

July 1, 2015

Dear Editor,

Please find enclosed the edited manuscript in word format (file name: 19108-review.doc).

Title: New Insights into Tumor Dormancy: Targeting DNA Repair

Authors: Elizabeth B. Evans and Shiaw-Yih Lin

Name of Journal: *World Journal of Clinical Oncology*

ESPS Manuscript NO: 19108

The authors would like to thank reviewer 00033009 for the kind comments about the manuscript. In addition, the manuscript has been improved according to the suggestions of reviewer 00505473:
1 Format has been updated

2 Revision has been made according to the suggestions of the reviewer

(1) The molecular mechanism of tumor dormancy should be discussed based on Figure 1.

Figure 1 was included in the introduction to represent the overall mechanisms that lead to tumor dormancy. The authors believe that they have used this figure to discuss these molecular mechanisms; however we have referenced figure 1 throughout the manuscript to address this comment.

(2) There are several proteins in G0-G1/S phase of the cell cycle associated with the development of different tumors. The importance of these proteins along with p21, p27 should be discussed in tumor dormancy and recurrence.

The authors agree with the reviewer, and we have modified the cellular dormancy section to include the importance of the cell cycle proteins in the formation, dormancy, and recurrence of different tumors. Specifically, we have added an additional paragraph describing how aberrant regulation of the cell cycle leads to tumor development. The paragraph states: "Mis-regulation of cell cycle proteins can result in tumor formation, dormancy, and recurrence. Prostate cancer, breast cancer, and renal cell carcinoma are linked to the loss of p27 (Kip1) [40-42]. In addition, reduction in p27 (Kip1) is used as a strong prognostic marker for recurrence and poor outcomes in renal cell carcinoma patients [42]. Loss of p53, the upstream regulator of p21, was correlated with drug resistance and recurrence in colorectal cancer [43]. Overexpression of cyclin D is associated with recurrence of multiple neoplasms including breast, lymphomas, prostate, and non-small cell lung cancers [44-46]. Overexpression of cyclin D1 can occur via a multitude of

different mechanisms including genetic rearrangements, amplification of the gene locus, oncogenic signaling, and mutation in the gene that result in the inability to degrade the protein [44]. Recently, Kim et al. (2014) reported that overexpression of the cell cycle regulators CDK4, CDK6, pRB, and cyclin D1 was correlated with the recurrence of atypical meningioma [47]. Furthermore, some evidence suggested that overexpression of CDK4 may be connected to nasopharyngeal carcinoma tumor aggression and serve as a diagnostic biomarker [48]. Clearly, these results demonstrate the importance in controlling the cell cycle and how aberrant regulation may lead to tumor recurrence and poor prognosis."

(3) In case of angiogenesis and immunological tolerance regarding tumor dormancy and recurrence several genes are involved. These should be discussed in details.

The authors agree with the reviewer and we have revised a part within the angiogenic dormancy section to include, "Tumor dormancy via angiogenesis requires the interaction between the microenvironment and cell cycle regulators including p21, p27, Myc, u-PAR, ERK, and p38 [49]. Blockage of the metastasis-associated urokinase receptor (u-PAR), integrins, focal adhesion kinase (FAK) or EGF receptor (EGFR) can result in tumor suppression and induction of tumor dormancy [49]. U-PAR can also regulate tumor dormancy by favoring p38 activation over ERK activation [50]. In addition, the activation of the PI3K/c-Myc pathway controls the level of thrombospondin (TSP), a vital factor of tumor dormancy [16]. Troyanovsky et al. (2001) also discovered that the expression of angiostatin can control tumor dormancy by suppressing tumor growth, and one mediator of angiostatin, angiomin, was highly elevated in dormant cells [51]." In addition, we have modified the immunologic dormancy section to state, "Direct tumor immunosuppression can mediate the escape from dormancy by driving the overexpression of B7 homolog 1 (B7-H1) which inhibits T-cell activation and the cytotoxic T lymphocyte (CTL) response [63]. In addition, cancer cells can escape tumor dormancy by inhibiting antigen presentation and by methylating cytokine signaling 1 (SOCS1) thus leading to resistance to CTL-induced apoptosis [63]. Furthermore, loss of CD4⁺ or CD8⁺ T-cells can result in tumor cell dormancy escape [64]. Several cell types within the immune system can indirectly regulate the escape from dormancy by secreting proteins that promote angiogenesis. Interleukin 23, produced by macrophages, suppresses anti-tumor effectors responses, whereas interleukin 12 represses tumor growth [65, 66]. The glycoprotein, macrophage stimulating 1 (MS1) can bind to its receptor, macrophage stimulating 1 receptor (MST1R), thus suppressing antitumor immune response and promoting cell proliferation, survival, and chemotaxis. The loss of MST1R increases antitumor CD8⁺ T-cell responses resulting in higher levels of secreted tumor necrosis factor α (TNF α) subsequently leading to the inability of micrometastatic cancer cells to generate macrometastases [67, 68]. In addition, myeloid-derived suppressor cells (MDSCs), regulatory T-cells, and tumor-associated macrophages (TAMs) can also indirectly promote tumor cells to escape dormancy [63]. These cells can secrete mitogens and proangiogenic molecules which promote cell proliferation, angiogenesis and immunosuppression causing the cells to exit dormancy [63]. These results demonstrate the importance in controlling the immune system to prevent tumor recurrence and metastasis."

(4) The importance of DNA repair mechanism in tumor dormancy and recurrence was not reviewed well. Some of the information seems to be confusing like over-expression of BRCA1 in tumors.

The authors have significantly expanded the section on DNA repair mechanisms and tumor dormancy to include a section on single-stranded break repair as well as elaborating on the genes involved in the double-stranded break repair pathways. In addition, we have clarified the sentence referred to in comment 4 to state, "The -misregulation of genes associated with HR, RAD51, BRCA1, ERCC1, APE1, and PARP1, are also observed in various cancers and are associated with resistance to chemotherapies."

(5) Conclusion should be made based on discussion made above. The importance of nano-particle should not be discussed in conclusion. It should be discussed in specific sub-heading, if needed.

The authors have revised the conclusion and have deleted the section on nano-particles. Specifically, the conclusion discusses the role of DNA repair mechanisms in tumor dormancy.

3 References and typesetting were corrected

4 Spelling and grammatical mistakes were corrected

Thank you again for publishing our manuscript in the *World Journal of Clinical Oncology*.

Sincerely,



Shiaw-Yih Lin, Ph.D.