Am J Transl Res 2021;13(6):6211-6220
www.ajtr.org /ISSN:1943-8141/AJTR0132276

Original Article

Acrid-release and bitter-downbearing therapy and banxia xiexin decoction regulate Wnt/β-catenin pathway, inhibit proliferation and invasion, and induce apoptosis in gastric cancer cells

Xiaofen Sun[1], Dewen Xue[2], Kanru Zhang[1], Fang Jiang[3], Duoqiao Li[1]

Departments of [1]Gastroenterology, [2]Nephrology, [3]Outpatient, The 943 Hospital of The Joint Logistics Support Unit of The Chinese People's Liberation Army, Wuwei, Gansu Province, China

Received February 22, 2021; Accepted March 7, 2021; Epub June 15, 2021; Published June 30, 2021

Abstract: Objective: To explore the efficacy of the acrid-release and bitter-downbearing therapy and Banxia Xiexin Decoction (BXD) in treating gastric cancer (GC). Methods: BXD was decocted, and serum containing medicine was prepared from rats. The SNU-16 cells were cultured with different concentrations of BXD serum (25, 50, 100 μL/mL). Then, those were treated with BXD and Wnt/β-catenin pathway activator (LiCl) and divided into three groups: Control group, BXD group and BXD+LiCl group. Activation of the Wnt/β-catenin pathway was detected by immunofluorescence staining, qRT-PCR, and western blot. Cell activity, clone formation, invasion, metastasis and apoptosis in each group were examined by MTT, clone formation test, Transwell and flow cytometry. The oxidative stress measures in cells of each group were tested by an oxidative stress kit. Results: With increasing BXD concentration, the clonogenic ability of cells was inhibited. BXD can inhibit cell activity, clone formation, invasion and metastasis, promote oxidative stress, and induce apoptosis. It can also inhibit the activation of Wnt/β-catenin signaling pathway. A Wnt/β-catenin signaling pathway activator could partially inhibit the action of BXD. Conclusion: BXD participates in GC treatment by inhibiting Wnt/β-catenin signaling pathway, thus inhibiting GC cell activity and clone formation, promoting oxidative stress, and inducing apoptosis.

Keywords: BXD, gastric cancer, Wnt/β-catenin, apoptosis, oxidative stress, activity, invasion and metastasis

Introduction

Gastric cancer (GC) ranks first among malignancies in China, and it occurs in middle-aged people over 50 years old. However, due to the changes of work pressure and lifestyle, the affected population has become younger in recent years [1]. Most GC has no obvious symptoms at an early stage and is easily confused with gastritis, so the early diagnosis rate is low [2]. GC can be treated by surgery, chemotherapy, or targeted therapy. It has been found that many natural compounds can effectively treat GC [3]. Research has shown that pine nut extract can inhibit the proliferation of GC and prostate cancer cells [4]. Acrid-release and bitter-downbearing therapy are used to treat stomach ache caused by mixed cold and heat. Banxia Xiexin decoction (BXD) is the agent of this therapy, and is often used in the treatment of gastrointestinal diseases [5]. Many studies have shown that BXD has a good effect in the treatment of gastrointestinal diseases [6, 7]. Some studies have found that BXD also plays a role in cancer treatment. Wang *et al.* found that BXD alone had a good effect on advanced liver cancer, and there is no adverse reaction in the treatment [8]. However, its role in GC is still vague.

Wnt/β-catenin is a common signaling pathway, that is involved in the development of many diseases. Research has shown that oncolytic adenovirus can inhibit the migration and invasion of colorectal cancer cells and the differentiation of tumor stem cells by inhibiting the Wnt/β-catenin/epithelial-mesenchymal transition signaling pathway [9]. Wnt/β-catenin signaling

pathway is usually activated in prostate cancer, and is related to cancer cell metastasis and drug resistance [10]. Up-regulating miR-6838-5p can inhibit the malignant behavior of GC cells by targeting GPRIN3 to inhibit the Wnt/β-catenin signaling pathway [11]. The GGT gene of Helicobactor pylori promotes GC by up-regulating TET1 and activating the Wnt/β-catenin signaling pathway [12]. Key components of BXD, such as jatrorrhizine and rutin, have been proven to play an anticancer part by inhibiting Wnt pathway activation [13, 14].

The purpose of this research is to probe the mechanism of BXD regulation of GC cell proliferation and apoptosis through the Wnt/β-catenin signaling pathway, and find a new treatment for GC.

Methods and materials

BXD decoction

BXD was prepared according to the prescription dosage of decoction given in *Treatise on Febrile Diseases*, and the dosage was converted in light of *Science of Prescription*. The required medicines are as follows: Pinellia ternata 12 g, Scutellaria baicalensis, ginseng, dried ginger, licorice, four Chinese-dates each 9 g, and Coptis chinensis 3 g. First of all, 8 times volume of water was added, decocted for 1.5 h, and filtered. Next, 6 times volume of water was added, decocted for 1 h, and filtered. Next, the decoctions were collected. Finally, they were processed into lyophilized powder and dissolved.

Preparation of medicated serum

The drug-containing serum was prepared from 25 Wistar rats, weighing 223.15±12.24 g, with an average age of 6 weeks. Rats were fed adaptively for 1 day, and then medicine was injected. Those in the BXD group were given 10 mL/kg BXD twice a day for 3 consecutive days, while the control group was given the same dose of normal saline. Afterwards, they were fasted 12 h before the last administration. On the 4th day, blood was taken from the aorta 1 h later. The operation is as follows: Rats were anesthetized with 40 mg/kg 3% pentobarbital. They were supine on the plate, limbs were fixed, and the abdomen was disinfected. The abdominal skin was cut, and the internal organs were separated. The back was exposed, and a film was separated. The abdominal aorta was exposed, and the blood collection needle was used to collect blood from the abdominal aorta. Blood was allowed to stand for 4 h, centrifuged at 3,500 rpm for 30 min, and serum was collected, filtered, sterilized, and stored at -80°C [15]. This animal experiment was approved by the Ethics Committee. It follows the principles of the guidelines for the care and use of experimental animals.

Cell culture and grouping

Human GC cell line SNU-16 was from the cell bank of Beijing Academy of Sciences (China). The cells were placed in a RPMI1640 medium containing 10% fetal bovine serum (PM1501-10B, Procell, Wuhan, China), and placed in an incubator containing 5% CO_2 at 37°C. The medium was changed every 3 days. When the cells were growing logarithmically, they were taken out for the next experiment.

The experimental groups were divided into Control group (SNU-16 cells were cultured without any treatment), BXD group (SNU-16 cells were cultured with 25 μL BXD medicated serum), and BXD+LiCl group (SNU-16 cells were cultured with 25 μL BXD medicated serum and 25 μL LiCl).

Detection of β-catenin nuclear translocation by immunofluorescence staining

The expression of β-catenin was tested by immunofluorescence staining. The cells were inoculated in a 24-well culture plate, cultivated in an incubator for 24 h, taken out and washed with PBS. After that, they were permeabilized with 0.2% TritionX-100 (Solarbio, Beijing, China), sealed with 5% sheep serum (Yi Lai Sa Biology, China) for 30 min, then added dropwise with primary antibody β-catenin (8480S, Cell Signaling Technology, 1:100), and finally cultivated at 4°C all night. The cells were washed with PBS, incubated with second antibody IgG (ab172730, Abcam, 1:2,000) for 90 min, stained with DAPI (Solarbio, Beijing, China) and cleaned with distilled water. Subsequently, the stained cells were observed by fluorescence microscopy (IX71, Olympus, Japan).

Clone formation assay

The cells were inoculated on a 6-well plate, and the control and experimental wells of the plate

were set, and 500 cells were inoculated in each hole. Subsequently, 0, 25, 50 and 100 μL/mL BXD were added to the experimental wells, and then the cells were cultured in an incubator for 14 days. The incubator was set at 37°C and contained 5% CO_2. After that, the cells were fixed with 4% paraformaldehyde and stained with 0.5% crystal violet (G1065, Solarbio, Beijing, China). The number of cell clones was observed using an inverted microscope (GX53, OLYMPUS, China).

MTT

The 96-well plates were planted with 5×10^3 cells per well, and the control wells were set. The experimental wells were divided into three groups; each group was set with 5 replicate wells. The BXD group and BXD+LiCl group were added with 25 μL BXD, 25 μL BXD+25 μL LiCl (40 mmol/L) into the wells respectively. Then, they were incubated in an incubator containing 5% CO_2 at 37°C for 2 h. Altogether 10 μL MTT solution (M1020, Solarbio, Beijing, China) was added to each well and incubated for 4 h, then the supernatant was discarded, and finally 110 μL Formazan solution was added to each well and shaken well. The absorption value (OD value) was measured with the help of a multifunctional microplate reader, the emission wavelength was 490 nm, and the cell survival rate was calculated.

Flow cytometry

Cell apoptosis was detected by the AnnexinV Alexa Fluor448/PI apoptosis detection kit (CA1040, Solarbio, Beijing, China). After being cultured with the above drugs for 24 h, the cells were digested with 0.25% trypsin to prepare a single cell suspension. Then, they were washed with PBS and centrifuged. Afterwards, the supernatant was discarded, and the cells were resuspended with PBS. Next, 200 μL binding buffer was added, and the cells were put in a dark room. Subsequently, 5 μL Annexin V/Alexa Fluor 488 and 10 μL 20 μg/mL propidium iodide solution were added, mixed well and incubated for 5 min. Apoptosis was tested and the apoptosis rate was analyzed by flow cytometry (CytoFLEX, Beckman, USA).

Transwell

Cell invasion and metastasis was tested by the Transwell method. At first, the cells were treated with drugs according to the same method as above, and then they were inoculated into the upper chamber. RPMI1640 medium containing 10% fetal bovine serum (PM150110B, Procell, Wuhan, China) was put into the lower chamber, placed in the incubator for 24 h, taken out, fixed with paraformaldehyde, and stained with crystal violet. The cell metastasis was observed under an inverted microscope. The upper chamber was coated with Matrigel (356234, Shanghai Qcbio Science&Technologies Co., Ltd., China) before the cell invasion test, and other steps were the same as for detection of metastasis.

qRT-PCR

The mRNA expression of β-catenin was detected by qRT-PCR after the cells were cultured with the above drugs. Total RNA was extracted from cells by TRIzol reagent (155960216, Thermo Fisher, USA), and then synthesized into cDNA by high capacity cDNA reverse transcription kit (4374967, Thermo Fisher, USA), and 7500 Fast System (A30299, Thermo Fisher, USA) and PowerUpSYBR Green Master mix (A25741, Thermo Fisher, USA) to complete RNA detection. It was calculated by using $2^{-\Delta\Delta CT}$, and GAPDH was used as an internal reference. Primers were synthesized by Shanghai GenePharma Co., Ltd (China), upstream of β-catenin: CGCTAGGCTAGCAGGATAAAGA and downstream: AATCGCTCGCGCTAGCGCTAGCGC, upstream of GAPDH: CTGATCAGCGATCAGCGCGATCATCG, and downstream: CGCTCGGGTCGGGAACGCCGCTA.

Western blot

The expression of β-catenin protein in cells treated with the above drugs was detected. Each group of cells were lysed by RIPA cell lysate (SW104-01, SevenBiotech, Beijing, China), and the protein concentration was tested by the BCA protein assay kit (PC0020, Solarbio, Beijing, China). The proteins were separated by 12% sodium dodecyl sulfate polyacrylamide gel electrophoresis. Then, they were transferred to PVDF membrane (ISEQ00010, Solarbio, Beijing, China) and separated by 5% skim milk. The primary antibodies β-catenin (1:1,000, ab32572, Abcam, UK) and GAPDH (1:1000, ab8245, Abcam, UK) were incubated with the membrane overnight at 4°C. Horseradish peroxidase labeled goat anti-rabbit IgG (1:1,000, ab172730, Abcam, UK) was incu-

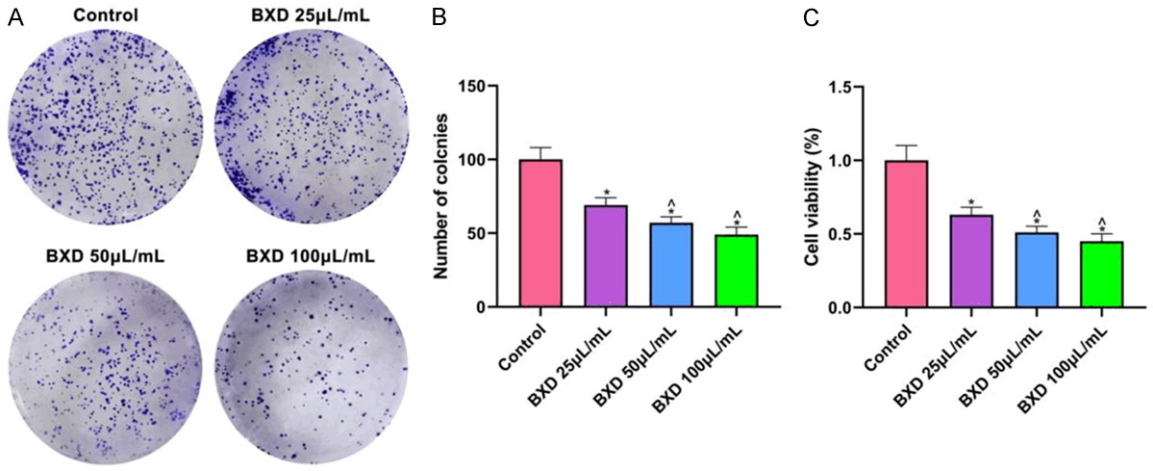

Figure 1. BXD inhibits cell clone formation and activity. A: Results of cell clonal formation ability in each group; B: Quantification of cell clone number in each group; C: Detection of cell activity in each group. Compared with Control group, *P<0.05, and compared with BXD 25 μL/mL group, ^P<0.05. BXD: Banxia Xiexin Decoction.

bated with the membrane for 2 h. TBST was washed for 10 min, chemiluminescence (ECL) (SW2030, Solarbio, Beijing, China) was added to the membrane, and the protein was observed by gel imaging system. The protein expression was counted by Image J software, and the result was expressed by the ratio of target protein band gray value/GAPDH protein band gray value.

Detection of oxidative stress parameters

After the cells were treated with the above drugs for 24 h, the contents of MDA, SOD and GSH-PX were measured by malondialdehyde (MDA) content detection kit (BC0025, Solarbio, Beijing, China), superoxide dismutase (SOD) activity detection kit (BC0170, Solarbio, Beijing, China) and glutathione peroxidase (GSH-PX) activity detection kit (BC1190, Solarbio, Beijing, China). Detection was carried out according to kit instructions.

Statistical analysis

The experiments were repeated three times. The data were analyzed by SPSS23.0 software and expressed with mean ± standard deviation. The differences between the two groups were compared by Student's *t* test, and those between multiple groups were compared by one-way ANOVA combined with Tukey test. P<0.05 indicated a significant difference.

Results

With increasing BXD concentration, cell clone formation ability decreases

The SNU-16 cells were treated with BXD medicated serum with different concentrations, and their clone formation and activity were observed by clone formation test and MTT assay. Compared to cells cultured without BXD medicated serum, the clonogenic ability and activity in 25, 50 and 100 μL/mL groups were inhibited, and there was a dose-response relationship at 25 μL and 50 μL (**Figure 1**, all P<0.05). After that, 25 μL/mL BXD began to show obvious differences, so it was selected for subsequent experiments. These results indicate that BXD can effectively inhibit the cloning of GC cells.

BXD can inhibit activation of the Wnt/β-catenin signaling pathway and inhibit ability of cell clone formation. Adding Wnt/β-catenin signaling pathway activator can partially reverse BXD's effect

First, the activation effect of LiCl on β-catenin was verified. The results manifested that LiCl could promote the nuclear implantation expression of β-catenin and activate the Wnt/β-catenin signal pathway (**Figure 2**). After that, the expression of β-catenin in cells of each group was detected by immunofluorescence. The results revealed that compared with the

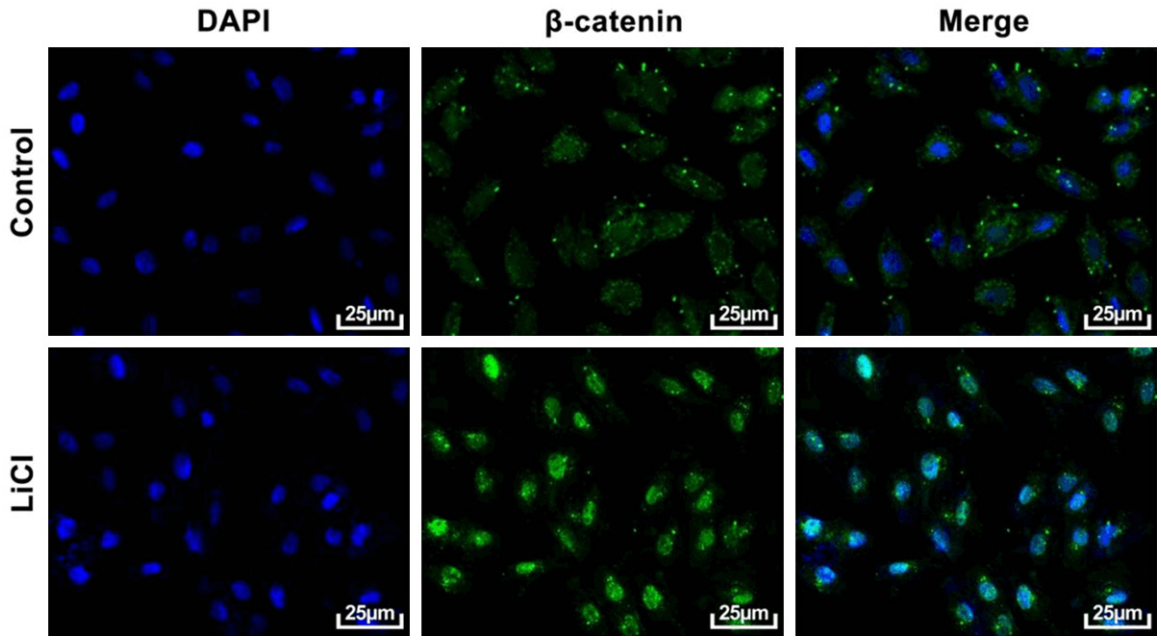

Figure 2. Verification of activation effect of LiCl on β-catenin (400×).

Control group, the fluorescence intensity of β-catenin in BXD group was inhibited, and this effect was partially reversed by adding LiCl (**Figure 3A**). Compared with the Control group, the clonal formation of cells in BXD group decreased, while that in BXD+LiCl group increased (**Figure 3B**, all P<0.05). The experiment indicated that BXD could inhibit the formation of cell clones, while LiCl could partially reverse the effect of BXD. The above experiments indicate that BXD may inhibit cell clone formation by inhibiting the Wnt/β-catenin signaling pathway.

BXD can inhibit cell activity and induce apoptosis, and Wnt/β-catenin signaling pathway activator can partially reverse BXD's effect

MTT assay showed that compared with the Control group, cell activity of the BXD group was decreased, and compared with BXD group, the activity of the BXD+LiCl group was increased (**Figure 4A**, all P<0.05). The apoptosis in each group was detected by flow cytometry. The experimental results manifested that compared with the Control group, apoptosis in the BXD group increased, while compared with BXD group, apoptosis in the BXD+LiCl group was decreased (**Figure 4B**, all P<0.05). The above experiments signify that BXD can inhibit apoptosis induced by cell activity, while LiCl partially reverses this effect.

BXD can inhibit cell invasion and metastasis, and Wnt/β-catenin signaling pathway activator can partially reverse BXD's effect

Transwell test revealed that compared with the Control group, invasion and metastasis of cells in the BXD group were inhibited, and compared with the BXD group, those in the BXD+LiCl group were promoted (**Figure 5A**, **5B**, all P<0.05). The above experiments reveal that BXD can inhibit cell invasion and metastasis, while LiCl can partially reverse this effect.

BXD can promote oxidative stress in cells, and Wnt/β-catenin signaling pathway activator can partially reverse BXD's effect

The contents of MDA, SOD, and GSH-PX in cells of each group were detected. The experiment showed that compared with the Control group, the contents of MDA, SOD and GSH-PX of the BXD group increased, and compared with the BXD group, the above measures of the BXD+LiCl group were partially reversed (**Figure 6**, all P<0.05). This shows that BXD can promote the oxidative stress response of cells, while Wnt/β-catenin signaling pathway activator partially reverses this effect.

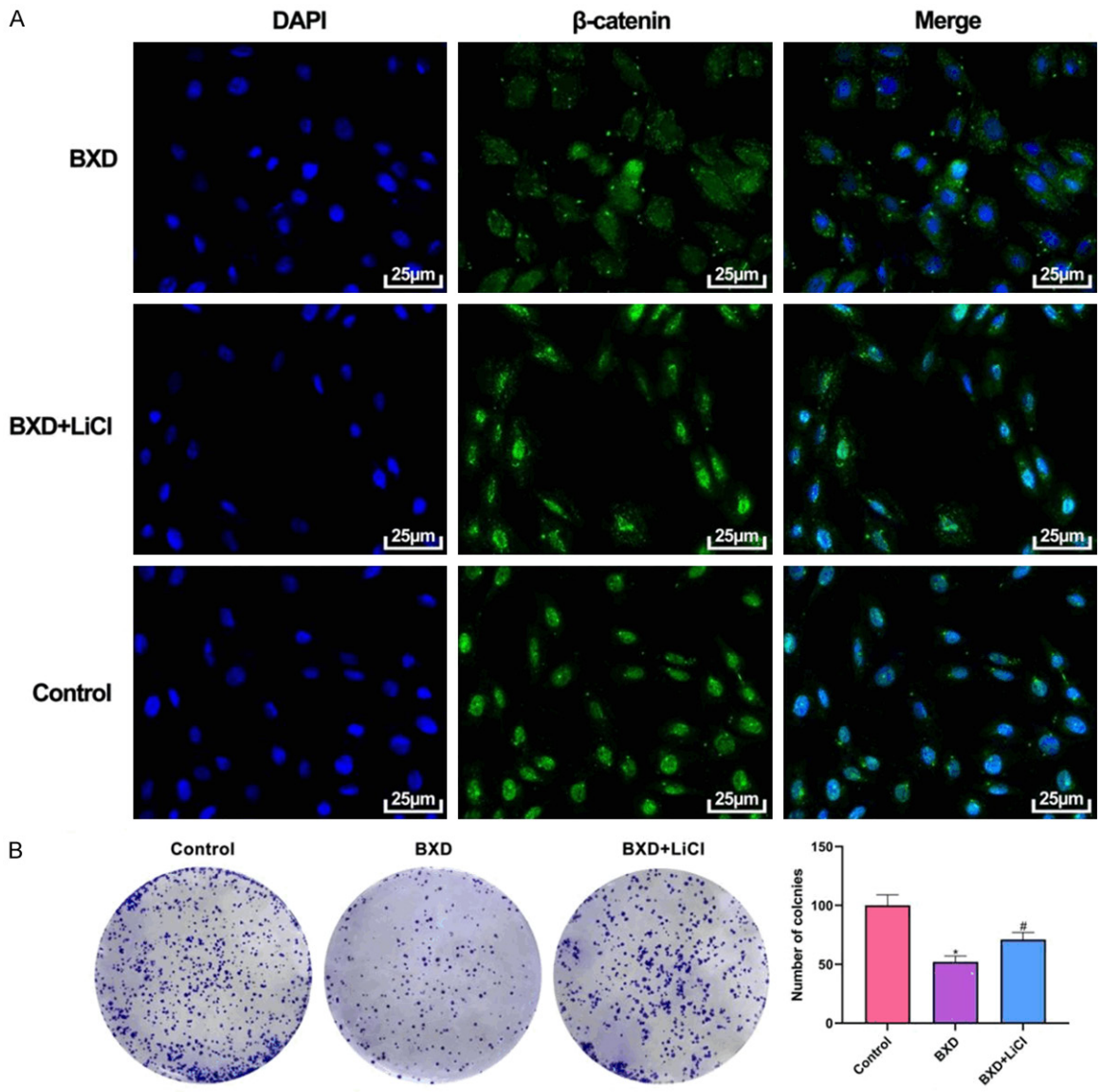

Figure 3. BXD effectively inhibits cell clone formation. A: Detection of nuclear translocation expression of β-catenin by immunofluorescence (400×); B: Test results of cell clone formation ability of each group. Compared with Control group, *P<0.05; compared with BXD group, #P<0.05. BXD: Banxia Xiexin Decoction.

Discussion

BXD, recorded in *Treatise on Febrile Diseases*, is a representative drug of acrid-release and bitter-downbearing therapy. It can eliminate phlegm, strengthen the spleen, clear heat, and relieve dampness. Research has found that BXD can activate the PI3K/Akt signaling pathway, plays a neuroprotective role in APPswe/PS1dE9 transgenic mice, and may have an effect on the treatment of Alzheimer's disease [16]. Another study found that BXD combined with cisplatin in the treatment of lung cancer can improve cisplatin-induced apoptosis of cancer cells [17]. The therapeutic effect of BXD on gastrointestinal diseases was confirmed. This research explored the specific mechanism of BXD in GC [18]. Oxidative stress refers to a state in which the oxidation reaction is more than antioxidation, and oxidation and antioxidation are out of balance, which will lead to an increase in secretion of inflammatory factors and promote cell aging and body diseases. This is a feature of tumor cells and can run through the whole process of tumor development [19]. MDA is a common indicator of oxida-

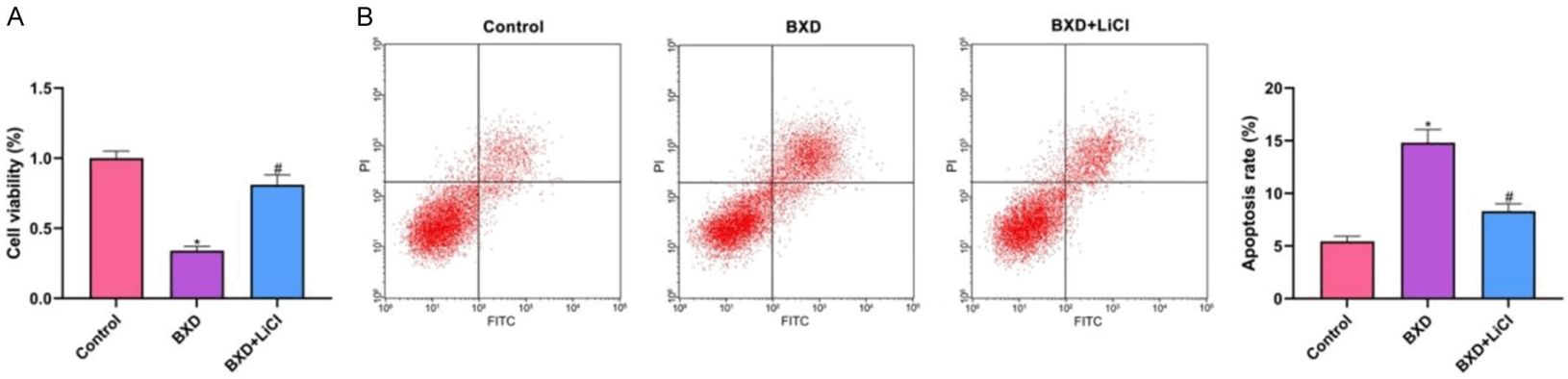

Figure 4. BXD can inhibit cell activity and induce apoptosis. A: Cell activity of each group is tested by MTT assay; B: Apoptosis in each group is tested by flow cytometry. Compared with Control group, *P<0.05; compared with BXD group, #P<0.05. BXD: Banxia Xiexin Decoction.

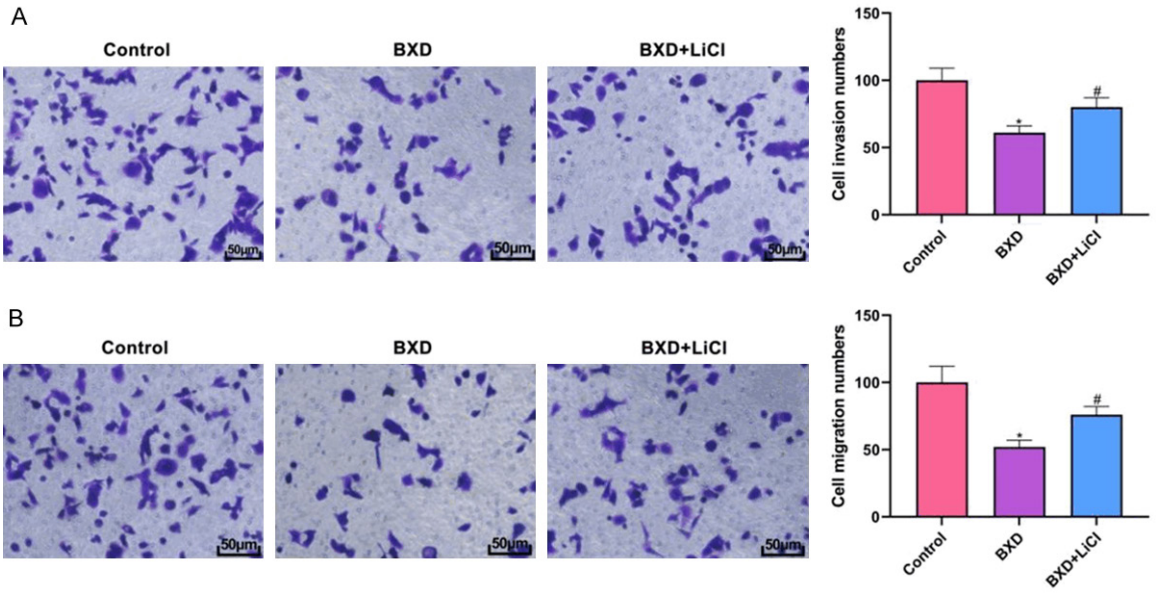

Figure 5. BXD combined with 5-FU can inhibit cell invasion and metastasis. A: Cell invasion of each group analyzed by Transwell test (200×); B: Cell metastasis in each group is assessed by Transwell test (200×). Compared with Control group, *P<0.05; compared with BXD group, #P<0.05. BXD: Banxia Xiexin Decoction.

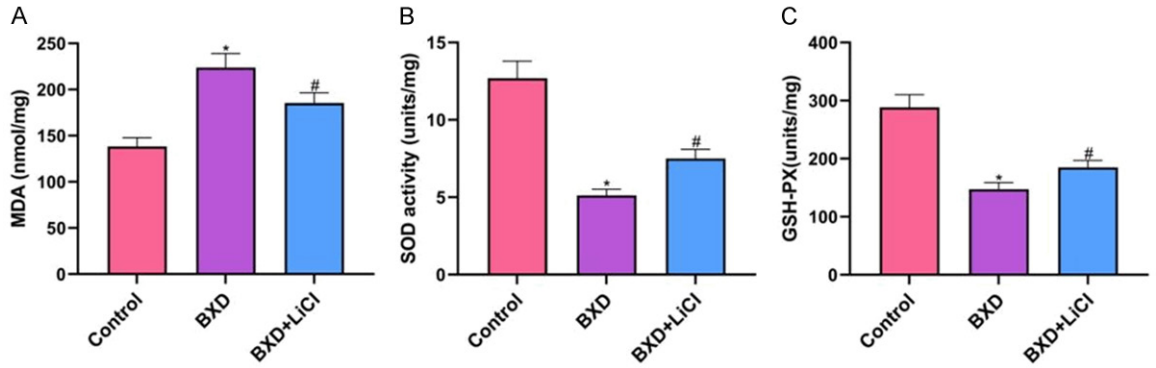

Figure 6. The Wnt/β-catenin signaling pathway activator partially reverses BXD's effect. A: MDA content test results; B: SOD activity test results; C: Detection results of GSH-PX activity. Compared with Control group, *P<0.05; compared with BXD group, #P<0.05. BXD: Banxia Xiexin Decoction; MDA: malondialdehyde; SOD: superoxide dismutase; GSH-PX: glutathione peroxidase.

tion, and can reflect the degree of peroxidation in vivo. SOD and GSH-PX are antioxidant enzymes that can protect the structure and function of the cell membrane [20]. This research found that BXD could promote the oxidative stress response of GC cells. BXD can inhibit cell vitality, clone formation, proliferation and invasion, promote oxidative stress, and induce apoptosis.

Wnt/β-catenin is a highly conserved signaling pathway. Wnt gene was first found in mouse breast cancer, which could induce β-catenin to accumulate in cytoplasm, form a classic pathway with β-catenin, and participate in disease development [21]. Knockout of mll1 gene can reportedly inhibit the activation of Wnt/β-catenin pathway and further prevent the formation of intestinal stem cell adenoma of Lgr5 [22]. Circ_104348 promotes the development of liver cancer by regulating the miR-187-3p/RTKN2 axis and activating the Wnt/β-catenin pathway [23]. Research has also found that Wnt/β-catenin pathway can promote the proliferation and invasion of non-small cell lung cancer cells [24]. Therefore, activation of the Wnt/β-catenin pathway generally plays a role in tumor promotion. In this research, we found

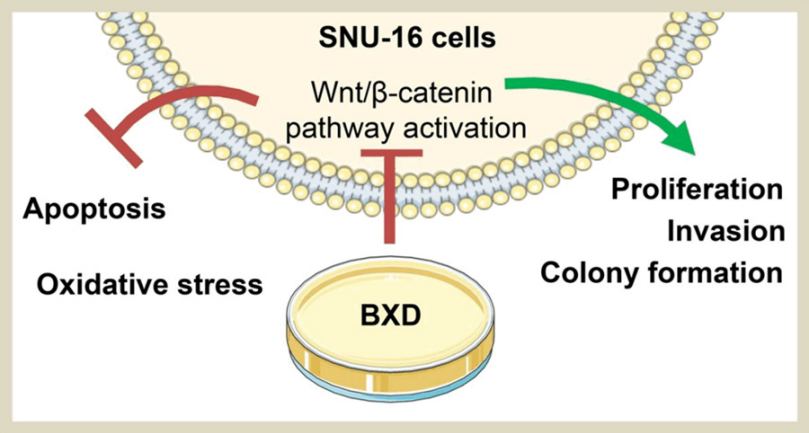

Figure 7. Mechanism diagram of suppressing malignant biologic behavior of gastric cancer through Wnt/β-catenin signaling pathway by acrid-release and bitter-downbearing therapy and Banxia Xiexin Decoction.

that the Wnt/β-catenin pathway is highly activated in GC cells. BXD can inhibit Wnt/β-catenin pathway activation, inhibit GC cell activity, invasion and metastasis, clone formation, and promote oxidative stress-induced apoptosis. On the basis of BXD, Wnt/β-catenin pathway activator can improve GC cell activity, invasion and metastasis, clone formation, and inhibit oxidative stress and apoptosis. The activation of Wnt/β-catenin pathway may be related to a series of malignant behaviors such as GC cell activity, clone formation, invasion and metastasis, and oxidative stress, and the inhibition of BXD on GC cells may be realized by blocking Wnt/β-catenin pathway.

Nevertheless, the study has limitations. At present, there are no clinical cases or further verification of BXD's role in animals. In addition, there are differences between animal and human experiments. Only the mechanism of action in vitro model is discussed, and the above defects need to be improved. However, the study does provides a basis for BXD to treat GC. The mechanism diagram is shown in **Figure 7**.

Disclosure of conflict of interest

None.

Address correspondence to: Xiaofen Sun, Department of Gastroenterology, The 943 Hospital of The Joint Logistics Support Unit of The Chinese People's Liberation Army, No. 3 Beiguan East Road, Liangzhou District, Wuwei 733000, Gansu Province, China. Tel: +86-1879738-9896; E-mail: sunxiaofen-7979@163.com

References

[1] Kono Y, Kanzaki H, Iwamuro M, Kawano S, Kawahara Y and Okada H. Reality of gastric cancer in young patients: the importance and difficulty of the early diagnosis, prevention and treatment. Acta Med Okayama 2020; 74: 461-466.

[2] Li S, Zhang F, Li J, Hu X, Zhao W, Zhang K and Li J. The role of the Epstein-Barr virus-encoded BARF1 gene expressed in human gastric epithelial cells. Turk J Gastroenterol 2020; 31: 775-781.

[3] Nakonieczna S, Grabarska A and Kukula-Koch W. The potential anticancer activity of phytoconstituents against gastric cancer-a review on in vitro, in vivo, and clinical studies. Int J Mol Sci 2020; 21: 8307.

[4] Sallai R, Salu B, Silva-Lucca R, Alves F, Napoleão T, Maria P, Paiva G, Da R, Ferreira S, Sampaio M, Oliva M and Oliva V. Biotechnological potential of araucaria angustifolia pine nuts extract and the cysteine protease inhibitor AaCI-2S. Plants 2020; 9: 1676.

[5] Cao Y, Zheng Y, Niu J, Zhu C, Yang D, Rong F and Liu G. Efficacy of Banxia Xiexin decoction for chronic atrophic gastritis: a systematic review and meta-analysis. PLoS One 2020; 15: e0241202.

[6] Yuan T, Li P and Jia B. A meta-analysis of Xin Kai bitter method in the treatment of functional dyspepsia. Ann Palliat Med 2020; 9: 993-1003.

[7] Ji Q, Yang Y, Song X, Han X and Wang W. Banxia Xiexin decoction in the treatment of chronic atrophic gastritis: a protocol for systematic review and meta-analysis. Medicine (Baltimore) 2020; 99: e22110.

[8] Wang L, Ke J, Wang C, Li Y, Wu G, Ding Q, Luo Q, Cai R, Lv P, Song T and Xiong S. Efficacy and safety of banxia xiexin decoction, a blended traditional chinese medicine, as monotherapy for patients with advanced hepatocellular carcinoma. Integr Cancer Ther 2020; 19: 1534735420942587.

[9] Xiao B, Zhang L, Liu H, Fang H, Wang C, Huang B, Liu X, Zhou X and Wang Y. Oncolytic adenovirus CD55-Smad4 suppresses cell proliferation, metastasis, and tumor stemness in

colorectal cancer by regulating wnt/β-catenin signaling pathway. Biomedicines 2020; 8: 593.
[10] Pan KF, Lee WJ, Chou CC, Yang YC, Chang YC, Chien MH, Hsiao M and Hua KT. Direct interaction of β-catenin with nuclear ESM1 supports stemness of metastatic prostate cancer. EMBO J 2021; 40: e105450.
[11] Zhou W, Ding X, Jin P and Li P. miR-6838-5p affects cell growth, migration, and invasion by targeting GPRIN3 via the Wnt/β-catenin signaling pathway in gastric cancer. Pathobiology 2020; 87: 327-337.
[12] Meng L, Shi H, Wang Z, Fan M, Pang S and Lin R. The gamma-glutamyltransferase gene of Helicobacter pylori can promote gastric carcinogenesis by activating Wnt signal pathway through up-regulating TET1. Life Sci 2021; 267: 118921.
[13] Wang P, Gao XY, Yang SQ, Sun ZX, Dian LL, Qasim M, Phyo AT, Liang ZS and Sun YF. Jatrorrhizine inhibits colorectal carcinoma proliferation and metastasis through Wnt/β-catenin signaling pathway and epithelial-mesenchymal transition. Drug Des Devel Ther 2019; 13: 2235-2247.
[14] Jia Y, Chen L, Guo S and Li Y. Baicalin induced colon cancer cells apoptosis through miR-217/DKK1-mediated inhibition of Wnt signaling pathway. Mol Biol Rep 2019; 46: 1693-1700.
[15] Li DF, Zhao WP, Liu XP, Ming HX, Sun SB and Zhang W. Effects of BanXia XieXin tang medicated serum on the proliferation of human gastric cancer cells through peritoneal metastasis. West J Tradit Chin 2014; 27: 14-16.
[16] Chen F, He Y, Wang P, Wei P, Feng H, Rao Y, Shi J and Tian J. Banxia Xiexin decoction ameliorated cognition via the regulation of insulin pathways and glucose transporters in the hippocampus of APPswe/PS1dE9 mice. Int J Immunopathol Pharmacol 2018; 32: 2058738418780066.
[17] Kim HR, Lee GS, Kim MS, Ryu DG, So HS, Moon HC, Lee YR, Yang SH and Kwon KB. Effects of banxia xiexin decoction on cisplatin-induced apoptosis of human A549 lung cancer cells. Chin J Integr Med 2018; 24: 436-441.
[18] Chen G, Yang Y, Liu M, Teng Z, Ye J, Xu Y, Cai X, Cheng X, Yang J, Hu C, Wang M and Cao P. Banxia xiexin decoction protects against dextran sulfate sodium-induced chronic ulcerative colitis in mice. J Ethnopharmacol 2015; 166: 149-156.
[19] de Oliveira Silva E and Batista R. Ferulic acid and naturally occurring compounds bearing a feruloyl moiety: a review on their structures, occurrence, and potential health benefits. Compr Rev Food Sci Food Saf 2017; 16: 580-616.
[20] Wang Y, Chen X, Huang Z, Chen D, Yu B, Yu J, Chen H, He J, Luo Y and Zheng P. Dietary ferulic acid supplementation improves antioxidant capacity and lipid metabolism in weaned piglets. Nutrients 2020; 12: 3811.
[21] Tao C, Luo J, Tang J, Zhou D, Feng S, Qiu Z, Putti TC, Xiang T, Tao Q, Li L and Ren G. The tumor suppressor Zinc finger protein 471 suppresses breast cancer growth and metastasis through inhibiting AKT and Wnt/β-catenin signaling. Clin Epigenetics 2020; 12: 173.
[22] Grinat J, Heuberger J, Vidal RO, Goveas N, Kosel F, Berenguer-Llergo A, Kranz A, Wulf-Goldenberg A, Behrens D, Melcher B, Sauer S, Vieth M, Batlle E, Stewart AF and Birchmeier W. The epigenetic regulator Mll1 is required for Wnt-driven intestinal tumorigenesis and cancer stemness. Nat Commun 2020; 11: 6422.
[23] Huang G, Liang M, Liu H, Huang J, Li P, Wang C, Zhang Y, Lin Y and Jiang X. CircRNA hsa_circRNA_104348 promotes hepatocellular carcinoma progression through modulating miR-187-3p/RTKN2 axis and activating Wnt/β-catenin pathway. Cell Death Dis 2020; 11: 1065.
[24] Yu JE, Ju JA, Musacchio N, Mathias TJ and Vitolo MI. Long noncoding RNA DANCR activates Wnt/β-catenin signaling through MiR-216a inhibition in non-small cell lung cancer. Biomolecules 2020; 10: 1646.