Send Orders for Reprints to reprints@benthamscience.ae

Current Aging Science, 2016, *9*, 279-283

RESEARCH ARTICLE

S100b Induces Expression of Myoglobin in APβ Treated Neuronal Cells *In Vitro*: A Possible Neuroprotective Mechanism

Maria E. Clementi[1,*], Beatrice Sampaolese[1] and Bruno Giardina[1,2]

[1]*CNR-ICRM Institute of "Chimica del Riconoscimento Molecolare", c/o Institute of Biochemistry and Clinical Biochemistry, Catholic University School of Medicine Largo F. Vito 1, 00168 Rome, Italy;* [2]*Institute of Biochemistry and Clinical Biochemistry, Catholic University School of Medicine, Largo F. Vito 1, 00168 Rome, Italy*

Maria E. Clementi

Abstract: ***Background*:** In this study, human neuroblastoma cells (IMR32) treated with Amyloid Beta Peptide (APβ), were used as model to evaluate the molecular basis of protective role of S100b, a neurotrophic factor and neuronal survival protein, highly expressed by reactive astrocytes close to amyloid deposition in the cortex of Alzheimer's patients. The aim of this work is to value the effect of S100b on ROS production in cells treated with Amyloid Beta Peptide and the subsequent influence on globin gene expression.

***Method*:** In this study we investigated the effect of S100b on ROS production and on globin gene expression in human neuroblastoma cells (IMR32) treated with Amyloid Beta Peptide (APβ).

***Results*:** Our results have shown that at nanomolar concentrations, S100b protects cells against APβ mediated cytotoxicity and the protective mechanism could be related, almost in part, to the control of ROS production through an over expression of Myoglobin gene.

***Conclusion*:** In light of our results, we speculate that over-expression of the Myoglobin gene could be read as a possible attempt of the cell to increase the scavengers of reactive oxygen species (ROS).

ARTICLE HISTORY

Received: November 26, 2015
Revised: January 26, 2016
Accepted: February 19, 2016

DOI:
10.2174/1874609809666160222112850

Keywords: S100b, oxidative stress, myoglobin.

1. INTRODUCTION

Alzheimer's disease (AD) is one of the most common neurodegenerative disease characterized by the accumulation in the brain of patients, due to misfolding of the protein Amyloid Beta (APβ), a peptide of 42 amino acids. Another important pathogenetic event in AD is the relevant oxidative stress; it is well known in fact that APβ can insert into the lipid bilayer and initiate lipid peroxidation and oxidative damage to proteins and other molecules.

In this complex pathogenetic network recent observations have shown that, during the early stages of AD, microglia produces numerous pro-inflammatory molecules (such as IL1 and S100b) whose expression is highly modulated by reactive astrocytes in close proximity of beta-amyloid deposits; in fact our recent results showed that S100b exerts a protective role interfering with some APβ-mediated apoptotic events [1]. In present study we examined the effect of S100b on IMR32, a human neuroblastoma cellular line, treated with Amyloid beta peptide as model of Alzheimer Disease. In particular, we investigated the role of this neuroprotective effector on ROS production and on expression of some globin gene expression with particular regard to neuroglobin (NGb), myoglobin (Mb) and cytoglobin (Cygb). The choice of the three globins, particularly neuroglobin, has been suggested by recent experimental works where emerges their neuroprotective role against the damage to neurons and brain as they work as scavengers of reactive oxygen species (ROS), and as molecules involved in signal transduction neuroprotective pathways.

In this short paper, we reported preliminary but relevant data, evidencing as S100b defends neuron by ROS production induced by beta amyloid peptide, and increases the expression of Mb but not of Ngb and Cygb. We have assumed that this effect may be considered a defensive attempt of the neurons by using the Mb as a scavenger of reactive oxygen species and speculate that, in a similar manner to hemoglobin [2], Myoglobin can bind to APβ to enhance its aggregation.

2. MATERIALS AND METHODS

2.1. Cellular Culture and Treatments

Human neuroblastoma IMR32 cells were grown in minimum essential medium supplemented with 10% heat inactivated fetal bovine serum, 100 IU/ml penicillin, and 100 μg/ml streptomycin, and cultured at 37°C in an atmosphere

*Address correspondence to this author at the CNR-ICRM Institute of "Chimica del Riconoscimento Molecolare", c/o Institute of Biochemistry and Clinical Biochemistry, Catholic University Medical School, Largo F. Vito 1, 00168 Rome, Italy; Tel: ++39 6 30154215; Fax: ++39 6 30154309; E-mail: e.clementi@icrm.cnr.it

1874-6098/16 $58.00+.00

Send Orders for Reprints to reprints@benthamscience.ae

Current Aging Science, 2016, *9*, 279-283

RESEARCH ARTICLE

S100b Induces Expression of Myoglobin in APβ Treated Neuronal Cells *In Vitro*: A Possible Neuroprotective Mechanism

Maria E. Clementi[1,*], Beatrice Sampaolese[1] and Bruno Giardina[1,2]

[1]*CNR-ICRM Institute of "Chimica del Riconoscimento Molecolare", c/o Institute of Biochemistry and Clinical Biochemistry, Catholic University School of Medicine Largo F. Vito 1, 00168 Rome, Italy;* [2]*Institute of Biochemistry and Clinical Biochemistry, Catholic University School of Medicine, Largo F. Vito 1, 00168 Rome, Italy*

Maria E. Clementi

Abstract: ***Background*:** In this study, human neuroblastoma cells (IMR32) treated with Amyloid Beta Peptide (APβ), were used as model to evaluate the molecular basis of protective role of S100b, a neurotrophic factor and neuronal survival protein, highly expressed by reactive astrocytes close to amyloid deposition in the cortex of Alzheimer's patients. The aim of this work is to value the effect of S100b on ROS production in cells treated with Amyloid Beta Peptide and the subsequent influence on globin gene expression.

***Method*:** In this study we investigated the effect of S100b on ROS production and on globin gene expression in human neuroblastoma cells (IMR32) treated with Amyloid Beta Peptide (APβ).

***Results*:** Our results have shown that at nanomolar concentrations, S100b protects cells against APβ mediated cytotoxicity and the protective mechanism could be related, almost in part, to the control of ROS production through an over expression of Myoglobin gene.

***Conclusion*:** In light of our results, we speculate that over-expression of the Myoglobin gene could be read as a possible attempt of the cell to increase the scavengers of reactive oxygen species (ROS).

ARTICLE HISTORY

Received: November 26, 2015
Revised: January 26, 2016
Accepted: February 19, 2016

DOI:
10.2174/1874609809666160222112850

Keywords: S100b, oxidative stress, myoglobin.

1. INTRODUCTION

Alzheimer's disease (AD) is one of the most common neurodegenerative disease characterized by the accumulation in the brain of patients, due to misfolding of the protein Amyloid Beta (APβ), a peptide of 42 amino acids. Another important pathogenetic event in AD is the relevant oxidative stress; it is well known in fact that APβ can insert into the lipid bilayer and initiate lipid peroxidation and oxidative damage to proteins and other molecules.

In this complex pathogenetic network recent observations have shown that, during the early stages of AD, microglia produces numerous pro-inflammatory molecules (such as IL1 and S100b) whose expression is highly modulated by reactive astrocytes in close proximity of beta-amyloid deposits; in fact our recent results showed that S100b exerts a protective role interfering with some APβ-mediated apoptotic events [1]. In present study we examined the effect of S100b on IMR32, a human neuroblastoma cellular line, treated with Amyloid beta peptide as model of Alzheimer Disease. In particular, we investigated the role of this neuroprotective effector on ROS production and on expression of some globin gene expression with particular regard to neuroglobin (NGb), myoglobin (Mb) and cytoglobin (Cygb). The choice of the three globins, particularly neuroglobin, has been suggested by recent experimental works where emerges their neuroprotective role against the damage to neurons and brain as they work as scavengers of reactive oxygen species (ROS), and as molecules involved in signal transduction neuroprotective pathways.

In this short paper, we reported preliminary but relevant data, evidencing as S100b defends neuron by ROS production induced by beta amyloid peptide, and increases the expression of Mb but not of Ngb and Cygb. We have assumed that this effect may be considered a defensive attempt of the neurons by using the Mb as a scavenger of reactive oxygen species and speculate that, in a similar manner to hemoglobin [2], Myoglobin can bind to APβ to enhance its aggregation.

2. MATERIALS AND METHODS

2.1. Cellular Culture and Treatments

Human neuroblastoma IMR32 cells were grown in minimum essential medium supplemented with 10% heat inactivated fetal bovine serum, 100 IU/ml penicillin, and 100 μg/ml streptomycin, and cultured at 37°C in an atmosphere

*Address correspondence to this author at the CNR-ICRM Institute of "Chimica del Riconoscimento Molecolare", c/o Institute of Biochemistry and Clinical Biochemistry, Catholic University Medical School, Largo F. Vito 1, 00168 Rome, Italy; Tel: ++39 6 30154215; Fax: ++39 6 30154309; E-mail: e.clementi@icrm.cnr.it

1874-6098/16 $58.00+.00

of 5% CO2 in air. Cell differentiation was induced by 1 mM dibutyryl cAMP and 2.5 μM 5-bromodeoxyuridine (Sigma, St. Louis, MO, USA), which were added to the culture medium three times per week, starting from the day after plating. After a week, the differentiated cells were plated at an appropriate density according to each experimental procedure (25.000 cells for ROS detection and 1000000 cells for RNA isolation).

Stock solution of APβ peptide was solubilised in DMSO according to the manufacturer's instructions and stored at −80 °C. In the previous studies [3] these conditions have been shown to lead to the predominance of the soluble monomeric form of this peptide. Human recombinant S100b was solved in minimum essential medium, filtered and stored at –20 °C. S100b (5 nM) was added to the cultures 12 hr before exposure to APβ (10 μM). The cells were then incubated for 24 h before different experimental procedures. In all experiments, DMSO was added to cell cultures at the same concentrations present in the beta amyloid peptide solutions. The concentrations of APβ peptide and S100b are chosen by previous time-course studies (data not shown).

2.2. Detection of ROS

The detection of ROS was performed after staining cells with DCFDA Cellular ROS Detection Assay Kit. Briefly, cells treated with different experimental conditions, were grown in 96-well microplate with 25,000 cells per well and treated successively with 2',7'–dichlorofluorescein diacetate (DCFDA) that is converted, by oxidation, to the fluorescent molecular DCF. DCF was then quantified using a CytoFluor Multi-well Plate Reader, with 485 nm excitation and 538 nm emission filters.

2.3. RNA Isolation and Semi-quantitative PCR

Total RNA was isolated with PureLink RNA mini Kit (Life technologies) and the elimination of any genomic DNA was performed by on-column DNAse treatment. RNA concentration was evaluated by spectrophotometric reading at 280 and 260 nm. Total RNA was used for first strand cDNA synthesis with SCRIPT cDNA Synthesis Kit and Oligo-dT, as random primer (Jena Bioscience). The PCR was performed according to the manufacturer's instructions. The following primer sequences were used for amplification:

GAPDH forward 5'-AACGGATTTGGTCGTATTG-3',

GAPDH reverse 5'-GGAAGATGGTGATGGGATT-3' (208 bp);

Myoglobin forward 5'-GGGTCTGATCTCGTGTAGCC-3',

Myoglobin reverse 5'-CCAAACCATGCAGAACACAG-3' (199 bp);

Neuroglobin forward 5'-CAGGAAGGTGATGCTCGTGA-3',

Neuroglobin reverse 5'-GTAGAGTTGGCTCCAGGCAG-3' (217 bp) and

Cytoglobin forward 5'- CCAACTGCGAGGACGTG-3',

Cytoglobin reverse 5'-ACTGGCTGAAGTACTGCT-3'(81 bp)

PCR products were then analysed by 1.5 % agarose gels electrophoresis. Image acquisition from gels and quantitative elaboration was carried out by Bio-Rad Quantity One so ware. Band density of PCRs have been developed in relatic in percentage, with that of the housekeeping gene consider as control.

2.4. Myoglobin Preparation and Cell Treatment

Horse heart Myoglobin was dissolved in phosphate bu ered saline (PBS, 100 mM, pH 7.4), reduced by adding s dium dithionite and purified on a Sephadex G-25 colum The concentration of the ferrous Myoglobin (Mb) generat was calculated from the absorbance of the Soret band at 4 nm). Mb solution was filtered through 0,2 mm and diluted essential medium supplemented at the opportune concentr tions. The Mb solution was added to cellular cultures at di ferent concentrations (0.5, 1, 5 and 10 μM) simultaneous with beta amyloid peptide (10 μM). The ROS measuremen were performed 24 h afterward.

2.5. Statistical Analysis

The data were analysed by one-way ANOVA, followe by post hoc Newman–Keul test for multiple comparison among group means, using a Prism TM computer prograr (Graph-Pad, San Diego, CA, USA), and differences wer considered statistically significant if $P < 0.01$. All results ar presented as the mean ± S.E.M. of at least eight differer experiments.

3. RESULTS

Starting from our recent paper [1] where the effect ex erted by S100b on modulation of apoptotic genes in IMR3 treated with beta amyloid peptide was evidenced, in this con test we investigated the role of stress oxidative in this mode of Alzheimer Disease. Since reactive oxygen species produc tion has been strongly implicated in the patho-physiology o this neurodegenerative disorders, in this paper the RO measurement is reported for IMR32 in different experimenta conditions. It is evident, see Fig. (**1**) that exposure of neuroblastoma cells with beta amyloid peptide (10 μM) signifi-

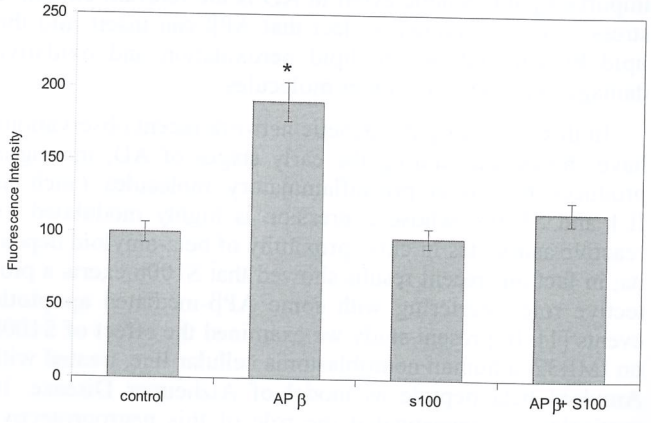


Fig. (1). ROS production (expressed as Fluorescence Intensity) in IMR32 cells treated with beta amyloid peptide (APβ), with S100b and with both S100b and APβ. Human recombinant S100b (5 nM) was added to the cultures 12 hr before exposure to APβ (10 μM) and successively all the cells were incubated for 24h before the ROS detection. All values indicate means ± S.E. of eight independent experiments: significantly different from cells untreated: *$P < 0.01$.

cantly ($P < 0.01$) increases (of 90% ± 10%) production of ROS whereas a 12 hr of pretreatment with S100b (5 nM) defends the cells by oxidation processes showing ROS values similar to the control. To note that the treatment with S100b alone does not modify the control values. Because the expression of Neuroglobin, Myoglobin and Cytoglobin genes in neurons is likely to act as a scavenger of reactive species, as well as a signal transduction molecule involved in neuroprotective pathways, we investigated the expression of these three globins in neuroblastoma cells treated with S100b and beta amyloid peptide. As evident (see Fig. **2**) neither Ngb and Cygb are expressed (panel B and C respectively) while the expression of Myoglobin (panel D) seems to be length to the presence of S100b. In particular, while the RNA for Mb is blank for cells treated with beta amyloid peptide alone, such as in the controls, the presence of S100b stimulates significantly ($P < 0.01$) the expression of this gene reaching an increase of 100% (± 10%). To value the effective role of Mb on reactive oxygen species production, we tested the effect of different concentrations of this hemoprotein on IMR32 treated with beta amyloid peptide (see Fig. **3**). As is evident, treatment with Myoglobin keeps ROS values lower than cells treated with Amyloid beta peptide alone in a dependent-dose manner, reaching a significant protective effect to a 5 μM. It must also underline that higher concentrations of Mb are toxic on cell culture (data not shown), according to literature data [4].

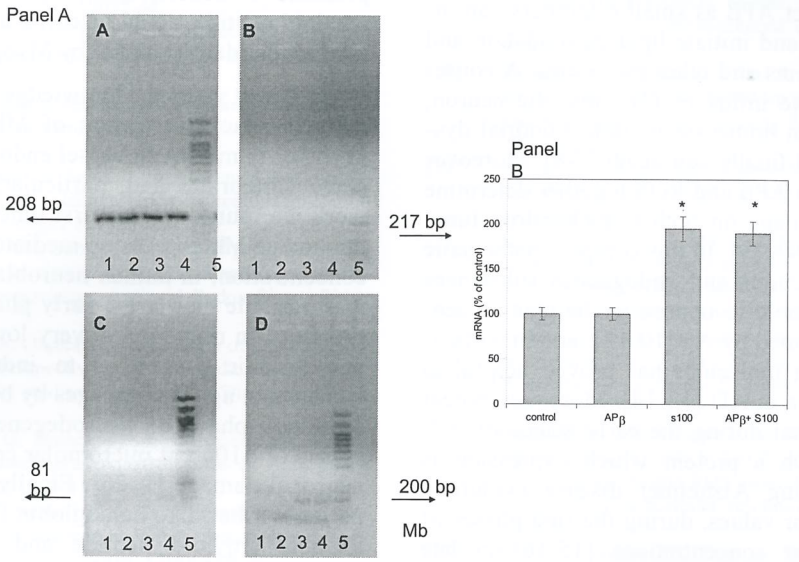


Fig. (2). Panel **A**. Agarose gels representing mRNA expression levels for GAPDH (**A**), Ngb (**B**), Cyt (**C**) and Mb (**D**) genes in IMR32 cells. Legend of lines: 1) Cell untreated, 2) Cells treated with APβ, 3) Cells treated with S100b, 4) Cells treated with APβ and S100b, 5) markers. In detail: human recombinant S100b (5 nM) was added to the cultures 12 hr before exposure to APβ (10 μM). All the cells were incubated, after treatments for 24h before RNA isolation. Panel **B**. The density of the gel bands for Mb was divided by that of the GADPH (housekeeping gene) and expressed as percent of the control band density. Results are from eight independent experiments. Significantly different from controls *$P < 0.01$.

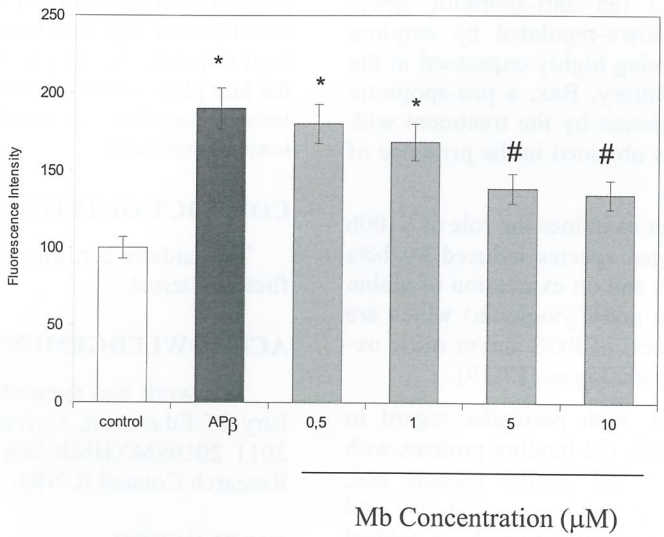


Fig. (3). ROS production (expressed as Fluorescence Intensity) in IMR32 cells treated with APβ (10 μM) and in the presence of different concentrations of Myoglobin (0.5,1,5 and 10 μM). The Mb solution was added to cellular cultures simultaneously with APβ and incubated for 24 h before the ROS measurement. All values indicate means ± S.E. of eight independent experiments: significantly different from cells untreated: *$P < 0.01$ and respect to cells treated with APβ alone #$P < 0.01$.

4. DISCUSSION

Alzheimer's disease (AD) is an irreversible neurodegenerative disorder affecting mainly individuals over the age of 65. It is characterized clinically by progressive dementia and histopathologically, by the presence of extracellular deposits of-senile plaques consisting of amyloid fibrils, intracellularly neurofibrillary tangles and neuronal cell loss [5]. The molecular mechanisms of AD has been identified as a protein misfolding disease due to the accumulation of abnormally folded amyloid beta protein (APβ), a peptide of 1-42 amino-acids, which deposits in dense formations known as senile plaques or neuritic plaque in the brain.

Another important pathogenetic event in AD is a relevant oxidative stress [6]; in fact APβ as small oligomers can insert into the lipid bilayer and initiate lipid peroxidation and oxidative damage to proteins and other molecules. A consequence of this event is the influx of Ca^{2+} into the neuron, resulting in the loss of ion homeostasis, mitochondrial dysfunction, synapse loss and finally cell death [7-8]. Moreover it is well evidenced which APβ and ROS together determine an intracellular direct damage on both mitochondrial function and mitochondrial DNA [9]. In this complex pathogenic network, numerous exogenous and endogenous substances are been individuated as able to oppose to the neurodegenerative mechanisms generated by Aβ [10-12]; and in particular the use of antioxidant molecules has proved helpful to slow the onset of dementia in AD [13-14]. Moreover, recent observations evidenced that during the early stages of AD, microglia produces S100b a protein which expression is carefully modulated during Alzheimer disease evolution, increasing from nanomolar values, during the first phases of pathology, to micromolar concentrations [15-16] in late phases of AD.

The role of S100b in AD was investigated in our recent study where we showed that this neurotrophic factor protects, at nanomolar concentration, cells against APβ mediated cytotoxicity. The protective mechanism seems to be related to the effect on bcl-2 (an anti-apoptotic gene) expression, which is highly down-regulated by amyloid beta peptide treatment, while being highly expressed in the presence of S100b. On the contrary, Bax, a pro-apoptotic gene, resulted in to down-regulation by the treatment with S100 as compared to the results obtained in the presence of APβ.

In this light, the present paper examines the role of S100b on production of reactive oxygen species induced by beta amyloid peptide on IMR32 cells and on expression of globin genes (Neuroglobin, Myoglobin and Cytoglobin) which are considered as inducible scavengers of ROS and/or nitric oxide (NO) in neurodegenerative pathologies [17-19].

Mammalian globins in fact, with particular regard to Neuroglobin are hypoxia-inducible O2-binding proteins with neuroprotective effects in cell and animal models and, though were identified firstly in relation to hypoxic and ischemic injury [20], subsequent investigations have pointed to a possible connection with Alzheimer's disease [21]. The mechanism underlying heme-protein protective action is unknown, although several possibilities have been proposed, suggesting an oxygen storage similar to myoglobin in muscles; however, the low concentrations (in micromolar range) of globin expression in brain tissues suggests a more persuasive role of scavengers by free radicals [22].

Our results evidenced that the presence of S100b at nanomolar concentration, reduces ROS production in cells treated with beta amyloid peptide which, according to previous report, induces a significant presence of reactive oxygen species [23]. The study of the possible correlation between ROS production and the expression of heme-globin RNA, highlighted that neither Neuroglobin and Cytoglobin are inducible in this model of AD, while Myoglobin appears modulated by the presence of S100b. Hence the lower ROS presence evidenced in cells treated with S100b and beta amyloid peptide, could be correlated with an increase in cellular antioxidant potential by Myoglobin.

In recent years the knowledge on the extra muscular and extra cardiac localization of Mb has been evidenced by Myoglobin mRNA in vessel endothelium, liver and brain of some vertebrates and, particularly, in fishes [24]. In this work we found, for the first time, a new mechanism of induction of Mb expression mediated by S100b, at nanomolar concentration, in human neuroblastoma cellular line. Thus, it is possible that in the early phase of AD, when S100b is produced in microglia at very low values, the effect of this neurotrophic protein is to induce protective molecular mechanism against damages by beta amyloid. Instead in the advanced phase of neurodegenerative pathology, the increase of S100b at micromolar concentration contributes to neuronal damage [1, 25]. Finally, it is possible also to hypothesize that, like hemoglobin [2, 19] Mb could also bind to beta amyloid peptide and co-localizers in amyloid plaques.

CONCLUSION

In the light of the present study, we would like to propose the following scenario: in the early phases of AD, S100b produces very low values which could induce Mb synthesis, thereby reducing able to reduce the oxidant injury and perhaps sequestering Amyloid Beta Peptide molecules. While in the late phases both S100b and Mb at more elevated concentration contribute to accelerate the aggregation and neurotoxic phenomena.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

ACKNOWLEDGEMENTS

This work was supported by grants from the Italian Ministry of Education, University, and Research (PRIN 2010-2011 20109MXHMR-006 to B.G.) and by Italian National Research Council (CNR).

DISCLOSURE

"Part of this article has been previously published in Advances in Alzheimer's Disease Vol. 2 No. 3 (2013), Article ID:37285, 10 pages DOI:10.4236/aad.2013.23013".

PATIENT'S CONSENT

Declared None.

REFERENCES

[1] Clementi ME, Sampaolese B, Triggiani D, Tiezzi A, Giardina B. S100b protects IMR-32 cells against Aβ(1-42) induced neurotoxicity *via* modulation of apoptotic genes expression. Adv Alzheimer's Dis 2013; 2(3): 99-108.

[2] Chuang JY, Lee CW, Shih YH, Yang T, Yu L, Kuo YM. Interactions between amyloid-β and hemoglobin: implications for amyloid plaque formation in Alzheimer's disease. PLoS One 2012; 7(3): 1-10.

[3] Boland K, Behrens M, Choi D, Manias K, Perlmutter DH. The serpin-enzyme complex receptor recognizes soluble, nontoxic amyloid-beta peptide but not aggregated, cytotoxic amyloid-beta peptide. J Biol Chem 1996; 271(30): 18032-44.

[4] D'Agnillo F, Alayash AI. A role for the myoglobin redox cycle in the induction of endothelial cell apoptosis. Free Radic Biol Med 2002; 33(8): 1153-64.

[5] Selkoe DJ. Alzheimer's disease results from the cerebral accumulation and cytotoxicity of amyloid beta-protein. J Alzheimers Dis 2001; 3(1): 75-80.

[6] Miller E, Morel A, Saso L, Saluk J. Isoprostanes and neuroprostanes as biomarkers of oxidative stress in neurodegenerative diseases. Oxid Med Cell Longev 2014; 2014: 1-10.

[7] Butterfield A, Swomley AM, Sultana R. Amyloid β-peptide 1-42-induced oxidative stress in alzheimer disease: Importance in disease pathogenesis and progression. Antioxid Redox Signal 2013; 19(8): 823-35.

[8] Sultana R, Butterfield DA. Oxidative modification of brain proteins in Alzheimer's disease: Perspective on future studies based on results of redox proteomics studies. J Alzheimers Dis 2013; 33(1): 243-51.

[9] Ajith TA, Padmajanair G. Mitochondrial pharmaceutics: A new therapeutic strategy to ameliorate oxidative stress in Alzheimer's disease. Curr Aging Sci 2015; 8(3): 235-40.

[10] Ansari N, Khodagholi F. Natural products as promising drug candidates for the treatment of Alzheimer's disease: molecular mechanism aspect. Curr Neuropharmacol 2013; 11(4): 414-29.

[11] Soosanabadi M, Bayat H, Kamali K, *et al.* Association study of IL-4 -590 C/T and DDX39B -22 G/C polymorphisms with the risk of late-onset Alzheimer's disease in Iranian Population. Curr Aging Sci 2015; 8(3): 276-81.

[12] Wang WY, Tan MS, Yu JT, Tan L. Role of pro-inflammatory cytokines released from microglia in Alzheimer's disease. Ann Transl Med 2015; 3(10): 1-15.

[13] de Oliveira BF, Veloso CA, Nogueira-Machado JA, Martins CM. High doses of *in vitro* beta-carotene, alpha-tocopherol and ascorbic acid induce oxidative stress and secretion of IL-6 in peripheral blood mononuclear cells from healthy donors. Curr Aging Sci 2012; 5(2): 148-56.

[14] Çoban J, Bingül I, Yesil-Mizrak K, Dogru-Abbasoglu S, Oztezcan S, Uysal M. Effects of carnosine plus vitamin E and betaine treatments on oxidative stress in some tissues of aged rats. Curr Aging Sci 2013; 6(2): 199-205.

[15] Jesse S, Steinacker P, Cepek L, *et al.* Glial fibrillary acidic protein and protein S-100B: different concentration pattern of glial proteins in cerebrospinal fluid of patients with Alzheimer's disease and Creutzfeldt-Jakob disease. J Alzheimers Dis 2009; 17(3): 541-51.

[16] Chaves ML, Camozzato AL, Ferreira ED, *et al.* Serum levels of S100B and NSE proteins in Alzheimer's disease patients. J Neuroinflammation 2010; 7(6): 1-6.

[17] Fordel E, Thijs L, Martinet W, *et al.* Neuroglobin and cytoglobin overexpression protects human SH-SY5Y neuroblastoma cells against oxidative stress-induced cell death. Neurosci Lett 2006; 410(2): 146-51.

[18] Sirangelo I, Iannuzzi C, Vilasi S, *et al.* W7FW14F apomyoglobin amyloid aggregates-mediated apoptosis is due to oxidative stress and AKT inactivation caused by Ras and Rac. J Cell Physiol 2009; 221(2): 412-23.

[19] Haines BA, Davis DA, Zykovich A, *et al.* Comparative protein interactomics of neuroglobin and myoglobin. J Neurochem 2012; 123(1): 192-8.

[20] Sun Y, Jin K, Peel A, Mao XO, Xie L, Greenberg DA. Neuroglobin protects the brain from experimental stroke *in vivo*. Proc Natl Acad Sci USA 2003; 100(6): 3497-500.

[21] Chen LM, Xiong YS, Kong FL, *et al.* Neuroglobin attenuates Alzheimer-like tau hyperphosphorylation by activating Akt signaling. J Neurochem 2012; 120(1): 157-64.

[22] Watanabe S, Takahashi N, Uchida H, Wakasugi K. Human neuroglobin functions as an oxidative stress-responsive sensor for neuroprotection. J Biol Chem 2012; 287(36): 30128-38.

[23] Jiao Y, Zhang Y, Wei Y, Liu Z, An W, Guo M. Direct observation of internalization and ROS generation of amyloid β-peptide in neuronal cells at subcellular resolution. Chembiochem 2012; 13(16): 2335-8.

[24] Helbo S, Weber RE, Fago A. Expression patterns and adaptive functional diversity of vertebrate myoglobins. Biochim Biophys Acta 2013; 1834(9): 1832-9.

[25] Mori T, Koyama N, Arendash GW, *et al.* Overexpression of human S100b exacerbates cerebral amyloidosis and gliosis in the Tg2576 mouse model of Alzheimer's disease. Glia 2010; 58(3): 300-14.

ISSN: 1874-6098
eISSN: 1874-6128

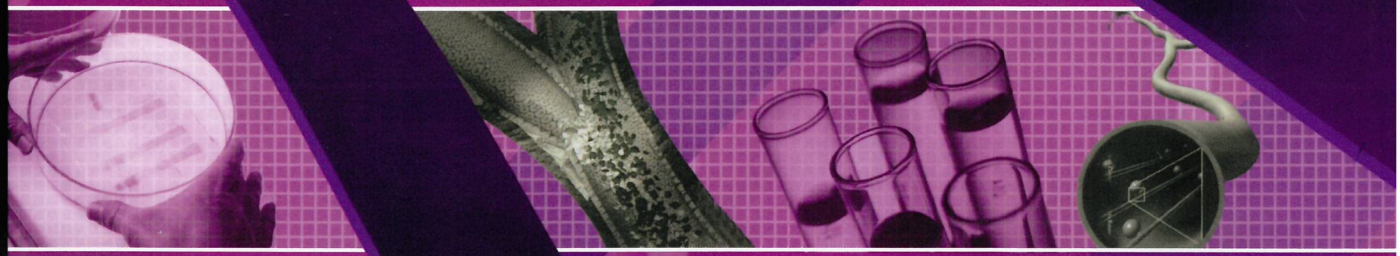
Volume 9, Number 4

Current Aging Science

S100b Induces Expression of Myoglobin in APβ Treated Neuronal Cells *In Vitro*: A Possible Neuroprotective Mechanism

Maria E. Clementi[1,*], Beatrice Sampaolese[1] and Bruno Giardina[1,2]

[1]CNR-ICRM Institute of "Chimica del Riconoscimento Molecolare", c/o Institute of Biochemistry and Clinical Biochemistry, Catholic University School of Medicine Largo F. Vito 1, 00168 Rome, Italy; [2]Institute of Biochemistry and Clinical Biochemistry, Catholic University School of Medicine, Largo F. Vito 1, 00168 Rome, Italy



BENTHAM SCIENCE