



Antioxidant extracts from micro-algae for cardiometabolic health: an in vitro pilot study

A collaboration between UHI Active Health and Wellbeing (Prof Ian Megson, Dr Andy Treweek) and Cinoalgae



Background

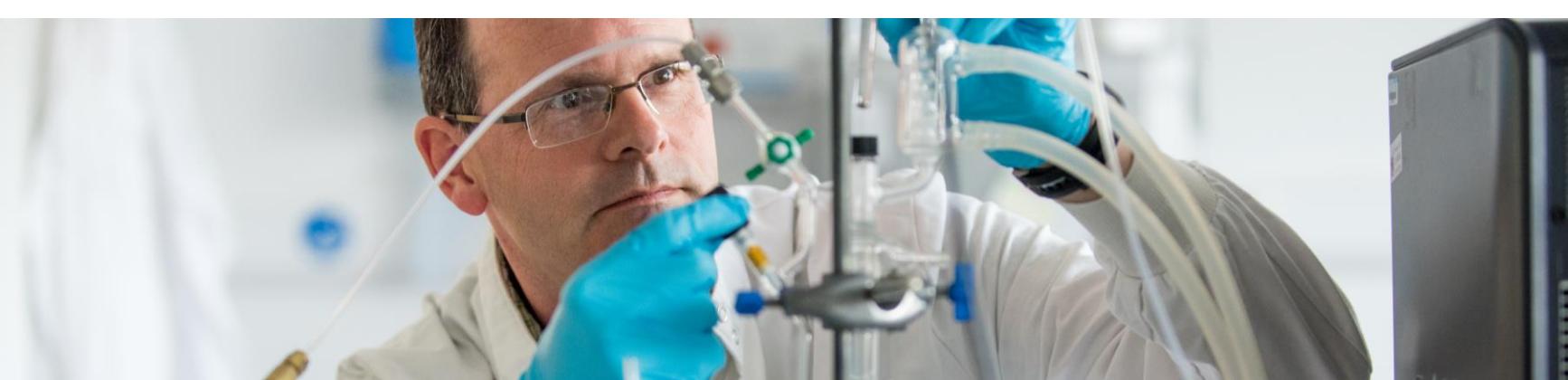
Oxidative stress is a key feature of cardiometabolic disease. Antioxidants have the potential to help reduce the impact of oxidative stress in this setting. Polyphenols are a class of antioxidants that have long been suggested for this purpose, but low absorption by the body has limited their therapeutic benefit. Nevertheless, some polyphenols have been shown to have antioxidant effects, even at the very low concentrations that match those associated with oral

availability (6). It seems that these compounds might work by stimulating the body's own antioxidant defences.

Microalgae represent a plentiful source of a wide variety of polyphenols which could have potential in this regard. By way of a forerunner to clinical studies to investigate their potential, it is first essential to use *in vitro* models to probe their efficacy.

Purpose

To test the ability of four microalgal extracts provided by Cinoalgae to protect the cells that line our blood vessels via an antioxidant effect



Methods

Four extracts were tested: *tetraselmis chair* (TSC), *nannochloropsis gaditana* (NCG), *haematococcus pluvialis* (HCP) and *phycocyanin* (PC)

Experiment 1:

Human umbilical vein endothelial cells (HUVECs) were grown for 24 h (37°C, 5% CO₂ and a humidified atmosphere).

Cells were then treated in two different ways:

- with addition of a series of extract dilutions alone (1 ng/ml, 10 ng/ml, 100 ng/ml, 1 µg/ml, 10 µg/ml and 100 µg/ml), and
- with the same extract dilutions + 200 µM pyrogallol (a potentially toxic oxidant), with appropriate controls.

Each plate was placed in the incubator for ~45 h under standard conditions prior to treatment with MTT and incubated a further 4 hours.

Absorbance was then measured to assess viability of cells. .

Experiment 2:

Under the same conditions as Experiment 1, but introduced a third treatment condition in which some cells were pre-incubated with extract dilutions for 24 h prior to treatment with pyrogallol. After the pre-incubation period, the extracts were removed and replaced with a combination of fresh extract plus pyrogallol (200 µM).

Each plate was placed in the incubator for ~45 h under standard conditions prior to treatment with MTT and incubated for a further 4 hours.

Absorbance was then measured to assess viability of cells.

Results

Experiment 1 (Figure 1):

- Phycocyanin induced a small (5-10%), but consistent and concentration-independent increase in metabolic rate.
- TSC, NCG and HCP had no effect up to a concentration of at least 1 µg/ml, whereupon they induced a noticeable reduction in metabolism, likely due to cell death.
- The superoxide generator (oxidant), pyrogallol, produced a highly consistent, sub-maximal level of suppression of metabolism (~30% reduction; dotted lines), likely due to cell death.
- None of the extracts provided protection against the pyrogallol-induced effects and NCG and HCP induced an additional suppression of metabolism at 100 µg/ml concentration.

Experiment 2 produced a similar pattern of results

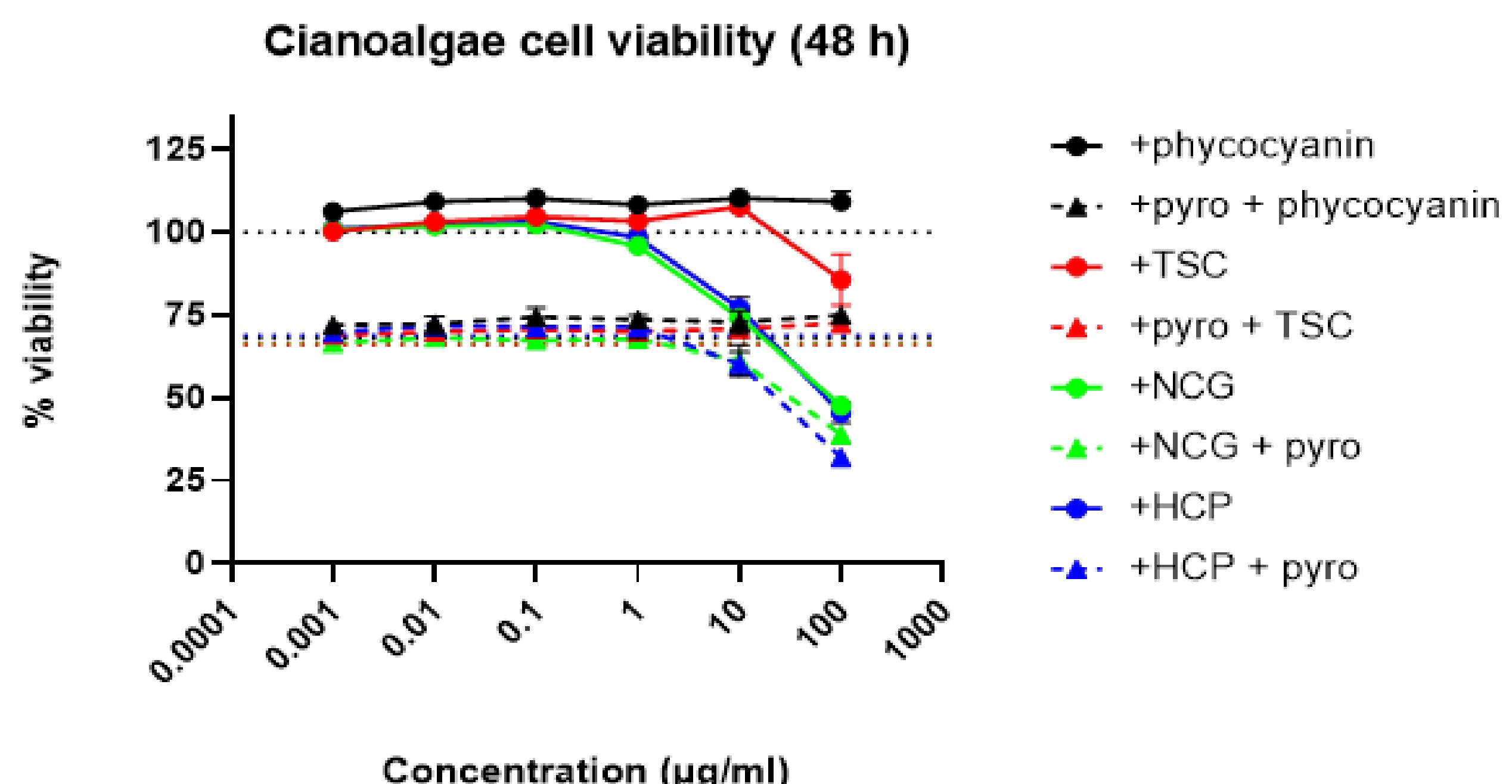


Fig 1: Effect of microalgal extracts on HUVEC cells in the presence and absence of the superoxide generator, pyrogallol. Coloured dotted lines at ~78% viability indicate the viability of cells, as measured by MTT assay, after treatment for 48 h with pyrogallol alone. Dotted lines with triangular symbols indicate the viability in cells co-cultured throughout with pyrogallol and varying concentrations of microalgal extracts. Solid lines represent the viability of cells in the presence of extracts alone (n=3 for all treatments).

Conclusion

Whilst the four extracts failed to protect against cell death or dysfunction induced by oxidative stress, phycocyanin induced an increase in cellular proliferation or metabolism which might have benefit in modulating cellular energy use

