Alzheimer’s disease is the most prevalent type of dementia today, discovered and described by Alois Alzheimer in 1907. According to the World Alzheimer Report 2021, 75% of people with dementia worldwide are undiagnosed, equivalent to 41 million people (Gauthier et al., 2021). With each passing year, the number of people affected by these diseases is increasing, and the estimates of suffering from them in the future are growing. There are currently only four drugs used against Alzheimer’s disease: donepezil, rivastigmine, galantamine, and memantine. The first three, based on the cholinergic hypothesis, aim to inhibit the acetylcholinesterase (AChE) and butyrylcholinesterase (BuChE) to prevent the reduction of acetylcholine levels in the brain. Memantine, on the other hand, is based on the glutamatergic hypothesis, according to which in Alzheimer’s disease there are higher than normal levels of glutamate which, through interaction with the glutamatergic N-methyl-D-aspartate receptor, lead to abnormally high levels of calcium which cause neuronal damage. Blocking N-methyl-D-aspartate receptors to reduce these calcium levels is a therapeutic target to control this disease, and this is the mechanism of action of memantine (Rubin, 2021). Recently, the drug Aducanumab has also been approved in the United States. This is a human IgG1 monoclonal antibody that primarily binds to amyloid-beta (Aβ) aggregates, soluble oligomers, and also insoluble fibrils, limiting the toxicity of these species.

Different molecular mechanisms that could be involved in the development of Alzheimer’s disease have been proposed. The main ones include oxidative stress, excessive levels of glutamate, neuroinflammation, hyperphosphorylation of tau proteins, decreased levels of acetylcholine and abnormal accumulation of Aβ plaques (Lindstrom et al., 2021). Ca²⁺ dysregulation is also another important factor in Alzheimer’s disease. Under normal conditions, extracellular Ca²⁺ concentration is higher than intracellular Ca²⁺ concentration. Molecular interactions of beta-amyloid molecules with neuronal membranes can significantly alter the calcium channels in the membrane, increase the influx and cause an imbalance of this ion. High intracellular calcium concentrations result in considerable toxicity and subsequently cell death. This disruption of calcium homeostasis can have other secondary consequences, such as lipid peroxidation and generation of reactive oxygen species, which can lead, in the long term, to a significant reduction in synaptic integrity (Garwood et al., 2013). Therefore, to treat this disease, we cannot only focus our strategies on targeting a single therapeutic target, but we must also address more than one for its treatment. Such a strategy is central to the development of new drugs and is now being pursued by a variety of researchers and holds great promise in the fight against Alzheimer’s disease.

Understanding the molecular mechanisms of membrane protection is what motivated us to study one of the multitarget compounds whose in vitro and in vivo characteristics have been of interest (Zambrano et al., 2021). AVCRI104P4 (Figure 1), is a donepezil-huprine hybrid that possesses marked in vitro inhibitory activity of human acetylcholinesterase and human butyrylcholinesterase. In addition, AVCRI104P4 has a strong ability to cross the blood-brain barrier, which has been confirmed in several studies. In experiments in mice (APP SL) AVCRI104P4 was able to improve short-term memory (Sola et al., 2015). Furthermore, the effect of AVCRI104P4 on Aβ aggregation has been studied in vitro and in vivo. Using a thioflavin T fluorescence method (Bartolini et al., 2007), AVCRI104P4 was found to inhibit AChE-induced Aβ₁₋₄₂ aggregation in vitro by 41% at 100 µM and inhibited spontaneous Aβ₁₋₄₂ aggregation by 29% at a concentration of 10 µM (Viama et al., 2010). However, our interest was focused on the study of its possible membrane-protective capacity. For this purpose, in our experiments, we used molecular models of cell membranes constructed with dimyristoyl (DM) phosphatidylcholine (PC) and DM phosphatidylethanolamine (PE) bilayers and human erythrocytes, which were exposed to different concentrations of Aβ₁₋₄₂. It has been shown by different methods that the two dominant phospholipid classes in all brain areas of patients with Alzheimer’s disease and dementia are PE and PC (Söderberg et al., 1992). This approach was based on the importance attributed to Aβ-neuronal membrane interaction in the development and progression of the disease which can have important consequences at the biochemical level, such as alterations in membrane fluidity and ion channel formation (Zambrano et al., 2021). For this reason, we considered it of interest to understand the molecular mechanism of the interaction of Aβ₁₋₄₂ with cell membranes and to determine the possible protective effect of AVCRI104P4.

Our experimental results showed a protective effect of AVCRI104P4 against Aβ₁₋₄₂ peptide-induced toxicity in human erythrocytes and membrane molecular models. Using X-ray diffraction studies, we found that increasing concentrations of AVCRI104P4 neutralize the disruptive effect of Aβ₁₋₄₀ on DMPC bilayers. Furthermore, results obtained by scanning electron microscopy showed that pre-incubation of erythrocytes with AVCRI104P4 at increasing concentrations prevents Aβ₁₋₄₀-induced red blood cell morphological alterations and cell lysis.

However, the detailed mechanism of action of this phenomenon has not yet been elucidated. Considering our experimental results, it is possible to conclude that there may be different mechanisms through which AVCRI104P4 may protect the plasma membrane from the toxic effects of Aβ₁₋₄₀. One of them could be due to the direct action of AVCRI104P4 on membrane models. We found that the hybrid was able to bind to the phospholipids DMPC and DMPE, which may indicate a mechanism to protect the membrane, either by reinforcing it or by preventing the disruptive action of the amyloid peptide itself. Another possible mechanism could be attributed to the dual effect of AVCRI104P4 in binding to AChE and preventing Aβ₁₋₄₀ aggregation (AChE-induced).

A more interesting mechanism to explore is the direct interaction of free AVCRI104P4 molecules and Aβ₁₋₄₀. This approach presents itself as a much more complex intellectual challenge because many of the currently tested Aβ₁₋₄₀ anti-aggregation molecules are not completely safe to project into in vivo studies (Pagano et al., 2020). Undesirable effects or parallel molecular interactions of these molecules do not allow them to be considered as safe alternatives. However, elucidation of the exact three-dimensional conformations of Aβ₁₋₄₀ peptides by cryo-electromicroscopy or another structure prediction technique could lead to a more precise identification of residues interacting with neuronal membrane proteins, lipids, and sugars. These questions can be further...
research into the origins of neurodegenerative diseases advances, new metabolic pathways are being discovered and new hypotheses are being tested experimentally, which will allow future research strategies and new families of molecules with potential therapeutic capabilities to be established.

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Date of submission: January 3, 2022
Date of decision: March 3, 2022
Date of acceptance: March 16, 2022
Date of web publication: July 1, 2022

https://doi.org/10.4103/1673-5374.343903

How to cite this article: Zambrano P (2023) On the interaction of a donepezil-huprine hybrid with synthetic membrane models. Neural Regen Res 18(2):333-334.

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References
Bartolini M, Bertucci C, Bolognesi ML, Cavalli A, Melchiorre C, Andrisano V (2007) Insight into the kinetic of amyloid beta (1-42) peptide self-aggregation: elucidation of inhibitors’ mechanism of action. ChemBiochem 8:2152-2161.
Garwood C, Faizullahbooy A, Wharton SB, Ince PG, Heath PR, Shaw RJ, Baxter L, Gelthorpe C, Forster G, Matthews FE, Brayne C, Simpson JE; MRC Cognitive Function and Ageing Neuropathology Study Group (2013) Calcium dysregulation in relation to Alzheimer-type pathology in the ageing brain. Neurophatol Appl Neurobiol 39:788-799.
Gauthier, Serge, Pedro Rosa-Neto, José Morais, and Claire Webster (2021) World Alzheimer Report 2021: Journey through the Diagnosis of Dementia. https://www.alzint.org/resource/world-alzheimer-report-2021/ Accessed December 30, 2021.