

Extracellular microRNA 3' end modification across diverse body fluids

Kikuye Koyano ^a, Jae Hoon Bahn ^b, and Xinshu Xiao ^{a, b, c, d, e}

^a Bioinformatics Interdepartmental Program, UCLA, Los Angeles, California, USA ^b Department of Integrative Biology and Physiology, UCLA, Los Angeles, California, USA ^c Molecular Biology Institute, UCLA, Los Angeles, California, USA ^d Institute for Quantitative and Computational Sciences, UCLA, California, CA, USA ^e Jonsson Comprehensive Cancer Center, UCLA, California, CA, US

ABSTRACT


microRNAs (miRNAs) are small non-coding RNAs that play critical roles in gene regulation. The presence of miRNAs in extracellular biofluids is increasingly recognized. However, most previous characterization of extracellular miRNAs focused on their overall expression levels. Alternative sequence isoforms and modifications of miRNAs were rarely considered in the extracellular space. Here, we developed a highly accurate bioinformatic method, called miNTA, to identify 3' non-templated additions (NTAs) of miRNAs using small RNA-sequencing data. Using miNTA, we conducted an in-depth analysis of miRNA 3' NTA profiles in 1047 extracellular RNA-sequencing data sets of 4 types of biofluids. This analysis identified hundreds of miRNAs with 3' uridylation or adenylation, with the former being more prevalent. Among these miRNAs, up to 53% (22%) had an average 3' uridylation (adenylation) level of at least 10% in a specific biofluid. Strikingly, we found that 3' uridylation levels enabled segregation of different types of biofluids, more effectively than overall miRNA expression levels. This observation suggests that 3' NTA levels possess fluid-specific information relatively robust to batch effects. In addition, we observed that extracellular miRNAs with 3' uridylations are enriched in processes related to angiogenesis, apoptosis, and inflammatory response, and this type of modification may stabilize base-pairing between miRNAs and their target genes. Together, our study provides a comprehensive landscape of miRNA NTAs in human biofluids, which paves way for further biomarker discoveries. The insights generated in our work built a foundation for future functional, mechanistic, and translational discoveries.

ARTICLE HISTORY

Received 10 June 2020
Revised 3 August 2020
Accepted 21 August 2020

KEYWORDS

microRNA, non-templated additions, extracellular RNA, computational analysis, biofluids

CONTACT Xinshu Xiao  gxxiao@ucla.edu 610 Charles E. Young Drive S Terasaki Life Sciences Building, Room 2000E
Los Angeles, CA 90095-1570

© 2020 Informa UK Limited, trading as Taylor & Francis Group

Introduction

Recent studies revealed the existence of extracellular RNAs (exRNAs) in many types of biofluids [1,2]. exRNAs are mostly packaged in small extracellular vesicles, microvesicles or in complex with lipoproteins or ribonucleoproteins [3], which protect them from degradation by ribonucleases. exRNA expression could be highly cell type- or disease-specific [4,5], thus affording potential values as disease biomarkers [6]. Importantly, the functional roles of exRNAs are also starting to unfold [7–9]. For example, several studies reported the involvement of exRNAs in cell-to-cell communication in the local tumour microenvironment and during angiogenesis [7,10,11].

The most-often studied exRNAs are microRNAs (miRNAs), small 18–22nt noncoding RNAs that are potent regulators of mRNA and protein expression levels [12]. Most previous studies on extracellular miRNAs focused on interrogating their overall expression levels. Nonetheless, many miRNAs assume multiple sequence forms resulted from alternative miRNA processing or post-transcriptional modification [13]. Specifically, a well-known class of post-transcriptional modification of miRNAs is non-templated addition (NTA) [14]. Two types of 3' miRNA NTAs have been reported [15,16]. 3' adenylation catalysed by GLD2 and 3' uridylation by the terminal uridylyltransferase 4 and 7