Effects of diallyl trisulfide-induced oxidative stress on proliferation, morphology and cell death in cancer cells

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Background

Breast cancer has become the most frequently diagnosed cancer worldwide accounting for 11.7% of new cancer diagnoses in 2020 followed by lung cancer (11.4%). Current cancer treatment strategies can have severe side effects which may result in treatment non-compliance and patient discomfort causing a shift towards phytomedicine. In this study we look at the effects of a garlic derived organosulphur compound called diallyl trisulphide (DATS) which has been shown to have antiproliferative and antimitotic effects on triple negative breast (MDA-MB-231) and lung (A549) cancer cells.

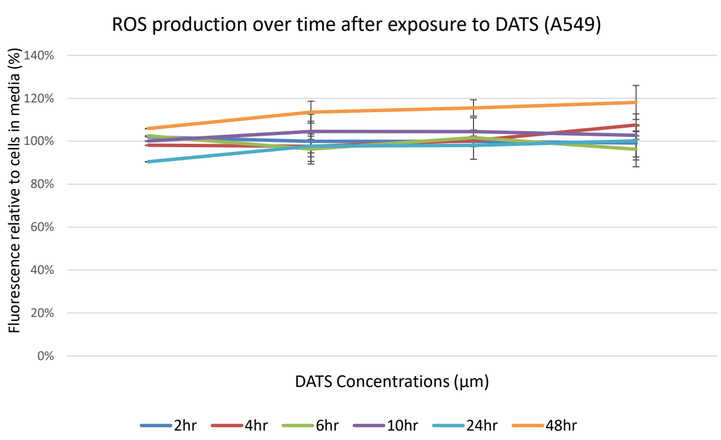
Materials and methods

The role of oxidative stress was investigated by means of a scavenger of reactive oxygen species (N-acetyl cystein (NAC)) and 6 inhibitors for specific ROS (table 1) for 24 and 48 hours in MDA-MB-231 and A549 cell lines, on proliferation (crystal violet staining and spectrophotometry), cell rounding (light microscopy), ROS production (2,7-dichlorofluoresceindiacetate (DCFDA) staining and fluorescent microscopy) and cell cycle progression and cell death induction (flow cytometry, ethanol fixation and propidium iodide staining).

Table 1: Inhibitors for specific reactive oxygen species

Reactive oxygen species	Inhibitor
Hydrogen peroxide (H_2O_2)	N, N - dimethyl thiourea (DMTU)
Hydroxyl radical (•OH)	D – Mannitol
Nitric oxide (·NO)	2-(4-Carboxyphenyl)-4,4,5,5-tetramethylimidazoline-1-oxyl-3-oxide (Carboxy-PTIO)
Peroxyl radical ($HO_2^•$)	Trolox
Singlet oxygen ($O_2^•$)	Sodium azide
Superoxide anion (O_2^-)	Tiron

Results



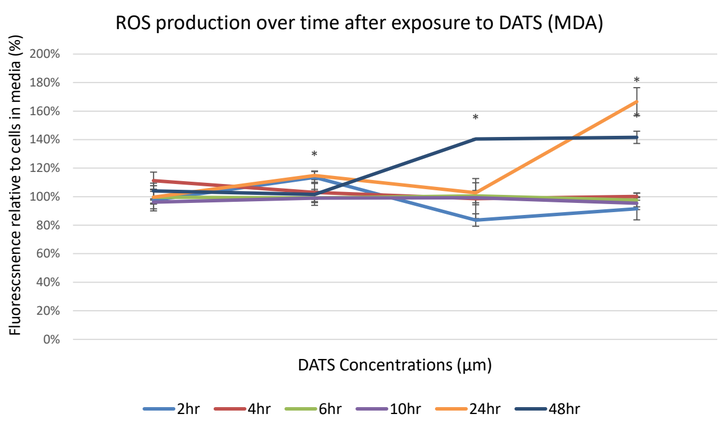


Figure 1: Fluorescent microscopy showing DCFDA staining results after exposure to DATS at various time intervals (2h, 4h, 6h, 10h, 24h, and 48h).

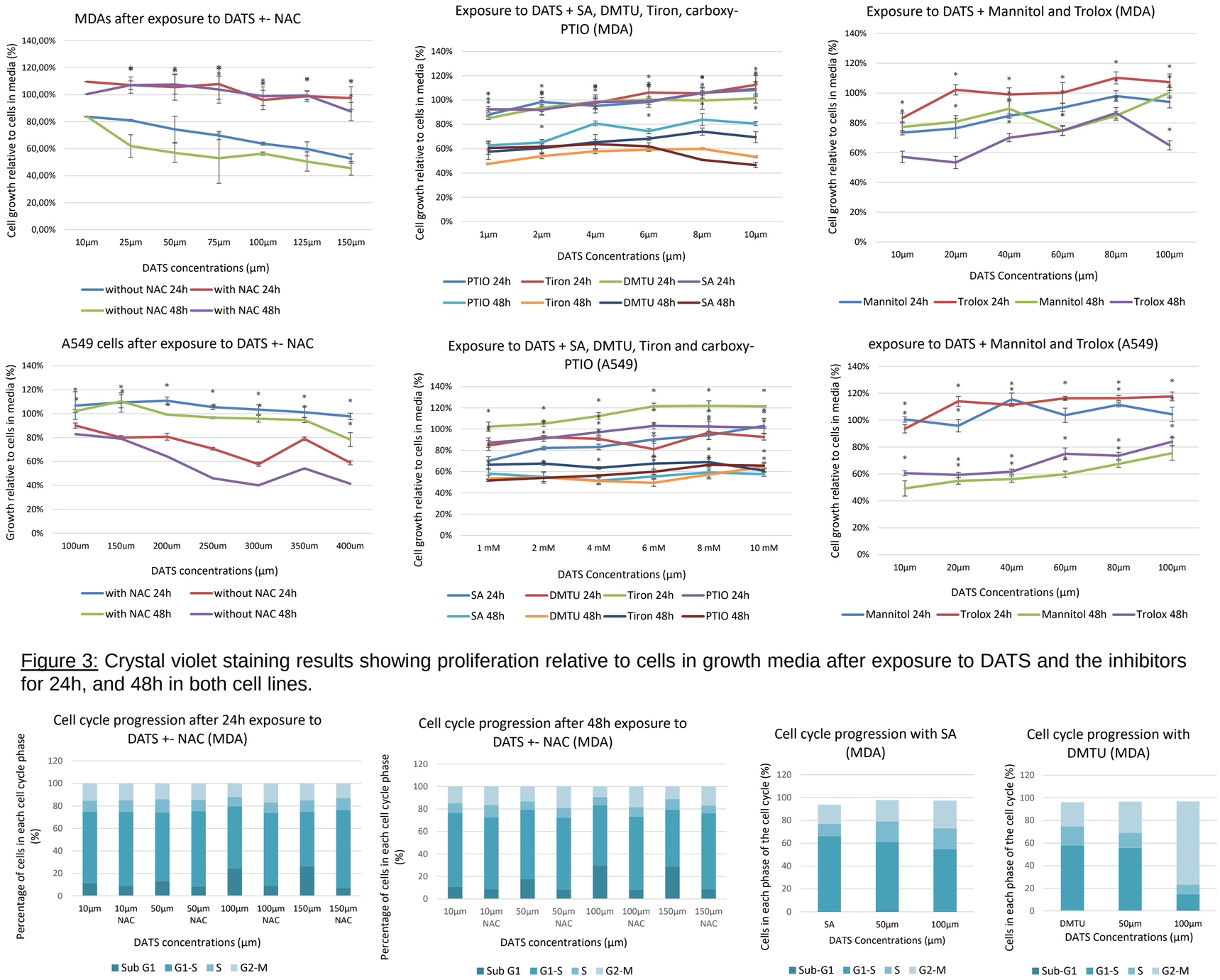
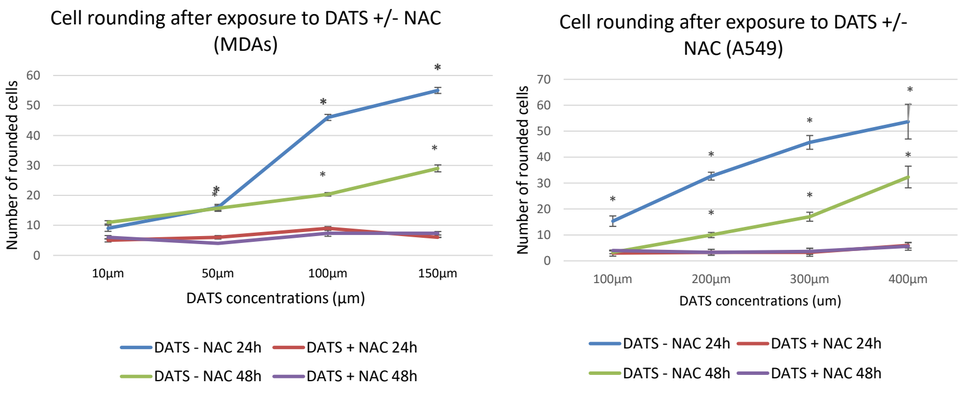


Figure 3: Crystal violet staining results showing proliferation relative to cells in growth media after exposure to DATS and the inhibitors for 24h, and 48h in both cell lines.



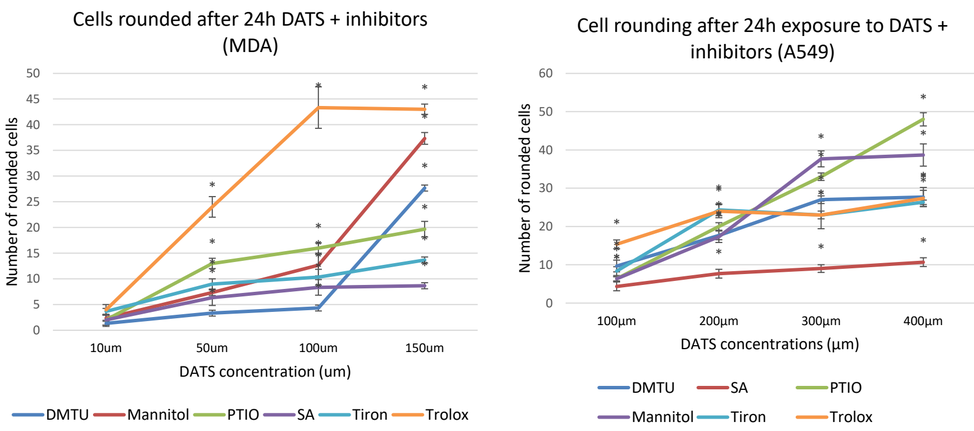


Figure 2: Light microscopy results showing the amount of cell rounding after exposure to DATS with the inhibitors for 24h, and 48h in both cell lines.

Figure 4: Flow cytometry results showing the amount of cells in each cell cycle phase after exposure to DATS with the inhibitors for 24h, and 48h in both cell lines.

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