Reviewer 3

The Authors are grateful to the reviewer for their kind suggestions and queries; we addressed most of them.

The main goal of the present paper is to develop easy and reproducible method to get a stable and biocompatible AuNR with promising application as photothermal agent. The authors used thiolated pectin (SH-PEC) to replace the CTAB, which is toxic. They demonstrated that SH-PEC is capable to replace the CTAB adsorbed on the surface of AuNRs. However, there some points to be analyzed:

1.Mine main concern is regarding the novelty of the paper, because the thiolated pectin (SH-PEC) have already used on gold nanoparticles, therefore the novelty will be limited to the shape (nanorods) what is not enough, see for example the following articles:

a) Artif Cells Nanomed Biotechnol

Shaivee Borker , Varsha Pokharkar .

2018;46:826-835.

doi: 10.1080/21691401.2018.1470525.

Engineering of pectin-capped gold nanoparticles for delivery of doxorubicin to hepatocarcinoma cells: an insight into mechanism of cellular uptake.

Thank you for your query and for the given references. Absolutely yes, there are many reports in which pectin has been used as stabilizer of plasmonic gold nanoparticular. About the novelty of paper, we would like to expose that SH-PEC has been widely used to coating and stabilize plasmonic gold nanoparticles of spherical geometry. In this regard, we proposed the use of SH-PEC to coat and stabilize gold nanorods, since there are no research documents (at least in the consulted bibliography by this research group) in which SH-PEC has been used with this purpose. To attend this query, the Authors rewrite the information as follow:

These stabilization techniques enable precise control over the physicochemical attributes of gold nanorods (AuNRs) while augmenting their functionality. This broadens the spectrum of potential applications for AuNRs, encompassing areas such as biomedicine, electronics, and beyond (Fan, Cheng, & Sun, 2020; Goodwin, 2021; Hakimian & Ghourchian, 2019; Madani et al., 2018; Mahalunkar et al., 2019). Polysaccharides emerges as alternative due to their low cost of production, biocompatibility, and reactivity. Anionic and cationic polysaccharides, such as alginic acid (Figueiredo, Rodrigues, Fernandes, Correia, & Moreira, 2023), , hyaluronic acid (L. S. Li et al., 2021), chitosan (de Barros et al., 2016), gum arabic (Gonçalves et al., 2021, Guzmán-Rocha, Córdova-Fraga, Bernal-Alvarado, & Cano-González, 2022) cellulose (Doghish et al., 2022), starch (You et al., 2020) thiolated dextran (Choi et al., 2010)and PEC (Attallah, Al-Ghobashy, Nebsen, El-Kholy, & Salem, 2018; Cisneros et al., 2021; Nemiwal, Zhang, & Kumar, 2021) have been used to coat and stabilize spherical gold nanoparticles (D. A. de Almeida et al., 2020; Y. Te Liao et al., 2019; Xu et al., 2017; Yoon et al., 2019; Zhang & Lin, 2014). In the present report, pectin (PEC)

has been proposed to stabilize gold nanorods. This biopolymer is an anionic heteropolysaccharide, which, like to other biopolymers, has been widely considered in the pharmaceutical area as an encapsulation agent for drugs and active compounds thanks to its solubility, biocompatibility, and biodegradability properties (Martau, Mihai, & Vodnar, 2019, Kedir, Deresa, & Diriba, 2022). Additionally, PEC has a grafted structure, distinct from other polysaccharides of linear structure used in the stabilization of gold nanoparticles and gold nanorods (Attallah, Al-ghobashy, Nebsen, El-kholy, & Salem, 2018; Beltrán et al., 2022). Furthermore, PEC has been the subject of numerous studies and applications due to its chemical composition facilitates its functionalization or chemical modification with different types of molecules (Borker & Pokharkar, 2018; Cisneros et al., 2021; D. qiang Li et al., 2021). It is important to note that at least in the consulted bibliography by this research group, there are no reports in which the PEC was used to stabilize AuNRs.

2. The removal of CTAB from the surface of Au Nps was demonstrated, but not from the solution. After removal form surface, CTAB should be not detectable by NMR spectroscopy, surface-enhanced Raman spectroscopy, or using pH dependent ζ-potential measurements. This test is essential because the use of PEC for removal of CTAB is the main aim of the present work.

Thanks for the comment. We would like to give highlights about the strategy followed to get AuNR-CTAB and AuNR@SH-PEC. Firstly, the AuNRs were synthesized using a 0.1 M solution of CTAB. After the growth process and get the gold nanorods, these were centrifuged and resuspended in a 10 mM CTAB solution to keep them in suspension until the coating process with SH-PEC. It is important to note, that AuNRs in 10 mM of CTAB solution, the shape of AuNR remains preserved. After the SH-PEC polymer coating process, an exhaustive washing process was conducted. This involved three washing with water (mili-Q) by centrifugation to ensure the majority of CTAB molecules were removed from the solution. Furthermore, the observed charge inversion in zeta potential measurements and the enhanced cell viability suggest that the amount of CTAB molecules remaining in the solution is negligible.

3) The literature review is missing important works, for example the article mentioned in the point 2.

To attend this suggestion, additional information and references was added.

In the present report, pectin (PEC) has been proposed to stabilize gold nanorods. This biopolymer is an anionic heteropolysaccharide, which, like to other biopolymers, has been widely considered in the pharmaceutical area as an encapsulation agent for drugs and active compounds thanks to its solubility, biocompatibility, and biodegradability properties (Martau, Mihai, & Vodnar, 2019, Kedir, Deresa, & Diriba, 2022). Additionally, PEC has a grafted structure, distinct from other polysaccharides of linear structure used in the stabilization of gold nanoparticles and gold nanorods (Attallah, Al-ghobashy, Nebsen, El-kholy, & Salem, 2018; Beltrán et al., 2022). Furthermore, PEC has been the subject of numerous studies and applications due to its chemical composition facilitates its functionalization or chemical modification with different types of molecules (Borker & Pokharkar, 2018; Cisneros et al., 2021; D. qiang Li et al., 2021). It is important to note that at least in the consulted bibliography by this research group, there are no reports in which the PEC was used to stabilize AuNRs.