

Evaluation of some synthetic chemical compounds against Caspase-3 by luciferase whole-cell biosensor

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ABSTRACT

Excessive amount of apoptosis unleashes undesirable amounts of special cysteine proteases known as caspases in normal cells, and this can lead to the emergence of neurodegenerative diseases. Activated Caspase-3 as an executioner caspase is considered a key indicator of initiation of apoptosis. Thus identification of compounds that can affect Caspase-3 activity is of significant importance. Drugs able to activate or inhibit apoptosis through modulation of Caspase-3 activity, exemplify such compounds and can be potential candidates in treatment of particular apoptosis associated diseases. In this study HEK293 cell line engineered to stably express luciferase, acts as a whole-cell biosensor to screen a number of potential drug compounds against Caspase-3. Concentration- and time-dependent experiments revealed the appropriate working concentration for each compound using death curves obtained from MTT assay. Luciferase activity is measured following stable HEK293 co-treatment with Doxorubicin as an apoptosis inducer and the drug compounds, eventually the difference in luminescent signals show how each compound affects Caspase-3. Also post-treatment morphological features of stable HEK293 cells compared to HEK293 cells as control, indicate how efficiently each compound acts. In order to achieve more confident results, Caspase-3 activity was assayed in cell extracts using Caspase-3 activity assay kit. Here we show that the stable luciferase cell line can indicate whether a compound inhibits or stimulates Caspase-3.

Keywords: Luciferase; Apoptosis; Caspase-3 inhibitors; Drug-protein interaction