Fatty acid metabolism and desaturation in the pathogenesis of leukemic stem cells in Acute Myeloid Leukemia

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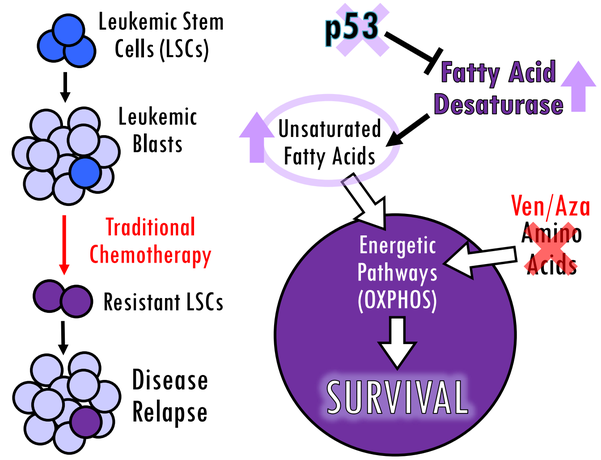
Abstract

Background: Acute myeloid leukemia (AML) is a cancer of bone marrow-derived blood cells, where leukemic blasts build up and block function and development of myeloid progenitors. Conventional therapy eliminates bulk tumor cells but leukemic stem cells (LSCs) survive, leading to disease progression and relapse. LSCs uniquely rely on oxidative phosphorylation (OXPHOS), metabolically driven by amino acid and fatty acid metabolism.

Aim: We have successfully targeted amino acid metabolism in LSCs, but the mechanisms controlling fatty acid metabolism are yet unknown. Our primary objective is to understand how fatty acids fuel OXPHOS in LSCs.

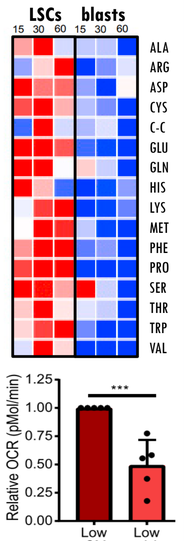
Results and Discussion: LSCs in relapsed/refractory patients display increased fatty acid metabolism, driving OXPHOS and LSC survival. Unsaturated fatty acids are oxidized more rapidly than saturated, so increased fatty acid desaturase (FADS) activity fuels OXPHOS more than overall fatty acid metabolism. Similar increases in fatty acid desaturation occur in cases of p53 loss in AML. Successful inhibition of OXPHOS is dependent on p53-driven apoptotic pathways, and p53 is a tight regulator of lipid metabolism. Therefore, loss of p53 function in AML may result in loss of FADS inhibition and promotion of fatty acid desaturation.

Conclusion: Relapsed/refractory LSCs upregulate fatty acid desaturation through increased FADS activity to maintain OXPHOS as a mechanism for survival. Additionally, loss of p53 function in AML may result in loss of inhibition of FADS1, increasing fatty acid desaturation. As unsaturated fatty acids are oxidized more quickly than saturated, this may allow relapse LSCs to compensate for a loss of amino acids resulting from Ven/Aza.



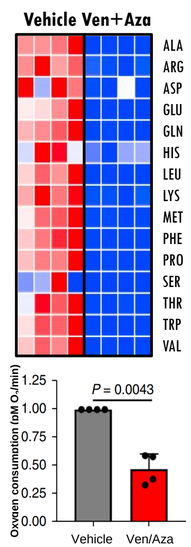
1. Jones CL, et al. Inhibition of Amino Acid Metabolism Selectively Targets Human Leukemia Stem Cells. Cancer Cell. 2019.
2. Pollyea DA, et al. Venetoclax with azacitidine disrupts energy metabolism and targets leukemia stem cells in patients with acute myeloid leukemia. Nature Medicine. 2018.
3. Nechiporuk T, et al. Cancer Discovery. The TP53 Apoptotic Network Is a Primary Mediator of Resistance to BCL2 Inhibition in AML Cells. 2019.
4. Moon et al. Cell. p53 Represses the Mevalonate Pathway to Mediate Tumor Suppression. 2019.

Ven/Aza targets *de novo* LSC metabolism…

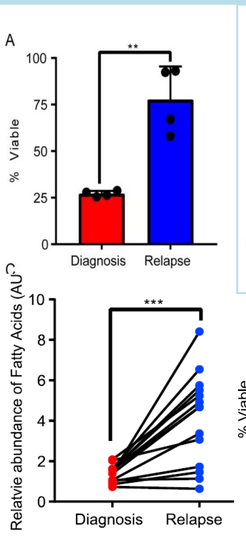


Newly diagnosed (*de novo*) LSCs uptake amino acids more quickly than leukemic blasts, and when amino acids are removed, OXPHOS is reduced. ***De novo* LSCs use amino acids to fuel OXPHOS.**

When *de novo* LSCs are treated with a combination of Venetoclax (BCL-2 inhibitor) and Azacitidine (DNA hypomethylator), amino acid depletion is recapitulated. **Ven/Aza metabolically targets *de novo* LSCs by impairing amino acid import and metabolism.**

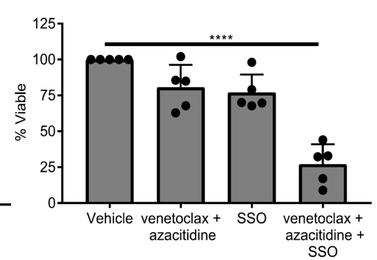


… but relapse LSCs can metabolically compensate

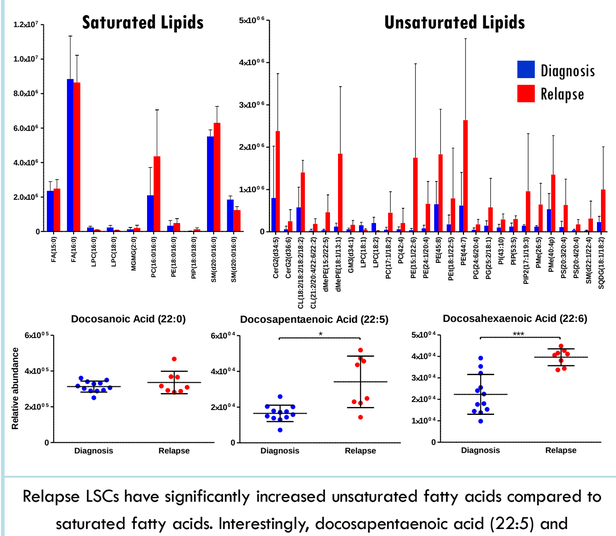


Upon treatment with Ven/Aza, relapse LSCs are significantly more viable than *de novo* LSCs. **Ven/Aza isn't affecting relapse LSCs in the same way as *de novo* LSCs.**

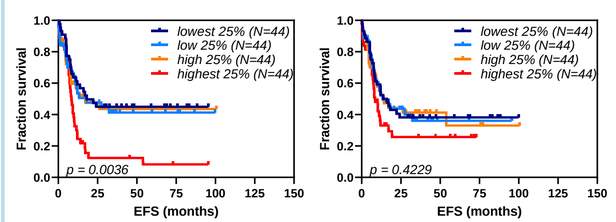
Upon depletion of AAs, relapse LSCs show increased levels of fatty acids. Addition of fatty acid translocase (CD36) inhibitor SSO re-sensitizes relapse LSCs to Ven/Aza. **Relapse LSCs may compensate for amino acid loss by upregulating fatty acid metabolism.**



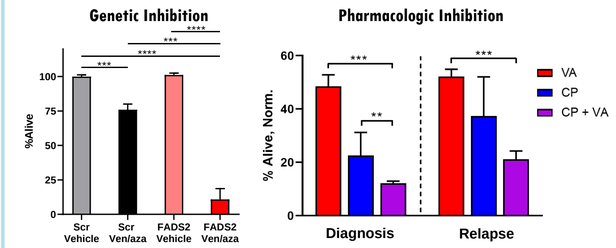
Dysregulation of fatty acid desaturation in relapse



Relapse LSCs have significantly increased unsaturated fatty acids compared to saturated fatty acids. Interestingly, docosapentaenoic acid (22:5) and docosahexaenoic acid (22:6) are the end products of a pathway to produce highly unsaturated fatty acids using **fatty acid desaturases FADS1 and FADS2.**

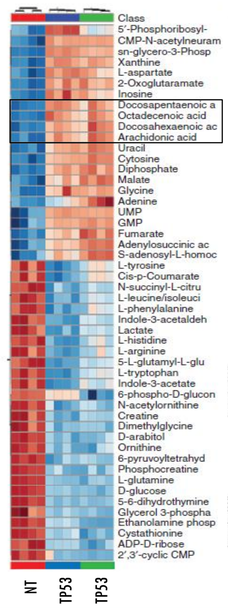


Increased expression of FADS1 and FADS2 is associated with poor prognosis in patients with AML, suggesting **FADS activity contributes to LSC survival.**



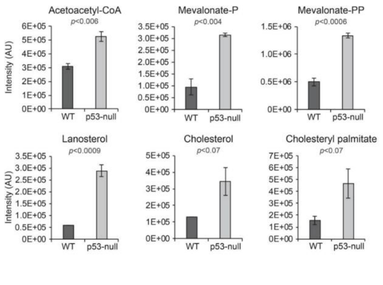
Both genetic and pharmacologic inhibition of FADS, when combined with ven/aza, successfully eliminate LSCs from relapse AML patients.

p53 controls FA metabolism and desaturation

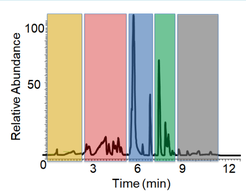


p53 knockout in an AML cell line results in increased unsaturated fatty acids, including docosapentaenoic acid (22:5) and docosahexaenoic acid (22:6). **Loss of p53 increases unsaturated fatty acids, similarly to the relapsed AML phenotype.**

p53 knockout also results in increases in all intermediates of the mevalonate pathway, resulting from production of Acetyl-CoA from fatty acids.



Methods



Vanquish UHPLC
Kinetex C18 column - 2.1x150mm, 1.7µ
- 3 min. isocratic elution
- 5/9 min. gradient elution

Acquity HSS T3 column - 2.1x150mm, 1.8µ
- 17 min. gradient elution

Q Exactive Mass Spectrometer
Full MS mode, 60-900m/z, 125-1500m/z

Future Directions

- Determine the effects of Ven/Aza on lipid desaturation and fatty acid metabolism
- Perform p53 knockdown in primary patient LSCs to confirm aberrant lipid desaturation
- Genetically and pharmacologically inhibit FADS in the context of p53 loss
- Further explore the role of p53 and the proteins controlling its function in the mechanism of survival for relapse LSCs
- Explore metabolic progression from MDS to AML