C. Nday, et all. Journal of Agroalimentary Processes and Technologies, Volume XIII, No.1 (2007), 217-220
Communication – Food Science

INVESTIGATION OF NEUROTOXIC EFFECTS OF Al(III) ON NEURONAL AND GLIAL CELLS THROUGH N-METHYL D-ASPARTIC ACID RECEPTORS

Christiane Nday[1], Alison J. Drysdale[2], Bettina Platt[2], A. Salifoglou[1]
[1] Department of Chemical Engineering, Laboratory of Inorganic Chemistry, Aristotle University of Thessaloniki, Thessaloniki 54124, Greece
[2] School of Medical Sciences, College of Life Sciences and Medicine, University of Aberdeen, Institute of Medical Sciences, Foresterhill,Aberdeen AB25 2ZD Scotland, UK.

Abstract

N-methyl D-aspartic acid (NMDA) is one of the five known stimulating amino acid receptor categories, which consist of chemical regulators of Ca(II) channels on neurocellular membranes, playing an important role in memory process. What kind of metallotoxin effects, however, can affect the NMDA function of neuronal cells? We investigated the potential biological activity of well-characterized Al(III) forms in neuronal and glial cellular environment. This effort constitutes a challenge, because of the neurotoxic potential of the metal interacting with the cellular receptors and the evidence linking aluminum to Alzheimer Disease.
***Keywords*:** *N-methyl D-spartic acid, Alzheimer, auminum, neurons*

Introduction

N-methyl D-aspartic acid (NMDA) is one of the five known stimulating amino acid receptor categories, which consists of chemical regulators of Ca(II) channels on neurocellular membranes, playing an important role in memory process. What kind of metallotoxins effects, however, can influence NMDA function of neurons? Elevated intracellular levels of Al have pleiotropic effects including disruption of calcium homeostasis (Flaten, 2001), which probably affect early onset in Alzheimer Disease (McLachlan, 1984).

Alzheimer's disease (AD), as the most common type of dementia, is a neurodegenerative disease characterized by progressive cognitive deterioration (Berne, 1996), together with declining activities of daily

living, neuropsychiatric symptoms or behavioral changes and progressive loss of memory.

The investigation of the potential biological activity of well-characterized Al(III) forms in neuronal and glial cellular environment constitutes a challenge, because of the neurotoxic potential of the metal interacting with the cellular receptors and the epidemiological evidence linking aluminum to Alzheimer Disease.

Experimental

In the course of this research, neuronal and glial cell cultures of neonate Prague-Dawley rats were used for the experiment. Also, two novel of Al(III) compounds: Aluminium citrate $K_4[Al(C_6H_4O_7)(C_6H_5O_7)]\cdot 4H_2O$ and Aluminium quinate $K[Al(C_7H_{11}O_6)_3]\cdot(OH)\cdot 5H_2O$ at concentration of:

a) 10 μM,
b) 100 μM,
c) 500 μM or
d) the combination of 10, 100, 500 μM.

The cells were incubated for one hour with Fura-2 AM at room temperature in the dark. Fura-2 is a ratiometric fluorescent dye, which binds to free intracellular calcium (Grynkiewicz, 1985). Fura-2 is excited at 340 nm and 380 nm of light, and the ratio of the emissions at those wavelengths is directly correlated to the amount of intracellular calcium.

The samples were run to the FURA-2 AM Ca(II) IMAGING system by perfusing before and after each Al(III) exposure to the employed concentrations, 15 μM NMDA for 2 minutes. Interval perfusion media for 10 minutes were applied before and after all concentrations for washing (figure 1).

After statistical analysis, we found the results from table 1. It can be observed that more than 60% of the neurons respond to the first and the second NMDA application, but there was no significant response of neurons to Al(III) exposure during the one hour of the experiment, without influencing the hippocampal cell viability or the last NMDA response of neurons.

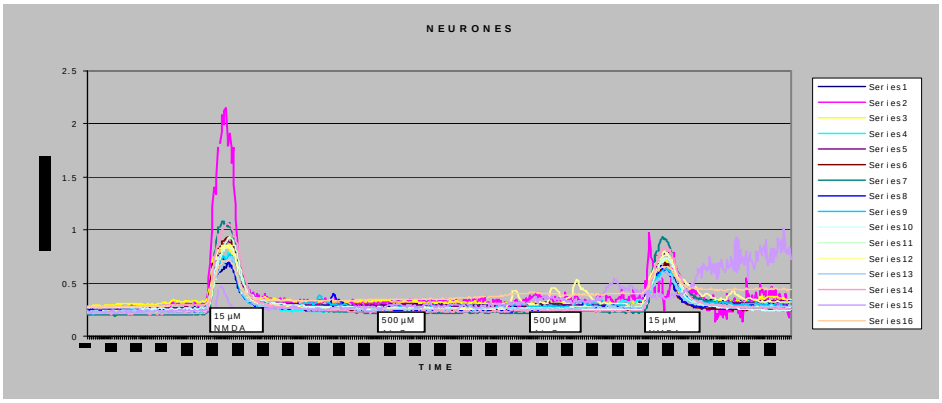

Figure 1. Variation of optical density

Table 1. Neurons responders in NMDA receptors and in Al(III) compounds

AC[1]	**67%** responders in **NMDA 15 µM** 3**%** from 67% responders applied also to **Al(III) 10 µM**
	65% responders in **NMDA 15 µM** **5%** from 65% responders applied also to **Al(III) 100 µM**
	58% responders in **NMDA 15 µM** **4%** from 58% responders applied also to **Al(III) 500 µM**
	60% responders in **NMDA 15 µM** **5%** from 60% responders applied also to **Al(III) 10, 100, 500 µM**
AQ[2]	**70%** responders in **NMDA 15 µM** **6%** from 70% responders applied also to **Al(III) 10 µM**
	66% responders in **NMDA 15 µM** No responder to **Al(III) 100 µM**
	65% responders in **NMDA 15 µM** **5%** from 65% responders applied also to **Al(III) 500 µM**
	69% responders in **NMDA 15 µM** **7%** from 69% responders applied also to **Al(III) 10, 100, 500 µM**

[1]AC = Aluminium citrate
[2]AQ = Aluminium quinate

Conclusions

After statistical analysis, we found that more than 60% of the neurons respond to the first and the second NMDA application, but there was no significant response of neurons to Al(III) exposure during the one hour of the experiment, without influencing the hippocampal cell viability or the last NMDA response of neurons. Also, the neuroapoptotic effect of the metal ion Al(III) is time dependent. No specific role emerged for the physiological ligands of Al(III) compounds, with quite same results for Aluminum citrate and Aluminum quinate, too. As expected, there was no response of glial cells.

Acknowledgments

The present study is co-funded by European Union-European Social Fund and National fund PYTHAGORAS-EPEAEK II. The authors would also like to acknowledge the financial support to this project by a "PENED" grant from the General Secretariat of Research and Technology, Greece.

References

Berne, R.M., Levy M.N. (1996). *Principles of Physiology*. Second Edition, Publisher: C.V. Mosby.

Flaten, T.P. (2001). *Brain Research Bulletin*, 55, 187-196.

Grynkiewicz, G., Poenie, M. Tsien, R.Y. (1985). *J.Biol.Chem.*, 260, 3440-3450.

McLachlan, D.R.C. (1984). *Environmetrics*, 6, 233-275.