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Research on the Protective Effects of Different Chinese Medicine Compounds on Gefitinib-Induced Hepatotoxicity

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Abstract

Objective Our objective was to screen drugs with good protective effects on gefitinib-induced hepatotoxicity.

Methods Fifty-four specific pathogen-free-grade male mice of the Institute of Cancer Research were randomly divided into a normal group, gefitinib group, glutathione group, ligustrazine group, silymarin group, glycyrrhizic acid group, baicalin group, paeoniflorin group, and matrine group, with six mice in each group. Except for the normal group, the remaining groups of mice were intragastrically administered 400 $mg \cdot kg^{-1}$ of gefitinib for 16 days to induce liver injury. Mice in each treatment group were intragastrically administered 100 $mg \cdot kg^{-1}$ of the corresponding drug 30 minutes after gefitinib administration each day. The normal group and model group mice were intragastrically administered with an equal volume of 0.5% carboxymethylcellulose sodium (CMC-Na), and the administration volume was 10 $mg \cdot kg^{-1}$. Then, 30 minutes after the last administration, blood was collected from the retro-orbital venous plexus, and the mice were killed by cervical dislocation to obtain liver weight and calculate the liver index. Liver pathological changes were observed by hematoxylin–eosin (HE) staining; the levels of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) in serum were detected using biochemical kits. AML12 cells were cultured in a medium containing drugs for 30 minutes. Except for the normal group, the remaining groups were induced cell damage with 20 $\mu mol \cdot L^{-1}$ gefitinib for 24 hours. Cell viability was detected using a cell counting kit-8, and the levels of ALT, AST, and lactate dehydrogenase (LDH) in the cell culture supernatant were measured using biochemical kits.

Results Animal experiments showed that compared with the gefitinib group, glycyrrhizic acid and baicalin significantly increased the body weight of mice ($p < 0.01$) and decreased the liver index and levels of ALT and AST ($p < 0.05$); ligustrazine and silymarin

Keywords
- hepatotoxicity
- gefitinib
- hepatoprotective effect
- traditional Chinese medicine
- glycyrrhizic acid

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significantly increased the body weight of mice ($p < 0.01$) and decreased the level of AST ($p < 0.05$); paeoniflorin and matrine significantly decreased the levels of ALT and AST ($p < 0.01$). HE staining showed that the liver tissue of mice in the gefitinib group presented a large number of inflammatory cell infiltrations and disordered arrangement of hepatic cords; glutathione, glycyrrhizic acid, baicalin, paeoniflorin, and matrine significantly alleviated pathological damage to mouse liver tissue. Cell experiments showed that all drugs could alleviate gefitinib-induced damage to AML12 cells to varying degrees, among which glycyrrhizic acid, baicalin, and paeoniflorin had better protective effects, with cell survival rates increased to 96.4, 81.1, and 78.2%, respectively. Compared with the gefitinib group, glycyrrhizic acid significantly reduced the levels of ALT, AST, and LDH ($p < 0.05$); silymarin, baicalin, and paeoniflorin significantly reduced the levels of ALT and LDH ($p < 0.05$).

Conclusion Glycyrrhizic acid, baicalin, and paeoniflorin all have good protective effects against gefitinib-induced hepatotoxicity, among which glycyrrhizic acid has the best effect.

Introduction

Drug-induced liver injury (DILI) has gradually become an important clinical challenge, with anticancer drugs being one of the main causes of drug-induced liver damage. Gefitinib is the first epidermal growth factor receptor-tyrosine kinase inhibitor, which can block tumor cell proliferation signaling and induce tumor cell apoptosis. It was approved by the Food and Drug Administration in May 2003 for the treatment of advanced non-small cell lung carcinoma (NSCLC), which can significantly prolong patients' progression-free survival for up to 11 years.[1,2] Despite the significant anticancer efficacy of gefitinib, its hepatotoxicity is an important reason limiting its clinical application.[3] Glutathione is commonly used clinically for hepatoprotection against gefitinib-induced liver injury, but due to the unclear hepatotoxic mechanism of gefitinib, it still lacks hepatoprotective drugs with better targeting.

Traditional Chinese medicine (TCM) has played a significant role in hepatoprotection for a long time. Various Chinese herbs such as Chaihu (Bupleuri Radix), Jinyinhua (Lonicerae Japonicae Flos), Gegen (Puerariae Radix), Wuweizhi (Schisandrae Chinensis Fructus), Huangqin (Scutellariae Radix), and Baishao (Paeoniae Radix Alba) have good hepatoprotective effects.[4] Moreover, the hepatoprotective effects of many natural compounds isolated from Chinese herbs such as silymarin,[5] glycyrrhizic acid,[6] and baicalin[7] have been widely confirmed, becoming an important source for the development of new hepatoprotective drugs in clinical practice. In order to screen for drugs with better protective effects against gefitinib-induced hepatotoxicity, this study selected six natural compounds, including ligustrazine, silymarin, glycyrrhizic acid, baicalin, paeoniflorin, and matrine, which have been clearly demonstrated to have hepatoprotective effects in numerous studies. The protective effects of these compounds on gefitinib-induced hepatotoxicity were comprehensively compared through in vivo and in vitro experiments, providing experimental evidence for the clinical application of related hepatoprotective drugs.

Materials

Experimental Animals and Cells

A total of 54 specific pathogen-free-grade male mice of the Institute of Cancer Research, aged 6 to 8 weeks with a body weight of 18 to 22 g, were purchased from Beijing Vital River Laboratory Animal Technology Co., Ltd., with production license number: SCXK (Beijing) 2016-0006. The mice were housed under standard conditions, with a temperature of 23 to 25 °C, humidity of 40 to 60%, and a 12-hour light-dark cycle (8:30–20:30), with free access to water and food. All animal experimental procedures were approved by the Animal Ethics Committee of Henan University of Chinese Medicine (DWLL201903018).

12 normal liver cells of Alpha mouse liver (American Type Culture Collection, Catalog No: CRL-2254).

Drugs and Reagents

Following are the details of drugs and reagents: gefitinib (purity 99%), ligustrazine (purity 98%), baicalin (purity 95%), and reduced glutathione (purity 98%) (Shanghai Aladdin Biochemical Technology Co., Ltd., China, Catalog No: G125799, T111263, B110211, and G105426); glycyrrhizic acid (purity 98%), paeoniflorin (purity 95%), matrine (purity 98%), and silymarin (purity 80%) (Dalian Meilun Biotech Co., Ltd., China, Catalog No: MB6163-1, MB1712-1, MB5477, MB5976); alanine aminotransferase (ALT) assay kit and aspartate aminotransferase (AST) assay kit (Nanjing Jiancheng Bioengineering Institute, China, Catalog No: C009, C010); DMEM/F12 culture medium and fetal bovine serum (Thermo Fisher Scientific, United States, Catalog No: 11320033, A5669701); penicillin-streptomycin solution and trypsin (Hyclone, United States, Catalog No: SV30010,

SV30031); dimethylsulfoxide (DMSO), dexamethasone, and insulin-transferrin-selenium liquid culture supplement (Sigma, United States, Catalog No: 34943, 265005, 13146); and cell counting kit-8 (CCK-8) cell proliferation and cytotoxicity assay kit (Dojindo Laboratories, Japan, Catalog No: CK04).

Instruments

Imark enzyme-linked immunosorbent assay analyzer (Bio-Rad, United States); RM2125 slicer (Leica, Germany); 90i microscope (Nikon, Japan); FRESCO21 centrifuge, Countes 3 cell counter, 3111 CO_2 incubator (Thermo Fisher Scientific, United States).

Methods

Establishment of Animal Models and Drug Administration

After 1 week of adaptation to the environment, mice were randomly divided into nine groups: normal group, gefitinib group, glutathione group, ligustrazine group, silymarin group, glycyrrhizic acid group, baicalin group, paeoniflorin group, and matrine group, with six mice in each group. The drugs were suspended in 0.5% CMC-Na. The mice in the normal group were intragastrically administered an equal volume of 0.5% CMC-Na, while the remaining groups were intragastrically administered gefitinib solution at a dose of 400 $mg{\cdot}kg^{-1}$ for 16 consecutive days to establish a liver injury model. Additionally, 30 minutes after gefitinib administration each day, the corresponding drug was intragastrically administered at a dose of 100 $mg{\cdot}kg^{-1}$ to each group, with a dosage volume of 10 $mg{\cdot}kg^{-1}$. Both normal and model groups were intragastrically administered an equal volume of 0.5% CMC-Na.

Measurement of Liver Index

Thirty minutes after the final drug administration, blood was collected from the retro-orbital venous plexus, and the mice were killed by cervical dislocation. Liver tissues were collected on ice, washed with normal saline, dried, and weighed to calculate the liver index.

$$\text{Liver index} = (\text{liver weight}/\text{body weight}) \times 100\%.$$

Liver Function Tests

Blood was collected from the mice, centrifuged at $4{,}000 \times g$ for 10 minutes, and the upper serum layer was obtained. The levels of ALT and AST in the serum were measured using a biochemical assay kit according to the manufacturer's instructions.

Hematoxylin–Eosin Staining

After collecting blood from the mice, they were killed by cervical dislocation and liver tissues were collected. Liver tissues of approximately 0.5 cm × 0.5 cm were fixed in 10% formalin, embedded in paraffin, sectioned (5 μm), and subjected to HE staining to observe pathological changes in liver tissue under a light microscope.

Cell Culture

AML12 mouse normal liver cells were cultured in DMEM/F12 medium supplemented with 10% fetal bovine serum, 1% penicillin-streptomycin solution, 1% ITS liquid medium supplement, and 0.2 $g{\cdot}L^{-1}$ dexamethasone at 37 °C in a 5% CO_2 cell culture incubator. All drugs were dissolved in 0.1% DMSO and diluted to the appropriate concentrations in a serum-free medium.

Determination of Cytotoxicity of Drugs

AML12 cells were seeded in a 96-well plate at a density of 8×10^3 cells per well. After 24 hours of culture, the old medium was removed, and medium containing drugs was added to achieve final concentrations of 0, 10, 20, 40, 80, 160, 320, and 640 $\mu mol{\cdot}L^{-1}$. The cells were further cultured for 24 hours, and cell viability was measured using a CCK-8 assay kit. Each group had six replicate wells, and the experiment was repeated three times.

Protective Effects of Drugs on Gefitinib-Induced Cell Damage

AML12 cells were seeded in a 96-well plate at a density of 8×10^3 cells per well. After 24 hours of culture, the culture medium was replaced. The normal group and the gefitinib group were replaced with fresh medium, while the other groups were replaced with medium containing drugs at final concentrations of 10, 20, 40, 80, 160, 320, and 640 $\mu mol{\cdot}L^{-1}$. After 30 minutes of culture, except for the normal group, all other groups were supplemented with 20 $\mu mol{\cdot}L^{-1}$ gefitinib and cultured for 24 hours. Cell viability was measured using a CCK-8 assay, and ALT, AST, and lactate dehydrogenase (LDH) levels in the cell supernatant were determined using an assay kit. Each group had six replicate wells, and the experiment was repeated three times.

Statistical Analysis

Statistical analysis was performed using SPSS 25.0 software, and all data were expressed as mean ± standard deviation ($\bar{X} \pm s$). One-way analysis of variance combined with Dunnett's multiple comparison test was used for data analysis, with $p < 0.05$ indicating statistical significance.

Results

Effects of Different Drugs on the Body Weight and the Liver Index of Mice

As shown in ►**Table 1**, compared with the normal group, gefitinib significantly reduced the body weight of mice ($p < 0.01$) and significantly increased the liver weight ($p < 0.01$) and the liver index ($p < 0.01$). Compared with the gefitinib group, glycyrrhizic acid and baicalin significantly increased the body weight of mice ($p < 0.01$) and significantly decreased the liver index ($p < 0.05$). Ligustrazine and silymarin significantly increased the body weight and the liver weight of mice ($p < 0.01$). However, glutathione, paeoniflorin, and matrine had no significant effect on the body weight and the liver index of mice.

Table 1 Effects of different drugs on the body weight and the liver index of mice ($\bar{X} \pm s$, $n = 6$)

Groups	*n*	Dose (mg·kg^{-1})	Body weight (m/g)	Liver weight (m/g)	Liver index/%
Normal group	6	–	31.40 ± 0.60	1.33 ± 0.05	4.23 ± 0.15
Gefitinib group	6	400	26.50 ± 1.00^{b}	1.73 ± 0.19^{b}	6.65 ± 0.32^{b}
Glutathione group	6	100	26.30 ± 1.30	1.87 ± 0.13	7.02 ± 0.26
Ligustrazine group	6	100	30.90 ± 2.50^{d}	2.21 ± 0.29^{d}	7.14 ± 0.55^{c}
Silymarin group	6	100	34.30 ± 2.50^{d}	2.41 ± 0.26^{d}	7.02 ± 0.42
Glycyrrhizic acid group	6	100	34.90 ± 1.90^{d}	2.16 ± 0.18^{d}	6.05 ± 0.15^{c}
Baicalin group	6	100	32.40 ± 0.90^{d}	1.95 ± 0.19	6.01 ± 0.45^{c}
Paeoniflorin group	6	100	27.00 ± 2.80	1.80 ± 1.32	6.45 ± 0.42
Matrine group	6	100	24.60 ± 1.50	1.67 ± 0.21	6.78 ± 0.58

Note: Compared with the normal group, $^{a}P<0.05$, $^{b}P<0.01$; compared with the gefitinib group, $^{c}P<0.05$, $^{d}P<0.01$.

Table 2 Effects of different drugs on serum ALT and AST levels in mice ($\bar{X} \pm s$, $n = 6$)

Groups	*n*	Dose (mg·kg^{-1})	ALT (U·L^{-1})	AST (U·L^{-1})
Normal group	6	–	14.80 ± 2.40	27.50 ± 0.30
Gefitinib group	6	400	153.00 ± 10.80^{b}	84.70 ± 20.80^{b}
Glutathione group	6	100	104.90 ± 12.60^{d}	55.70 ± 15.10^{d}
Ligustrazine group	6	100	140.40 ± 16.40	63.60 ± 14.20^{c}
Silymarin group	6	100	150.10 ± 10.50	41.90 ± 22.50^{d}
Glycyrrhizic acid group	6	100	92.10 ± 16.20^{d}	31.7 ± 4.10^{d}
Baicalin group	6	100	102.00 ± 8.90^{d}	37.5 ± 9.60^{d}
Paeoniflorin group	6	100	105.90 ± 29.40^{d}	48.7 ± 8.20^{d}
Matrine group	6	100	116.70 ± 14.2^{d}	46.4 ± 15.9^{d}

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase.
Note: Compared with the normal group, $^{a}P<0.05$, $^{b}P<0.01$; compared with the gefitinib group, $^{c}P<0.05$, $^{d}P<0.01$.

Effects of Different Drugs on Serum Alanine Aminotransferase and Aspartate Aminotransferase Levels in Mice

As shown in ►**Table 2**, compared with the normal group, gefitinib significantly increased serum ALT and AST levels in mice ($p < 0.01$). Compared with the gefitinib group, glutathione, glycyrrhizic acid, baicalin, paeoniflorin, and matrine all significantly decreased ALT and AST levels ($p < 0.01$); ligustrazine and silymarin only significantly decreased AST levels ($p < 0.05$, $p < 0.01$). Among them, glycyrrhizic acid had the best effect in reducing ALT and AST levels, followed by baicalin, both significantly better than glutathione, while the effect of paeoniflorin is roughly equivalent to that of glutathione.

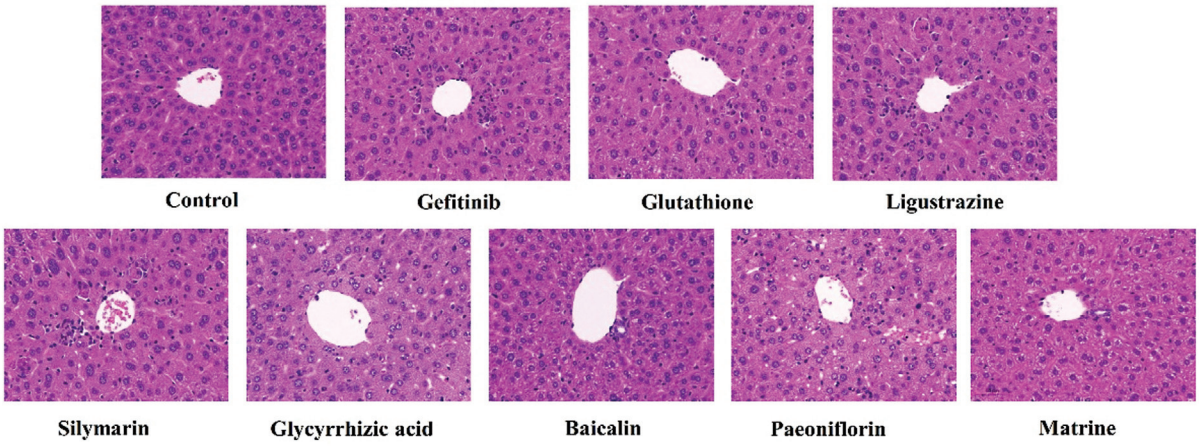

Fig. 1 Effects of different drugs on hepatic pathological changes in mice (bar = 50 μm, × 200)

Effects of Different Drugs on Hepatic Pathological Changes in Mice

As shown in ►**Fig. 1**, the liver cell cords of mice in the normal group were arranged radially, evenly, and orderly, and the liver cells showed no edema, degeneration, or necrosis; mice in the gefitinib group showed a large number of inflammatory cell infiltrations in the liver tissue, and the arrangement of liver cords was disorganized; mice in the glutathione group, glycyrrhizic acid group, baicalin group, paeoniflorin group, and matrine group showed a significant reduction in inflammatory cell infiltration in the liver tissue.

Toxic Effects of Different Drugs on AML12 Cells

As shown in ►**Table 3**, within the concentration range of 0 to 640 μmoL·L^{-1}, glutathione, ligustrazine, silymarin, glycyrrhizic acid, paeoniflorin, and matrine did not affect the viability of AML12 cells. When the concentration of baicalin

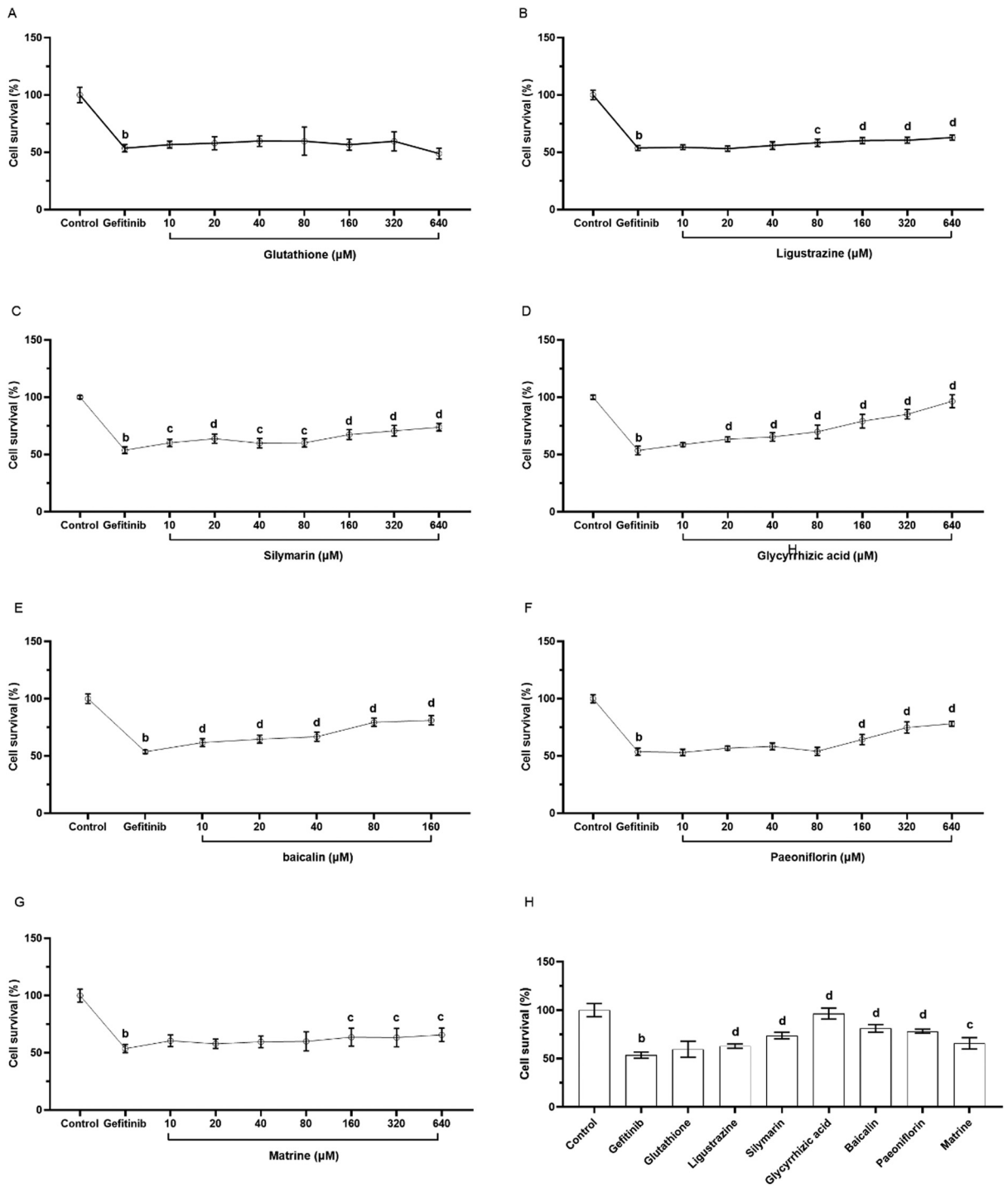

Fig. 2 Protective effects of different drugs on gefitinib-induced damage to AML12 cells. Notes: Compared with the normal group, $^{b}p < 0.01$; compared with the gefitinib group, $^{c}p < 0.05$, $^{d}p < 0.01$.

was ≥320 μmol·L^{-1}, it significantly reduced cell viability ($p < 0.01$), exhibiting cytotoxic effects. The highest concentration of baicalin selected for subsequent experiments was 160 μmol·L^{-1}.

Protective Effects of Different Drugs on Gefitinib-Induced Damage to AML12 Cells

As shown in ►**Fig. 2**, ligustrazine, silymarin, glycyrrhizic acid, baicalin, paeoniflorin, and matrine all could alleviate gefitinib-induced damage to AML12 cells to varying degrees (►**Fig. 2A–G**) except for glutathione. Among them, glycyrrhizic acid showed the best protective effect, increasing cell survival rate to 96.4% at 640 μmol·L^{-1}, significantly higher than other drugs (►**Fig. 2H**); baicalin and paeoniflorin were next effective, increasing cell survival rate to 81.1 and 78.2%, respectively; glutathione showed no protective effect on gefitinib-induced damage to AML12 cells in vitro (►**Fig. 2A**).

Effects of Different Drugs on the Levels of Alanine Aminotransferase, Aspartate Aminotransferase, and Lactate Dehydrogenase in the Supernatant of AML12 Cells

As shown in ►**Table 4**, compared with the normal group, gefitinib significantly increased the levels of ALT, AST, and LDH in the cell supernatant ($p < 0.01$); compared with the gefitinib group, glycyrrhizic acid significantly decreased the levels of ALT, AST, and LDH ($p < 0.05$); silymarin, baicalin, and paeoniflorin significantly decreased ALT and LDH levels ($p < 0.05$); ligustrazine and matrine only significantly decreased LDH levels ($p < 0.05$).

Discussion

Gefitinib is one of the first-line targeted drugs for the treatment of advanced NSCLC, but long-term use can lead to varying degrees of liver damage in patients. Clinical investigations have shown that among NSCLC patients treated with gefitinib, 16 to 26% experienced grade 2 to grade 3 liver damage. Once liver damage occurs, patients must discontinue the medication for hepatoprotective treatment, which significantly impacts the treatment process for cancer patients.[8] Therefore, preventing and treating gefitinib-induced hepatotoxicity is of great importance for the treatment of cancer patients. The results of this study indicate that ligustrazine, silybin, baicalin, glycyrrhizic acid, paeoniflorin, and matrine all have varying degrees of protective effects against gefitinib-induced liver damage, with glycyrrhizic acid showing the best protective effect, significantly better than glutathione.

Gancao (Glycyrrhizae Radix et Rhizoma) is the root and rhizome of *Glycyrrhiza uralensis Fisch*, with the effects of tonifying the spleen and replenishing qi, resolving phlegm and relieving cough, mitigating urgency and alleviating pain, clearing heat and removing toxin, and harmonizing various medicines, widely used in TCM prescriptions. Modern research studies have shown that glycyrrhizic acid is one of the main active components of Gancao (Glycyrrhizae Radix et Rhizoma), possessing various pharmacological activities such as antioxidant, anti-inflammatory, antiviral, hepatoprotective, and

Table 3 Influence of different drugs on the viability of AML12 cells ($\bar{X} \pm s$, %)

Groups	0 μmol·L^{-1}	10 μmol·L^{-1}	20 μmol·L^{-1}	40 μmol·L^{-1}	80 μmol·L^{-1}	160 μmol·L^{-1}	320 μmol·L^{-1}	640 μmol·L^{-1}
Normal group	100.00 ± 3.80	94.60 ± 5.10	99.90 ± 5.90	97.70 ± 4.90	99.50 ± 7.80	99.50 ± 7.80	101.60 ± 3.50	97.40 ± 8.10
Gefitinib group	100.00 ± 4.20	96.50 ± 3.40	94.70 ± 6.60	99.50 ± 5.20	96.80 ± 5.80	105.30 ± 4.50	98.90 ± 3.10	101.60 ± 2.60
Glutathione group	100.00 ± 2.80	97.70 ± 4.10	96.40 ± 10.30	96.20 ± 6.20	95.70 ± 3.70	99.90 ± 6.40	99.60 ± 7.30	97.40 ± 1.40
Ligustrazine group	100.00 ± 3.30	109.60 ± 4.90	98.90 ± 10.00	100.00 ± 9.90	96.10 ± 8.40	96.00 ± 5.00	103.50 ± 6.00	100.40 ± 3.90
Silymarin group	100.00 ± 6.50	93.50 ± 7.00	100.40 ± 6.30	99.70 ± 9.30	95.10 ± 4.20	95.50 ± 1.50	84.70 ± 1.50[b]	69.40 ± 4.00[b]
Glycyrrhizic acid group	100.00 ± 6.60	99.00 ± 2.60	100.00 ± 6.30	98.80 ± 5.20	100.90 ± 4.10	103.50 ± 6.60	105.30 ± 3.60	104.50 ± 6.80
Baicalin group	100.00 ± 5.50	100.70 ± 5.30	99.10 ± 4.60	98.50 ± 5.80	97.30 ± 4.20	108.30 ± 7.00	105.60 ± 5.10	100.90 ± 5.30

Note: Compared with 0μmol·L^{-1}, [b]$P<0.01$.

Table 4 Effects of different drugs on the levels of ALT, AST, and LDH in the supernatant of AML12 cells ($\bar{X} \pm s$, $n = 6$)

Groups	Dosage (c/μmoL·L^{-1})	ALT (U·L^{-1})	AST (U·L^{-1})	LDH (U·L^{-1})
Normal group	–	1.35 ± 0.44	1.72 ± 0.35	14.3 ± 3.6
Gefitinib group	320	4.27 ± 0.38^{b}	3.31 ± 0.38^{b}	44.5 ± 8.2^{b}
Glutathione group	640	4.18 ± 0.47	3.55 ± 0.47	45.7 ± 6.5
Ligustrazine group	640	3.75 ± 0.36	3.28 ± 0.44	33.6 ± 4.7^{c}
Silymarin group	640	2.72 ± 0.25^{c}	2.94 ± 0.36	31.5 ± 5.6^{c}
Glycyrrhizic acid group	640	2.21 ± 0.46^{d}	2.56 ± 0.34^{c}	25.6 ± 6.4^{d}
Baicalin group	160	2.60 ± 0.39^{c}	2.74 ± 0.46	30.3 ± 5.9^{c}
Groups	640	3.15 ± 0.54^{c}	2.98 ± 0.48	33.8 ± 8.1^{c}
Normal group	640	3.52 ± 0.44	3.11 ± 0.35	36.4 ± 7.5^{d}

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; LDH, lactate dehydrogenase.
Note: Compared with the normal group, $^{a}P<0.05$, $^{b}P<0.01$; compared with the gefitinib group, $^{c}P<0.05$, $^{d}P<0.01$.

antitumor effects.[9] Glycyrrhizic acid can significantly alleviate liver damage induced by carbon tetrachloride,[10] alcohol,[6] phaseolus vulgaris lectin,[11] and oxaliplatin.[12] The results of this study indicate that glycyrrhizic acid has the best protective effect against gefitinib-induced liver damage, significantly increasing the body weight of mice, reducing the liver index, lowering serum ALT and AST levels, and alleviating liver pathological damage, and baicalin follows, both of which are superior to glutathione, while the protective effect of paeoniflorin is roughly equivalent to that of glutathione.

AML12 is a kind of mouse normal liver cells commonly used in vitro evaluation studies of hepatotoxicity. Previous studies have found that gefitinib induces AML12 cell damage in a time- and concentration-dependent manner.[13] In this study, treatment of AML12 cells with 20 μmol·L^{-1} gefitinib for 24 hours reduced cell viability to 53%. Ligustrazine, silybin, glycyrrhizic acid, baicalin, paeoniflorin, and matrine all dose-dependently reduced cell damage and increased cell viability. Comparatively, glycyrrhizic acid restored cell viability to 96.4% at 640 μmol·L^{-1}, and significantly reduced the levels of ALT, AST, and LDH in the supernatant, with effects superior to other drugs. Baicalin and paeoniflorin followed, restoring cell viability to 81.1 and 78.2%, respectively, and significantly reducing ALT and LDH levels in the supernatant. Although glutathione has a good protective effect in vivo, it has no protective effect in vitro, which may be related to the redox cycle of glutathione in vivo.[14]

Conclusion

In summary, glycyrrhizic acid, baicalin, and paeoniflorin all have good protective effects against gefitinib-induced hepatotoxicity, with glycyrrhizic acid being the most effective, providing experimental evidence for the clinical application of related hepatoprotective drug preparation.

CRediT Authorship Contribution Statement

Xiaoting Yin: Investigation, methodology, validation, visualization, and writing—original draft. Min Li: Conceptualization, data curation, funding acquisition, supervision, and writing—review and editing. Yucheng Li: Conceptualization, funding acquisition, resources, supervision, and writing—review and editing.

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Conflict of Interest

The authors declare no conflict of interest.

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