Cell Reports Medicine

CellPress
OPEN ACCESS

Spotlight

MEK inhibition invigorates chemoimmunotherapy by tumor mitophagy-induced CXCL10 expression

B. Leticia Rodriguez[1] and Don L. Gibbons[1,2,*]
[1]Department of Thoracic/Head and Neck Medical Oncology, The University of Texas MD Anderson Cancer Center, Houston, TX 77030, USA
[2]Department of Molecular and Cellular Oncology, The University of Texas MD Anderson Cancer Center, Houston, TX 77030, USA
*Correspondence: dlgibbon@mdanderson.org
https://doi.org/10.1016/j.xcrm.2021.100506

A recent study by Limagne et al.[1] in *Cancer Cell* demonstrates that addition of MEK inhibitor to standard-of-care platinum/pemetrexed promotes mitophagy-dependent CXCL10 expression via optineurin and TLR9. Tumor cell secretion of CXCL10 produces T cell recruitment and enhances immunotherapy efficacy.

In various tumor types, including non-small cell lung cancer (NSCLC), the MAPK signaling pathway is frequently mutated at one of multiple points and plays a driver role in tumorigenesis. However, except in combination with RAF inhibitors for BRAF mutant NSCLC, MEK inhibitors have failed to demonstrate clinical activity for oncogene-selected or broader groups of NSCLC as single-agents, or with either chemotherapy or immunotherapy. Front-line treatment options for NSCLC patients include treatment with immune checkpoint blockade (ICB) alone or with a platinum-based doublet chemotherapy (taxane or pemetrexed), depending on histology. Despite high response rates and improved progression-free or overall survival, durable responses indicative of effective immune engagement are observed in less than 50% of patients receiving the ICB combination with platinum/pemextrexed.[2] The lack of effective response is most pronounced in patients with low levels of PD-L1 staining, suggesting that patients without significant tumor immune infiltration at baseline are less likely to respond. Identifying combination strategies that may enhance the durable immunologic response will improve clinical outcomes for NSCLC patients. Recent pre-clinical studies have highlighted the potential role of MEK inhibitor combinations that affect the immune cell landscape[3,4] or where tumor MAPK activation drives an immune-evasive tumor microenvironment,[5] indicating a potential role for MEK inhibitors outside of the targeted agent landscape.

Using the syngeneic Lewis lung cancer (LLC1) model, Limagne et al.[1] initially investigated the optimal chemotherapy treatment strategy to induce immunological cell death (ICD), aiming to identify combinations that might work best with ICB. Standard chemotherapy platinum-doublet regimens were tested and cisplatin/pemetrexed was found to induce the highest levels of ten validated ICD markers. However, they further observed that the addition of immunotherapy (anti-PD-L1) did not improve the therapeutic outcome of cisplatin/pemetrexed. The authors hypothesized that despite the synergistic stimulation of ICD, the chemo-immunotherapy combination was unable to induce T cell recruitment and that augmentation of the CXCL10 levels may reinvigorate the T cell recruitment and immune response.

The authors performed a drug screen with clinically available kinase inhibitors to rationally identify a combination capable of restoring CXCL10 production upon cisplatin/pemetrexed treatment. They observed that three MEK inhibitors (MEKi) and an ERK inhibitor enhanced CXCL10 production in combination with cisplatin/pemetrexed treatment. Utilizing the MEK inhibitor, trametinib, they found that the CXCL10 levels increased in a dose-dependent manner only in combination with cisplatin/pemetrexed and enhanced mouse survival in the LLC1 model in an immune-dependent manner. Addition of the MEK inhibitor enhanced the T cell infiltration, consistent with findings by multiple groups.[6,7] The addition of the MEK inhibitor to cisplatin/pemetrexed also induced a prominent upregulation of PD-L1. Therefore, the authors investigated the addition of PD-L1 checkpoint blockade to the MEKi/cisplatin/pemetrexed combination in the LLC1 model and observed enhanced survival and improved tumor growth inhibition. They further confirmed the enhanced *in vivo* effects of MEK inhibition on chemoimmunotherapy across multiple models, including a urethane-induced lung cancer model and both the syngeneic 4T1 breast and the CT26 colorectal cancer models.

The authors investigated the mechanism underlying the improved efficacy of MEK inhibitor in the cisplatin/pemetrexed combination and observed that the increased CXCL10 expression was from the tumor cells rather than the CD45 population, suggesting a tumor-intrinsic response specifically due to MEK inhibition. To gain a deeper understanding of the tumor cell changes contributing to the increased CXCL10, the authors performed mRNA sequencing, and differential expression analysis revealed an increase in pathways associated with autophagy/mitophagy processes. Knockout of the essential autophagy related gene 5 (Atg5) decreased CXCL10 induction and eliminated the synergistic effect of MEKi within the cisplatin/pemetrexed combination. Interestingly these results are consistent with recent findings by Kinsey et al.[8] which also found that inhibition of the MEK/ERK signaling pathway elicits autophagy. The authors further found that the selective mitophagy adaptor optineurin was induced by MEK inhibition and was necessary for both CXCL10 production and the antitumor effect of the MEKi/cisplatin/pemetrexed combination. Optineurin appears to be the driver of autophagy, as its knockdown

eliminates the ability of MEKi to induce CXCL10. The authors also demonstrate that mitochondrial damage released mitochondrial DNA and activated TLR9 in the LLC1 cells to trigger CXCL10 production. Taking these results together, the authors have shown in the syngeneic LLC1 tumors that the addition of MEKi to cisplatin/pemetrexed drives mitochondrial damage and mitophagy via optineurin, resulting in mitochondrial DNA degradation that is recognized by TLR9 and leads to CXCL10 expression. CXCL10 expression by the tumor cells increases CD8 T cell infiltration into the microenvironment and sensitizes them to checkpoint blockade.

The authors explored the translational impact of their preclinical findings by evaluating human lung cancer cell lines and performing retrospective analysis of either soluble plasma factors or mRNA seq from four small, independent cohorts of advanced NSCLC patients treated with immunotherapy alone or platinum-based doublet chemotherapy. Increased CXCL10 expression was observed in human NSCLC cell lines (including A549, H358, and H1975) after exposure to MEKi/cisplatin/pemetrexed as compared to cisplatin/pemetrexed. Patients that had response to first-line platinum-based doublet chemotherapy had higher levels of circulating CXCL10, while higher plasma levels of CXCL10 were found to be associated with better progression-free survival rates upon subsequent anti-PD-1 treatment. In the Cancer Genome Atlas (TCGA) and other cohorts, the authors observed a correlation between the activation of the MAPK pathway and the expression of OPTN, TLR9, CXCL10, and the levels of CD8A as a marker of effector T cells. They additionally showed that the combination of OPTN, CXCL10, and TLR9 levels predicts for better progression-free survival with anti-PD-1 treatment.

Although their original observations of synergy with MEKi/chemoimmunotherapy treatment were noted across tumor models, one of the study limitations arises from the authors primarily using the LLC1 model for their mechanistic studies outlining the role for mitophagy, which they acknowledge may not entirely recapitulate human cancer biology. Nonetheless, the studies with human cell lines provide additional support for some of the preclinical results. Another limitation is that the retrospective translational analyses were performed in advanced NSCLC samples from patients who did not receive MEK inhibitor treatment. These patients were treated with immunotherapy as second-line treatment after progression on initial platinum-based doublet chemotherapy, but were not treated with MEK inhibitors, providing a caveat to the applicability of the results. Clinical trials in which a MEK inhibitor was added to standard-of-care chemotherapy reveal limited efficacy and a high incidence of toxicity.[9,10] As such, addition of MEK inhibitor to standard-of-care chemoimmunotherapy may need to be considered on a temporal sequential basis guided by biomarkers and/or indicators of resistance. Recent pre-clinical work by Wang et al.[4] found that initial treatment with immunotherapy followed by MAPK inhibition optimized anti-tumor response and lead to immune cell reinvigoration. Future studies should focus on the dosing of MEKi that is necessary for CXCL10 induction and further assess which biomarkers best indicate the time point and order when immunotherapy should be given versus MEKi.

In summary, the study by Laminge et al.[1] has shown that CXCL10 is critical for CD8 T cell recruitment and necessary to restore immune checkpoint blockade efficacy within cisplatin/pemetrexed-treated tumors. The authors demonstrate that addition of MEK inhibitor restored CXCL10 expression, which was accomplished via upregulation of the optineurin and TLR9 axis. This work highlights the need to utilize effective combination approaches that are guided by biomarkers, such as CXCL10, in order to enhance the efficacy of currently available immune checkpoint therapies. A focused effort is needed to determine and define biomarkers of response to MEK inhibitors, chemotherapy/immunotherapy, and the optimal drug dosing strategy that will produce better outcomes for NSCLC patients.

DECLARATION OF INTERESTS

D.L.G. has received research funding from AstraZeneca, Astellas, Janssen, Ribon Therapeutics, Takeda, and NGM Biopharmaceuticals; reports advisory role/consulting fees from AstraZeneca, Sanofi, Menarini Ricerche, and Eli Lilly; and research funding to MD Anderson Cancer Center from Boehringer Ingelheim. B.L.R. declares no competing interests.

REFERENCES

1. Limagne, E., Nuttin, L., Thibaudin, M., Jacquin, E., Aucagne, R., Bon, M., Revy, S., Barnestein, R., Ballot, E., Truntzer, C., et al. (2021). MEK inhibition overcomes chemoimmunotherapy resistance by inducing CXCL10 in cancer cells. Cancer Cell. Published January 13, 2022. https://doi.org/10.1016/j.ccell.2021.12.009.
2. Gandhi, L., Rodríguez-Abreu, D., Gadgeel, S., Esteban, E., Felip, E., De Angelis, F., Domine, M., Clingan, P., Hochmair, M.J., Powell, S.F., et al.; KEYNOTE-189 Investigators (2018). Pembrolizumab plus Chemotherapy in Metastatic Non-Small-Cell Lung Cancer. N. Engl. J. Med. *378*, 2078–2092. https://doi.org/10.1056/NEJMoa1801005.
3. Peng, D.H., Rodriguez, B.L., Diao, L., Gaudreau, P.O., Padhye, A., Konen, J.M., Ochieng, J.K., Class, C.A., Fradette, J.J., Gibson, L., et al. (2021). Th17 cells contribute to combination MEK inhibitor and anti-PD-L1 therapy resistance in KRAS/p53 mutant lung cancers. Nat. Commun. *12*, 2606. https://doi.org/10.1038/s41467-021-22875-w.
4. Wang, Y., Liu, S., Yang, Z., Algazi, A.P., Lomeli, S.H., Wang, Y., Othus, M., Hong, A., Wang, X., Randolph, C.E., et al. (2021). Anti-PD-1/L1 lead-in before MAPK inhibitor combination maximizes antitumor immunity and efficacy. Cancer Cell *39*, 1375–1387.e6. https://doi.org/10.1016/j.ccell.2021.07.023.
5. Haas, L., Elewaut, A., Gerard, C.L., Umkehrer, C., Leiendecker, L., Pedersen, M., Krecioch, I., Hoffmann, D., Novatchkova, M., Kuttke, M., et al. (2021). Acquired resistance to anti-MAPK targeted therapy confers an immune-evasive tumor microenvironment and cross-resistance to immunotherapy in melanoma. Nat. Can. *2*, 693–708. https://doi.org/10.1038/s43018-021-00221-9.
6. Verma, V., Jafarzadeh, N., Boi, S., Kundu, S., Jiang, Z., Fan, Y., Lopez, J., Nandre, R., Zeng, P., Alolaqi, F., et al. (2021). MEK inhibition reprograms $CD8^+$ T lymphocytes into memory stem cells with potent antitumor effects. Nat. Immunol. *22*, 53–66. https://doi.org/10.1038/s41590-020-00818-9.
7. Ebert, P.J.R., Cheung, J., Yang, Y., McNamara, E., Hong, R., Moskalenko, M., Gould, S.E., Maecker, H., Irving, B.A., Kim, J.M., et al. (2016). MAP Kinase Inhibition Promotes T Cell and Anti-tumor Activity in Combination with PD-L1 Checkpoint Blockade. Immunity *44*, 609–621. https://doi.org/10.1016/j.immuni.2016.01.024.
8. Kinsey, C.G., Camolotto, S.A., Boespflug, A.M., Guillen, K.P., Foth, M., Truong, A.,

Schuman, S.S., Shea, J.E., Seipp, M.T., Yap, J.T., et al. (2019). Protective autophagy elicited by RAF®MEK®ERK inhibition suggests a treatment strategy for RAS-driven cancers. Nat. Med. *25*, 620–627. https://doi.org/10.1038/s41591-019-0367-9.

9. Melosky, B., Bradbury, P., Tu, D., Florescu, M., Reiman, A., Nicholas, G., Basappa, N., Rothenstein, J., Goffin, J.R., Laurie, S.A., et al. (2019). Selumetinib in patients receiving standard pemetrexed and platinum-based chemotherapy for advanced or metastatic KRAS wildtype or unknown non-squamous non-small cell lung cancer: A randomized, multicenter, phase II study. Canadian Cancer Trials Group (CCTG) IND.219. Lung Cancer *133*, 48–55. https://doi.org/10.1016/j.lungcan.2019.04.027.

10. Froesch, P., Mark, M., Rothschild, S.I., Li, Q., Godar, G., Rusterholz, C., Oppliger Leibundgut, E., Schmid, S., Colombo, I., Metaxas, Y., et al.; Swiss Group for Clinical Cancer Research (SAKK) (2021). Binimetinib, pemetrexed and cisplatin, followed by maintenance of binimetinib and pemetrexed in patients with advanced non-small cell lung cancer (NSCLC) and KRAS mutations. The phase 1B SAKK 19/16 trial. Lung Cancer *156*, 91–99. https://doi.org/10.1016/j.lungcan.2021.04.002.