

December 31, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 7280-review.doc).

**Title:** ISCHEMIC HEART DISEASE, FACTOR PREDISPOSING TO BARRETT'S ADENOCARCINOMA: A CASE CONTROL STUDY.

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**Name of Journal:** *World Journal of Pharmacology and Therapeutics*

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

1 Format has been updated

2. Dr Isaacs revised English language

3. Revision has been made according to the suggestions of the reviewer

1) A) The phrase has change to 'IHD was more prevalent in **AdE than BE** patients.' B) The phrase has changed to 'Logistic regression analysis in the whole study population has shown that age (per decade), duration of reflux (in decades) and prevalence of IHD (Odds ratio: 1.987, 95% Confidence intervals: 1.241-3.182, p=0.004) was significantly more frequent in AdE patients, **while use of statins was less frequent** (table 5).' C) the phrase has been removed. D) the phrase has changed to 'Nevertheless, we found that nitrates had a beneficial role in BE patients with IHD.' E) we have entered the phrase '**Beneficial role of nitrates/ sphincter relaxing medication in BE patients with IHD could be incidental, mirroring not a truly protective relationship but being the result of the small number of patients studied.**' F) We have entered the phrases '**Old age is more prevalent in AdE patients [28] and IHD is a disease of old age [29]. Thus it is possible that higher IHD incidence in AdE patients is solely a result of old age. Nevertheless, IHD was an independent risk factor for AdE in multiple regression analysis and pathogenetic mechanisms support a deleterious effect of IHD in BE patients.**' and 'More studies are needed to **show if IHD is more frequent in BE patients because they are older or verify that IHD is deleterious for BE patients and unveil the pathogenetic mechanisms (increase of angiogenetic and growth factors) beneath it.**'

2) We changed abstract conclusions to present tense and introduced several recent bibliographies concerning the role of angiogenetic factors and mucosal ischemia on Barrett esophagus evolution towards esophageal adenocarcinoma. Thus the second paragraph of introduction has changed to 'One of the main macroscopic features of BE is the net-like blood vessel structure lining the esophageal mucosa. Barrett's epithelium derives its increased blood supply from the preexisting vasculature in submucosal lamina propria and these new blood vessels occupy the mucosa layer, giving it its ominous salmon-pink color. Neovascularization emphasizes why BE is a precancerous lesion and how it can give rise to the more cancerous dysplasia and AdE [4]. Although, oxygen saturation of the microvascular blood remains high (approximately 90%) throughout the metaplasia-dysplasia-AdE sequence in BE patients[5], **microvasculature density raises stepwise as BE evolves towards AdE[6]. Esophageal inflammation enriches stromal angiogenesis [7] and the erosive environment of acid-reflux causes periodic hypoxic events [8]. Several markers of hypoxia, including oxygen-regulated transcription factor subunit hypoxia inducible factor-1alpha and vascular endothelial growth factor, have been related with**

advanced BE [9,10], while neovascularization markers, such as endoglin (CD-105), have been reported to be up-regulated in patients with high-grade dysplasia and AdE [11]. ' while the 5<sup>th</sup> paragraph of discussion has changed to 'One of the first adaptations of human myocardium to a deprivation of blood in patients with IHD, is over-expression of various angiogenetic factors, such as hypoxia-inducible factor or vascular endothelial growth factor [30]. Increased angiogenesis in pre-malignant lesions, such as BE, may serve as a surrogate marker for tumor development [4]. Moreover, hypoxia is a common state in cancers [31]. Finally, epidermal growth factor up-regulation in IHD [32], increases BE malignant potential [33], while oxidative phosphorylation upregulation [34] and subsequent reactive oxygen species overproduction, increases the mutagenic pressure and raises genetic instability [35]. Thus, we expected and we found that IHD is more frequently acquired in AdE than BE patients, especially those suffered an MI.' We also introduced a bibliography (Solaymani-Dodaran M, Card TR, West J. Cause-specific mortality of people with Barrett's esophagus compared with the general population: a population-based cohort study. *Gastroenterology*. 2013;144:1375-83, 1383.e1. doi: 10.1053/j.gastro.2013.02.050.PMID: 23583429) further emphasizing that the main cause of mortality in patients with Barrett esophagus is ischemic heart disease.

3) We introduced the following sentence in the conclusions to emphasize study drawbacks: The study drawbacks are that it is retrospective and that the number of the studied patients is still too exiguous to come to strong conclusions. We cannot overcome all the drawbacks. We increased the number of patients by entering patients from a study ran in Trikala General Hospital. Nevertheless the study is still retrospective and the number of patients is low. Thus we called in the last sentence for a study as requested : 'Our results are encouraging to design larger multicentric prospective studies in this high-risk group.'

(3) There were no more comments apart from English revision

4. References and typesetting were corrected

Thank you again for publishing our manuscript in the *World Journal of Pharmacology and Therapeutics*.



Sincerely yours,

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