

Peer Review File

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Comment 1: In the Figure 1C legend, most of the peptides were 5 to 26 amino acids in length and 700 to 3199 daltons in molecular weight. Please check the horizontal coordinates and vertical coordinates in the Figure 1C.

Reply 1: We have checked the horizontal coordinates and vertical coordinates in the Figure 1C, and the horizontal coordinates have been corrected as “Monoisotopic Mass (Daltons).

Change in the text: None.

Comment 2: The colors of Figure 3A and 3B were different. Why? And scale bar is needed to be added in the Figure 3A and 3B.

Reply 2: The colors of Figure 3A and 3B were different because the white balance value adjustment is inconsistent between the two figures and we have readjusted them to be consistent. The scale bar has been added in Figure 3A and 3B.

Change in the text: None.

Comment 3: In the experiments of transwell assay, wound healing assay and apoptosis testing, the control group is negative control or normal control? In these experiments, both negative control and normal control are need.

Reply 3: In the experiments of transwell assay, wound healing assay and apoptosis testing, the control group is normal control. Since we found that the phenotype of normal control and negative control was similar in our preliminary study, the synthesized peptide is expensive, so we only used normal control in the functional study and used the negative control in the mechanism study.

Change in the text: None.

Comment 4: In the Figure 3F, the cells were not clear. Please replace it with a new.

Reply 4: The old figure has been replaced.

Change in the text: None.

Comment 5: The illustrations of E and F were missing in the Figure 4 legend.

Reply 5: The illustrations of E and F in the Figure 4 legend have been added. E. The apoptosis cell percentage (percentage of both the upper and lower right quadrant cells) was compared between the ZYX₃₆₋₅₈ and control treated SKOV3 cells. F. The apoptosis cell percentage (percentage of both the upper and lower right quadrant cells) was compared between the ZYX₃₆₋₅₈ and control treated HO8910 cells.

Change in the text: E. Quantitative analysis of the apoptotic cells of ZYX₃₆₋₅₈ peptide and control solvent treated SKOV3 cells. F. Quantitative analysis of the apoptotic cells of ZYX₃₆₋₅₈ peptide and control solvent treated A2780 cells. (See Page 25, line 590-592)

Comment 6: In the Figure 5A, the arrowhead was indicated for what?

Reply 6: The arrowhead indicated the different band between ZYX₃₆₋₅₈ and scrambled peptide treated group.

Change in the text: The arrowhead indicated the different band between ZYX₃₆₋₅₈ and scrambled peptide treated group. (See Page 26, line 597-598)

Comment 7: Why to choose ZYX₃₆₋₅₈ to research in the paper? Please elaborate in detail in the introduction.

Reply 7: In this study, three peptides derived from the precursor proteins F13A1, ZYX and SNP23, respectively, which were down-regulated in the serum of ovarian cancer patients were chosen for the functional analysis. It was found that only ZYX₃₆₋₅₈ could significantly inhibit the invasion and migration of ovarian cancer cells. Therefore, we chose ZYX₃₆₋₅₈ for further research. Our results show that the endogenous peptide ZYX₃₆₋₅₈ has great promise in the treatment of ovarian cancer. We have elaborated this in detail in the introduction.

Change in the text: (See Page 4, line 70-76)

Comment 8: Why to choose TSP1 in the investigation of mechanism? Please elaborate in detail in the discussion.

Reply 8: Firstly, TSP1 can be specifically detected in the peptide pull down lysate of ZYX₃₆₋₅₈ treated SKOV3 cells by mass spectrometry. Second, only TSP1 was specifically detected in the eluent of the biotin-labeled ZYX₃₆₋₅₈ compared with the biotin-labeled control peptide by Western blot verification experiment, indicating that TSP1 was the most abundant protein in the eluent of biotin-labeled ZYX₃₆₋₅₈. Third, TSP1 have been reported to be a tumor suppressor of ovarian cancer and TSP1 protein was elevated in the ZYX₃₆₋₅₈ treated SKOV3 cells. Due to the reasons above, TSP1 was chosen for the investigation of the mechanism

Change in the text: The reasons have been explained in the manuscript.

Comment 9: There are several severe mistakes in the manuscript. Please read the manuscript carefully, and correct it.

Reply 9: We have read the manuscript carefully and the mistakes have been corrected.

Change in the text: The changed text was marked in red.

Comment 10: How about the effect of ZYX₃₆₋₅₈ in cancer animal models?

Reply 10: The first author Xusu Wang needs to graduate using this article, however, due to the epidemic of SARS-CoV-2, animal experiments have not been conducted.

Change in the text: None.