SOCIETÀ ITALIANA DI FARMACOLOGIA

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TIPOLOGIA DI BORSA RICEVUTA: Borsa per soggiorno all'estero

TIPOLOGIA DI RELAZIONE: relazione finale

TITOLO DELLA RELAZIONE: Protective effects of nutraceuticals in cell models of neurodegenerative damage: roles of HSF/HSPs and NRF2/ARE pathways.

RELAZIONE:

Cellular stress is the basis of a dose-dependent continuum of responses leading to adaptive health or pathogenesis. The cell ability to counteract stressful conditions, known as cellular stress response, requires the activation of pro-survival pathways and the production of molecules with anti-oxidant as well as anti-apoptotic or pro-apoptotic activities. Among the cellular pathways conferring protection against oxidative stress, a key role is played by vitagenes, which include heat shock proteins (Hsps) heme oxygenase-1 and Hsp70, sirtuins and thioredoxin/thioredoxin reductase system (1). Transcriptional, translational, and post-translational regulation of cellular stress response occur with the intervention of heat shock transcription factors (HSFs) normally expressed and maintained in an inactive state. Heat shock response contributes to establish a cytoprotective state in a wide variety of human diseases, including inflammation, cancer, aging and neurodegenerative disorders. Given the broad cytoprotective properties of the heat shock response, there is now strong interest in discovering and developing pharmacological agents capable of inducing stress responses (2-4). Moreover, the strong evidence that the vitagene network operates as a defense system in the brain during oxidative and nitrosative stress open new perspectives in the treatment of neurodegenerative disorders. Two central regulators, nuclear factor-erythroid 2 p45-related factor 2 (NRF2) and heat shock factor 1 (HSF1), control the NRF2/ARE pathway and the heat shock response, two essential cellular defense mechanisms (5). Both systems are highly inducible under conditions of stress. Many small molecules, including

certain phytochemicals, such as isothiocyanates and phenylpropanoids, and/or their metabolites, have the capacity to induce the NRF2/ARE pathway. Celastrol, withaferin A, gedunin, curcumin, and sulforaphane are examples of structurally diverse phytochemicals for which protective effects have been documented in numerous animal models of human disease and which include induction of large networks of transcriptional programs regulated by transcription factors NRF2 and HSF1.

Nutritional manipulation of endogenous cellular defense mechanisms may represents an innovative approach for the prevention or treatment of neurodegeneration and propose novel potential therapeutic strategies relying upon the simultaneous activation of cytoprotective genes of the cell life program and down-regulation of proinflammatory and pro-oxidative genes involved in programmed cell death (6).

These pathways represent a promising drug target for various compounds with protective effects in neurodegenerative diseases. *Citrus* fruits and their juices are the main food sources of flavonoids and have been extensively studied for their cardiovascular, anti-inflammatory and anti-cancer activities. In recent years, scientific interest toward *Citrus bergamia* (bergamot) has gained ground. We have reported the anti-cancer properties of bergamot juice (BJ) in different *in vitro* and *in vivo* models, and proposed the flavonoid fraction of BJ (BJe) to be responsible for this action (7-10). We have also shown that low concentrations of BJe reduce the LPS-induced inflammatory response in THP-1 cells through SIRT1-mediated NF-κB inhibition (11), and exerts anti-inflammatory effects *in vivo* (12). In addition, we studied the ability of BJe to reduce β-amyloid-induced pro-inflammatory response in THP-1 cells through a mechanism involving both MAPKs and AP-1 pathways (13).

Based on these evidences, the main goal of my research was to assess the ability of BJe and its major flavonoids to modulate the cellular stress response in an *in vitro* model of neurodegeneration.

During the first part of the research I evaluated the ability of BJe and flavonoids to activate both NRF2 and HSF1 pathways in two different cell lines (Hepa-1c1c7 and MDA-MB 231 cells).

The second part of my research period was focused on the set up of the *in vitro* model of neurodegeneration. To this aim, I used primary cultures of *wild-type* and NRF2-*knockout* astrocytes exposed or not to menadione in presence/absence of BJe or flavonoids.

Preliminary results show as BJe could have some effects on the investigated pathways and only some of tested flavonoids appear to be effectiveness.

This project is currently in progress, and the ongoing experiments will provide further prompt on the potential neuroprotective efficacy of BJe, clarifying what flavonoids are responsible for modulating the cellular cytoprotection pathways.

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