Name of Journal: World Journal of Gastroenterology

ESPS Manuscript NO: 32489

Manuscript Type: ORIGINAL ARTICLE

Title: Tumor-associated autoantibodies are useful biomarkers in immunodiagnosis of α-fetoprotein-negative hepatocellular carcinoma

Authors: Ting Wang, Mei Liu, Su-Jun Zheng, Dan-Dan Bian, Jin-Yan Zhang, Jia Yao, Qing-Fen Zheng, A-Meng Shi, Wen-Han Li, Lu Li, Yu Chen, Jin-Hai Wang, Zhong-Ping Duan, Lei Dong

Correspondence to: Lei Dong, Professor, Department of Gastroenterology, Second Affiliated Hospital of Xi'an Jiaotong University, Xi'an 710004, Shaanxi Province, China; donglei6655@126.com; Telephone: +86-29-87679272; Fax: +86-29-87678758.

1) What did this study explore?

   To determine the prevalence and diagnostic value of autoantibodies in α-fetoprotein (AFP) negative hepatocellular carcinoma (HCC).

2) How did the authors perform all experiments?

   Fifty-six serum samples from AFP-negative HCC, 86 from AFP-positive HCC, 168 from chronic liver diseases, and 59 from normal human controls were included in this study.

   Autoantibodies to nucleophosmin (NPM)1, 14-3-3zeta and mouse double minute 2 homolog (MDM2) proteins in AFP-negative HCC serum were evaluated by ELISA. Partially positive sera were further evaluated by Western blotting.

   Immunohistochemistry was used to detect the expression of three tumor-associated antigens (TAAs) in AFP-negative HCC and normal control

tissues.

3) How did the authors process all experimental data?

A $\chi 2$ test with Yates' correction was used to determine whether the frequency of autoantibodies to three TAAs in each cohort of patient sera was significantly higher than that in sera from normal individuals. Two significant levels (0.05 and 0.01) were used.

Methods for calculating the sensitivity, specificity and accuracy were based on the methodology provided in Epidemiology (6th edition, edited by Dr. Ray M. Merrill, Jones & Bartlett Learning Company, Burlington, 2012).

4) How did the authors deal with the pre-study hypothesis?

Many autoantibodies to tumor-associated antigens (TAAs) have been reported in hepatocellular carcinoma (HCC), which are suggested to be useful tools for immunodiagnosis of HCC. However, no previous study has specifically evaluated the diagnostic value of TAA autoantibodies in α-fetoprotein (AFP)-negative HCC. Thus, we specifically evaluated the diagnostic value of TAA autoantibodies in AFP-negative HCC.

5) What are the novel findings of this study?

This study demonstrated that autoantibodies to nucleophosmin 1, 14-3-3zeta and mouse double minute 2 homolog may be useful biomarkers for immunodiagnosis of AFP-negative HCC.

Lei Dong

2017.2.20