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Review Article

IsomiRs: A New Approach in Cancer Study

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ABSTRACT

microRNAs are regulatory non-coding RNA molecules that containing about 18-24 nucleotides which involved in critical steps of many cellular processes, specifically gene expression, therefore, their deregulation can lead to human tumorigenesis. some kinds of variants of microRNAs are isomiRs that their length or sequence varies from miRNAs and they commonly a raised from diverse cleavage by the ribonucleases Drosha and Dicer. Recent reports confirm that some of isomiRs may be yielded from a miRNA locus, and these physiological mature isomers have important roles in miRNA evolution. In this research, we reviewed new field of miRNAs structural /sequence diversity that can be involved in cancer biology. Also describe the evolutionary approaches and the functional significance of the miRNAs isoforms; however, there are lots of isomiR/miRNA molecular mechanisms that remain unclear. More studies to reveal more insights of functionality isomiRs into the carcinogenesis will provide informative strategies to be used in prognosis, diagnosis or treatment.

Keywords: IsomiR, miRNA, Cancer

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In the past decade miRNAs regulatory role have been highlighted in physiological and pathological processes and indicated their misregulation lead to tumorigenesis. Genetic variations on the sequences of human miRNAs have impact on various phenotypes including cancers and other genetic diseases. One of the newest length variant in miRNAs so called isomiRs identified by Morin et al in 2008 in their cloning study. IsomiRs are structural variation in miRNAs which some of them have been found in cancers (1). These forms of heterogeneity in miRNAs sequence and length may affect the selecting target, miRNAs steadiness, or loading into the RNA-induced silencing complex (RISC) (2), miRNA half-life, subcellular localization (1). According to Cloonan and etal, isomiRs in biological and functional pathway such as gene targeting collaborate with canonical miRNAs (3) , therefore, suggested every changes in isomiRs proportions have functional effect on gene regulation (4). IsomiRs can be the extension of the miRNAs, which will contribute to flexible and powerful

References

1. Ryan BM, Robles AI, Harris CC. Genetic variation in microRNA networks: the implications for cancer research. Nature reviews Cancer. 2010 Jun;10(6):389-402. PubMed PMID: 20495573. Pubmed Central PMCID: 2950312.
2. Neilsen CT, Goodall GJ, Bracken CP. IsomiRs--the overlooked repertoire in the dynamic microRNAome. Trends in genetics : TIG. 2012 Nov;28(11):544-9. PubMed PMID: 22883467.
3. Cloonan N, Wani S, Xu Q, Gu J, Lea K, Heater S, et al. MicroRNAs and their isomiRs function cooperatively to target common biological pathways. Genome biology. 2011 Dec 30;12(12):R126. PubMed PMID: 22208850. Pubmed Central PMCID: 3334621.

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coding–non-coding RNA regulatory network (5). Research on isomiRs distribution on number of cancerous tissues such as breast, bladder, colon (6) and lung tissue (7) and also identification of isomiRs epidermal growth factor signaling pathways (8) showed different expression patterns of isomiRs in human cancer (7) even in cell types (8) that suggest the comparative abundance of isomiRs may be intrinsically different between normal and tumor tissues (6). In addition, research on human lymphocyte cell lines showed different expressions of isomiR depends on the type of population and gender (9).

Isomir origin

Initial investigations in solid cancer showed isomiRs can be stem from DNA variation like(SNPs and cancer-related mutations) or post-transcriptional modifications (6). Advanced research from NGS (next-generation sequencing) suggests isomiRs are multiple distinct mature miRNAs that arise from the same hairpin arm and 5' or 3' ends of their sequences and are different from canonical miRNAs in public databases such as miRBase (9). Moreover, recent studies have shown variation in cleavage site for DROSHA and DICER1 enzymes or even other miRNAs processing enzymes or post-transcriptional editing which lead to sequence modification in miRNAs (1) that are classified as "trimming variants" (10). Further evidences have shown there are another group of isomiRs that stem from Argonaute 2(AGO2) cleavage independent of Dicer. For example, the human miR-451 gene cleavages by Argonaut 2 (AGO2) to generate a molecule-intermediate called as 'ac-pre-miRNA' that are processed to mature miRNAs by exonucleolytic trimming (3). Data received from lymphoblastoid cell lines and analysis of Argonaute verify connection of many isomiRs with the Argonaute silencing complex and their important role in RNA interference pathway (9). There are three main types of isomiRs including 3′ isomiRs, 5' isomiRs and internal modifications (1) so called polymorphic isomiRs and 3′, 5' miR variants subclassified to 'templated' and 'non-templated' modification (figure 1). During the imprecise cutting by Dicer or Drosha and/or exonuclease activity, despite the fact that miRNAs and parent gene sequences length heterogeneity is created on referred to template.

References

4. Siddle KJ, Tailleux L, Deschamps M, Loh YH, Deluen C, Gicquel B, et al. bacterial infection drives the expression dynamics of microRNAs and their isomiRs. PLoS genetics. 2015 Mar;11(3):e1005064. PubMed PMID: 25793259. Pubmed Central PMCID: 4368565.
5. Wu X, Zeng R, Wu S, Zhong J, Yang L, Xu J. Comprehensive expression analysis of miRNA in breast cancer at the miRNA and isomiR levels. Gene. 2015 Feb 25;557(2):195-200. PubMed PMID: 25523096.
6. Meiri E, Levy A, Benjamin H, Ben-David M, Cohen L, Dov A, et al. Discovery of microRNAs and other small RNAs in solid tumors. Nucleic acids research. 2010 Oct;38(18):6234-46. PubMed PMID: 20483914. Pubmed Central PMCID: 2952848.
7. Li SC, Liao YL, Ho MR, Tsai KW, Lai CH, Lin WC. miRNA arm selection and isomiR distribution in gastric cancer. BMC genomics. 2012;13 Suppl 1:S13. PubMed PMID: 22369582. Pubmed Central PMCID: 3303722.
8. Llorens F, Hummel M, Pantano L, Pastor X, Vivancos A, Castillo E, et al. Microarray and deep sequencing cross-platform analysis of the mirRNome and isomiR variation in response to epidermal growth factor. BMC genomics. 2013 Jun 01;14:371. PubMed PMID: 23724959. Pubmed Central PMCID: 3680220.
9. Loher P, Londin ER, Rigoutsos I. IsomiR expression profiles in human lymphoblastoid cell lines exhibit population and gender dependencies. Oncotarget. 2014 Sep 30;5(18):8790-802. PubMed PMID: 25229428. Pubmed Central PMCID: 4226722.
10. Llorens F, Banez-Coronel M, Pantano L, del Rio JA, Ferrer I, Estivill X, et al. A highly expressed miR-101 isomiR is a functional silencing small RNA. BMC genomics. 2013 Feb 15;14:104. PubMed PMID: 23414127. Pubmed Central PMCID: 3751341.

CANCER PRESS
Vol. 3, No. 2, June, 2017

In non-template, in addition to the length heterogenity, sequences may not match with parent gene due to post-transcriptional base addition (2).

Three main forms of isomiR variations are described as follows: 3′ deletion/addition(3' modifications),5'deletion/addition (5' modifications), internal modifications: 3' modifications: The most common sequence variation of mature miRNA is single nucleotid extention in 3' end (1). For example, nucleotidyl transferases that are 5'–3' polymerases by adding nucleotides causing more "nontemplated" nucleotide extensions at 3' compared to 5'. Seven numbers of Nucleotidyl transferases involved in isomiR biogenesis have Adenyl transferase and Uridyl transferase function (2) which suggest the most prevalent nucleotides added are adenine and uridine (1). In this regard, Meiri et.al's research on the 4 types of cancer have been detected 3' uridylation of hsa-miR-143 and 3' adenylation of hsa -miR-100. However, isomiRs caused by C or G 3' addition have not been observed (6). Probably part of the nucleotide changes occur due to deamination of cytosine to uracil by cytidine deaminases or deamination of an adenosine to inosine by adenosine deaminases (1). Some 3'–5' exoribonucleases which generate "templated" 3' isomir are Nibbler and QIP reported in Drosophila and Neurospora crassa, respectively (2).

New research indicated that viral (11) and bacterial infections and also their cellular responses by changing cellular process and involving in post-transcriptional modifications like 3’ uridylation / adenylation and nucleolytic trimming regulate miRNAs expression and isomiRs generation (4).

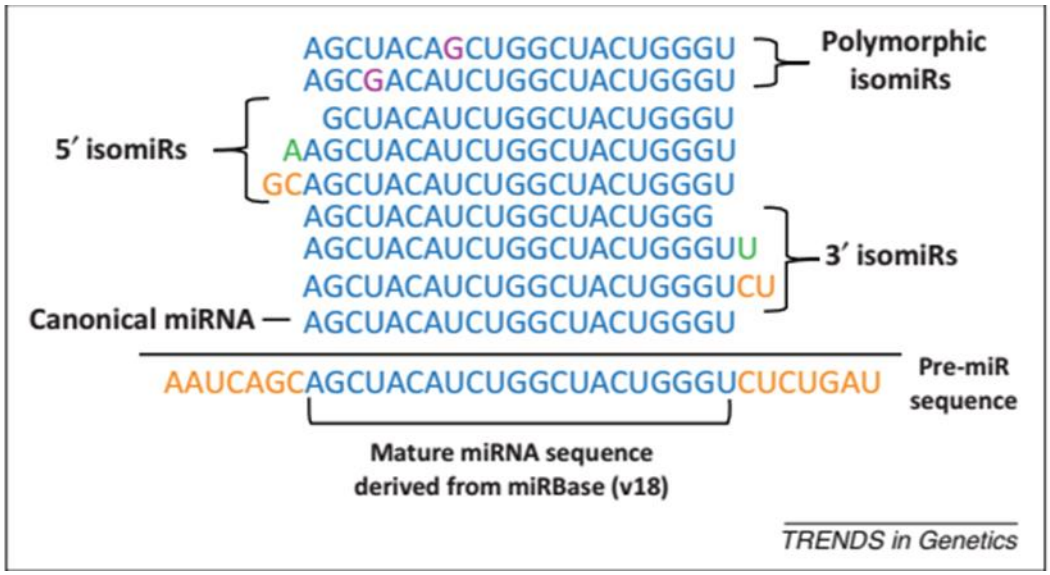

Figure1: Example of various isomiRs species (2)

5' modifications: despite of this notion that pre-miRNA processing leads to a mature miRNA sequence with a constant nucleotide at the 5′ end, recent experiment suggest that isomiRs may also result from alteration at the 5′end. Although the isomiRs arising from 5' variation of mature miRNAs are generally rarer, having more interest because possess distinct seed sequence rather than reference miRNA and then prone to bind different target transcripts (1). Seed-shifting is an example of editorial changes at the 5' end of the miRNA that generated by nucleotide extension, deletion, or

CANCER PRESS
Vol. 3, No. 2, June, 2017

variable cleavage (2)(figure 2). This kind of variant in seed region (nucleotides 2–8 at the 5' end of the miRNA responsible for binding to targets) in addition to resulting in novel target repertoire (12), involved in determining miRNA/miRNA* strand for connecting with AGO proteins and subsequently RISC complex activation (2).Variation of seed shifting in some miRNAs including 5' isomiR-142 (13), 5'isomiR-133a (14) sequences lead to finding different gene target (2). There also have been reported the bacterial infection leading to seed shifting process in miR-191-3p and miR-342-5p (4).

Internal modification: or polymorphic isomiRs contain different internal combinations compared with mature canonical miRNA sequences (2). Previous studies have shown appearance of the internal variations which arise from some specific mutation such as internal insertion, deletions and substitutions in murine let-7a (15). In addition to, research conducted on 20% primary and precursor miRNA transcripts subjected by A-to-I editing suggest that the process is important driver in the production of polymorphic isomiRs. For example the single nucleoid variations arising from RNA editing on mature miR-376 by modifying the 'seed region' change repertoire of targets (16). Another example of the occurrence of internal editing in miRNAs is hsa-miR-1274b (6). Probably internal modification in nucleotide sequence can increase gene targets and also by changing the degree of complementarity enhance mRNA decay over translational repression (15).

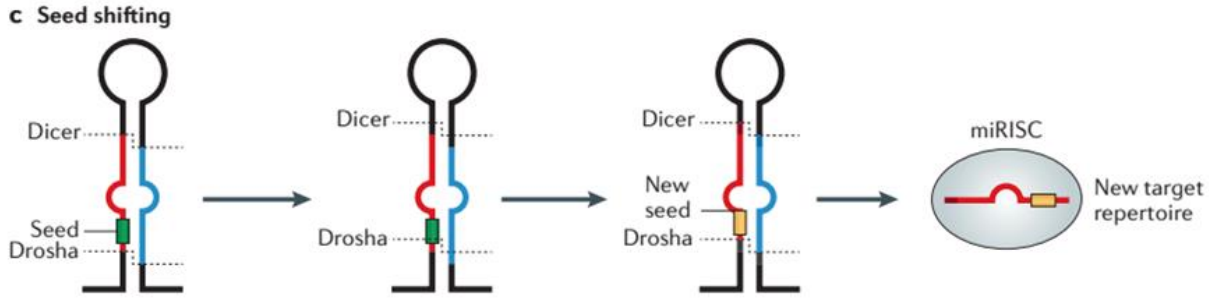

Figure 2: seed shifting changed 5′ end of the mature miRNAs, resulting in novel target set (12)

References

11. Backes S, Shapiro JS, Sabin LR, Pham AM, Reyes I, Moss B, et al. Degradation of host microRNAs by poxvirus poly(A) polymerase reveals terminal RNA methylation as a protective antiviral mechanism. Cell host & microbe. 2012 Aug 16;12(2):200-10. PubMed PMID: 22901540. Pubmed Central PMCID: 3782087.
12. Berezikov E. Evolution of microRNA diversity and regulation in animals. Nature reviews Genetics. 2011 Nov 18;12 (12):846-60. PubMed PMID: 22094948.
13. Azuma-Mukai A, Oguri H, Mituyama T, Qian ZR, Asai K, Siomi H, et al. Characterization of endogenous human Argonautes and their miRNA partners in RNA silencing. Proceedings of the National Academy of Sciences of the United States of America. 2008 Jun 10;105(23):7964-9. PubMed PMID: 18524951. PubMed Central PMCID: 2430345.
14. Humphreys DT, Hynes CJ, Patel HR, Wei GH, Cannon L, Fatkin D, et al. Complexity of murine cardiomyocyte miRNA biogenesis, sequence variant expression and function. PloS one. 2012;7(2):e30933. PubMed PMID: 22319597. Pubmed Central PMCID: 3272019.
15. Reid JG, Nagaraja AK, Lynn FC, Drabek RB, Muzny DM, Shaw CA, et al. Mouse let-7 miRNA populations exhibit RNA editing that is constrained in the 5'-seed/ cleavage/anchor regions and stabilize predicted mmu-let-7a:mRNA duplexes. Genome research. 2008 Oct;18(10):1571-81. PubMed PMID: 18614752. Pubmed Central PMCID: 2556275.

The functional significance of isomiRs in cancer: As mention above the most of isomiRs are made through variations in 3' end of miRNAs, so, further isomiRs may possess same 5' end and same seed sequences compared to canonical miRNAs. The results shown in expression analysis of Iso/miR level in breast cancer confirm cooperative function between isomiRs and miRNAs in regulation of similar target genes and biological pathways (5). Meiri and colleagues in their comprehensive study have indicated relative frequency of isomiRs in tumor tissues were much more than normal tissue and suggested that these sequence variants of miRNAs could be connected to tumorigenesis or derived from other genetic variation such as single nucleotide polymorphism(SNP) and mutations (6). Some researches evaluated isomiRs role in cancers like breast (5),(17) melanoma (18) and stomach (7) which indicated biological roles of isomiRs in the miRNA–mRNA regulatory networks. A lot of these miRNA structural variants were down regulated in the tumor cells (1) which suggest tumor suppressor activity for isomiRs. In this regard, Salem and et.al have found evidence of tumor suppressor role of 5'isomiR-140-3p and its functional synergy by canonical hsa-miR-140-3p in inhibition of development of breast tumor via targeting genes associated with differentiation, proliferation, and migration (17). Moreover, the isoform of miR-451a called miR-451a.1 leads to reducing migration and suppressing cell invasion in melanoma cell lines (18).

Conclusion

This review has focused on isomiRs as a genetic sequence /structural variations occurred in miRNAs, their types, and the ways of biogenesis. New form of genetic variations simultaneously developing miRNAs related to cancer susceptibility have emerged that is deserving further investigation. Due to delicate and complex collaboration of isomiRs and miRNAs in regulation of various cellular processes such as cancer, better understanding of miRNA biology particularly in cancer, isomiRs unique properties and determining association between isomiRs and specific cancer phenotype will be important. In fact, according to the recent report of functional regulatory role and biological significance of isomiRs, their comprehensive study are an important step towards an identification more characterization of the canonical miRNAs in cancer cell /tissue and could lead to growing new therapeutic targets like more specific therapy for targeting cancer cells.

References

16. Kawahara Y, Zinshteyn B, Sethupathy P, Iizasa H, Hatzigeorgiou AG, Nishikura K. Redirection of silencing targets by adenosine-to-inosine editing of miRNAs. Science. 2007 Feb 23;315(5815):1137-40. PubMed PMID: 17322061. Pubmed Central PMCID: 2953418.

17. Salem O, Erdem N, Jung J, Munstermann E, Worner A, Wilhelm H, et al. The highly expressed 5'isomiR of hsa-miR-140-3p contributes to the tumor-suppressive effects of miR-140 by reducing breast cancer proliferation and migration. BMC genomics. 2016 Aug 08;17:566. PubMed PMID: 27502506. PubMed Central PMCID: 4977694.

18. Babapoor S, Fleming E, Wu R, Dadras SS. A novel miR-451a isomiR, associated with amelanotypic phenotype, acts as a tumor suppressor in melanoma by retarding cell migration and invasion. PloS one. 2014;9(9):e107502. PubMed PMID: 25237911. Pubmed Central PMCID: 4169564.