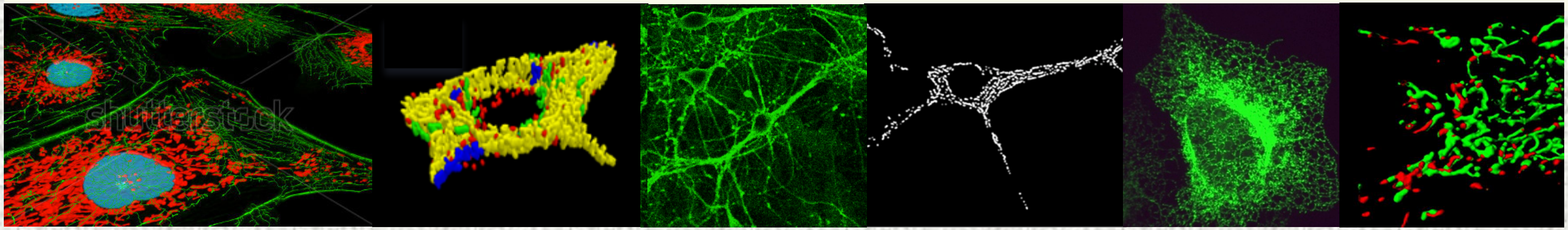
BIOMEDICINA I– 2014



ESTRUCTURA Y SEÑALIZACIÓN CELULAR III

MEMBRANA, RETÍCULO, MITOCONDRIA Y NÚCLEO EN LA SEÑALIZACIÓN EN PATOLOGÍAS NEURODEGENERATIVAS

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Postdoctorado – Instituto de Ciencias Biomédicas, Facultad de Medicina, Universidad de Chile

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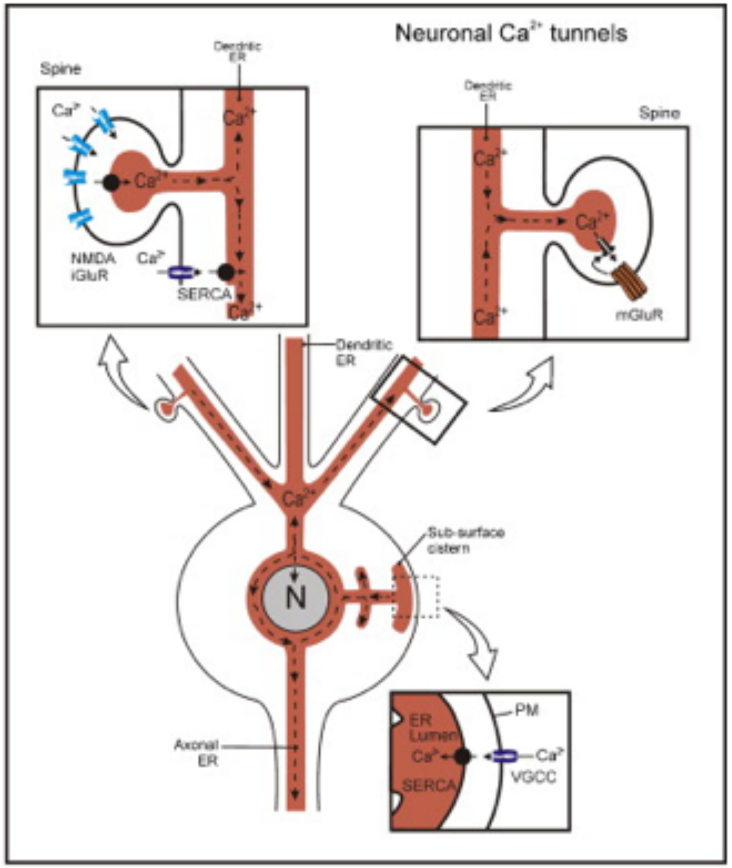
La continuidad del REL es importante para la propagación de señales de Ca^{2+} a través de largas distancias y para el transporte de lípidos y proteínas

A model of Ca^{2+} tunnels in neurons



Continuous ER lumen allows Ca^{2+} movements between neuronal compartments, which could be important for internal reloading of locally activated Ca^{2+} release and for long-range Ca^{2+} transport from the sites of entry located in distal dendrites towards the soma and axon.

Petersen, O.H. and Verkhratsky, A., 2007) Cell Calcium 42, 373–378

Señales de liberación de Ca^{2+}

- *La liberación de calcio es rápida y circunscrita a microdominios, determinados por las múltiples proteínas que lo unen.*

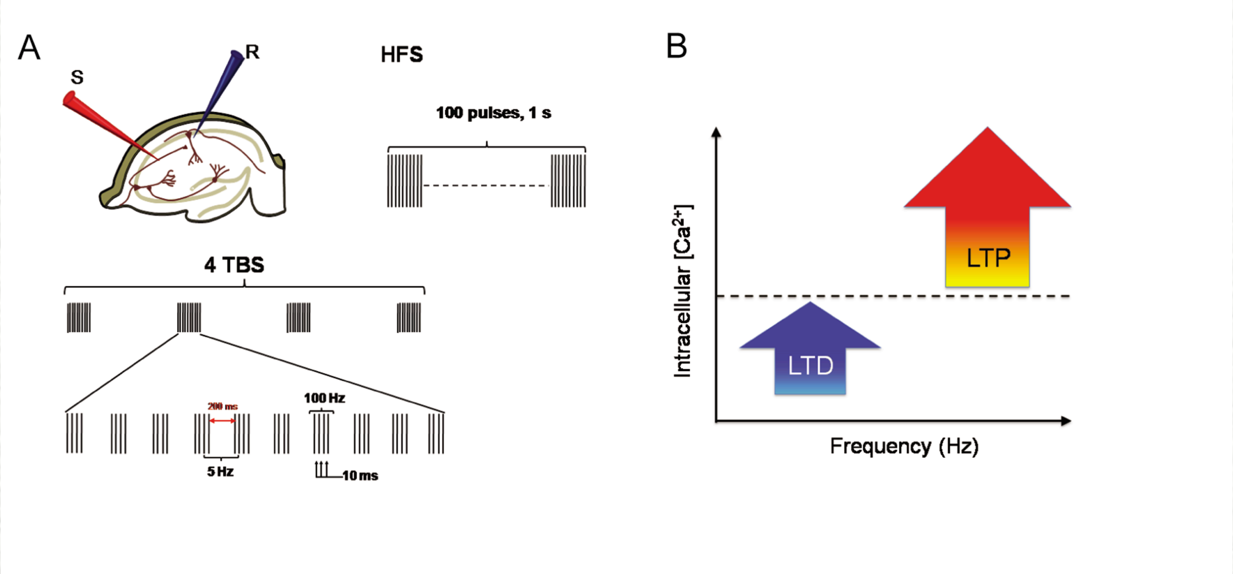
- *Las señales pueden ser generadas por eventos de liberación muy localizados, hasta eventos más complejos que se propagan como ondas.*

Señales de Ca^{2+} distintas: LTP X LTD

High neuronal activity (HFS) triggers the emergence of transient, high magnitude postsynaptic calcium signals.

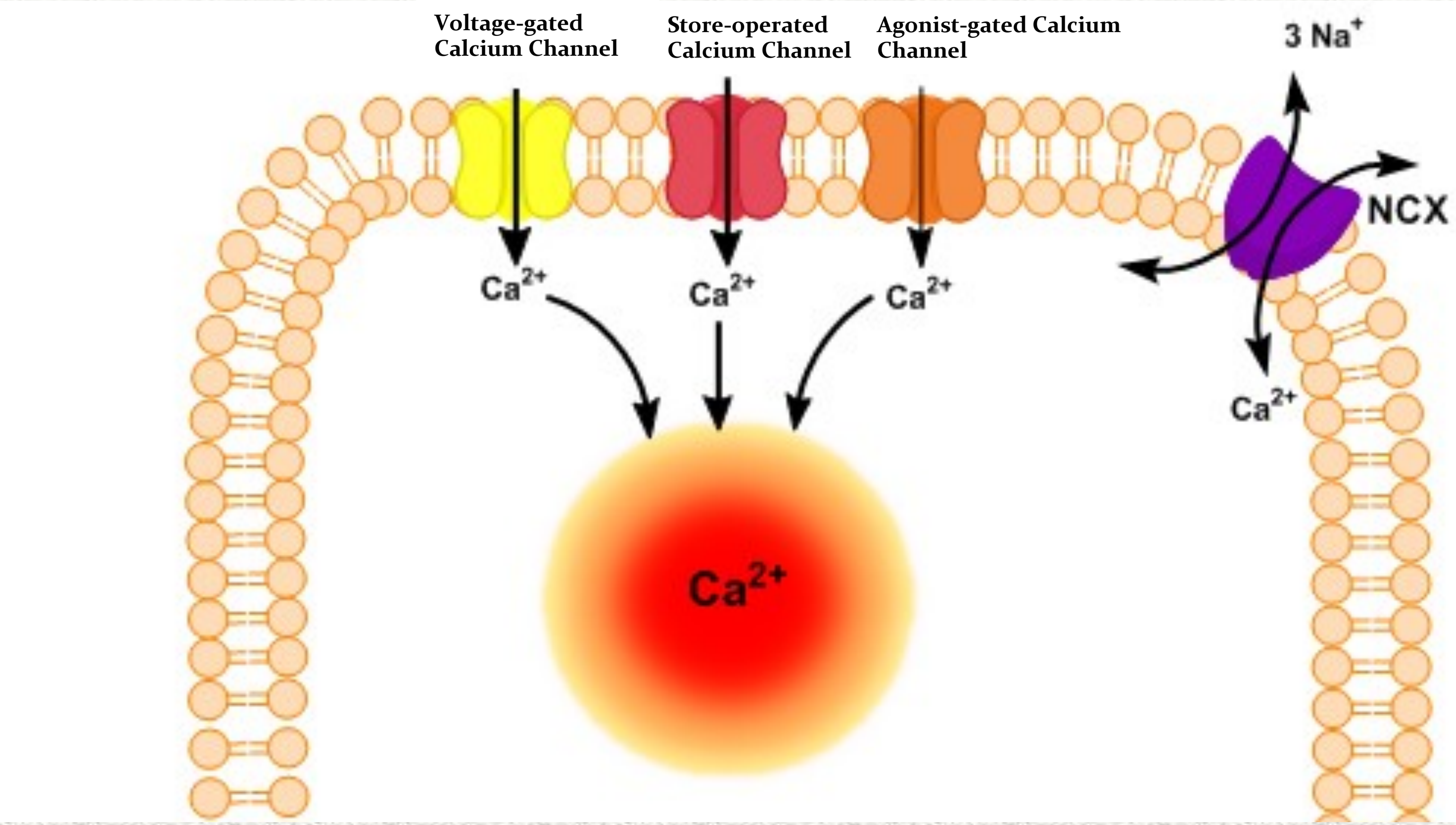
These calcium signals induce LTP, and stimulate kinases that induce the expression of genes, promoting dendritic spine growth and triggering cellular cascades that contribute to the integration and storage of neuronal information (learning and memory).

In contrast, low neuronal activity (low frequency stimulation) generates low magnitude but persistent calcium signals that activate phosphatases which inhibit gene expression (forgetting).



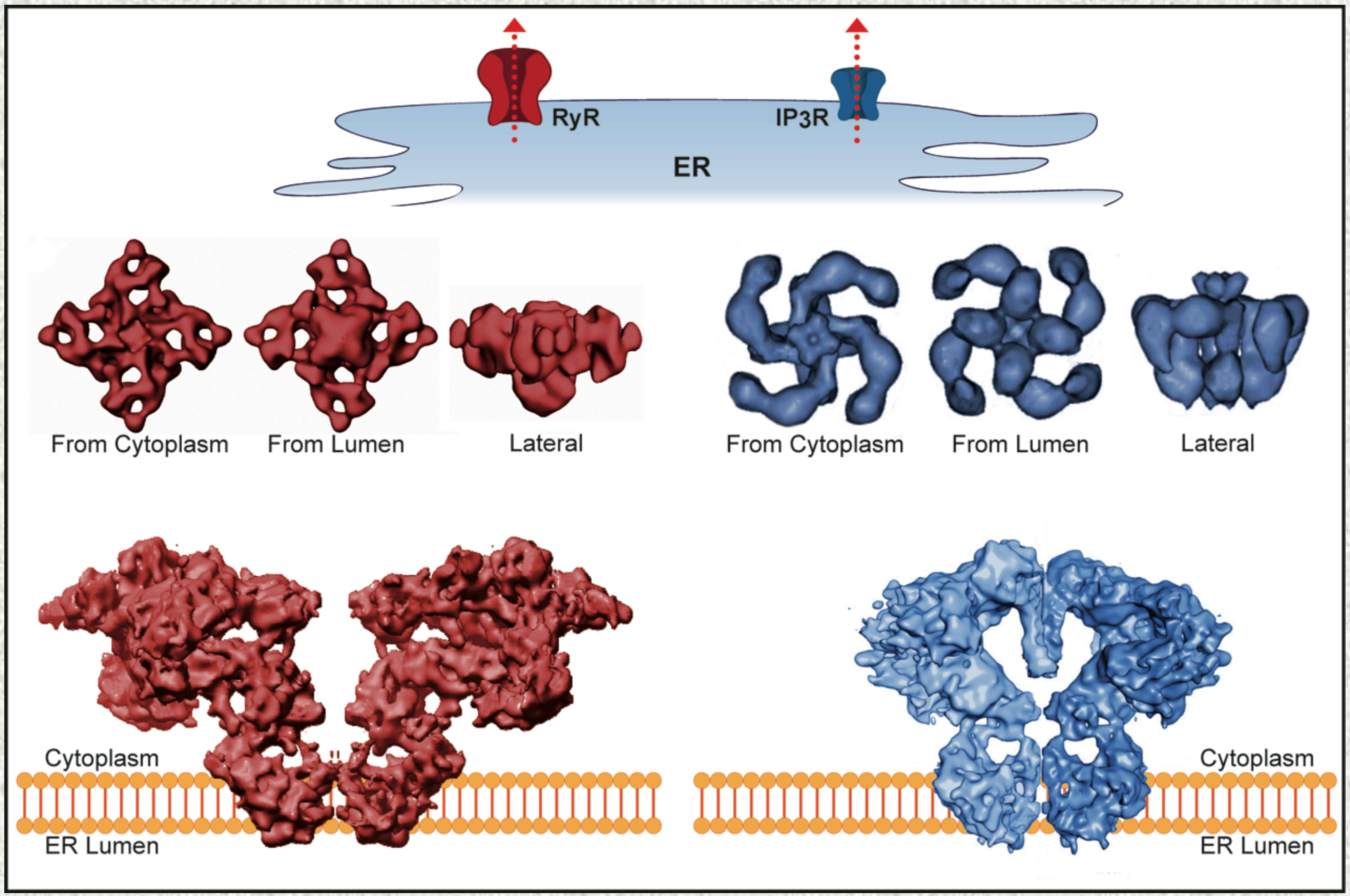
Paula-Lima et al., 2014
Antiox. & Redox. Sig.

Señales de Ca^{2+} generadas por entrada de Ca^{2+} desde el medio extracelular



Modificado de Paula-Lima et al., 2014 Antiox. & Redox. Sig.

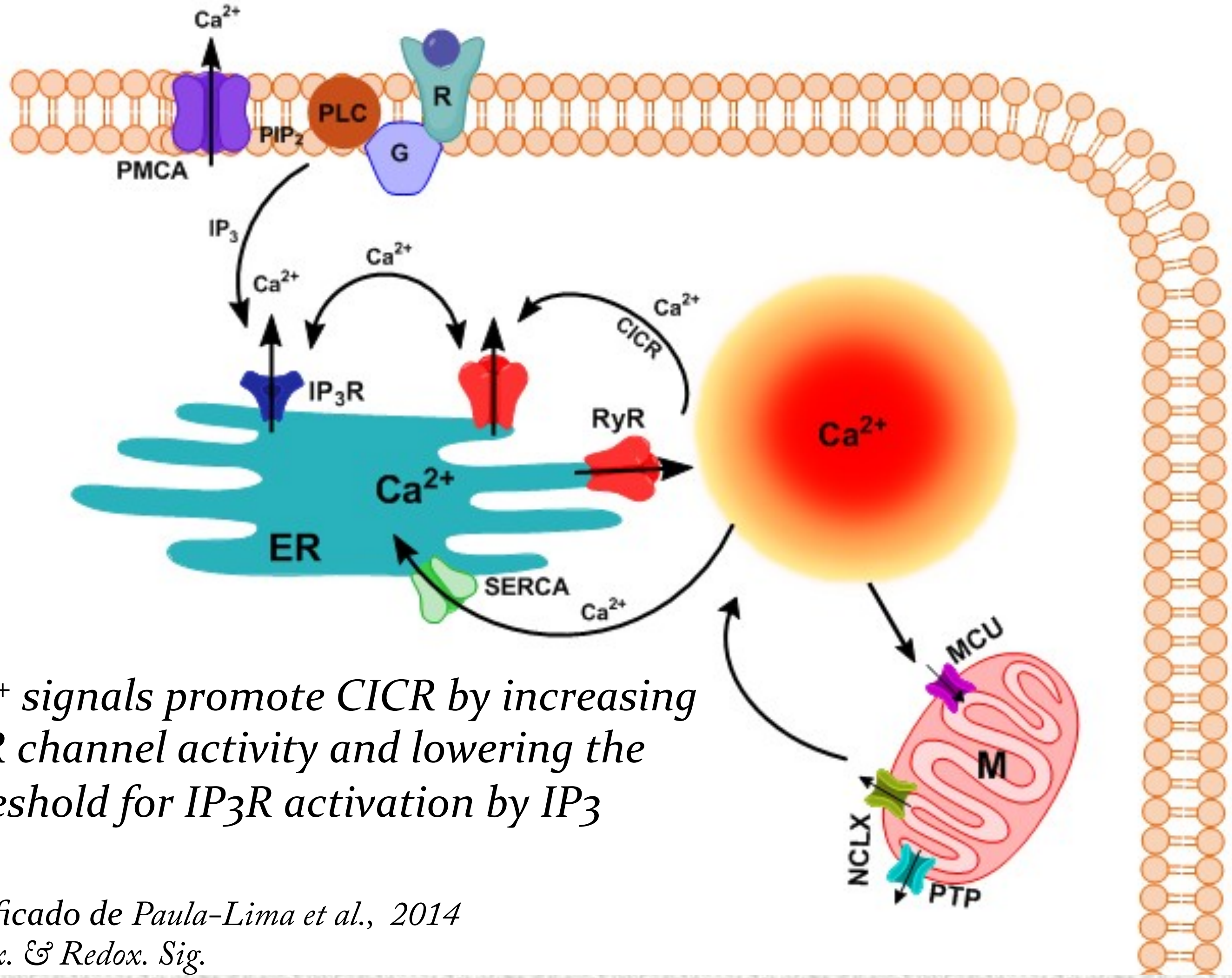
El retículo endoplasmico tiene dos tipos de canales de liberación de Ca^{2+}: IP3R y RyR



Ambos generan señales de Ca^{2+}

Paula-Lima et al., 2014
Antiox. & Redox. Sig.

Señales de Ca^{2+} generados por liberación de Ca^{2+} del retículo endoplasmico o la mitocondria



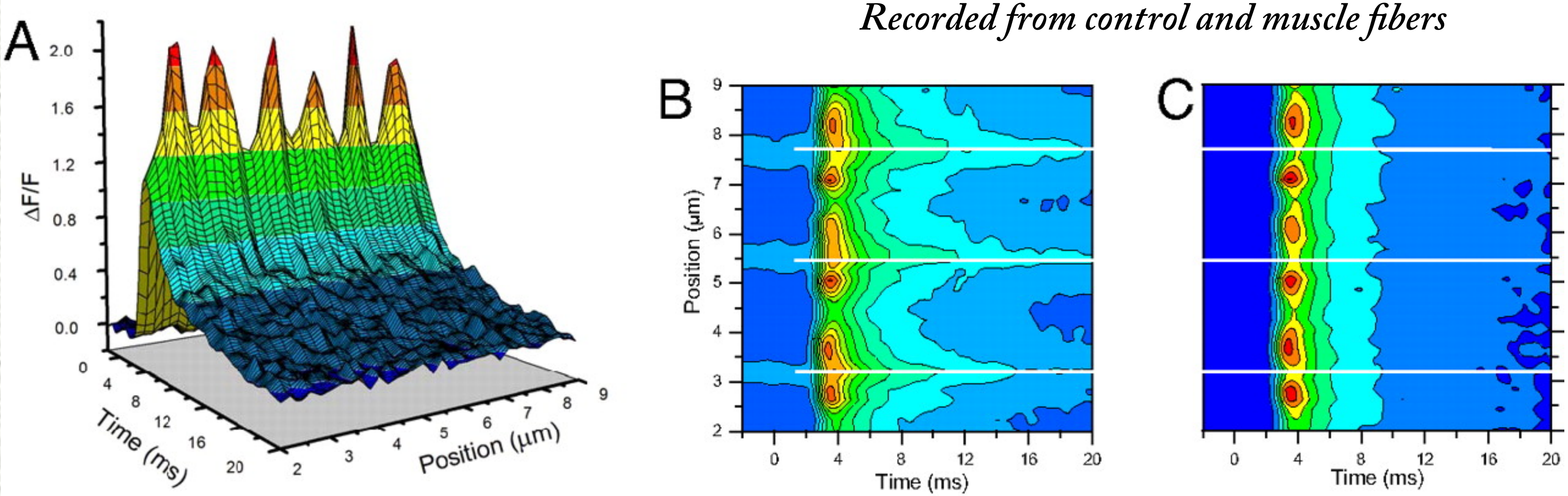
Ca^{2+} signals promote CICR by increasing RyR channel activity and lowering the threshold for IP3R activation by IP3

Modificado de *Paula-Lima et al., 2014 Antiox. & Redox. Sig.*

Microdomínios de Ca^{2+}

3-D plots (A) and contour maps (C) of Ca^{2+} microdomains constructed from $\Delta F/F$ records acquired from a control muscle fiber.

Recorded from control and muscle fibers



DiFranco M et al. PNAS 2008;105:14698-14703

PNAS

La misma población de canales de Ca²⁺ puede producir diversidad en los microdomínios

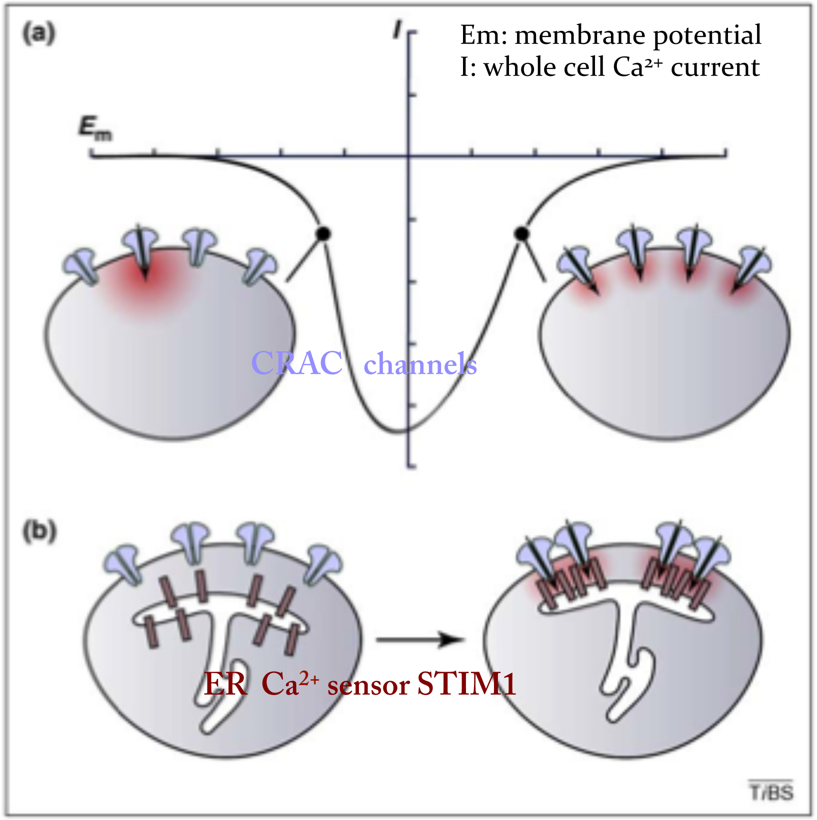


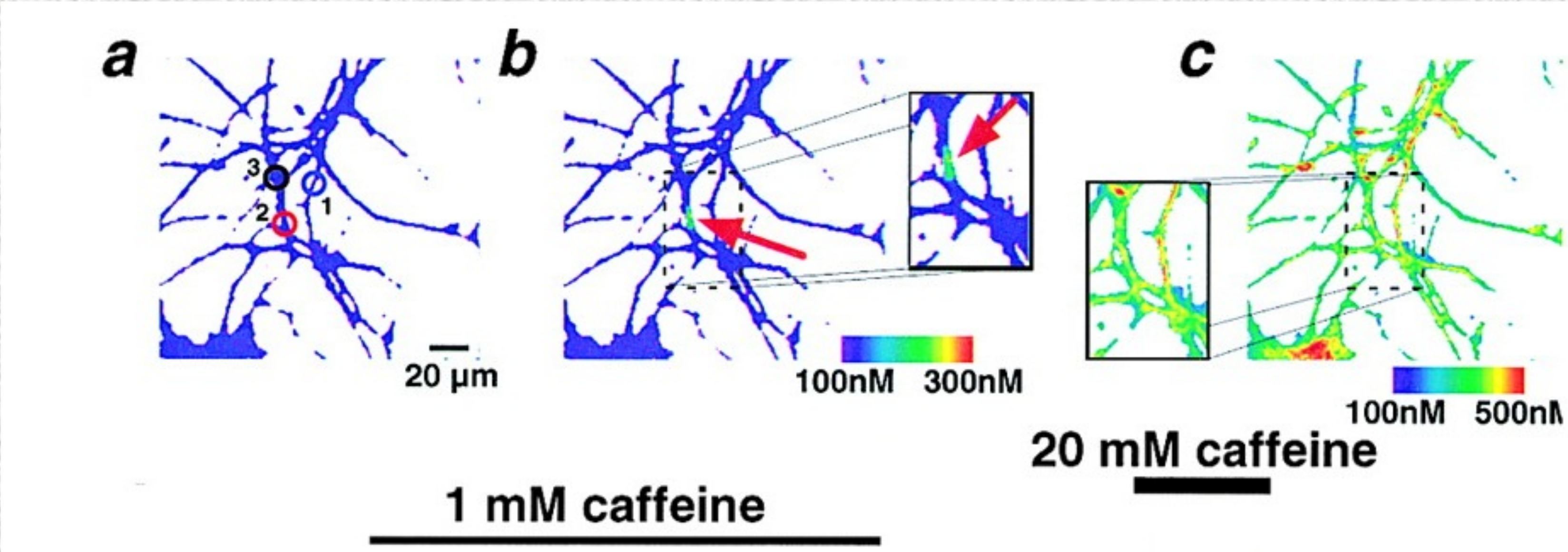
Figure 4. The same population of Ca^{2+} channels can generate Ca^{2+} microdomain diversity. **(a)** Modest depolarisations open few voltage-gated Ca^{2+} channels but the driving force for Ca^{2+} entry is relatively large and results in a large Ca^{2+} microdomain. Strong depolarisations, to give the same amount of whole cell current, recruit many more channels, but the subsequent microdomains are smaller. E_m denotes membrane potential and I represents whole cell Ca^{2+} current. **(b)** Clustering of plasmalemmal CRAC channels following store depletion by the ER Ca^{2+} sensor STIM1 produces hotspots of overlapping Ca^{2+} microdomains that will increase local $[Ca^{2+}]$ considerably in the restricted subplasmalemmal space.

Modest depolarization: large Ca2+ microdomain
Strong depolarizations: for the same I, more channels, smaller microdomains

Ca^{2+} microdomains: considerable increase in local Ca^{2+} in the restricted subplasmalemal space.

Parek, 2011
Cell

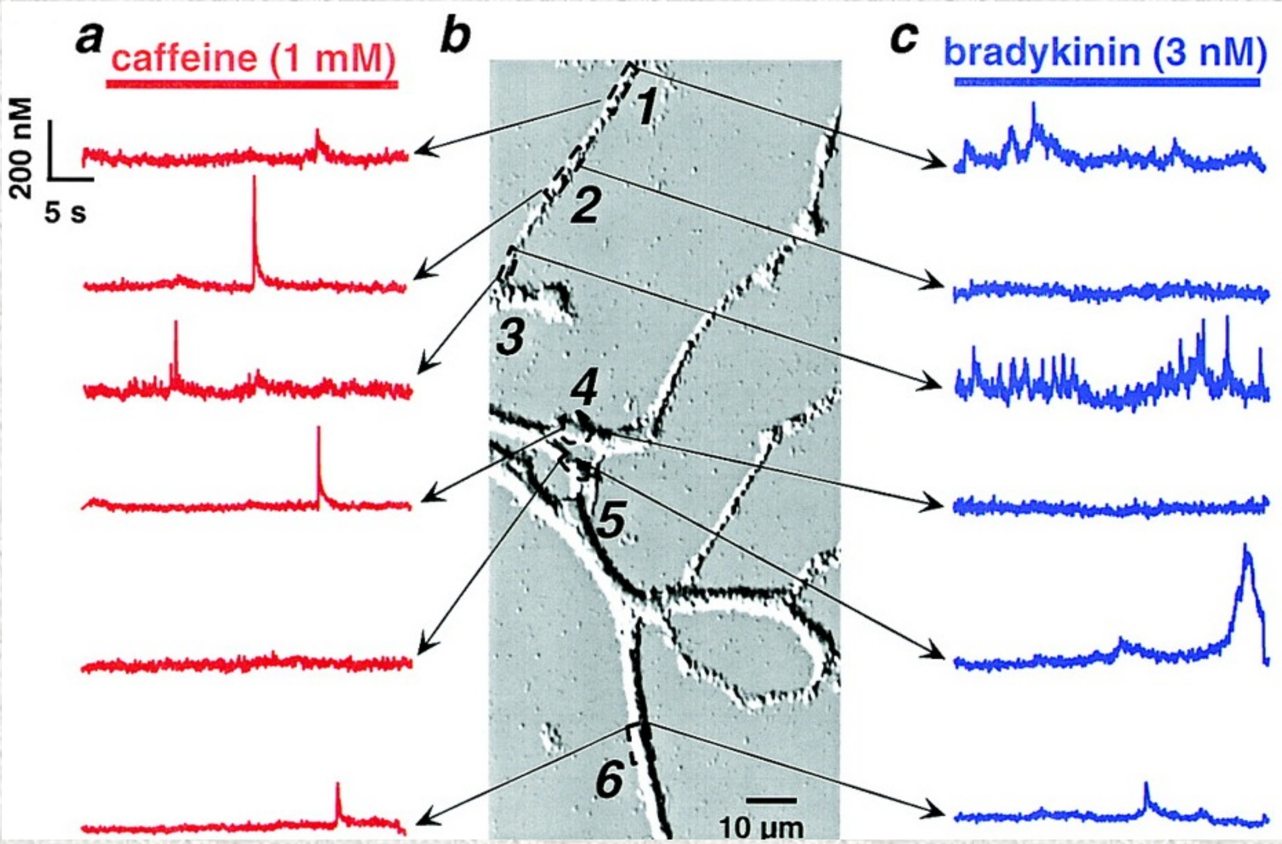
Liberación de Ca^{2+} mediada por RyR en neuronas en cultivo



Characterization of Elementary Ca^{2+} Release Signals in NGF-Differentiated PC12 Cells and Hippocampal Neurons

Koizumi, Bootman, Bobanovi, Schell, Berridge, Lipp (1999). Neuron 22, 125-137

Señales de Ca^{+2} mediadas por RyR e IP3R registradas en distintas regiones de las neuritas



Characterization of Elementary Ca^{2+} Release Signals in NGF-Differentiated PC12 Cells and Hippocampal Neurons
Koizumi, Bootman, Bobanovi, Schell, Berridge, Lipp (1999). Neuron 22, 125-137

Ondas de Ca^{2+} mediadas por receptores IP3 en medio libre de Ca^{2+}

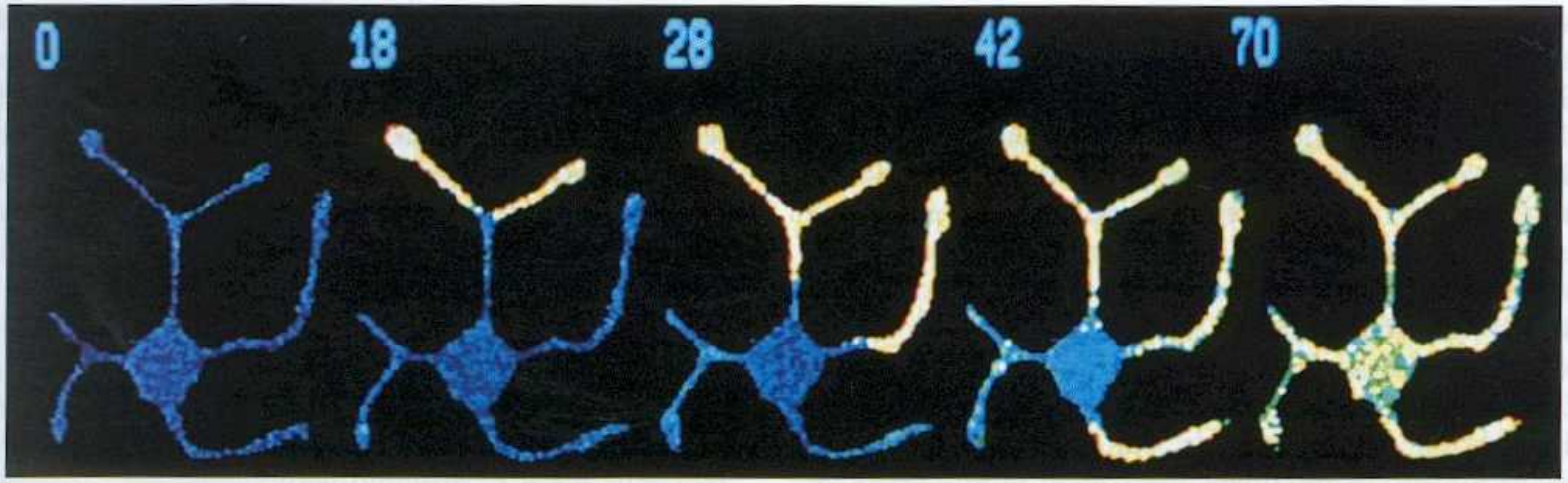
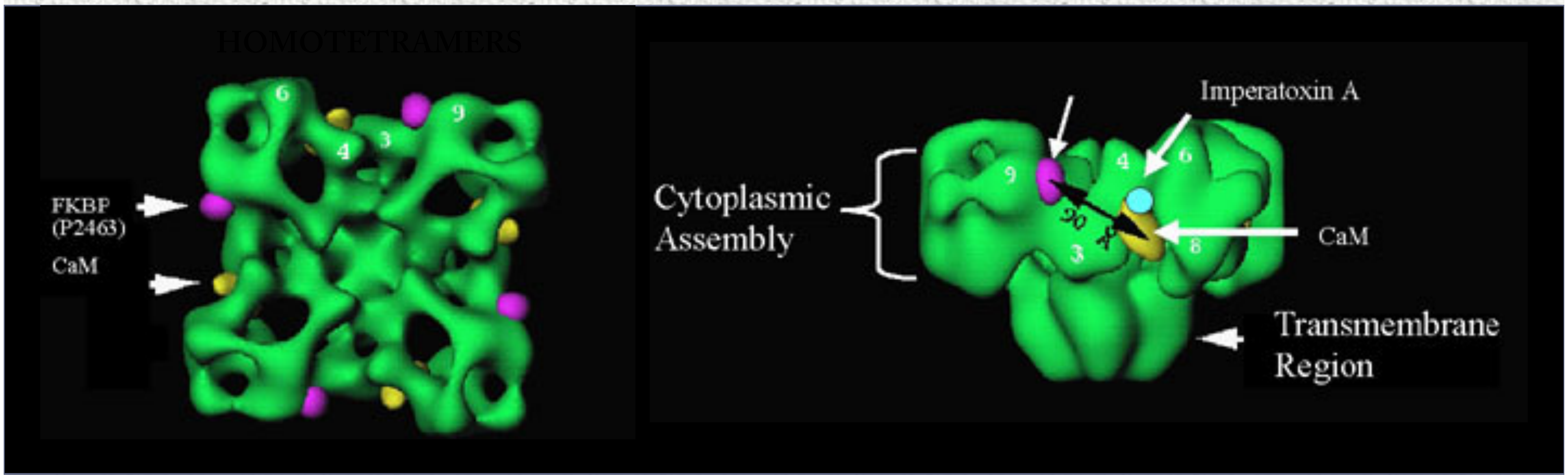


Fig. 4. Ca^{2+} waves in differentiated PC12 cells stimulated with bradikynin in a Ca^{2+} free medium. Notice that the store-dependent waves initiate asynchronously at the tip of the various neurites and then proceed non-decrementally towards soma. Reproduced from the J. Cell Biol., from Lorenzon et al. (1995) by copyright permission of The Rockefeller University Press.

Receptor de ryanodina (RyR)



- *Subunits - 5000 amino acids, 565 kDa, 3 isoforms*

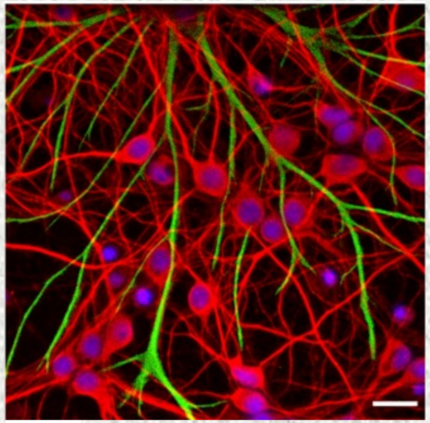
- *RyR activity ▸ muscle contraction, secretion, fecundation, and apoptosis.*

- *RyR1 activity is enhanced in vitro by ROS and Ca^{+2}*

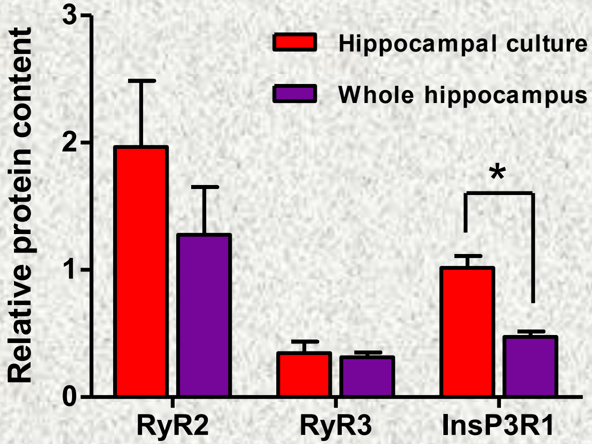
– *Role in neurons: RyR is a coincident receptor of the intracellular calcium and ROS increases resultant from the activation of the NMDA receptors in neurons (Hidalgo C., 2005) and RyR-mediated CICR is involved in changes in gene expression.*

RyR2 y RyR3 en neuronas de hipocampo maduras

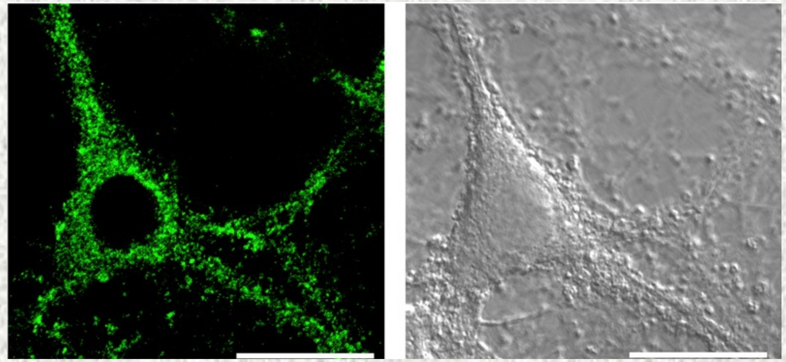
MAP2 GFAP Hoechst



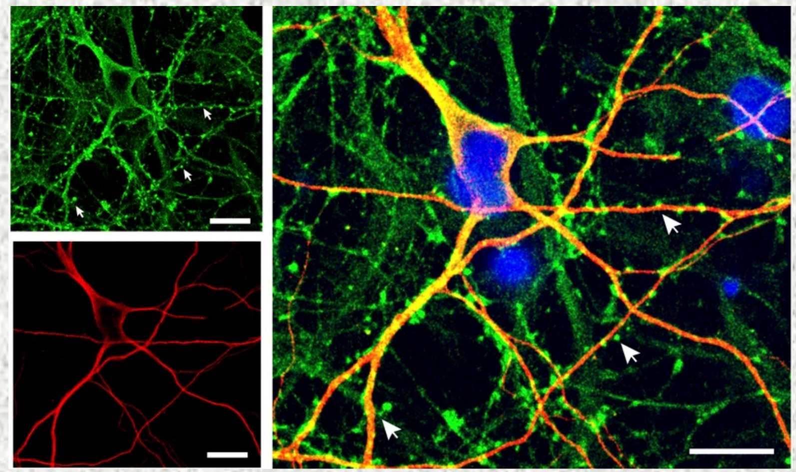
Cultivo primario de hipocampo, enriquecido en neuronas 14 DIV



RyR2

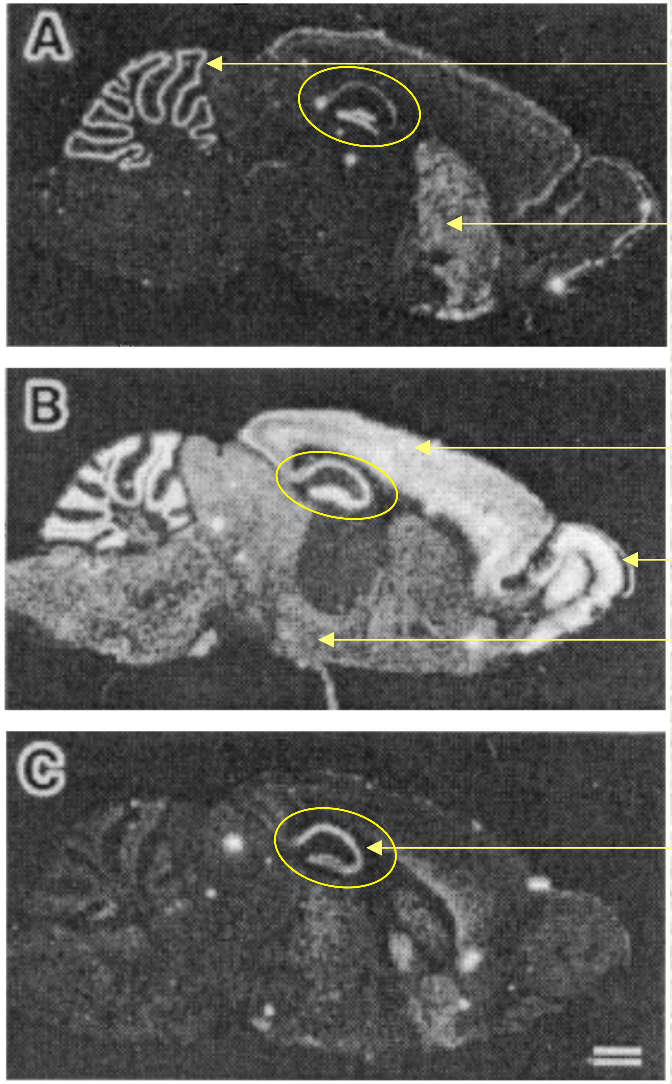


RyR3



MAP2

Distribution of RyR mRNA isoforms in adult mouse brain



RyR1

Purkinje/granular cells, cerebellum

Caudate putamen

RyR2

Brain cortex

Olfactory bulb

Hypothalamus

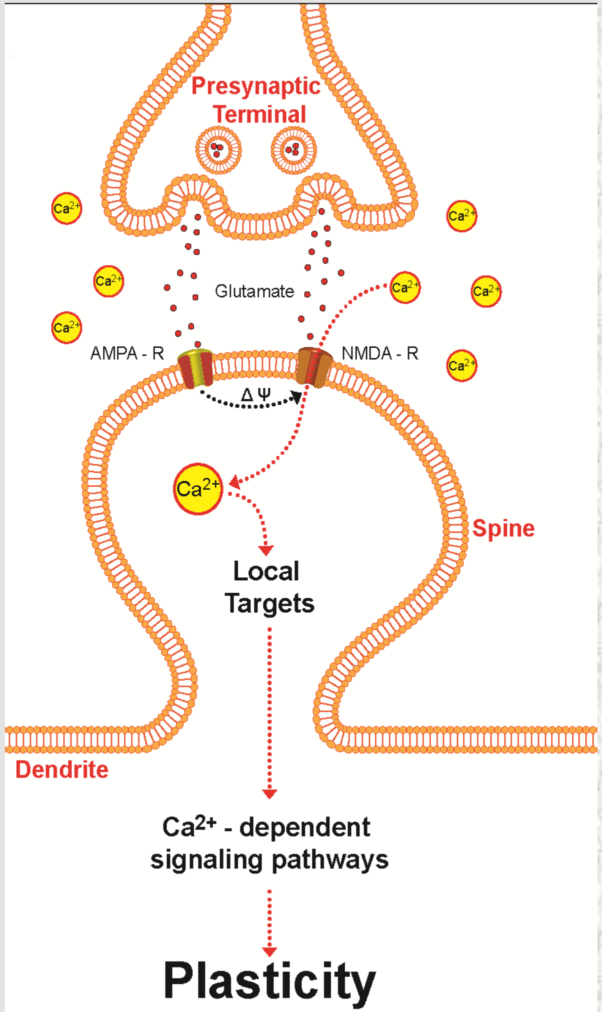
RyR3

Hippocampus

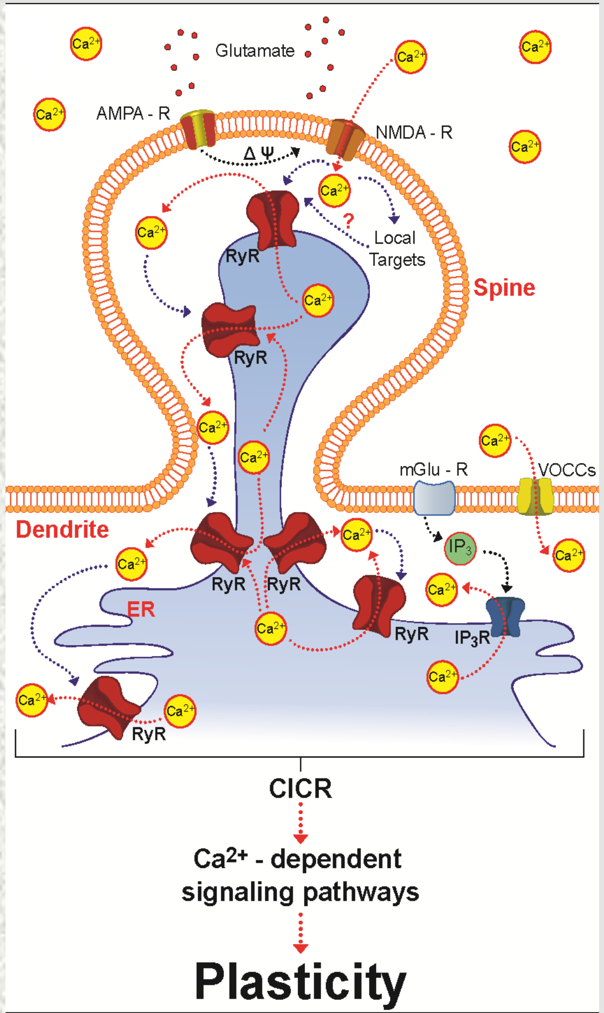
Mori et al. 2000.Neurosc. Lett.82: 57-60.

Nuestro modelo de plasticidade sináptica incluye el CICR

Widespread View



Our model



Paula-Lima et al., 2014
Antiox. & Redox. Sig.

RyR y plasticidade sináptica

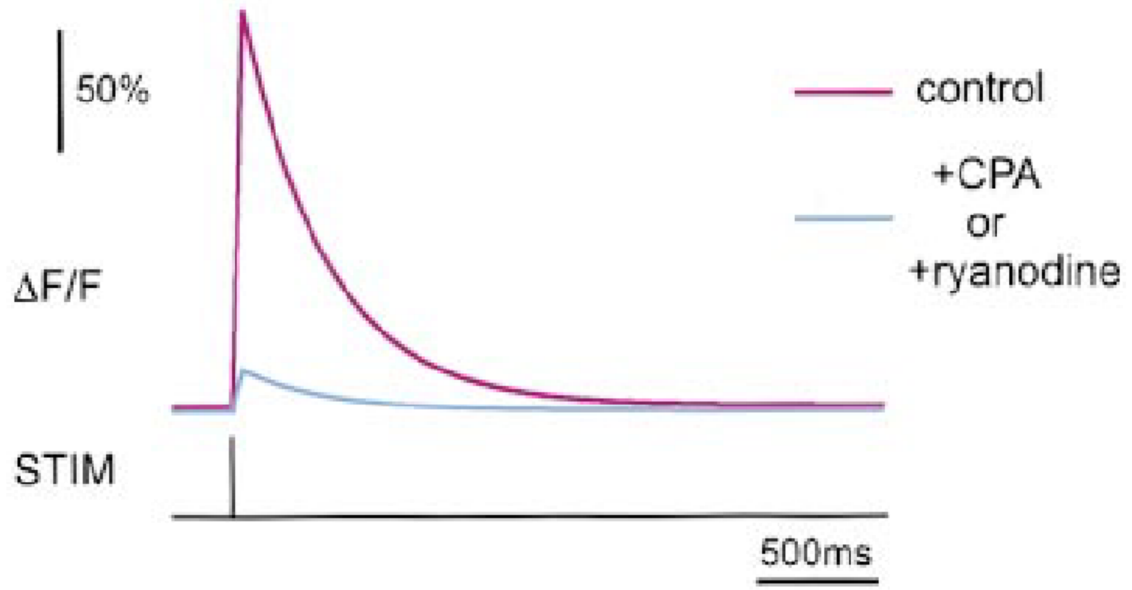
- RyR-mediated Ca^{+2} induced Ca^{+2} release (CICR) promotes spontaneous transmitter release in hippocampal synaptic boutons (Emptage et al., 2001).

- Caffeine-induced RyR-mediated CICR promotes dendritic spine elongation in hippocampal neurons in primary culture (Korkotian and Segal, 1998; Korkotian and Segal, 1999).

- Hippocampal expression of the RyR2 isoform increases after spatial memory training (Zhao et al., 2000)

- Selective knockdown of RyR2 and RyR3 impairs memory processes (Galeotti et al.,2008).

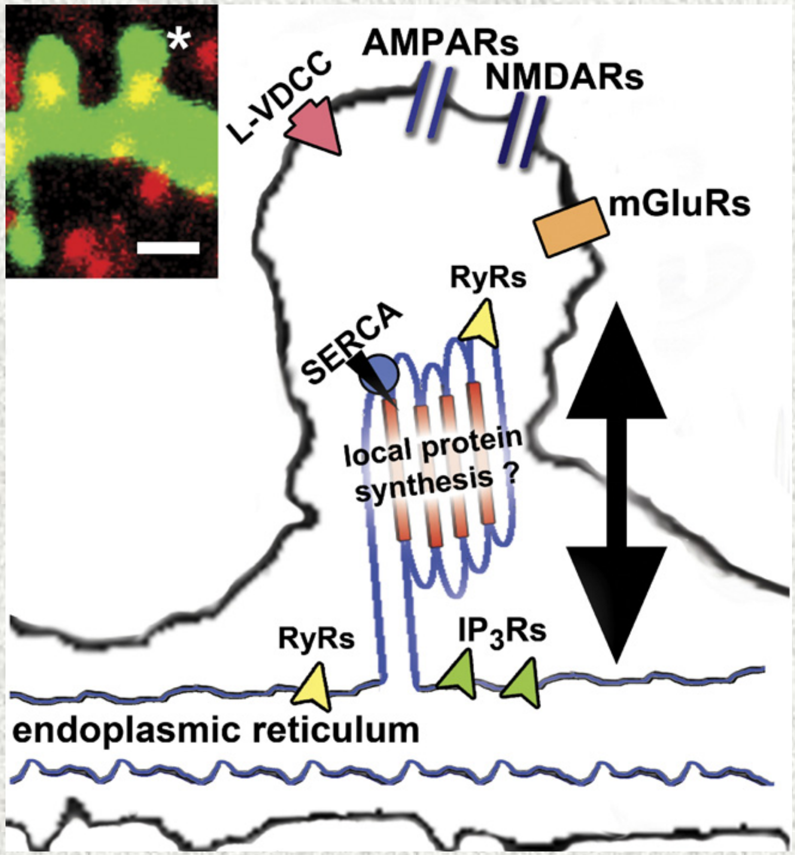
La señales de Ca^{2+} generadas por estímulos sinápticos es dependiente de la liberación de Ca^{2+} mediada por Ca^{2+} (CICR) desde el retículo endoplásmico



(Bottom) Schematic time course of [Ca^{2+}] transients (ΔF/F) evoked by synaptic stimuli (STIM) in the presence and absence of blockers of CICR (Emptage et al., 1999).

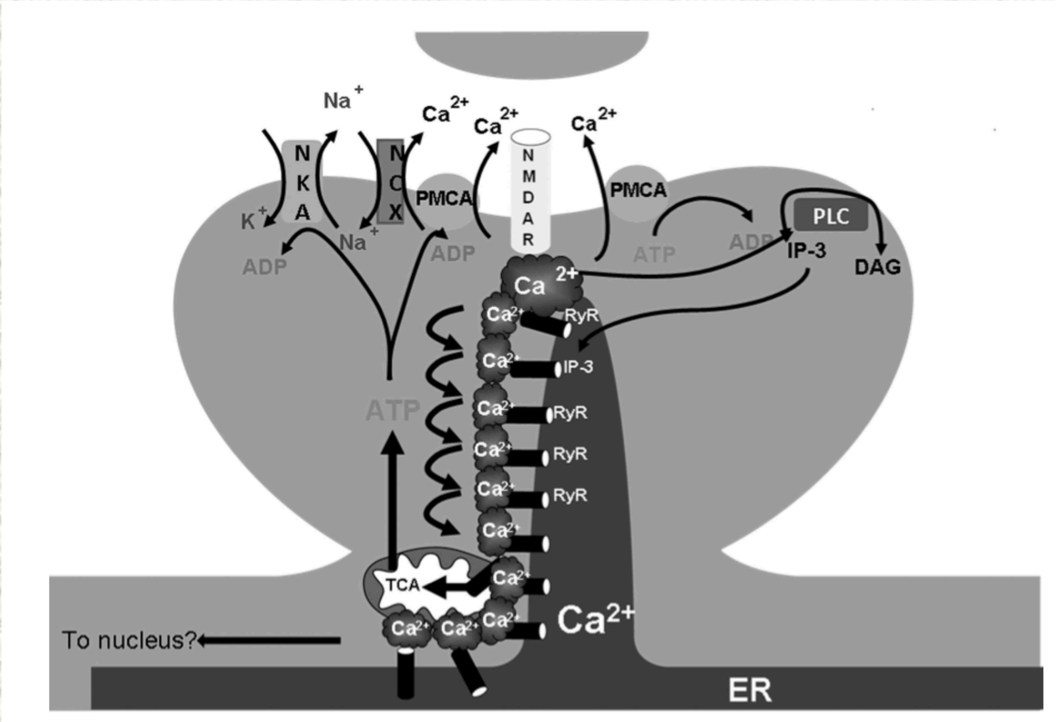
Svoboda & Mainem, Neuron 22, 427, 1999

Las señales de Ca^{2+} generados por las isoformas RyR2-RyR3 estimulan las vías celulares que subyacen a los procesos de memoria.



Jedlicka et al., 2008 | Behav Brain Res

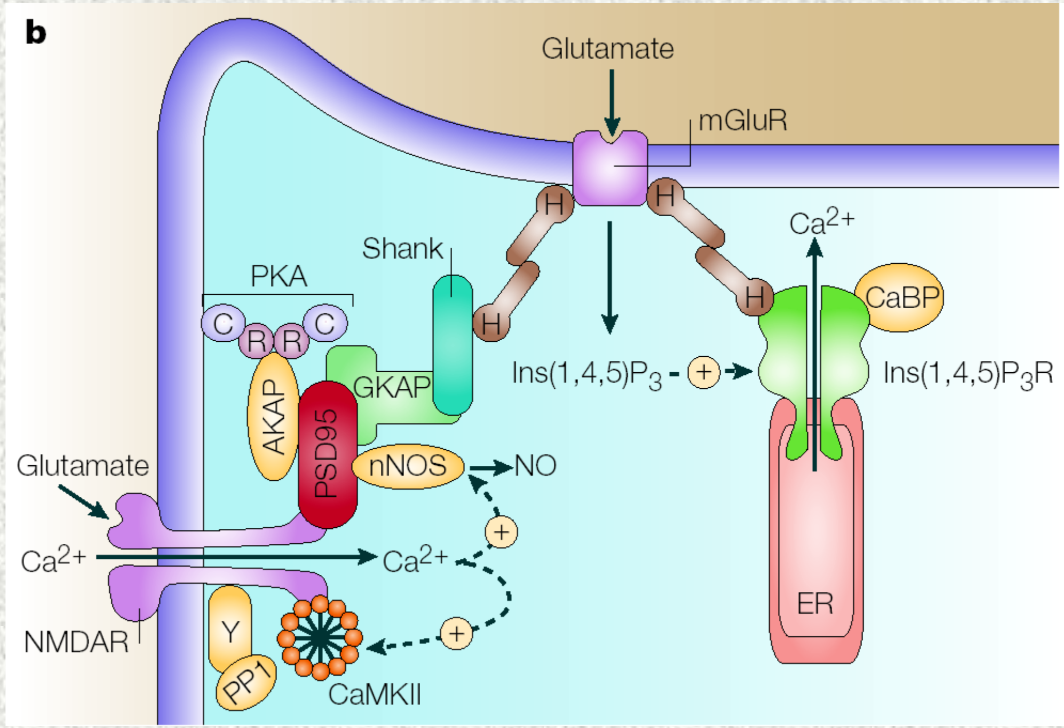
La señales de Ca^{2+} generadas por la activación de receptores NMDA es amplificada por CICR



Gleishmann and Mattson, 2011
Antiox. & Redox Sig.

Figure 3. **Ca^{2+}-induced Ca^{2+} release (CICR).** A postsynaptic spine is depicted. In CICR Ca^{2+} enters the cytosol through channels in the plasma membrane. The increased $[Ca^{2+}]_i$ will cause the opening of nearby ryanodine receptors (RyR) in the ER membrane. Additionally, increased $[Ca^{2+}]_i$ levels result in increased IP_3 levels through phospholipase C (PLC) activation. Opening of RyRs and IP_3 receptors, causes "hotspots" of $Ca^{2+}]_i$ that may cause further opening of ER Ca^{2+} channels and also promote mitochondrial Ca^{2+} uptake.

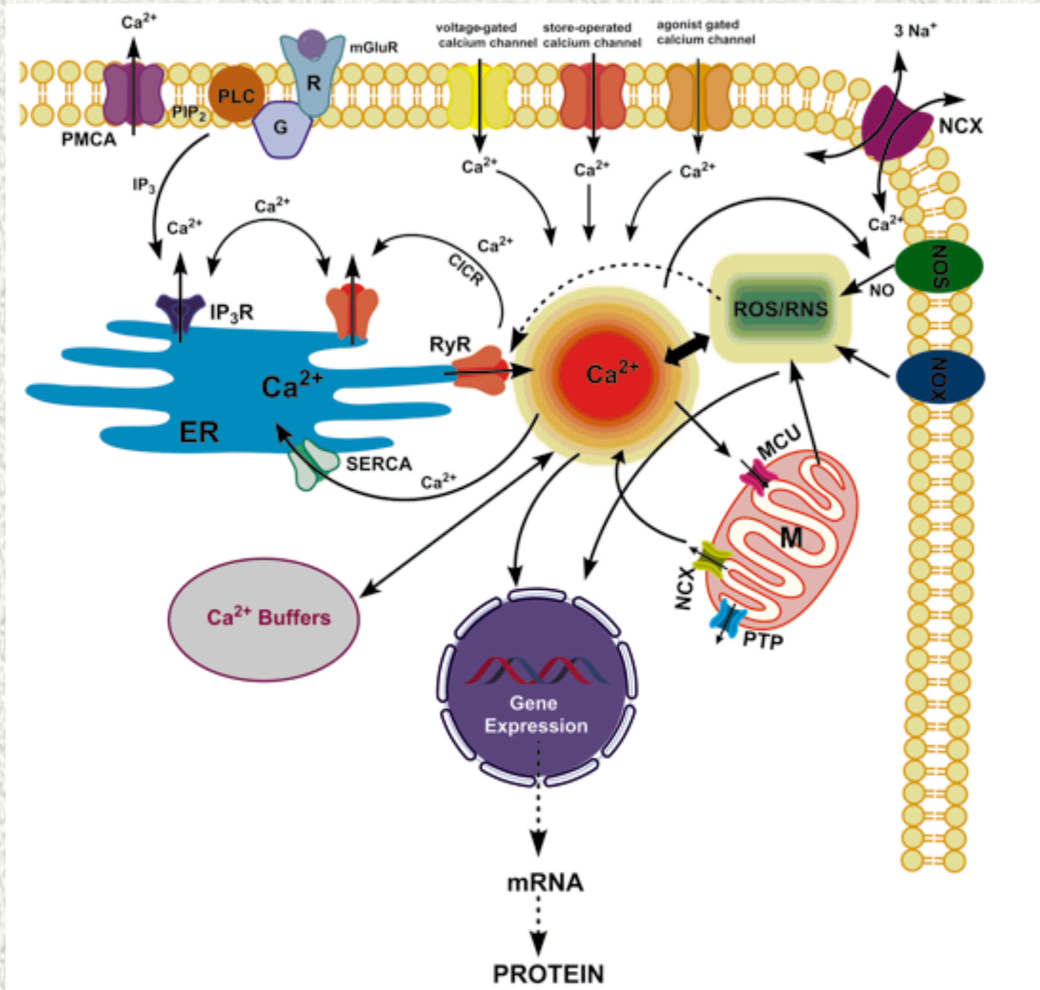
Complejos Moleculares que conectan la membrana plasmática con el retículo endoplasmico



Glutamate-induced Ca^{2+} entry is carried out by NMDA (N-methyl-D-aspartate) receptors (NMDARs) that are linked to other signalling components, some of which are Ca2+ sensitive, such as Ca2+/CaM-dependent kinase II (CaMKII) and neuronal nitric oxide synthase (nNOS).NMDARs are also associated with other proteins such as yotiao (Y), which binds PP1,and the scaffolding protein PSD95, which links into other signalling components such as the AKAP–PKA complex and guanylate kinase-associated protein (GKAP). ***Proteins such as shank and Homer (H) might link the metabotropic glutamate receptor (mGluR) to both the NMDAR and the inositol-1,4,5- trisphospate receptor (Ins(1,4,5)P3R), which is also associated with a Ca2+-binding protein (CaBP).***

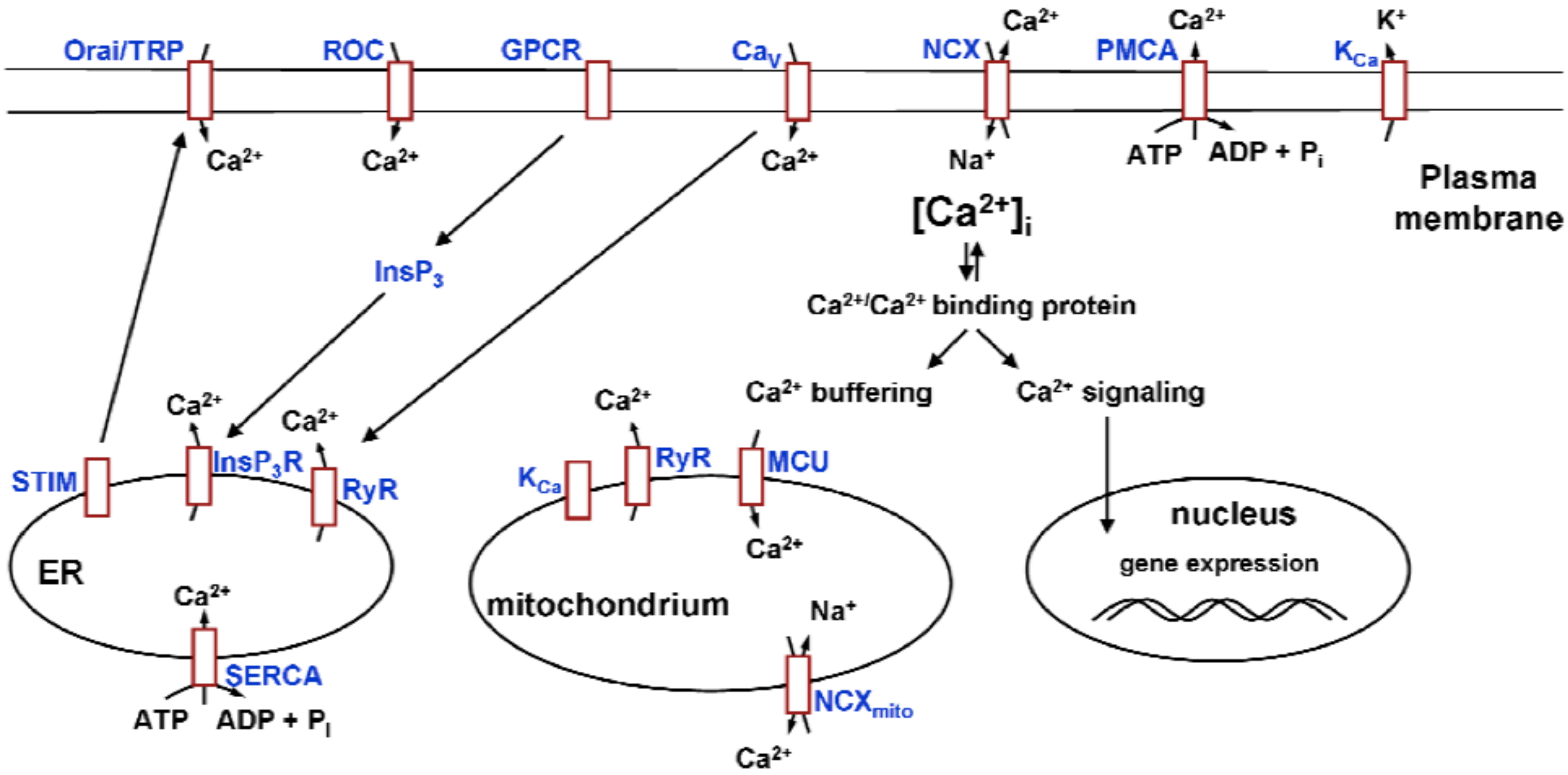
Berridge et al, 2003 Nature Revs Mol Cell Biol 4, 517

Esquema general de las entidades moleculares responsables de la generación de señales intracelulares Ca^{2+} en neuronas

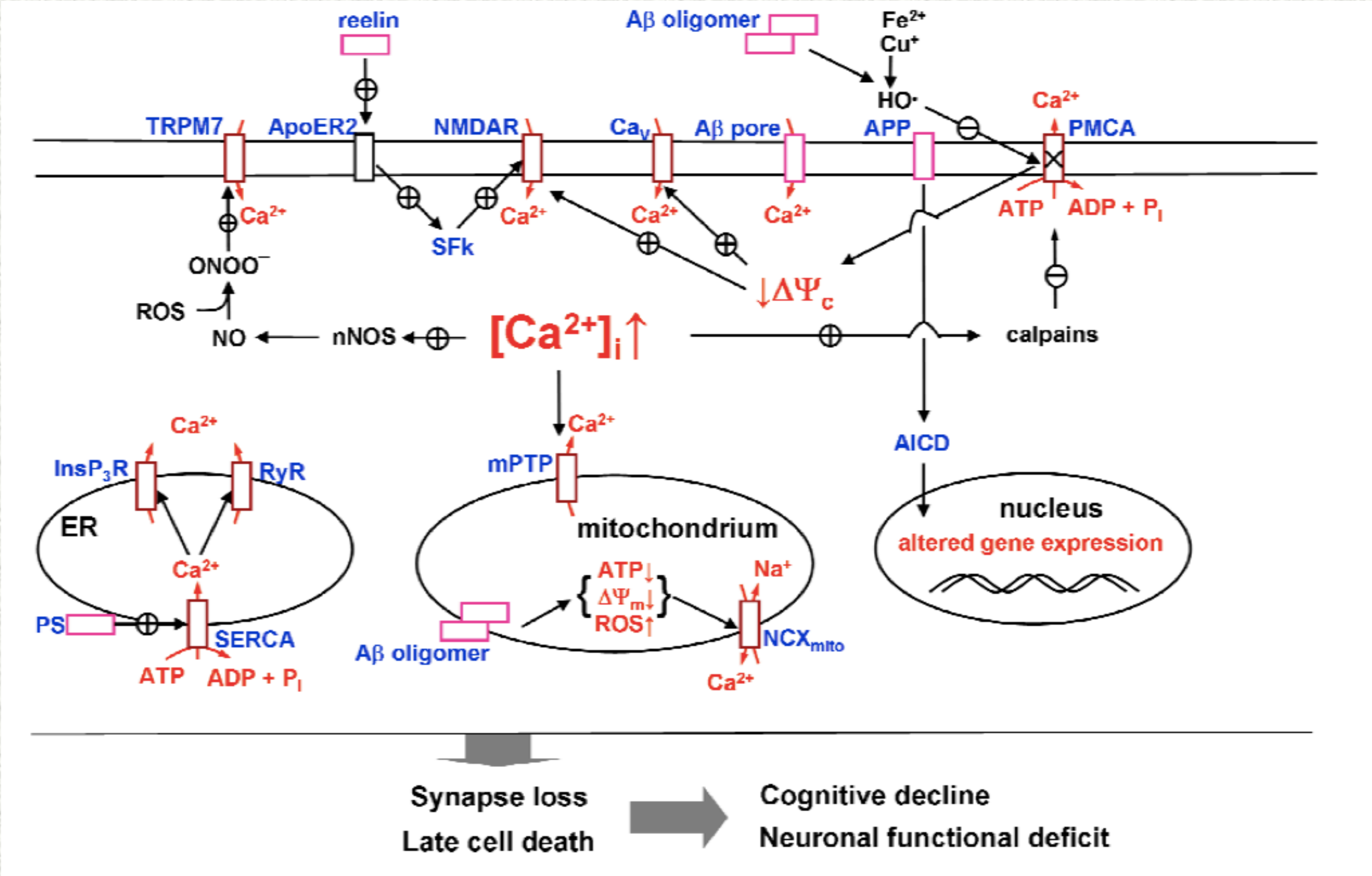


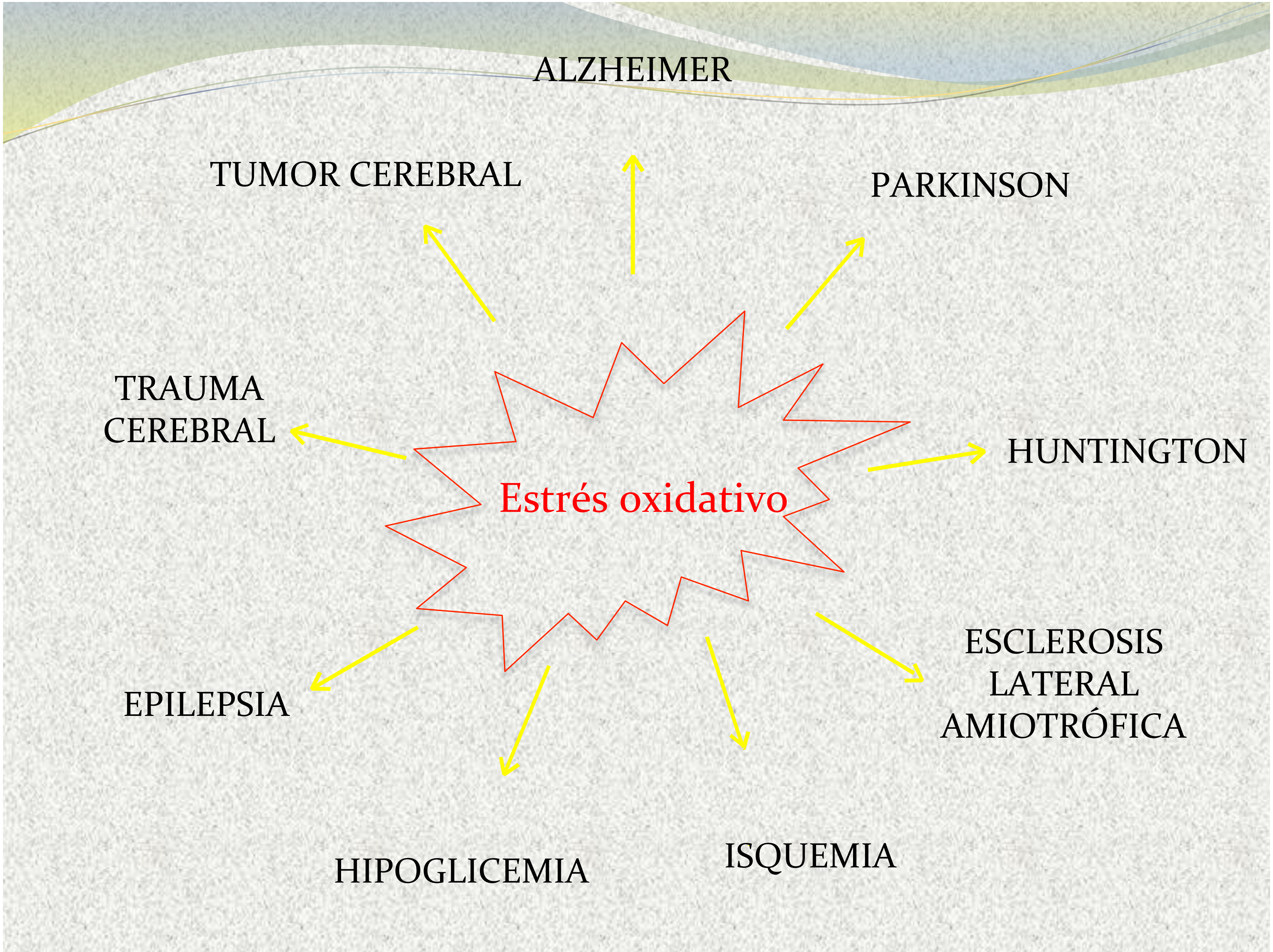
Paula-Lima et al., 2014
Antiox. & Redox. Sig.

Homeostasis de Ca^{2+} en el cerebro en condiciones fisiológicas normales

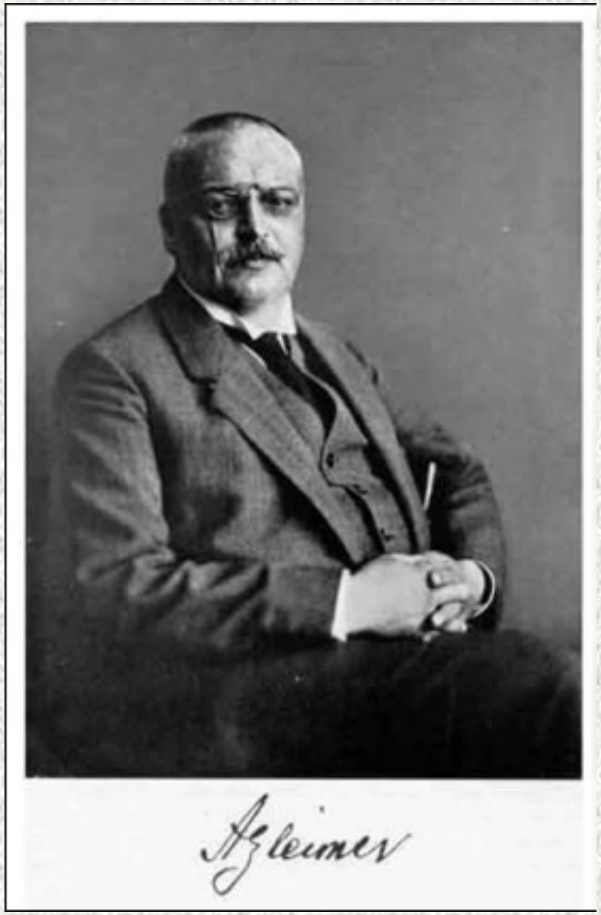


Alteraciones en la homeostasis de Ca^{2+} en la enfermedad de Alzheimer




ALZHEIMER
TUMOR CEREBRAL
PARKINSON
TRAUMA CEREBRAL
Estrés oxidativo
HUNTINGTON
ESCLEROSIS LATERAL AMIOTRÓFICA
EPILEPSIA
HIPOGLICEMIA
ISQUEMIA

Enfermedad de Alzheimer: enfermedad neurodegenerativa caracterizada por pérdida de memoria y demencia catatrófica

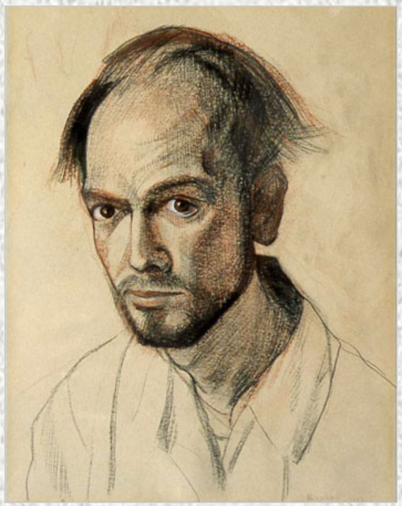


Alzheimer et al.,1907
"Uber eine eigenartige Erkankung der Hirnrinde”
Allgemeine Zeitschrift fur Psychiatrie und Psychisch-gerichtliche Medizin. 64:146-8.

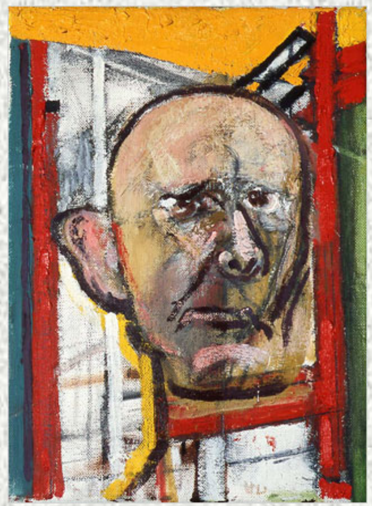
Signos de demencia y problemática

- Pérdida de memoria espacial
- *Pérdida cognitiva*
- *Transtornos del lenguaje*
- *Transtornos de humor*

Auto retrato
(William Utermohlens)



1967

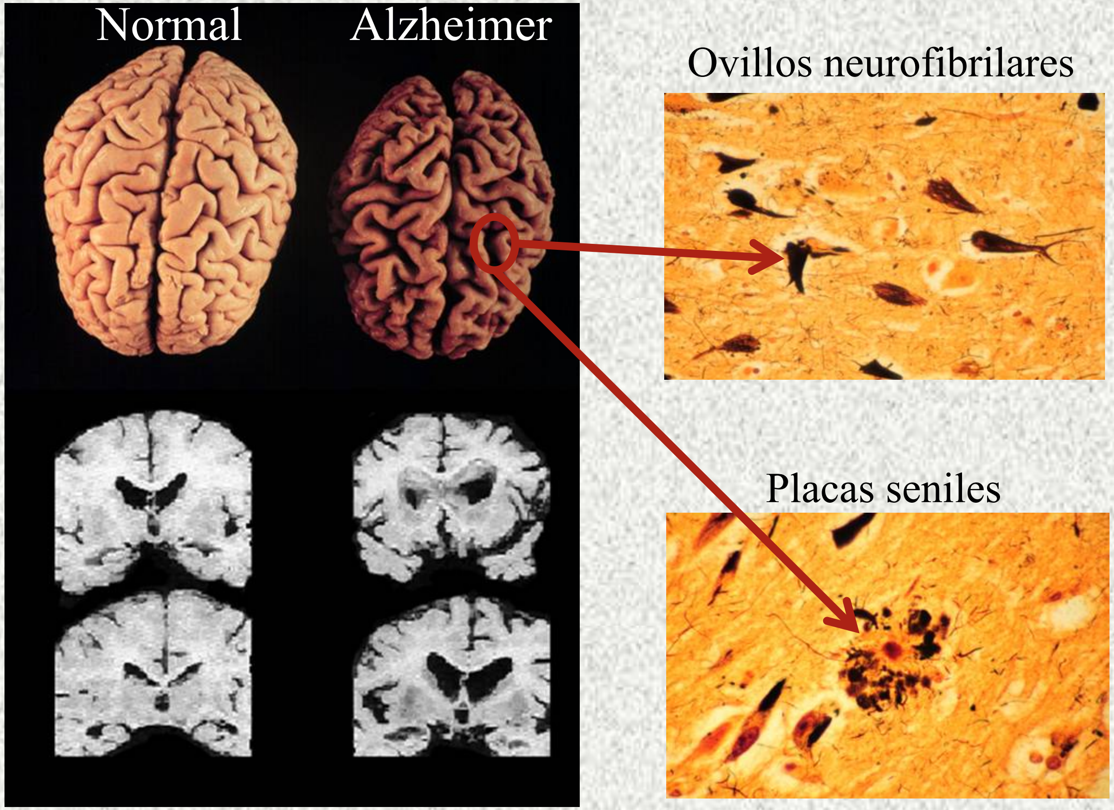


1998

2000

- 1 en cada 10 sobre 65; 1 en cada 2 sobre 85
- Aumento esperado de 5x en 2050
- **Inexistencia de tratamiento curativo**
- **Curso promedio de 10 años desde el diagnóstico**

Marcadores histopatológicos

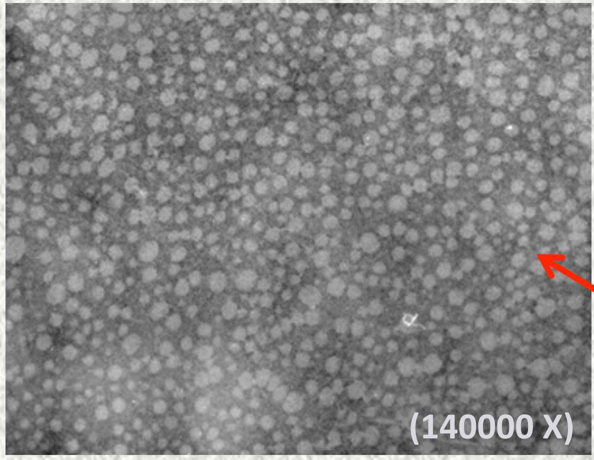


Progresión de las lesiones

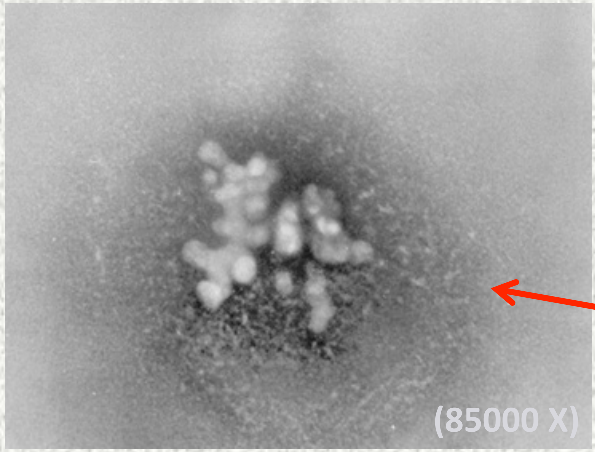
Agregación de Aβ y ganancia de función tóxica

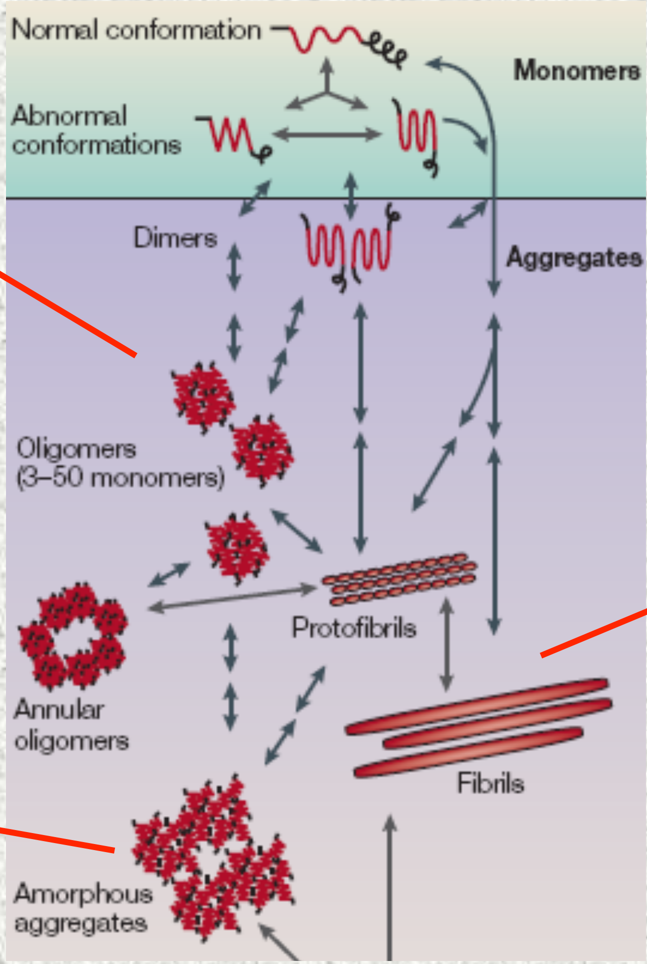
Transmition eletronic microscopy
CESAT
Uchile

Oligômeros de Aβ o AβOs

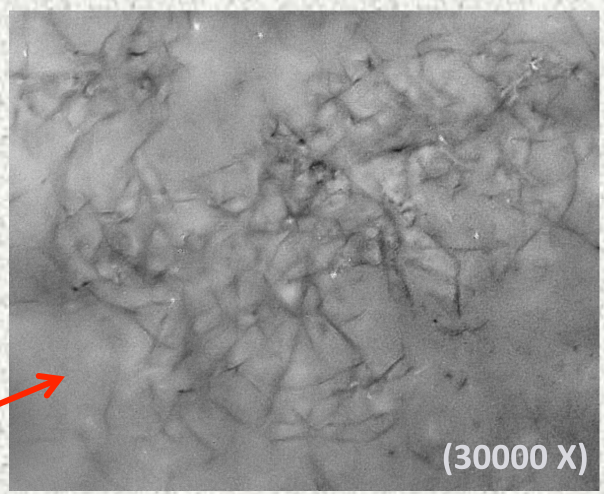


Agregados amorfos





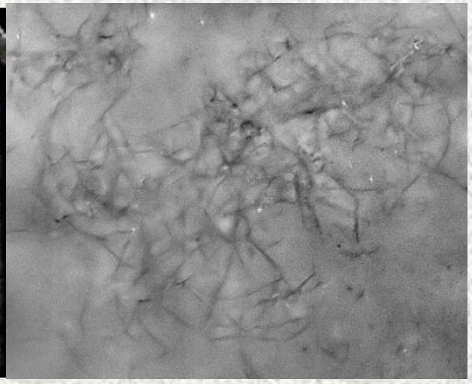
Fibrilas



Ross and Portier, 2005
Nature Reviews in Molecular Cell Biology

Agregados de Aβ, disfunción y muerte neuronal

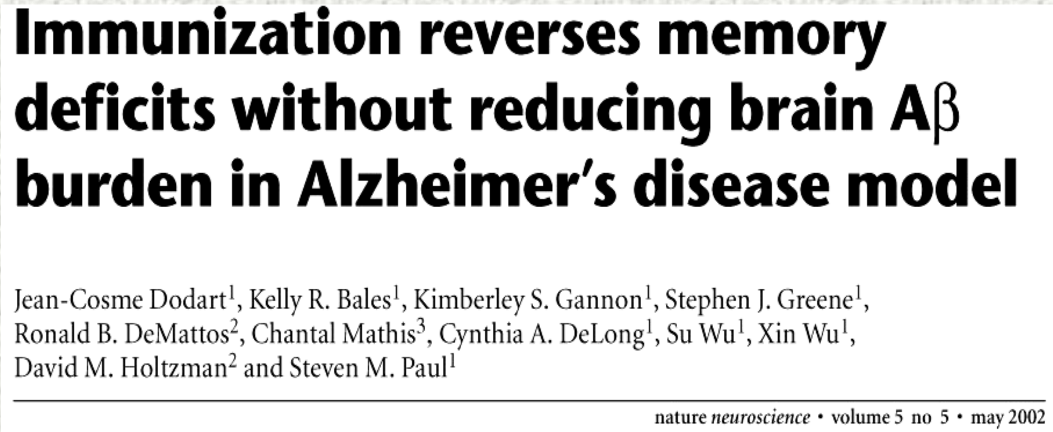
Fibrillas amiloides son neurotóxicas



- Louzada, Paula-Lima et al., 2001, *Neurosci. Letters*
- Paula-Lima et al., 2003, *Neurotoxicity Res.*
- Louzada, Paula-Lima et al., 2004, *FASEB J.*
- Paula-Lima et al., 2005, *Neuropharmacology*.
- Paula-Lima et al., 2009, *Int. J. Biochem. Cell Biol.*

AD = *muerte neuronal causada por depósitos fibrillas amiloides.*

Sin embargo…



Immunization reverses memory deficits without reducing brain Aβ burden in Alzheimer's disease model

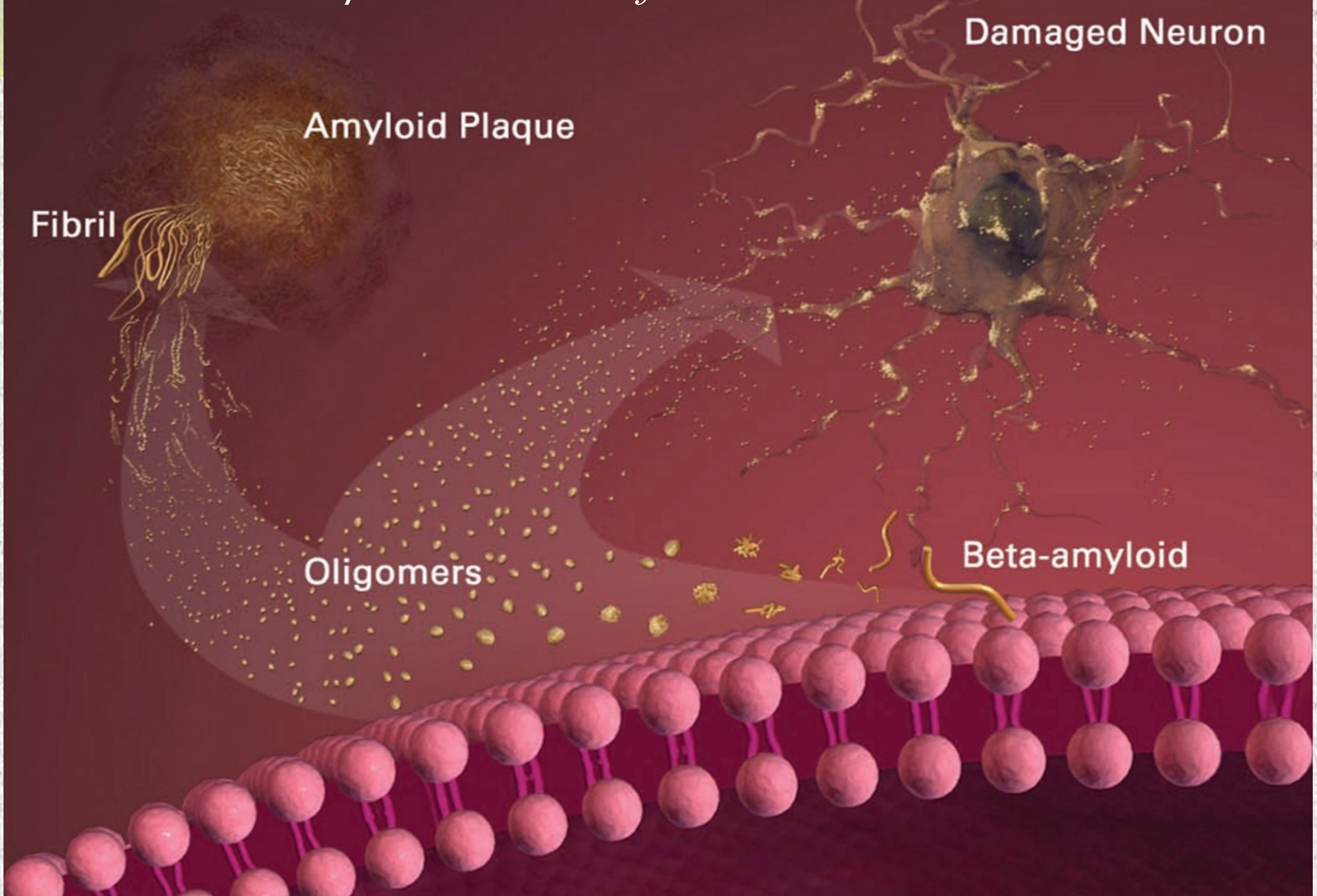
Jean-Cosme Dodart[1], Kelly R. Bales[1], Kimberley S. Gannon[1], Stephen J. Greene[1], Ronald B. DeMattos[2], Chantal Mathis[3], Cynthia A. DeLong[1], Su Wu[1], Xin Wu[1], David M. Holtzman[2] and Steven M. Paul[1]

nature *neuroscience* • volume 5 no 5 • may 2002

Reversible => *¿muerte neuronal?*

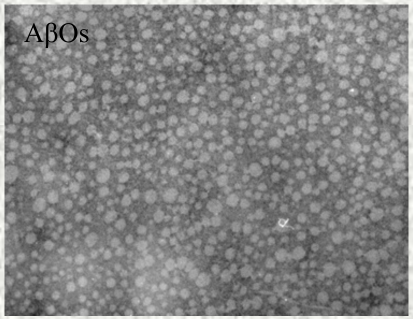
¿Cómo explicar la recuperación de la memoria con la imunoterapia?

AβOs dañan la función neuronal

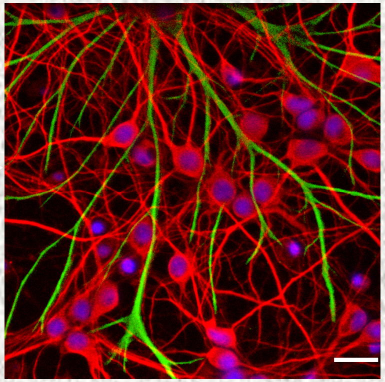


Health & Human Service Progress Report on AD, 2004-2005

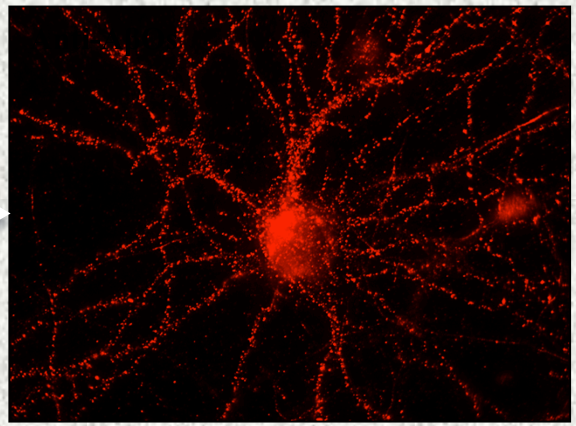
Los AβOs se unen a la superficie neuronal



Cultivo primario de neuronas hipocampales

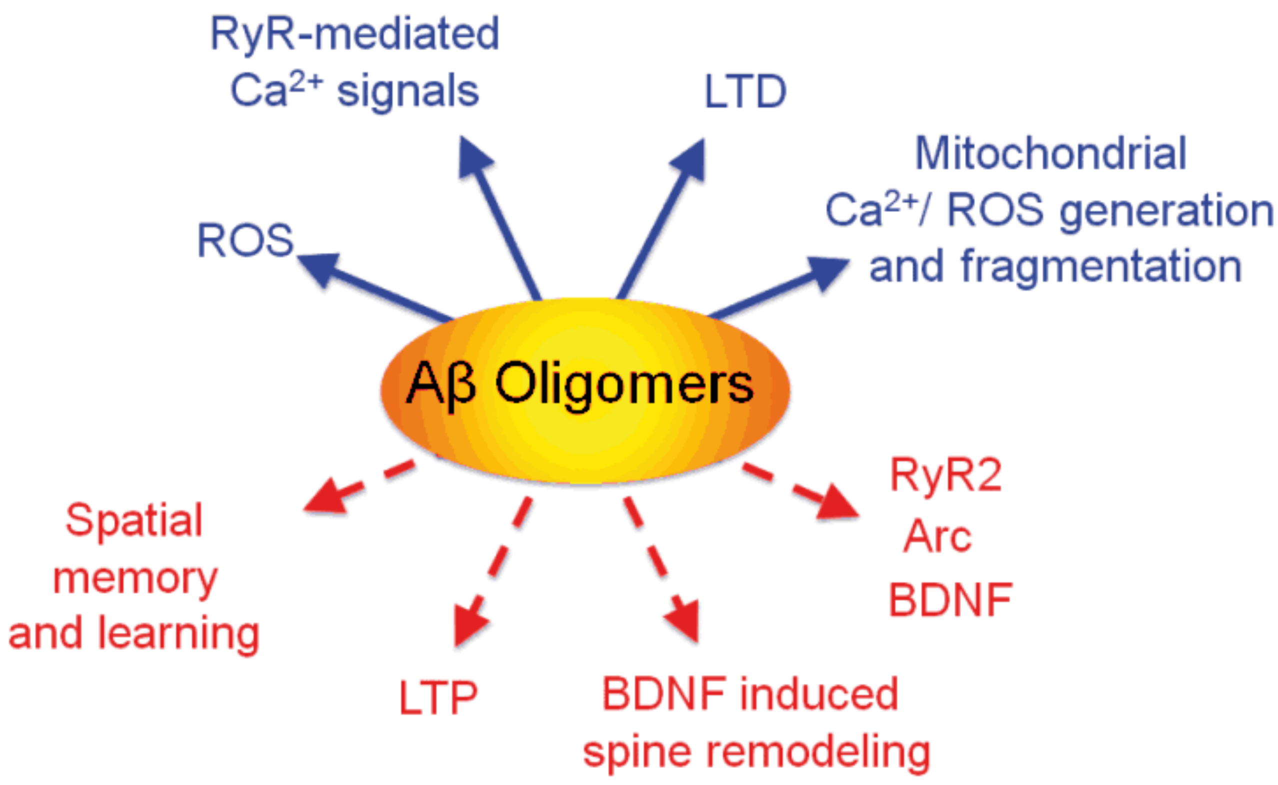


Adasme et al., 2011



De Felice et al., 2009

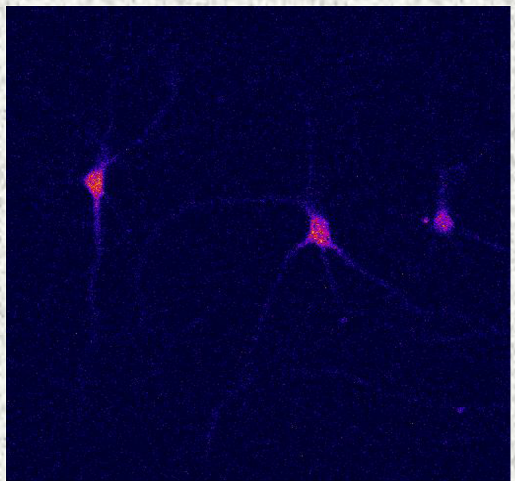
Efectos de los AβOs sobre diferentes funciones hipocampales

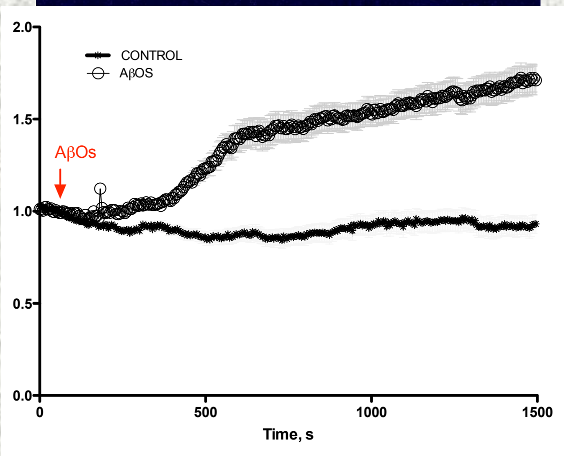


Paula-Lima et al., *Accepted*
Antiox. & Redox Signaling

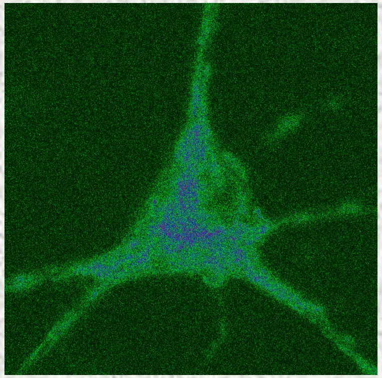
Los AβOs inducen la generación de señales de Ca^{2+} y de ROS

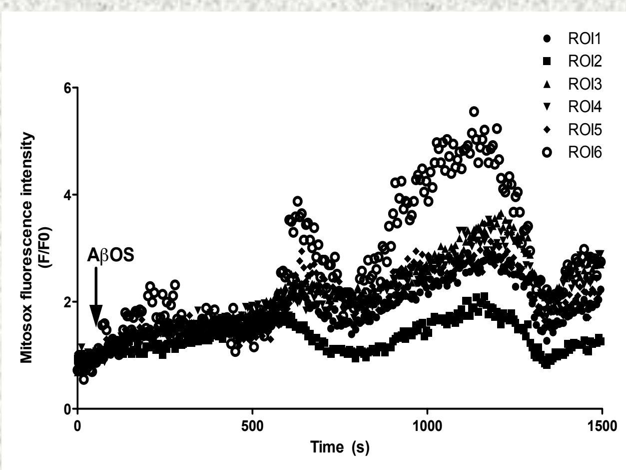
Video: Ca^{2+}-signals elicited by AβOs
Fluo 4 fluorescence



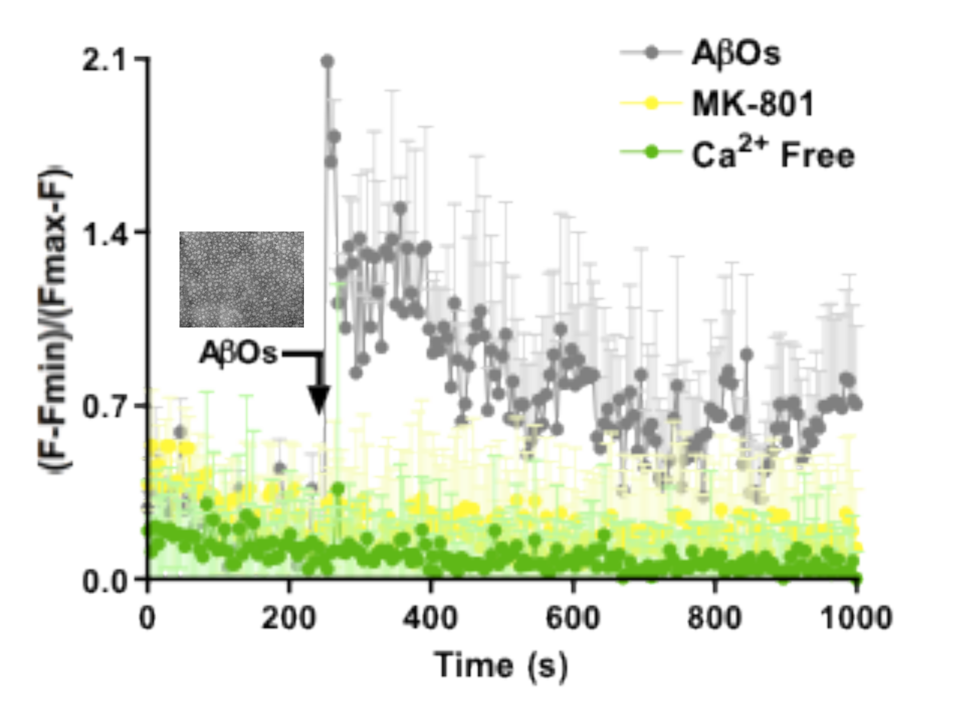


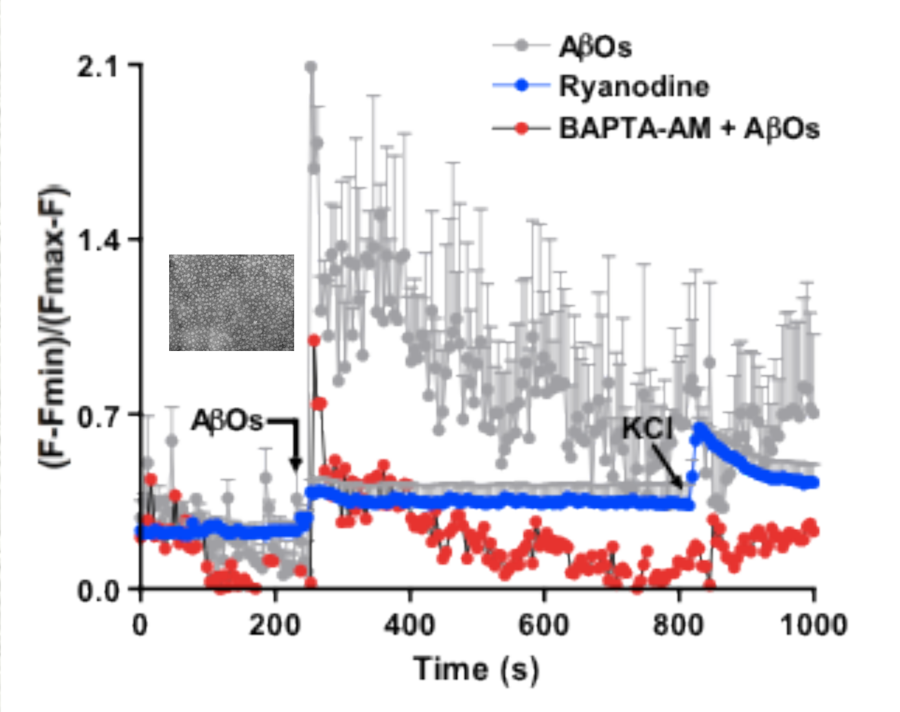
Video: O_2^--signals elicited by AβOs
Mitosox fluorescence





Las señales de Ca^{2+} generadas por AβOs requieren NMDAR y RyR funcionales



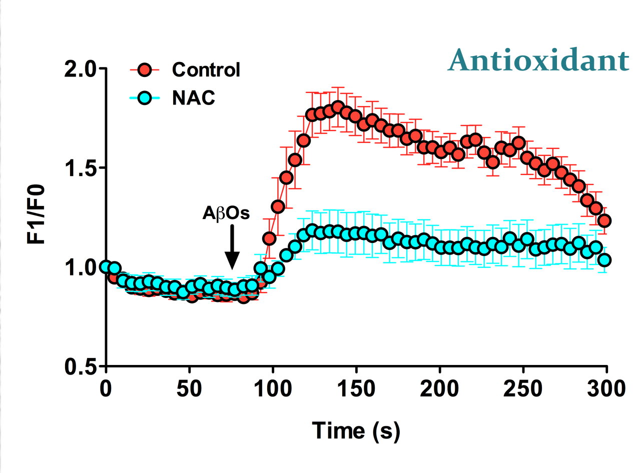


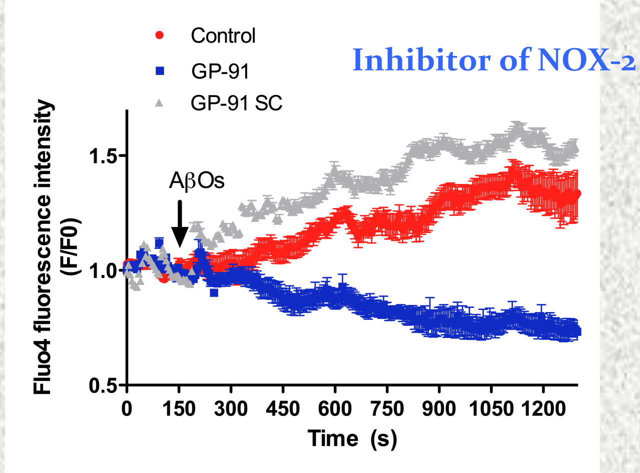
Paula-Lima et al,2011
Antiox. & Redox Sig.

Los AβOs inducen generación de Ca^{2+} dependente de ROS y generación de ROS dependente de señales de Ca^{2+}

ROS-dependent citoplasmic Ca^{2+}signals

Fluo4



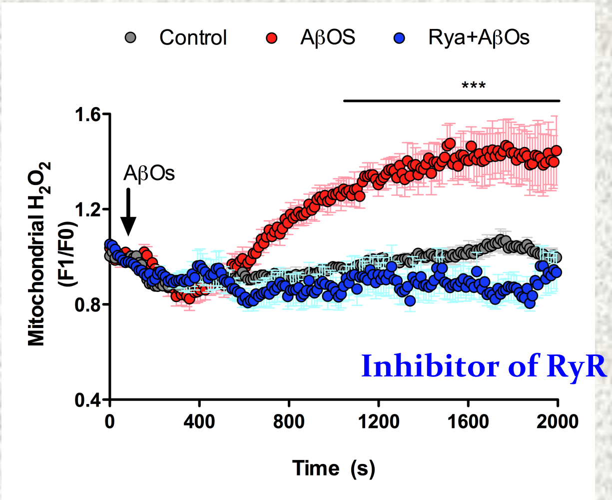


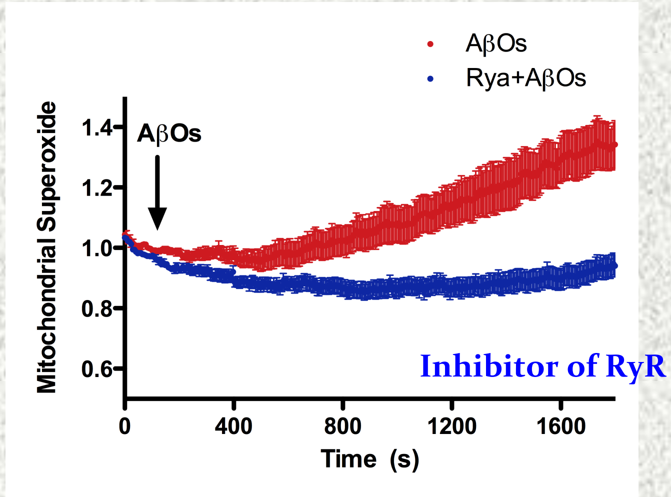
Paula-Lima et al., 2011, *Antiox. & Redox Sig.*,
San Martin et al., *Neurodegenerative diseases*

Ca^{2+}-dependent mitochondrial ROS generation

Hypermito protein

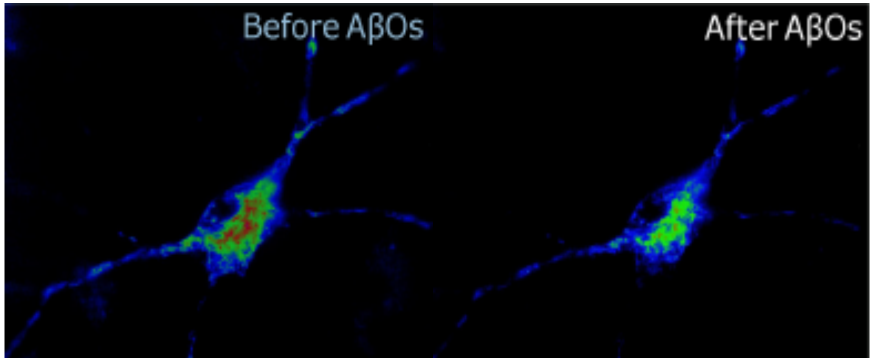
Mitosox

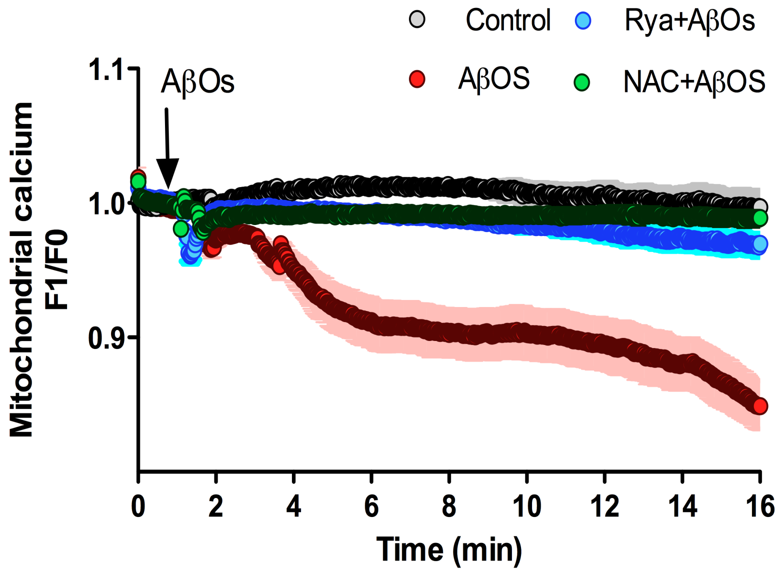




Los AβOs inducen ingreso de Ca^{2+} a la mitocondria en forma dependente de RyR

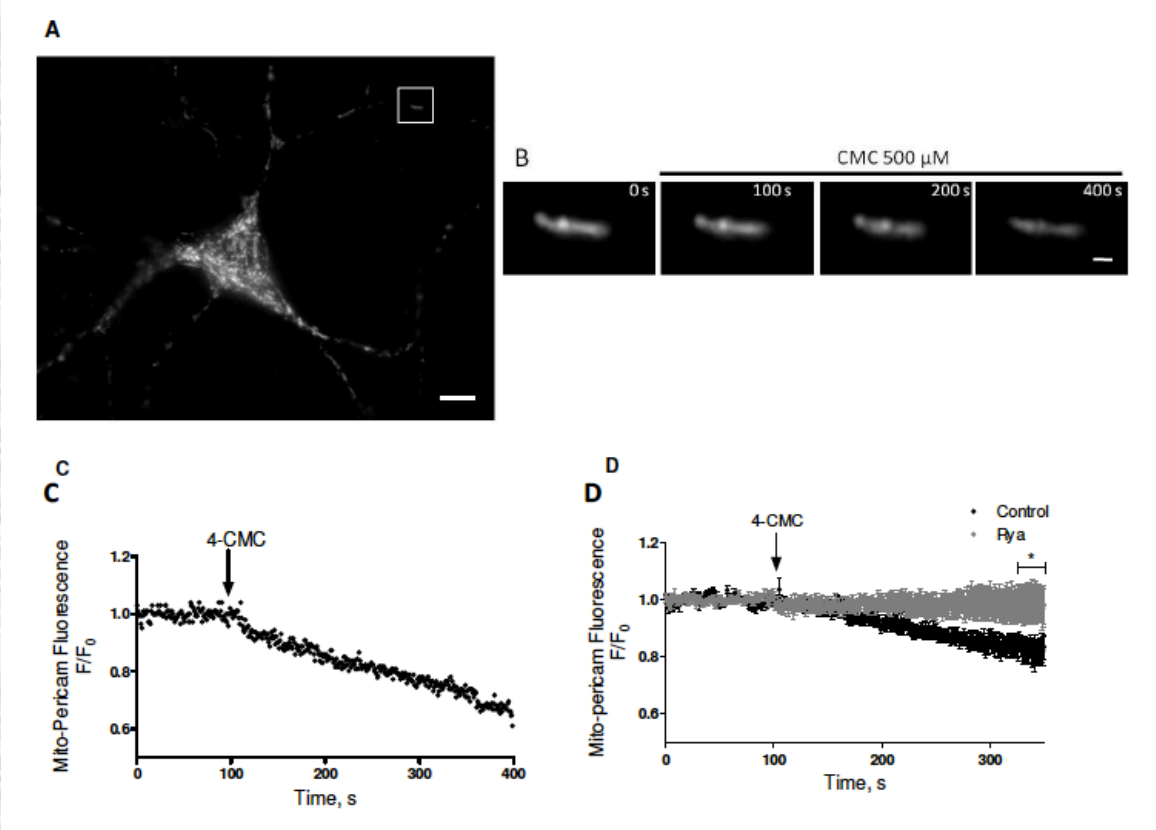
Mitopericam:
Mitochondrial calcium uptake





Unpublished results

Los niveles de Ca^{2+} mitocondrial aumentan en respuesta a la activación directa de RyR



San Martin, Paula-Lima et al, , 2014
Frontiers in Molecular Neurosciences

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Published in final edited form as:
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Mitochondria, calcium and cell death: A deadly triad in neurodegeneration

Fulvio Celsi[a,b], **Paola Pizzo**[c], **Marisa Brini**[d,e], **Sara Leo**[a,b], **Carmen Fotino**[a,b], **Paolo Pinton**[a,b], and **Rosario Rizzuto**[c,*]

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Am J Physiol Cell Physiol 292: C641–C657, 2007.
First published November 8, 2006; doi:10.1152/ajpcell.00222.2006.

Invited Review

Mitochondria and neuronal activity

Oliver Kann and Richard Kovács
Institute for Neurophysiology, Charité-Medical University of Berlin, Germany

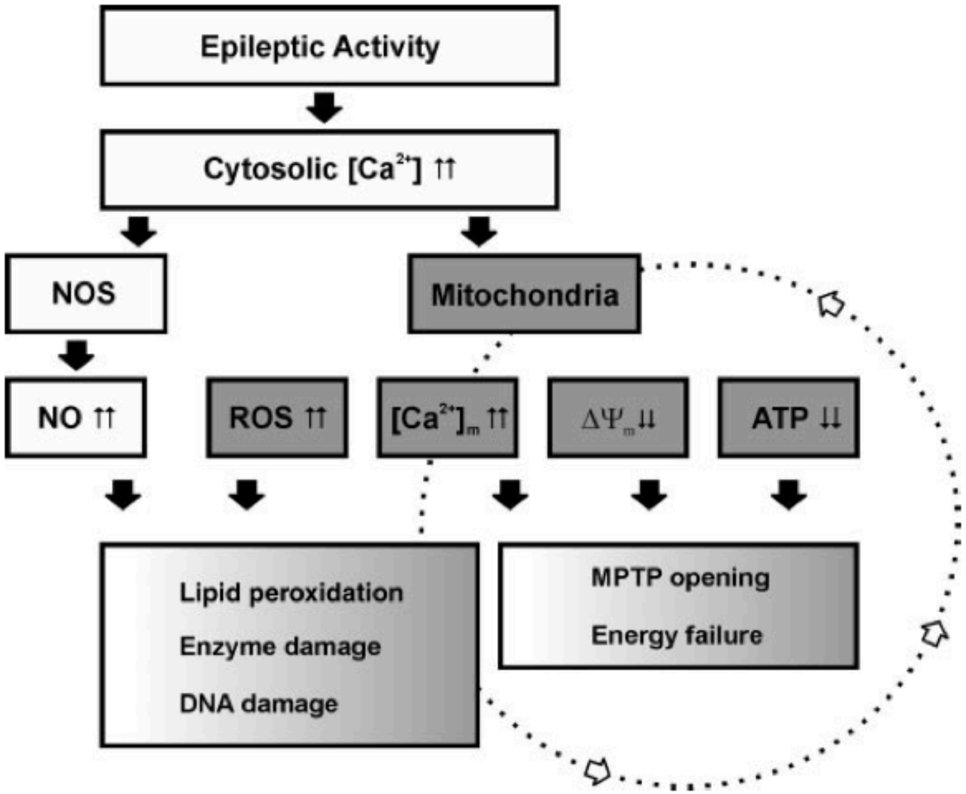
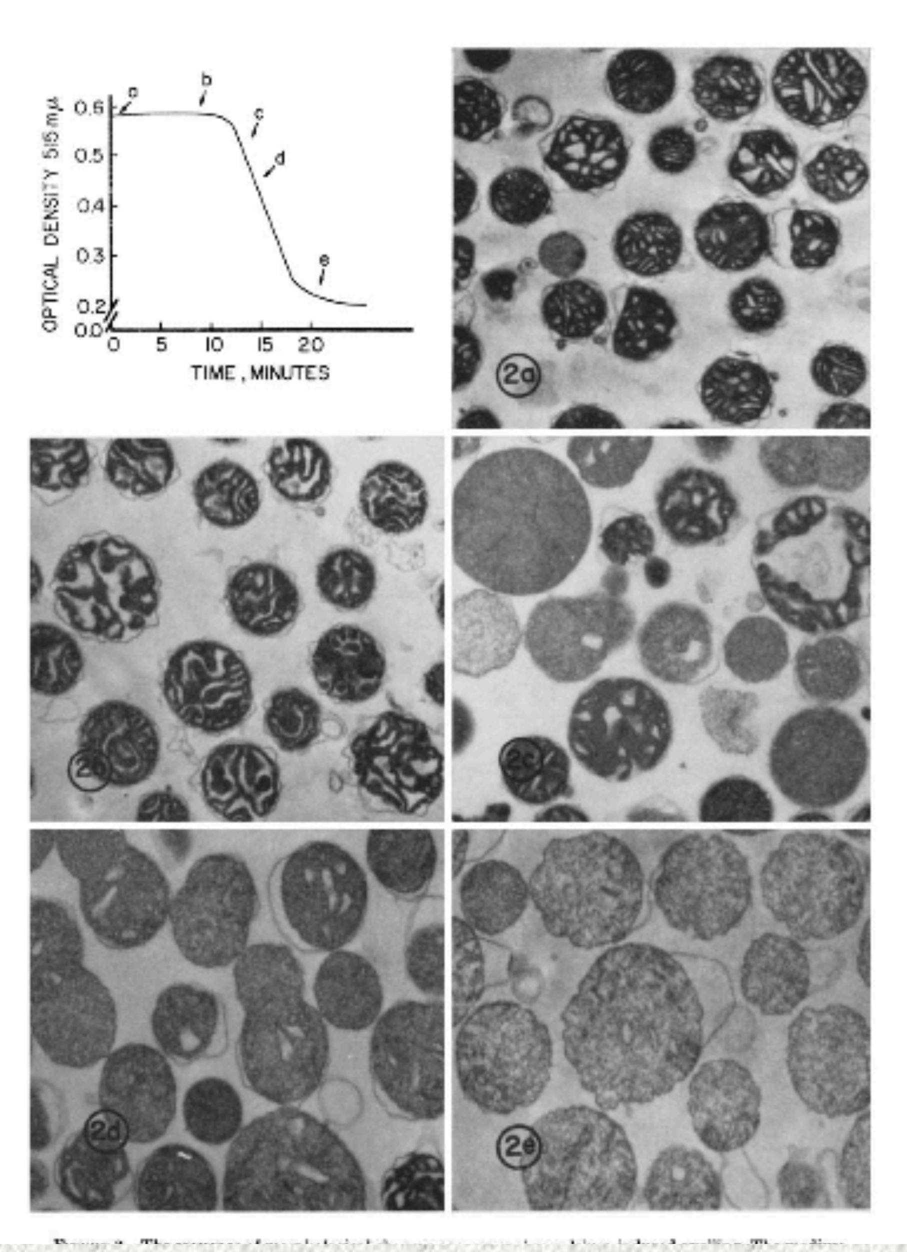
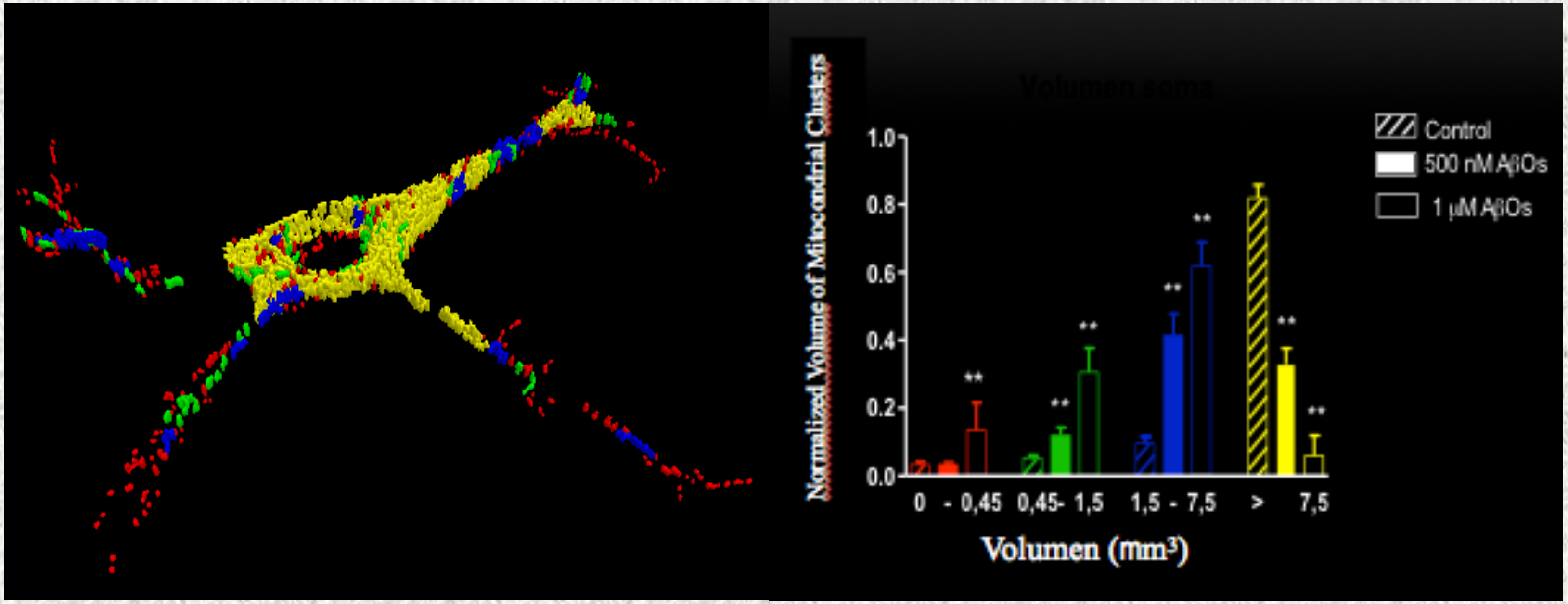


Fig. 2. Scheme illustrating the pathological mechanisms in cytoplasm (light gray) and mitochondria (dark gray), as well as their consequences that might contribute to neuronal cell injury and death in temporal lobe epilepsy. Cellular and mitochondrial DNA and/or enzyme damage might enhance mitochondrial vulnerability in a vicious cycle (dotted circle). NO (nitric oxide), NOS (NO synthase).

Cambios morfológicos mitocondriales que acompañan el aumento de los niveles de Ca^{2+} mitocondriales



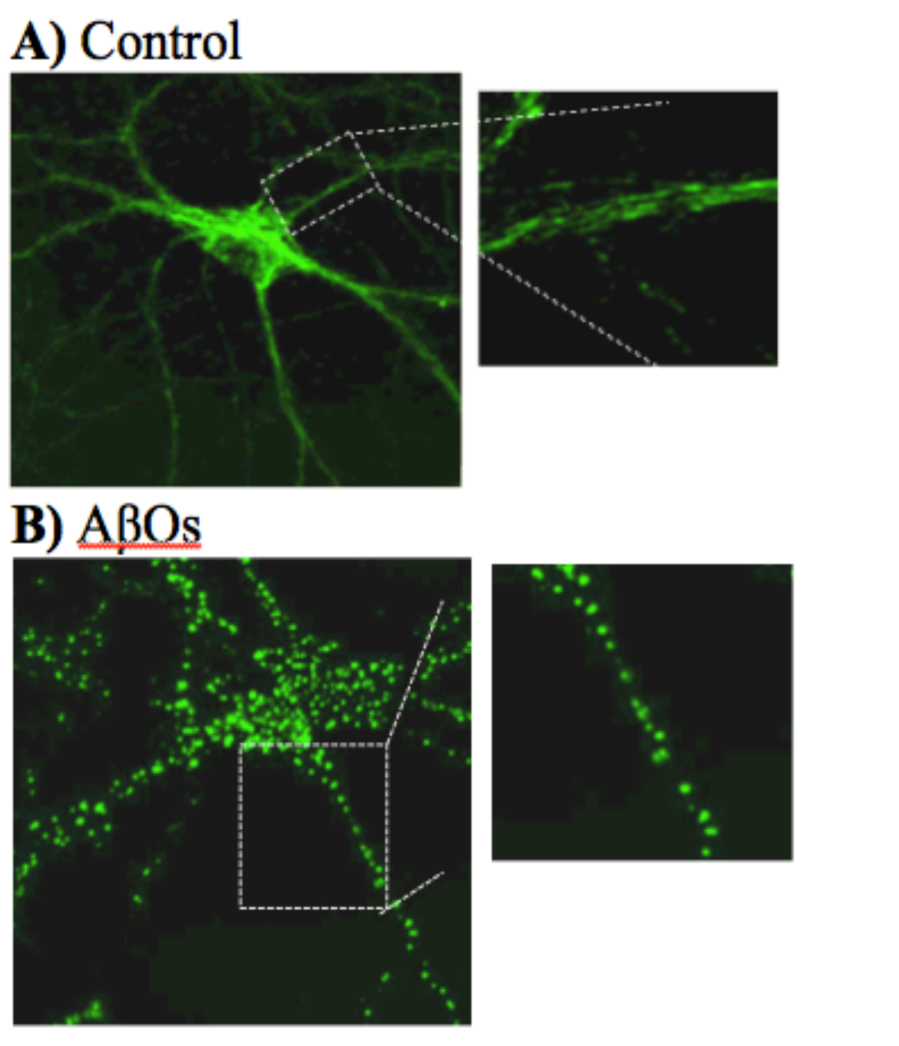
Los AβOs promueven la fragmentación de la red mitocondrial



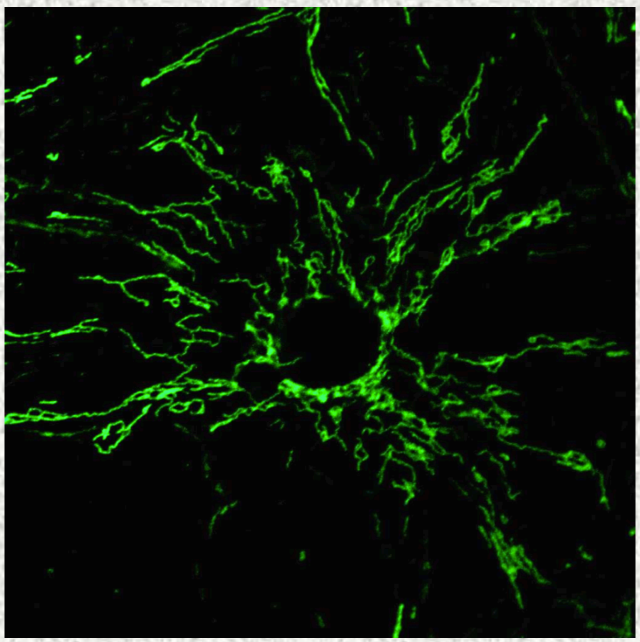
Red : 0- 0,45 mm³
Blue: 1,5- 7,5 mm³
Green: 0,45-1,5 mm³
Yellow: >7,5 mm³

Los AβOs inducen fragmentación de la red mitocondrial neuronal

Neuronal mitochondrial fragmentation

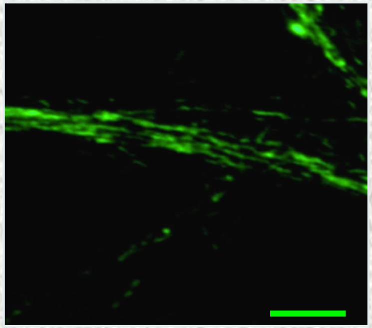


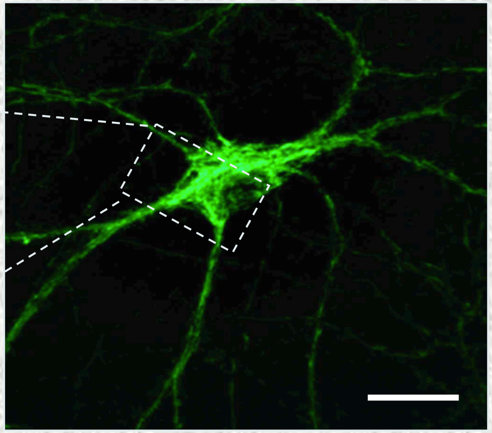
Glial mitochondrial network remains filamentous after treatment with AβOs



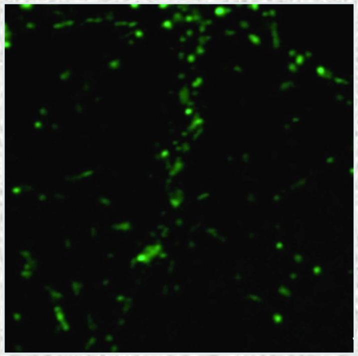
Fragmentación mitocondrial en cuerpo celular y neuronas

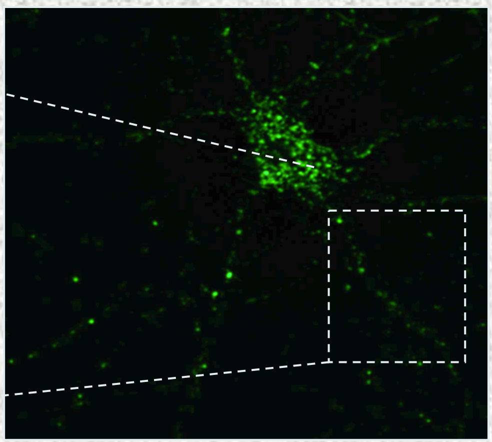
Control





500 nM AβOs 24 h

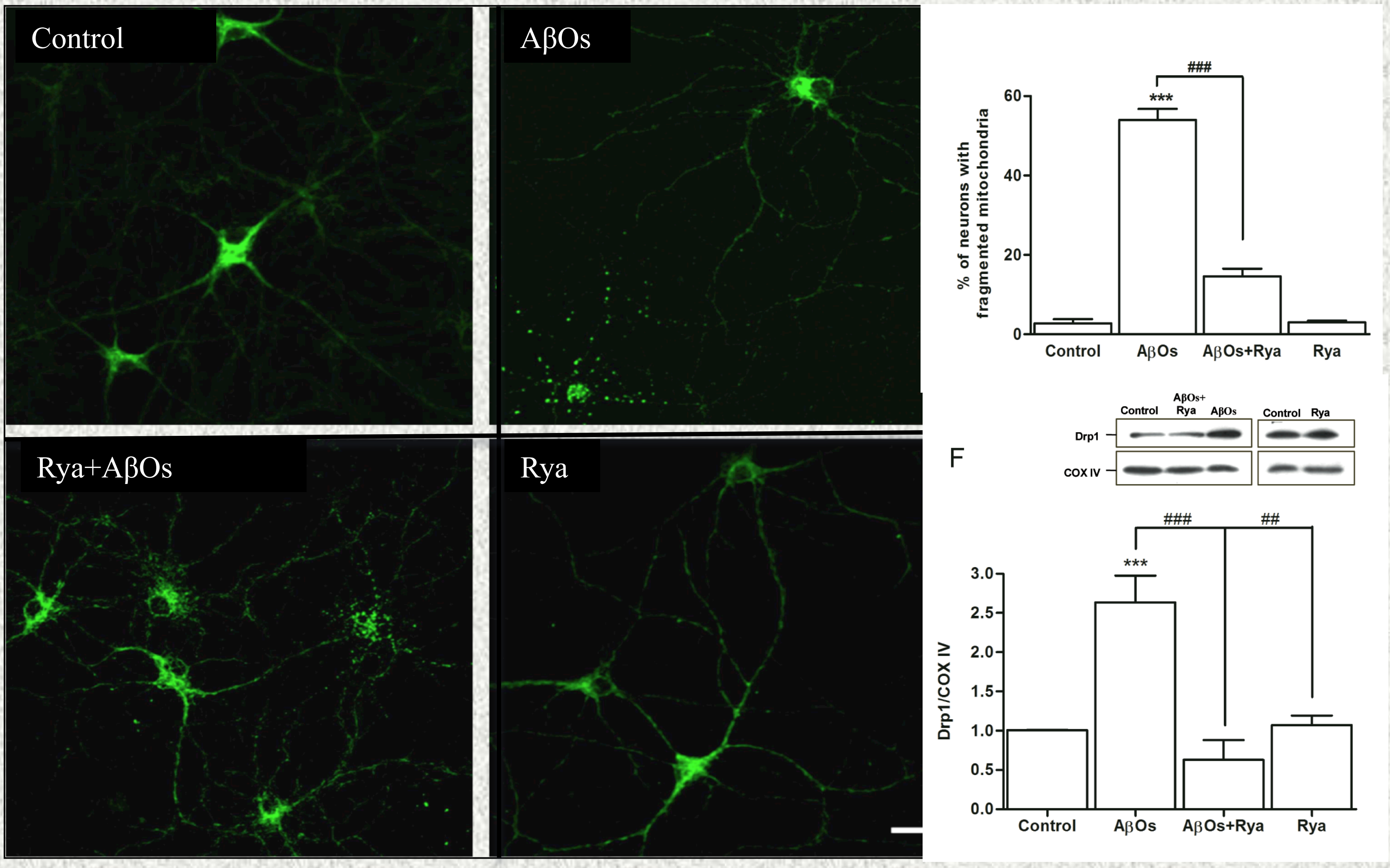




10 um

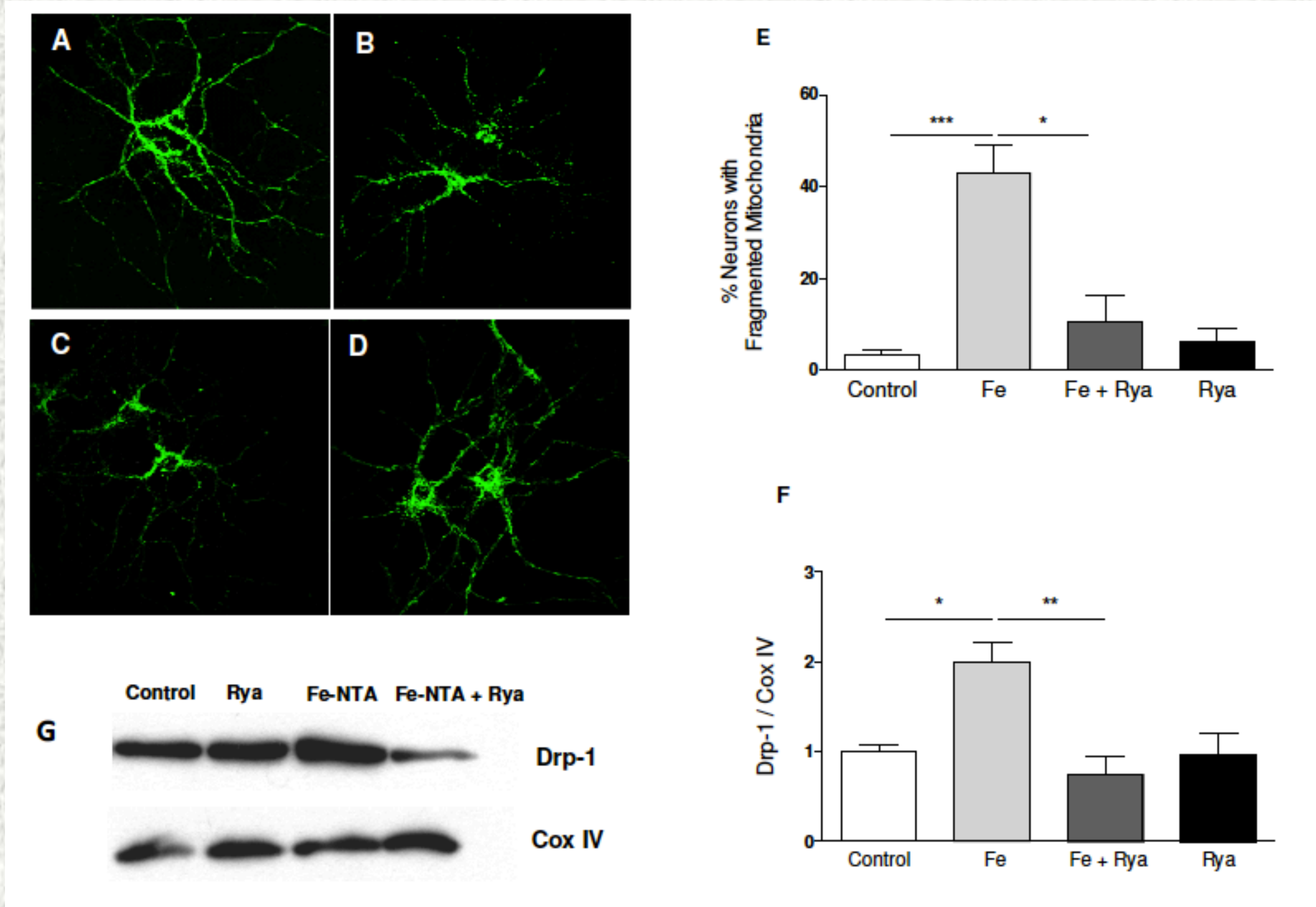
5 um

RyR inhibition prevents AβOs-induced mitochondrial network fragmentation



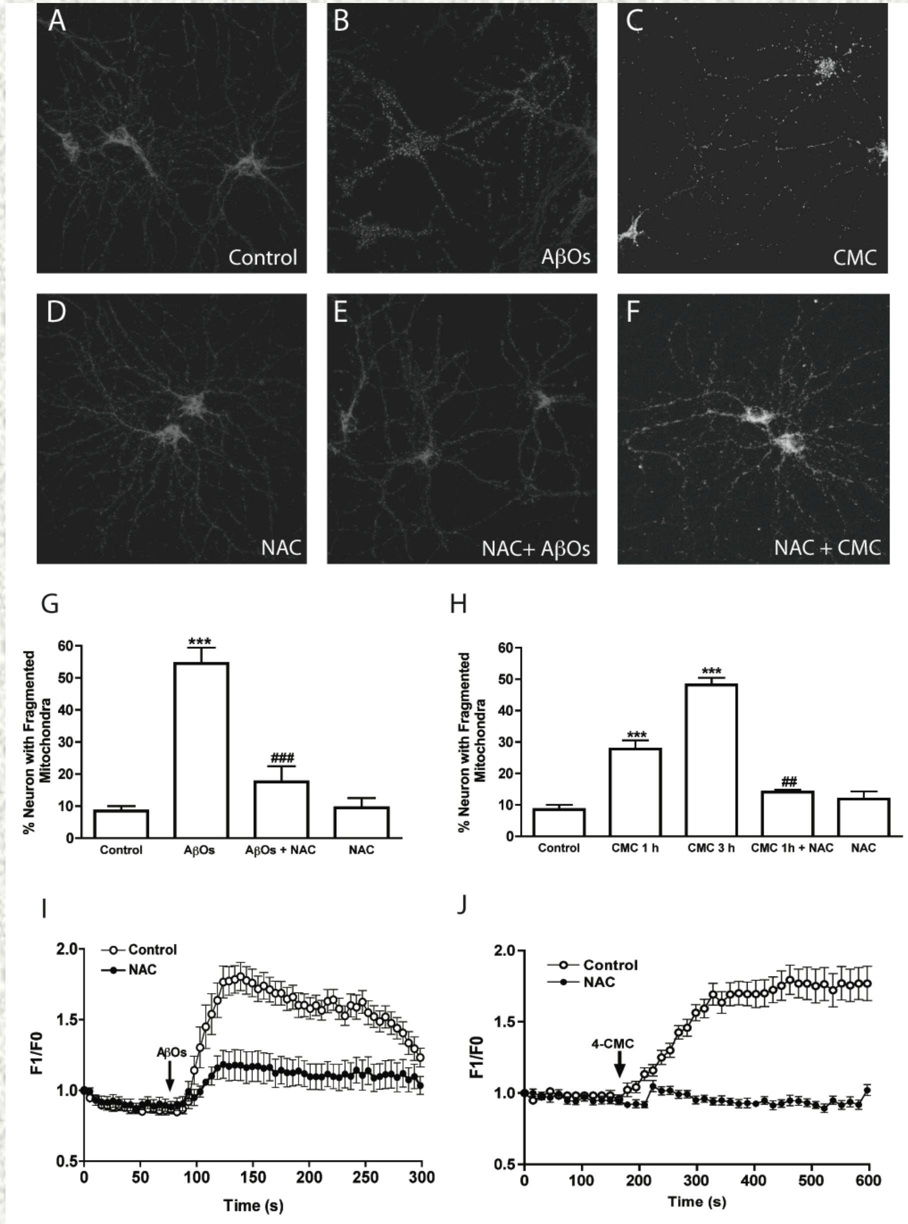
Paula-Lima et al., 2011
Antiox. & Redox. Sig.

El hierro iduce fragmentación de la red mitocondrial de forma dependente de la liberación de Ca^{2+}



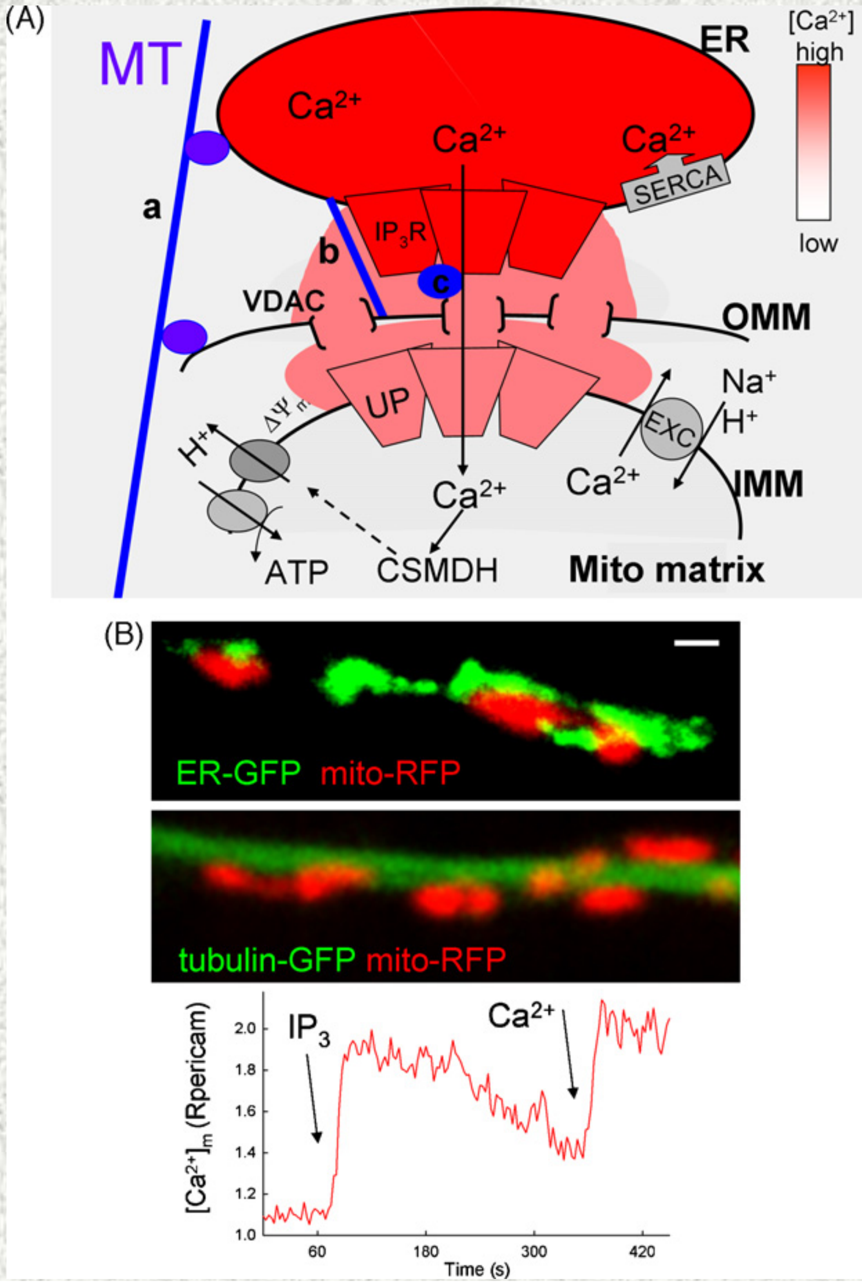
San Martin, Paula-Lima et al, , 2014
Frontiers in Molecular Neurosciences

El antioxidante NAC previene la fragmentación de la red mitocondrial inducidas por los AβOs



San Martin et al., 2012
Neuorodegenerative Dis.

Acoplamiento local del ER y mitocondria



Skeletal muscle

Spat et al, 2008
Cell Calcium

Gleishmann and Mattson, 2011
Antiox. & Redox Sig.

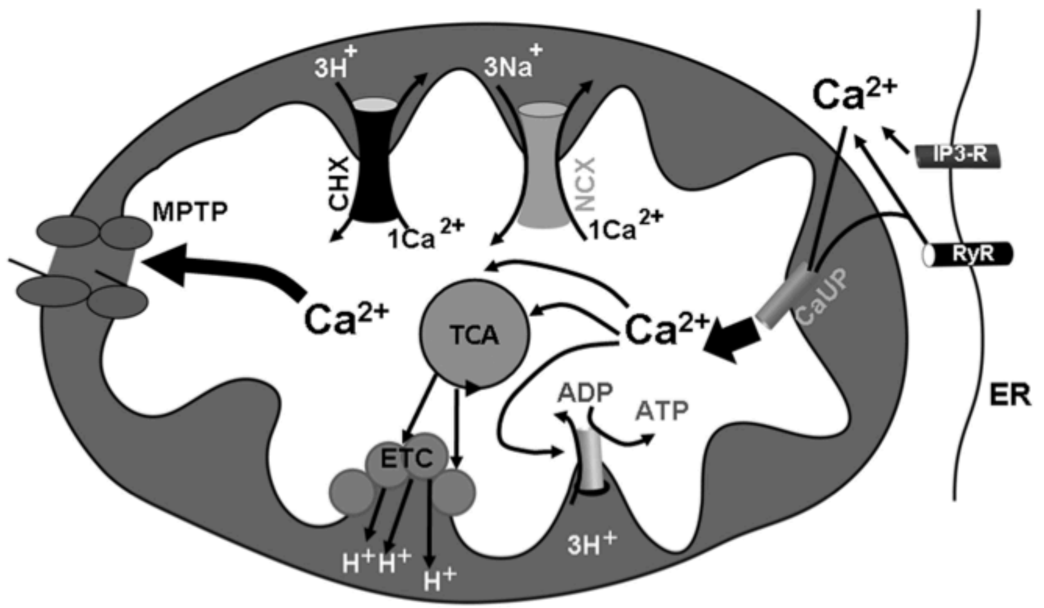
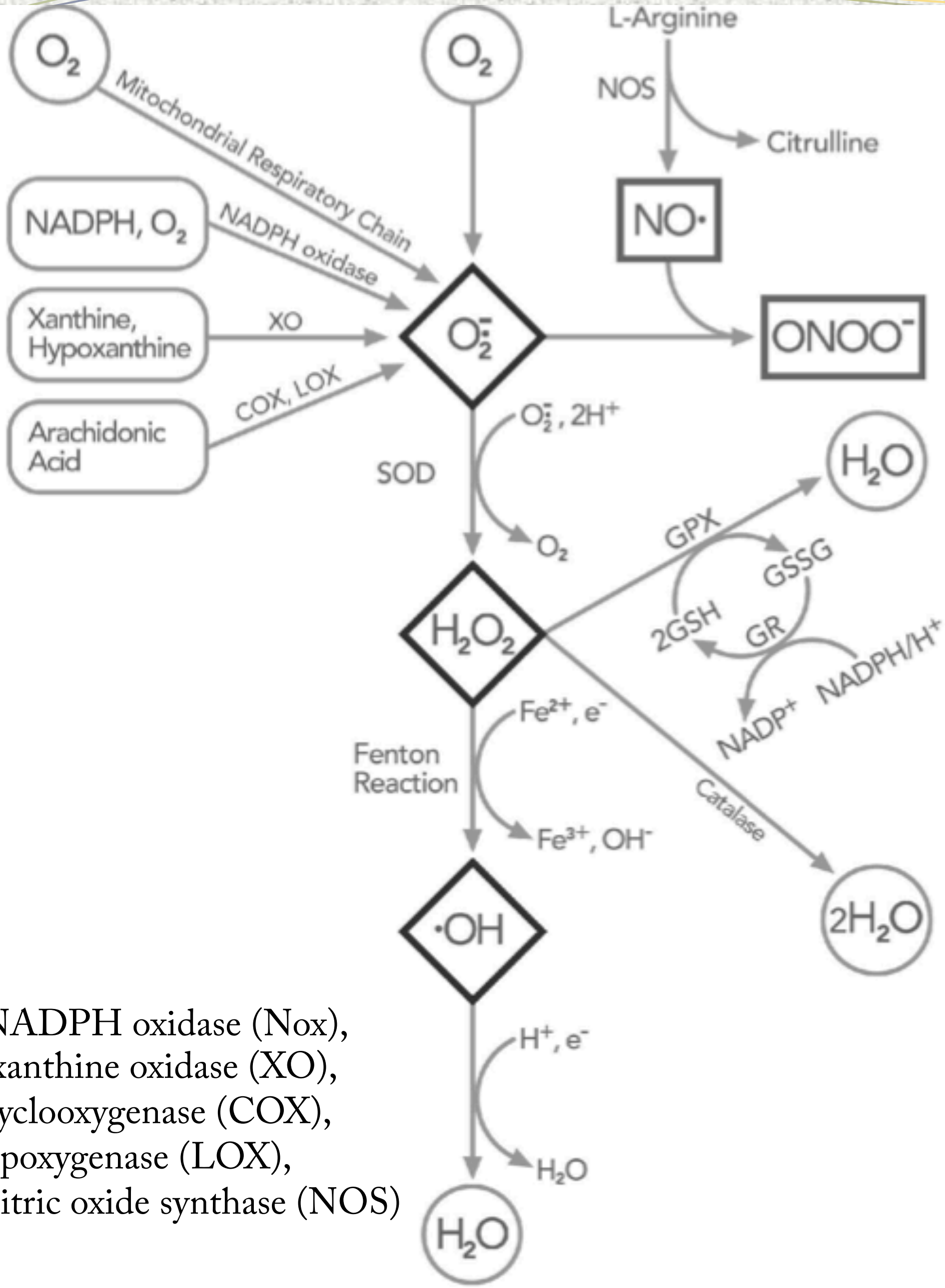


Figure 1. **Mechansims of mitochondrial Ca^{2+} influx and efflux.** Ca^{2+} enters the mitochondrial through the Ca^{2+} uniporter (CaUP). Close apposition of ER Ca^{2+} release channels and Ca^{2+} uniporter are likely to result in enhanced mitochondrial Ca^{2+} uptake. Inside the mitochondrial matrix Ca^{2+} can increase the activity of dehydrogenases of the tricarboxic acid cycle (TCA), leading to enhanced feeding of the electron transport chain (ETC) and increased transfer of protons to the intermembrane space. Mitochondrial Ca^{2+} also activates the F1F0 ATPase to produce more ATP. Ca^{2+} is extruded from the mitochondrial matrix through a sodium-dependent mechanism (sodium calcium exchanger, NCX) and a membrane potential-dependent mechanism (calcium proton exchanger, CHX) as well as the permeability transition pore (mPTP). Note that the molecular identities of CaUP, mitochondrial NCX and CHX are unclear.

Formación de ROS/RNS



NADPH oxidase (Nox),
xanthine oxidase (XO),
cyclooxygenase (COX),
lipoxygenase (LOX),
nitric oxide synthase (NOS)

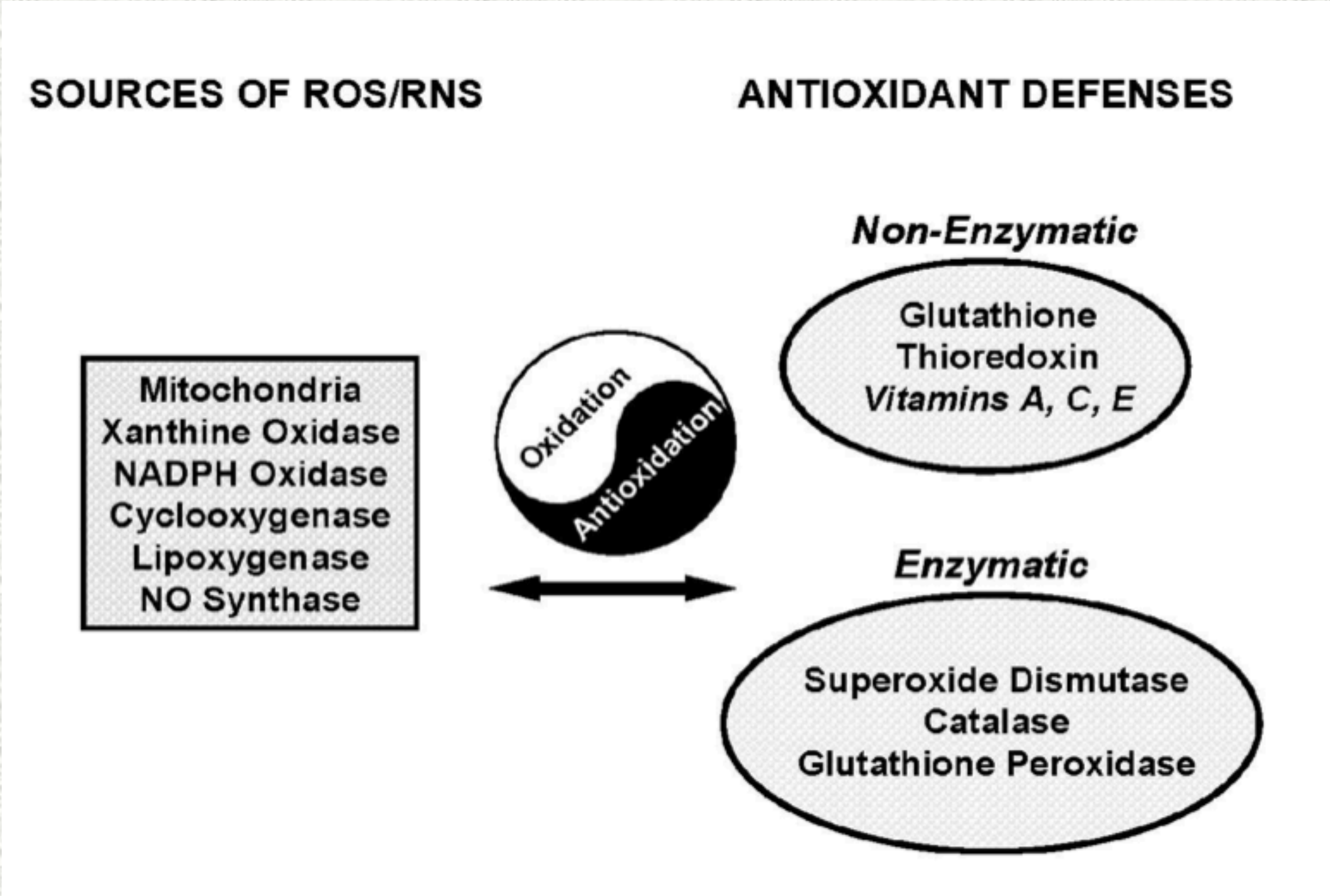
The mitochondrial respiratory chain and enzymes systems that include Nox, XO, COX and LOX, generate superoxide anion from molecular O2.

Superoxide can dismutate into H2O2 spontaneously or enzymatically through the action of SOD, or can react with NOS-produced NO to generate peroxynitrite.

The enzymes GPX and catalase can convert H2O2 into water. Alternatively, H2O2 can react with redox active metal ions, such as Fe2+, to generate the hydroxyl radical.

Hidalgo & Donoso, 2008
Antiox. & Redox Sig.

Homeostasis redox



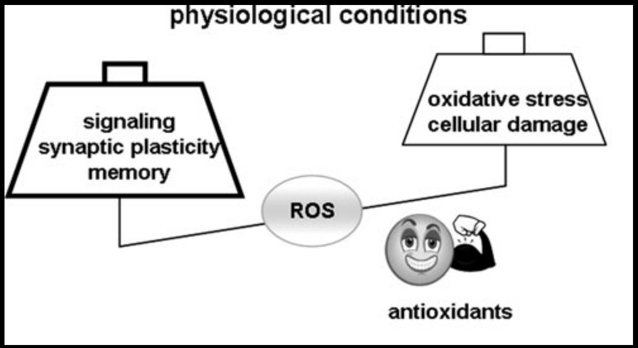
Hidalgo & Donoso, 2008
Antiox. & Redox Sig.

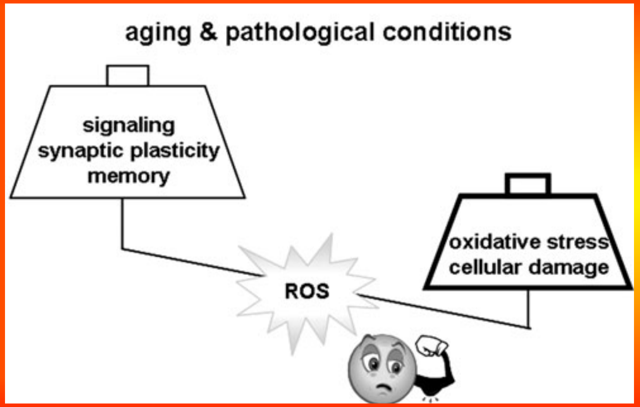
ROS

The brain has high oxygen consumption, with the consequent generation of significant amounts of reactive oxygen species (ROS).

At physiological concentrations, ROS are involved in functional changes necessary for synaptic plasticity and hence, for normal cognitive function.

ROS are an essential signaling component for memory formation





Excessive ROS levels, however, produce oxidative stress and are associated with decreased performance in cognitive function.

Massaad and Klann, ARS 2011

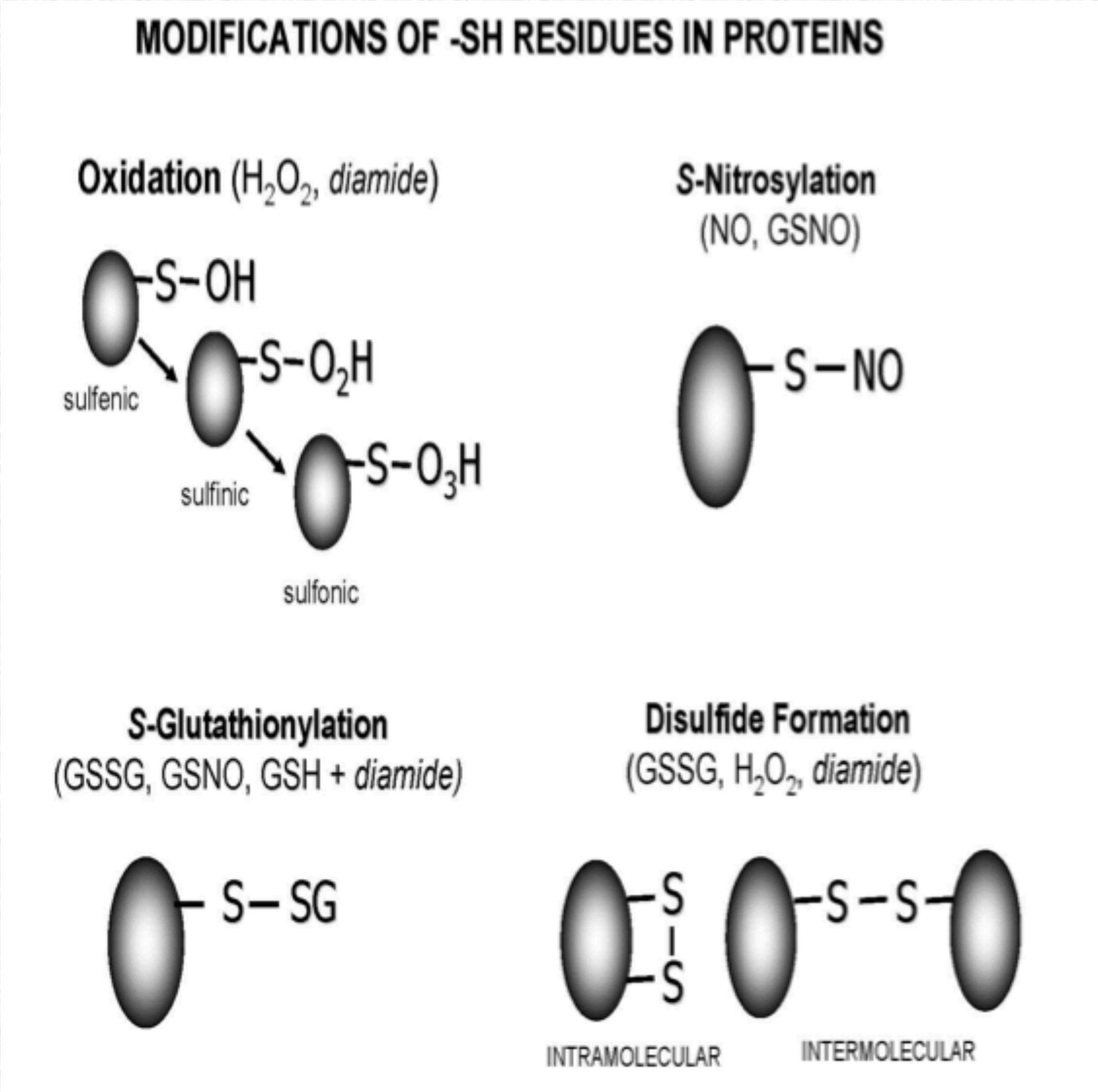
ROS y RNS: función and disfunción

- *ROS and RNS, including nitric oxide (NO) gas, are short-lived small molecules, which depending on their reactivity can reach only immediate targets, as happens with the hydroxyl radical, or can diffuse freely within cells or even outside cells as does NO gas.*

- *It has become increasingly apparent that ROS/RNS serve a function as signaling molecules in various biological responses, which include gene expression and cell death, among others.*

- *Posttranslational modifications by ROS/RNS modify the function of many cellular proteins involved in signal transduction, such as protein phosphatases, protein/lipid kinases, and transcription factors calcium channels.*

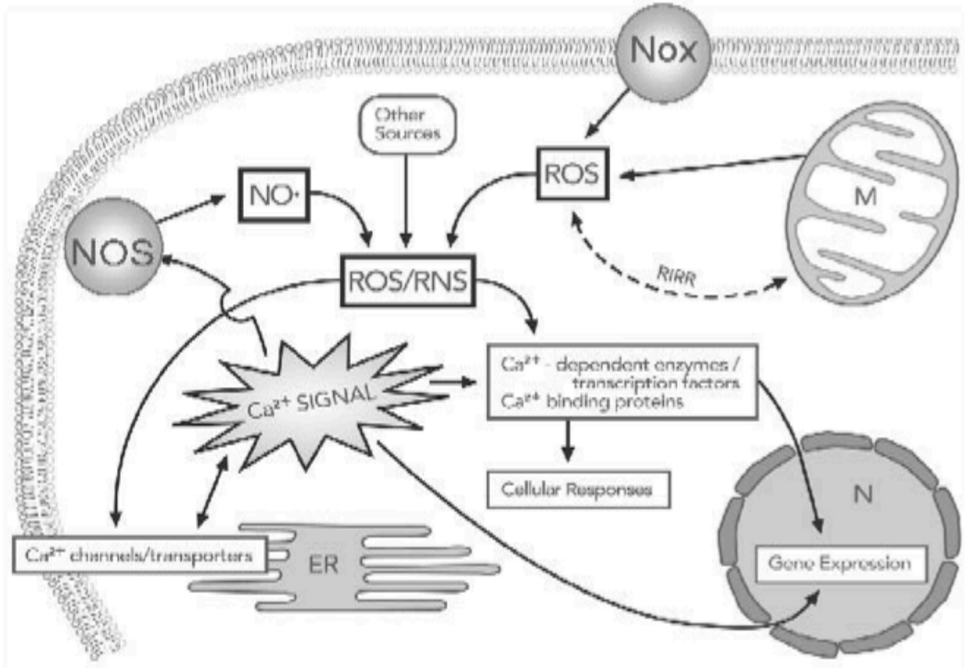
Modificaciones de los residuos en proteínas por ROS/RNS



A range of different **modifications of protein thiol groups**, which include *oxidation to sulfenic, sulfinic and sulfonic derivatives, S-nitrosylation, S-glutathionylation and the formation of intra or intermolecular disulfide bonds.*

Hidalgo & Donoso, 2008
Antiox. & Redox Sig.

Esquema ilustrando la conversación cruzada entre las vías de señalización por Ca^{2+} y ROS/RNS



Hidalgo & Donoso, 2008
Antiox. & Redox Sig.

Localized or global increments in ([Ca^{2+}]i) have the potential to modulate redox signaling pathways in eukaryotic cells.

Similarly, variations in local or global concentrations of reactive oxygen species (ROS) or nitrogen species (RNS) can affect cellular Ca^{2+}-signaling pathways through different mechanisms.

The relevance of this cross talk between Ca^{2+} and redox signaling for normal or pathological cell function is becoming increasingly apparent (Hidalgo & Donoso, 2008).

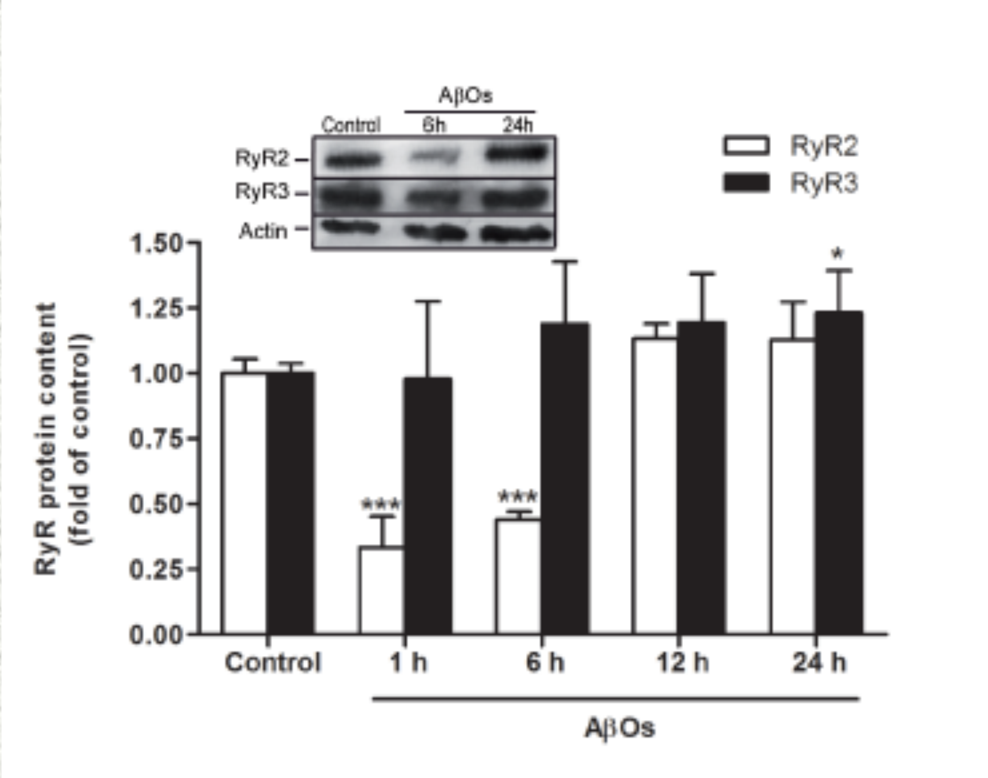
The duality of ROS/RNS as both signaling and stress molecules is exemplified by the effects of these reactive species on synaptic plasticity. At high concentrations, ROS and RNS, such as hydrogen peroxide and hydroxyl radical, attenuate long-term potentiation (LTP) and synaptic neurotransmission (Colton et al., 1989; Pellmar et al., 1991; Gahtan et al., 1998; Avshalumov et al., 2000). This might be partially due to the inhibition of glutamatergic *N*-methyl-D-aspartate (NMDA) receptors by ROS/RNS through excessive oxidation of the extracellular redox-sensitive sites of these receptors (Steullet et al., 2006). On the other hand, at lower ("physiological") levels, ROS such as superoxide and hydrogen peroxide enhance LTP and synaptic neurotransmission (Thiels et al., 2000; Thiels and Klann, 2002; Kamsler and Segal, 2003, 2004).

Hidalgo & Donoso, 2008
Antiox. & Redox Sig.

Oxidative stress (OS), caused by the imbalance between the generation and detoxification of reactive oxygen and nitrogen species (ROS/RNS), plays an important role in brain aging, neurodegenerative diseases, and other related adverse conditions, such as ischemia.

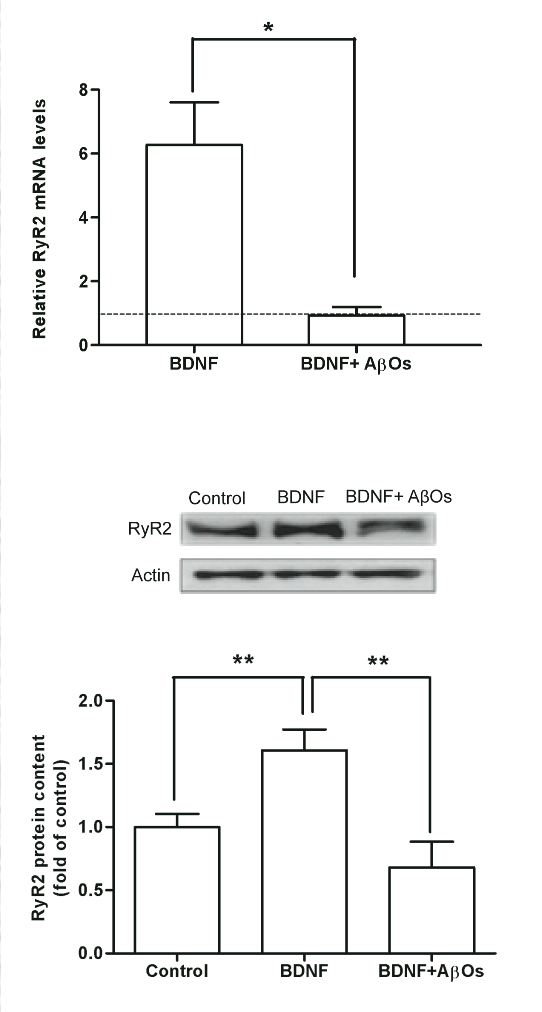
Los AβOs también afectan la expresión génica dependiente de Ca^{2+}

Los AβOs inducen una downregulación rapida y transiente del contenido de la proteína RyR2



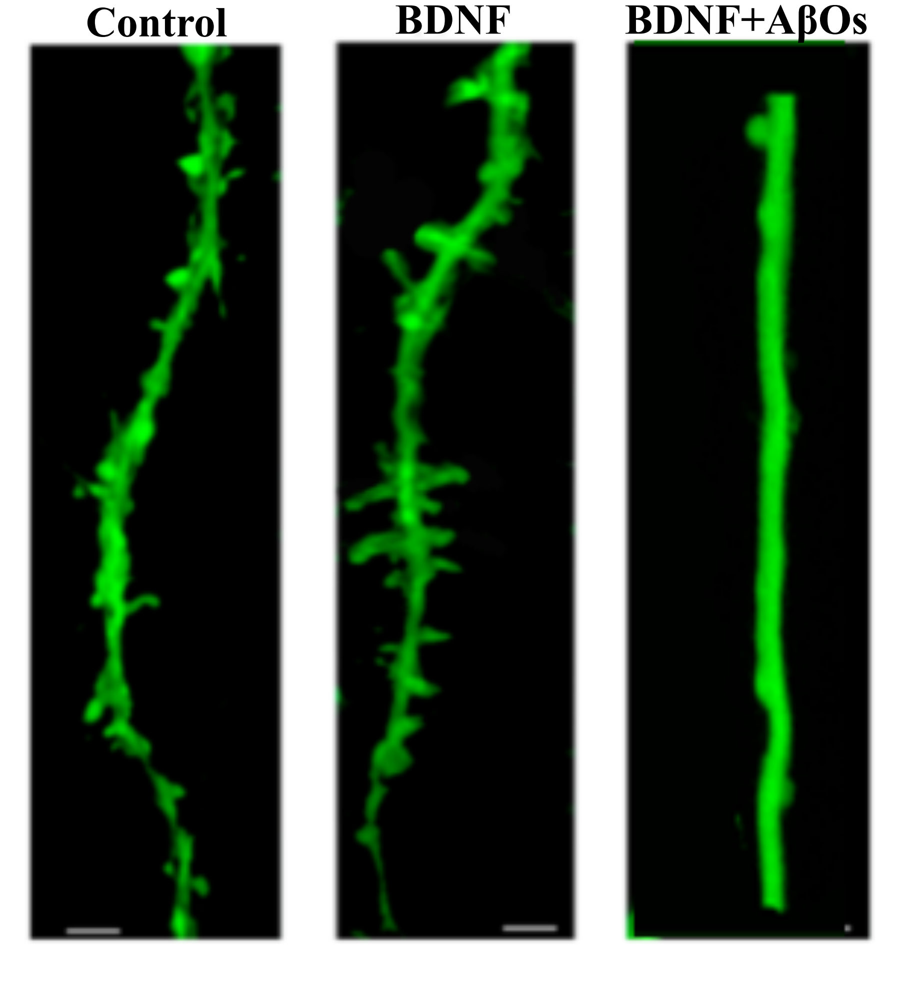
Paula-Lima et al., 2011
Antiox. & Redox. Sig.

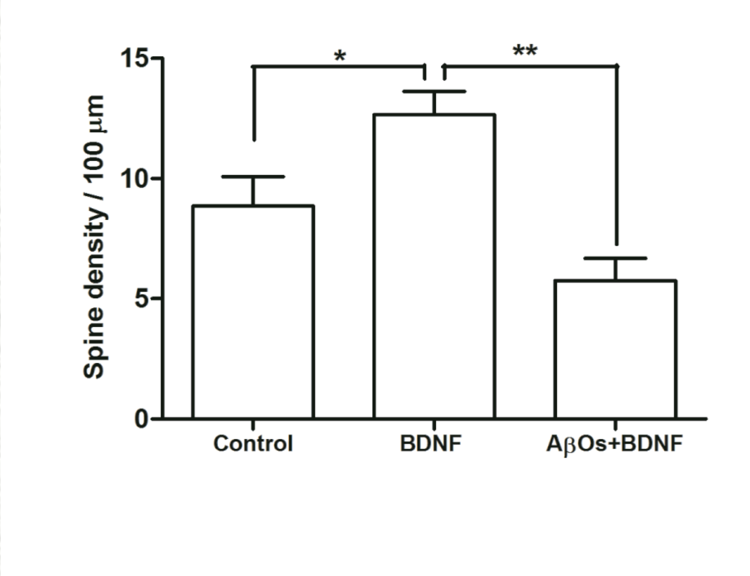
Los AβOs previenen el aumento de la expresión de RyR2 inducida por BDNF



Paula-Lima et al., 2010
Antiox. & Redox. Sig.

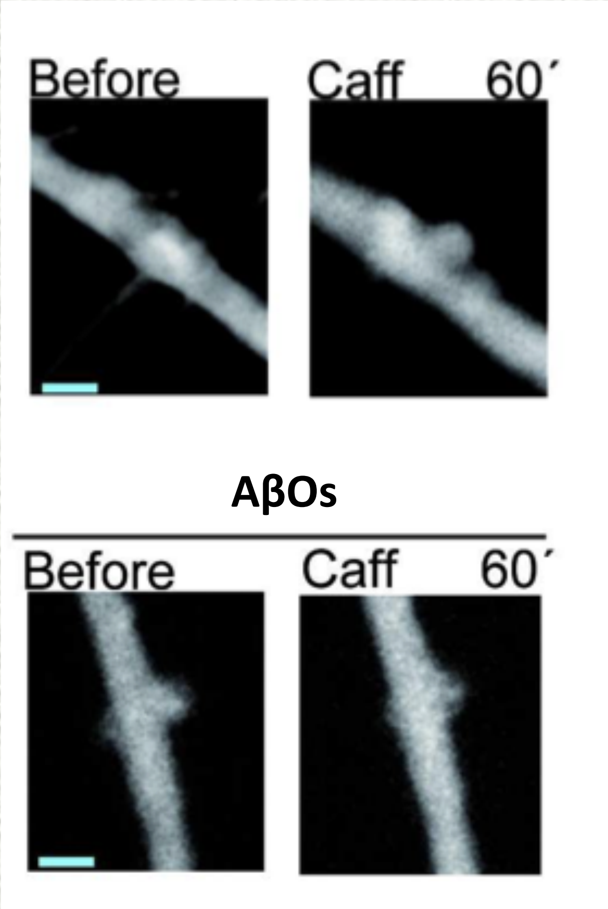
Los A β Os previenen la plasticidad sináptica inducida por BDNF

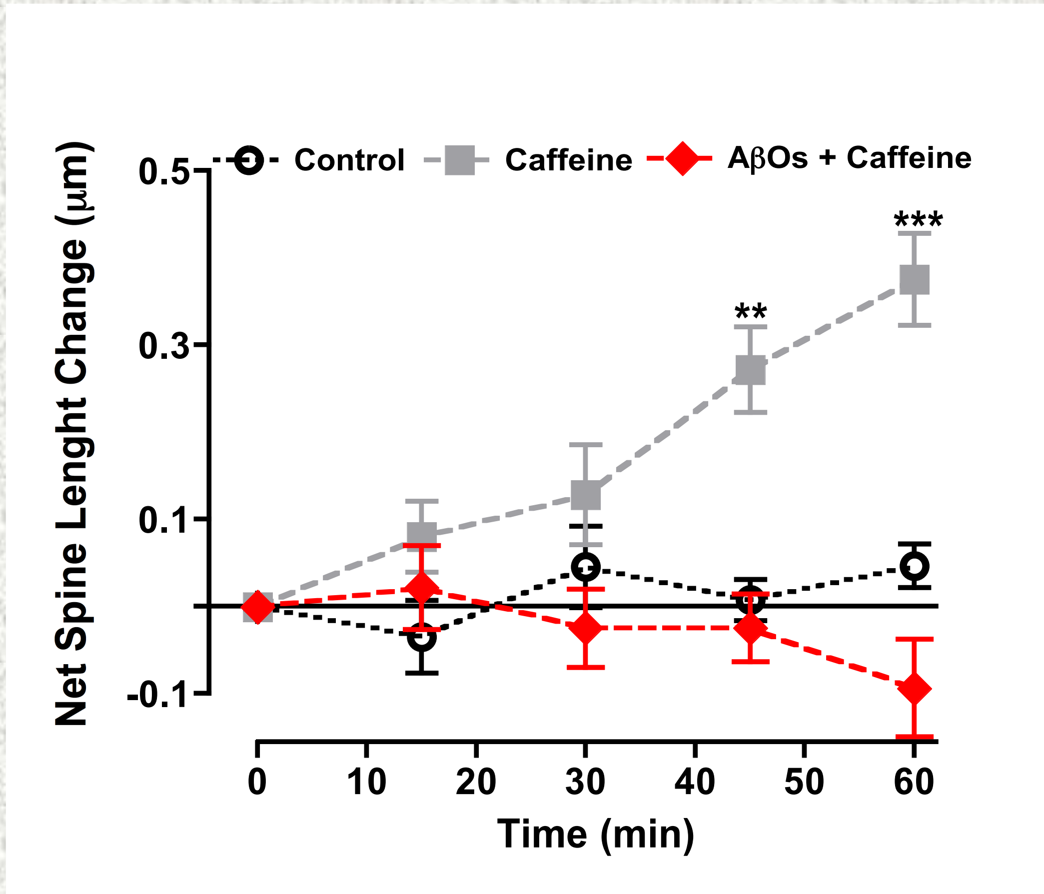




Paula-Lima et al., 2011
Antiox, Redox Sig.

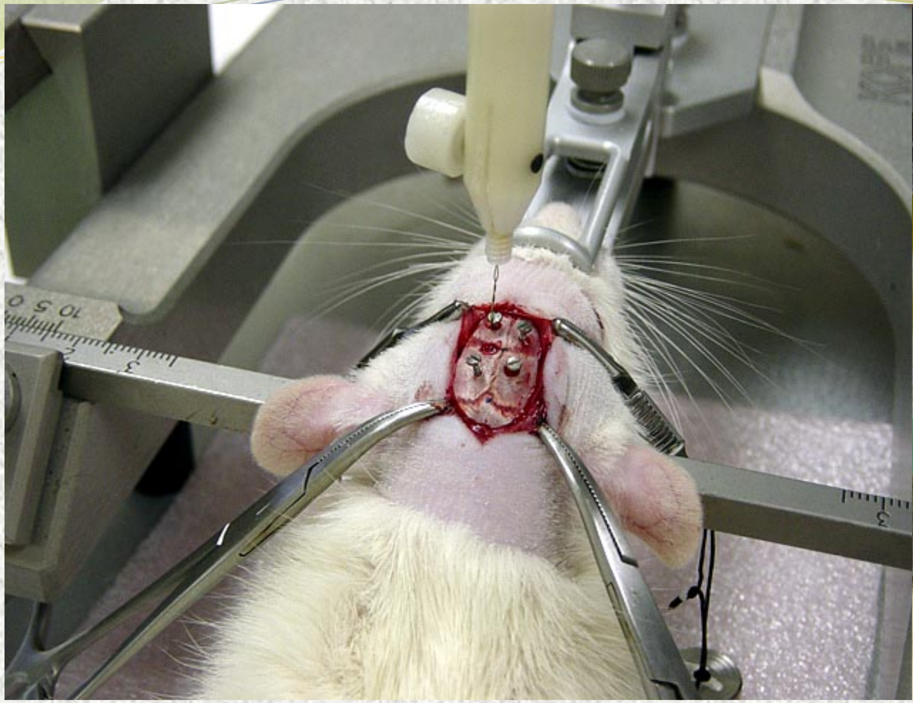
RyR-mediated spine remodeling is inhibited by AβOs





Paula-Lima et al., 2011
Antiox, Redox Sig.

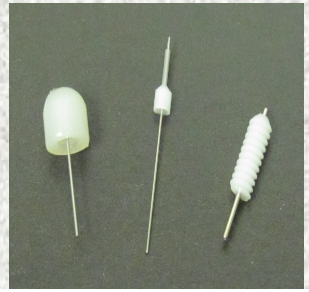
Inserción de cánulas guía para inyección en hipocampo



En nuestro caso inyección **bilateral**

Coordinadas: Bregma -3,30 mm; línea media +/-2,6 mm; profundidad 1,22 mm desde la superficie craneal.

Cánulas de 2,7 mm de largo
(26 gauge, Plastics One, VA)



Oasis Maze

- Es una tarea utilizada para evaluar memoria espacial, corresponde a una versión seca del *Morris Water Maze*, en la cual un roedor debe buscar un pocillo con recompensa (agua o alimento), originalmente alrededor de 400 pocillos y uno sólo con agua ("el oasis")

- En esta tarea se presume que los sujetos aprenden la ubicación del pocillo recompensado, utilizando puntos de referencia como claves visuales, espaciales.

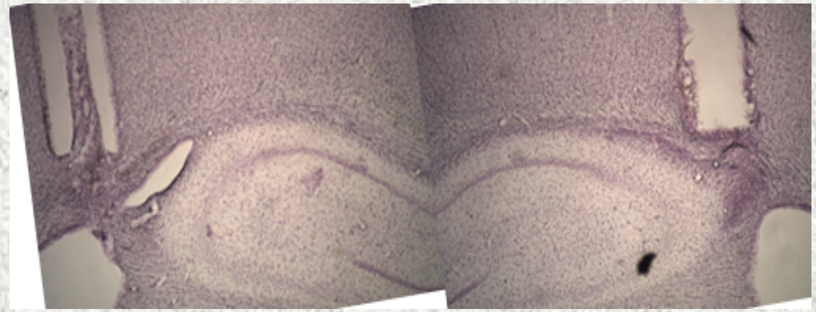
- Se ha sugerido que el Oasis maze tendría menor grado de componentes estresores que el *Morris water maze*.

Jamileth More

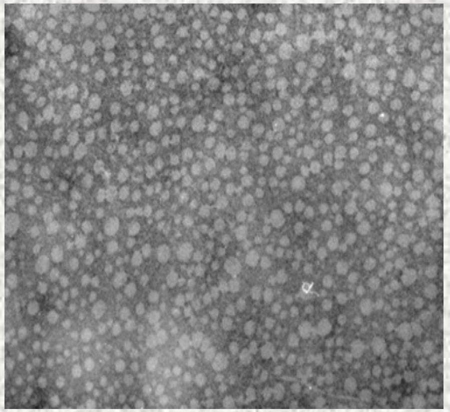
BILATERAL INJECTION OF AβOS IN CA3

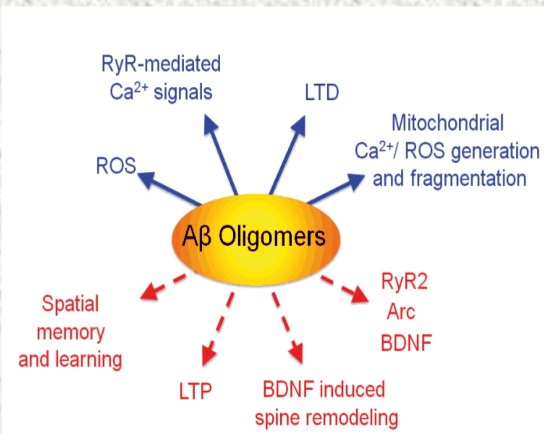
Coordinates: Bregma -3,30 mm; Madium Line+/-2,6 mm; Deep 1,22 mm from craneal surface.

Cresyl Violet



AβOs



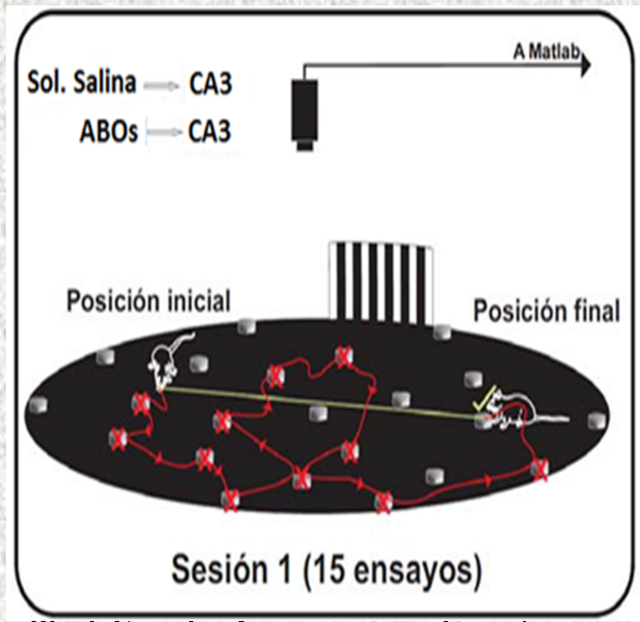


Jamileth More

José Luis Valdés

Paula-Lima et al., 2011, 2012, In press

OASIS MAZE EXPERIMENTAL PROTOCOL

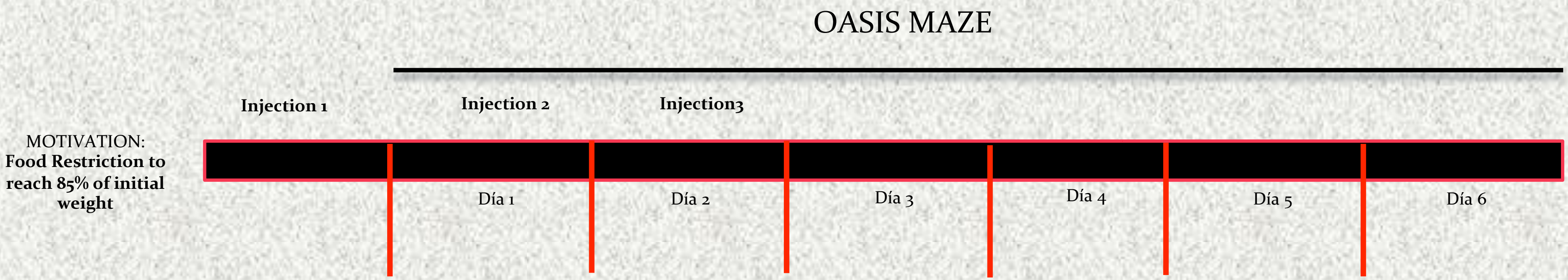


Black Circular Arena 1.4 m in diameter

The rat must find one well with a reward (water or food) among 21 wells ("the oasis") using visual spatial cues.

Individuals learn the spatial location of reward.

- ✓ Each session consists of 15 trials.
- ✓ One daily session for 6 days.



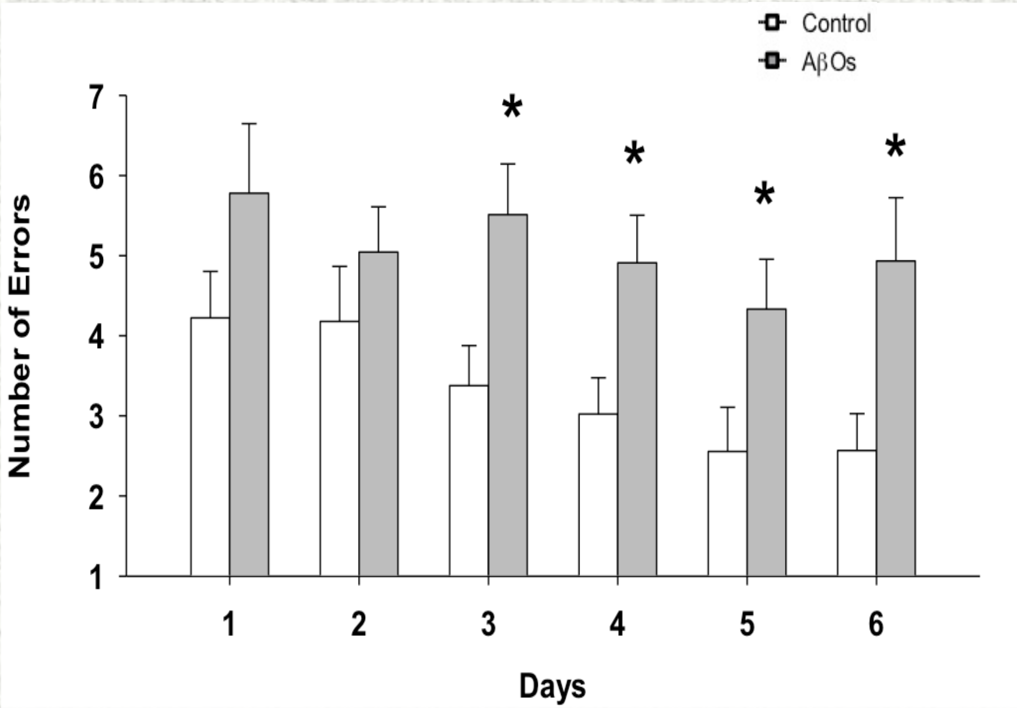
NUMBER OF ERRORS

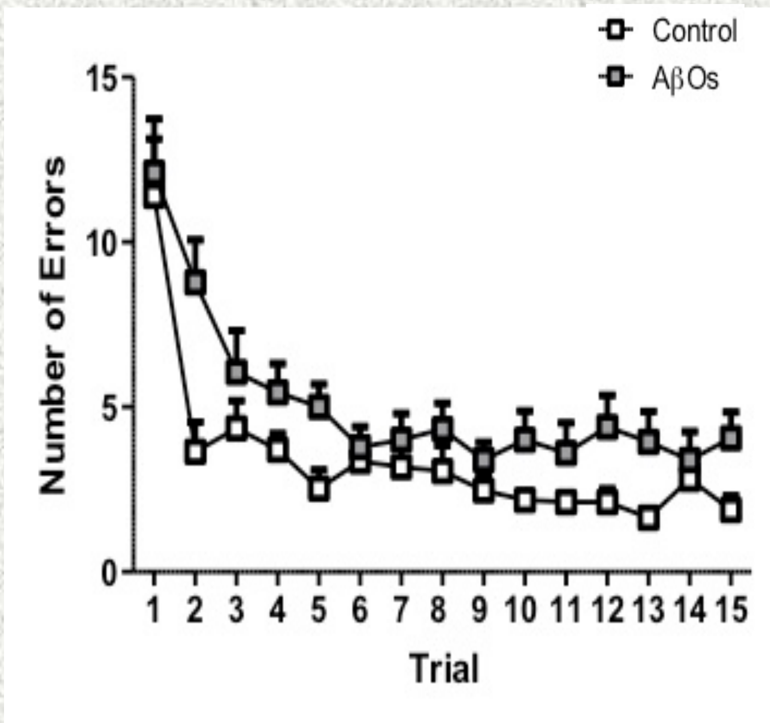
.

Corresponds to the times rat visit a well unrewarded in each trial

Number of errors per day

Number of errors per trial / 6 days





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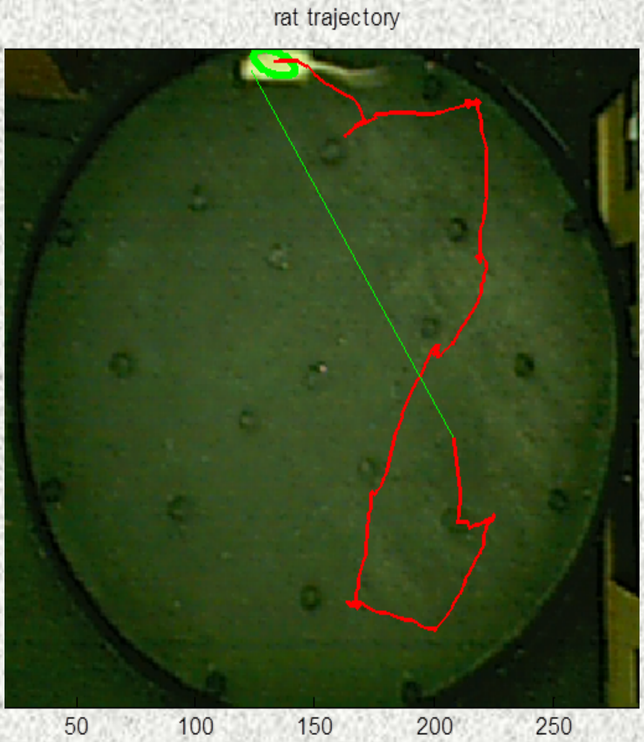
Rats injected with AβOs exhibit impaired spatial learning

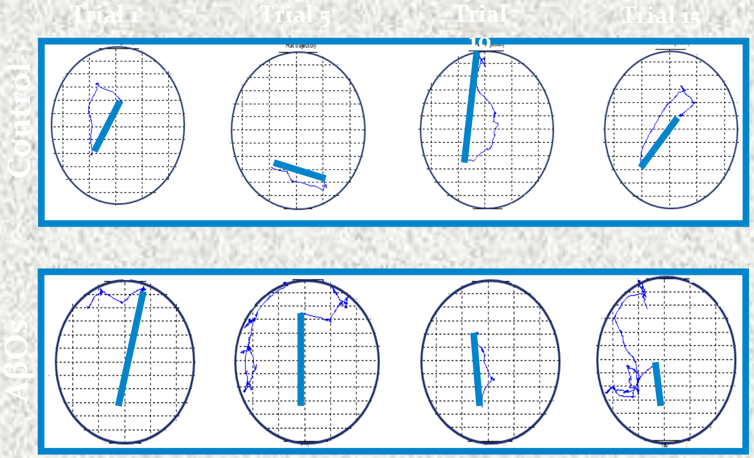
OBSERVED AND EXPECTED DISTANCE

OBSERVED ROUTE
EXPECTED ROUTE

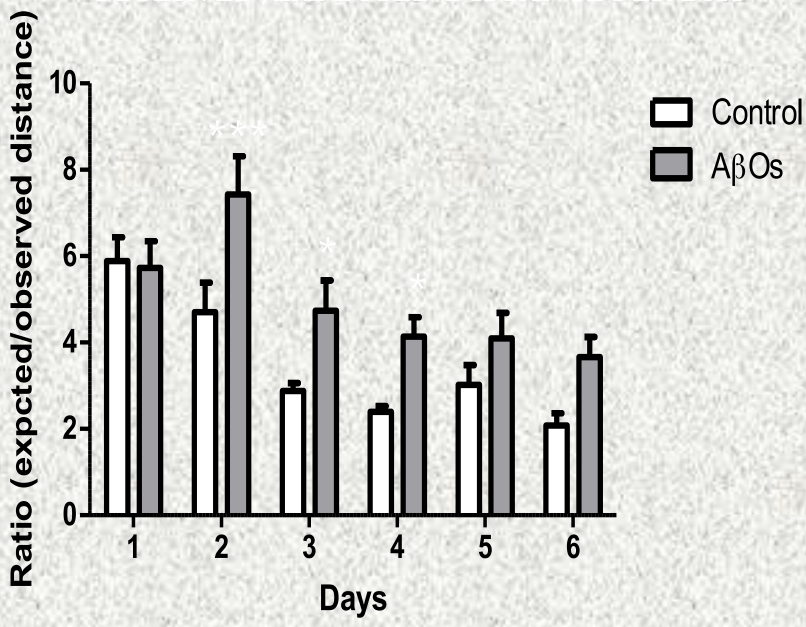


For each animal 15 movies per section, 6 section

OBSERVED DISTANCE (thin line) AND
EXPECTED DISTANCE (thick line) IN TRIALS 1, 5, 10 AND 15

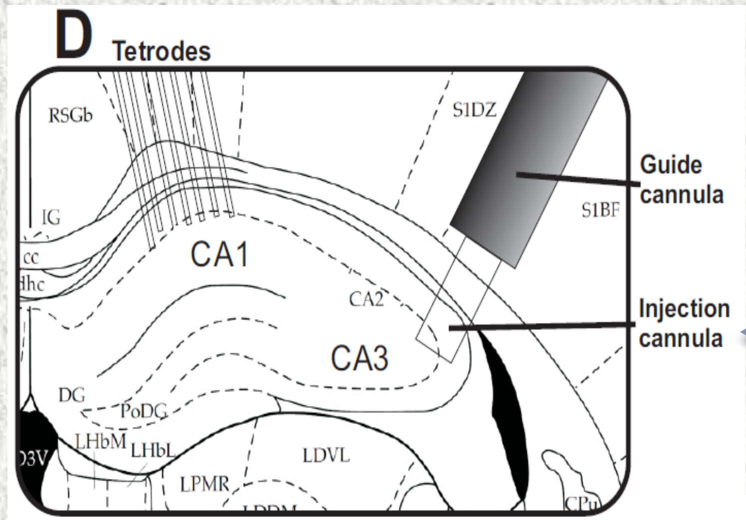


OBSERVED/EXPECTED DISTANCE PER DAY



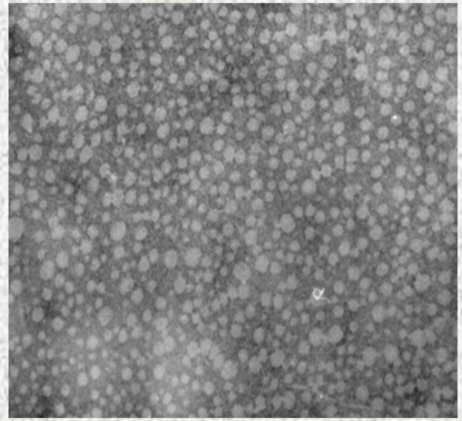
Rats injected with AβOs exhibit impaired spatial learning

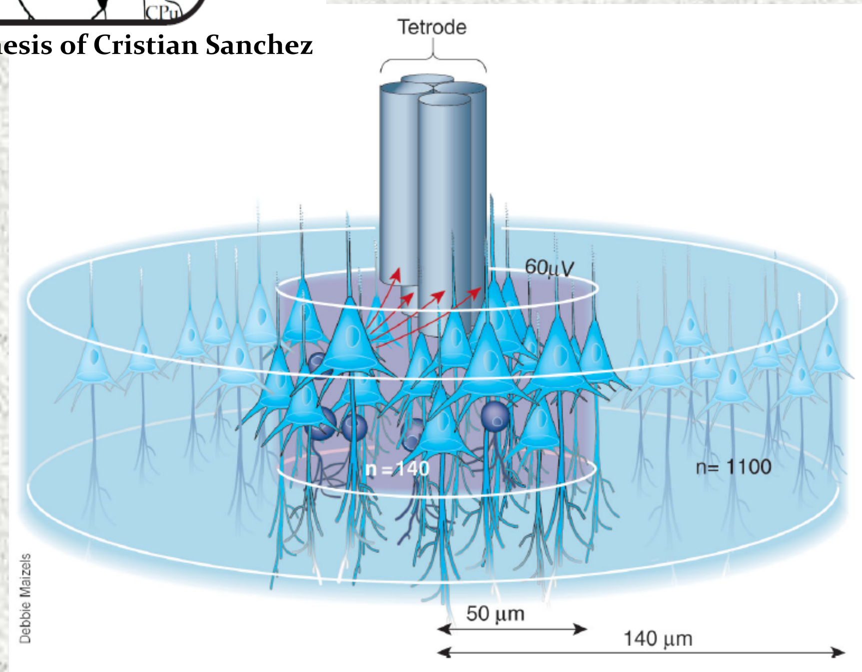
EXTRACELLULAR RECORDS IN AWAKE FREELY MOVING ANIMALS USING TETRODES



Scheme taken from the PhD thesis of Cristian Sanchez

AβOs

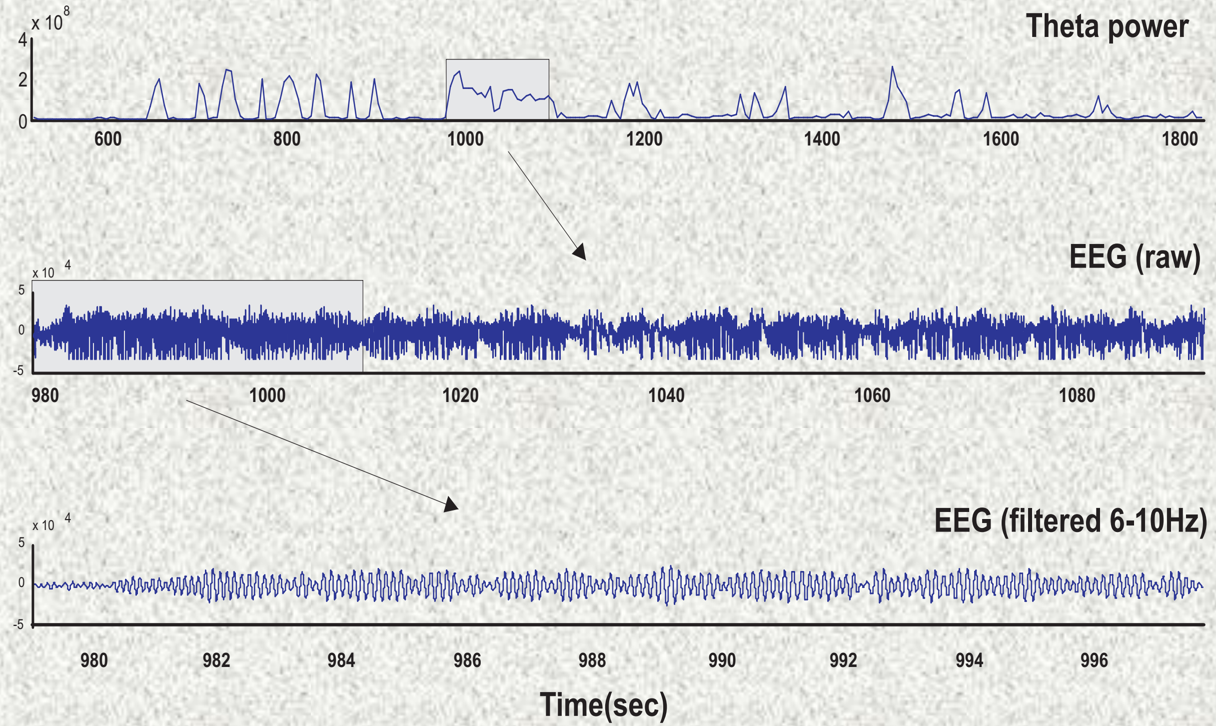




Yerko Fuentealba

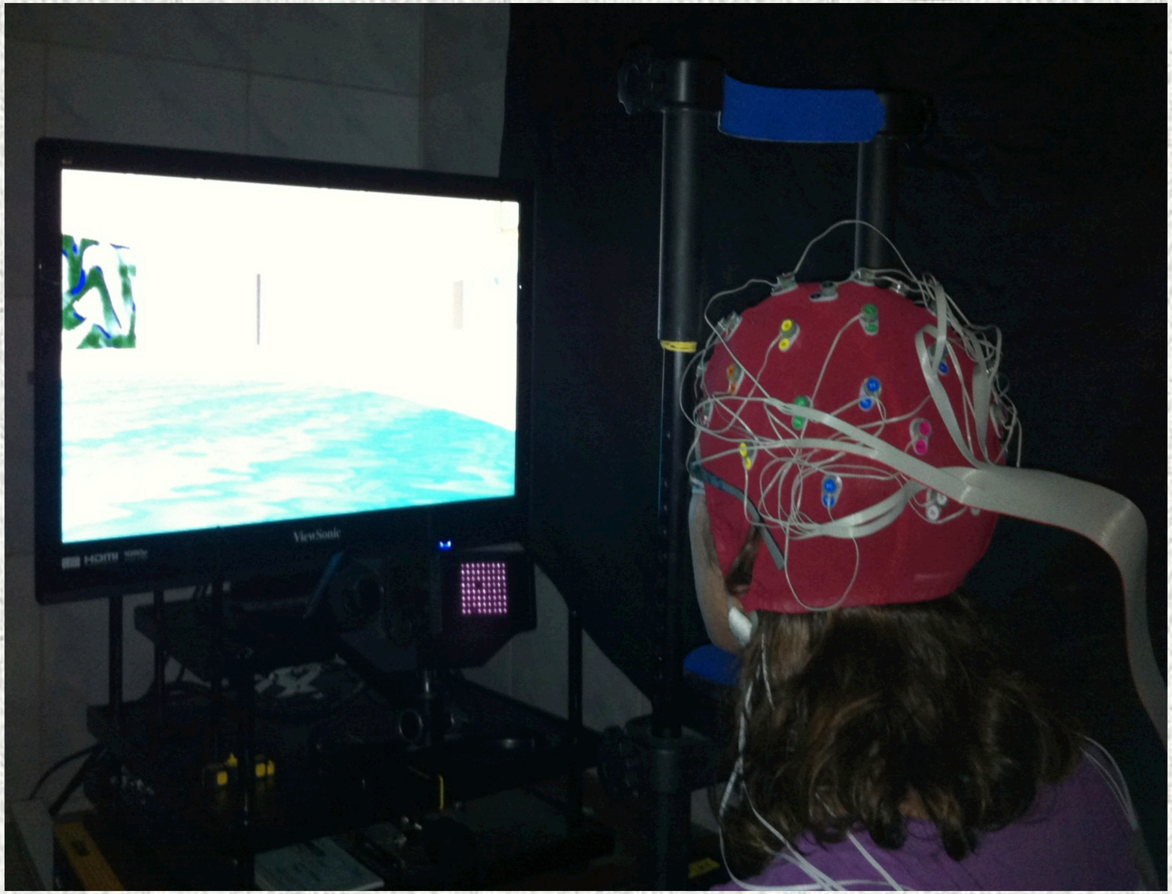
José Luis Valdés

HIPPOCAMPAL THETA POWER



- The *hippocampal theta activity* is a strong oscillation between 6–10 Hz
- It appears when the rat is engaged in spatial exploration

VIRTUAL MAZE NAVEGATION TASK



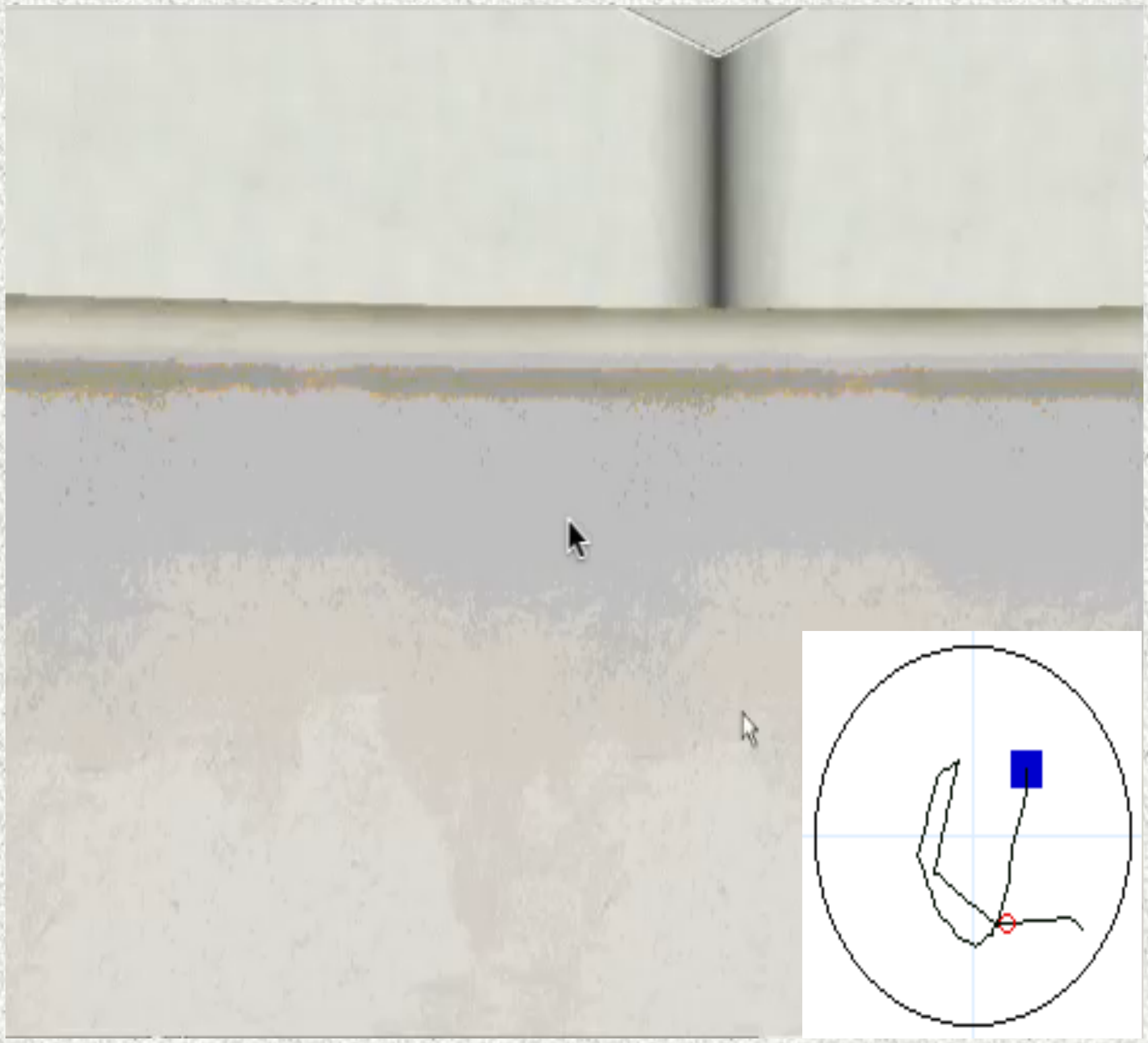
Enzo Brunetti

Rodrigo Häfelin

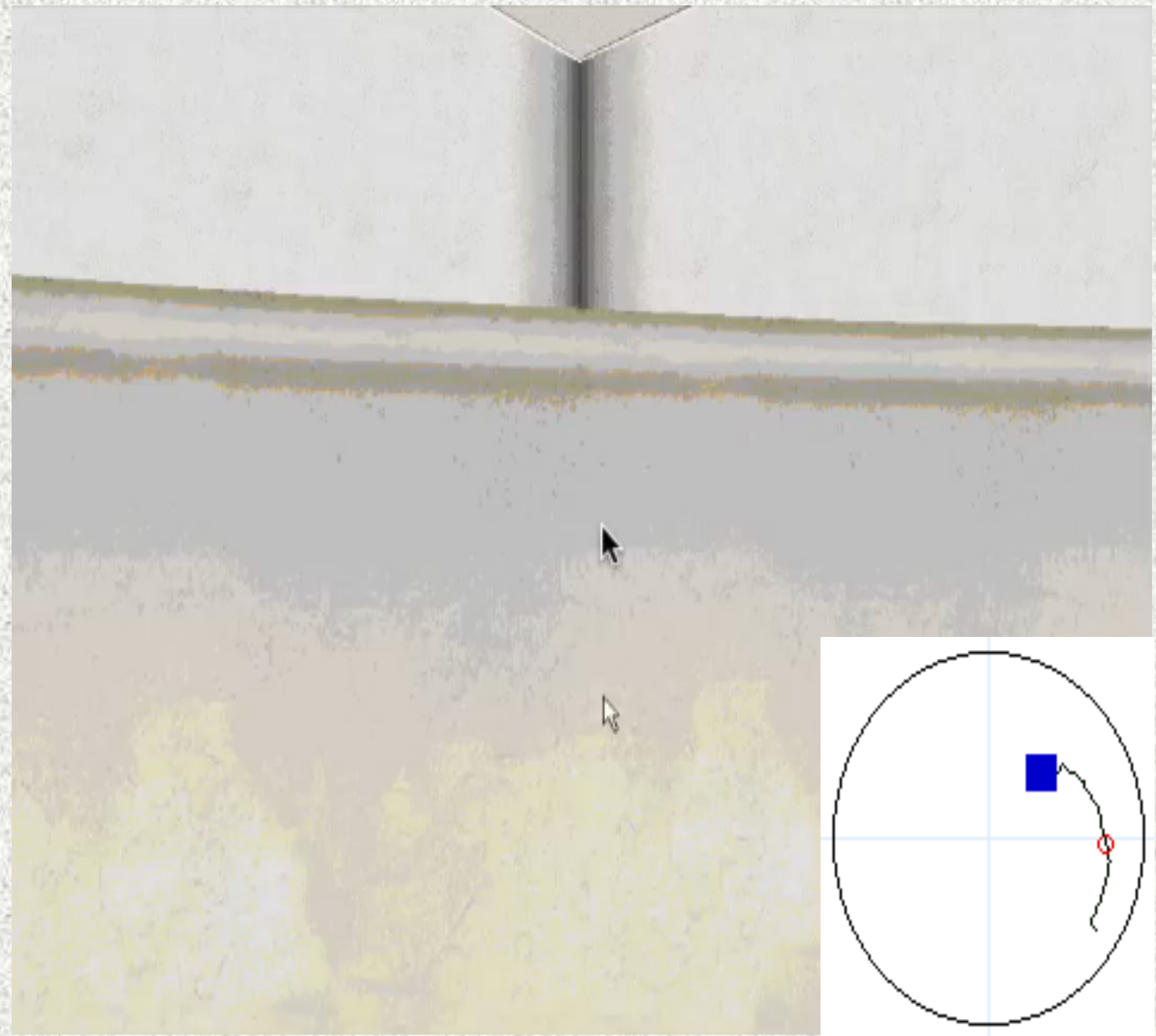
Maria Isabel Behrens, PhD

Daniela Ponce

Tarea de memoria espacial virtual water maze

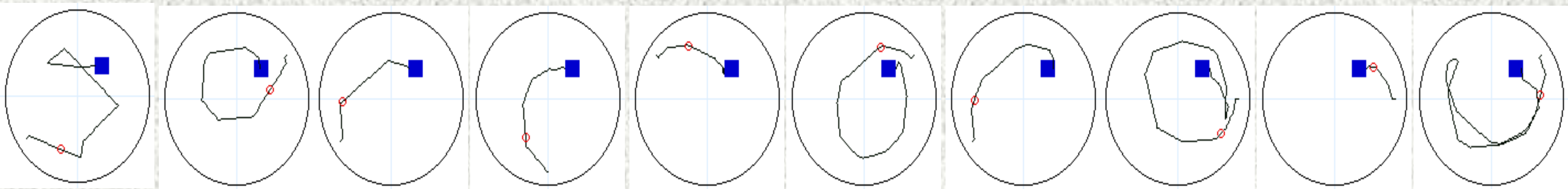


Ejemplo de aprendizaje de la tarea de memoria espacial virtual water maze

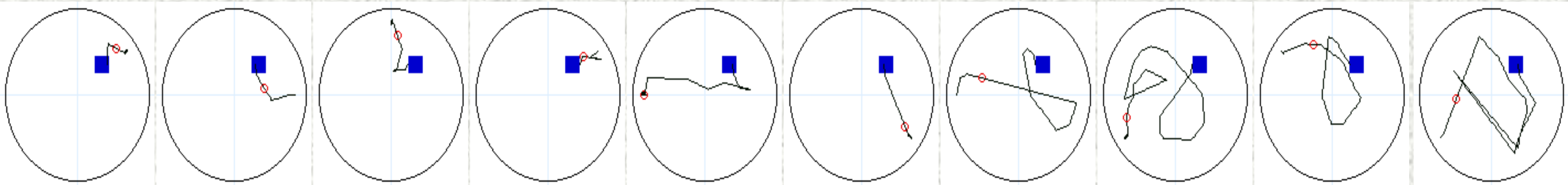


Conducta exploración espacial

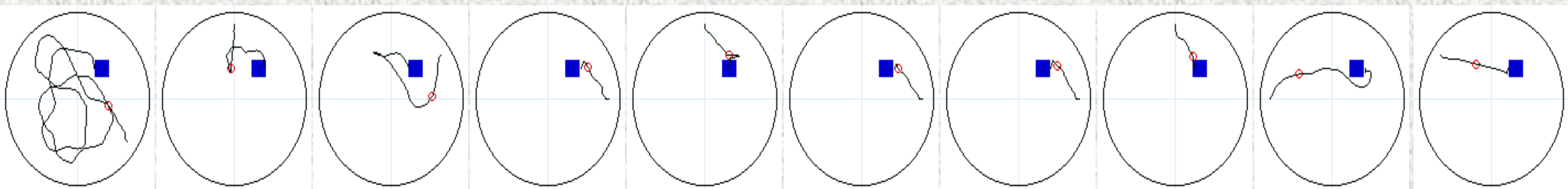
Alzheimer



Control edad



Control joven



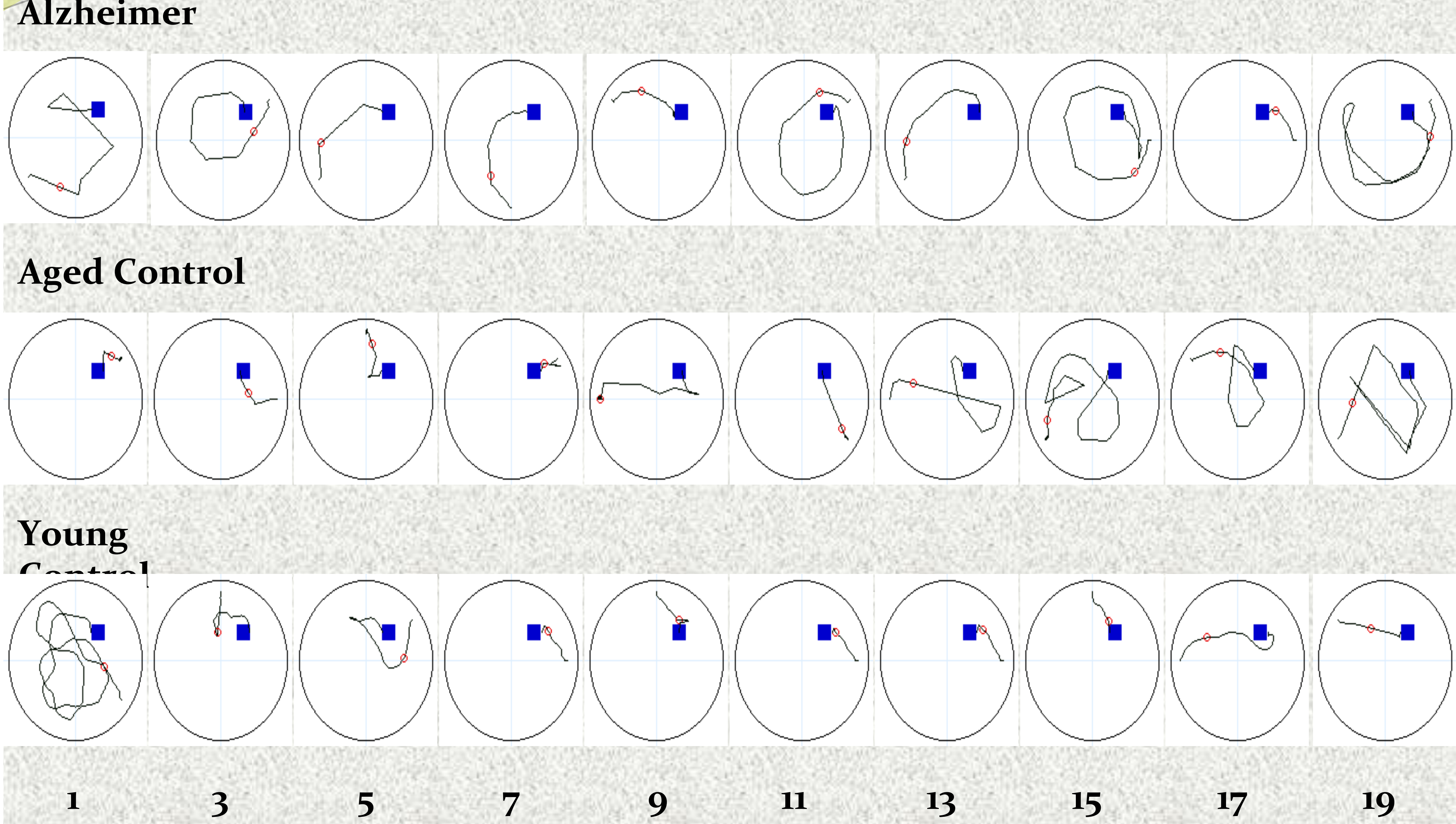
1 3 5 7 9 11 13 15 17 19

VIRTUAL MAZE PROTOCOL

HIDDEN PLATFORM
Number of trials: 20
Duration of each trial: 60 sec

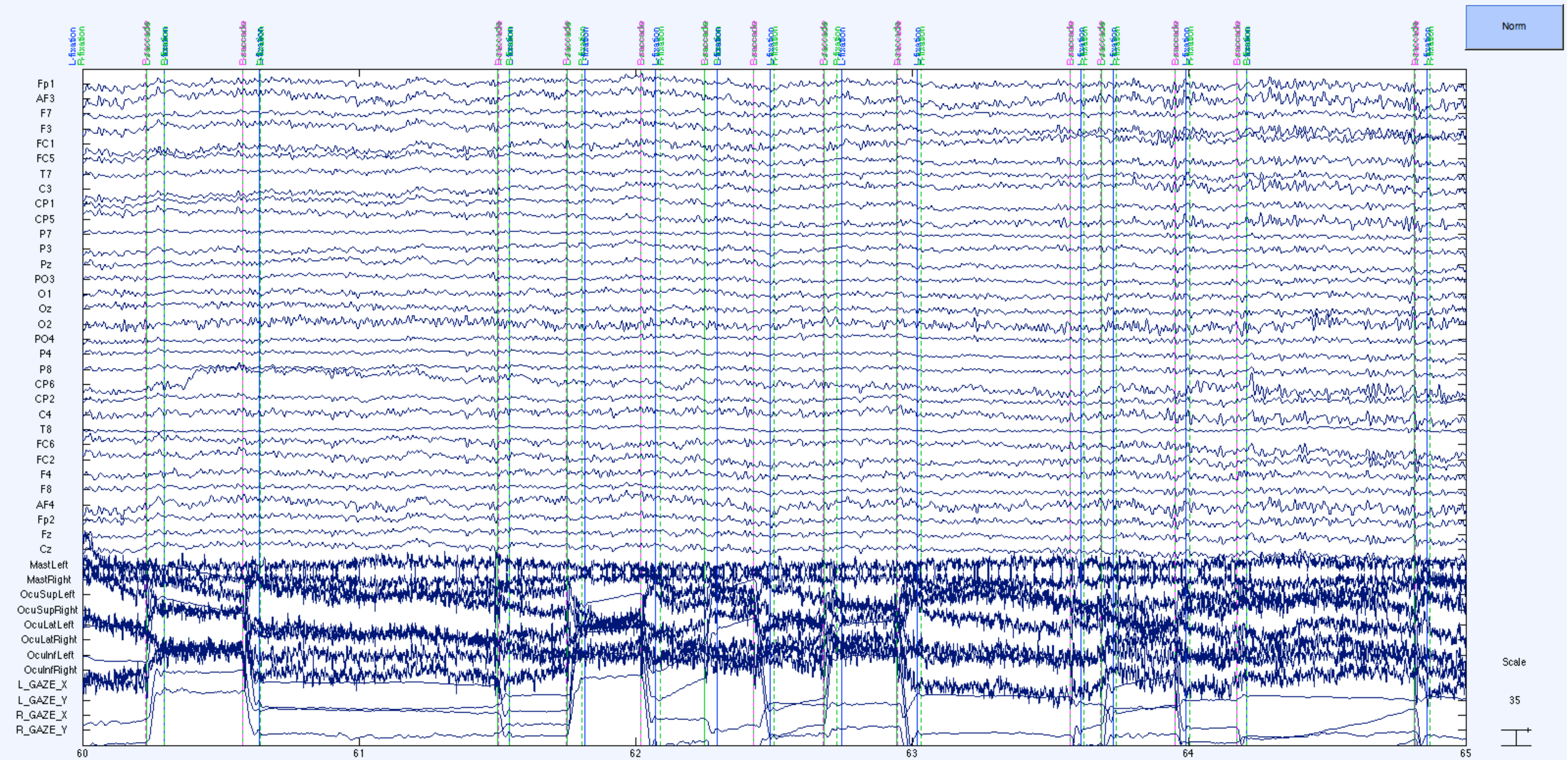
ALREADY RECORDED:
5 YOUNG CONTROL SUBJECTS (OVER 65 YEAR-OLD)
9 AGED CONTROL SUBJECTS (20-40 YEAR-OLD)
3 AD PATIENTS (OVER 65 YEAR-OLD)

PERFORMANCE IN THE SPATIAL EXPLORATION



Both young and old subjects learn the task, but with different patterns. AD patients never learn the task

CONTINUOUS EEG RECORD DURING VWMT

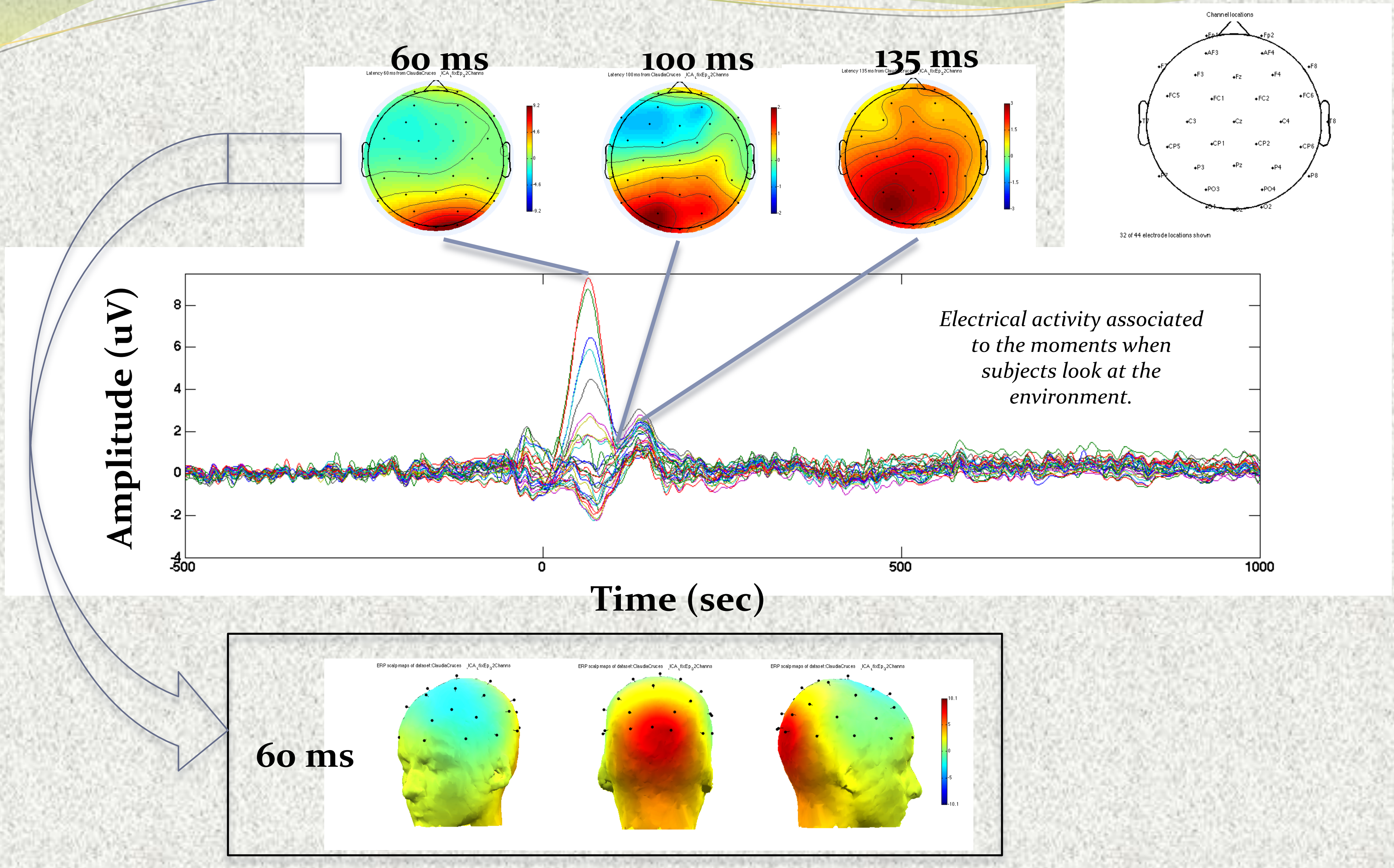


Brunetti et al., 2013

Time (sec)

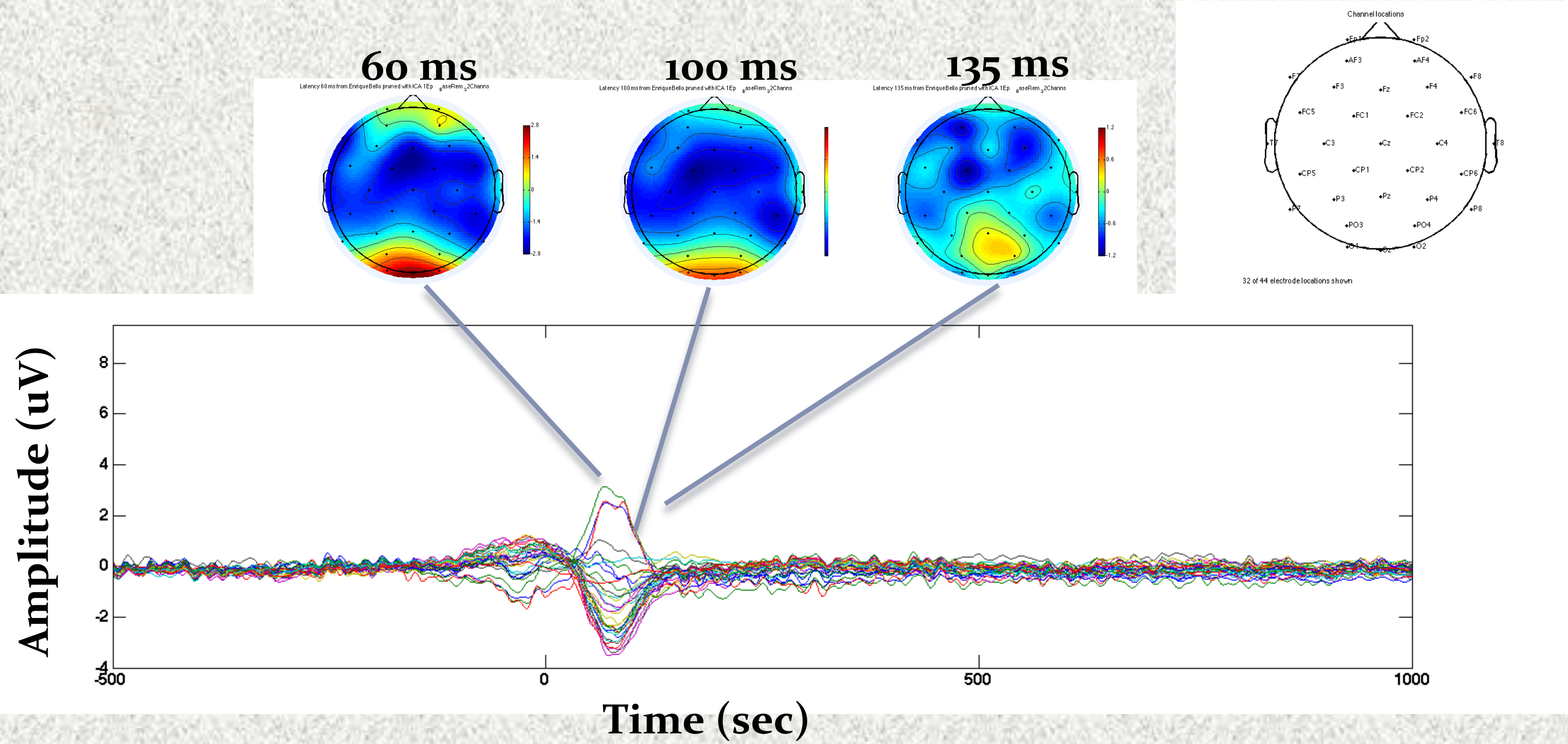
By the combination of eye tracker with EEG we are able to study the periods in which the subjects are looking to the

EYE-FIXATION RELATED POTENTIALS



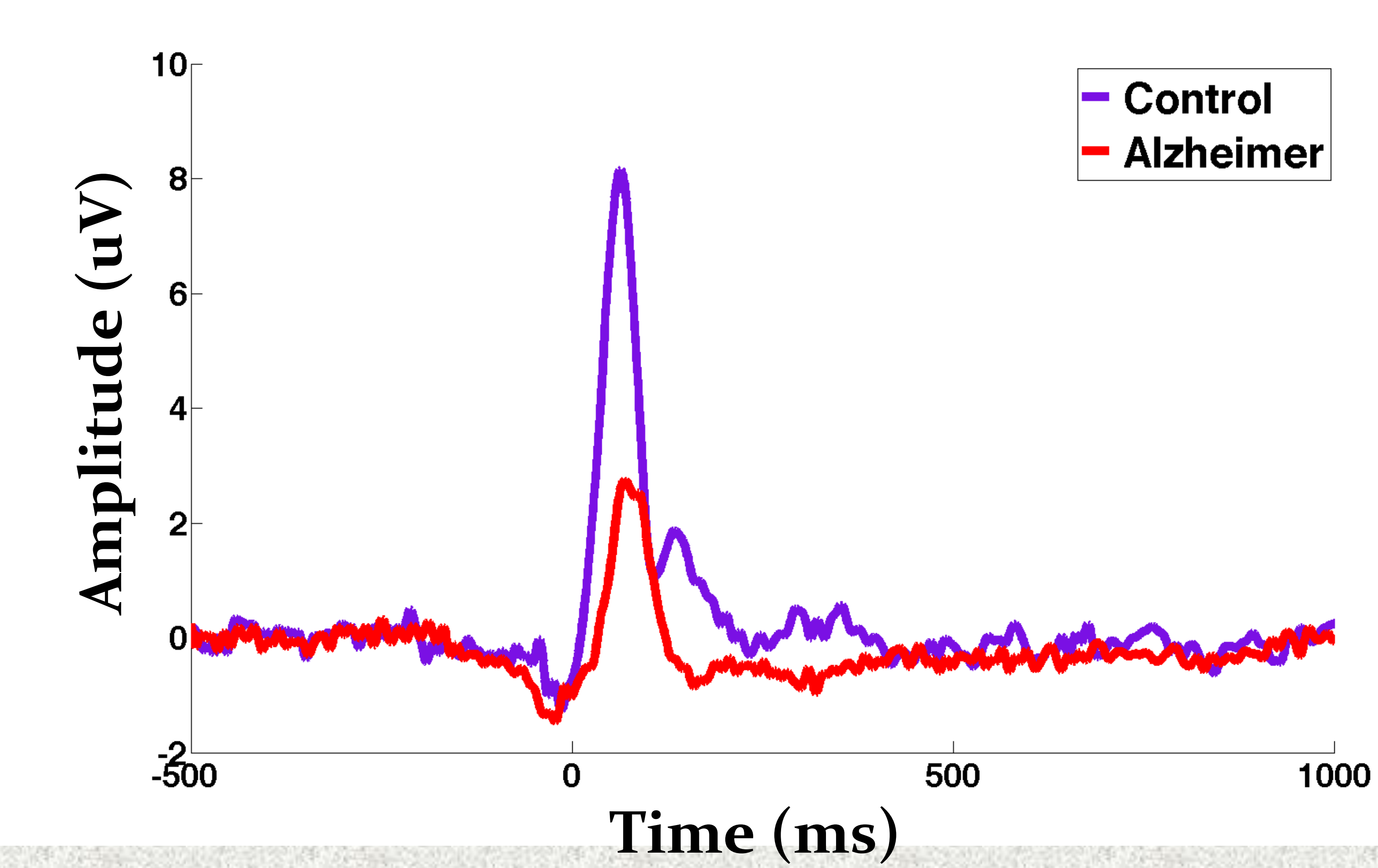
The activity propagates from visual to parietal cortex

EARLY ALZHEIMER´s DISEASE



In AD patients the activity is remarkably reduced and does not spread from visual to the parietal co

DIFFERENCE IN THETA ACTIVITY AT EARLY SENSORY PROCESSING



This is the difference of the theta activity's power between controls and AD

Gracias!

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