Format for ANSWERING REVIEWERS

November 7, 2014

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 13166-review.doc).

Title: Epigenetics and DNA methylation in cancer

Author: Laura Lattanzio and Cristiana Lo Nigro

Name of Journal: *World Journal of Translational Medicine*

ESPS Manuscript NO: 13166

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

1. Format has been updated

2. Revision has been made according to the suggestions of the reviewers:

Reviewer 1 (00031349) **commented to authors: "The review article entitled "Epigenetics and Methylation in Cancer" is not only well written, but also provides insights into tumor development. It is well-known that angiogenesis plays a central role in the progression of cancer. Therefore, it is important to include the role of DNA methylation in the initiation and progression of angiogenesis in cancer in this review".**

We agree with reviewer 1 on the importance of angiogenesis in cancer progression. For this reason, we followed reviewer' suggestion and we amplified the paragraph "Methylation landscape of tumour metastasis" reporting several examples strictly related to angiogenesis with the corresponding references. The new paragraph appears:

"**DNA methylation in angiogenesis and metastasis.**

During tumorigenesis, cells acquire metastatic potential following angiogenesis, induction of cell surface metalloproteases, decrease in the expression of cell–cell adhesion molecules, and increased expression of cell surface receptors that aid in motility. E-cadherin and alpha-4 integrins, two of the most common cell adhesion receptors, are silenced by methylation in several cancers[12]. Similarly, intracellular basement membrane proteins (i.e., NID1 and NID2) are also silenced by methylation in cancer. Therefore, it is evident that epigenetics could also play a critical role in the metastatic process[13]. The phenomenon of metastasis is a complex process involving several distinct steps: tumor cells, supported by angiogenesis, infiltrate the basement membrane.

Aberrant methylation of metastasis initiation genes could be responsible of tumor invasiveness (for a detailed review, refer to Cock-Rada and Weitzman, 2013[8]).

Several genes have been identified which regulate the metastatic process, can predict prognosis and metastasis in cancer patients, and are routinely used in clinical practice[8]. These genes are usually involved in regulation of ECM and angiogenesis, regulation of cell adhesion and invasion, and repressive and activating histone modifications.

In particular, in the early stages of cancer progression, tumour cells induce degradation of the ECM by matrix metalloproteinases (MMPs) for angiogenesis. Downregulation of the genes encoding tissue

inhibitor of metalloproteinases contributes to this process by loss ofMMP regulation and release of angiogenic factors such as fibroblast growth factor-2 (FGF–2) and vascular endothelial growth factor (VEGF)[14]. TIMP-2 is suppressed in some solid tumours and lymphoid malignancies by promoter CpGI hypermethylation[15, 16]. TIMP-3 was also found to be suppressed by DNA methylation during initial, local progression of gastric and oesophageal cancers, correlating with poor patient survival[17].
Cells then migrate and traverse the extracellular matrix (ECM), invading nearby structures and intravasating into lymphatic or blood vessels. Cells that survive hostile circulatory environment disseminate to distant sites and form micrometastases. These cells then proliferate and colonise the new organ, becoming macrometastases (Figure 6)".

Reviewer 2 (00061154) **commented to authors: "1. More primary references are needed. 2. I have specific comments in what I have uploaded. 3. The manuscript is long and many "the" can be deleted without loss of meaning. I have done so in many places. 4. A few sentences are confusing. 5. Have you obtained permission to publish all the tables and figures? 6. I have taken the liberty of editing in a number of places, using more standard English".**
We thanks reviewer 2 for his precious comments and we have corrected the manuscript according to his suggestions. In particular:

1. Many references have been added to the text and in the References section (there are now 45 references),
2. We agree with all comments and we have accepted them in the revised manuscript,
3. We agree and we have deleted, under suggestion, many "the" that were in surplus,
4. We tried to reformulate the sentences that reviewer 2 considered confused (they are highlighted in the revised manuscript),
5. All tables and figures have been adapted by others published work and it is specify in all tables and figures legend.
6. We thanks reviewer 2 for its help in correcting our English and we have accepted all the changes.

Reviewer 3 (00541708) **commented to authors: "The authors provide an excellent work clearly reviewing state of the art in the field of epigenetic of cancer".**
We thanks reviewer 3 for his comment and we really appreciate his positive judgement.

3. References and typesetting were corrected and are highlighted in yellow in the revised manuscript.

Thank you again for publishing our manuscript in the *World Journal of Translational Medicine*.

Sincerely yours,

Cristiana Lo Nigro, PhD
Medical Oncology
S. Croce University Hospital
Via Carle 25, 12100 Cuneo, Italy
Phone: +39 0171 616338, Fax: +39 0171 616331
e-mail: lonigro.c@ospedale.cuneo.it