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Abstract P-16: Cryo-Electron Microscopy Study of Vesicles from Various Species

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Background: Plant-derived extracellular vesicles (PEVs) are studied as a natural carrier of functional biomolecules and as a potential system of targeted delivery of therapeutic agents. One of the urgent tasks in this direction is the selection of the carrier with optimal physicochemical parameters and morphology from a variety of plant sources. To date, vesicles from only a few plants were visualized using cryo-electron microscopy (cryo-EM). Here we investigated the morphology and physical parameters of extracellular vesicles from plant sources not previously studied utilizing this method.

Methods: PEVs derived by ultracentrifugation from juice and cultural medium of 11 plants and mushrooms were studied using methods of cryo-EM. Samples were plunge frozen in liquid ethane with Vitrobot Mark IV and examined under cryogenic transmission electron microscope Titan Krios 60-300 (ThermoFisher Scientific, USA) in low dose mode using EPU software.

Results: Most of the observed particles in each sample were classified as extracellular vesicles due to the presence of the lipid bilayer. Morphology and size characteristics of PEVs were determined and compared with each other. A variety of morphological configurations of PEVs were found: with single and multiple membranes, with different conformations and integrity state. Most of the isolated PEVs were single, round-shaped, and in a size range from 30 to 150 nm.

Conclusion: Cryo-EM allowed us to obtain high-quality images of PEVs isolated from 11 plants and mushrooms (blueberry, chanterelle, cowberry, fly agaric, garlic, redcurrant, *chlamydomonas*, cucumber, shadberry, viburnum, gooseberry) which have been characterized by their size and morphology. From the data obtained, the most promising sources of vesicles were proposed. The approbation of the selected vesicles as effective delivery systems requires further research.

Key Words: cryo-EM • extracellular vesicles • plant-derived extracellular vesicles

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