湖南中醫藥大學

HUNAN UNIVERSITY OF CHINESE MEDICINE

博士学位论文

Specialized subject	:	Integrative Chinese and Western Medicine
Research field	:	Integrative Chinese and Western Medicine in Cancer
Ph.D. student	:	Fenny Yunita
Principal supervisor	:	Professor Xuefei Tian
Degree type	:	Scientific degree

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Hunan University of Chinese Medicine

Ph.D. Dissertation

The evaluation of ginsenoside and curcumin inhibitory effect on PD-1 / PD-L1 pathway in nude mice model and its immune mechanism

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Name : Fenny Yunita

Student number : 2014321023

Major : Integrative Chinese and Western Medicine

No.	Name	Signature	Date
1	Professor Xuefei Tian (Promotor)		5/31/2017
2	Professor Lizhong Huang (Chairman)		5/26/2017
3	Professor Tang Tao (Member)		5/26/2017
4	Professor Cai Xiong (Member)		5/26/2017
5	Professor Wang Zhe (Member)		5/26/2017
6	Professor Jiekun Luo (Member)		5/26/2017
7	Doctor Sha Tian (Secretary)		5/26/2017

Abstract

Objective : This research is aim to investigate the synergistic effect of ginsenoside and curcumin as PD-1/PD-L1 pathway blocker and NF-κB, MMP-9 inhibitor as anticancer in hepatocellular carcinoma and therefore could boost the immune system, suppress the inflammation, and prevent chemoresistancy and metastasis in animal HCC model.

Methods : 46 HepG2 cells xenograft nude mice were randomly divided into a model group of 7 nude mice, a 104mg/kg bodyweight low dose ginsenoside group of 7 nude mice, a 520mg/kg high dose ginsenoside group of 6 nude mice, a 200mg/kg curcumin group of 7 nude mice, a curcumin+low dose ginsenoside group of 6 nude mice, a curcumin+high dose ginsenoside group of 7 nude mice, and a 24mg/kg 5FU + 4mg/kg Cisplatin chemotherapy group of 6 nude mice, raised in SPF environment. HepG2 cells were transplanted into nude mice subcutaneous. Curcumin and ginsenoside were given orally once daily for 18 consecutive days, while 5FU and Cisplatin were injected intraperitoneally once a week. Tumor volume and weight were measured. PD-1, PD-L1, NF-κB, and MMP-9 expression were detected by western blotting. Data would be analyzed to proof the effectivity of these compounds.

Results : The tumor volume significantly inhibited by curcumin , ginsenoside, and combination of both, compared with the model group ($P<0.05$). There are no significant difference of tumor growth between combination of curcumin and ginsenoside with chemotherapy group ($P>0.05$). There are significant anti PD-1 effect of ginsenoside compared with other groups ($P<0.05$) without dose dependent manner. There are significant anti PD-L1 effect in low dose ginsenoside , curcumin, also in curcumin and high dose ginsenoside group ($P<0.05$). There are significant NF-κB inhibitory effect in the group treated with combination of curcumin and ginsenoside without dose dependent manner ($P<0.05$). There are significant MMP-9 inhibitory effect in high dose ginsenoside and curcumin group, and the inhibition increased in the combination of curcumin and ginsenoside group without dose dependent manner ($P<0.05$).

Conclusion : Ginsenoside and curcumin significantly inhibit tumor growth in HCC *in vivo*. Ginsenoside significantly downregulated the PD-1 expression in HCC. Combination of

ginsenoside and curcumin have significant anti PD-L1, also MMP-9 and NF-κB inhibitory effect. The effect ginsenoside and curcumin *in vivo* may be related to their mechanism to increase immune system, anti-inflammatory and prevent invasion and chemoresistancy

Keywords : Curcumin, Ginsenoside, PD-1, PD-L1, NF-κB , MMP-9