Commentary

# Moving miRNAs to therapeutic targets in colorectal cancer

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Understanding the prognosis and the search for additional targets for advanced colorectal cancer remains an important focus of research. MicroRNAs (miRNAs) are short, non-coding RNA molecules that act as regulators of post-transcription gene expression. Their dysregulation has been associated with the diagnosis and altered prognosis of a number of malignancies, including colorectal cancer (CRC). Furthermore, circulating blood miRNAs are shielded from endogenous ribonucleases providing stability thus making them potential cancer biomarkers.

MiR-338-5p, a miRNA on chromosome 17, has been shown to be up-regulated in metastatic CRC tumours. It was one of 3 miRNA's found to be measurable in circulating blood for early detection of CRC, with good correlation between tissue and serum expression and high sensitivity and specificity for distinguishing colorectal cancer over those with benign polyps or healthy controls [9]. Interestingly, increased expression was found in more advanced early stage disease, indicating this may also be a measurable option in the prognostic assessment of advanced CRC. Other authors also assessed miR-338-5p as a diagnostic marker using CEA as the comparator, reporting a greater ability of serum miR-338-5p to differentiate those with CRC from non-CRC controls [1]. These findings potentially offer additional, less invasive screening modalities to detect colorectal cancer.

Chu and colleagues have further explored the role of miR-338-5p in the pathogenesis and behaviour of CRC in this edition of EBioMedicine [3]. They have built on their earlier work, first confirming that phosphatidylinositol-3-kinase catalytic subunit type 3 (PIK3C3) was the target gene for miR-338-5p and that over-expression of miR-338-5p and increased miR-338-5p/PIK3C3 ratios in tumour tissue were associated with more advanced stage and poorer prognosis in those with colorectal cancer, suggesting that this ratio could become a prognostic biomarker for CRC patients. The mechanism for this was found to be induction of cancer cell migration and invasion, key features of advanced disease, through inhibition of the PIK3C3-related autophagy pathway.

These results supported a number of prior studies identifying the association of miR-338-5p over-expression with CRC and more specifically its role in inhibiting PIK3C3, a known autophagy promoter. The authors suggest that potentially inducing autophagy could be a valid treatment strategy for CRC. However, there is widely conflicting prior evidence as to whether autophagy functions as a tumour suppressor or promoter.

The true role of autophagy- the mechanism of packaging, degradation and recycling of cytosolic components- and its interplay with cancer is not fully understood. Current evidence suggests the effect of autophagy is likely to be both tumour-dependent and context dependent.

As summarized in a review by Wilde et al. [6], genetic mutations causing deletion of autophagy proteins, such as Beclin-1, have been noted in many solid organ cancers including CRC, whilst knockout-mice deficient in autophagy genes have had tumour development from accumulated reactive oxygen species in defective mitochondria. Induction of autophagy has been successful in a number of cancers with the use of mTOR inhibitors, whilst arsenic trioxide, another autophagy inducer, is now a standard of care treatment for acute promyelocytic leukaemia, at least in part supporting a potential therapeutic role via this mechanism.

The opposite impact of autophagy has been demonstrated in a number of settings however, suggesting a need for caution when considering autophagy induction as a treatment. Multiple cancers have been shown to be reliant on autophagy for growth and proliferation and it can contribute to treatment resistance, particularly in those with RAS mutations [6]. Chloroquine and hydroxychloroquine are known to directly inhibit autophagy, with their use in colon cancer cell lines potentiating the effect of fluorouracil, in contrast to the findings of Chu and colleagues [5]. A further study inhibiting autophagy in colon cancer, through a PIK3C3 independent mechanism, also successfully inhibited colon cancer progression [10].

Evidence for autophagy promoting malignant progression is also demonstrated by its impact on cancer stem cells, allowing continued self-renewal and promoting treatment resistance, as well as its impact on stromal fibroblasts in the tumour microenvironment (TME) where upregulation of autophagy indirectly promotes proliferation of adjacent cancer cells through aerobic glycolysis [6]. Inhibiting autophagy in the TME of CRC through inhibition of miR-31 has resulted in decreased tumour cell proliferation [8].

Given the conflicting evidence thus far, it is unclear if targeting autophagy is a valid and reproducible treatment strategy for CRC. It is noteworthy even in this study, re-activating autophagy did not completely reverse the miR-338-5p-induced cancer cell invasion and migration, suggesting other potential pathways that could limit the effectiveness of targeting autophagy alone. Other difficulties include uncertainty as to how individual drugs will affect autophagy in different tissues, as evidenced by older studies for an investigational drug that successfully inhibited PIK3C3-induced autophagy in glioma, despite

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being found to induce autophagy through alternate pathways in earlier studies [2,7]. Given the ongoing uncertainty, studies combining an autophagy inducer with inhibitor have also been performed, with a phase 1 trial demonstrating safety despite it being a somewhat counter-intuitive approach [4].

Studies such as that reported by Chu et al. could identify when miRNA's may be appropriate biomarkers used to guide targeted treatment, eg using autophagy inducers in cancers with high miR-338-5p. However further research is necessary to allow a greater understanding of the interplay between autophagy, cancer cells and the TME, guiding future decisions on whether inhibiting or inducing autophagy will be the correct approach. Targeting miRNA's directly may be a better alternative to ensure treatment of all cancer promoting pathways, other than PIK3C3-induced autophagy, that may be yet unknown. Despite its uncertainty as a therapeutic target, the use of serum miR-338-5p as a prognostic or diagnostic biomarker for colorectal cancer has promise for use in the near future.

## Disclosure

The authors declared no conflicts of interest.

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