

Study of metformin effects in mTOR/S6Ks signal transduction pathway in lung cancer cells

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Abstract

Over the past few years, the mTOR signal transduction pathway has been associated to the control of body energy balance, acting as a sensor of the amount of nutrients available or a trigger for the expense or storage of energy. The S6Ks proteins have shown an important role in the mTOR signaling, working as effectors of this signaling pathway, although there are still questions not answered. Among the diseases related to obesity, as well as to the control or dysfunction of the mTOR/S6Ks pathway, cancer may be highlighted, whose incidence in obese is clearly higher. Metformin has been used in the treatment of type II diabetes, but many reports in the literature indicate its importance also as a drug against cancer. This research project aimed to study the effects of metformin treatment in the mTOR/S6Ks signaling pathway in A549 lung cancer cells. Our results show that metformin is able to reduce the mTOR pathway signaling even after treatment of cells with a low dose of cisplatin, which may induce lung cancer resistance.

Key words:

Cancer, metformin, mTOR.

Introduction

Lung adenocarcinoma is one of the main causes of cancer mortality worldwide and its prognosis remains poor, with a five-year survival rate of 15%. Therefore, the effective prevention and treatment have a high importance for this type of cancer. The metformin, a biguanide drug used in the treatment of diabetes type II, known to decrease blood glucose by mean liver and mitochondrial metabolism, has been shown to also reduce cancer effects¹. This was discovered after previous epidemiological studies on patients with diabetes who were treated with metformin, which presented lower cancer mortality than patients who received other drugs. Besides, the prevalence of diabetes in newly diagnosed cancer patients is near 20%, suggesting a bidirectional association between these two diseases².

Metformin is also able to increase the cytotoxicity effects of chemotherapy treatments of cancer, including the cisplatin in different lung cancer cell lines, but the molecular mechanisms are still unclear. Given the important role of the mTOR/S6K pathway in diseases related to metabolism such as obesity, diabetes and cancer, a relationship of the positive effects of metformin in cancer and this pathway may be characterized.

For this purpose, we investigated whether A549 lung cancer cells in a combination of treatments between cisplatin and metformin present a modulation of the mTOR/S6Ks signaling pathway.

Results and Discussion

Firstly, A549 cells were cultured in a 6 well plate and treated with metformin 10 μ M, cisplatin 10 μ M or metformin plus cisplatin for 72 hours. We found that metformin is able to decrease the phosphorylation of S6K1 and S6 proteins and thus decrease the signaling of mTOR pathway. Moreover, metformin was able to improve the effects of cisplatin, since the increased of the mTOR/S6Ks signaling by cisplatin was reversed by the combination with metformin (Figure 1).

According to previous studies³, the cell line A549 has a nonsense LKB1 mutation at codon 37. The LKB1 has been reported to negatively regulate mTOR pathway. One of the actions of metformin is to upregulate AMPK

via LKB1, thus decreasing mTOR signaling. We then decided to compare the activation of AMPK in A549 and PC3, a prostate cancer cell line that presents functional LKB1, in order to check a different response of the metformin treatment. As expected, we have found a change in AMPK phosphorylation after 4 hours of metformin treatment only in PC3 cells, but not in A549.

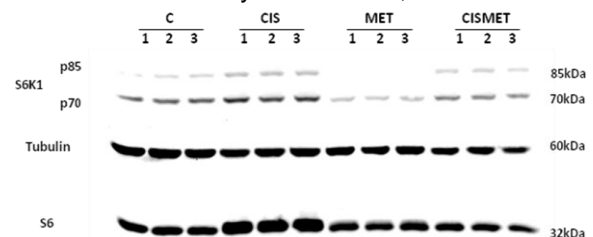


Figure 1. Regulation of S6K1 and S6, components of the mTOR signaling pathway, after treatment with cisplatin (CIS), metformin (MET) or their combination (CISMET).

Conclusions

In this study we have observed a reduction of the mTOR/S6K signaling pathway after treatment with metformin in A549 lung cancer cell line. This reduction was also achieved after a low dose of cisplatin treatment, with may induce lung cancer resistance. Besides, this effect has been characterized independent of the fact that A549 are not functional for LKB1/AMPK pathway, which is known to inhibit mTOR/S6Ks signaling pathway.

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