

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: GNAS1 T393TC manuscript.doc).

Title:Single nucleotide polymorphisms of *GNAS1 T393C TT* predicts better outcome of advanced non-small cell lung cancer patients. A case report and review of the literature

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Name of Journal: *World Journal of Gastrointestinal Oncology*

ESPS Manuscript NO: 9917

The manuscript has been improved according to the suggestions of reviewers:

1. Format has been updated and we have polished the language as requested by reviewers.
2. Revision has been made according to the suggestions of the reviewer. The following are the detailed information:

Reviewer 1

a. There are many typos in the manuscript. For instance; nueleotide (in the title), genotype (in the abstract), feasibl (in the introduction), sstubstitution(in the discussion)..... Should be reconsidered.

We have carefully checked all the typo errors and made corresponding changes in the manuscript.

Page 1: we changed “nueleotide” to “nucleotide” in the title.

Page 2: “genotype ” was changed to “genotype” in the abstract.

Page 3: “feasibl” was changed to “feasible” in the introduction.

b. There is no information regarding the aim of study in the summary section.

We appreciate the reviewer’s point. We have added the aim of the study in the abstract section.

Page 1: Added “The aim of this study was to evaluate the potential prognostic value of GNAS1 T393C polymorphism in advanced non-small cell lung cancer (NSCLC).”

c. Similar results about TT genotype had been given by Xie et al (see below). The authors must add the results of this article. Xie FJ, Zhao P, Kou JY, Hong W, Fu L, Hu L, Hong D, Su D, Gao Y, Zhang YP. The T393C polymorphism of GNAS1 as a predictor for chemotherapy sensitivity and survival in advanced non-small-cell lung cancer patients treated with gemcitabine plus platinum. Cancer Chemother Pharmacol. 2012. Jun; 69(6):1443-8.

We have cited the reference (Reference 20) in the manuscript and compared the results with our data in the discussion section(in page 10).

d. What is the meaning of the abbreviation of “PS” in the results section? This meaning should be added.

We feel sorry for this mistake. “PS” stands for “performance status”. We have added the full name of “PS” in the manuscript.

Page 6: Added “performance status (PS)”.

e. What is the meaning of “vitro” in the discussion section? I think it can be “in vitro”

We changed “vitro” to “in vitro”.

Page 8: Changed “vitro” to “in vitro”.

Page 9: Changed “vitro” to “in vitro”.

f. The results of other studies related to lung cancer should also be given in Table 2.

We have included other studies related to lung cancer in the Table 3 (Page 15).

Reviewer 2

a. There is some clinical value in this study. Whereas this content of MS was about NSCLC, and not suitable for the world Journal of Gastrointestinal Oncology. Abstract: Purpose: the author was not explained the aim of the MS.

We have added the aim of the study in the abstract section.

Page 1: Added “The aim of this study was to evaluate the potential prognostic value of GNAS1 T393C polymorphism in advanced non-small cell lung cancer (NSCLC).”

b. Introduction: 1. some references need to verify. For example, “The 5-year survival rate is still only at 10%” is not correct.

We appreciated the reviewer’s comments. We have changed “The 5-year survival rate is still only at 10%” to “only 16.6% of all lung cancer patients are alive 5 years or more after diagnosis” according to the NCCN guidelines Version3.2014.

Page 3: "The 5-year survival rate is still only at 10%" to "only 16.6% of all lung cancer patients are alive 5 years or more after diagnosis".

c. Please explain the association between GNAS1 gene and lung cancer, and the reason of select GNAS1 as prognosis factor?

The association between GNAS1 gene and lung cancer is that the GNAS1 T393C polymorphism correlates with lung cancer. To make this clear, we made changes in the abstract: "Recent studies have shown that the GNAS1 T393C polymorphism correlates with some cancers, such as lung cancer, colorectal cancer etc." Also, the reason we chose GNAS1 gene as prognosis factor is the correlation between GNAS1 T393C polymorphism and cancer clinical outcome. To make the rationale clear, we added the sentence "GNAS1 gene encodes the Gαs subunit of heterotrimeric G proteins and recent studies have shown that the GNAS1 T393C polymorphism correlates with lung cancer".

Page 2: Changed "Recent studies have shown that the GNAS1 T393C polymorphism correlates with some cancers, such as bladder cancer, colorectal cancer etc." to "Recent studies have shown that the GNAS1 T393C polymorphism correlates with some cancers, such as lung cancer, colorectal cancer etc.".

Page 8: Added "GNAS1 gene encodes the Gαs subunit of heterotrimeric G proteins and recent studies have shown that the GNAS1 T393C polymorphism correlates with lung cancer".

d. Patients and methods 1. What were the exclusion criteria? There are only 94 patients with NSCLC in the hospital in China, which was not credible.

That is a good point. We have added the inclusion criteria in the methods parts. During our study period, there were only 94 patients available. Although there were only 94 patients, our data is still convincing given all the data were analyzed by statistical tools.

Page 4: Added "Patients were enrolled in this study according following criteria: (1) histologically or cytologically confirmed NSCLC; (2) stage IIIB or IV disease; (3) Eastern Cooperative Oncology Group PS of 2 or less; and (4) life expectancy of more than 3 months. Patients were excluded if they had received chemotherapy or radiotherapy previously."

e. No details were given on using serum or plasma samples Statistical analysis. What was the Statistical methods for Hardy-Weinberg equilibrium? Result Author need to make a concrete analysis of association between GNAS1 SNP and clinical TNM stages, male and female, synergistic effect of GNAS1 SNP and smoke state.

We have added the relevant information for statistical analysis in the manuscript. We have also added a table (Table 2) showing the association between GNAS1 SNP and clinical TNM stages, male and female, synergistic effect of GNAS1 SNP and smoke state.

Page 4: DNA was extracted from whole blood samples using a commercially available kit.

Page 5: Compatibility with the Hardy-Weinberg equilibrium was calculated with the public domain program HWE (<http://linkage.rockefeller.edu/ott/linkutil.htm>).

f. Discussion 1. The rationale for the present study is not well discussed and need to improve.

That is a good point. We have added the rationale of this study in the first paragraph of the discussion section.

Page 7: Added "Lung cancer is still the leading cause of cancer death worldwide. Although clinicopathological parameters such as UICC stages may serve as prognostic markers in lung cancer, markers facilitating a more precise prediction of the clinical outcome of individual patients are still desirable. The majority of prognostic markers are based on features of the tumor tissue itself. GNAS1 gene encodes the Gas subunit of heterotrimeric G proteins and recent studies have shown that the GNAS1 T393C polymorphism correlates with lung cancer. Hence, we investigated whether GNAS1 T393C polymorphism may be predictive for clinical outcome in patients with NSCLC."

Reviewer 3

Major points:

a. The major problem with the manuscript is the relatively small number of patients utilized in the study to back up the conclusions made. In addition, the association between the GNAS1 T393C polymorphism needs to be better discussed.

We appreciate the reviewer's point here. During our study period, there were only 94 patients available according to our selection criteria. To acknowledge the small number of patients utilized in this study, we clearly stated the following sentence in the discussion section: "it must be emphasized that in this study a limited number of patients were investigated. Although our findings support the concept of a role of genetic host factors in tumor progression, consistent with the result published previously [20], further independent studies of large cohorts are necessary to confirm their validity."

b. The authors need to elaborate the discussion comprehensively to discuss their findings and to distinguish between different cancer survival rate and TT genotype.

We have added relevant information in the discussion section.

Page 8: Added" For many tumors, TT genotype was significantly correlated with better OS compared with CT or CC genotypes. For example, five-year survival rates were 76% for TT, 49% for TC, and 43.5% for CC in patients with advanced squamous cell carcinoma of the larynx[10]. And in patients with sporadic colorectal cancer, the five-year survival rate was also significantly higher in TT genotypes (87.8%) compared with TC (71.0%) and CC genotypes (50.0%) [15]. On the other hand, in intrahepatic cholangio-carcinoma [9], esophageal cancer[12] and breast cancer [16], CC genotype patients had a more favorable clinical course (Table 2). "

c. Finally, the quality of writing is extremely poor. The manuscript contains numerous spelling and grammatically errors throughout.

We have carefully checked all the typo errors and made corresponding changes in the manuscript.

Minor points:

a. Please insure all abbreviations are expressed in full the first time they appear in the manuscript
We have made corresponding changes.

b. Change vitro to in vitro

We have made corresponding changes.

c. Please stay consistent i.e. Gas vs G alpha s

We have made corresponding changes.

d. Check spacing between words

We have made corresponding changes.

Reviewer 4

We also made changes as suggested by reviewer 4 in the main text.

3. References and typesetting were corrected.

Thank you again for publishing our manuscript in the *World Journal of Gastrointestinal Oncology*. If you have any questions, please do not hesitate to contact with me.

Sincerely yours,

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