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Extracellular Vesicle-based Delivery: Biotechnological Approaches for Exogenous Loading

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Extracellular vesicles (EVs) are naturally derived nanoparticles released by different cells to mediate cell-to-cell communication. Their inherent ability to transport biological materials and information makes them highly promising as drug delivery systems. We set up a biotechnological platform for lab-scale vesicle production and exogenous loading of relevant molecules. Here we use EVs derived from microalgae Tetraselmis chuii, named nanoalgosomes, focusing on the loading of both small drugs and large-size macromolecules, namely proteins and ds-dna, the latter still posing a significant challenge. Our process optimization includes the following steps: EV isolation from cell cultures by using TFF; perturbation of EV membrane by different methods to induce molecule encapsulation; purification of loaded product by different chromatographic techniques; structural and physico-chemical characterization by using different biophysical techniques; assessment of EVs biological activities in order to validate and control their function and their mode of action. The biophysical and biochemical characterization of loaded EVs allows to validate the biotechnological approach for efficient loading of both small and large cargoes into extracellular vesicles, confirming that nanoalgosomes are biocompatible EVs suited to carry not only small cargoes but also large-size macromolecules.