



PEER-REVIEW REPORT

Name of journal: *World Journal of Gastrointestinal Oncology*

Manuscript NO: 110736

Title: TRPM6 and TRPM6/7 antagonists suppress colon adenocarcinoma HT-29 cells

Provenance and peer review: Invited manuscript; Externally peer reviewed

Peer-review model: Single blind

Reviewer's code: 07885321

Position: Peer Reviewer

Academic degree and professional title: PhD, Professor, Senior Researcher

Reviewer's Country/Territory: China

Author's Country/Territory: Thailand

Manuscript submission date: 2025-06-13

Reviewer chosen by: AI Editor

Reviewer accepted review: 2025-06-17 00:52

Reviewer performed review: 2025-06-17 09:02

Review time: 8 Hours

Content to be reviewed	Does the manuscript's content fall within the scope of the journal? Yes Is there any Key Word that is not included in the manuscript title? No Do authors' affiliations correspond to the content of the manuscript? Yes Does the Abstract contain the contents of each part of the manuscript (IMRaD)? Yes Are the Key Words complete? Yes Is the content of the Introduction adequate? Yes
-------------------------------	---



Baishideng Publishing Group

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

	<p>Is the content of the Materials and Methods complete? Yes</p> <p>Is the description of the experiments clear and complete? Yes</p> <p>Are the experimental data presented in the manuscript's biostatistics content reliable? Yes</p> <p>Are the experimental data of the Results true and reliable? Yes</p> <p>Are the quality and resolution of the images up to standard? Yes</p> <p>Do the selection and design of the figures and tables follow the principles of necessity and clarity? Yes</p> <p>Is there any duplication between various parts of the manuscript and between the main text and the content presented in the figures and tables? No</p> <p>Are the figures and tables numbered consecutively in the order in which they appear in the manuscript? Yes</p> <p>Is the content of the Discussion reasonable? Yes</p> <p>Is the Conclusion reasonable? Yes</p> <p>Are all references necessary and reasonable? No</p> <p>Do authors omit important references? Yes</p> <p>Are all references related to the topic of the manuscript? Yes</p> <p>Do authors only cite their own earlier publications? No</p> <p>Is the manuscript's text correct, concise, and clear? Yes</p> <p>Will the manuscript's content be of interest to readers? Yes</p> <p>Are additional experiments needed for the study? Yes</p> <p>Does the research scope comply with ethics? Yes</p>
<p>Scientific quality</p>	<p>Grade B (Very good)</p>



Novelty of this manuscript	Grade A (Excellent)
Creativity or innovation of this manuscript	Grade A (Excellent)
Scientific significance of the conclusion in this manuscript	Grade B (Very Good)
Language quality	Grade B (Very good)
Does this manuscript describe a study of the existing knowledge system?	No
Does this manuscript report a revolutionary innovation?	No
Does this manuscript report an unconventional innovation?	No
Conclusion	Major revision
Re-review	Yes
Peer-reviewer statements	Peer-Review: Anonymous
	Conflicts-of-Interest: No
Are your review comments generated by AI tools?	

SPECIFIC COMMENTS TO AUTHORS

This study investigates the importance of membrane-localized TRPM6 and TRPM6/7 channels in Mg²⁺ influx regulation, CRC spheroid formation, cancer stem-like cell maintenance, and cell migration. The manuscript is well-structured, and the experiments are meticulously designed. The findings suggest that targeting TRPM6/7-mediated Mg²⁺ transport could be a novel therapeutic strategy for CRC. However, several issues need to be addressed to strengthen the manuscript.

1. Inhibitor Specificity and Mechanistic Validation

The inhibitors used (Co(III)hex, 2-APB, ME) may have off-target effects. To



**Baishideng
Publishing
Group**

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

unequivocally link the observed phenotypes to TRPM6/7 inhibition, the authors should: Perform genetic knockdown or knockout experiments (e.g., siRNA, shRNA, or CRISPR-Cas9) targeting TRPM6 and/or TRPM7. This would confirm that the effects on Mg^{2+} influx, spheroid formation, and migration are indeed mediated by these channels. Include rescue experiments where TRPM6/7 expression is restored in knockdown cells to verify phenotype reversal.

Provide dose-response curves for the inhibitors to establish their specificity and potency in the experimental system.

2. In Vivo Validation

The current study is limited to in vitro models, which restricts its translational relevance. To address this:

The authors should consider incorporating xenograft experiments using HT-29 spheroids in immunocompromised mice to evaluate the effects of TRPM6/7 inhibition on tumor growth and metastasis.

If in vivo experiments are not feasible, the authors should at least analyze TRPM6/7 expression in CRC patient datasets (e.g., TCGA or GEO) to correlate channel expression with clinical outcomes (e.g., survival, tumor stage). This would strengthen the clinical relevance of the findings.

3. Discussion Refinement

The Discussion section is overly speculative in places, particularly regarding the roles of Mg^{2+} in mitochondrial function and PI3K signaling.

The authors should tighten the narrative around their key findings and avoid extensive speculation unsupported by their data.

If the authors wish to discuss potential mechanisms (e.g., PI3K signaling), they should either provide experimental evidence (e.g., Western blots of pathway components) or explicitly state that these are hypotheses for future study.



**Baishideng
Publishing
Group**

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

4. Updated References

A significant number of references are outdated (pre-2015). The authors should:

Cite recent studies (2020–2024) on TRPM6/7 in cancer, particularly those exploring their roles in stemness and metastasis.

Include clinical studies linking Mg^{2+} homeostasis to CRC progression or treatment resistance.

Final Decision:

Major Revision Required.



PEER-REVIEW REPORT

Name of journal: *World Journal of Gastrointestinal Oncology*

Manuscript NO: 110736

Title: TRPM6 and TRPM6/7 antagonists suppress colon adenocarcinoma HT-29 cells

Provenance and peer review: Invited manuscript; Externally peer reviewed

Peer-review model: Single blind

Reviewer’s code: 08447451

Position: Peer Reviewer

Academic degree and professional title: Academic Fellow, MD, PhD, Professor

Reviewer’s Country/Territory: China

Author’s Country/Territory: Thailand

Manuscript submission date: 2025-06-13

Reviewer chosen by: Hong-Xin Jiang

Reviewer accepted review: 2025-06-24 14:35

Reviewer performed review: 2025-06-24 14:49

Review time: 1 Hour

Content to be reviewed	<p>Does the manuscript’s content fall within the scope of the journal? Yes</p> <p>Is there any Key Word that is not included in the manuscript title? Yes</p> <p>Do authors’ affiliations correspond to the content of the manuscript? Yes</p> <p>Does the Abstract contain the contents of each part of the manuscript (IMRaD)? Yes</p> <p>Are the Key Words complete? Yes</p> <p>Is the content of the Introduction adequate? Yes</p>
-------------------------------	---



Baishideng Publishing Group

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

	<p>Is the content of the Materials and Methods complete? Yes</p> <p>Is the description of the experiments clear and complete? Yes</p> <p>Are the experimental data presented in the manuscript's biostatistics content reliable? Yes</p> <p>Are the experimental data of the Results true and reliable? Yes</p> <p>Are the quality and resolution of the images up to standard? Yes</p> <p>Do the selection and design of the figures and tables follow the principles of necessity and clarity? Yes</p> <p>Is there any duplication between various parts of the manuscript and between the main text and the content presented in the figures and tables? Yes</p> <p>Are the figures and tables numbered consecutively in the order in which they appear in the manuscript? Yes</p> <p>Is the content of the Discussion reasonable? Yes</p> <p>Is the Conclusion reasonable? Yes</p> <p>Are all references necessary and reasonable? Yes</p> <p>Do authors omit important references? Yes</p> <p>Are all references related to the topic of the manuscript? Yes</p> <p>Do authors only cite their own earlier publications? Yes</p> <p>Is the manuscript's text correct, concise, and clear? Yes</p> <p>Will the manuscript's content be of interest to readers? Yes</p> <p>Are additional experiments needed for the study? Yes</p> <p>Does the research scope comply with ethics? Yes</p>
<p>Scientific quality</p>	<p>Grade C (Good)</p>



Novelty of this manuscript	Grade B (Very Good)
Creativity or innovation of this manuscript	Grade B (Very Good)
Scientific significance of the conclusion in this manuscript	Grade B (Very Good)
Language quality	Grade A (Excellent)
Does this manuscript describe a study of the existing knowledge system?	Yes
Does this manuscript report a revolutionary innovation?	Yes
Does this manuscript report an unconventional innovation?	Yes
Conclusion	Major revision
Re-review	Yes
Peer-reviewer statements	Peer-Review: Anonymous
	Conflicts-of-Interest: No
Are your review comments generated by AI tools?	

SPECIFIC COMMENTS TO AUTHORS

Overall, this is an interesting study showing that blocking TRPM6 and TRPM6/7 channels disrupts magnesium uptake, spheroid formation, stem-like traits and migration in HT-29 colorectal cancer cells. The authors combine biochemical assays, mass spectrometry and functional experiments to make a solid case that these channels help CRC spheroids survive and move. The data are convincing and the topic is timely, since targeting ion channels in cancer stem cells is an emerging area. However, the manuscript could use a bit more polish and deeper context, especially around how these findings fit with dietary magnesium studies and known signaling pathways.



**Baishideng
Publishing
Group**

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

In the Abstract, the background and aim are clear, but a few sentences feel overloaded. For example, the line “Although TRPM7 has been implicated... remains largely unexplored” is a mouthful; you might split it or trim it down. Under the Methods subtitle, I spotted “Parental HT-29 cells and spheroid-derived (SP) HT-29 cells at comparable passages were used as in vitro models.” It would read more smoothly as “We used parental and spheroid-derived HT-29 cells at comparable passages as our in vitro models.” Also, the phrase “quantified using fluorescent imaging and biochemical assays” could be more specific, what kind of biochemical assays? A tiny clarification helps. This is a cancer study, In the introduction, I recommend opening your report with a cohesive paragraph that introduces cancer biology and treatment modalities, cite review by Chief of US National Cancer Institute D. Sonkin, A. Thomas “Cancer Treatments: Past, Present, and Future (2024)” . The you need to cite previous TRPM7 studies in cancer, and emphasize the contribution of Lin’s lab in Stony Brook, such as “Lidocaine Suppresses Viability and Migration of Human Breast Cancer Cells: TRPM7 as A Target for Some Breast Cancer Cell Lines, 2021”and “The Role of Transient Receptor Potential Melastatin 7 (TRPM7) in Cell Viability: A Potential Target to Suppress Breast Cancer Cell Cycle, 2020”

In the Results section, a few grammar glitches stand out. Under “TRPM6 and TRPM7 expression in cancer spheroids,” the phrase “both total and membrane TRPM7 expression levels were markedly higher in SP cells than in parental cells” repeats “expression levels” twice. Maybe change the second to “both total and membrane TRPM7 were markedly higher...” In the Nano-LC-MS/MS subtitle, the sentence “After determining the total number of phosphorylated residues..., we found that the parental and SP groups had 159 and 185 phosphorylated residues, respectively” is clear, but the next sentence tacks on too much detail – consider breaking it for readability.

Throughout the manuscript, some statements would benefit from extra citations. For



**Baishideng
Publishing
Group**

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

instance, when you say that magnesium is crucial for mitochondrial ATP production and oxidative phosphorylation, add one or two primary references on Mg^{2+} in bioenergetics. Also, the idea that PI3K signaling links Mg^{2+} influx to Nanog, COX-2 and MMP-9 expression needs stronger backing, there's literature on PI3K regulation of stemness markers and metalloproteinases that could be cited.

In the Discussion, it would strengthen your paper to more thoroughly compare your findings with the meta-analysis suggesting a U-shaped relationship between dietary magnesium and CRC risk, and the mouse-model data showing Mg^{2+} supplementation can suppress tumors. A brief paragraph considering why intracellular Mg^{2+} from TRPM channels might behave differently than systemic Mg^{2+} would add depth. You should also discuss TRPM channels in other cancer types rather than only colon cancer, should cite pan-cancer study in TRPM7 "A pan-cancer-bioinformatic-based literature review of TRPM7 in cancers, 2022"

Finally, the manuscript occasionally leans heavily on passive voice and dense phrasing—shortening a few complex sentences and switching to active voice will make it flow better. I recommend citing "Different Strategies for Cancer Treatment: Targeting Cancer Cells or Their Neighbors? (2025)", which offers a comprehensive framework comparing the merits and limitations of directly targeting malignant cells versus modulating the surrounding stroma and immune components, discuss how the mechanisms discussed in the present work can help develop cancer treatment Strategies. the authors can more clearly illustrate how their findings might inform the design of next-generation treatment regimens that optimally balance cell-autonomous and microenvironmental interventions. Try to discuss how TRPM7 can be applied. For example, Recent advances in circulating tumor DNA (ctDNA) analysis have enabled highly sensitive and specific detection of oncogenic mutations in plasma. For example, the study "Ultra-sensitive detection of melanoma NRAS mutant ctDNA based on



**Baishideng
Publishing
Group**

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

programmable endonucleases” demonstrates how programmable endonuclease-mediated cleavage can enrich rare NRAS mutant fragments from a background of wild-type DNA, achieving detection limits below 0.1% mutant allele frequency. Citing this work is important because you candiscuss if TRPM gene can be used in liquid biopsy.

When discussing the translational potential of the TRPM function identified in this study. it is worth exploring their candidacy as novel drug targets through the lens of traditional medicine. For instance, “Identification of Molecular Targets of Hypericum perforatum in Blood for Major Depressive Disorder: A Machine-Learning Pharmacological Study (2024)” demonstrates how network-pharmacological analyses can systematically map active herbal compounds to human disease targets, offering a blueprint for similar investigations in oncology. Moreover, clinical and preclinical reports – such as “Effect of Traditional Medicine on Clinical Cancer (2020)” and “Harnessing Traditional Medicine and Biomarker-Driven Approaches to Counteract Trichostatin A-Induced Esophageal Cancer Progression (2025)” – provide compelling evidence that phytochemicals and complex herbal formulations can modulate tumor growth and treatment response. Mechanistic studies like “Toxic Medicine Used in Traditional Chinese Medicine for Cancer Treatment: Are Ion Channels Involved? (2022)” this is especially need to be discussed as it is talking about ion channel targeting for cancer, and a follow up study “An Example of Toxic Medicine Used in Traditional Chinese Medicine for Cancer Treatment (2023)” where targets ion channels further highlight the importance of dissecting molecular pathways to ensure both efficacy and safety.

Overall, a solid piece of work that just needs a bit more context, some extra citations, and light editing to polish up grammar and clarity. I look forward to seeing this improved version.



RE-REVIEW REPORT OF REVISED MANUSCRIPT

Name of journal: *World Journal of Gastrointestinal Oncology*

Manuscript NO: 110736

Title: TRPM6 and TRPM6/7 antagonists suppress colon adenocarcinoma HT-29 cells

Provenance and peer review: Invited manuscript; Externally peer reviewed

Peer-review model: Single blind

Reviewer’s code: 08447451

Position: Peer Reviewer

Academic degree and professional title: Academic Fellow, MD, PhD, Professor

Reviewer’s Country/Territory: China

Author’s Country/Territory: Thailand

Manuscript submission date: 2025-06-13

Reviewer chosen by: Shang Wu

Reviewer accepted review: 2025-08-01 13:16

Reviewer performed review: 2025-08-01 13:19

Review time: 1 Hour

Content to be reviewed	<p>Does the manuscript’s content fall within the scope of the journal? Yes</p> <p>Is there any Key Word that is not included in the manuscript title? Yes</p> <p>Do authors’ affiliations correspond to the content of the manuscript? Yes</p> <p>Does the Abstract contain the contents of each part of the manuscript (IMRaD)? Yes</p> <p>Are the Key Words complete? Yes</p> <p>Is the content of the Introduction adequate? Yes</p>
-------------------------------	---



Baishideng Publishing Group

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

	<p>Is the content of the Materials and Methods complete? Yes</p> <p>Is the description of the experiments clear and complete? Yes</p> <p>Are the experimental data presented in the manuscript's biostatistics content reliable? Yes</p> <p>Are the experimental data of the Results true and reliable? Yes</p> <p>Are the quality and resolution of the images up to standard? Yes</p> <p>Do the selection and design of the figures and tables follow the principles of necessity and clarity? Yes</p> <p>Is there any duplication between various parts of the manuscript and between the main text and the content presented in the figures and tables? Yes</p> <p>Are the figures and tables numbered consecutively in the order in which they appear in the manuscript? Yes</p> <p>Is the content of the Discussion reasonable? Yes</p> <p>Is the Conclusion reasonable? Yes</p> <p>Are all references necessary and reasonable? Yes</p> <p>Do authors omit important references? Yes</p> <p>Are all references related to the topic of the manuscript? Yes</p> <p>Do authors only cite their own earlier publications? Yes</p> <p>Is the manuscript's text correct, concise, and clear? Yes</p> <p>Will the manuscript's content be of interest to readers? Yes</p> <p>Are additional experiments needed for the study? Yes</p> <p>Does the research scope comply with ethics? Yes</p>
<p>Scientific quality</p>	<p>Grade B (Very good)</p>



Baishideng Publishing Group

7041 Koll Center Parkway, Suite
160, Pleasanton, CA 94566, USA
Telephone: +1-925-399-1568
E-mail: office@baishideng.com
https://www.wjgnet.com

Novelty of this manuscript	Grade B (Very Good)
Creativity or innovation of this manuscript	Grade B (Very Good)
Scientific significance of the conclusion in this manuscript	Grade B (Very Good)
Language quality	Grade B (Very good)
Does this manuscript describe a study of the existing knowledge system?	Yes
Does this manuscript report a revolutionary innovation?	Yes
Does this manuscript report an unconventional innovation?	Yes
Conclusion	Accept
Peer-reviewer statements	Peer-Review: Anonymous
	Conflicts-of-Interest: No
Are your review comments generated by AI tools?	No

SPECIFIC COMMENTS TO AUTHORS

ok