Editorial

# Serum microRNA guiding personalized radiation therapy in non-small cell lung cancer

**Neha P. Amin[1], Pranshu Mohindra[1], Salma K. Jabbour[2]**

[1]Department of Radiation Oncology, University of Maryland School of Medicine, Baltimore, MD, USA; [2]Department of Radiation Oncology, Rutgers Cancer Institute of New Jersey, Rutgers Robert Wood Johnson Medical School, Rutgers University, New Brunswick, NJ, USA

*Correspondence to:* Neha P. Amin. Department of Radiation Oncology, 500 Upper Chesapeake Drive, Bel Air, MD 21014, USA.
Email: npamin@gmail.com.

*Provenance:* This is an invited Editorial commissioned by the Executive Editor-in-Chief Jianxing He (Department of Cardiothoracic Surgery, The First Affiliated Hospital of Guangzhou Medical University, Guangzhou, China).

*Comment on:* Sun Y, Hawkins PG, Bi N, *et al.* Serum MicroRNA Signature Predicts Response to High-Dose Radiation Therapy in Locally Advanced Non-Small Cell Lung Cancer. Int J Radiat Oncol Biol Phys 2018;100:107-14.

Submitted Sep 21, 2018. Accepted for publication Sep 26, 2018.
doi: 10.21037/jtd.2018.09.143
**View this article at:** http://dx.doi.org/10.21037/jtd.2018.09.143

Lung cancer is the most commonly diagnosed cancer and leading cause of cancer death worldwide, with an estimated 2,093,876 new lung cancers and 1,761,007 deaths from lung cancer in 2018 (1). Out of the 85% of lung cancers that are non-small cell lung cancer (NSCLC), approximately 30% of patients will present with unresectable Stage III disease for which combination chemotherapy and radiation therapy (RT) is the recommended treatment (2,3). Despite advancements in RT technology that have improved accuracy and therapeutic index of RT (4-6), local recurrence in NSCLC is still a key problem fueling research interests in RT dose escalation approaches (7-9). Even though higher RT doses have resulted in better tumor control when delivering stereotactic body radiation therapy (SBRT) using 48–55 Gy in 3–5 fractions to Stage I–II lung tumors, when conventionally fractionated 60 *vs.* 74 Gy for locally advanced NSCLC patients was studied in a large phase III dose-escalation study (RTOG 0617), patients in the 74 Gy arm had worse outcomes (8). Due to the heterogeneity of NSCLC, it is very possible that there may be a subset of tumors that would benefit from either dose-escalation or dose de-escalation. If it were possible to identify certain NSCLC tumors or patients who would benefit from more specific RT doses, then we could better customize RT plans to provide optimal tumor control while minimizing toxicities. This type of biological precision would elevate radiation oncology into a new dimension of personalized medicine.

NSCLC's are a highly heterogeneous group of cancers for which targeted therapies for mutations of epidermal growth factor receptor (*EGFR*) and the translocation of anaplastic lymphoma kinase (*ALK*) have shown promise. Other frequently mutated genes in NSCLC tumors include BRAF, KRAS, and TP53 (10). In contrast to systemic therapy delivery for lung cancer that often is based on driver mutational status, radiation oncology has not yet incorporated personalized knowledge of tumor genomics or molecular signatures into RT administration. Instead, the historical knowledge of the tumor type's radiosensitivity, adjacent normal tissue tolerances, and the results from clinical trials have resulted in a recommended standard RT dose of ≥60 Gy at 1.8–2 Gy per fraction for NSCLC regardless of the tumor's unique genetic profile. Torres-Roca and team have developed a tumor gene signature-focused method of personalizing radiation dose by utilizing the tumors radiosensitivity index (RSI) and genomically adjusted radiation dose (GARD), but it is not yet ready for clinical implementation (11-15).

Another area of active research is serum circulating microRNAs (c-miRNAs), a non-coding RNA that regulates gene expression. MiRNA's may serve as biomarkers for tumor detection and prognosis, have a broad spectrum of effects including tumor suppression and proliferation, and have a distinct profile in different tumors and normal tissues (16). Biomarkers play a vital role in diagnosis and prognosis of many cancers, and serum miRNA's have been

recognized as markers of early lung cancer detection (17,18). A study that analyzed 904 serum miRNAs at pre- and post-lung cancer diagnosis time points observed changes in a specific c-miRNA signature during tumor development, and proposed that lung cancer may be detectable years prior to diagnosis (19). What if c-miRNAs could also be used as a serum biomarker that could identify patients who would benefit from higher doses of RT?

In "Serum MicroRNA Signature Predicts Response to High-Dose Radiation Therapy in Locally Advanced Non-Small Cell Lung Cancer (NSCLC)" by Sun *et al.*, the authors sought to determine if c-miRNAs could be used as a biomarker to predict outcomes with high-dose RT for NSCLC. This study was a novel endeavor as there are currently no clinically utilized serum biomarkers to differentiate NSCLC radiation sensitivity. Pre-treatment serum from 80 patients, who were prospectively treated from 2004 to 2013 on 4 different dose-escalation clinical trials for Stage II-III NSCLC using three-dimensional conformal RT to deliver 66–86 Gy in 30–37 fractions of ≥2 Gy per fraction, were analyzed with a panel of 62 c-miRNAs out of which 11 miRNAs were identified as dose-effect modifiers. A model to calculate the dose-response score (DRS) to predict for overall survival (OS) after high- *vs.* standard-dose RT incorporated the 11 miRNAs (identified as dose-effect modifiers), chemotherapy, RT dose, normal tissue toxicities, and patient characteristics to describe a c-miRNA signature that could identify a subset of NSCLC patients who may benefit from RT dose-escalation. Patients with low and high DRS (split at median) were further stratified into 2 subgroups of high- *vs.* standard RT dose using a cutoff median biological effective dose (BED) of 87.1 $Gy_{10}$. For reference, using a tumor $\alpha/\beta$ ratio =10, BED of a typical definitive dose of 60, 66, 70 and 74 Gy is equivalent to 72, 79.2, 84 and 88.8 $Gy_{10}$, respectively (all in 2 Gy fractions). While rates of OS in patients with high-DRS were not affected by RT dose, patients with low-DRS benefited from high-dose RT compared to standard dose, resulting in improved OS and lower risk of distant metastasis. The DRS also predicted RT dose effect on local control (LC), but the relationship was not statistically significant. The authors concluded that the DRS model they developed could identify a subset of NSCLC patients who could obtain an OS benefit from high-dose RT, but the model would need external validation prior to implementing it clinically (20).

While this study's approach and conclusions are original and hypothesis-generating, there are some limitations pertaining to the sample size, type of sample, and choice of the miRNA panel that should be considered before drawing conclusions. There may be a possible selection bias in the patients that were included in the dose escalation trials, which could skew the results of this study. For example, patients with smaller tumors and less lymph node involvement are often easier to dose-escalate safely and may have made up the majority of the trial patients. These less advanced tumors may also have a distinct genetic profile that could predict favorable response to systemic therapy or RT. Since there was no validation cohort and only 80 (46%) serum samples could be evaluated out of the total 173 NSCLC patients treated on the 4 RT dose escalation studies, there may not be enough samples to account for the potential variations in the tumors genetic profiles, treatment variables, and patient's characteristics in this study, thus making it difficult to validate results or generalize from this study.

There are also standard inherent weaknesses when retrospectively analyzing specimens, including the integrity of the serum sample and possible loss of c-miRNA material during the freezing, isolation, or RNA quantification processes. Additionally, serum provides a less invasive sample to use for miRNA testing, however serum c-miRNA profiles may not correlate to the tumor's miRNA profile (21). Thus serum c-miRNA may not be as helpful when trying to determine if a tumor would be more responsive to higher RT doses since the serum may not be circulating tumor-specific miRNA in high enough quantity to be detected, and c-miRNA may not capture changes in the tumor microenvironment that play a more direct role in RT-induced tumor kill mechanisms.

Another limitation is the choice of miRNA panel for this study. While over 2,000 human miRNAs have been identified (16), only 62 (~3%) miRNAs were evaluated in this study. These specific 62 miRNA were previously studied in melanoma and zinc depletion, not NSCLC. During the time period of this study's miRNA panel analysis, there had already been published miRNA studies in NSCLC. A study published in 2010 in the Journal of Clinical Oncology investigated the role of serum miRNA in predicting prognosis of NSCLC patients and identified a four-miRNA signature (miR-486, miR-30d, miR-1 and miR-499) that was an independent predictor of OS in their cohort of patients (22). None of these 4 miRNAs were included in the 62-miRNA panelevaluated in Sun *et al.*'s study. Another study identified miR-374a as a prognostic marker for NSCLC progression (23); miR-374a was included within the 62-miRNA panel but

 

not found to be correlated to dose-response. While ideally future studies on the relationship between c-miRNA and RT dose-escalation would have the funding to utilize big data resources (24) and test a comprehensive panel of serum miRNAs, an obvious limitation will be the lack of samples from patients who received RT dose-escalation. Thus, while there were only 80 samples in this study, it is likely one of the largest sources of data available for answering questions about dose escalation in conventionally fractionated RT treatments for locally advanced NSCLC.

There are also concerns about the data cut-offs and endpoints used in the DRS model that could have influenced the final analysis. By selecting a cut-off at BED 87.1 $Gy_{10}$ between high- and standard-RT doses, the study considered 70 Gy as standard RT dose, which though acceptable per the National Comprehensive Cancer Network (NCCN) guidelines (3), would be considered as high dose by most radiation oncologists. Could the analysis be different if the dose was distributed into three strata: 60, >60 to ≤70 and >70 Gy? The authors recognize the impact of this dose cut-off choice on generalizability of their findings. The choice of OS as the endpoint could have also influenced the final analysis. The goal of using higher RT doses is to improve LC, which would hopefully translate to fewer distant metastases and improved OS. With this in mind, it is unclear if the model outcomes would have been different if LC was chosen as the DRS end-point rather than OS. In fact, while the use of high-dose RT in patients with low DRS was statistically significant in predicting lower risk of distant metastases, it was not statistically significant when predicting dose effect on LC. It is possible that the higher RT doses killed off the cells that would be more likely to metastasize, but larger sample sizes would be needed to validate this hypothesis.

The DRS was estimated by using a variety of clinical factors and 11 miRNA measurements as potential dose modifiers for the hazard ratio of death. In the absence of mechanistic information regarding the role of the selected miRNAs, a clear explanation for this effect is difficult to ascertain. The authors admitted that it was beyond the scope of this study to determine the mechanisms of the 11 c-miRNAs and how they were involved with the improved outcomes after high-dose RT. Of the 11 miRNAs identified in their final subset, many are associated with tumor suppressor activities while others may promote tumorigenesis and progression. There are inherent challenges with correlating miRNA with outcomes since knowledge regarding miRNAs are still evolving.

The association of miRNAs with OS and distant metastases without significantly impacting dose response effect on LC may open a window of opportunity to select patients who might benefit from trimodality therapy, which even in the subset of locally advanced NSCLC disease has been linked to high OS (25). In single institutional experiences, while mediastinal nodal clearance after neoadjuvant chemoradiation was predictive of improved survival, a pathological complete response at the primary site in patients who achieved mediastinal nodal clearance did not further improve outcomes (26). Such patients with low DRS and at lower risk of distant metastases may be better served with neoadjuvant chemoradiation followed by surgical resection of primary disease, thereby reducing loco-regional recurrence risks, based on the results of the Sun *et al.* study.

Despite the limitations in this current study, examining c-miRNA is an important step towards personalizing RT and oncological care. To improve outcomes in the subset of patients who fail standard doses of RT for NSCLC, we need to understand differences in radiosensitivity based on genetic differences between tumors of the same histology. While the role of c-miRNA in determining RT dose is in its nascent stages and serum c-miRNA may not be specific to the tumor, it does open a window to the milleau of other processes going on in the body that may affect outcomes. Future studies would ideally include a serum component along with tumor samples for genetic profiling using big data platforms so that stronger correlations could be made between peripheral blood and tumor samples. Progress may be limited by lack of funding or results that show worse or marginal differences in outcomes. This hypotheses-generating work on c-miRNA predicting response to RT-dose escalation in NSCLC could influence how we determine RT dose for NSCLC patients, however; additional data validating these results will be necessary before changing practice. We continue to rely on methodical study of locally advanced NSCLC to guide future treatment paradigms to continue to increase cure rates.

## Acknowledgements

None.

## Footnote

*Conflicts of Interest:* The authors have no conflicts of interest to declare.

 

## References


1. Bray F, Ferlay J, Soerjomataram I, et al. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 2018. [Epub ahead of print].
2. Morgensztern D, Ng SH, Gao F, et al. Trends in stage distribution for patients with non-small cell lung cancer: A national cancer database survey. J Thorac Oncol 2010;5:29-33.
3. National Comprehensive Cancer Network. Non-Small Cell Lung Cancer (Version 5.2018). Available online: https://www.nccn.org/professionals/physician_gls/PDF/nscl.pdf. Accessed June 30, 2018.
4. Yegya-Raman N, Zou W, Nie K, et al. Advanced radiation techniques for locally advanced non-small cell lung cancer: Intensity-modulated radiation therapy and proton therapy. J Thorac Dis 2018;10:S2474-91.
5. Vyfhuis MAL, Onyeuku N, Diwanji T, et al. Advances in proton therapy in lung cancer. Ther Adv Respir Dis 2018;12:1753466618783878.
6. Molitoris JK, Diwanji T, Snider JW 3rd, et al. Advances in the use of motion management and image guidance in radiation therapy treatment for lung cancer. J Thorac Dis 2018;10:S2437-50.
7. Cannon DM, Mehta MP, Adkison JB, et al. Dose-limiting toxicity after hypofractionated dose-escalated radiotherapy in non-small-cell lung cancer. J Clin Oncol 2013;31:4343-8.
8. Bradley JD, Paulus R, Komaki R, et al. Standard-dose versus high-dose conformal radiotherapy with concurrent and consolidation carboplatin plus paclitaxel with or without cetuximab for patients with stage IIIA or IIIB non-small-cell lung cancer (RTOG 0617): A randomised, two-by-two factorial phase 3 study. Lancet Oncol 2015;16:187-99.
9. Urbanic JJ, Wang X, Bogart JA, et al. Phase 1 Study of Accelerated Hypofractionated Radiation Therapy With Concurrent Chemotherapy for Stage III Non-Small Cell Lung Cancer: CALGB 31102 (Alliance). Int J Radiat Oncol Biol Phys 2018;101:177-85.
10. Chang JT, Lee YM, Huang RS. The impact of the cancer genome atlas on lung cancer. Transl Res 2015;166:568-85.
11. Torres-Roca JF. A molecular assay of tumor radiosensitivity: a roadmap towards biology-based personalized radiation therapy. Per Med 2012;9:547-57.
12. Scott JG, Berglund A, Schell MJ, et al. A genome-based model for adjusting radiotherapy dose (GARD): a retrospective, cohort-based study. Lancet Oncol 2017;18:202-11.
13. Ahmed KA, Scott JG, Arrington JA, et al. Radiosensitivity of lung metastases by primary histology and implications for stereotactic body radiation therapy using the genomically adjusted radiation dose. J Thorac Oncol 2018;13:1121-7.
14. Ahmed KA, Grass GD, Orman AG, et al. Personalizing Radiation Treatment Delivery in the Management of Breast Cancer. Int J Breast Cancer 2018;2018:6729802.
15. Hall WA, Bergom C, Thompson RF, et al. Precision Oncology and Genomically Guided Radiation Therapy: A Report From the American Society for Radiation Oncology/American Association of Physicists in Medicine/National Cancer Institute Precision Medicine Conference. Int J Radiat Oncol Biol Phys 2018;101:274-84.
16. Hammond SM. An Overview of MicroRNAs. Adv Drug Deliv Rev 2015;87:3-14.
17. Boeri M, Verri C, Conte D, et al. MicroRNA signatures in tissues and plasma predict development and prognosis of computed tomography detected lung cancer. Proc Natl Acad Sci U S A 2011;108:3713-8.
18. Castro D, Moreira M, Gouveia AM, et al. MicroRNAs in Lung Cancer. Oncotarget 2017;8:81679-85.
19. Keller A, Leidinger P, Gislefoss R, et al. Stable serum miRNA profiles as potential tool for non-invasive lung cancer diagnosis. RNA Biol 2011;8:506-16.
20. Sun Y, Schipper MJ, Hawkins PG, et al. Serum MicroRNA signature predicts response to high-dose radiation therapy in locally advanced non-small cell lung cancer. Int J Radiat Oncol Biol Phys 2018;100:107-14.
21. Leng Q, Lin Y, Jiang F, et al. A plasma miRNA signature for lung cancer early detection. Oncotarget 2017;8:111902-11.
22. Hu Z, Chen X, Zhao Y, et al. Serum MicroRNA signatures identified in a genome-wide serum MicroRNA expression profiling predict survival of non-small-cell lung cancer. J Clin Oncol 2010;28:1721-6.
23. Võsa U, Vooder T, Kolde R, et al. Identification of miR-374a as a prognostic marker for survival in patients with early-stage nonsmall cell lung cancer. Genes Chromosomes Cancer 2011;50:812-22.
24. Hinkson IV, Davidsen TM, Klemm JD, et al. A Comprehensive Infrastructure for Big Data in Cancer Research: Accelerating Cancer Research and Precision Medicine. Front Cell Dev Biol 2017;5:83.
25. Vyfhuis MAL, Bhooshan N, Burrows WM, et al. Oncological outcomes from trimodality therapy receiving definitive doses of neoadjuvant chemoradiation (≥60 Gy)


 

and factors influencing consideration for surgery in stage III non-small cell lung cancer. Adv Radiat Oncol 2017;2:259-69.
26. Vyfhuis MAL, Burrows WM, Bhooshan N, et al. Implications of pathologic complete response beyond mediastinal nodal clearance with high-dose neoadjuvant chemoradiation therapy in locally advanced, non-small cell lung cancer. Int J Radiat Oncol Biol Phys 2018;101:445-52.

**Cite this article as:** Amin NP, Mohindra P, Jabbour SK. Serum microRNA guiding personalized radiation therapy in non-small cell lung cancer. J Thorac Dis 2018;10(Suppl 33):S4108-S4112. doi: 10.21037/jtd.2018.09.143