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جامعة الملك سعود
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Role of N-Acetylcysteine on 5-Fluorouracil Anti-Cancer Activity in Triple-Negative Breast Cancer

Aljoharah O. AlShabanah[1], Abeer M. Bin Mohareb[1], Layla A. Alkharashi[2], Qamraa H. Alqahtani[2]

[1] Undergraduate Pharm.D Student, College of Pharmacy, King Saud University, Riyadh, Saudi Arabia
[2] Department of Pharmacology and Toxicology, College of Pharmacy, King Saud University, Riyadh, Saudi Arabia

INTRODUCTION

Breast cancer (BC) is one of the most prevalent cancer types in Saudi females , and it is the first leading cause of death among women in our kingdom(1). The triple negative breast cancer (TNBC) is characterized by the absence of Estrogen (ER), Progesterone receptors (PR), and Human epidermal growth factor receptor 2 (HER2)(2)(3). TNBC is very aggressive, contain high proportion of mutagenicity and difficult to treat with chemotherapeutic drugs including 5-Fluorouracil (5-FU)(3). To overcome these limitations, we combined N-acetylcysteine (NAC), a membrane-permeable antioxidant molecule, with 5-FU to synergize 5-FU-mediated cytotoxicity(4). The effects of NAC on TNBC cell lines have previously been observed, but not on luminal cells(5). Furthermore, NAC was previously shown to decrease breast cancer (BC) cell proliferation(4). Therefore, we are aiming to investigate whether NAC exhibits additive anti-cancer effects against TNBC cells and synergize the cytotoxic activity of 5-FU.

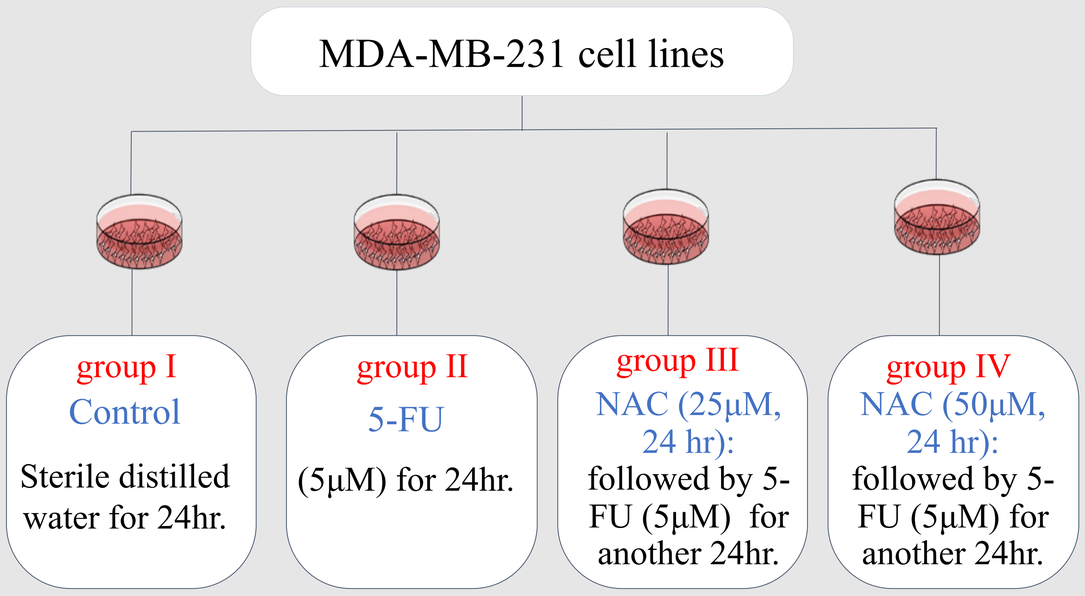
OBJECTIVES

1. Investigate the possible synergistic cytotoxic activity of NAC and 5-FU against TNBC.
2. Evaluate the efficacy of NAC as an adjunct to 5-FU on the expression of apoptotic/proliferative regulatory proteins.

METHODS

CELLS AND CELL CULTURE; MDA-MB-231 (human breast cancer) cells were cultured with RPMI medium supplemented with 10% fetal bovine serum (FBS) and 1% of Penicillin-Streptomycin in a T-75 tissue culture flask and maintained under 5% CO2 at 37°C and 95% of relative humidity.

STUDY DESIGN;



CELL VIABILITY; At the cellular level, the viability of the treated cells was evaluated using MTT assay.

IMMUNOBLOTTING NALYSIS; At the molecular level, the treated cells were subjected to immunoblotting analysis to investigate the differences in the expression level of several oncoproteins.

RESULTS

Our results showed that NAC markedly reduced the IC50 of 5-FU from 12μM to 5μM. Also, NAC potentiates the capacity of 5-FU to inhibit the expression of several oncoproteins including; NF-κB, MMP-9, p-STAT, AKT, p-AKT.

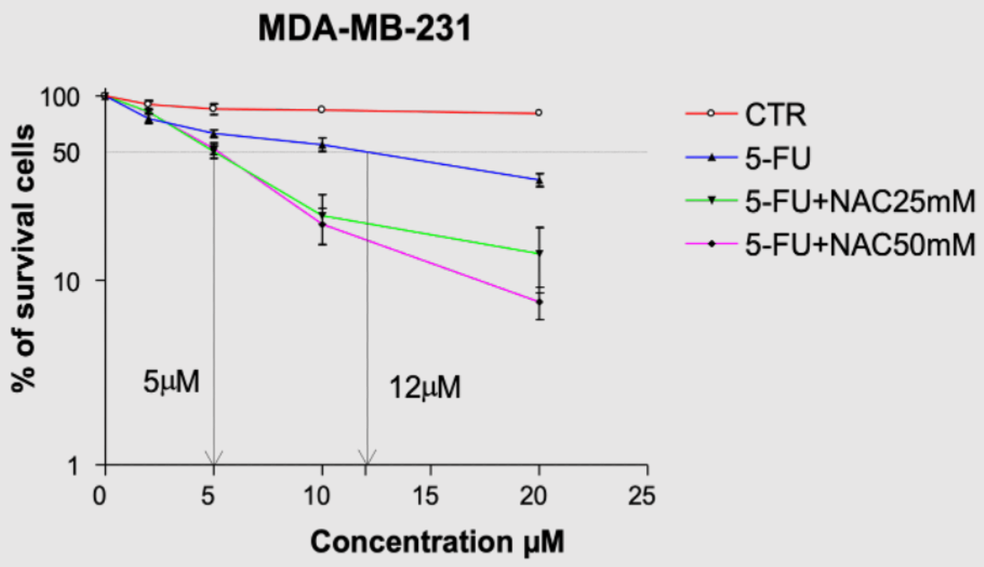


Figure 1; N-acetylcysteine (NAC) sensitizes MDA-MB-231 cells to 5-Flurouracil. MDA-MB-231 cells were treated with NAC "25& 50 mM" for 24 hrs, then the exponentially growing cells were cultured in 96 well plates and treated with the indicated concentrations of 5-FU for 48hrs.

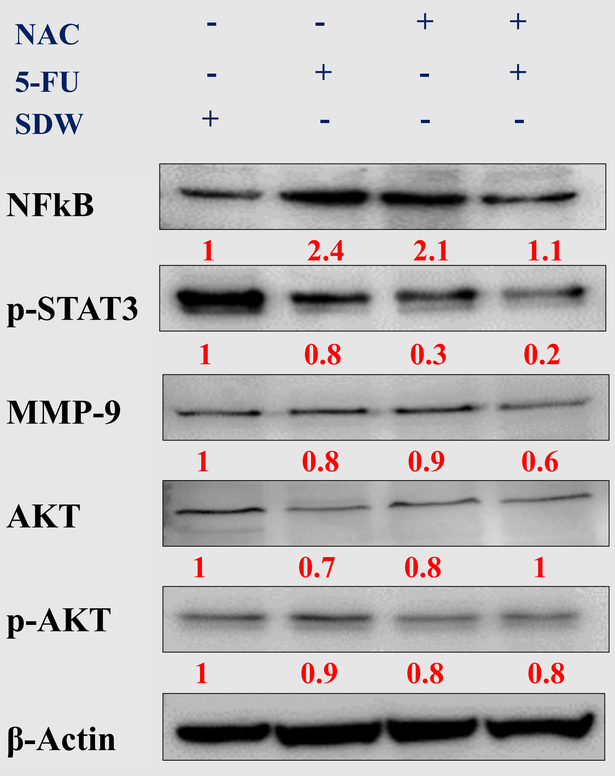


Figure 2; Combination treatment synergistically suppresses the expression of several oncoproteins. Cells were either DMSO -treated (CTR) or challenged with NAC "25mM" alone or in combination for 24 hrs prior to 5-FU (5 μM) dose. Subsequently, cells were harvested, and proteins were used for western blot analysis using the indicated antibodies. The numbers under the bands represent the corresponding expression levels as compared to time 0 and after normalization β-Actin. SDW= Sterile distilled water.

DISCUSSION AND CONCLOSIONS

In vitro, when NAC and 5-FU were pre-sequentially combined, we observed that NAC potentiated 5-FU's ability to inhibit the expression of several onco-proteins compared with 5-FU or NAC alone in TNBC.

REFERENCES

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