{mol/L}$, 50.89 $\mu\text{mol/L}$, or 101.79 $\mu\text{mol/L}$ oxocorebanine, the cells were stained with FITC-labeled Annexin V and PI staining solution, and then analyzed by flow

cytometry. A: Control; B: VP16; C: 25.45 $\mu\text{mol/L}$ oxocorebanine; D: 50.89 $\mu\text{mol/L}$ oxocorebanine; E: 101.78 $\mu\text{mol/L}$ oxocorebanine.

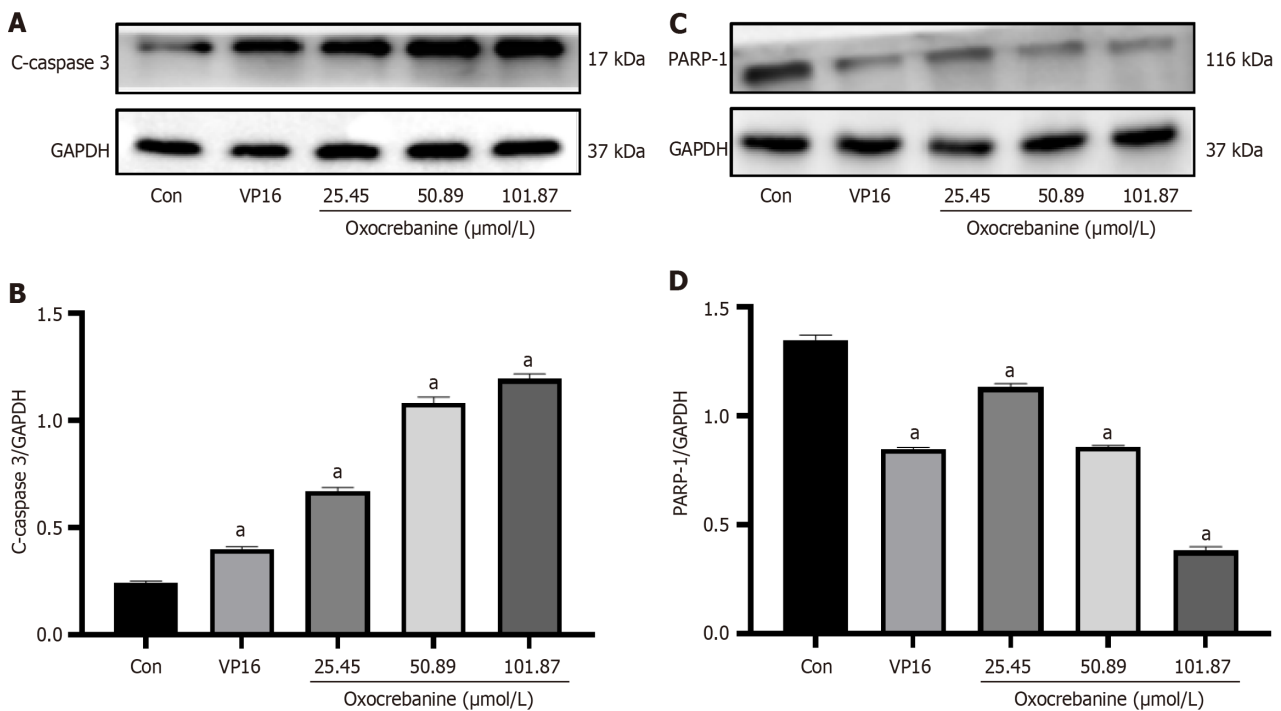


Figure 3 Protein expression levels of cleaved caspase3 and PARP in Hep3B2.1-7 cells. A: Western blot analysis of cleaved caspase3 (c-caspase3) protein level in Hep3B2.1-7 cells after treatment with oxocorebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; B: Western blot analysis data indicated that the expression of c-caspase3 was upregulated with oxocorebanine treatments; C: Level of PARP1 in Hep3B2.1-7 cells after treatment with oxocorebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; D: Western blot analysis data showed that the expression of PARP1 was downregulated by oxocorebanine treatments. Data are shown as the mean \pm SD. ^a $P < 0.01$ vs control ($n = 3$).

Inhibition of Hep3B2.1-7 cell growth and proliferation by oxocorebanine after addition of an autophagy inhibitor

To assess whether oxocorebanine affects the activity of Hep3B2.1-7 cells through the autophagy pathway, MTT and BrdU labeling assays were performed to evaluate the inhibition of HCC cell growth and proliferation after adding the autophagy inhibitor 3-MA. The results of the MTT assay indicated that, compared with the control group, the viability of Hep3B2.1-7 cells significantly decreased after treatment with oxocorebanine alone ($P < 0.01$; Table 5 and Figure 5A). Compared with the oxocorebanine group, the cell viability in the oxocorebanine + 3-MA group decreased significantly ($P < 0.01$; Table 5 and Figure 5A). Moreover, the results of the BrdU assay indicated that, compared with the control group, oxocorebanine significantly reduced the proliferation of Hep3B2.1-7 cells ($P < 0.01$; Table 5 and Figure 5B). Compared with the oxocorebanine group, the proliferation rate in the oxocorebanine + 3-MA group also decreased significantly ($P < 0.01$; Table 5 and Figure 5B). This indicates that after the addition of the autophagy inhibitor 3-MA, oxocorebanine significantly inhibited the growth and proliferation of Hep3B2.1-7 cells. These results indicate that the inhibition of Hep3B2.1-7 cell proliferation by oxocorebanine may be related to the induction of protective autophagy in HCC cells.

Effect of oxocorebanine on expression of autophagy marker LC3 protein in Hep3B2.1-7 cells after addition of an autophagy inhibitor

Western blot analysis was performed to measure the LC3-II/LC3-I protein expression ratio in Hep3B2.1-7 cells after administration of the autophagy inhibitor 3-MA and treatment with oxocorebanine. Compared with the control group, the LC3-II/LC3-I ratio in the oxocorebanine group significantly increased ($P < 0.01$). Compared with oxocorebanine alone, the LC3-II/LC3-I protein expression ratio in the oxocorebanine + 3-MA group significantly decreased ($P < 0.01$; Figure 6A and B). These results indicate that after treating Hep3B2.1-7 cells with oxocorebanine, the LC3-II/LC3-I ratio increases, and the autophagy inhibitor 3-MA can reverse this process.

Effect of oxocorebanine on expression of 4EBP1 and p-4EBP1 proteins in Hep3B2.1-7 cells after addition of an autophagy inhibitor

Western blot results demonstrated that, compared with the control group, the phosphorylation level of 4EBP1 in Hep3B2.1-7 cells significantly decreased in the oxocorebanine group ($P < 0.01$). Compared with oxocorebanine alone, the phosphorylation level of 4EBP1 in the oxocorebanine + 3-MA group significantly increased ($P < 0.01$; Figure 6C and D). The results suggest that oxocorebanine can reduce the phosphorylation level of 4EBP1 in Hep3B2.1-7 cells, and the

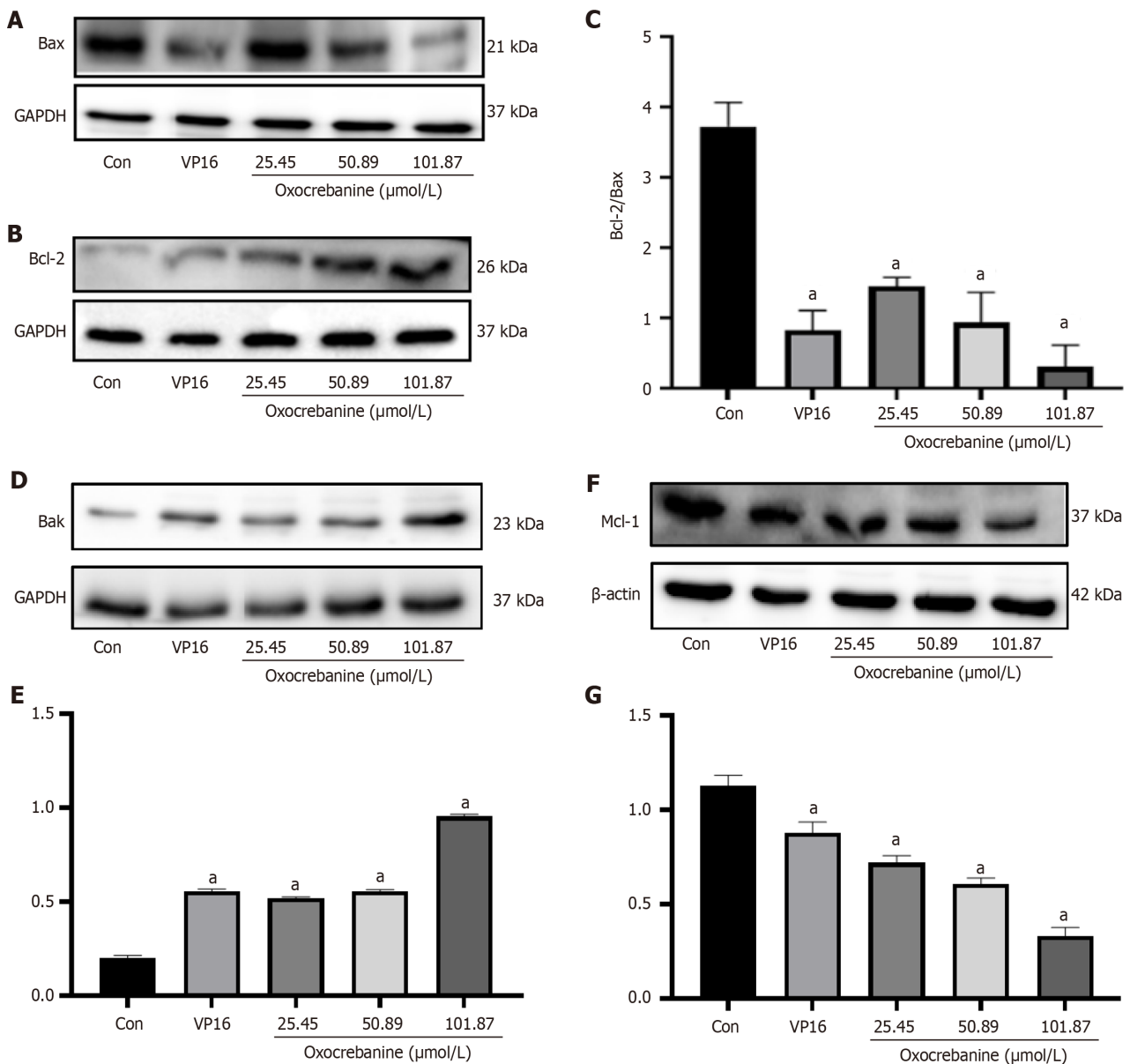


Figure 4 Protein expression levels of Bcl-2, Bax, Bak, and Mcl-1 in Hep3B2.1-7 cells. A: Western blot analysis of Bcl-2 protein level in Hep3B2.1-7 cells after treatment with oxocorebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; B: Western blot analysis of Bax protein level in Hep3B2.1-7 cells after treatment with oxocorebanine, with VP16 (32.54 $\mu\text{mol/L}$) used as a control; C: Western blot analysis data indicated that the ratio of Bcl-2 to Bax protein expression was decreased with oxocorebanine treatments; D: Level of Bak in Hep3B2.1-7 cells after treatment with oxocorebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; E: Western blot analysis data showed that the expression of Bak was downregulated by oxocorebanine treatments; F: Level of Mcl-1 in Hep3B2.1-7 cells after treatment with oxocorebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; G: Western blot analysis data showed that the expression of Mcl-1 was downregulated by oxocorebanine treatments. Data are shown as the mean \pm SD. ^a $P < 0.01$ vs control ($n = 3$).

autophagy inhibitor 3-MA can reverse this process.

Effect of oxocorebanine on expression of P70S6K and p-P70S6K proteins in Hep3B2.1-7 cells after addition of an autophagy inhibitor

Compared with the control group, the phosphorylation level of P70S6K in Hep3B2.1-7 cells significantly decreased in the oxocorebanine group ($P < 0.01$). Compared with oxocorebanine alone, the phosphorylation level of P70S6K in the oxocorebanine + 3-MA group significantly increased ($P < 0.01$; Figure 6E and F). These results indicate that oxocorebanine can reduce the phosphorylation level of P70S6K, and the autophagy inhibitor 3-MA can reverse this process.

Oxocorebanine inhibits tumor growth in vivo

The tumor volume decreased significantly in the oxocorebanine group compared with the control group, and oxocorebanine administration was associated with a significant inhibition of tumor growth *in vivo* (Figure 7A and B). The TUNEL results revealed significantly increased apoptosis in the oxocorebanine group, and the immunohistochemical staining results demonstrated that the expression level of LC3 in the tumor tissues of mice in the oxocorebanine group was much higher

Table 1 Growth inhibition rate of Hep3B2.1-7 cells in MTT assay (mean ± SD, n = 3)		
Group	Concentration (μmol/L)	Growth inhibition rate (%)
Oxocrebanine	5	4.18 ± 0.02
	10	10.24 ± 0.01 ^a
	20	25.09 ± 0.03 ^a
	40	35.59 ± 0.02 ^a
	80	76.6 ± 0.02 ^a

^a*P* < 0.01 *vs* control.

Table 2 Proliferation level of Hep3B2.1-7 cells in BrdU assay (mean ± SD, n = 3)		
Group	Concentration (μmol/L)	Proliferation rate (%)
Oxocrebanine	0	102.6 ± 0.07
	5	95.6 ± 0.04 ^a
	10	88.59 ± 0.05 ^a
	20	79.09 ± 0.02 ^a
	40	60.24 ± 0.02 ^a
	80	37.18 ± 0.06 ^a

^a*P* < 0.01 *vs* control.

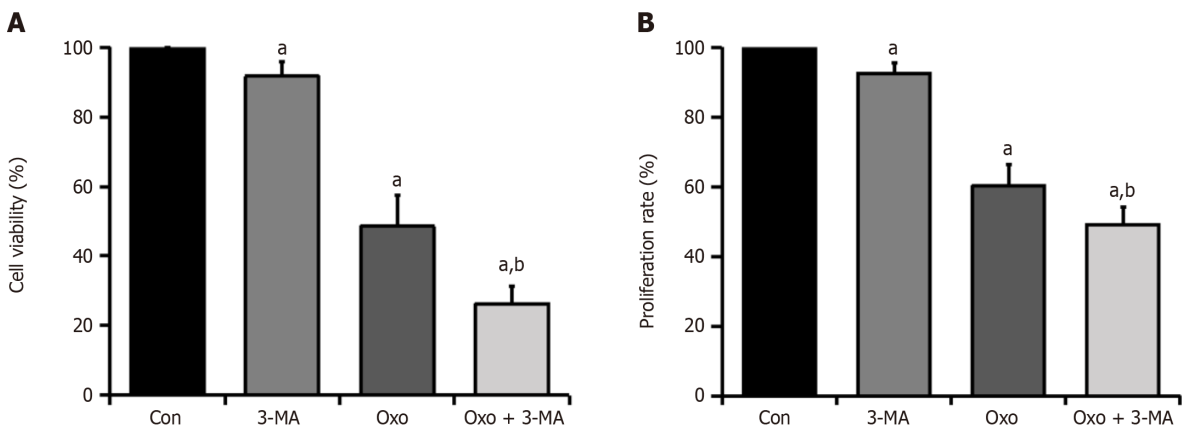


Figure 5 Hep3B2.1-7 cell viability and proliferation rate measured by MTT assay and BrdU test. A: Viability of cells treated with oxocrebanine and 3-MA as demonstrated by MTT assay; B: Proliferation rate of cells treated with oxocrebanine and 3-MA. Data are shown as the mean ± SD. ^a*P* < 0.01 *vs* control; ^b*P* < 0.01 *vs* oxocrebanine group (*n* = 3).

than that of the control group (Figure 7C). This indicates that oxocrebanine can reduce the proliferation of HCC cells in mice through apoptosis and autophagy. Mice in the control group died on days 24, 27, 37, 42, and 46 of treatment, whilst mice in the oxocrebanine group died on days 45 and 50. The remaining three mice survived for > 70 days. On day 42, the survival rate of mice in the oxocrebanine group was 100%, whilst the survival rate in the control group was 20% (Figure 7D). On day 46, the survival rate of mice in the oxocrebanine group was 60%, whilst the survival rate in the control group was 0%. The Kaplan-Meier survival curve demonstrated that oxocrebanine can significantly prolong the survival time of tumor-bearing nude mice (*P* < 0.01; Figure 7D).

DISCUSSION

Stephania hainanensis H.S. Lo et Y. Tsoong contains a notable concentration of aporphine alkaloids. Our previous research

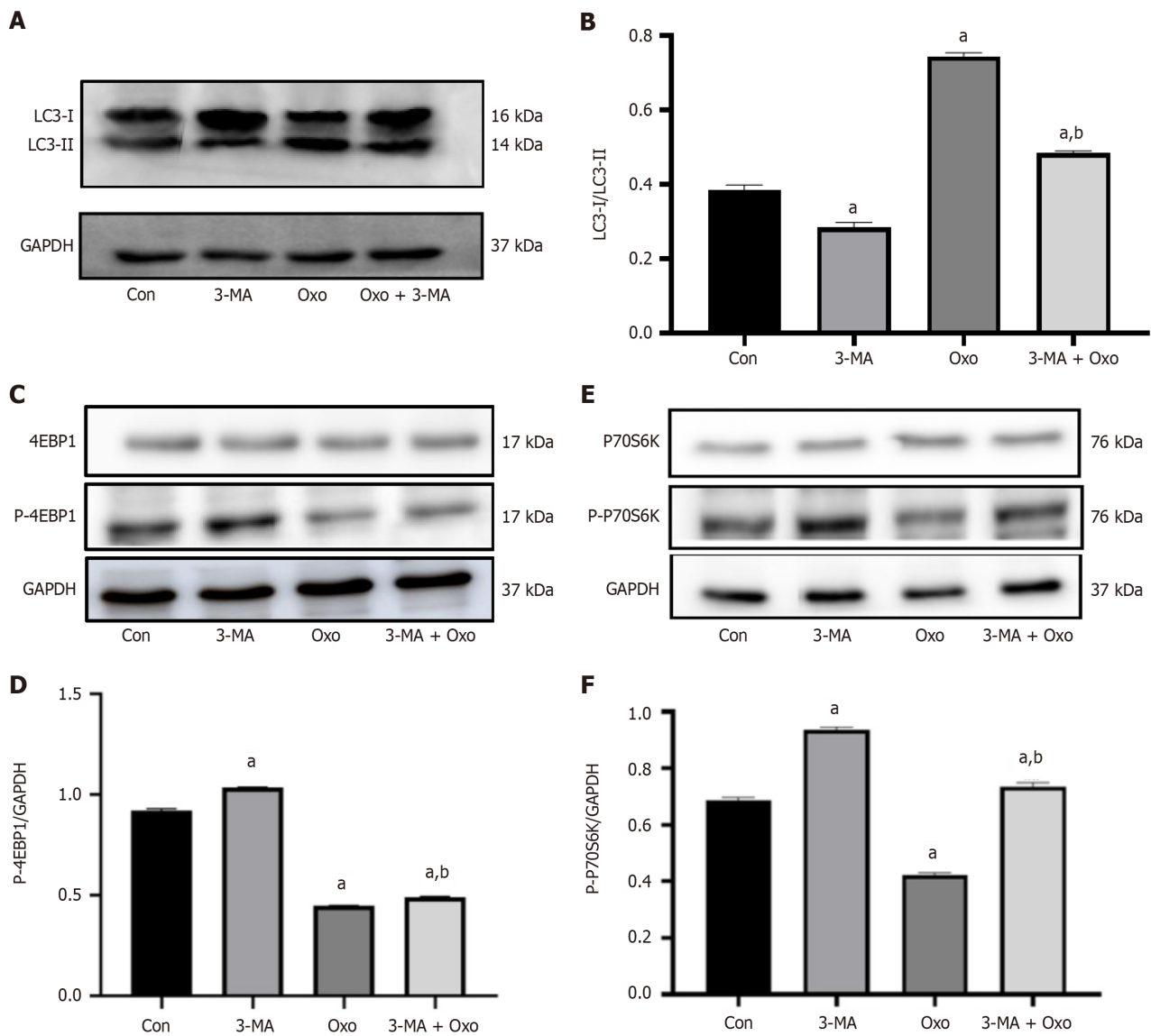


Figure 6 Expression level of LC3 protein and phosphorylation levels of 4EBP1 and P70S6K in Hep3B2.1-7 cells. A: Western blot analysis of LC3-I and LC3-II protein levels in Hep3B2.1-7 cells after treatment with oxocrebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; B: Western blot analysis data indicated that the ratio of LC3-II to LC3-I protein expression was increased with oxocrebanine treatments compared with the control group. Compared with oxocrebanine alone, the LC3-II/LC3-I ratio in the 3-MA + oxocrebanine group was significantly decreased; C: Phosphorylation level of 4EBP1 protein in Hep3B2.1-7 cells after treatment with oxocrebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; D: Western blot analysis data showed that the phosphorylation level of 4EBP1 was downregulated by oxocrebanine treatments compared with the control group. Compared with oxocrebanine alone, the phosphorylation level of 4EBP1 in the 3-MA + oxocrebanine group was significantly upregulated; E: Phosphorylation level of P70S6K in Hep3B2.1-7 cells after treatment with oxocrebanine. VP16 (32.54 $\mu\text{mol/L}$) was used as a control; F: Western blot analysis data showed that the phosphorylation level of P70S6K protein was downregulated by oxocrebanine treatments. Compared with oxocrebanine alone, the phosphorylation level of P70S6K protein in the 3-MA + oxocrebanine group was significantly upregulated. Data are shown as the mean \pm SD. ^a $P < 0.01$ vs control; ^b $P < 0.01$ vs oxocrebanine group ($n = 3$).

isolated the aporphine alkaloid oxocrebanine from *Stephania hainanensis* H.S. Lo et Y. Tsoong, with the molecular formula $\text{C}_{20}\text{H}_{21}\text{O}_5\text{N}$ and a molecular weight of 355.14, and demonstrated its promising antitumor activity[10,11]. The present study assessed the mechanism by which oxocrebanine inhibits the proliferation of HCC, a prevalent cancer type in tropical regions, such as Hainan Province, Leizhou Peninsula in Guangdong Province, and some areas in Guangxi Province. The results from the MTT, BrdU cell proliferation, and colony formation assays indicate that oxocrebanine effectively inhibits the growth and proliferation of Hep3B2.1-7 cells. The IC_{50} of oxocrebanine is slightly weaker than that of sorafenib, a targeted drug against HCC. However, sorafenib has cardiovascular toxicity and lead to adverse cardiovascular effects including hypertension, thromboembolism, cardiac ischemia, and left ventricular dysfunction. Unfortunately, the adverse cardiovascular effects caused by sorafenib not only affect solid tumor patients but also limit its application in curing other diseases[13]. Oxocrebanine has similar or slightly better activity compared with Chinese herbal ingredients with significant anti-HCC activity reported recently, such as ginsenosides[14], matrine[15], resveratrol [16], and demethylzeylasteral[17]. The commonly used decoction of root tuber from *Stephania hainanensis* H.S. Lo et Y. Tsoong has been used to treat liver cancer and precancerous lesions in hospitals of Li Autonomous Prefecture, Hainan Province, China[18]. Previous research results[10] show that oxocrebanine has the characteristics of low toxicity and high

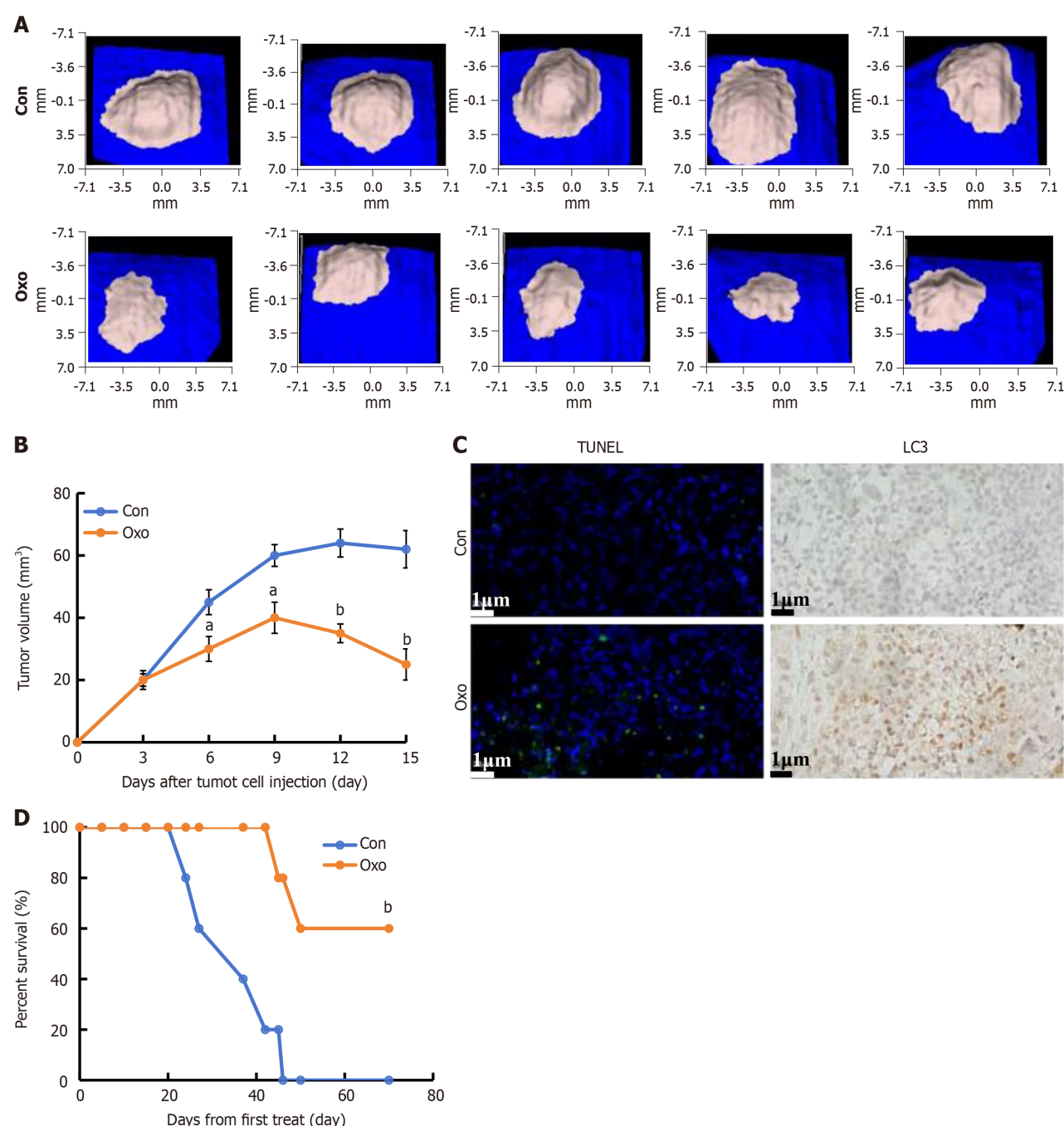


Figure 7 Inhibitory effect of oxocrebanine on growth of Hep3B2.1-7 cells *in vivo*. Hep3B2.1-7 cells were injected into the right flanks of Nu/Nu mice (5 mice/group). When tumor volumes reached 50 mm³, mice were treated with indicated formulations every 3 days for 5 times. A: Tumor mass images of Hep3B2.1-7 model mice at day 18; B: Tumor volumes at different time points; C: Apoptosis and LC3 expression in tumor xenografts detected by TUNEL and IHC, respectively; D: Survival curve. Data are shown as the mean \pm SD; ^a $P < 0.05$ vs control; ^b $P < 0.01$ vs control.

efficiency, and has good application prospects. Animal tumor inhibition experiments showed that the body weight of the experimental animals in the oxocrebanine group was slightly reduced, though there was no significant difference compared to the control group, indicating that the toxicity of oxocrebanine is lower. Therefore, as a component of Li ethnomedicine, oxocrebanine has lower toxicity and side effects compared with chemotherapy drugs[19]. Apoptosis and autophagy are two critical cellular pathways that are involved in cell survival or death. The balance between apoptosis and autophagy maintains intracellular homeostasis, but the balance is often dysregulated in many cancers, including HCC[7]. Flow cytometry analysis revealed that oxocrebanine treatment promoted apoptosis in Hep3B2.1-7 cells, suggesting that the inhibition of cell growth and proliferation by oxocrebanine may be related to its induction of apoptosis. Further assessment using Western blotting revealed that oxocrebanine upregulated the expression of the cleaved caspase3 protein whilst downregulating PARP1. Additionally, oxocrebanine upregulated the pro-apoptotic proteins, Bax and Bak, and downregulated the anti-apoptotic proteins, Bcl-2 and Mcl-1, whilst increasing the ratio of Bcl-2 to Bax. These findings suggest that oxocrebanine promotes apoptosis in Hep3B2.1-7 cells. Under several apoptotic stimuli, downstream effectors of p53, such as Bax and Bak, may activate the p53 pathway, increase Bax and Bak expression, and

Table 3 Proliferation level of Hep3B2.1-7 cells in plate colony formation assay (mean \pm SD, $n = 3$)

Group	Concentration (μ mol/L)	Proliferation rate (%)
Control	-	100
VP16	32.54	49.76 \pm 0.02 ^a
Oxocrebanine	25.45	76.30 \pm 0.02 ^a
	50.89	51.74 \pm 0.04 ^a
	101.78	23.55 \pm 0.03 ^a

^a $P < 0.01$ vs control.**Table 4 Apoptosis rate of Hep3B2.1-7 cells in different groups (mean \pm SD, $n = 3$)**

Group	Concentration (μ mol/L)	Apoptosis rate (%)
Control	-	22.40 \pm 0.02
VP16	32.54	49.76 \pm 0.02 ^b
Oxocrebanine	25.45	26.30 \pm 0.02 ^a
	50.89	31.74 \pm 0.04 ^b
	101.78	53.55 \pm 0.03 ^b

^a $P < 0.05$ vs control.^b $P < 0.01$ vs control.**Table 5 Hep3B2.1-7 cell viability and proliferation rate in MTT assay and BrdU test (mean \pm SD, $n = 3$)**

Group	Cell viability (%)	Proliferation rate (%)
Control	100	101.60 \pm 0.07
3-MA	91.68 \pm 0.04 ^a	92.60 \pm 0.03 ^a
Oxo	48.56 \pm 0.09 ^a	60.22 \pm 0.06 ^a
Oxo + 3-MA	26.37 \pm 0.05 ^{a,b}	49.09 \pm 0.05 ^{a,b}

^a $P < 0.01$ vs control.^b $P < 0.01$ vs oxocrebanine group.

enable their translocation to the cell membrane[20]. There, they can combine with Bcl-2 and Mcl-1, potentially increasing membrane permeability and facilitating the release of apoptosis-related factors, thus directly inducing apoptosis in HCC cells. On the other hand, the activation of the caspase pathway enables caspases to bind with PARP, further activating caspase 3 and promoting apoptosis in Hep3B2.1-7 cells[21]. Autophagy, which serves a major role in cell survival and tumor development, can inhibit or promote tumor proliferation[22]. It could inhibit the proliferation of cancer cells by eliminating tumorigenic protein substrates and damaging organelles[23]. The present study further demonstrated that the inhibitory effect of oxocrebanine on Hep3B2.1-7 cell proliferation may be related to the induction of protective autophagy in HCC cells through the addition of an autophagy inhibitor. The inhibition of autophagy markedly restored cell proliferation in oxocrebanine-treated cells. These results indicated that oxocrebanine induced autophagy in Hep3B2.1-7 cells to inhibit the proliferation of cancer cells. LC3 is a marker of autophagy. During autophagy, cytoplasmic LC3 (LC3-I) undergoes enzymatic cleavage of a small peptide and is converted to membrane-bound LC3-II, indicated by a notable increase in LC3-II in autophagic cells. The LC3-II/LC3-I ratio is commonly used to observe autophagic activity[24,25]. The results of the present study suggest that oxocrebanine increases the LC3-II/LC3-I ratio, indicating an increase in the expression of autophagy marker proteins. However, the autophagy inhibitor 3-MA can reverse this process. The PI3K/Akt/mTOR pathway is a critical signaling pathway in the regulation of autophagy. When inhibited, the phosphorylation of downstream effectors, 4EBP1 and P70S6K, decreases, inducing autophagy[26]. The present research also revealed that oxocrebanine significantly reduced the phosphorylation levels of 4EBP1 and P70S6K. This indicates that oxocrebanine, after acting on Hep3B2.1-7 cells, promotes the conversion of LC3-I to LC3-II, inhibits the phosphorylation of 4EBP1 and P70S6K proteins, and induces autophagy. We speculate that Beclin1, by activating PI3K, regulates the formation of autophagic vesicles, and then induces the lipidation of LC3-I to LC3-II, enabling its translocation to the autophagosome

membrane. The autophagosome expands and then induces autophagy in the HCC cells[27,28]. Additionally, oxocrebanine may inhibit the Akt/mTOR signaling pathway, thus reducing the phosphorylation levels of its downstream effectors, 4EBP1 and P70S6K. The autophagy inhibitor 3-MA can reverse this process, further indicating that oxocrebanine can inhibit the proliferation of Hep3B2.1-7 cells through the autophagy pathway. Based on the *in vitro* experimental results, the present study assessed the anticancer effect and the mechanism of action of oxocrebanine in a subcutaneous transplantation tumor model of human Hep3B2.1-7 cells in nude mice. Oxocrebanine suppressed the growth of Hep3B2.1-7 cells in the tumor-bearing nude mice. The tumor volume of the oxocrebanine group markedly decreased compared with that of the mock-treated and control groups ($P < 0.05$ and $P < 0.01$, respectively). Apoptosis is a programmed cell death process, which serves an important role in maintaining the balance between tissues and organs [29]. Most of the tumor tissue cells in the oxocrebanine-treated group underwent apoptosis that was detected using a TUNEL assay, revealing that the decrease in the viability of Hep3B2.1-7 cells was due to the induction of apoptosis. Immunohistochemical staining revealed that LC3 expression in the tumor tissue was reduced, which indicated that oxocrebanine significantly inhibited tumor growth by inducing cell autophagy. These results indicate that oxocrebanine inhibited tumor growth by inducing cell apoptosis and promoting cell autophagy *in vivo*, which are in line with those obtained in the cell experiments. In summary, the results of the present study demonstrate that oxocrebanine effectively inhibits the growth and proliferation of Hep3B2.1-7 cells. It upregulates the expression of cleaved caspase3, downregulates PARP1, upregulates Bax and Bak, and downregulates Bcl-2 and Mcl-1, leading to apoptosis in Hep3B2.1-7 cells and the inhibition of cell proliferation. The inhibition of Hep3B2.1-7 cell proliferation by oxocrebanine may be related to the induction of protective autophagy in HCC cells. Oxocrebanine also promotes the conversion of LC3-I to LC3-II in Hep3B2.1-7 cells, a process reversed by the autophagy inhibitor 3-MA, which reduces the phosphorylation levels of 4EBP1 and P70S6K, induces autophagy, and thus inhibits cell growth. In addition, there was also an increase in the number of TUNEL positive cells, a marker for apoptosis induction, in the oxocrebanine group. Moreover, immunohistochemistry revealed that the level of LC3 was markedly upregulated after the treatment with oxocrebanine. The present study revealed that oxocrebanine can inhibit the proliferation of HCC cells by promoting apoptosis and inducing autophagy both *in vitro* and *in vivo*. These findings raise the possibility of oxocrebanine as a potential new active ingredient of *tephania hainanensis* H.S. Lo et Y. Tsoong that has anti-HCC properties. However, additional studies are warranted to elucidate other molecular mechanisms which are responsible for the anticancer efficacy of oxocrebanine as a potent anticancer agent against HCC. The research findings may provide a drug option with improved efficacy and reduced toxicity for the clinical treatment of HCC.

CONCLUSION

In conclusion, oxocrebanine can inhibit the proliferation of human HCC cells by promoting apoptosis and inducing autophagy *in vitro* and *in vivo*.

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FOOTNOTES

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REFERENCES

- 1 **Younossi ZM**, Wong G, Anstee QM, Henry L. The Global Burden of Liver Disease. *Clin Gastroenterol Hepatol* 2023; **21**: 1978-1991 [RCA] [PMID: 37121527 DOI: 10.1016/j.cgh.2023.04.015] [FullText] [Citation(s) in RCA: 178] [Article Influence: 89.0]
- 2 **Singal AG**, Lampertico P, Nahon P. Epidemiology and surveillance for hepatocellular carcinoma: New trends. *J Hepatol* 2020; **72**: 250-261 [RCA] [PMID: 31954490 DOI: 10.1016/j.jhep.2019.08.025] [FullText] [Citation(s) in RCA: 757] [Article Influence: 151.4]
- 3 **Hepatocellular carcinoma**. *Nat Rev Dis Primers* 2021; **7**: 7 [RCA] [PMID: 33479233 DOI: 10.1038/s41572-021-00245-6] [FullText] [Citation(s) in RCA: 137] [Article Influence: 34.3]
- 4 **Rumgay H**, Arnold M, Ferlay J, Lesi O, Cabaasag CJ, Vignat J, Laversanne M, McGlynn KA, Soerjomataram I. Global burden of primary liver cancer in 2020 and predictions to 2040. *J Hepatol* 2022; **77**: 1598-1606 [RCA] [PMID: 36208844 DOI: 10.1016/j.jhep.2022.08.021] [FullText] [Full Text(PDF)] [Citation(s) in RCA: 973] [Article Influence: 324.3]
- 5 **Sung H**, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, Bray F. Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA Cancer J Clin* 2021; **71**: 209-249 [RCA] [PMID: 33538338 DOI: 10.3322/caac.21660] [FullText] [Citation(s) in RCA: 62863] [Article Influence: 15715.8]
- 6 **Gao C**, Xu J, Liu Y, Yang Y. Nutrition Policy and Healthy China 2030 Building. *Eur J Clin Nutr* 2021; **75**: 238-246 [RCA] [PMID: 33219269 DOI: 10.1038/s41430-020-00765-6] [FullText] [Citation(s) in RCA: 40] [Article Influence: 8.0]
- 7 **Kouroumalis E**, Tsomidis I, Voumvouraki A. Pathogenesis of Hepatocellular Carcinoma: The Interplay of Apoptosis and Autophagy. *Biomedicines* 2023; **11** [RCA] [PMID: 37189787 DOI: 10.3390/biomedicines11041166] [FullText] [Citation(s) in RCA: 23] [Article Influence: 11.5]
- 8 **de Mattos AZ**, Bombassaro IZ, Vogel A, Debes JD. Hepatocellular carcinoma-the role of the underlying liver disease in clinical practice. *World J Gastroenterol* 2024; **30**: 2488-2495 [RCA] [PMID: 38817660 DOI: 10.3748/wjg.v30.i19.2488] [FullText] [Full Text(PDF)] [Citation(s) in RCA: 4] [Article Influence: 4.0]
- 9 **Bergquist A**, Ekstedt M, Hagström H, Järnerot G, Lindgren S, Nilsson E, Nyhlin N, Rorsman F, Stål P, Werner M; Swehep, Kechagias S. Forty years of successful national research collaboration in liver disease - the Swedish experience. *Scand J Gastroenterol* 2024; **59**: 1314-1321 [RCA] [PMID: 39485016 DOI: 10.1080/00365521.2024.2421824] [FullText] [Citation(s) in RCA: 1] [Article Influence: 1.0]
- 10 **Yu L**, Han S, Lang L, Song H, Zhang C, Dong L, Jia S, Zhang Y, Xiao D, Liu J, Xu Y, Zhang X. Oxocrebanine: A Novel Dual Topoisomerase inhibitor, Suppressed the Proliferation of Breast Cancer Cells MCF-7 by Inducing DNA Damage and Mitotic Arrest. *Phytomedicine* 2021; **84**: 153504 [RCA] [PMID: 33611211 DOI: 10.1016/j.phymed.2021.153504] [FullText] [Citation(s) in RCA: 9] [Article Influence: 2.3]
- 11 **Yu L**, Jiang CY, Song DX, Yu M, Shang DY, Jiang B, Ji YB and Wang ZW. [Analysis of the Structure Activity Relationship and Inhibition of Tumor Cell Proliferation by Alkaloids in Stephania Hainannensis H.S. Lo et Y. Tsoong]. *Liaoning Zhongyi Zazhi* 2020; **47**: 145-154 [DOI: 10.13192/j.issn.1000-1719.2020.02.047] [FullText]
- 12 Guide for the Care and Use of Laboratory Animals. Washington (DC): National Academies Press (US); 2011– [PMID: 21595115] [FullText]
- 13 **Li J**, Zhang L, Ge T, Liu J, Wang C, Yu Q. Understanding Sorafenib-Induced Cardiovascular Toxicity: Mechanisms and Treatment Implications. *Drug Des Devel Ther* 2024; **18**: 829-843 [RCA] [PMID: 38524877 DOI: 10.2147/DDDT.S443107] [FullText] [Full Text(PDF)] [Citation(s) in RCA: 14] [Article Influence: 14.0]
- 14 **Zhou Y**, Wang Z, Ren S, Li W. Mechanism of action of protopanaxadiol ginsenosides on hepatocellular carcinoma and network pharmacological analysis. *Chin Herb Med* 2024; **16**: 548-557 [RCA] [PMID: 39606268 DOI: 10.1016/j.chmed.2024.06.002] [FullText] [Full Text(PDF)] [Citation(s) in RCA: 3] [Article Influence: 3.0]
- 15 **Chang X**, Li H, Huang Z, Song C, Zhang Z, Pan W. Matrine suppresses hepatocellular carcinoma tumorigenesis by modulating circ_0055976/miR-1179/lactate dehydrogenase A axis. *Environ Toxicol* 2024; **39**: 1481-1493 [RCA] [PMID: 37994612 DOI: 10.1002/tox.24041] [FullText] [Citation(s) in RCA: 5] [Article Influence: 5.0]
- 16 **Feng CY**, Cai CS, Shi XQ, Zhang ZJ, Su D, Qiu YQ. Resveratrol promotes mitophagy via the MALAT1/miR-143-3p/RRM2 axis and suppresses cancer progression in hepatocellular carcinoma. *J Integr Med* 2025; **23**: 79-92 [RCA] [PMID: 39675939 DOI: 10.1016/j.joim.2024.11.003] [FullText] [Citation(s) in RCA: 1] [Article Influence: 1.0]
- 17 **Pan L**, Feng F, Wu J, Fan S, Han J, Wang S, Yang L, Liu W, Wang C, Xu K. Demethylzeylasteral targets lactate by inhibiting histone lactylation to suppress the tumorigenicity of liver cancer stem cells. *Pharmacol Res* 2022; **181**: 106270 [RCA] [PMID: 35605812 DOI: 10.1016/j.phrs.2022.106270] [FullText] [Citation(s) in RCA: 190] [Article Influence: 63.3]
- 18 **Wang M**, Zhang XM, Fu X, Zhang P, Hu WJ, Yang BY, Kuang HX. Alkaloids in genus stephania (Menispermaceae): A comprehensive review of its ethnopharmacology, phytochemistry, pharmacology and toxicology. *J Ethnopharmacol* 2022; **293**: 115248 [RCA] [PMID: 35430287 DOI: 10.1016/j.jep.2022.115248] [FullText] [Citation(s) in RCA: 15] [Article Influence: 5.0]
- 19 **Yang XH**, Ren LS, Wang GP, Zhao LL, Zhang H, Mi ZG, Bai X. A new method of establishing orthotopic bladder transplantable tumor in mice. *Cancer Biol Med* 2012; **9**: 261-265 [RCA] [PMID: 23691487 DOI: 10.7497/j.issn.2095-3941.2012.04.007] [FullText] [Full Text(PDF)]

- [Citation(s) in *RCA*: 17] [Article Influence: 1.4]
- 20 **Wei MC**, Zong WX, Cheng EH, Lindsten T, Panoutsakopoulou V, Ross AJ, Roth KA, MacGregor GR, Thompson CB, Korsmeyer SJ. Proapoptotic BAX and BAK: a requisite gateway to mitochondrial dysfunction and death. *Science* 2001; **292**: 727-730 [*RCA*] [PMID: 11326099 DOI: 10.1126/science.1059108] [FullText] [Citation(s) in *RCA*: 3070] [Article Influence: 127.9]
 - 21 **Shimizu S**, Narita M, Tsujimoto Y. Bcl-2 family proteins regulate the release of apoptogenic cytochrome c by the mitochondrial channel VDAC. *Nature* 1999; **399**: 483-487 [*RCA*] [PMID: 10365962 DOI: 10.1038/20959] [FullText] [Citation(s) in *RCA*: 1655] [Article Influence: 63.7]
 - 22 **Wen X**, Klionsky DJ. At a glance: A history of autophagy and cancer. *Semin Cancer Biol* 2020; **66**: 3-11 [*RCA*] [PMID: 31707087 DOI: 10.1016/j.semcancer.2019.11.005] [FullText] [Citation(s) in *RCA*: 76] [Article Influence: 12.7]
 - 23 **Singh SS**, Vats S, Chia AY, Tan TZ, Deng S, Ong MS, Arfuso F, Yap CT, Goh BC, Sethi G, Huang RY, Shen HM, Manjithaya R, Kumar AP. Dual role of autophagy in hallmarks of cancer. *Oncogene* 2018; **37**: 1142-1158 [*RCA*] [PMID: 29255248 DOI: 10.1038/s41388-017-0046-6] [FullText] [Citation(s) in *RCA*: 415] [Article Influence: 51.9]
 - 24 **Marsden VS**, O'Connor L, O'Reilly LA, Silke J, Metcalf D, Ekert PG, Huang DC, Cecconi F, Kuida K, Tomaselli KJ, Roy S, Nicholson DW, Vaux DL, Bouillet P, Adams JM, Strasser A. Apoptosis initiated by Bcl-2-regulated caspase activation independently of the cytochrome c/ Apaf-1/caspase-9 apoptosome. *Nature* 2002; **419**: 634-637 [*RCA*] [PMID: 12374983 DOI: 10.1038/nature01101] [FullText] [Citation(s) in *RCA*: 449] [Article Influence: 19.5]
 - 25 **Levy JMM**, Towers CG, Thorburn A. Targeting autophagy in cancer. *Nat Rev Cancer* 2017; **17**: 528-542 [*RCA*] [PMID: 28751651 DOI: 10.1038/nrc.2017.53] [FullText] [Citation(s) in *RCA*: 1927] [Article Influence: 240.9]
 - 26 **Mizushima N**, Yoshimori T. How to interpret LC3 immunoblotting. *Autophagy* 2007; **3**: 542-545 [*RCA*] [PMID: 17611390 DOI: 10.4161/auto.4600] [FullText] [Citation(s) in *RCA*: 2132] [Article Influence: 118.4]
 - 27 **Porta C**, Paglino C, Mosca A. Targeting PI3K/Akt/mTOR Signaling in Cancer. *Front Oncol* 2014; **4**: 64 [*RCA*] [PMID: 24782981 DOI: 10.3389/fonc.2014.00064] [FullText] [Full Text(PDF)] [Citation(s) in *RCA*: 1128] [Article Influence: 102.5]
 - 28 **Kisen GO**, Tessitore L, Costelli P, Gordon PB, Schwarze PE, Baccino FM, Seglen PO. Reduced autophagic activity in primary rat hepatocellular carcinoma and ascites hepatoma cells. *Carcinogenesis* 1993; **14**: 2501-2505 [*RCA*] [PMID: 8269618 DOI: 10.1093/carcin/14.12.2501] [FullText] [Citation(s) in *RCA*: 96] [Article Influence: 3.0]
 - 29 **Cheung CS**, Chung KK, Lui JC, Lau CP, Hon PM, Chan JY, Fung KP, Au SW. Leachianone A as a potential anti-cancer drug by induction of apoptosis in human hepatoma HepG2 cells. *Cancer Lett* 2007; **253**: 224-235 [*RCA*] [PMID: 17379399 DOI: 10.1016/j.canlet.2007.01.025] [FullText] [Citation(s) in *RCA*: 28] [Article Influence: 1.6]



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