EFFECTS OF CLONIDINE ON HIPPOCAMPAL SYNAPTIC ENERGY METABOLISM: A FUNCTIONAL PROTEOMIC STUDY

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The anti-hypertensive Clonidine binds to pre-synaptic 2-adrenoreceptors and inhibits the release of norepinephrine. The liposoluble Clonidine crosses the blood-brain barrier producing many central pharmacological effects and some studies showed that 2 agonists improve the morphologic and functional outcome after experimental ischemia (Gupta & Sharma, 2014; Hoffman et al., 1991; Zhang et al., 2004), also preventing memory deficits and the lowering of acetylcholinesterase activity in Vascular Dementia. Nevertheless, Brede et al. (2011) failed to demonstrate a neuroprotective effect of Clonidine in a rat ischemic model and further studies are recommended.

In this study, the effects of acute Clonidine treatment (5 g x kg-1, 30 minutes) were assessed on the hippocampus of male Sprague-Dawley rats aged 3 months. The maximal rate (Vmax) of regulatory energy-linked enzyme activities (Functional Proteomics) of (a) Glycolysis, (b) Krebs' cycle, (c) Glutamate and related amino acids metabolism and (d) acetylcholine catabolism were evaluated as a measure of hippocampal energy transduction potentialities (Villa et al., 2016). Because of the heterogeneity of hippocampal synapses (Villa et al., 2013), this research was performed on (a) the "large" synaptosomes (LS), derived from glutamatergic mossy fiber endings connecting granule cells of dentate gyrus with apical dendrites of CA3 pyramidal cells, and (b) the "small" synaptosomes (SS), derived from the cholinergic small nerve endings of septo-hippocampal fibers, whose projections reach CA1 pyramidal cells.

In control animals, enzyme activities varied in a complex way respect to the type of synaptosomes. This micro-heterogeneity is an important factor, confirming that the different types of synapses possess specific and independent metabolic features coherently with the selective vulnerability to noxious stimuli and to pharmacological treatment of the respective hippocampal subfields (Ferrari et al., 2015; Moretti et al., 2015; Villa et al., 2013). In fact, Clonidine acute treatment modified differently some enzyme activities: (a) in LS, glutamate dehydrogenase, glutamate-oxaloacetate-transaminase and acetylcholinesterase activities were decreased by the drug; (b) in SS, the activities of phosphofructokinase and lactate dehydrogenase were increased, while citrate synthase and glutamate-pyruvate transaminase were decreased.

These diversified effects of Clonidine on LS and SS are the consequence of the different metabolic features of synaptosomes derived from specific hippocampal subfields. Moreover, Clonidine is also agonist of receptors other than 2 (Wang et al., 2005), and the different modification in the energy transduction potentialities are likely due to the different receptors' distribution and to the entailing heterogeneous intracellular events triggered by the drug.

The conclusion is that these results allow to better understand the molecular mechanism of action of this drug and this functional proteomic approach represents an original pharmacological

strategy to evaluate the diversified effects of Clonidine on brain energy metabolism at subcellular level, taking into account the selective vulnerability of different hippocampal subfields.

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Brede et al. (2011). J Cereb Blood Flow Metab. 31, 1-7.

Ferrari et al. (2015). Eur J Pharmacol. 756, 67-74.

Gupta & Sharma (2014). Eur J Pharmacol. 723, 80-90.

Hoffman et al. (1991). Anesth. Analg. 73, 460-4.

Moretti et al. (2015). Pharmacol Ther. 146, 23-34.

Villa et al. (2013). J Proteome Res. 12, 5422-35.

Villa et al. (2016). Neuroscience. 330, 326-34.

Wang et al. (2005). Eur J Pharmacol. 514, 191-9.

Zhang (2004). Neuroscience. 125, 625-31.