

## **Reviewer #2:**

This review article summarizes the resistance mechanisms for chemotherapy, radiotherapy, immunotherapy, and targeted therapy alternatives used in colorectal cancer. Additionally, recent advances in nanotechnology in CRC therapy were assessed in terms of therapeutic resistance and preventing metastasis in this review paper. Overall paper is well written and intellectually sound, however it could be improved by expanding on a few points and providing more details. Below are suggestions for improving the manuscript.

**Comment 1.** Pharmacokinetic effects (absorption, distribution, metabolism, and elimination) can also limit the amount of drug which reaches the tumor. Authors are requested to make mention of the pharmacokinetic effects in the chemotherapy resistance section of the manuscript.

**Response 1:** Thanks for the valuable suggestions. Modification has been made in the chemotherapy resistance section with the pharmacokinetic effects (Page 6, paragraph 2 of section "*Chemotherapy resistance*").

**Comment 2.** The phosphatidylinositol-3-kinase (PI3K)/protein kinase B (Akt) pathway is one of the well known pathways for the development and progression of many solid cancers. PI3K signalling and its downstream effector Akt are considered one of the important reasons of chemoresistance in colorectal cancer therapy. The reviewers should additionally mention the PI3K/AKT/mTOR signalling pathway importance in the chemotherapy resistance section of the manuscript.

**Response 2.** Thanks for the valuable suggestion. The importance of PI3K/AKT/mTOR signalling pathway has been added in the chemotherapy resistance section (Page 8, paragraph 5 of section "*Chemotherapy resistance*").

**Comment 3.** Recent studies have also showed a link between chemotherapy resistance and the epithelial–mesenchymal transition (EMT) phenotype. Authors indicated that the

importance of the EMT for radiotherapy resistance. Please, give a detail for the EMT-chemotherapy resistance link and also the other resistance types.

**Response 3:** Thanks for the valuable suggestion. The importance of the EMT for radiotherapy resistance has been added in the manuscript (Page 9, paragraph 7 of section "*Chemotherapy resistance*"; Page 14, paragraph 4 of section "*Immunotherapy resistance*"; Page 17, paragraph 7 of section "*Targeted therapy resistance*")

**Comment 4.** Hypoxia has a crucial role in radiotherapy resistance and it could be a cause for treatment failure after radiotherapy. The role of hypoxia in radiotherapy resistance should be addressed in the related part of the manuscript.

**Response 4:** Thanks for the valuable suggestion. The role of hypoxia in radiotherapy resistance has been addressed in the manuscript (Page 11, paragraph 2 of section "*Radiotherapy resistance*").

**Comment 5.** The authors should also mention Anti-VEGF alternative for the CRC therapy and resistance problem in the targeted therapy resistance section of the manuscript.

**Response 5:** Thanks for the valuable suggestion. Anti-VEGF alternative for the CRC therapy and resistance problem have been added in the targeted therapy resistance section of the manuscript (Page 17, paragraph 6 of section "*Targeted therapy resistance*").

**Comment 6.** Recently, extracellular vesicles (EVs), a heterogeneous group of vesicles involved in cell-to-cell communication, have been shown to contribute to drug therapy resistance as crucial modulators in a variety of cancer. The importance and role of EVs in therapy resistance for cancer should be addressed throughout the manuscript.

**Response 6:** Thanks for the valuable suggestion. The importance and role of EVs have been addressed in the manuscript (Page 9, paragraph 8 of section "*Chemotherapy resistance*").

**Comment 7.** Exosome has been reported as an important potential system that could be effectively used as a bioinspired, bioengineered, and biomimetic drug delivery solution considering its toxicity, immunogenicity, and rapid clearance by the mononuclear phagocyte system. Exosome-mimetic vesicles are receiving much interest for developing nano-sized delivery systems

**Response 7:** Thanks for the valuable suggestion. The importance of exosomes has been added in the manuscript (Page 20, paragraph 1 of section “APPLICATION OF NANOTECHNOLOGY IN CONQUERING THERAPEUTIC RESISTANCE AND METASTASIS”).

**Comment 8.** Recently, exosomes, one of the important subgroup of EVs, have been proposed as an important potential system that might be effectively used for drug delivery solution. They have advantages and disadvantages compared to liposomes and nanoparticles. Authors are requested to mention to exosomes as an alternative for drug delivery solution in this review article.

**Response 8:** Thanks for the valuable suggestion. The role of exosomes as an alternative for drug delivery solution has been addressed in the manuscript (Page 24, paragraph 6 of section “*Nanotechnology and chemotherapy*”; Page 26, paragraph 5 of section “*Nanotechnology and radiotherapy*”; Page 29, paragraph 5 of section “*Nanotechnology and gene therapy*”; Page 32, paragraph 6 of section “*Nanotechnology and immuno- and targeted therapies*”).

**Comment 9.** Resolution of figure 1 is not good. White color shouldn't be preferred in the text of the figure

**Response 9:** Thanks for the valuable suggestion. The figure has been modified accordingly (Figure 1).