Comments on the manuscript : ESPS No: 25573 “Exploring the role of molecular biomarkers as a potential weapon against gastric cancer” by Matboli et al.,. General work focusing on molecular pathways of GC that can guide therapeutic management of the disease.A review on molecular biomarkers predicting treatment response (Genetic and epigenetic, protein). Present a summary of molecular aberrations as potential target for novel agents (Ramucirumab, Bevacizumab, multi-tyrosine kinase inhibitors, EGFR inhibition based agents, Pi3K/AKT/mTor inhibitors, Met inhibitors, immune checkpoints inhibitors and insulin like growth factor monoclonal antibody). Finally, they review future candidates such as Gastrokine1, HDAC inhibitors and long non-coding RNAs.

**Comments**

-Title should state clearly that the report is a review.

Exploring the role of molecular biomarkers as a potential weapon against gastric cancer a review of the literature.

-Affiliations should not state author status, ex. Undergraduate student.

As suggested by the reviewer, Affiliations have been edited

-Although review is focused on the more adequate personalized treatments according to the altered molecular pathways in gastric cancer, some general recent articles could be also barely discussed, such as: - Cristescu et al., Nature Medicine, v21(5): 449-456, 2015 entitled: Molecular analysis of gastric cancer identifies subtypes associated with distinct clinical outcomes - The Cancer Genome Atlas Research Network: nature, 513: 202-209, 2014

As suggested by the reviewer, the recommended citations has been added to the main text

**A group of researchers used gene expression data to describe four molecular subtypes of GC linked disease progression and prognosis. The mesenchymal-like type with highest recurrence frequency (63%) of the four subtypes; Microsatellite-unstable tumors are hyper-mutated displaying the best overall prognosis and the lowest frequency of recurrence (22%) of the four subtypes;The tumor protein 53 (TP53)-active and TP53-inactive types include patients with intermediate prognosis and recurrence rates (with respect to the other two subtypes)[i] .**

**Scientists proposed a molecular classification dividing gastric cancer into four subtypes: for Epstein–Barr virus positive tumors with recurrent *PIK3CA* mutations, extreme DNA hypermethylation, and amplification of *JAK2*, *CD274* and *PDCD1LG2* ; microsatellite unstable tumourswith elevated mutation rates, including mutations of genes encoding targetable oncogenic signalling proteins; genomically stable tumours, with mutations of RHO-family GTPase-activating proteins; and tumours with chromosomal instability with marked aneuploidy and amplification of receptor tyrosine kinases. Identification of these molecular subtypes provides an efficient roadmap for patient stratification and targeted therapies [ii].**

Minor:

-Some long English sentences should be revised and shortened.

As suggested by reviewer, the whole text has been revised and shortened

-Pag 5 "… these signature genetic studies…" may be improved to "these genetic signature studies…"

As suggested by reviewer, these signature genetic studies…" has been changed to "these genetic signature studies

-Check errors such as: "monotherapyor" (pag 13) that should read as "monotherapy" and "or"…… "trastuzumabtherapy" (pag 14) should be "trastuzumab" and "therapy"…

As suggested by reviewer, : "monotherapyor" (pag 13) has been corrected to "monotherapy" and "or"…. "trastuzumabtherapy" (pag 14) has been corrected to "trastuzumab therapy"…

I found several through the whole text - Check misspellings: page 14: anatgonist, should be antagonist English should be revised thorough all text.

As suggested by reviewer, anatgonist has been corrected to antagonist. English has been revised thorough all text.

**Second comment**

The authors reviewed a role of biomarkers in gastric cancer. The manuscript is basically well written, however, the contents are a little bit different from the aim of this review.

In the first part of the manuscript, the authors actually reviewed the biomarkers that may predict response or prognosis of the treatment, however, the latter part of the manuscript the authors just listed the results of clinical trials not mentioned about the biomarkers.

As suggested by the reviewer,more details about the biomarkers in the main text were added in the latter part of the manuscript.

- **Several studies reported that The expression of VEGF and SSTR were associated with progression of GC [iii, iv]. A research group used a mouse model in which VEGF-A is expressed via adenovirus, enabling a stromal response marked by immune infiltration and angiogenesis, and**

identified specific stromal gene expression signatures to discover predictive biomarkers of therapeutic response, especially to immunotherapy and antiangiogenesis agents[v].

- Several research groups found that tyrosine kinase with immunoglobulin-like and EGF-like domains 1 (TIE-1) and mitogen-activated protein kinase kinase 4 (MKK4), serve as promising molecular biomarkers for gastric cancer prognosis. Overexpression of TIE-1 kinase in gastric cancer patients is associated with reduced survival rates[vi, vii, viii].
- Recent studies reported that serum HER2 levels are highly specific and demonstrated moderate diagnostic performance for HER2 tissue status in GC[ix, x, xi].
- Aberrant gastric MET activation can lead to increased mesenchymal characteristics and less epithelial features, and promote cancer cell stemness, invasion, metastasis, and chemo-resistance with repressed E-cadherin, which allows tumor cells to disseminate and spread throughout the body. Stress, and hypoxia could aggravate GC via MET, which is significantly correlated with prognosis[xii]..Many studies have suggested that MET protein GC patients[xiii, xiv.,xv].
- It has been suggested that while CTLA-4 may play a significant role in early immune response. Several checkpoints are involved in this process. CTLA-4 and PD-1 are both inhibitory receptors expressed by T cells. These molecules usually appear on the surface of T cells after their activation and send an inhibitory signal [xvi]. In GC, PD-1 expression on CD8+ lymphocytes is significantly higher than that of normal gastric mucosa and peripheral blood[xvii]. PD-L1 overexpression, may also serve as a predictive response biomarker in GC[xviii].
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The authors should revise the manuscript following above mentioned comments.

As suggested by the reviewer, the manuscript has been revised

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[iii]Zhao DQ, Chen J, Wu YF, Tian D, Zhou RX..Correlation between Vascular Endothelial Growth Factor and Somatostatin Receptor with Progression and Prognosis in Gastric Cancer.Hepatogastroenterology. 2014 Jun;61(132):1154-8.

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[v]Uhlik MT1, Liu J1, Falcon BL1, Iyer S2, Stewart J1, Celikkaya H2, O'Mahony M2, Sevinsky C, et al. Stromal-Based Signatures for the Classification of Gastric Cancer.Cancer Res. 2016 May 1;76(9):2573-86. doi: 10.1158/0008-5472.CAN-16-0022.

[vi]Lin W.C. et al. (1999) tie-1 protein tyrosine kinase: a novel independent prognostic marker for gastric cancer. Clinical Cancer Research: An Official Journal of the American Association for Cancer Research 5, 1745-1751

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J Gastrointest Surg. 2015 Sep;19(9):1565-71. doi: 10.1007/s11605-015-2888-y. Epub 2015 Jul 16.

[xii]Huang L, Wu R-L, Xu A-M. Epithelial-mesenchymal transition in gastric cancer.*American Journal of Translational Research*. 2015;7(11):2141-2158.
[xiii]Lee HE, Kim MA, Lee HS, Jung EJ, Yang HK, Lee BL, Bang YJ, Kim WH. MET in gastric carcinomas: comparison between protein expression and gene copy number and impact on clinical outcome. Br J Cancer. 2012;107:325–333.
[xiv]Ma J, Ma J, Meng Q, Zhao ZS, Xu WJ.Prognostic value and clinical pathology of MACC-1 and c-MET expression in gastric carcinoma. PatholOncol Res. 2013;19:821–832
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