Heliyon 8 (2022) e09192

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Research article

Cytotoxicity and ^{1}H NMR metabolomics analyses of microalgal extracts for synergistic application with Tamoxifen on breast cancer cells with reduced toxicity against Vero cells

Hanaa Ali Hussein [a,b], Murni Nur Islamiah Kassim [a], M. Maulidiani [c,**], Faridah Abas [d], Mohd Azmuddin Abdullah [a,e,*]

[a] Institute of Marine Biotechnology, Universiti Malaysia Terengganu, 21030 Kuala Nerus, Terengganu, Malaysia
[b] College of Dentistry, University of Basrah, Basrah, Iraq
[c] Faculty of Science and Marine Environment, Universiti Malaysia Terengganu, 21030 Kuala Nerus, Terengganu, Malaysia
[d] Laboratory of Natural Products, Institute of Bioscience, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia
[e] SIBCo Medical and Pharmaceuticals Sdn. Bhd., No. 2, Level 5, Jalan Tengku Ampuan Zabedah, D9/D, Seksyen 9, 40000 Shah Alam, Selangor, Malaysia

ARTICLE INFO

Keywords:
Cytotoxicity
^{1}H NMR metabolomics
Tamoxifen
Microalgal extracts
Synergistic application

ABSTRACT

This study evaluated the cytotoxic activity of Tamoxifen (TMX), an anti-estrogen drug, with microalgal crude extracts (MCEs) in single and synergistic application (TMX-MCEs) on MCF-7 and 4T1 breast cancer cells, and non-cancerous Vero cells. The MCEs of *Nannochloropsis oculata*, *Tetraselmis suecica* and *Chlorella* sp. from five different solvents (methanol, MET; ethanol, ETH; water, W; chloroform, CHL; and hexane, HEX) were developed. The TMX-MCEs-ETH and W at the 1:2 and 1:3 ratios, attained IC_{50} of 15.84–29.51 μg/mL against MCF-7; 13.8–31.62 μg/mL against 4T1; and 24.54–85.11 μg/mL against Vero cells. Higher late apoptosis was exhibited against MCF-7 by the TMX-*N. oculata*-ETH (41.15 %); and by the TMX-*T. suecica*-ETH (65.69 %) against 4T1 cells. The TMX-*T. suecica*-ETH also showed higher ADP/ATP ratios, but comparable Caspase activities to control. For Vero cells, overall apoptotic effects were lowered with synergistic application, and only early apoptosis was higher with TMX-*T. suecica*-ETH but at lower levels (29.84 %). The MCEs-W showed the presence of alanine, oleic acid, linoleic acid, lactic acid, and fumaric acid. Based on Principal Component Analysis (PCA), the spectral signals for polar solvents such as MET and ETH, were found in the same cluster, while the non-polar solvent CHL was with HEX, suggesting similar chemical profiles clustered for the same polarity. The CHL and HEX were more effective with *N. oculata* and *T. suecica* which were of the marine origin, while the ETH and MET were more effective with *Chlorella* sp., which was of the freshwater origin. The synergistic application of microalgal bioactive compounds with TMX can maintain the cytotoxicity against breast cancer cells whilst reducing the toxicity against non-cancerous Vero cells. These findings will benefit the biopharmaceutical, and functional and healthy food industries.

1. Introduction

The conventional treatments of breast cancer, as of today, face some drawbacks attributable to the associated side effects of the drugs, limited drug concentration and loss of specificity at the cancer site, and the development of chemo-resistance (Fanciullino et al., 2013; Singh et al., 2013; Hosseinia et al., 2017). Among the drugs commonly used to treat breast cancer are tamoxifen (TMX) (an anti-estrogen, hormone therapy drug), doxorubicin (a cytotoxic antibiotic), and paclitaxel (an anti-microtubule agent). These chemotherapy drugs are widely used to promote estrogen-dependent programmed breast cancer cell death (Cuzick et al., 2011). Paclitaxel causes mitotic arrest by stabilizing the cellular microtubule elements. Doxorubicin is used in a combination regimen where the cytotoxic activities have been attributed to free radical mechanism, lipid peroxidation, and direct membrane effects (Anjum et al., 2017). TMX or trans-1-[p-b-(dimethyl-amino) ethoxy-phenyl]1,2-diphenyl-1-butene is a substituted trans-isomer of triphenylethylene (Morrow and Jordan, 2000; Jordan, 2006), and a common

* Corresponding author.
** Corresponding author.
E-mail addresses: maulidiani@umt.edu.my (M. Maulidiani), azmuddin@sibcogroup.net, joule1602@gmail.com (M.A. Abdullah).

https://doi.org/10.1016/j.heliyon.2022.e09192
Received 29 September 2021; Received in revised form 22 January 2022; Accepted 22 March 2022