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College of Pharmacy Research Day

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Oxytocin Protects PC12 Cells Against β-Amyloid-Induced Cell Injury

INTRODUCTION

Alzheimer's disease (AD) is the most withering brain disorders among elderly population. AD lead to disturbances in the brain affecting the memory and language, visuospatial orientation and higher executive function. In fact, the primary cause of AD is an accumulation of abnormal disposition of amyloid-beta (Aβ) in brain tissue. Therefore, elimination of Aβ considered as an important role in AD treatment. The underlying mechanisms of Aβ-induced neurotoxicity include generation of reactive oxidative species (ROS), inflammation, excitotoxicity, and neurons loss. Antioxidants have shown a beneficial effect in neurodegenerative disorders treatment. The neuropeptide Oxytocin (OT) is known as antioxidant agent has been demonstrated to have a neuroprotective effect, anti-oxidant, anti-apoptotic, and anti-inflammatory effects in vivo and in vitro.

OBJECTIVES

- Investigating the protective effect of oxytocin on PC12 cells against β-amyloid-induced cell injury.
- Investigating the protective effect of oxytocin against β-amyloid-induced oxidative stress
- Investigating the protective effect of oxytocin against β-amyloid-induced changes in mitochondrial membrane potential

METHODS

- **Cell Culture**

Using neurons-like Pheochromocytoma cell line 12 (PC12**),** pre-treated with OT for 2hr followed by Aβ for 48hr.

- **MTT Assay**

measured cell proliferation and viability.

- **Measurement of Intracellular ROS Level**

The reactive oxygen species (ROS) level measured by DCFDA assay.

- **Measurement of Mitochondrial Membrane Potential (ΔΨM)**

The Mitochondrial Membrane Potential measured by JC-1 dye as an Indicator of early apoptosis.

RESULTS

Using PC12 cells, first we tested the cytotoxicity of Aβ25–35 (Most common active compound) on PC12 cells through MTT assay. As shown in Fig 1A cells exposed to different concentrations of Aβ25–35 for 48 h showed notable decline in the cell viability in a concentration dependent manner. Therefore, we found that OT pre-treatment significantly and in a dose dependent manner inhibited the Aβ toxicity effect. Furthermore, OT remarkably reduced the elevation of ROS level induced by Aβ. Furthermore, OT showed marked attenuation of early apoptosis.

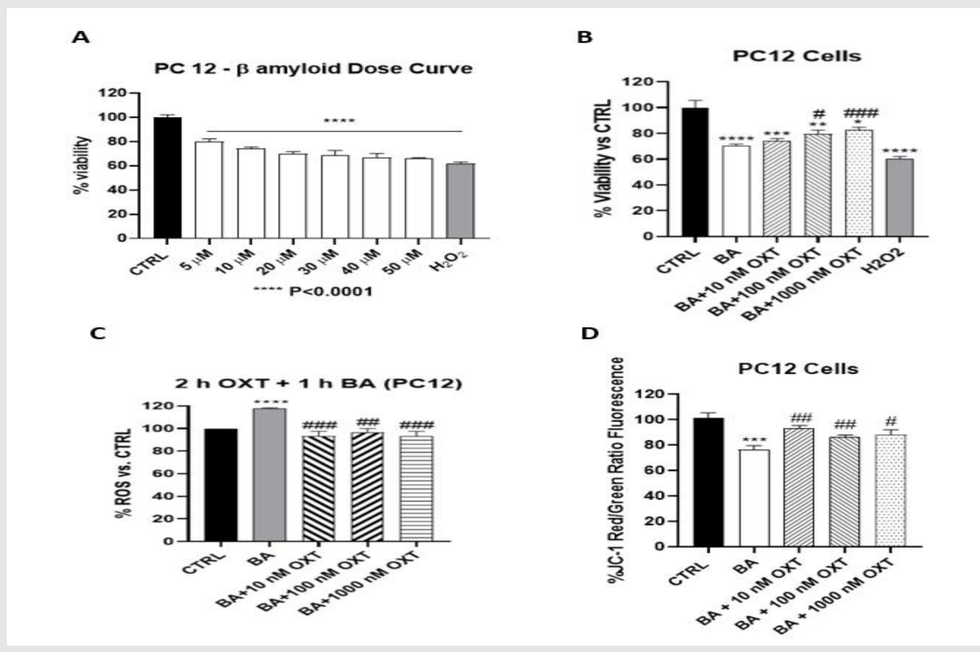

Fig. 1. Oxytocin in concentration dependently strangled the Aβ25-35-induced cell injury in PC12 cells. (A) Cells were treated with Aβ25-35 (5 – 50 μM) for 48 h and cell viability was measured by using the MTT assay. (B) Cells were pre-treated with oxytocin 2 h and then incubated with 20 μM of Aβ25-35 for a 48 h and cell viability were measured by MTT assay. (C) Oxytocin reduced the oxidative stress in PC12 cells that induced by Aβ25-35. After pre-treatment with oxytocin for 2 h, PC12 cells were incubated with 20 μM of Aβ25-35 for another 48 h. (D) Oxytocin attenuated PC12 loss of mitochondrial membrane potential (△ψm) induced by Aβ25-35. After pre-treatment with oxytocin for 2 h, PC12 cells were incubated with 20 μM Aβ25-35 for another 48 h, (△ψm) was confirmed by the JC-1 assay.

CONCLOSIONS

OT shows a protective effect against Aβ-induced cytotoxicity and oxidative stress that contributed to increase the cell proliferation and viability. Based on our findings, OT might serve as a potential therapeutic agent for treating AD and other neurodegenerative diseases.

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