論文 / 著書情報
Article / Book Information

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Title(English)	The Effect of 5-Aminolevulinic Acid and Sodium Ferrous Citrate on Mitochondrial Properties in Myoblast and Cancer Cells
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種別(和文)	論文要旨
Type(English)	Summary

（博士課程）
Doctoral Program

論 文 要 旨

THESIS SUMMARY

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要旨（英文 800 語程度）
Thesis Summary（approx.800 English Words ）

Mitochondria is the organelle that perform aerobic respiration and produce energy (ATP) through oxidative phosphorylation system which necessary for cellular activities (Bertram *et. al.,* 2006). It also plays an important role in controlling/signaling calcium (Ca^{2+}) which is essential for maintaining homeostasis of eukaryotic cells (Schatz, 1995). However, it has been found that the number and functions of mitochondria decrease in the process of aging and disease, then led to an abnormality in mitochondria. It is becoming very clear that the progress will be adversely affected, so improving the mitochondrial function and number are needed to eliminate the negative cycle due to mitochondrial abnormalities.

In the previous study, it has been investigated the differences of iron metabolism in tumor cells and in normal cells and found that the amount of mitochondrial labile iron ion in cancer cells was lower than that in normal cells (Hayasi *et. al.,* 2015). In addition, COX activity has been reported to decline in mitochondrial dysfunction, including in cancer cells (Rak *et. al.,* 2016). Therefore, in the present study, SFC was added for the free iron ions source, and the effect of ALA and SFC on cancer cells was analyzed.

This study has purpose to investigate the effect of ALA and SFC on mitochondrial activity in myoblast (C2C12) cells. Additionally, in cancer cells, this study also focused on the effect of ALA during PDT. Moreover, in the previous study, SFC is reported to increase COX activity. So, this study also aims to improve the COX activity by the addition of SFC that can promote ROS generation, which has a cytotoxic effect.

In summary, the findings in this study can be concluded as follows:

- The addition of ALA stimulates oxidative phosphorylation by upregulation COX in myoblast cells.
- The presence of ALA did not affect the mitochondrial content and mitochondrial biogenesis in myoblast cells.
- Mitochondrial activities were significantly improved by the addition of ALA in myoblast cells.
- The addition of ALA, SFC and ALA+SFC stimulates oxidative phosphorylation by upregulation of COX in myoblast cells.
- ALA markedly reduced the expression levels of the protein-containing Fe-S cluster. However, the abundance of free iron ions by SFC addition significantly up-regulated the expression levels of protein-containing Fe-S cluster in myoblast cells.
- The addition of ALA, SFC, and ALA+SFC stimulates oxidative phosphorylation by upregulation COX in cancer cells.
- ALA, SFC, and ALA+SFC markedly reduced the cell viability in cancer cells by increasing ROS production.

In this study, it demonstrated that the addition of ALA as the heme precursor, SFC as an iron source,

and the combination of ALA+SFC successfully increase heme production and contribute to the upregulation of heme proteins, resulting in the increase of COX activity and an increase in ATP levels in myoblast (C2C12) cells. Furthermore, the result showed that the addition of ALA, SFC, and the combination of ALA+SFC increased COX activity that shifts back the preferentially metabolized glucose by glycolysis in the cytosol to generate energy in the cancer cell to the usual citric acid cycle and OXPHOS in the mitochondria.

COX activity increases in both myoblast cells and cancer cells. Basal COX activity is higher in myoblast cells compared to in cancer cells 0.06 and 0.01 [units / mg-protein] consecutively. Then, with the addition of ALA, COX activity significantly increases in both cells 0.12 and 0.04 [units / mg-protein] consecutively. The administration of SFC also significantly increases the COX activity in both cells 0.11 and 0.16 [units / mg-protein] consecutively. Moreover, the combination of ALA+SFC obviously increase COX activity in both cells 0.21 and 0.18 [units / mg-protein] consecutively. Thus, the addition of ALA, SFC, and the combination ALA+SFC has more effective to increase the COX activity in myoblast cells compared to cancer cells.

The increasing of COX activity causes electron leakage during the electron transport within the mitochondria. Subsequently, excessive intracellular ROS production occurred, that leads to cell death. The basal level of COX activity is six times higher in myoblast compared to cancer cells. The ROS quenching system in myoblast is better compared to the cancer cells, even though ROS is produced in both cells. In cancer cells, the excessive amount of ROS that is accumulated that can cause the cell death. This phenomenon is not observed in myoblast cells.

To conclude, this study demonstrated that ALA and SFC improve mitochondrial properties in myoblast (C2C12) cells. Furthermore, the same results were obtained in the cancer cells, which reduced their cell viability by increasing ROS production. In conclusion, mitochondrial properties increase in myoblast, and this study also provides a novel treatment for mitochondrial dysfunction, especially cancer.