Examining Lactate as a Fuel Source in Human Non-Small Cell Lung Cancer

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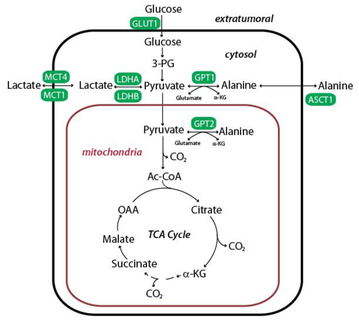
Abstract

Altered metabolism is a hallmark of cancer. Metabolic pathways such as glycolysis are enhanced in malignant tissue and can support the accelerated growth and proliferation of cancer cells. Using ^{13}C-stable isotope tracing, we found that glucose is highly oxidized in tumors compared to surrounding benign lung tissue. Next, we observed that lactate, in addition to glucose, can be used as a nutrient source by lung tumors. We primarily examine the molecular mechanisms of this phenomenon in cell and animal systems using siRNA and CRISPR-mediated knockdown of the lactate transporters MCT1 and MCT4. These clinical observations highlight new aspects of in vivo cancer metabolism, which may lead to the development of new biomarkers or therapeutic opportunities.

Introduction

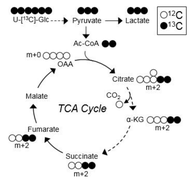
- Altered metabolism in tumors is a hallmark of cancer which is commonly characterized by increased glucose metabolism.
- Nutrient usage by tumors in vivo (beyond glucose), remains largely unknown
- Lactate is utilized as a fuel source by normal organ tissue such as the brain and liver.
- Lactate intake by tumors is controversial- it is conventionally thought that lactate is excreted as a waste product by tumor cells.
- High expression of the lactate importer MCT1 is correlated with poor prognosis in patients.



Methods

- Tumors resected from patients with NSCLC analyzed using Gas Chromatography-Mass Spectrometry (GCMS)

Mass Isotope Tracing by GCMS



- Inhibition of lactate transporters (MCT1 and MCT4) and lactate conversion enzymes (LDHA and LDHB) in NSCLC cell lines using siRNA, pharmacological agents, or CRISPR
  - Knockdown/out verified with Western Blot
  - NOVA and GCMS were used to analyze cell media and cell contents, respectively, to measure amounts of metabolites
- Positive CRISPR knockout cell lines can then be injected into mice and grown into tumors

Hypothesis

Tumor cells can utilize lactate as a fuel source in NSCLC tumors, and knocking out the lactate transporter MCT1 will inhibit the tumors ability to utilize lactate.

Results

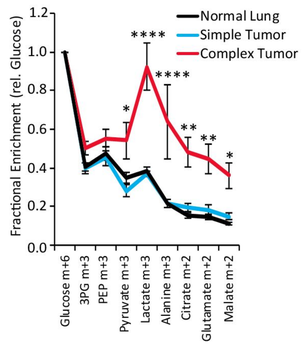


Figure 1: **Most human lung tumors display evidence of lactate uptake**. Data from n= 28 patients is shown (AVG +/- SD).

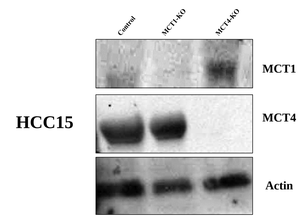


Figure 2: Western Blot demonstrates decreased MCT1 and MCT 4 protein expression when treated with siRNA

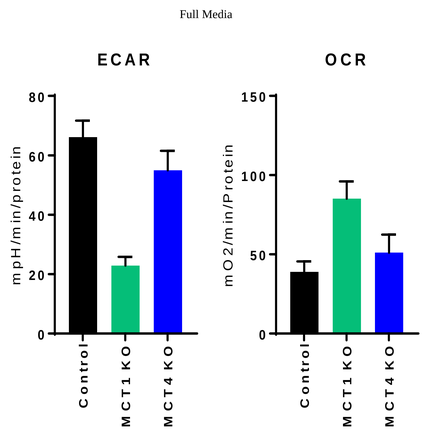


Figure 3: **Knocking out MCT1 severely reduced ECAR and increased OCR**

Results

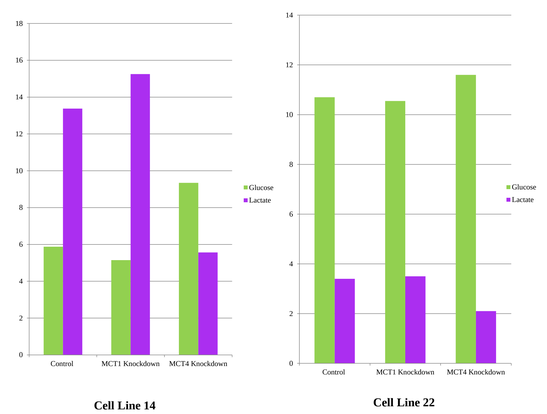


Figure 4: **NOVA Analysis. MCT1 Knockdown decreased lactate uptake in both cell lines**. The lactate decrease is larger in cell line 14 compared to cell line 22

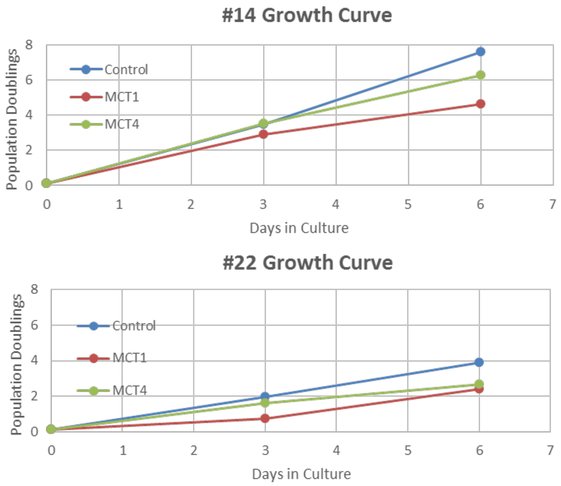


Figure 5: **MCT1 Knockdown decreased cell growth in both cell lines.** Cell growth decreased more in cell line 14 compared to cell line 22.

Summary

Inhibiting MCT1 has a much greater effect on lactate import than MCT4. However, some NSCLC cell lines are more susceptible to this inhibition that others. These susceptible cell lines also experience pronounced inhibition of cell growth when MCT1 is inhibited

Conclusion

- Human NSCLC tumors can utilize lactate as a fuel source.
- Inhibiting MCT1 in susceptible NSCLC lines significantly impacts a tumor's ability to uptake lactate and results in decreased growth.
- These observations could be used to develop more effective treatments for patients with NSCLC.

References

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