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Citation Classic

MicroRNAs Regulate Expression of Oncogenes

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Featured Article: Johnson SM, Grosshans H, Shingara J, Byrom M, Jarvis R, Cheng A, Labourier E, et al. RAS is regulated by the let-7 microRNA family. Cell 2005;120:635–47.[2]

Today, we fully appreciate that microRNAs (miRNAs)[3] represent a paradigm shift in our understanding of gene expression and disease, but it was not always this way. miRNAs are small noncoding RNAs belonging to a novel class of regulatory molecules in plants and animals that control the expression of hundreds of target mRNA transcripts. Thousands of known miRNAs regulate gene expression by binding to imperfect complementary sites within the 3′ untranslated regions (UTRs) of their target protein–coding mRNAs and repressing the expression of these genes at the level of mRNA stability and translation *(1)*. Recent progress has revealed that miRNAs are involved in the regulation of most biological functions, including development, life span, and metabolism, and their dysregulation contributes, not surprisingly, to many types of disease, notably cancer *(2)*. For example, certain miRNAs regulate processes important for cell growth, division, differentiation, survival, and migration—all processes that go awry in cancer. In addition, several key miRNA genes known to be amplified, deleted, or misexpressed in cancer *(3)* act in pathways important for cancer progression and metastasis. We now know that miRNAs in fact act as oncogenes and tumor suppressors.

let-7 was the first known human miRNA *(4)*. In 2005, when our *Cell* report was published, it was known that let-7 was required for the normal cell differentiation of stem cells in the model organism *Caenorhabditis elegans (5)* and that multiple human genes encoding let-7 family members mapped to regions of the genome with known deletions in cancer, especially lung and breast cancer *(3)*. This finding suggested that let-7 might be a tumor suppressor, but there was no mechanism of action for this miRNA (or any other) in cancer.

The results published in our *Cell* report, in collaboration with David Brown's group at Ambion, showed for the first time that miRNAs could regulate cellular oncogene mRNAs. We showed initially in *C. elegans* and subsequently in human cells that let-7 regulates the *RAS* oncogenes through complementary sites in their RNAs' 3′ UTRs. In *C. elegans*, we showed genetically that loss of *RAS* suppressed *let-7*[4] mutant phenotypes in the skin and vulval stem cells. The *RAS* 3′ UTR contained multiple let-7–complementary sites and restricted reporter gene expression in a let-7–dependent manner. We found that miR-84, a let-7 family member, was largely absent in the primitive primary vulval precursor cell at the time that *RAS* specified the primary vulval fate in that cell, and that miR-84 overproduction suppressed the multivulva phenotype of activating *RAS* mutations. Remarkably, the 3′ UTRs of the 3 human *RAS* genes [*HRAS* (v-Ha-ras Harvey rat sarcoma viral oncogene homolog), *KRAS* (v-Ki-ras2 Kirsten rat sarcoma viral oncogene homolog), and *NRAS* (neuroblastoma RAS viral (v-ras) oncogene homolog)] contained multiple let-7–complementary sites, allowing let-7 to regulate *RAS* expression. We also showed that let-7 was produced at low concentrations in lung tumor samples relative to those of normal lung tissue from the same patient, whereas the concentration of RAS protein was significantly higher in lung tumors. These results provided a likely mechanism for let-7 to function as a tumor suppressor in cancer via negative regulation of *RAS* oncogenes.

Subsequent work revealed that let-7 regulates a number of other oncogenes, including *CCND2* (cyclin D2), *CDK6* (cyclin-dependent kinase 6), *CDC25* [cell division cycle 25 homolog C (*S. pombe*)], and *MYC* [v-myc myelocytomatosis viral oncogene homolog (avian)], and the gene encoding let-7 is expressed at low levels in multiple different cancers, such as breast cancers and in breast cancer stem cells. Its misexpression patterns have also been shown to act as biomarkers of cancer outcome.

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[2] This article has been cited more than 1370 times since publication.
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[3] Nonstandard abbreviations: miRNA, microRNA; UTR, untranslated region; pre-miR, miRNA mimic used to increase miRNA function; antimiR, antisense molecule designed to inhibit miRNA function.

[4] Genes: *let-7*; *HRAS*, v-Ha-ras Harvey rat sarcoma viral oncogene homolog; *KRAS*, v-Ki-ras2 Kirsten rat sarcoma viral oncogene homolog; *NRAS*, neuroblastoma RAS viral (v-ras) oncogene homolog; *CCND2*, cyclin D2; *CDK6*, cyclin-dependent kinase 6; *CDC25*, cell division cycle 25 homolog C (*S. pombe*); *MYC*, v-myc myelocytomatosis viral oncogene homolog (avian).

I believe this publication is highly cited for several reasons. First, it introduced a number of useful tools for miRNA studies, including premiRs (miRNA mimics used to increase miRNA function) and antimiRs (antisense molecules designed to inhibit miRNA function). Second, it popularized the use of 3′ UTR reporter gene fusions for studying miRNA activity at 3′ UTRs. Third, and most importantly, it showed for the first time that miRNAs regulate important cancer genes and fit within cancer pathways. Consequently, this publication was partly responsible for launching a new field of cancer biology and cancer therapeutics that has culminated in the use of let-7 as a novel, natural anticancer agent *(6)*.

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