We demonstrated that ER stress may play a role in resveratrol induced inhibition of gastric cancer cell survival. And resveratrol may inhibit cell migration by affecting the expression of EMT related genes. Resveratrol- treated A549 cells showed decreased viability and increased H19 expression, while BGC823 cells also exhibited a high level of H19 expression and similar reduced viability. Therefore, we chose lncRNA H19 for further investigation of function in resveratrol-treated gastric cancer cells. In addition, phosphorylation of AKT was decreased in cells exposed to the combined treatment. However, there was virtually no increase in β- catenin nuclear translocate induced by 50 μM resveratrol alone or in combination with H19 knockdown, when compared to the control. Overall, the role played by the Wnt/β- catenin pathway in SGC7901 gastric cancer cells should be further determined.

There are still large numbers of natural compounds that are far from being applied in a clinical setting. Demonstration of an ability to regulate genetic and epigenetic factors would strengthen the case for using such compounds in cancer therapy. Results from the present study showed that H19 was over- expressed at low concentration of resveratrol and downregulated at high concentrations. Silencing of H19 has been reported to prevent cell proliferation and migration in lung cancer by reducing methylation of E- cadherin promoter. We therefore wanted to examine the impact of downregulated H19 expression on the anti- tumour effects of resveratrol. In fact, we have previously shown that knockdown of H19 contributes to the increased sensitivity of cancer cells to pterostilbene (a dimethyl ether analog of resveratrol), reducing cell proliferation and invasiveness.

УДК 2

调控 53BP1-P53 复合物提升紫檀芪抑制肺癌侵袭转移研究

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Summary. *Pterostilbene is a natural resveratrol dimethylated analogue that has anticancer effects against a variety of cancers. It has better and oral absorption than resveratrol. In this paper, the molecular mechanism of p53-53BP1 complex regulating pterostilbene on the proliferation of lung cancer cells was elaborated from the aspects of the regulatory mechanism of pterostilbene on lung cancer cells.*

Human beings have made certain progress in the fight against cancer, but the cancer prevention and treatment mechanism is still in the stage of gradually being cracked and improved. Pterostilbene is a natural resveratrol dimethylated analogue that has pleiopotent anticancer effects on a variety of cancer types, and has better lipophilicity and oral absorption than resveratrol, with higher cellular absorption, and longer half-life. The initiation of programmed cell death (PCD) is a key regulatory link for a variety of anticancer drugs, which involve two main types, apoptosis and autophagy. p53 accumulation is caused by phosphorylation (p-)AKT-mediated reduction in phosphorylation of specific MDM2. Once MDM2 is phosphorylated by p-Akt, p-MDM2 translocates to the nucleus and promotes ubiquitination of the target protein. MDM2 regulates the physiological level and function of p53 in normal cells through a continuous process of monoubiquitination.

The 53BP1 gene, located on chromosomes 15q15-q21, produces tumor suppressor p53-binding protein 1 (53BP1 protein), which interacts with the DNA-binding core domain of tumor suppressor p53 and enhances p53-mediated transcriptional activation. 53BP1 promotes non-homologous end-joining (NHEJ)-mediated DSB repair and prevents HR by counteracting BRCA1's function in homologous recombination (HR). This project intends to mine relevant databases through bioinformatics technology to analyze the target information of pterostilbene affecting the biological functions of lung cancer cell A549 cell line. A549 cells were treated with different concentrations of pterostilbene at different times, and cell proliferation was measured by CCK8 method. After that, soft agar experiment was used to further determine the proliferation of pterostilbene treated cells to explore the regulatory effect of 53BP1-p53 complex on pterostilbene and its role in

cancer suppression, aiming to propose a new idea for lung cancer treatment with better treatment effect and higher absorption rate.

After treating A549 cells with different concentrations of pterostilbene (0, 5, 25, 50, 100 μM) for 24 h, 48 h, and 72 h, the change in cell number was determined by CCK-8 method. After 24 h and 48 h of pterostilbene action cells, 50 and 100 μM pterostilbene significantly reduced the viability of A549 cells. After 72 h, 25, 50 and 100 μM pterostilbene treatment was able to significantly reduce cell viability.

The effect of 50 μM pterostilbene on apoptosis after 24 h, 48 h, and 72 h treatment of cells using annexin V-FITC/PI double staining method was determined. From which it can be seen that apoptosis of A549 cells can be significantly promoted after treatment with 50 μM pterostilbene.

A549 cells were treated with pterostilbene concentrations of 0, 5, 25 and 50 μM for 24 h, and then the cell cycle distribution was detected by PI method. After a certain concentration of pterostilbene treatment, lung cancer cells can be blocked in the S phase to inhibit cell proliferation.

A549 cells were treated with different concentrations of pterostilbene (0, 5, 25, 50, 75 μM) for 24 h, and the expression of the apoptotic gene was further determined using Western blot. It is revealed that compared with the control group, after treating lung cancer cells with pterostilbene, the pro-apoptotic protein in the cells can be upregulated.

This project was performed by Transwell to test A549 cells treated with pterostilbene. A549 cells were treated with 0, 5, 25, and 50 μM pterostilbene for 24 h, followed by Transwell to detect cell migration after treatment. After treatment of A549 cells with pterostilbene, cell mobility decreased significantly with concentration gradient. This result showed that pterostilbene can reduce the migration capacity of A549 cells.

1. Treatment of A549 cells with different concentrations of pterostilbene can significantly reduce cell viability; Some concentrations can significantly promote apoptosis of A549 cells; It can block lung cancer cells in the S phase and inhibit cell proliferation; It can also upregulate pro-apoptotic proteins in cells; pterostilbene can reduce the migration capacity of A549 cells; Pterostilbene treatment has a certain inhibitory effect on the increase of apoptotic protein levels in EMT cells, and has a certain inhibitory effect on cancer cell invasion and metastasis. From this, it has been proved that pterostilbene has demonstrated the inhibitory effect of pterostilbene on tumor progression.

2. After knocking down p53, the expression of p21 protein in A549 cells was inhibited, but the inhibitory effect caused by the decrease of p53 could be reversed to a certain extent after the addition of pterostilbene treatment; After the addition of pterostilbene, MDM2 expression levels decreased. After overexpression of p53, the expression of p21 protein did not change significantly, and the expression of MDM2 protein was significantly increased, while the expression of p21 and MDM2 proteins in the pterostilbene treatment group was significantly increased compared with that in the control group. This shows that p53 compound has a promoting effect on the tumor inhibitory effect of pterostilbene.

Very little is currently known about our 53BP1-p53 complex, so in future experiments, we will work to elucidate the regulatory effect of pterostilbene on the 53BP1-p53 complex, and the expected trials are as follows:

1. Cells were treated with pterostilbene and immunostained for proteins such as p53, 53BP1 and MDM2 to determine the colocalization of these genes.

2. Predict the target protein site of pterostilbene and perform 3D structural analysis on the target protein.

3. The interaction between p53 and BP531 after pterostilbene treatment was predicted by co-immunoprecipitation technology.

4. The pull-down experiment was used to further verify the binding of pterostilbene to the target protein.