

The sensitizing effects to Doxorubicin of a TIM16 inhibitor in human breast cancer depend on TIM16 expression

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BACKGROUND

TIM 16, a component of the translocase complex TIM 23 of the mitochondrial inner membrane is encoded by the Magma gene. Magma was found to be over-expressed in human pituitary adenomas. Silencing Magma in ACTH secreting rat pituitary adenoma cells enhance sensitivity to pro-apoptotic stimuli. Moreover Magma overexpression protects GH secreting rat pituitary adenoma cell lines towards apoptosis. Recently we found that compound 5, a TIM16 inhibitor, is not cytotoxic but enhances the proapoptotic effects of staurosporine by reducing mitochondrial membrane potential (MMP) activation in a medullary thyroid carcinoma cell line, suggesting that compound 5 may be useful for cancer treatment in association with chemotherapeutic drugs.

AIM

Since breast cancer (BC) display high chemoresistance the aim of our study was to investigate Magma expression in human breast cancer cell lines and to verify if compound 5 could increase the effects of a chemotherapeutic agent, such as Doxorubicin in these cells.

METHODS

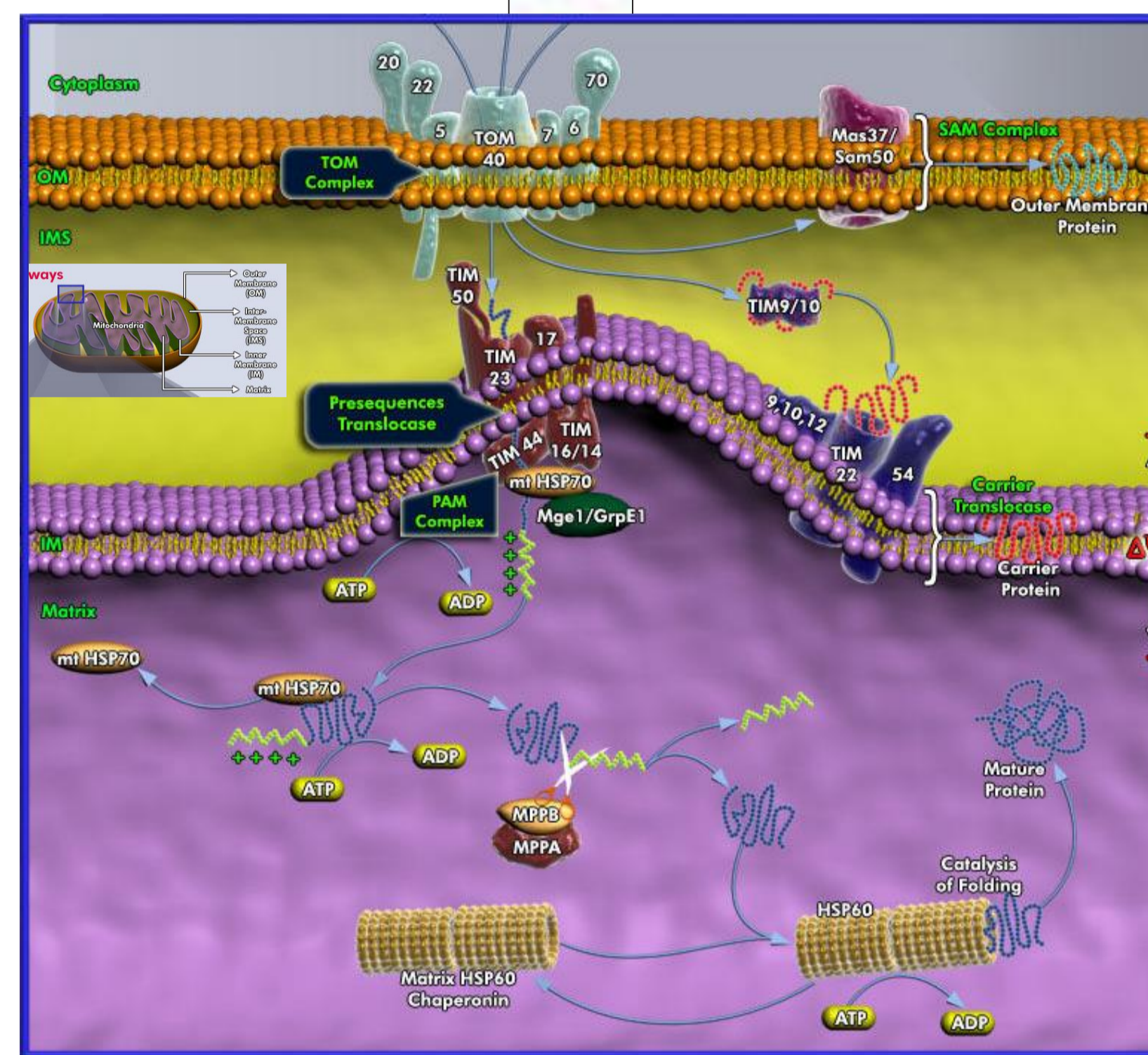
As an in-vitro model we use 3 breast tissue cell lines: MCF7 and MDA-MB231 human carcinoma cell lines and MCF12A normal breast tissue cell line.

TIM 16 expression was evaluated by western blot.

We evaluate cell viability by ATPlite assay.

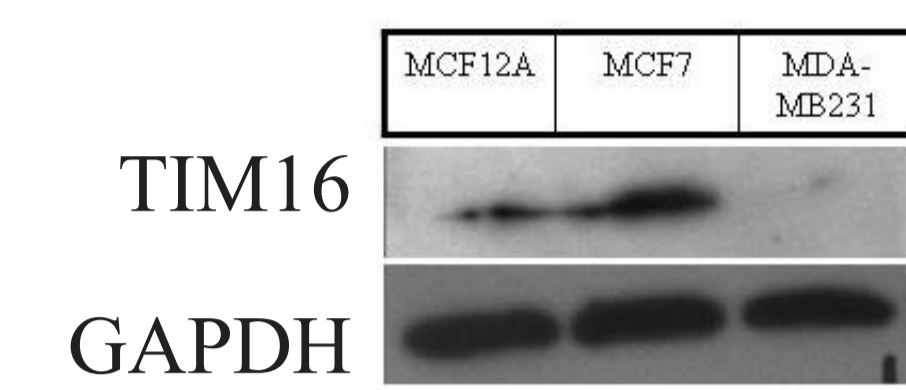
By a MitoTox Glo assay we determined mitochondrial toxicity.

To evaluate proliferation we use BrDu incorporation assay

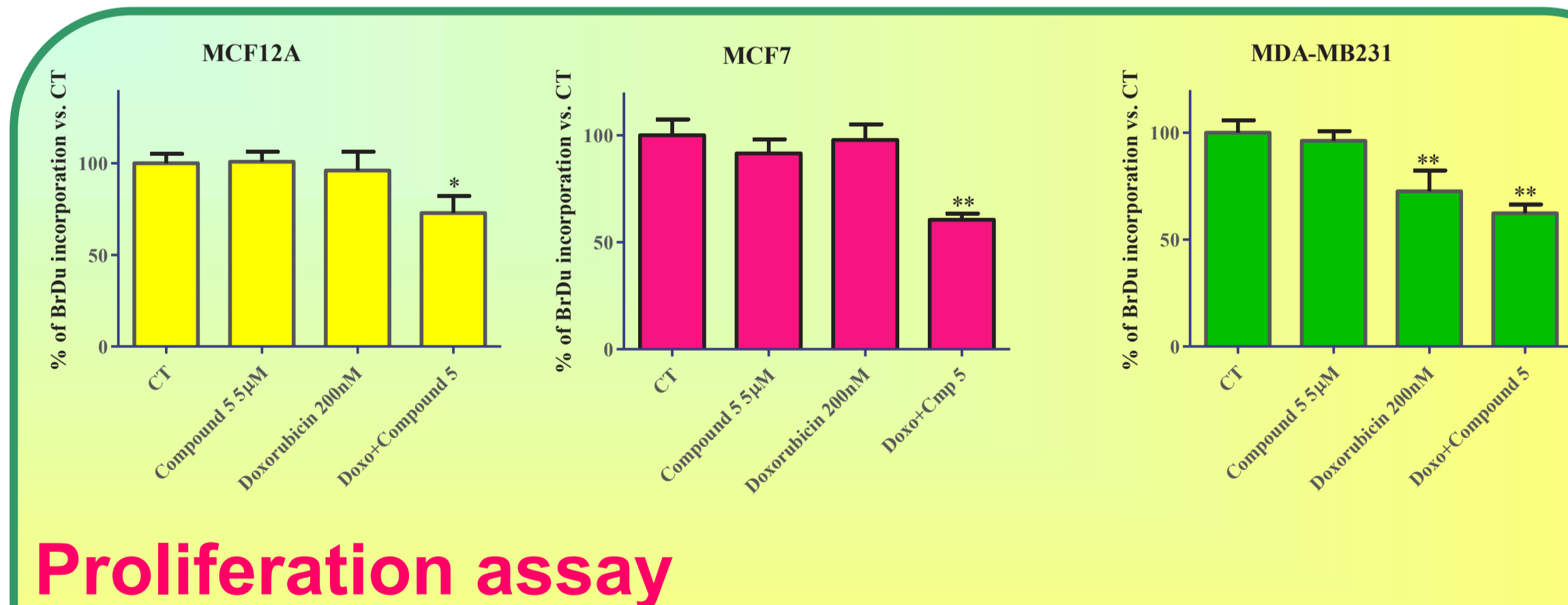
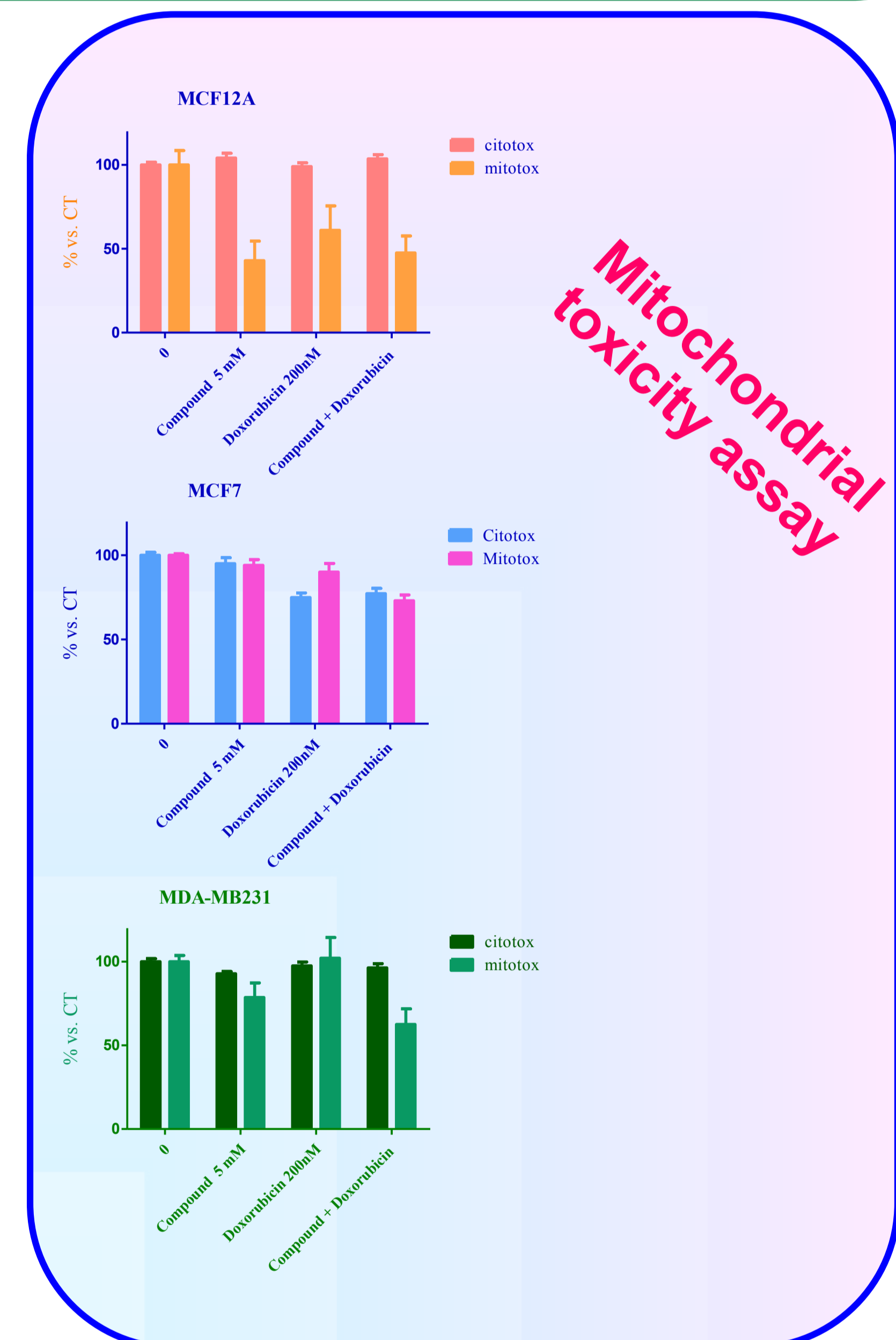
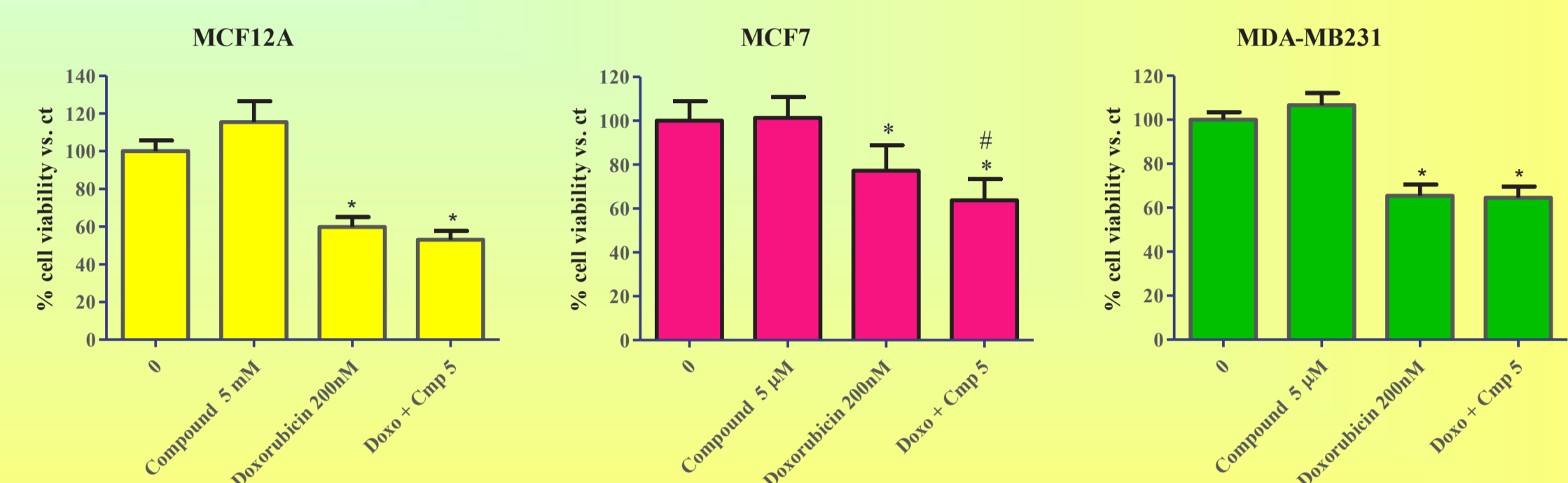


RESULTS

Tim16 protein expression



Cell viability evaluation



We found that Magma protein (Tim16) in the MCF7 cell line is highly expressed as compared to MDA-MB231 cells and MCF12A.

Our data show that treatment with compound 5 did not influence cell viability in the 3 cell lines, while Doxorubicin decreases this parameter by 20-30%.

Only in MCF7 **co-treatment with compound 5 enhances the antiproliferative effects of Doxorubicin.**

MitoTox Glo assay shows that in MCF7 cells the enhancing effects of compound 5 on Doxorubicin effects are due to mitochondrial toxicity. Finally we found that co-treatment with compound 5 and Doxorubicin in MCF7 significantly decreases BrDu incorporation, suggesting antiproliferative effects of the combination of these drugs.

Conclusion: these data suggest a role for the use of compound 5 as a sensitizing agent for chemoresistant breast cancer treatment in TIM16 over-expressing tumours.

