

Abstract

Extracellular MicroRNAs in Insects [†]

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[†] Presented at the 1st International Electronic Conference on Entomology (IECE 2021), 1–15 July 2021;

Available online: <https://iece.sciforum.net/>.

Citation: Van den Brande, S.; Remans, S.; Cordeiro dos Santos, D.; Broeck, J.V. Extracellular MicroRNAs in Insects, in Proceedings of the 1st International Electronic Conference on Entomology, 1–15 July 2021, MDPI: Basel, Switzerland, doi:10.3390/IECE-10538

Published: 1 July 2021

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In the last decade, extracellular RNAs have been unravelled as key players in cell-to-cell communication. In this respect, extracellular microRNAs have been thoroughly studied in mammalian systems; and RNA-based inter-cellular, inter-species and even inter-kingdom communication has been demonstrated. Nevertheless, studies reporting on extracellular (mi)RNAs in insects are scarce. Therefore, we have focused on unravelling the presence of ex-miRNAs in this group of animals. Specifically, we have demonstrated that ex-miRNAs are encapsulated in extracellular vesicles (EVs) or associated with Argonaute-1 in cell-free conditioned media of two *Drosophila* cell lines. In addition, we have found evidence for the selective secretion of miRNAs into EVs. Next, in order to start investigating these matters *in vivo*, we aimed at optimizing the isolation of EVs from hemolymph. EV purification from this complex biofluid is a challenging task, mainly due to the presence of contaminating (lipo-)proteins. We successfully developed a procedure to isolate EVs from locust hemolymph. Moreover, our results indicate that this approach can be adequate for EV isolation from a wide range of insect species. Given the current lack of standard and robust methods for EV isolation from insect hemolymph, our procedure constitutes an important step towards the advancement of this emerging research field. Our findings open new avenues to further research on insect extracellular RNAs, a so far underexplored but very fascinating topic.