メタボロミクス解析によるMalic enzyme 1のがん細胞における機能解析と創薬標的としての評価

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Abstract

Malic enzyme 1 (ME1) regulates one of the main pathways that provide nicotinamide adenine dinucleotide phosphate (NADPH), which is essential for cancer cell growth through maintenance of redox balance and biosynthesis processes in the cytoplasm. In this study, we found that ME1 inhibition disrupted metabolism in cancer cells and inhibited cancer cell growth with accompanying senescence. In glucose-restricted conditions, cancer cells increased ME1 expression, and tracer experiments with labelled glutamine revealed that the flux of ME1-derived pyruvate to citrate was enhanced. In addition, cancer cells showed higher sensitivity to ME1 depletion in glucose-restricted conditions compared to normal culture conditions. These results suggest that in a low-glucose environment, where glycolysis and the pentose-phosphate pathway (PPP) is attenuated, cancer cells become dependent on ME1 for the supply of NADPH and pyruvate. Our data demonstrate that ME1 is a promising target for cancer treatment, and a strategy using ME1 inhibitors combined with inhibition of glycolysis, PPP or redox balance regulators may provide an effective therapeutic option.

Result

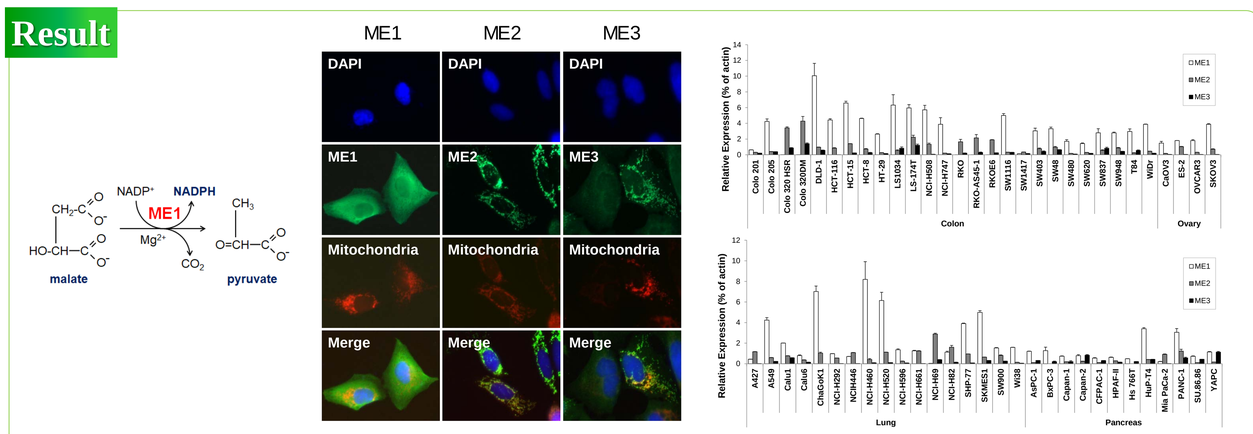

Figure 1. Malic enzyme 1, a NADPH-producing enzyme, was localized in cytosol and highly expressed in various cancer cell lines.

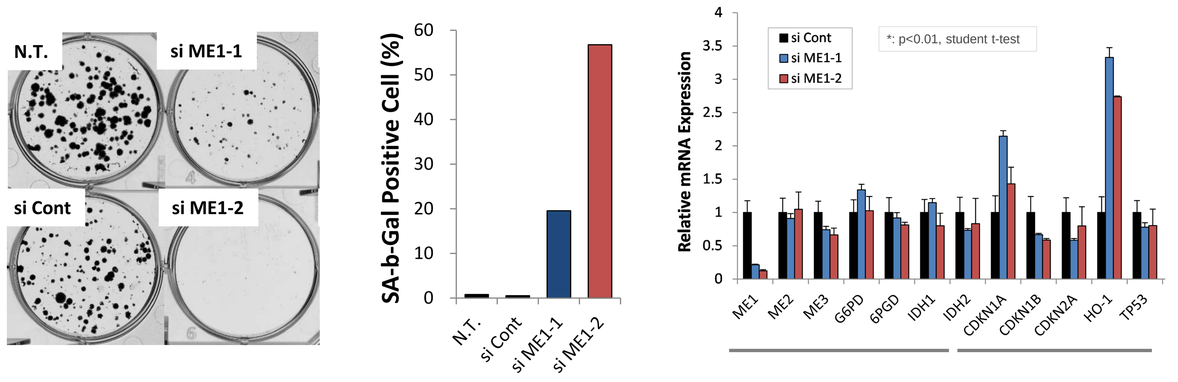

Figure 2. Knock down of ME1 suppressed growth of HCT116 cells and induced senescence and oxidative stress response.

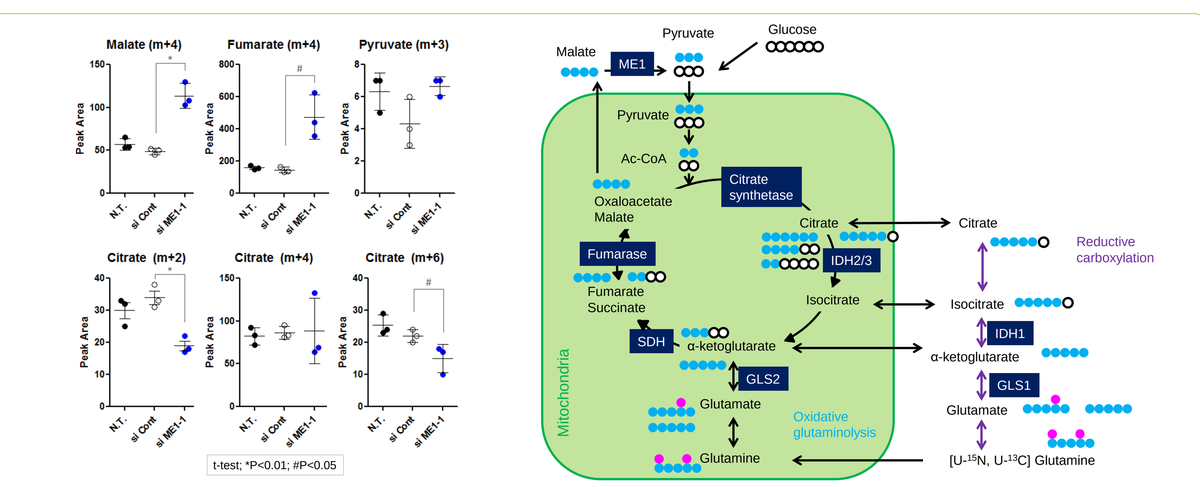

Figure 3. [U-^{13}C, U-^{15}N] L-glutamine flux analysis revealed ME1 knock down enhanced accumulation of malate and decreased downstream metabolites except for pyruvate.

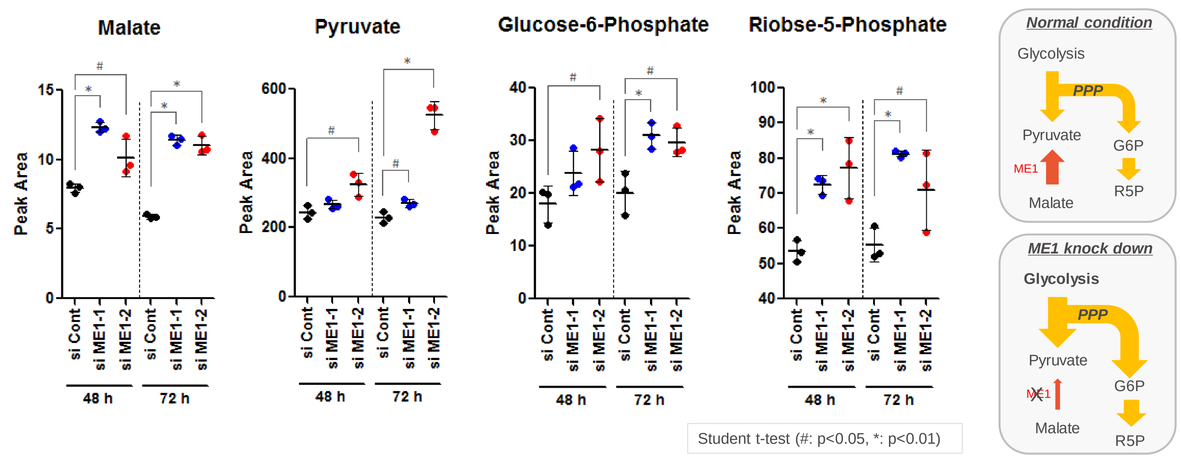

Figure 4. Pyruvate, G6P and R5P were increased by ME1 inhibition, suggesting ME1 inhibition activated glycolysis and pentose-phosphate pathway (PPP).

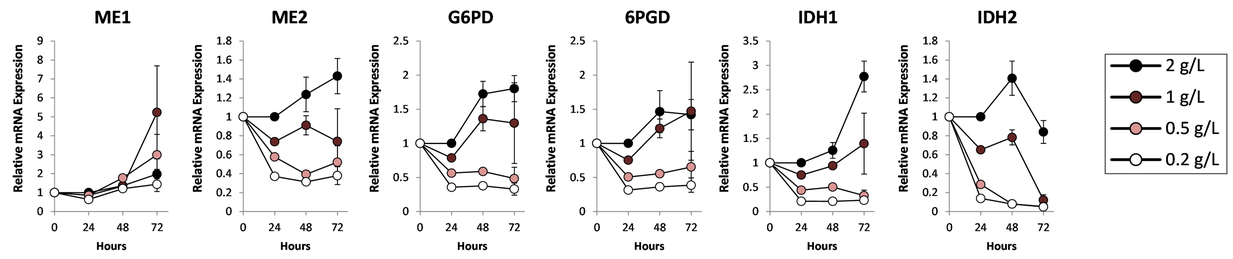

Figure 5. mRNA level of ME1 in HCT116 cells was augmented in lower concentration of glucose while that of other NADPH-producing enzymes were decreased.

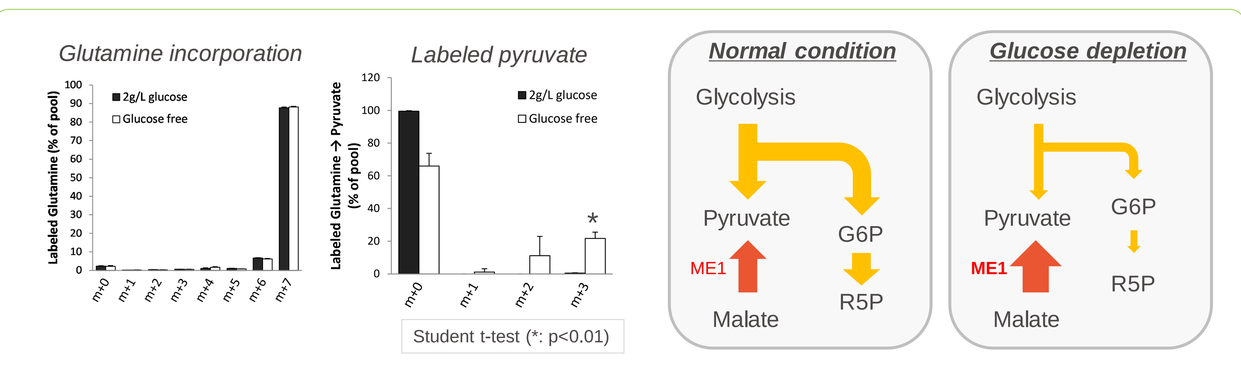

Figure 6. Pyruvate (m+2) and (m+3) were found to be increased in glucose-depleted condition by **[**U-^{13}C, U-^{15}N] L-glutamine flux analysis while glutamine incorporation was not changed, suggesting HCT116 cells utilized ME1 metabolic pathway under glucose-depleted condition.

Figure 7. ME1 gene knock-down by shRNA under glucose-depleted condition synergistically suppressed growth of HCT116 cells accompanied by NADPH depletion and oxidative stress.

Conclusion & Discussion

- In this study, we demonstrated that ME1 is an essential enzyme that regulates cellular metabolism and redox balance in cancer cells by metabolomics and labeled-glutamine flux analyses. We also showed that, in glucose-depleted conditions, cancer cells become dependent on the ME1 flux to produce NADPH and pyruvate and to manage redox homeostasis, suggesting that these cells become vulnerable to reduced ME1 activity. Taken together, our results demonstrated that ME1 is a promising target for treatment of tumors in a nutrient-limited microenvironment and that a strategy using ME1 inhibitors combined with inhibition of glycolysis or redox balance regulators may be an effective therapy for cancer patients.
- Further studies in *in vivo* setting should be conducted to evaluate ME1 as a therapeutic target disrupting cancer metabolism and redox homeostasis in tumor and to confirm combination effect on tumor growth.
- In addition, patient stratification marker should be elucidated for effective ME1 disrupting therapy. For example, ME1 expression is known to be regulated by Nrf2, and tumors with mutated Keap1 or Nrf2 genes could be dependent on ME1 and thus be sensitive to ME1 inhibition
- Currently, no ME1 inhibitors are being evaluated in clinical trials, although a few small-molecule ME2 inhibitors were discovered in pre-clinical trials. Therefore, ME1 inhibitor is a first-in-class target in oncology area and could provide a new therapeutic strategy and deliver a benefit for cancer patients.