DECIPHERING THE ROLE OF THE MITOCHONDRIAL CHAPERONINE MCJ IN OVARIAN CANCER

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INTRODUCTION

Ovarian cancer (OC) is the most lethal gynecological neoplasia due to its extremely silent invasive capacity. The high mortality of OC is often determined by a therapeutic failure caused by the development of chemoresistance[1,2]. In this context, recent studies have shown that the acquisition of resistance is influenced by the cellular metabolic state and mitochondrial bioenergetic efficiency[3,4]. Interestingly, it has been reported that in OC the onset of chemoresistance is associated with the epigenetic silencing of *DNAJC15* that encodes for the mitochondrial co-chaperonine MCJ[5,6]. The latter is reported as an endogenous negative regulator of the electron transport chain (ETC), capable of modifying the structure and function of mitochondrial supercomplexes[7]. Furthermore, it is known that the mitochondrial energy state may modulate the activation of oncogenic pathways such as Wnt/β-catenin axis [8,9].

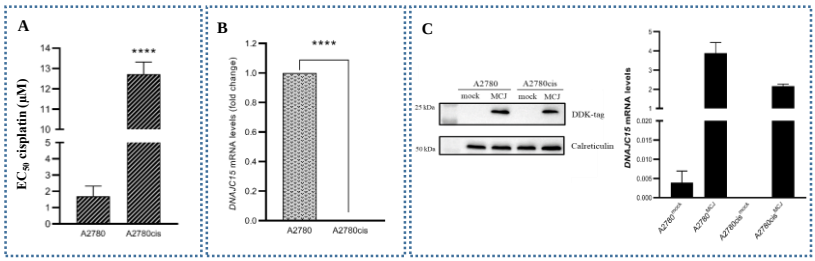
AIMS

The main aim of this project is to assess whether the expression of MCJ mitochondrial chaperone can modulates the OC growth and the chemoresistance occurrence, involving a mitochondria-Wnt/β-catenin axis.

MATERIAL & METHODS

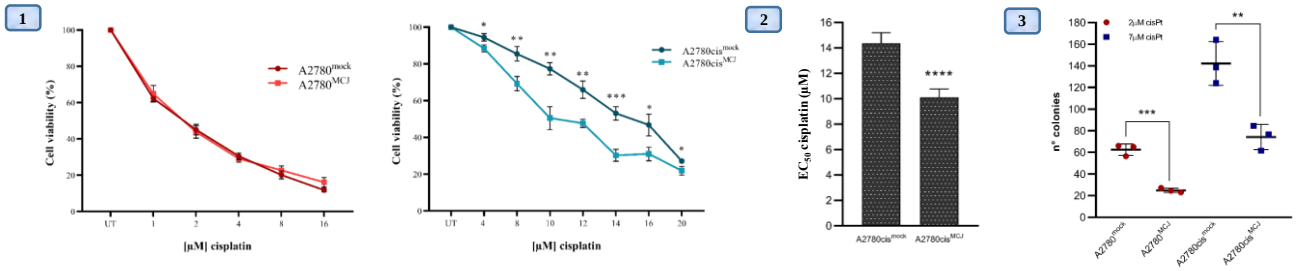
In order to investigate the role of MCJ in the molecular mechanisms underlying OC chemoresistance, we used chemosensitive (A2780) and its cisplatin-chemoresistant counterpart (A2780cis) OC cell lines, as shown in terms of cisplatin-related EC_{50} (Fig. A). According to the literature, the cisplatin chemoresistance is associated with the lack of MCJ expression (Fig. B). Further, we generated MCJ-overexpressing cell models of both chemosensitive and chemoresistant cells (Fig. C).

We used these OC cell lines to correlate the effect of MCJ expression with the onset of chemoresistance by evaluating the viability and the clonogenic ability of OC cells during cisplatin treatments. Futhermore, to evaluate the levels of expression of β-catenin and its downstream targets, Western blot and qRT-PCR analysis were performed. Moreover, in order to enroll the impact of MCJ on the bioenergetic profile of OC cells, we performed functional assays such as the measurement of spectrophotometric activity of ETC complexes, oxygen consumption (OCR) and the ROS production.

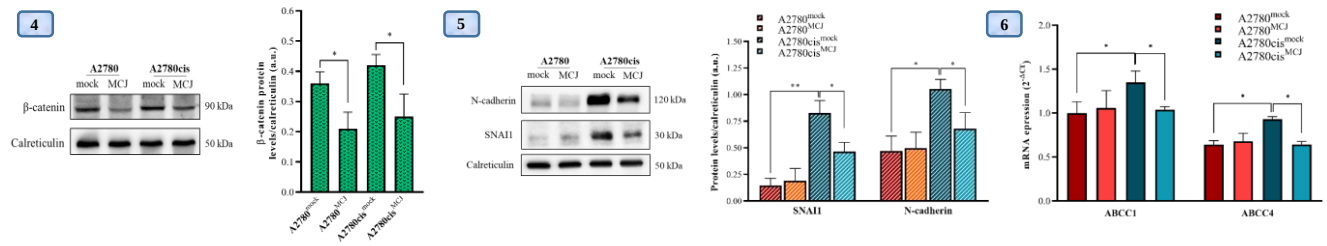


RESULTS

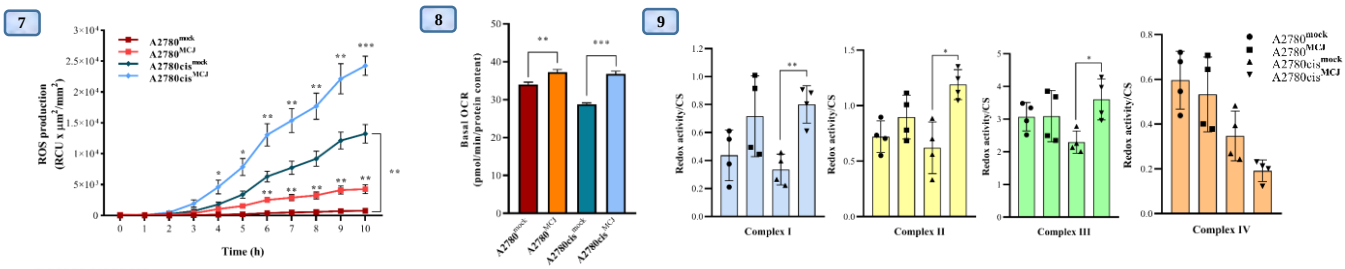
Interestingly, the MCJ overexpression was able to significantly restores the chemosensitivity only in A2780cis cell line (Fig 1, light blue curve) and, in turn, markedly decreases its cisplatin EC_{50} (Fig. 2). Moreover, MCJ is able to induces a significant reduction of the *in vitro* ability to form colonies, during cisplatin treatments of both OC chemosensitive and chemoresistant cell lines (Fig. 3).



Since Wnt/β-catenin pathway is able to transcribe downstream genes involved in multi-drug resistance (MDR), epithelial-mesenchymal transition (EMT) and cell proliferation, we investigate wheather MCJ could affects this regulatory axis. We found that the overexpression of MCJ is able to significantly reduce the β-catenin expression levels in both chemosensitive and chemoresistance cell lines (Fig. 4). Furthermore, MCJ significantly affects the expression levels of SNAI1 and N-cadherin, two β-catenin-downstream pro-EMT factors (Fig. 5) and MDRs mRNA levels (Fig. 6) in the chemoresistant cell line.



The Wnt/β-catenin signaling axis is a pathway that can be regulated by mitochondria in a ROS-related manner. In this regard, we analyze the ROS production and respiratory chain activity in cells overexpressing MCJ. We found that the both cell lines overexpressing MCJ show a significant increase of intracellular ROS levels (Fig. 7) and OCR (Fig. 8). However, the activity of the respiratory complexes I, II, III is increased only in MCJ overexpressing chemoresistant cell line (Fig. 9), suggesting a contribution of ETC activity in ROS production.



CONCLUSIONS

Based on these data, we suggest that MCJ may increase mitochondrial ETC activity, possibly leading to the intracellular ROS accumulation, which in turn may induce the degradation of β-catenin. The decrease in β-catenin may influence the expression levels of the main MDRs involved in cisplatin chemoresistance and EMT factors, rendering cells more responsive to the drug treatment and therefore less able to proliferate. These new findings set the bases for further investigation in order to define a possible role of MCJ in the modulation of mitochondrial function and chemoresistance in OC.

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