Name:	**UTTAR PRADESH JOURNAL OF ZOOLOGY**
Manuscript Number:	**Ms_UPJOZ_4555**
Title of the Manuscript:	**Mode of action of retinoic acid in the regulation of glucose metabolism in mud crab, Scylla serrata: evidence for the involvement of CHH**
Type of the Article	**Research**

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PART 1: Comments

	Reviewer's comment	**Author's Feedback** *(Please correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)*
Please write a few sentences regarding the importance of this manuscript for the scientific community. A minimum of 3-4 sentences may be required for this part.	This manuscript explores the role of retinoids in regulating carbohydrate metabolism in crustaceans, specifically the mud crab *Scylla serrata*. By demonstrating the hyperglycemic effects of 9-Cis retinoic acid (9CRA) through its influence on crustacean hyperglycemic hormone (CHH) release, the study aims at advancing the understanding of endocrine control in marine organisms. These findings have significant implications for aquaculture and ecological research, particularly in understanding metabolic responses to physiological and environmental stressors.	

	Moreover, the study contributes to broader knowledge of retinoid-mediated pathways and offers potential comparative insights into similar metabolic processes in other arthropods or even vertebrates.	
Is the title of the article suitable? **(If not please suggest an alternative title)**	The title effectively conveys the focus of the study. However, CHH (crustacean hyperglycemic hormone) could be clearly written in full to inform readers. Alternatively, the title could be slightly refined for clarity. Here is an alternative suggestion: **"Retinoic Acid Regulation of Glucose Metabolism in Mud Crab (*Scylla serrata*): Role of Crustacean Hyperglycemic Hormone**	
Is the abstract of the article comprehensive? Do you suggest the addition (or deletion) of some points in this section? Please write your suggestions here.	The abstract describes the experimental design (e.g., eyestalk ablation, retinoid injections) but does not clarify the number of crabs used or controls employed. Including this information briefly would improve scientific rigor. In addition, there are some repetitive phrases (e.g., "intact crabs") that could be streamlined for better readability.	
Is the manuscript scientifically, correct? Please write here.	Yes.	
Are the references sufficient and recent? If you have suggestions of additional references, please mention them in the review form.	Yes.	
Is the language/English quality of the article suitable for scholarly communications?	Yes.	
<u>Optional/General</u> comments	• While the introduction mentions gaps in understanding CHH regulation and RA's role in crustaceans, it could more explicitly highlight how the study addresses these gaps. **Suggestion:** Add a specific statement outlining the aspects of the study, such as "This study explores the underexamined role of RA isomers in carbohydrate metabolism and their potential regulatory interactions with CHH in crustaceans." • Some points are repeated, such as the presence of RA isoforms and RXR	

	receptors in crustaceans. These could be streamlined to avoid repetition. • Define terms like RA, ATRA, and 9CRA on their first mention to aid readers unfamiliar with these compounds. • The sentence "In this study, we selected *S. serrata* as an experimental model..." could be moved closer to the end of the introduction as a transition into the study's objectives • The introduction jumps between topics (e.g., CHH, RA effects, vitamin A supplementation), which disrupts the logical flow. A more structured approach could be: • Importance of glucose metabolism in crustaceans. • Role of CHH and endocrine regulation. • Known effects of RA in vertebrates and crustaceans. • Specific research gaps and study objectives. **In the methods section** • The section mentions injecting retinoic acid isomers into crabs at the base of the walking legs, but more detail could be provided on the exact volume of injection per animal (e.g., µL per leg) and how this might differ between treatments. Also, the potential effect of injection site variation (if any) on results could be considered or discussed briefly. • In the section detailing biochemical analysis, there are instances where measurement units are presented without proper space (e.g., "10 µL volumes" vs "10µL"). Standardizing this formatting for units would improve the readability. • The housing section provides good detail on the acclimatization process, but specifying the tank size and the number of crabs per tank would enhance the transparency of the experimental setup. Additionally, the temperature and salinity conditions are well-communicated but could be further explained if those parameters have a specific relevance to the experimental outcomes. In the discussions and conclusions • The statement that ATRA injection did not induce hyperglycemia contrasts with the effect of 9CRA. This is an important observation that could warrant further discussion regarding the specific roles of 9CRA and ATRA in glucose metabolism. The difference between these two isomers should be elaborated on, especially in terms of their molecular actions and receptor interactions in	

	crustaceans. • The **Conclusion** would benefit from a bit more context or a sentence reiterating the potential mechanism of action (i.e., the involvement of CHH and RXR in mediating this response). Specifically, - Provide more detail on how glucose is released into the hemolymph post-glycogen breakdown. - Provide a brief discussion of why ATRA did not induce hyperglycemia, including potential differences in molecular pathways. - Reinforce future research goals, particularly related to the RXR receptor, to clarify how the findings can be built upon in subsequent studies.	

PART 2:

	Reviewer's comment	**Author's comment** *(if agreed with reviewer, correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)*
Are there ethical issues in this manuscript?	*(If yes, Kindly please write down the ethical issues here in details)*	

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