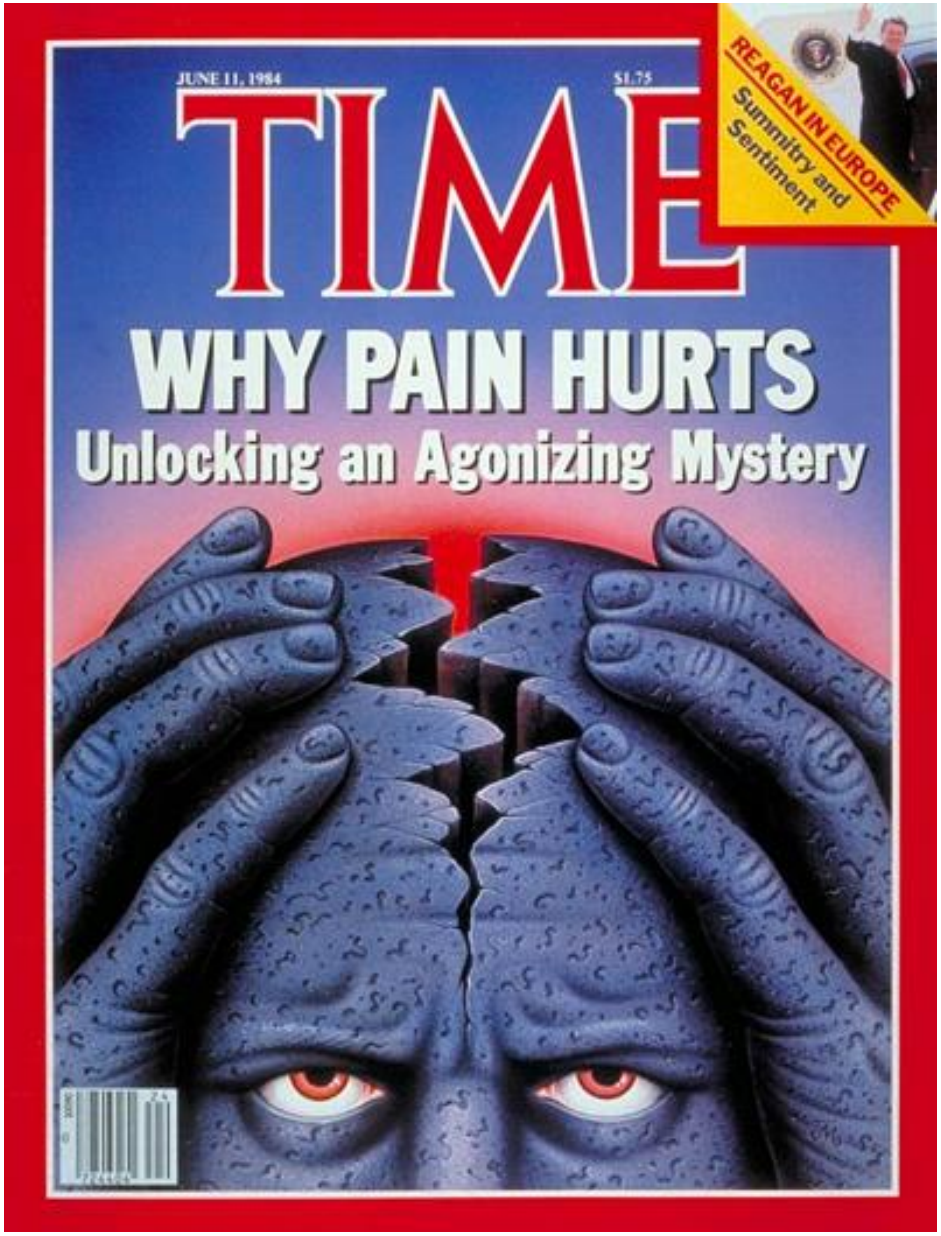
Il trattamento della neuroinfiammazione a livello del SNC mediante la modulazione delle cellule non neuronali

Salvatore Cuzzocrea, MD, PhD

MARCH 1, 2004
TIME
BLOOD, GUTS AND "THE PASSION"
THE E.U.'S IMMIGRATION FEARS
THE SECRET KILLER
The surprising link between INFLAMMATION and HEART ATTACKS, CANCER, ALZHEIMER'S and other diseases
What you can do to fight it
www.timeeurope.com AOL Keyword: TIME


JUNE 11, 1984
$1.75
TIME
REAGAN IN EUROPE
Summitry and Sentiment
WHY PAIN HURTS
Unlocking an Agonizing Mystery

What about Neuroinflammation….?

Clinical & Experimental Immunology
The Journal of Translational Immunology

Stüve &Zettl 2014; 175(3):333–335

British Society for immunology

Clinical and Experimental Immunology | EDITORIAL | doi:10.1111/cei.12260

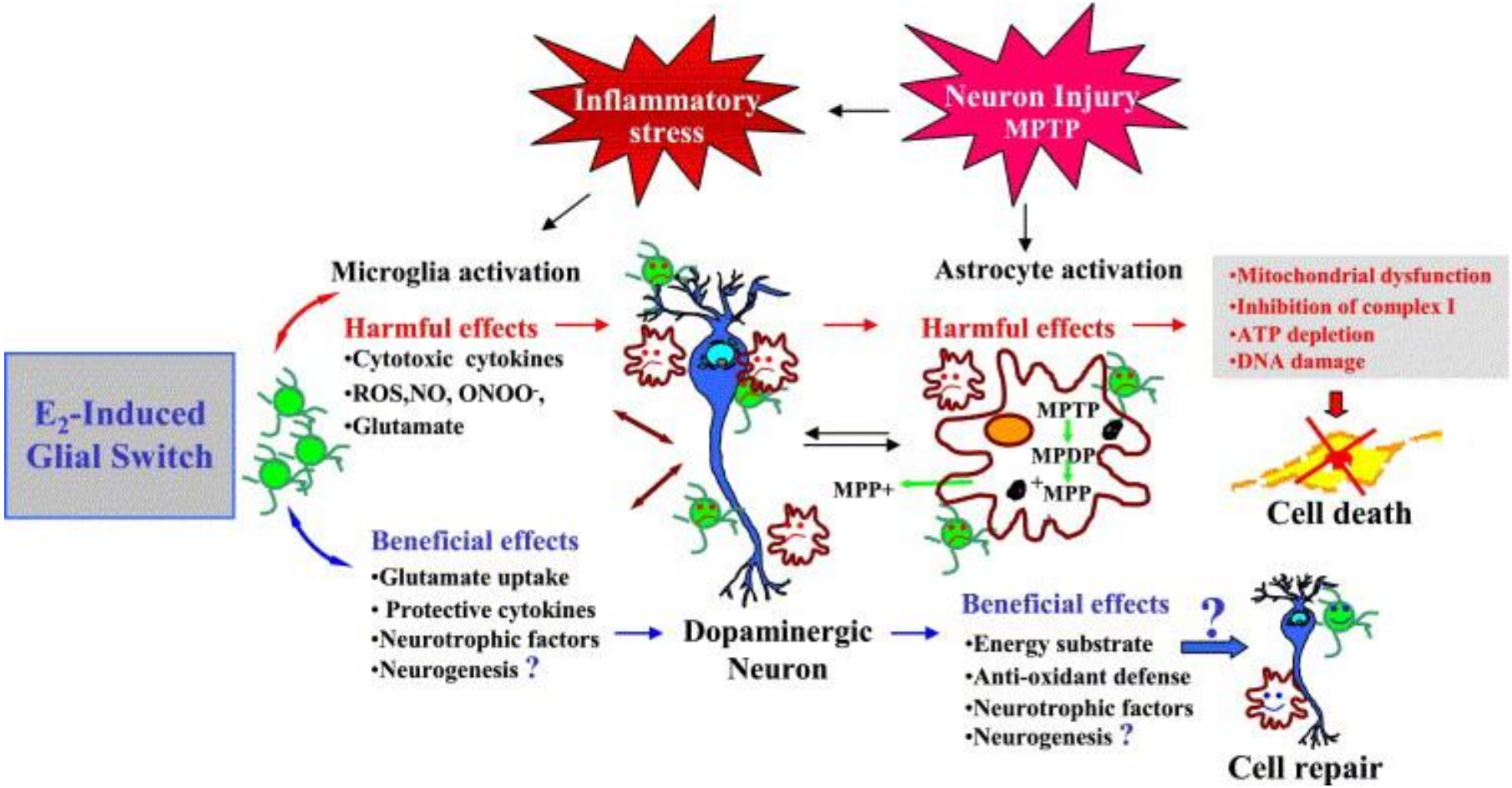
Neuroinflammation of the central and peripheral nervous system: an update

Inflammatory disorders of the peripheral nervous system and central nervous system are common, and contribute to the physical and emotional disability of affected individuals.

Key aspects of neuroinflammation

- Initiated from outside the CNS with subsequent leukocyte infiltration of the CNS.
- Also initiated from within the CNS with ensuing leukocyte infiltration.
- The CNS as an “immune privileged” organ (Medawar 1948).
- Interrelationship between neuroinflammation and neurodegeneration

(neural cell injury and death)



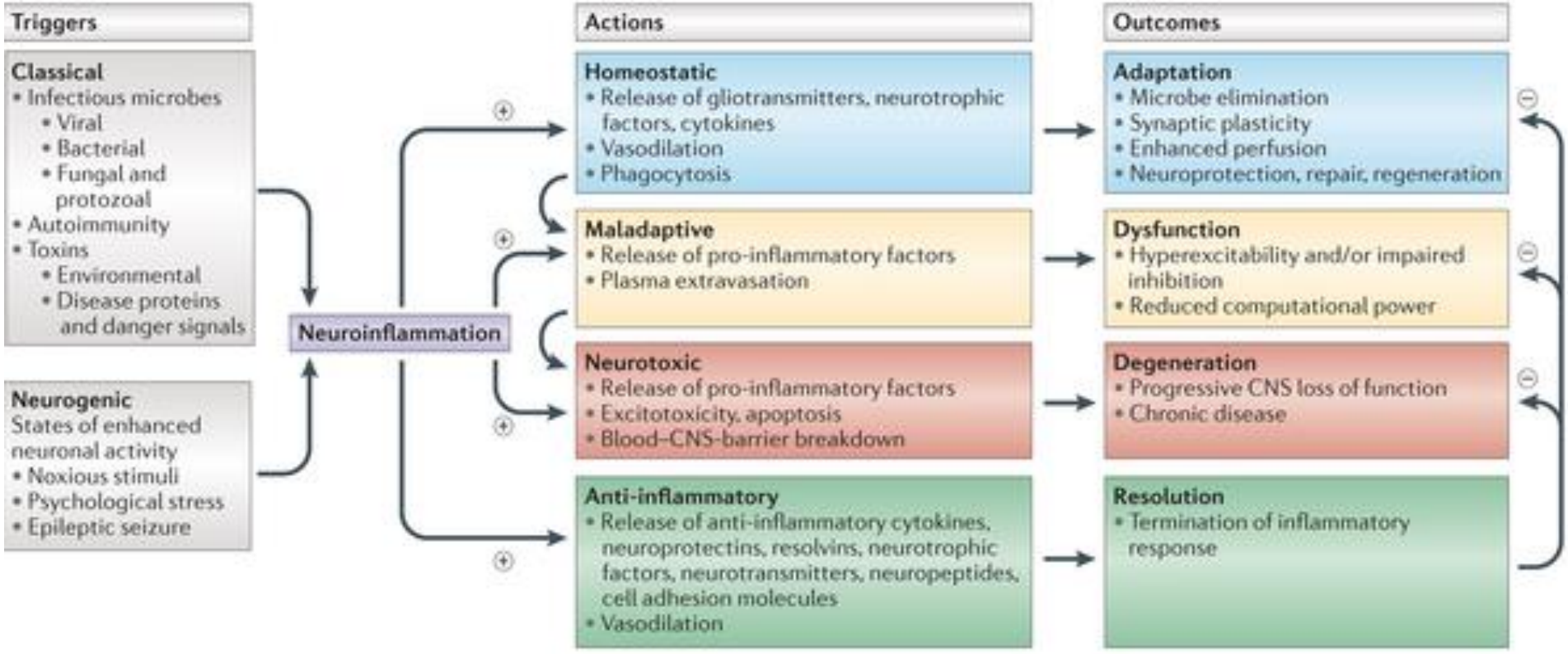
Neurogenic neuroinflammation: inflammatory CNS reactions in response to neuronal activity

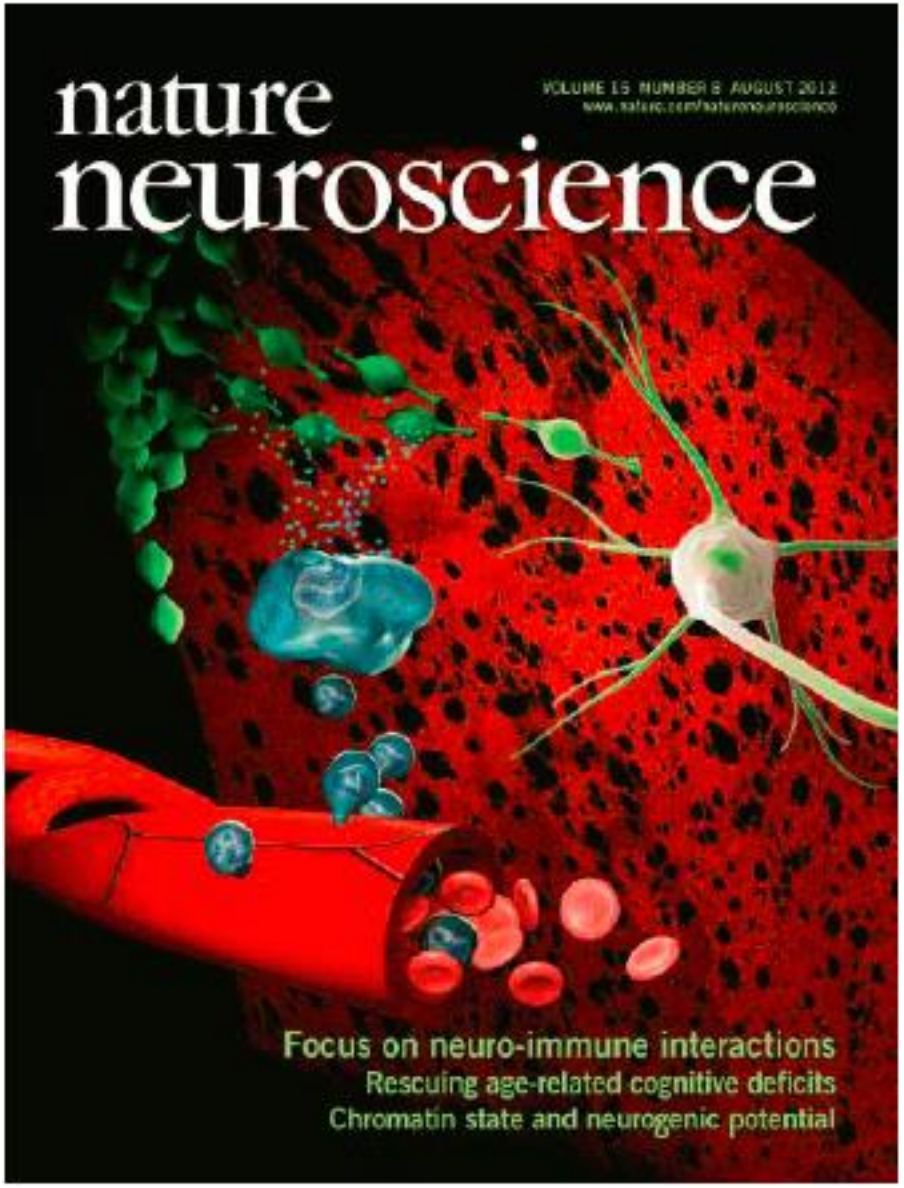
Dimitris N. Xanthos & Jürgen Sandkühler

Affiliations | **Corresponding author**

Nature Reviews Neuroscience **15**, 43–53 (2014) | doi:10.1038/nrn3617

Published online 27 November 2013





EDITORIAL

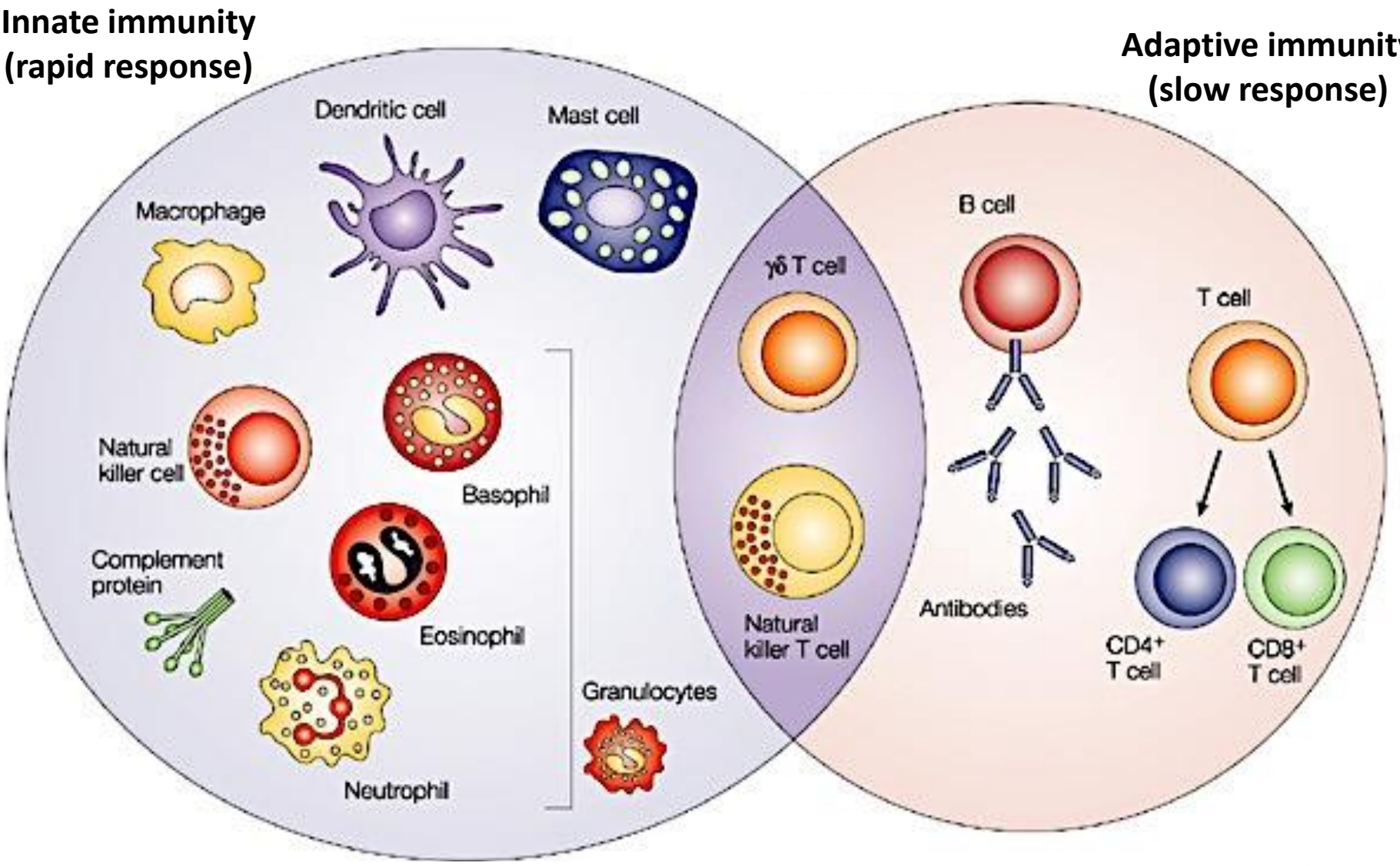
nature neuroscience

Focus on neuro-immune interactions

In this issue, *Nature Neuroscience* presents a focus on neuro-immune interactions.

"The work described by the reviews and perspectives presented in this focus issue has vastly improved our understanding of the **intricate and often reciprocal relationship between the neural and immune systems.** Once thought to be separate and distinct entities, **it is becoming increasingly clear that a number of diverse functions, including endogenous host-defense, disease response and postinfection or -injury repair depend on the activity and interplay of these systems**".

Mechanisms of immunity within the nervous system



Biochemical Pharmacology 103 (2016) 1–16

Contents lists available at ScienceDirect

Biochemical Pharmacology

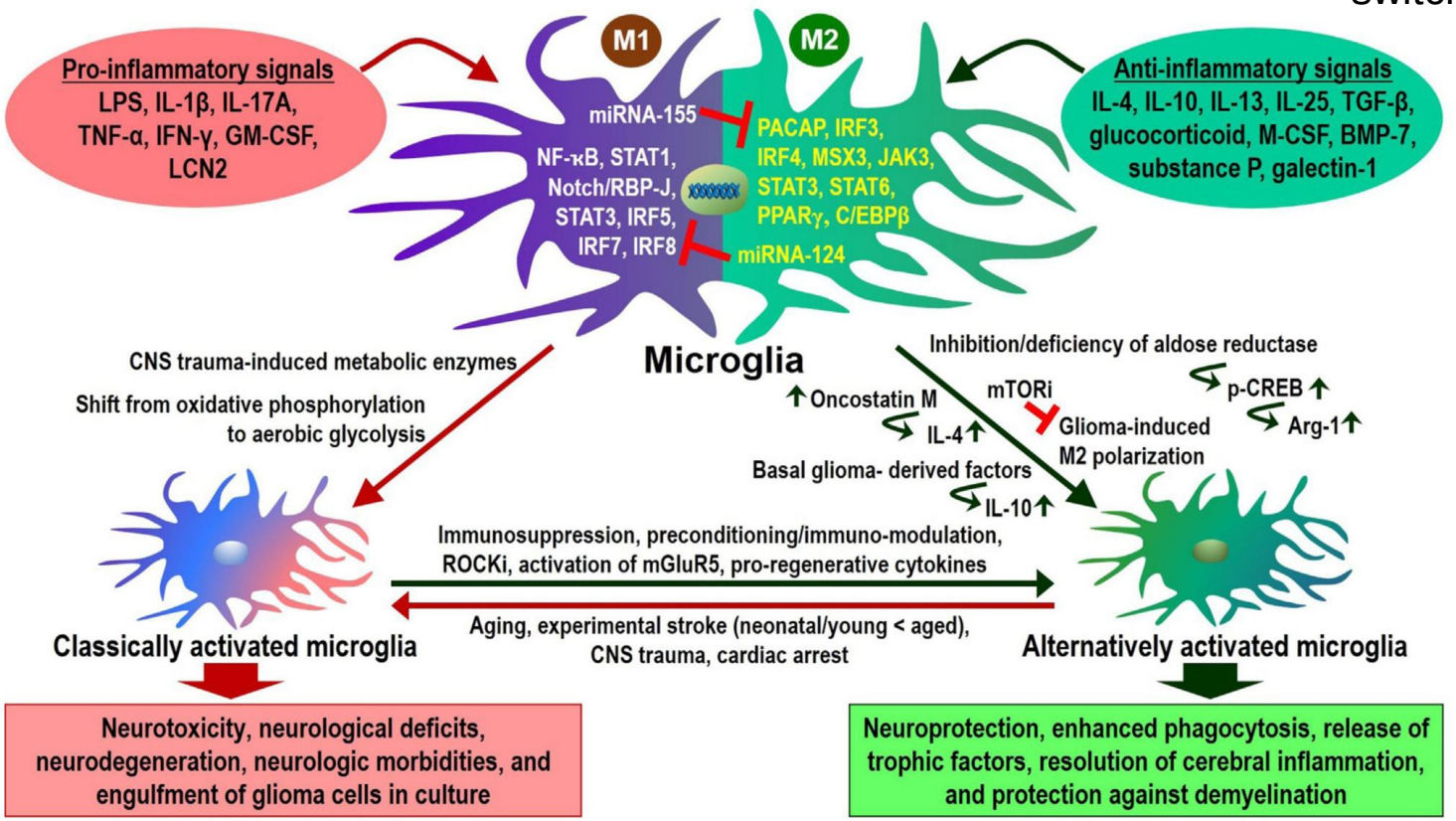
journal homepage: www.elsevier.com/locate/biochempharm

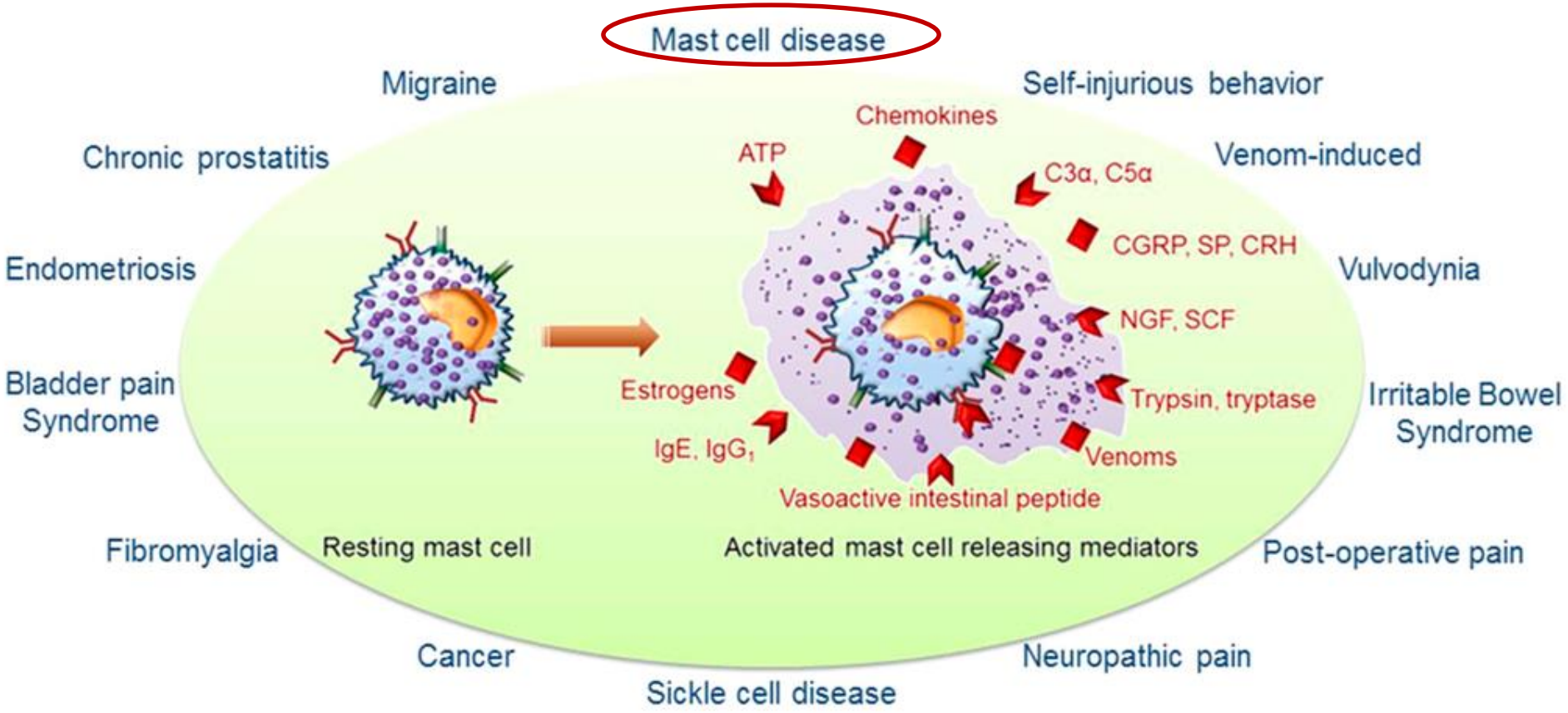
Research update

Functional polarization of neuroglia: Implications in neuroinflammation and neurological disorders

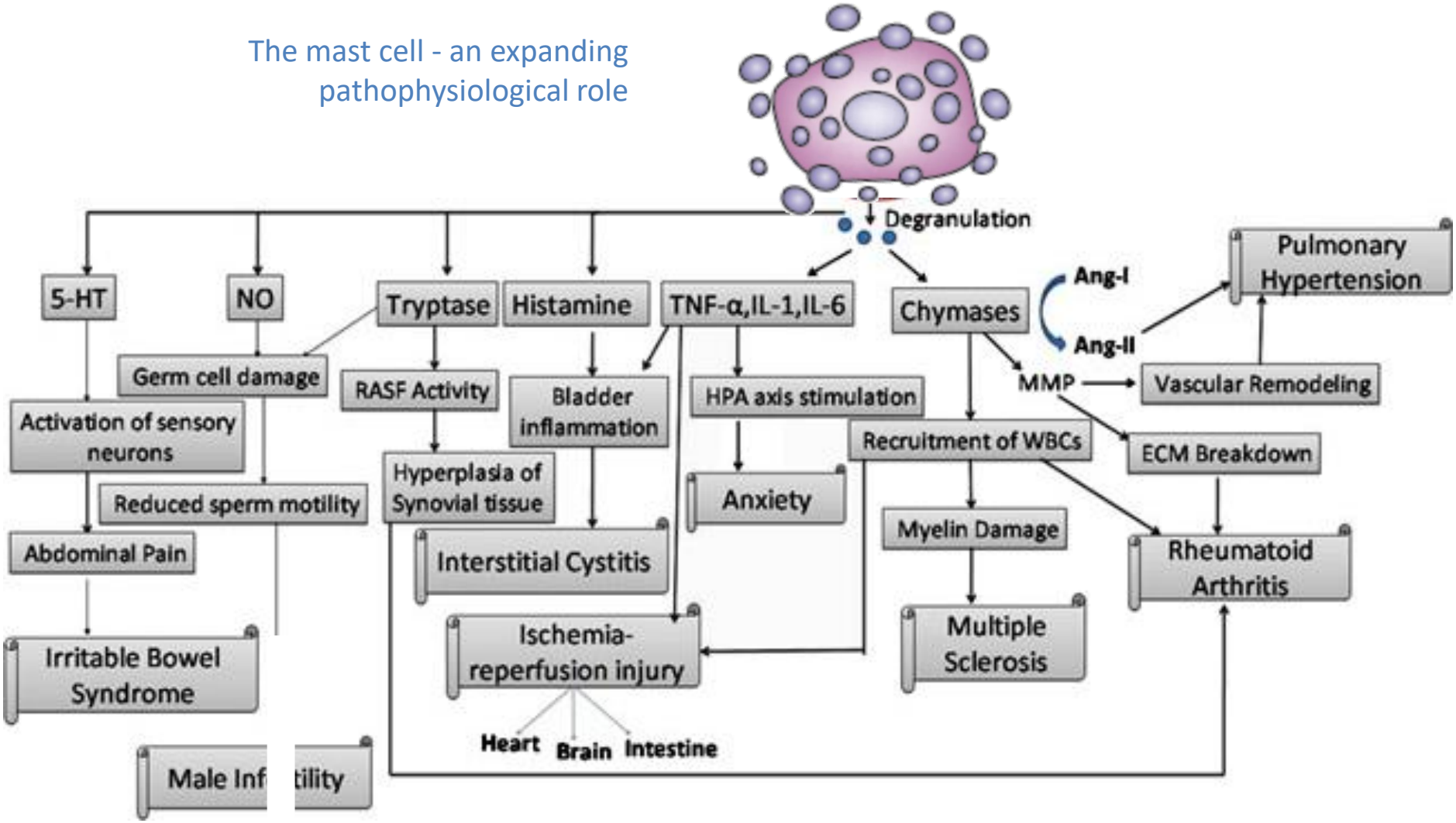
Mithilesh Kumar Jha[a], Won-Ha Lee[b], Kyoungho Suk[a,*]

Phenotypic polarization and switching of microglia



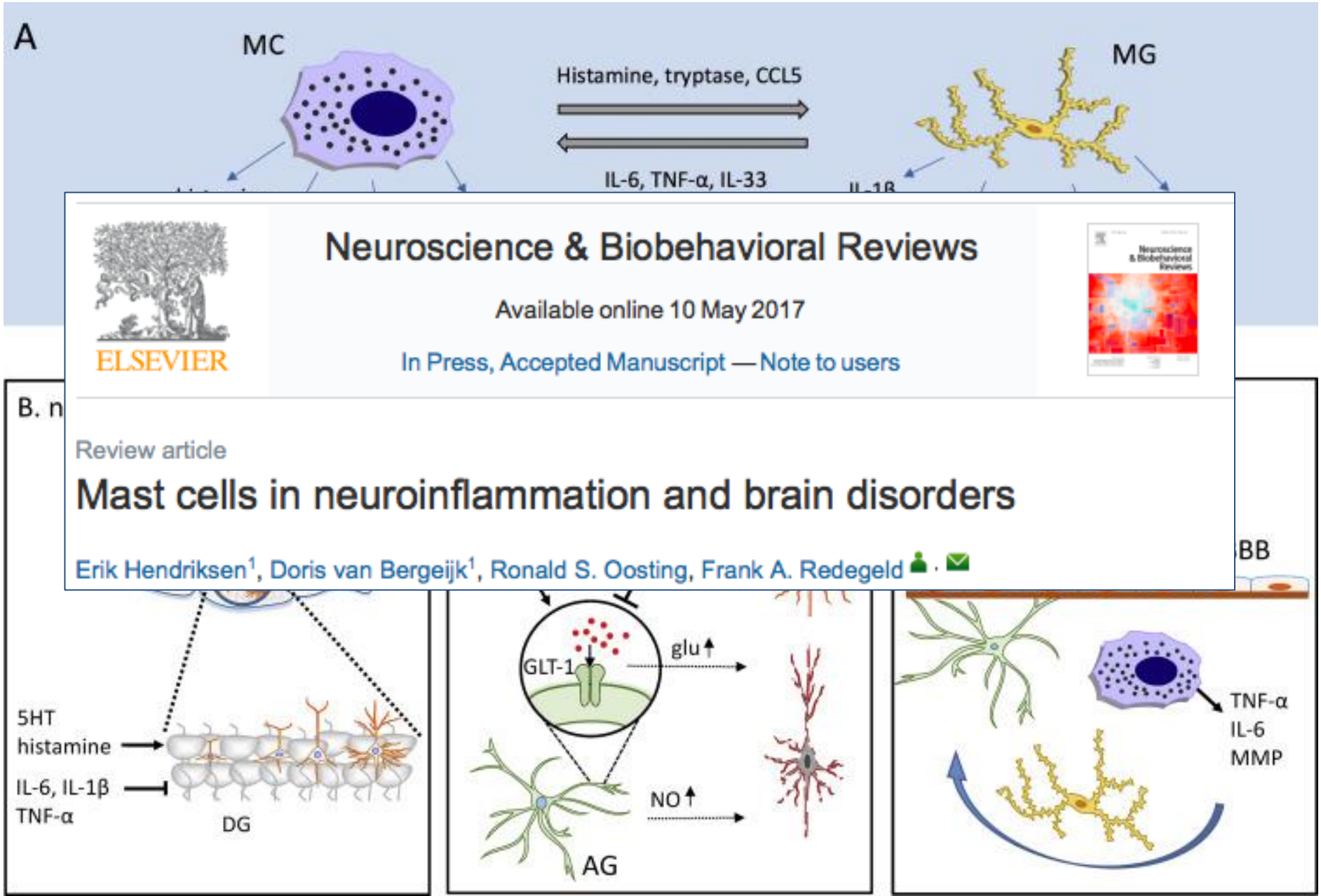

Mast cell disease
Migraine
Self-injurious behavior
Chronic prostatitis
Venom-induced
Endometriosis
Vulvodynia
Bladder pain Syndrome
Irritable Bowel Syndrome
Fibromyalgia
Post-operative pain
Cancer
Neuropathic pain
Sickle cell disease
Chemokines
ATP
C3α, C5α
CGRP, SP, CRH
NGF, SCF
Estrogens
Trypsin, tryptase
IgE, IgG1
Venoms
Vasoactive intestinal peptide
Resting mast cell
Activated mast cell releasing mediators

The mast cell - an expanding pathophysiological role



Naunyn Schmiedebergs Arch Pharmacol. 2012;385(7):657-70

Potential involvement of mast cells in physiological and pathological mechanisms involved in brain disorders



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Neuroscience & Biobehavioral Reviews

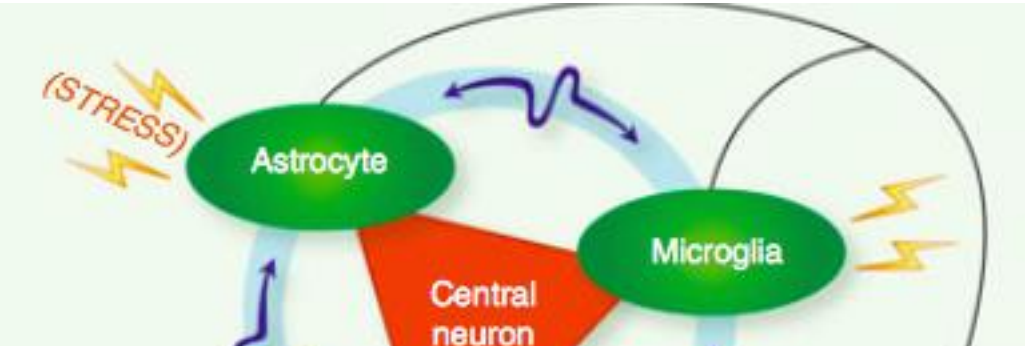
Available online 10 May 2017

In Press, Accepted Manuscript — Note to users

Review article

Mast cells in neuroinflammation and brain disorders

Erik Hendriksen[1], Doris van Bergeijk[1], Ronald S. Oosting, Frank A. Redegeld



Immunology
The Journal of cells, molecules, systems and technologies

British Society for immunology

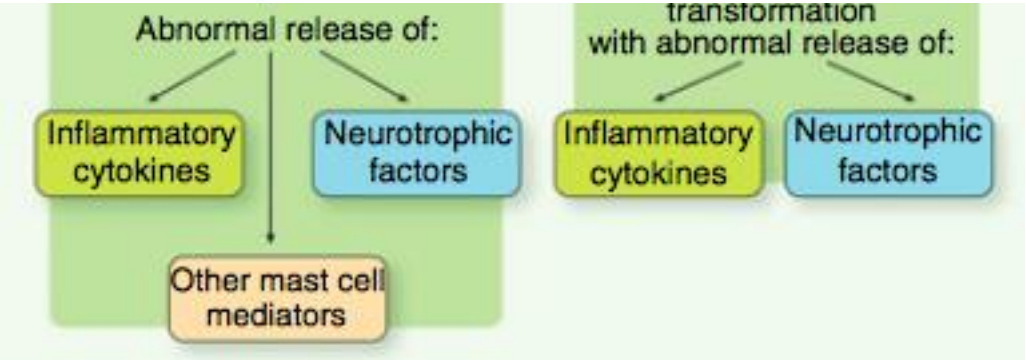
doi:10.1111/imm.12170

IMMUNOLOGY

REVIEW SERIES ON IMMUNE RESPONSES IN NEURODEGENERATIVE DISEASES

Series originated from a session at BSI Congress 2013

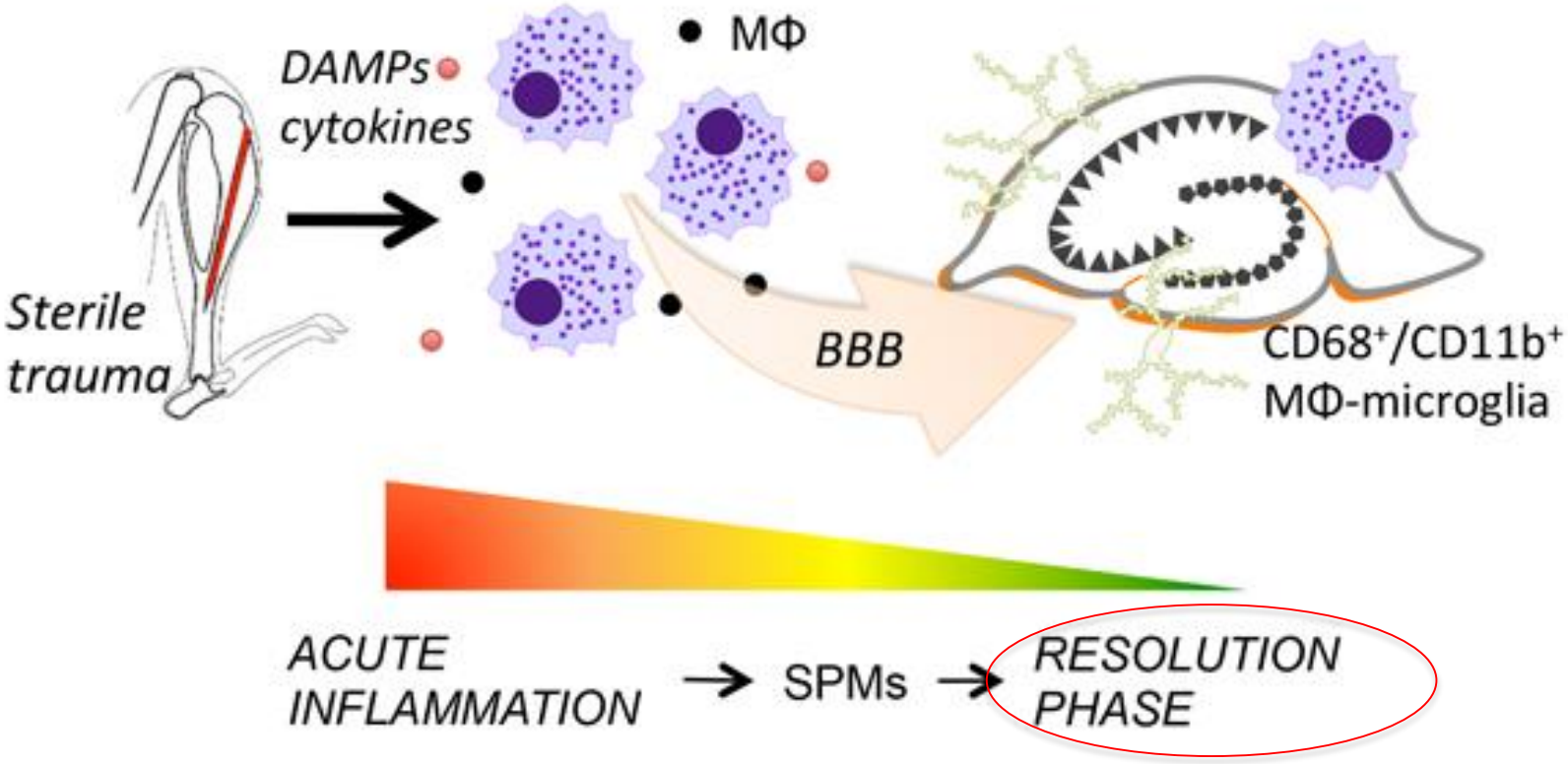
Mast cells, glia and neuroinflammation: partners in crime?



the deleterious signals acting on the neuron.

Immunology, 141, 314–327

The resolution of neuroinflammation is a process that allows for inflamed tissues to return to homeostasis



Trends in Pharmacological Sciences

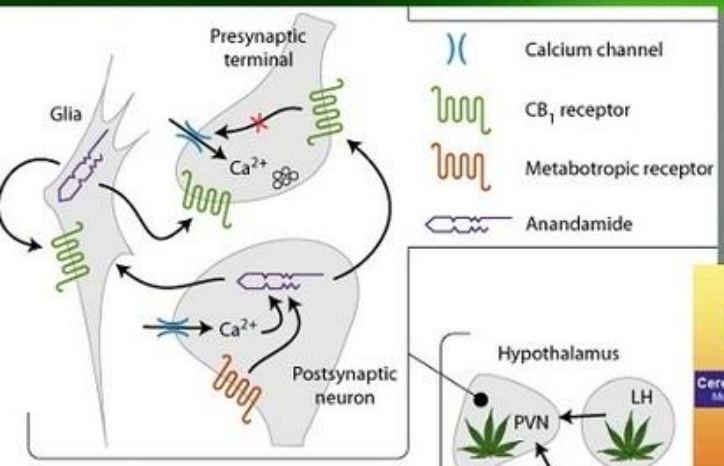
CellPress

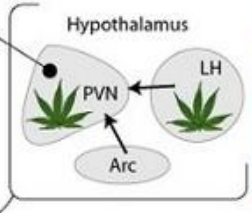
Review

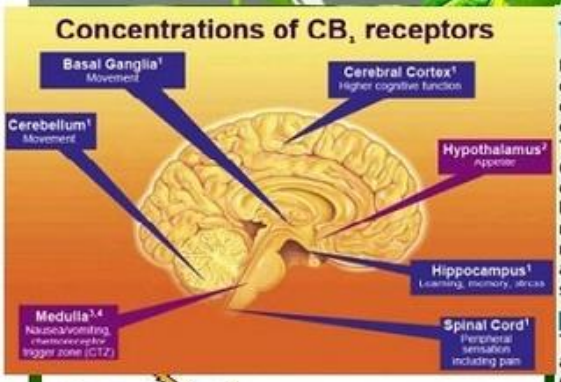
Resolution Pharmacology: Opportunities for Therapeutic Innovation in Inflammation

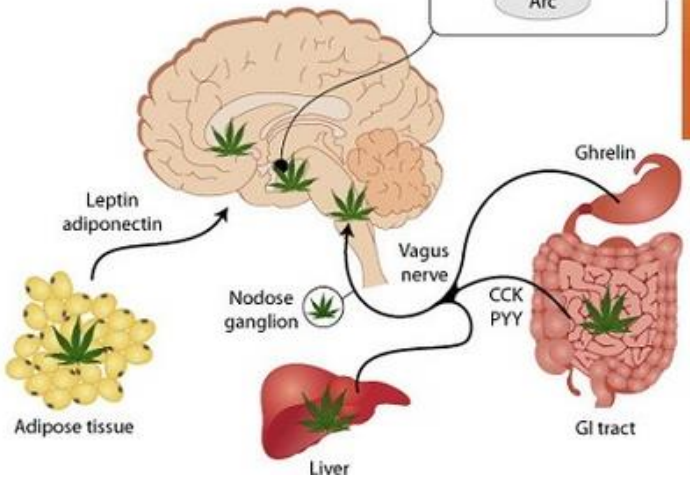
Mauro Perretti,[1,*] Xavier Leroy,[2] Elliot J. Bland,[3] and Trinidad Montero-Melendez[1]

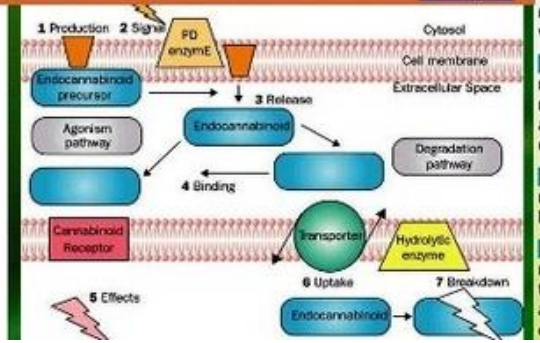
ENDOCANNABINOID SYSTEM











The Endocannabinoid System

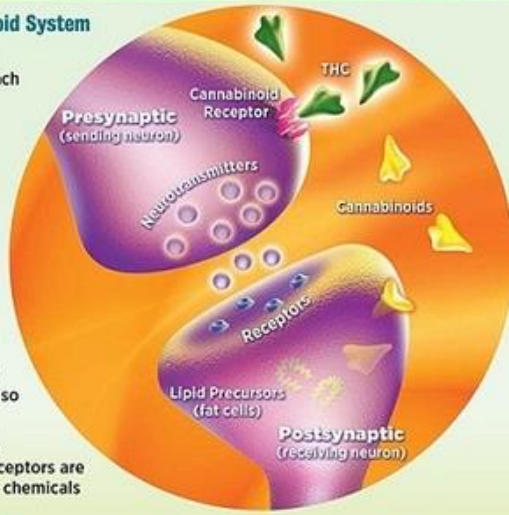
Brain cells (neurons) communicate with each other by sending chemical messages. The chemicals (neurotransmitters) cross a gap between neighboring neurons before attaching to their specific receptors.

Presynaptic: The neuron sending a message by releasing a chemical when signaled to do so

Postsynaptic: The neuron receiving the message when its receptors are activated by specific chemicals (neurotransmitters)

Neurotransmitters: The chemical messengers that travel from one brain cell to another

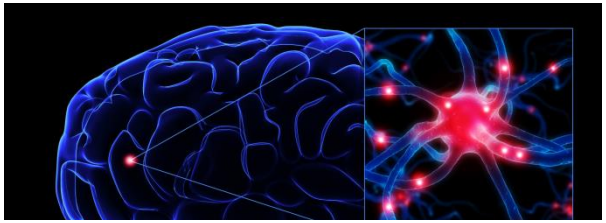
Receptors: Activated by neurotransmitters, receptors trigger a set of events that allows a message to be passed along to other neurons



Cannabinoids: Natural chemicals (anandamide and 2-AG) that bind to cannabinoid receptors in the brain and the body

THC: The main active ingredient in marijuana; THC, also a cannabinoid, interferes with the normal functioning of the endocannabinoid system

Endocannabinoid signalling



Boosting endocannabinoid tone might be a useful alternative therapeutic approach

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B

CrossMark click for updates

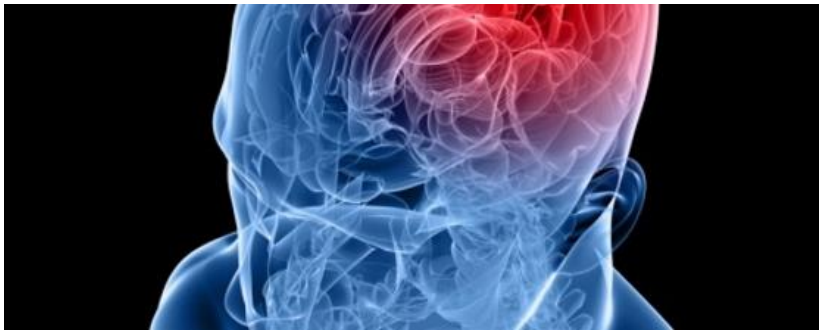
Phil. Trans. R. Soc. B (2012) **367**, 3193–3200
doi:10.1098/rstb.2012.0313

Introduction

Endocannabinoids in nervous system health and disease: the big picture in a nutshell

Stephen D. Skaper[1,*] and Vincenzo Di Marzo[2]

functionality might be considered a predisposing factor for neurological disorders



AEA levels in rat brain following acute and chronic drug administration

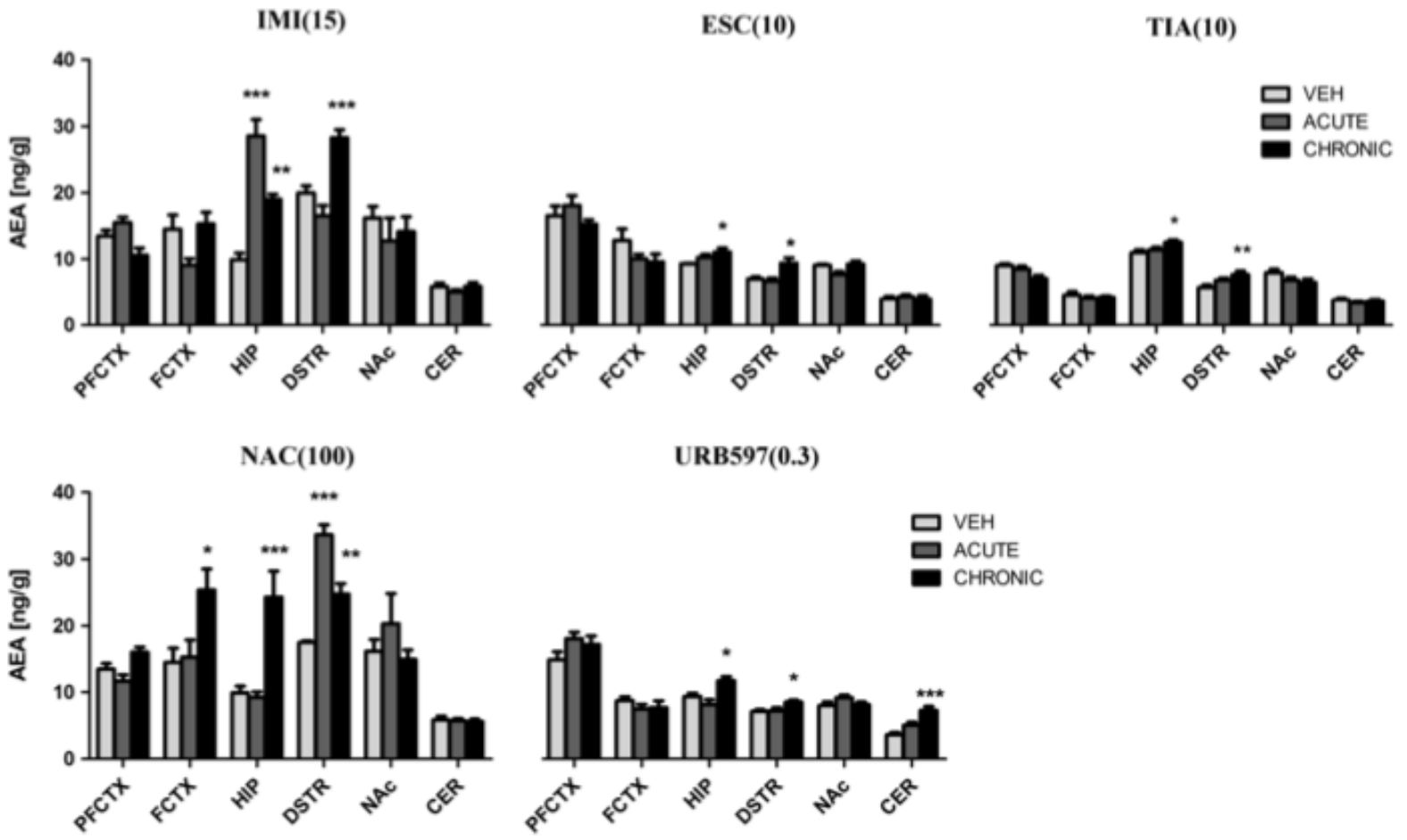


Fig. 1 AEA levels in rat brain structures following acute and chronic drug/compound administration. *AEA* Anandamide, *IMI(15)* imipramine hydrochloride (15 mg/kg), *ESC(10)* escitalopram oxalate, *TIA(10)* tianeptine sodium, *NAC(100)* *N*-acetylcysteine, *URB597(0.3)* cyclohexylcarbamic acid 3-carbamoylbiphenyl-3-yl ester, *PFCTX* prefrontal cortex, *FCTX* frontal cortex, *HIP* hippocampus, *DSTR* dorsal striatum, *NAc* nucleus accumbens, *CER* cerebellum. All data are expressed as the mean ± SEM. $N = 8$ rats/group. $^{*}p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$ versus corresponding vehicle

PEA levels in rat brain following acute and chronic drug administration

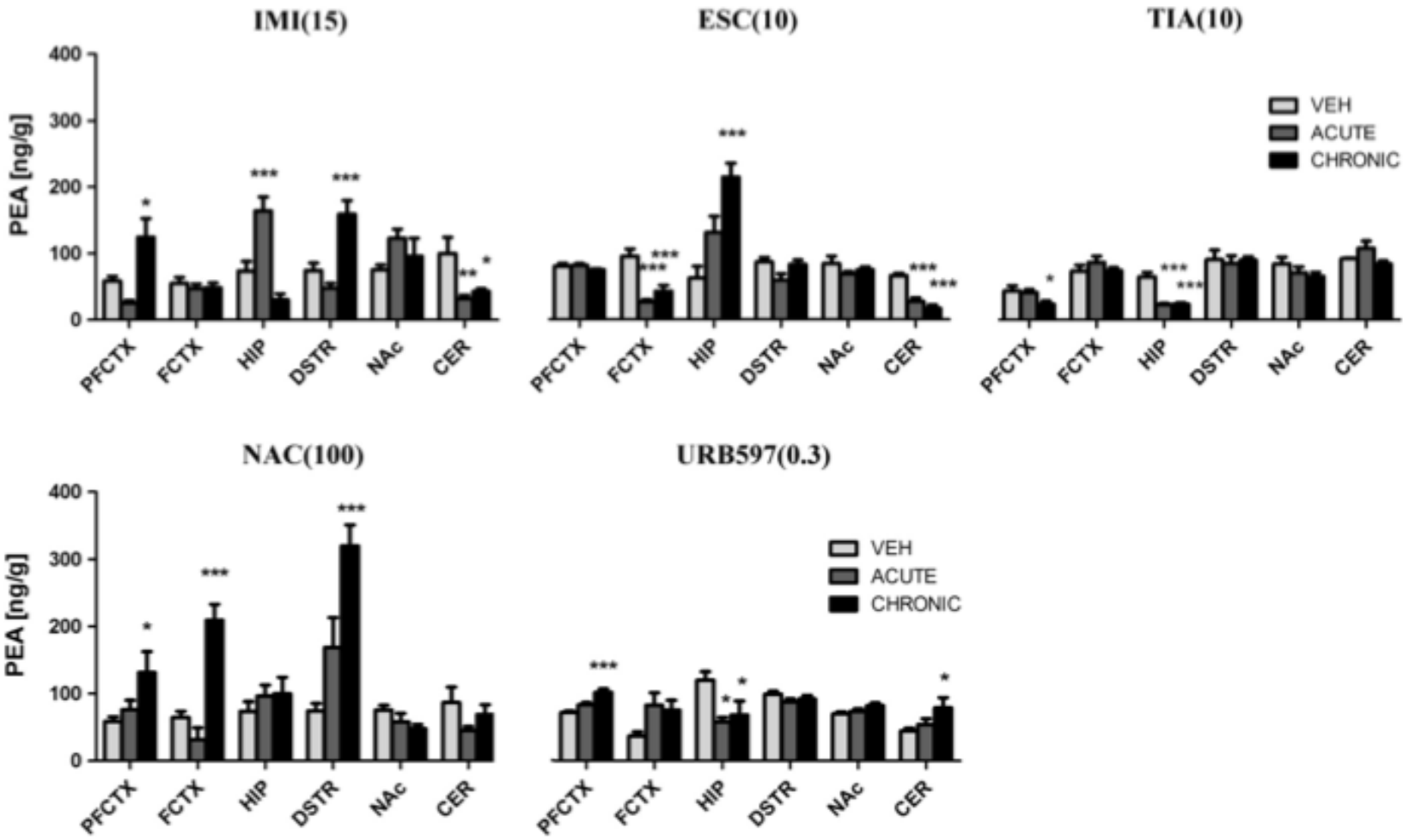


Fig. 5 PEA levels in rat brain structures following acute and chronic drug/compound administration. *PEA* Palmitoylethanolamide, *IMI(15)* imipramine hydrochloride (15 mg/kg), *ESC(10)* escitalopram oxalate, *TIA(10)* tianeptine sodium, *NAC(100)* *N*-acetylcysteine, *URB597(0.3)* cyclohexylcarbamic acid 3-carbamoylbiphenyl-3-yl ester, *PFCTX* prefrontal cortex, *FCTX* frontal cortex, *HIP* hippocampus, *DSTR* dorsal striatum, *NAc* nucleus accumbens, *CER* cerebellum. All data are expressed as the mean ± SEM. $N = 8$ rats/group. $^{*}p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$ versus corresponding vehicle

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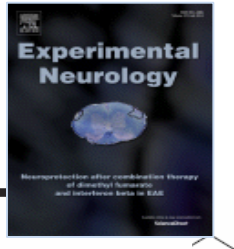
journal homepage: www.elsevier.com/locate/yexnr

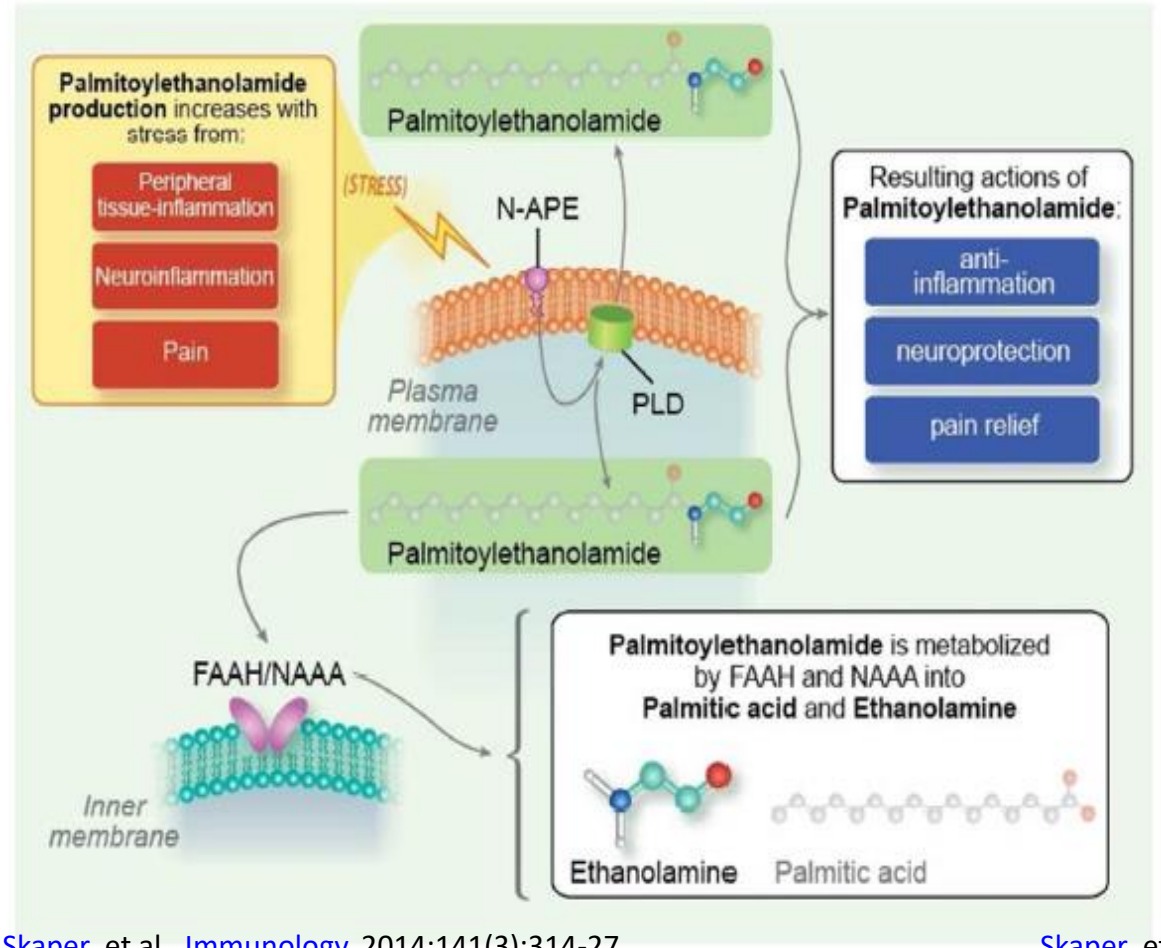


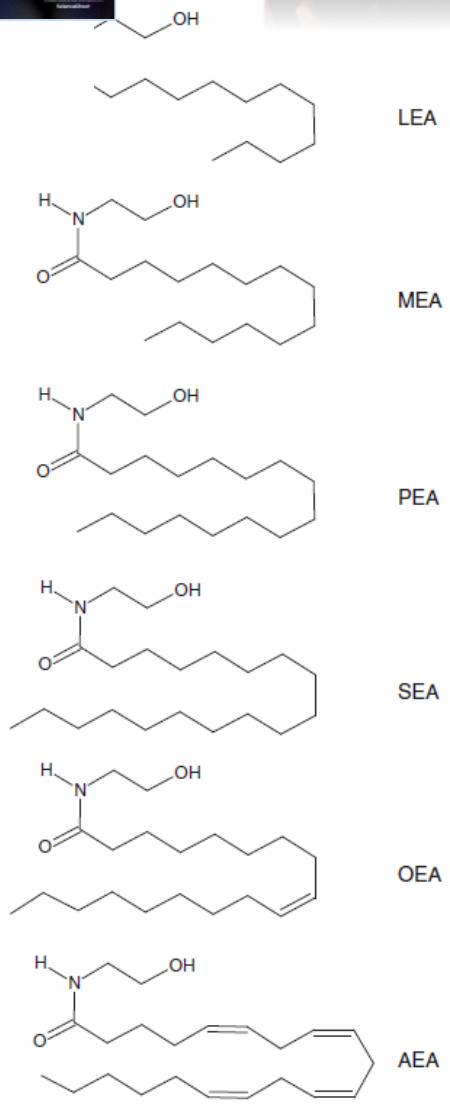
Review

Palmitoylethanolamide and other anandamide congeners. Proposed role in the diseased brain

Harald S. Hansen *

Amides of fatty acids and ethanolamine, called N-Acylethanolamines or acylethanolamides.

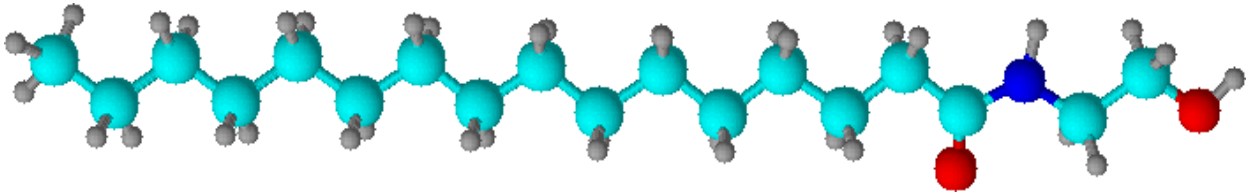




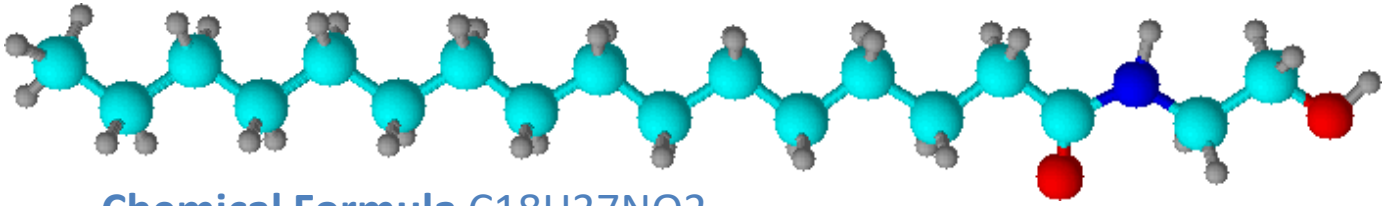
Skaper et al., Immunology. 2014;141(3):314-27.

Skaper et al., Inflammopharmacology. 2014;22(2):79-94.

What is PEA?



- **Naturally occurring fatty acid amide.**
  - Fatty acid derivative (C16:0 = palmitic acid) amidated by ethanolamine



Chemical Formula C18H37NO2

Molecular Weight 299.49

Nomenclature

Palmitoylethanolamide, N-Palmitoylethanolamine; Palmidrol [INN]; palmitylethanolamide; Hydroxyethylpalmitamide; LG 2110/1; N-(2-hydroxyethyl)hexadecanamide.

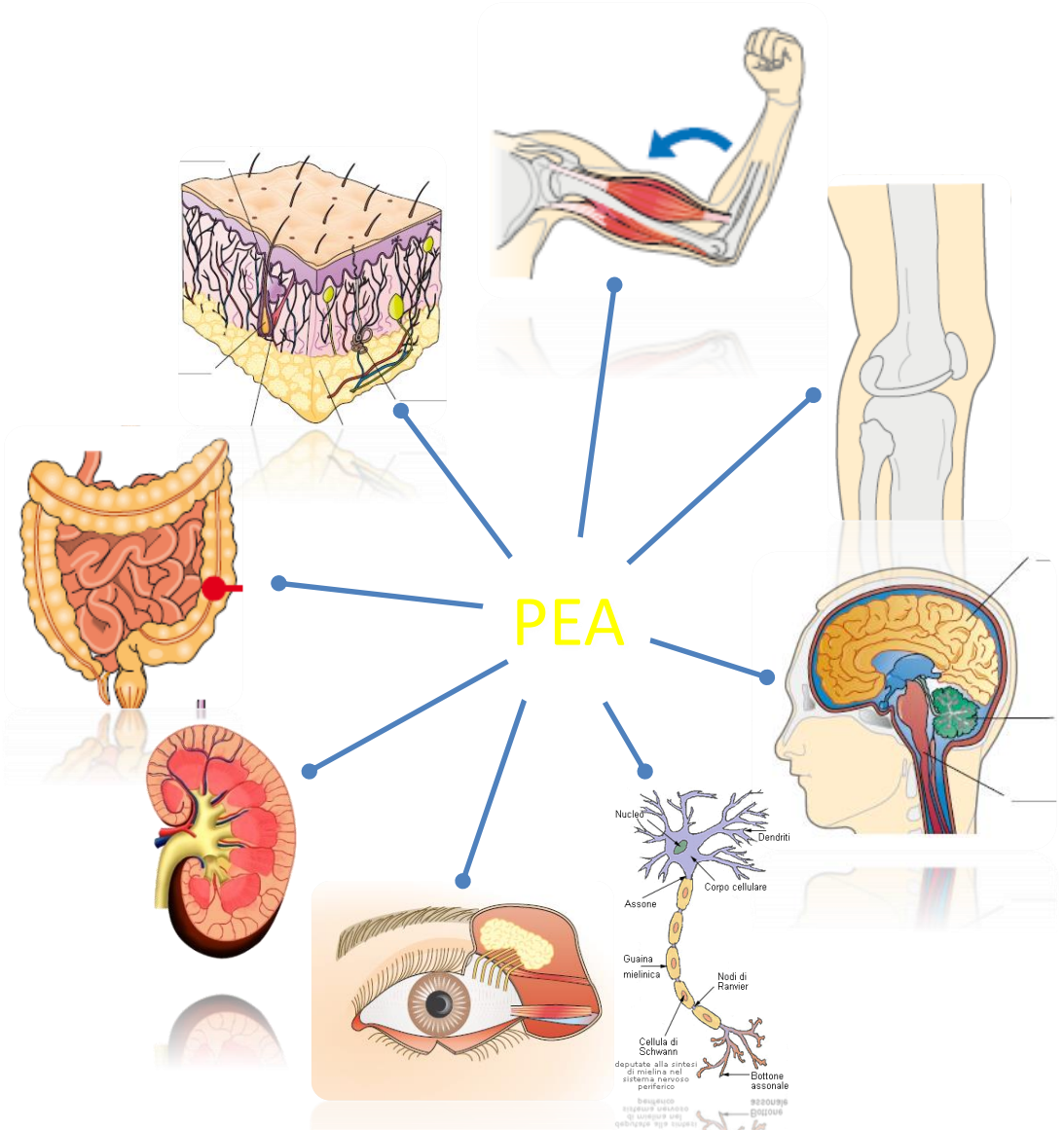
PEA levels in normal tissues

- In mammals, it has been found in many tissues and body fluids.

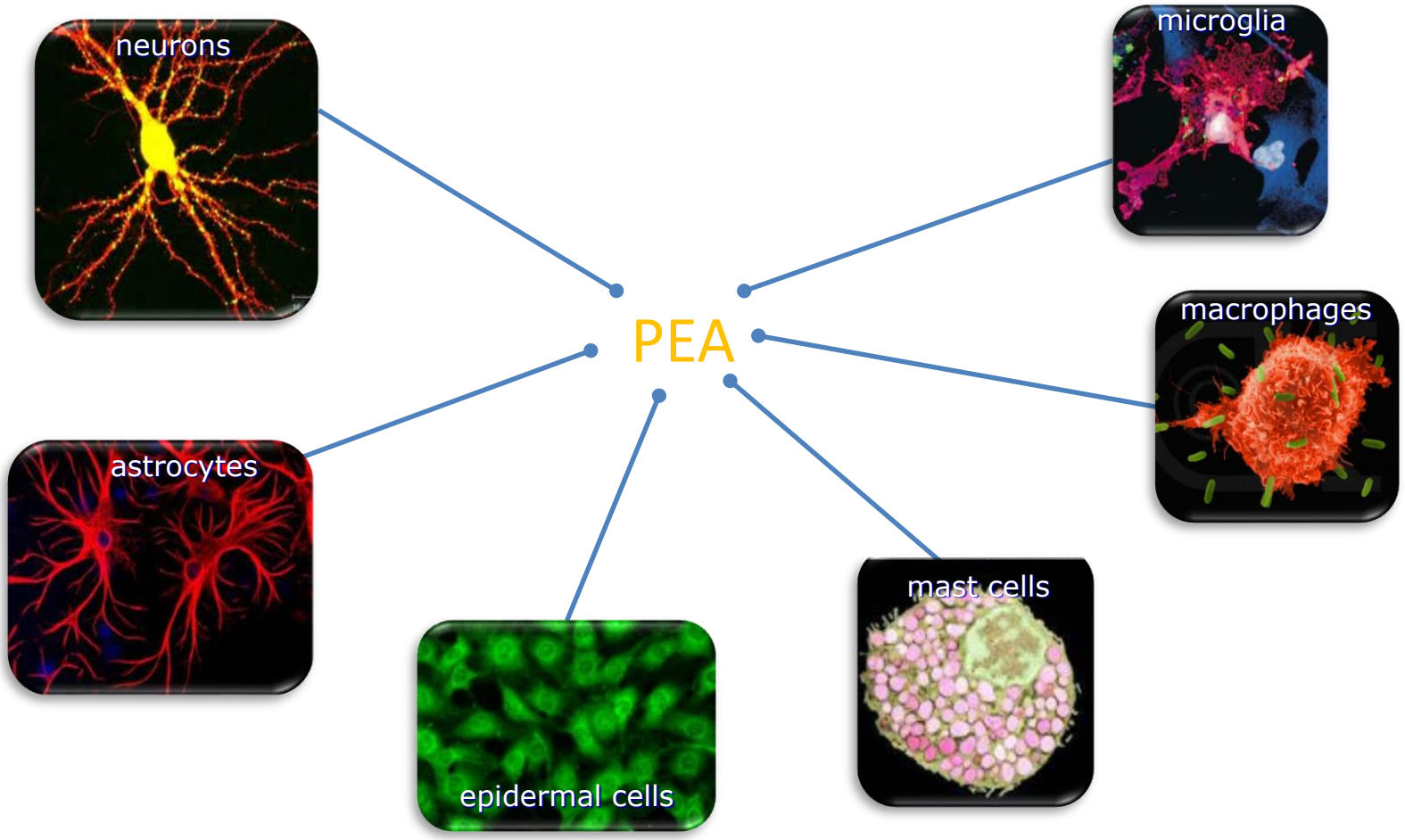
The levels of endogenous PEA in normal muscle and adipose tissues (0.04 - 6.00 pmol/mg)

are similar to those observed in the CNS and PNS

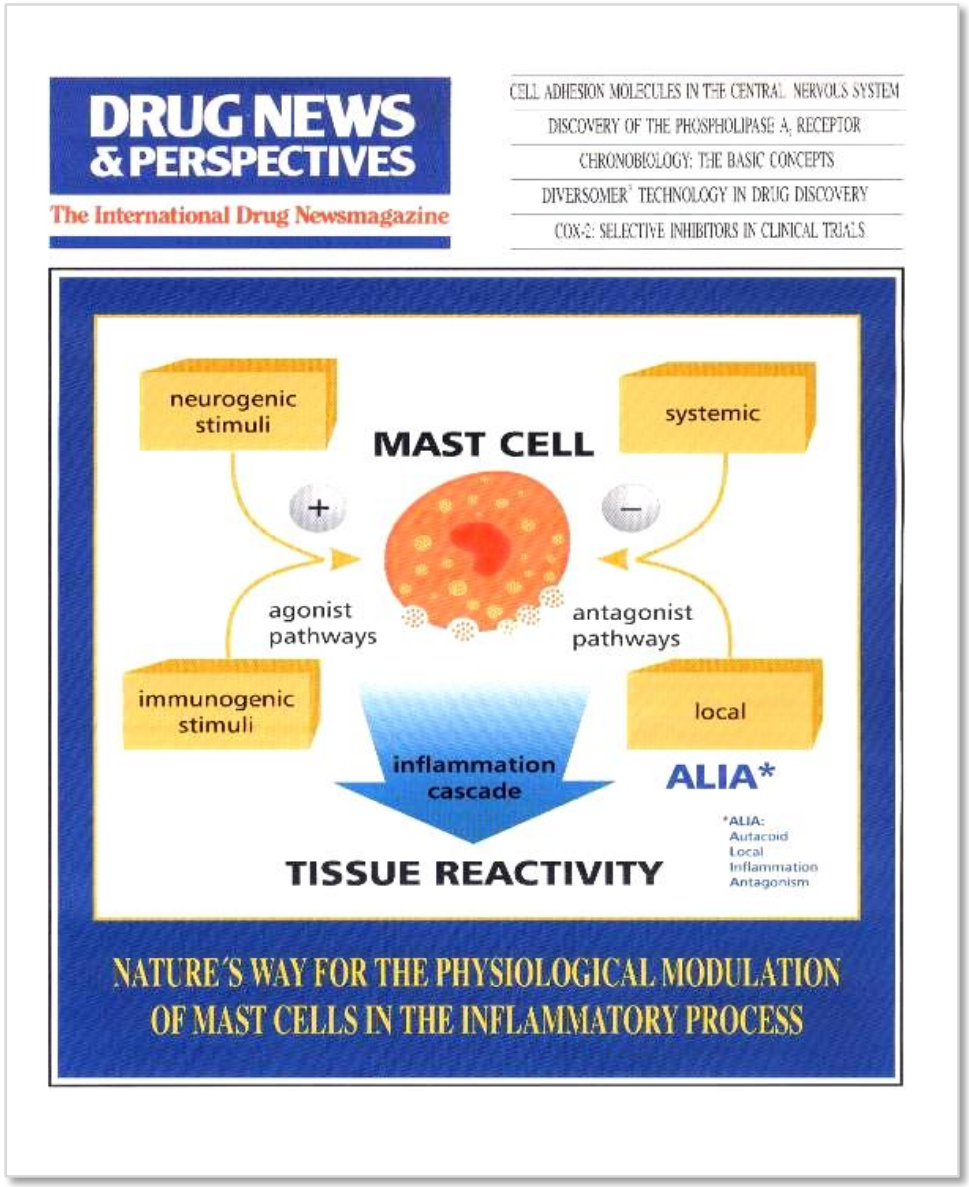
and something higher compared to the gastrointestinal tract (0.05-1.50 pmol/mg), skin (0.3- 0.69 pmol/mg) eye (0.10 - 0.64 pmol/mg) and kidney (2.5 – 7.1 pmol/mg).



Produced "*on demand*" by mast cells, microglia exposed to potential damage



ALIA mechanism



Autacoid
Local
Injury
Antagonism

"Aliamides
may represent the major
breakthrough
in inflammation we have been
waiting for."

Jack D. 1996 DN&P

PEA is formed locally in tissue suffering from inflammation

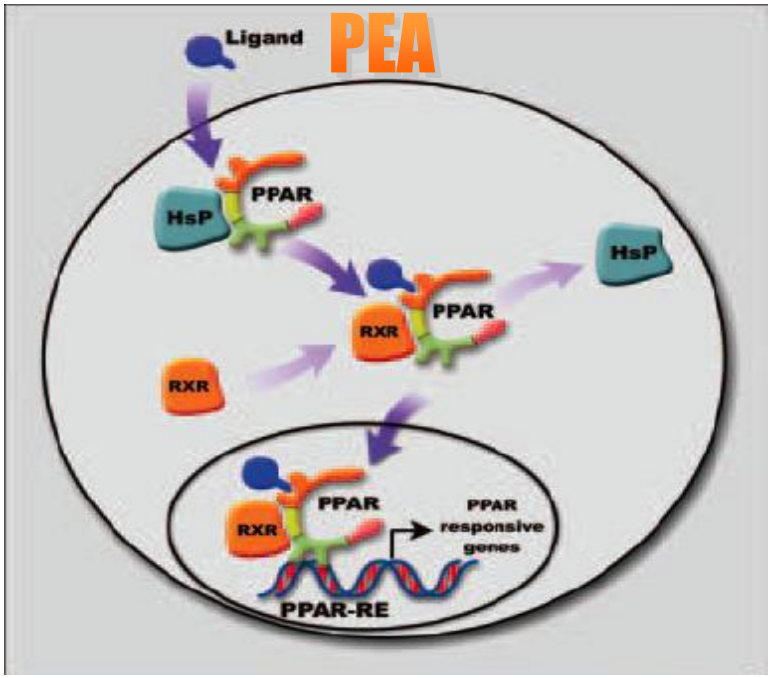
PALMITOYLETHANOLAMIDE : AN ENDOGENOUS PPAR-α AGONIST

MOL PHARM

ACCELERATED COMMUNICATION

The Nuclear Receptor Peroxisome Proliferator-Activated Receptor-α Mediates the Anti-Inflammatory Actions of Palmitoylethanolamide

Jesse Lo Verme, Jin Fu, Giuseppe Astarita, Giovanna La Rana, Roberto Russo, Antonio Calignano, and Daniele Piomelli



0022-3565/07/3223-1137–1143$20.00
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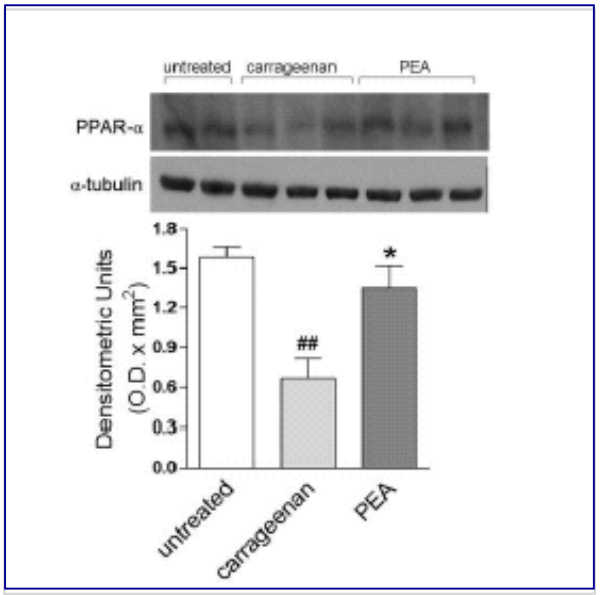
JPET 322:1137–1143, 2007
Vol. 322, No. 3
123265/3244537
Printed in U.S.A.

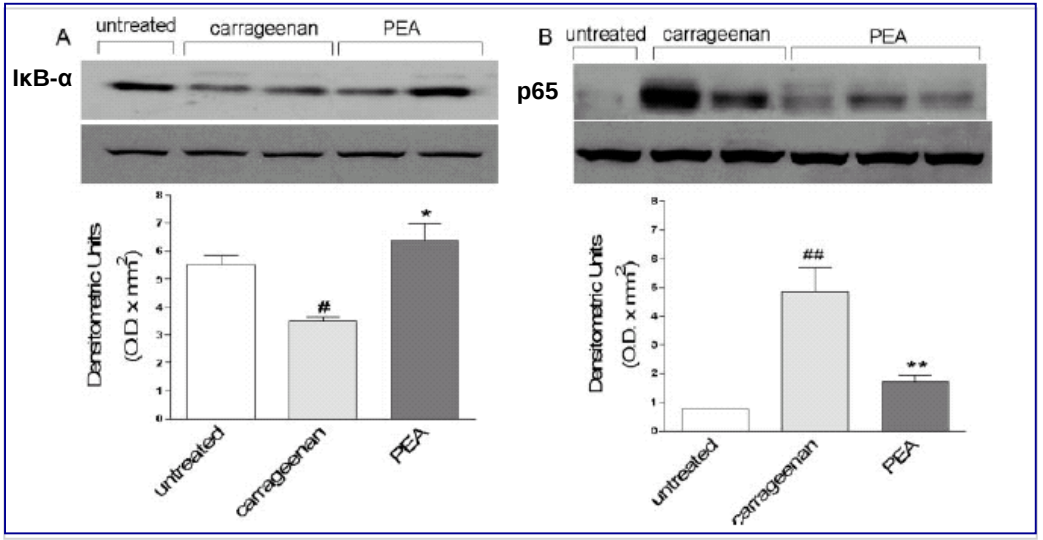
Acute Intracerebroventricular Administration of Palmitoylethanolamide, an Endogenous Peroxisome Proliferator-Activated Receptor-α Agonist, Modulates Carrageenan-Induced Paw Edema in Mice

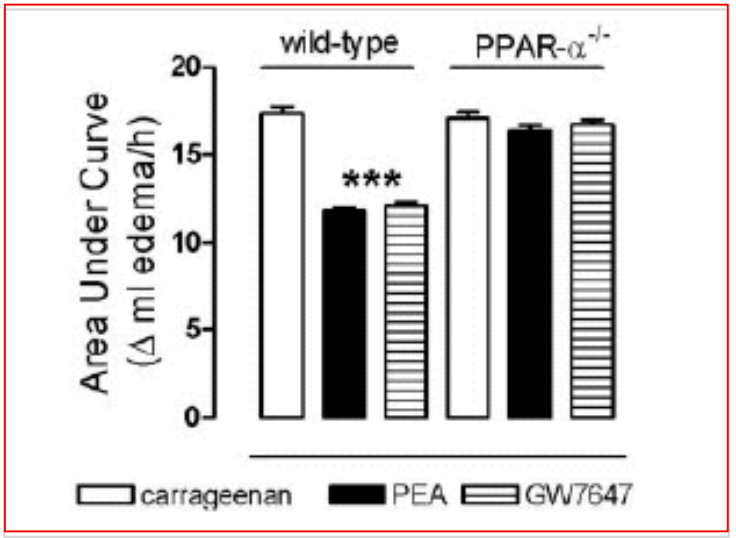
Giuseppe D'Agostino, Giovanna La Rana, Roberto Russo, Oscar Sasso, Anna Iacono, Emanuela Esposito, Giuseppina Mattace Raso, Salvatore Cuzzocrea, Jesse Lo Verme, Daniele Piomelli, Rosaria Meli, and Antonio Calignano

Department of Experimental Pharmacology, University of Naples "Federico II", Naples, Italy (G.D., G.L.R., R.R., O.S., A.I., E.E., G.M.R., R.M., A.C.); Istituti di Ricovero e Cura a Carattere Scientifico Centro Neurolesi "Bonino-Pulejo", University of Messina, Messina, Italy (E.E., S.C.); Department of Clinical and Experimental Medicine and Pharmacology, School of Medicine, University of Messina, Messina, Italy (S.C.); and Department of Pharmacology, University of California, Irvine, California (J.L.V., D.P.)

Received March 27, 2007; accepted June 11, 2007







N-palmitoylethanolamide (PEA)

Analgesic

IASP

PAIN® 153 (2012) 33–41

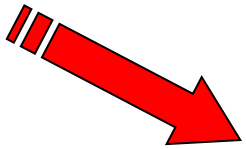
PAIN®

www.elsevier.com/locate/pain

Implication of allopregnanolone in the antinociceptive effect of *N*-palmitoylethanolamide in acute or persistent pain

Oscar Sasso [a], Roberto Russo [a], Sergio Vitiello [b,c], Giuseppina Mattace Raso [a], Giuseppe D'Agostino [a], Anna Iacono [a], Giovanna La Rana [a], Monique Vallée [b,c], Salvatore Cuzzocrea [d,e], Pier Vincenzo Piazza [b,c], Rosaria Meli [a,*], Antonio Calignano [a]

Anti-inflammatory



ELSEVIER

Brain, Behavior, and Immunity

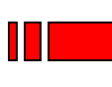
Available online 3 August 2012

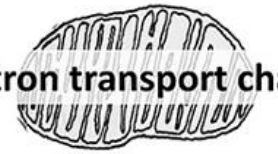
In Press, Uncorrected Proof — Note to users

Administration of palmitoylethanolamide (PEA) protects the neurovascular unit and reduces secondary injury after traumatic brain injury in mice

Akbar Ahmad[a], Rosalia Crupi[a], Daniela Impellizzeri[a], Michela Campolo[a], Angela Marino[b], Emanuela Esposito[a], Salvatore Cuzzocrea[a, c,]

Antioxidant

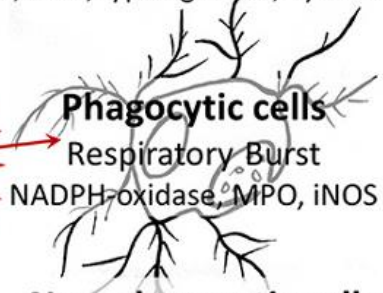




Electron transport chain

Cytosolic enzymes
XO, NOS, lypoxigenase, cyt P450

Free Radicals Producers
endogenous & exogenous



Phagocytic cells
Respiratory Burst
NADPH-oxidase, MPO, iNOS

Non-phagocytic cells
NADPH-oxidase

Environment
Radiation, pollution, chemicals, drugs

OXIDANT COMPOUNDS

Free Radicals
ROS & RNS

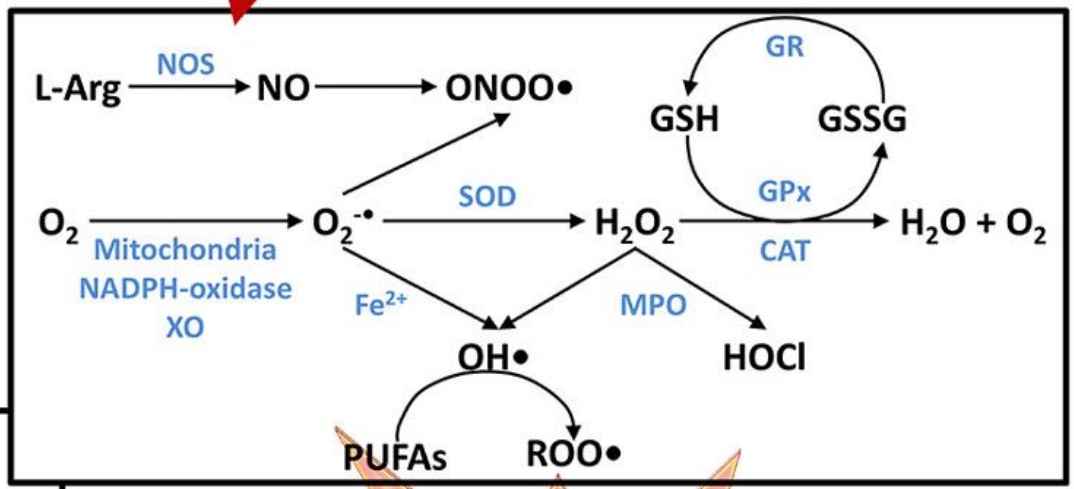
Transition metals
Fe^{+2}, Cu^{+2}

Others
$O_2^{-\bullet}$/OH•/ROO•/RO•
H_2O_2/ONOO•/NO/Fe^{2+}/Cu^{2+}

ANTIOXIDANT CAPACITY

Antioxidant Enzymes
CAT, SOD, GPx, GR

Non-enzymatic compounds
GSH, thioredoxin, Vits C & E, trace metals (Se, Zn)



Regulated

CELL SIGNALING

Gene Regulation
Signal Transduction Modulation

NFκβ (activation) → **Regulation of:**
- Cell proliferation
- Apoptosis
- Cognitive functions
- Immune functions

Unregulated

OXIDATIVE STRESS

Lipids - Lipid peroxidation (MDA)
Proteins - Oxidized proteins (3-NT)
Nucleic acids - DNA oxidation (8-0HdG) strand breaks

OXIDATIVE DAMAGE

Science
AAAS

A Nonpeptidyl Mimic of Superoxide Dismutase with Therapeutic Activity in Rats
Daniela Salvemini, *et al.*
Science **286**, 304 (1999);
DOI: 10.1126/science.286.5438.304

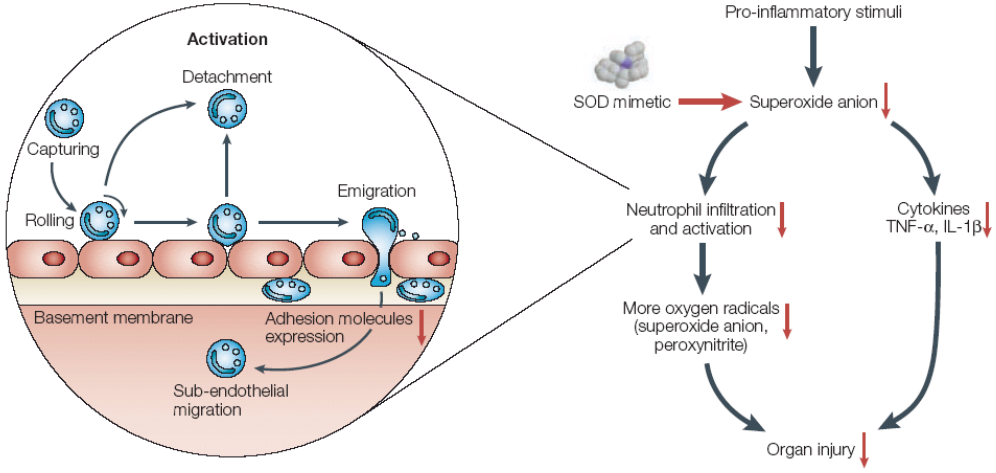
NATURE REVIEWS | DRUG DISCOVERY VOLUME 1 | MAY 2002 | 367

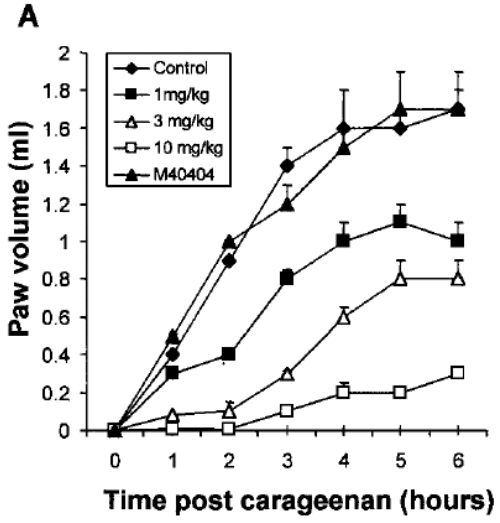
SOD MIMETICS ARE COMING OF AGE

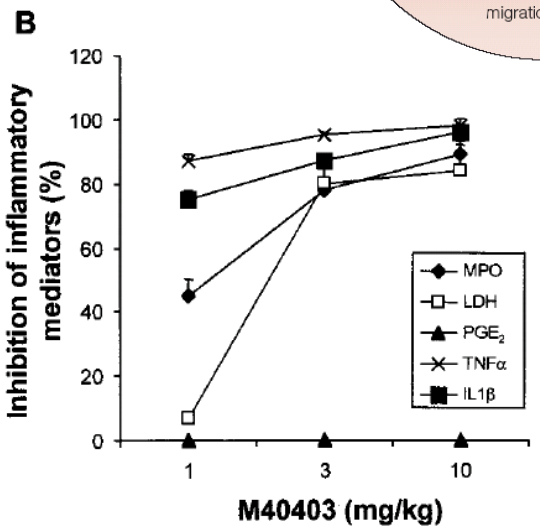
Daniela Salvemini, Dennis P. Riley* and Salvatore Cuzzocrea‡*

SODm

One of two subunits





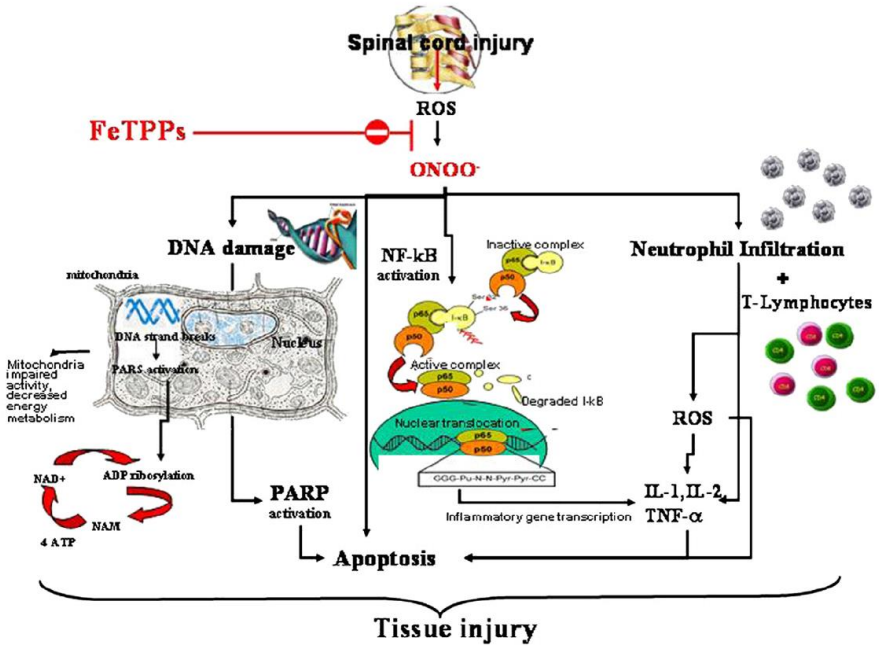


Available online at www.sciencedirect.com
ScienceDirect
Free Radical Biology & Medicine 43 (2007) 763–780
www.elsevier.com/locate/freeradbiomed

Original Contribution

Beneficial effects of FeTSPP, a peroxynitrite decomposition catalyst, in a mouse model of spinal cord injury

Tiziana Genovese [a,b], Emanuela Mazzon [a,b], Emanuela Esposito [b,c], Carmelo Muià [a], Rosanna Di Paola [a,b], Placido Bramanti [b], Salvatore Cuzzocrea [a,b,*]



NCBI Resources How To
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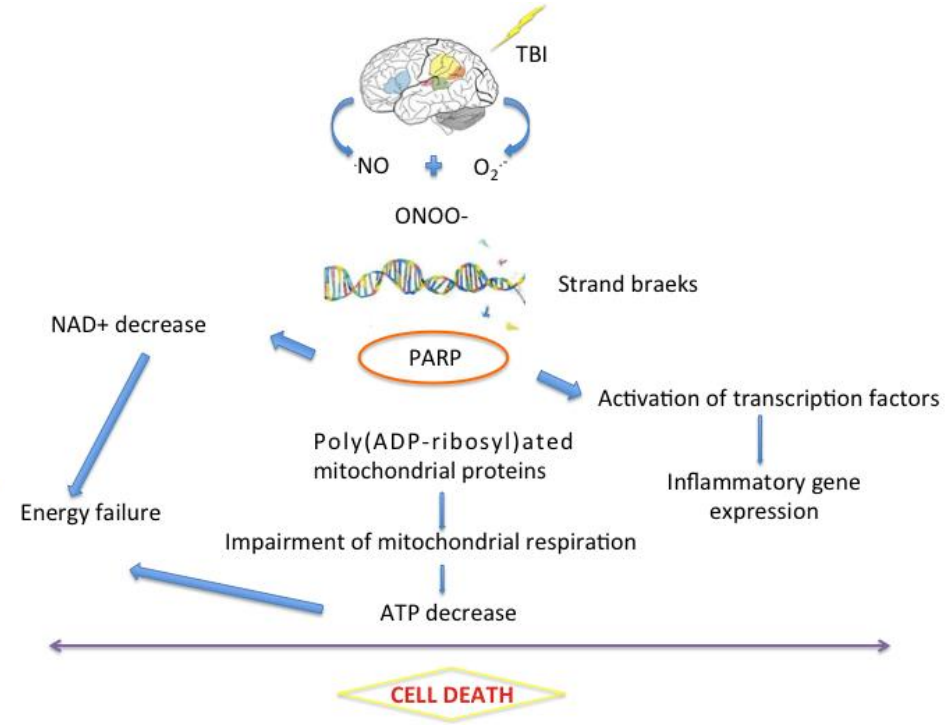
Advanced
Display Settings: Abstract
Antioxid Redox Signal. 2013 Apr 2. [Epub ahead of print]

TRAUMATIC BRAIN INJURY (TBI): OXIDATIVE STRESS AND NEUROPROTECTION.

Cornelius C, Crupi R, Calabrese V, Graziano A, Milone P, Pennisi G, Radak Z, Calabrese EJ, Cuzzocrea S.
University of Catania, Department of Chemistry, Catania, Italy ; carolin.cornelius@receptura.de.



Luteolin

Drug Design, Development and Therapy

Dovepress

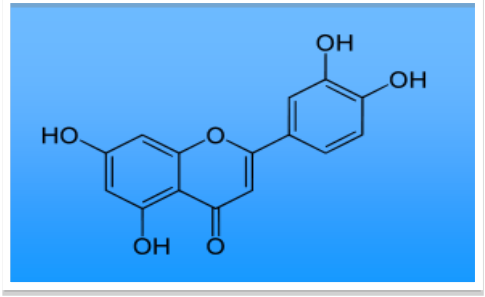
open access to scientific and medical research

Open Access Full Text Article

ORIGINAL RESEARCH

Flavonoids targeting of IκB phosphorylation abrogates carcinogen-induced MMP-9 and COX-2 expression in human brain endothelial cells

Pharmacological activity:



Journal of Neuroinflammation

Commentary

Luteolin as a therapeutic option for multiple sclerosis

Theoharis C Theoharides[1,2]

Int J Neurosci. 2011 Jun;121(6):329-36. doi: 10.3109/00207454.2011.569040.

Luteolin inhibits microglial inflammation and improves neuron survival against inflammation.

Zhu LH, Bi W, Qi RB, Wang HD, Lu DX.

Department of Pathophysiology, Institute of Brain Research, Key Laboratory of State Administration of Traditional Chinese Medicine of the People's Republic of China, School of Medicine, JiNan University, Guangzhou 510632, Guangdong, China.

Neurol Res. 2011 Nov;33(9):927-34. doi: 10.1179/1743132811Y.0000000023.

Luteolin reduces primary hippocampal neurons death induced by neuroinflammation.

Zhu LH, Bi W, Qi RB, Wang HD, Wang ZG, Zeng Q, Zhao YR, Lu DX.

Department of Pathophysiology, Institute of Brain Research, Key Laboratory of State Administration of Traditional Chinese Medicine of the People's Republic of China.

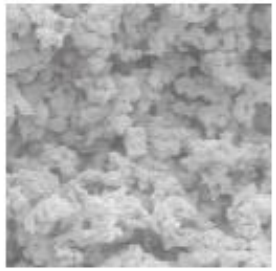
PEA

PEALUT

LUT

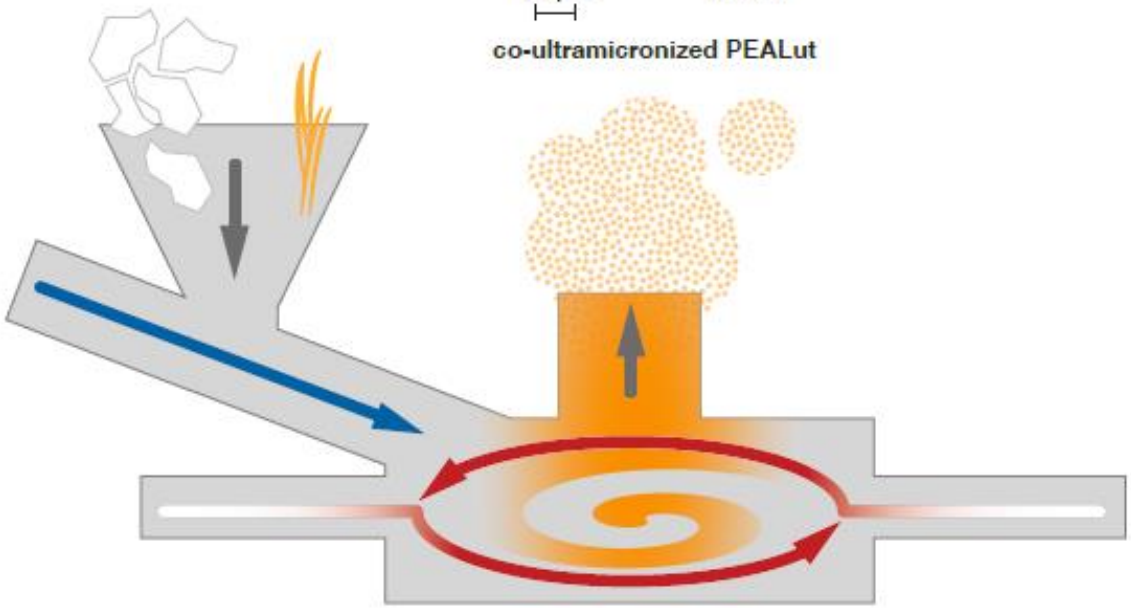
20 µm 500 X

PEA - Luteolin



10 µm 2000 X

co-ultramicronized PEALut



Micronized coultraPEALut association

ratio of 10:1 in mass

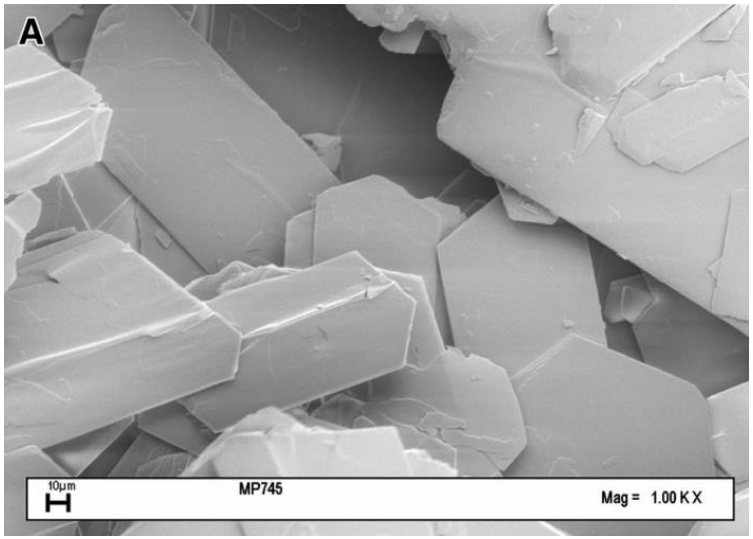
was obtained by a co-ultramicronization process in an jet-mill equipment

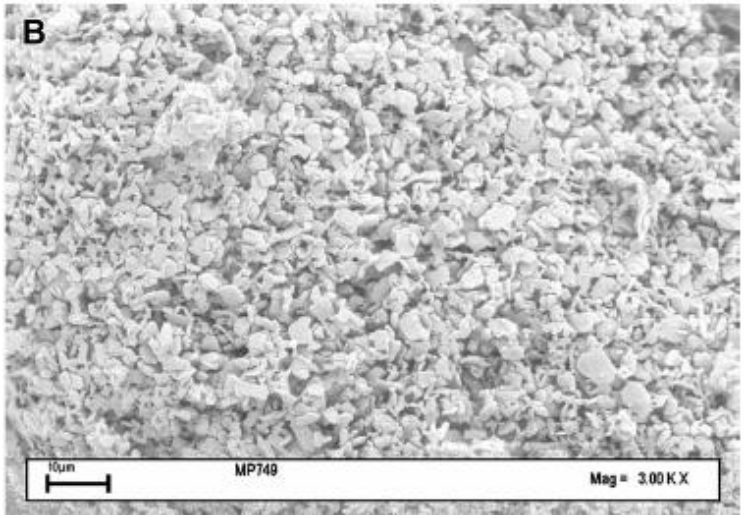
Jet milling is an innovative method that has many advantages such as the ability to produce micro-sized particles with a narrow particle size distribution and the absence of contamination

Skaper et al., Inflammopharmacology. 2014;22(2):79-94.

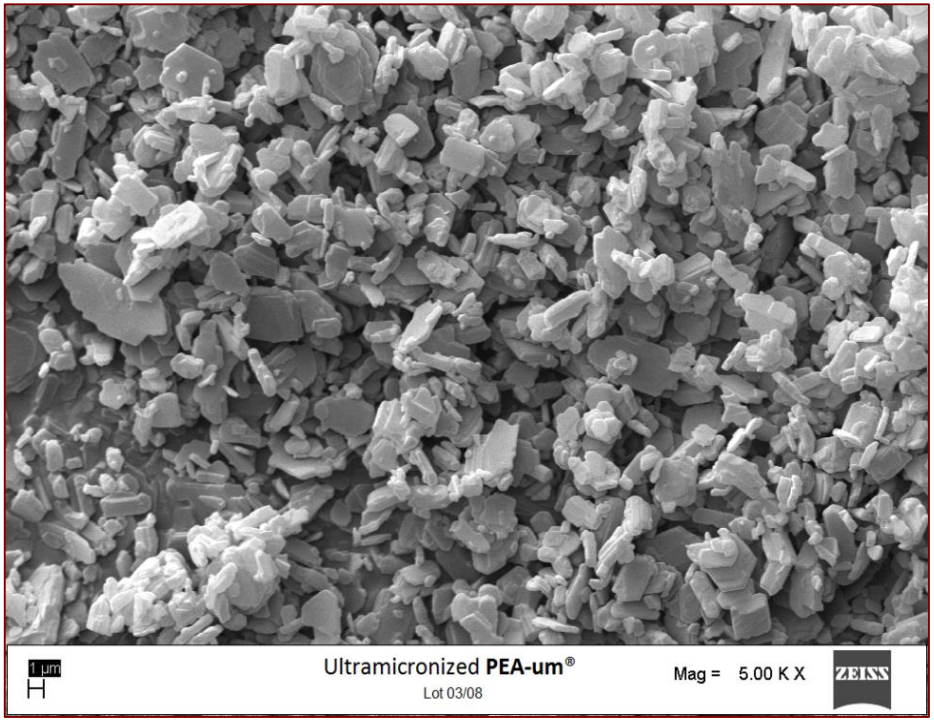
Table 3 Particle size profile of naive PEA compared to micronized and ultra-micronized PEA

Particle size	Naive PEA	Micronized PEA	Ultra-micronized PEA
100–700 microns	100 %	Absent	Absent
>14 microns	–	Trace	Absent
<10 microns	–	About 96 %	100 %
<6 microns	–	80 %	99.9 %
<2 microns	–	Not determined	59.6 %
<1 microns	–	Not determined	14.7 %
<0.6 micron	–	Not determined	2.0 %



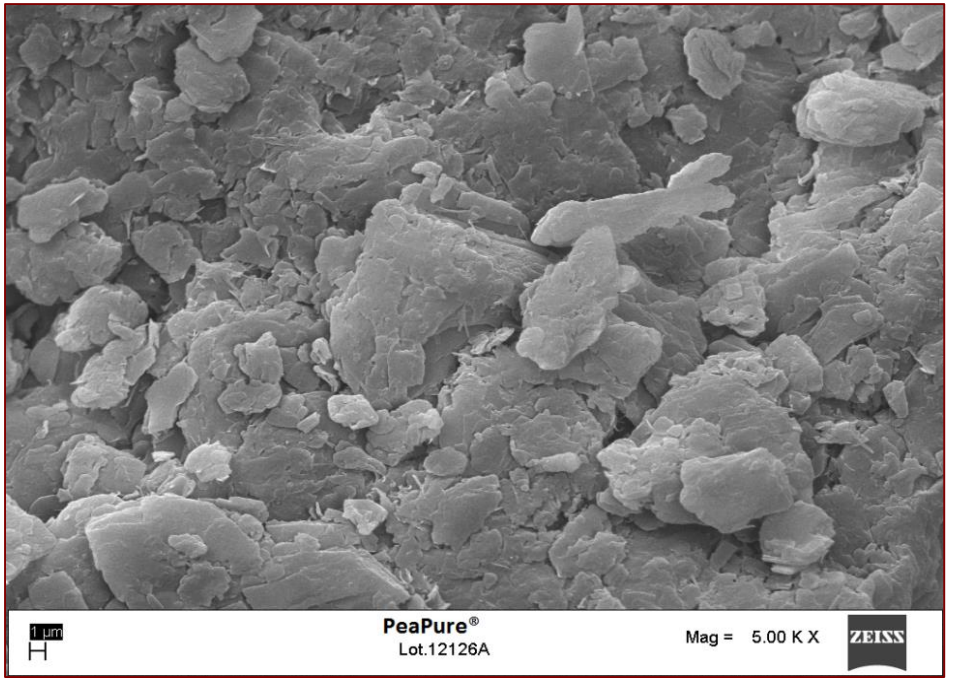


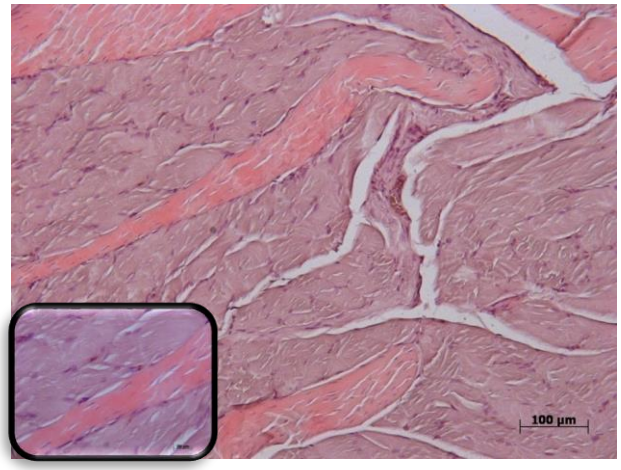
Photomicrographs of naïve PEA (A) and ultra-micronized (B) PEA obtained by scanning electron microscopy. Bar 10 microns

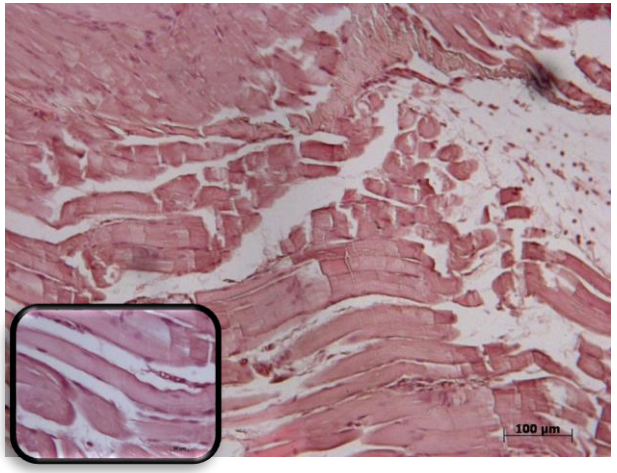

1 µm
Ultramicronized PEA-um®
Lot 03/08
Mag = 5.00 K X
ZEISS

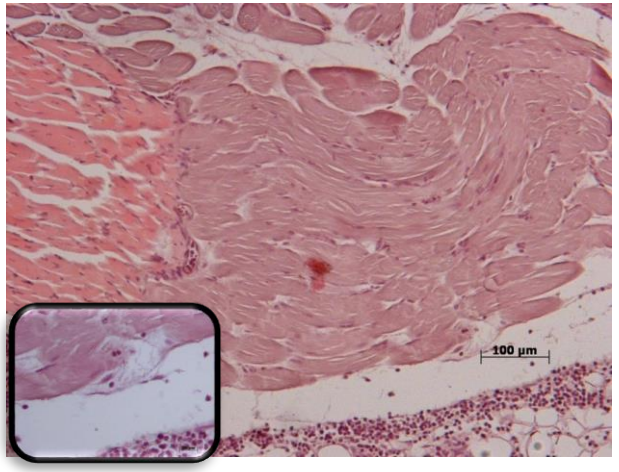
1 µm
Ultramicronized PEA-um®
Lot 03/08
Mag = 5.00 K X
ZEISS

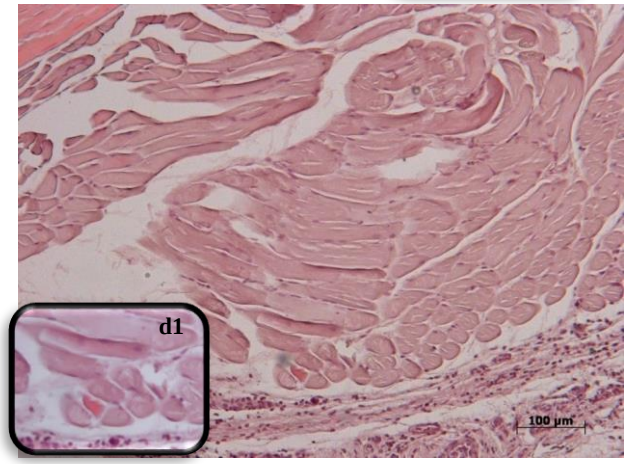
1 µm
PeaPure®
Lot.12126A
Mag = 5.00 K X
ZEISS

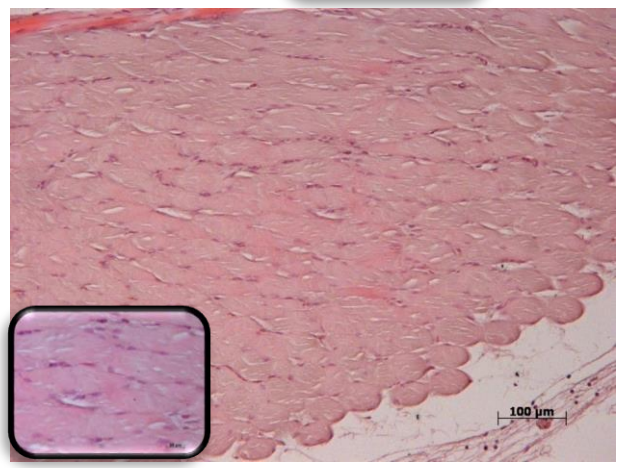

1 µm
PeaPure®
Lot.12126A
Mag = 5.00 K X
ZEISS

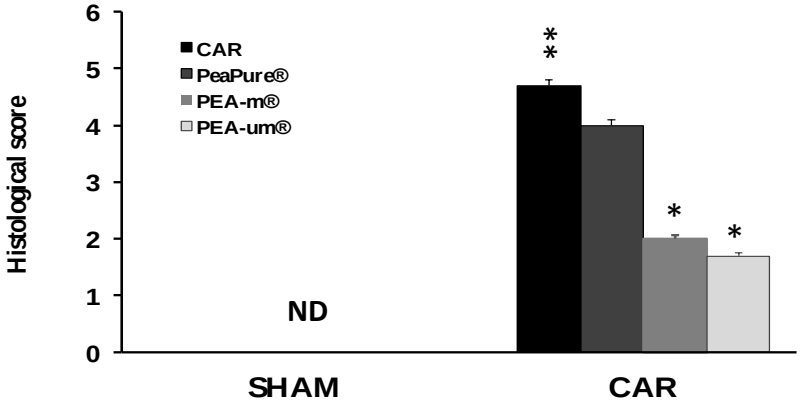

100 µm

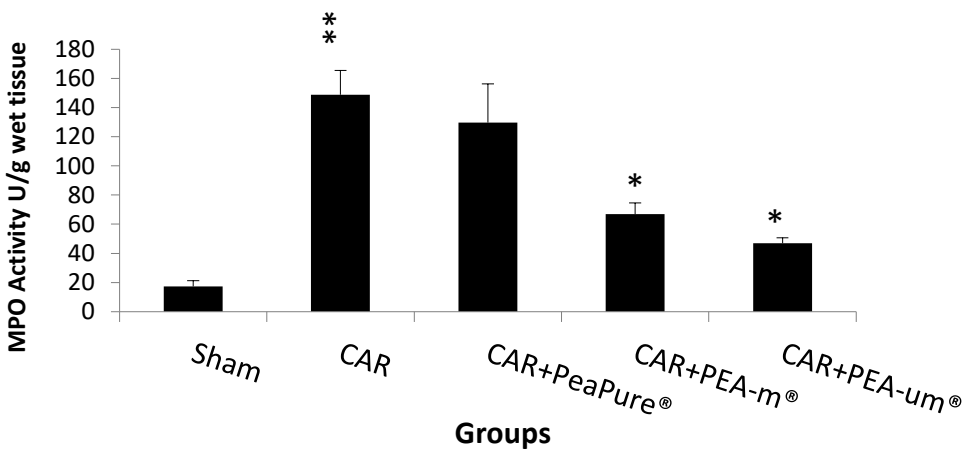

100 µm

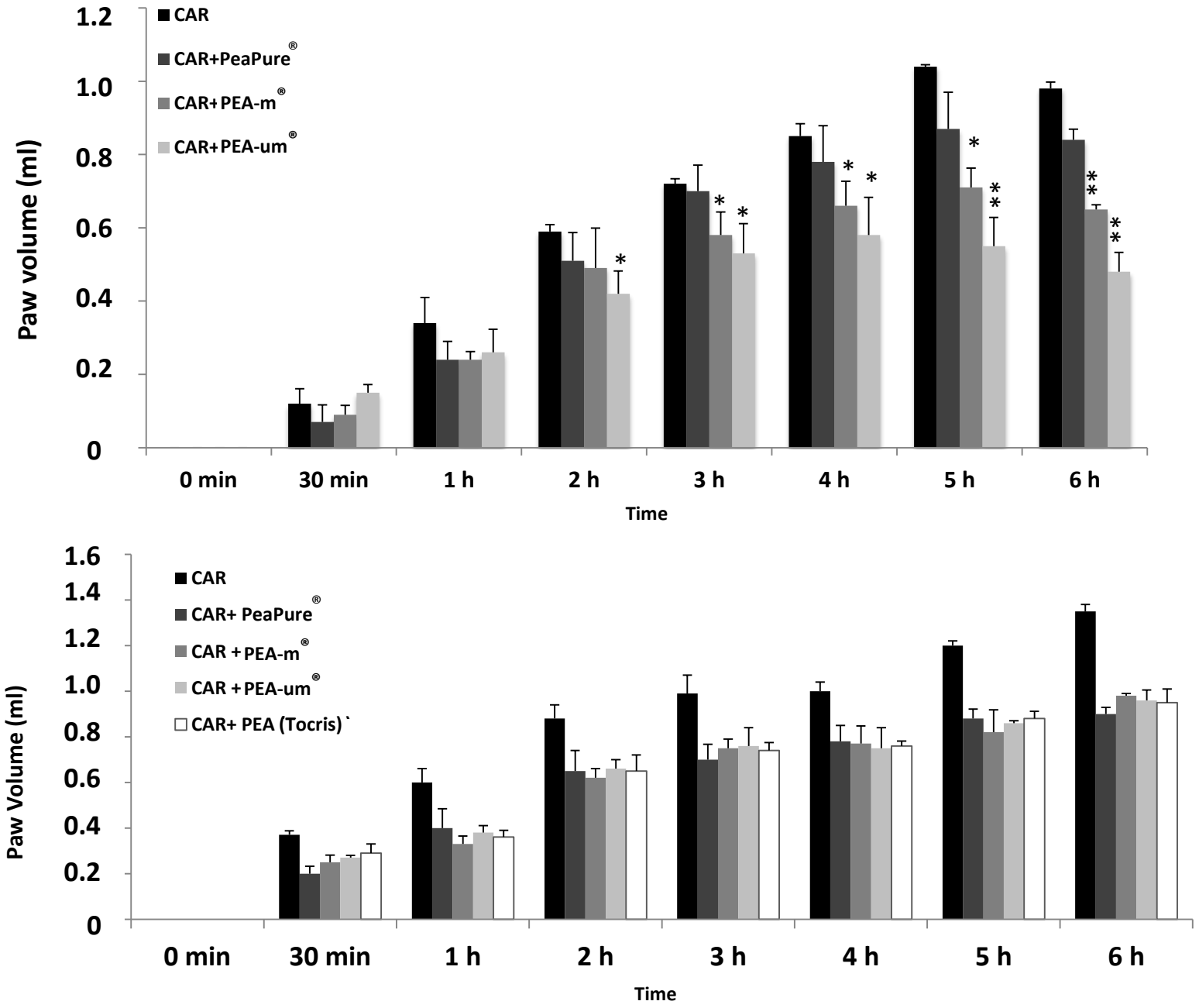

100 µm

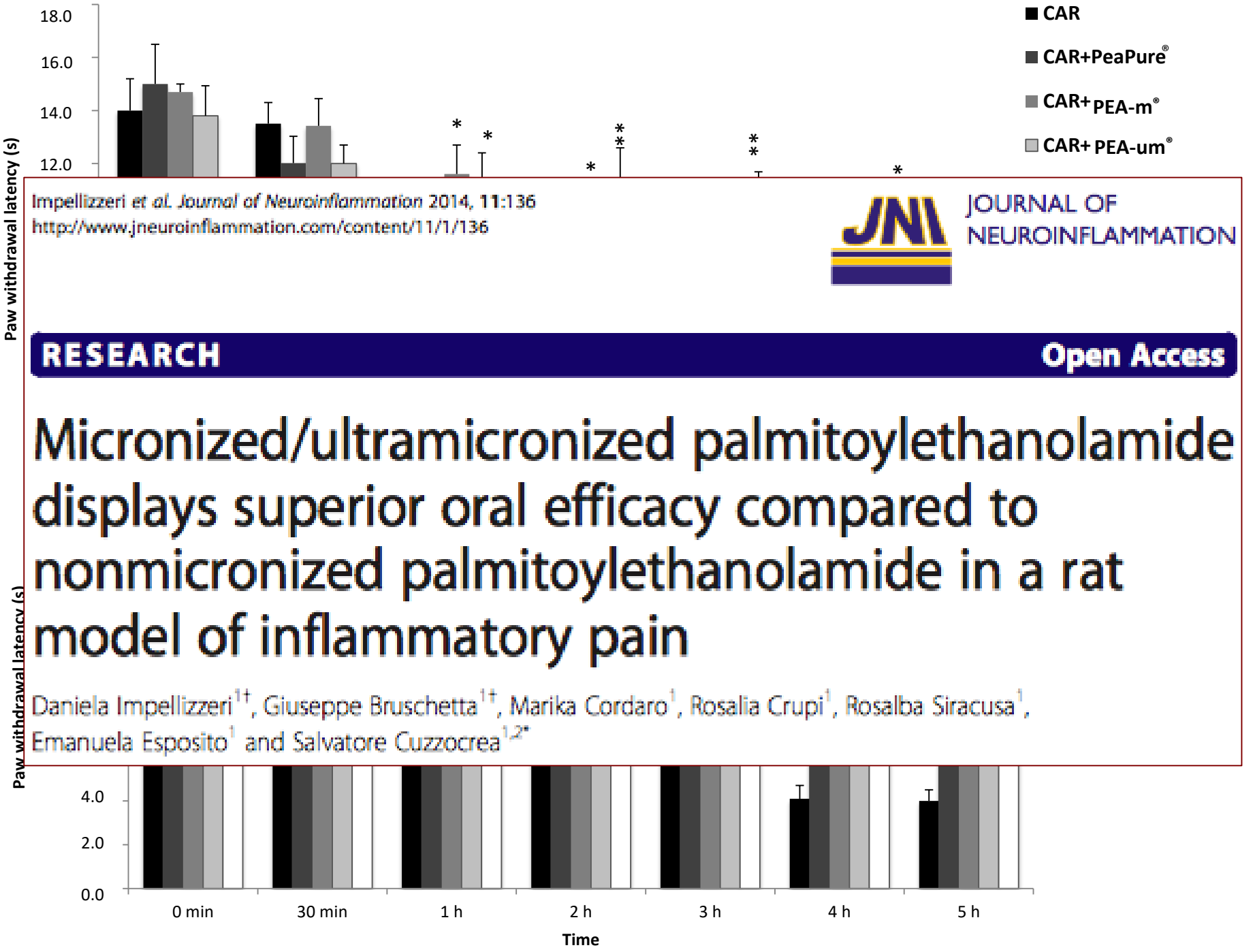

d1
100 µm


100 µm


Histological score
6
5
4
3
2
1
0
CAR
PeaPure®
PEA-m®
PEA-um®
**
*
*
*
ND
SHAM
CAR


MPO Activity U/g wet tissue
180
160
140
120
100
80
60
40
20
0
**
*
*
*
Sham
CAR
CAR+PeaPure®
CAR+PEA-m®
CAR+PEA-um®
Groups


CAR
CAR+PeaPure®
CAR+ PEA-m®
CAR+PEA-um®
Paw volume (ml)
1.2
1.0
0.8
0.6
0.4
0.2
0
0 min
30 min
1 h
2 h
3 h
4 h
5 h
6 h
Time
CAR
CAR+ PeaPure®
CAR + PEA-m®
CAR + PEA-um®
CAR+ PEA (Tocris)`
Paw Volume (ml)
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0
0 min
30 min
1 h
2 h
3 h
4 h
5 h
6 h
Time



Impellizzeri et al. Journal of Neuroinflammation 2014, 11:136
http://www.jneuroinflammation.com/content/11/1/136

JOURNAL OF NEUROINFLAMMATION

RESEARCH Open Access

Micronized/ultramicronized palmitoylethanolamide displays superior oral efficacy compared to nonmicronized palmitoylethanolamide in a rat model of inflammatory pain

Daniela Impellizzeri[1†], Giuseppe Bruschetta[1†], Marika Cordaro[1], Rosalia Crupi[1], Rosalba Siracusa[1], Emanuela Esposito[1] and Salvatore Cuzzocrea[1,2*]

ur country can
stead what you
nice can do for
what you can
take from a mi ice study"

Our Animal Model to Study PEALut

Vascular Dementia

Traumatic Brain Injury

Stroke

I° Hypothesis

Could co-ultraPEALut treatment reduces neuroinflammation associated with vascular dementia?

frontiers in Neurology

ORIGINAL RESEARCH
published: 06 June 2017
doi: 10.3389/fneur.2017.00233

Check for updates

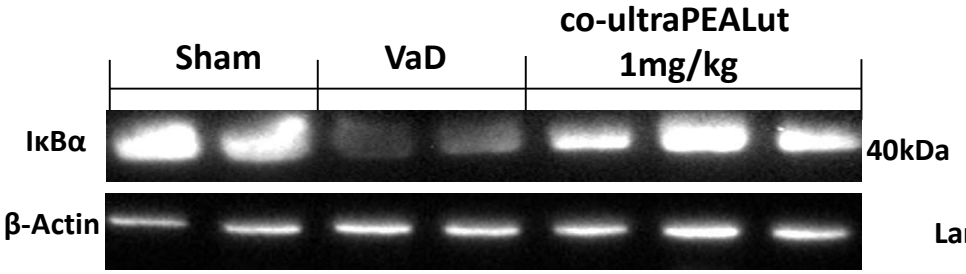
Anti-Inflammatory and Neuroprotective Effects of Co-UltraPEALut in a Mouse Model of Vascular Dementia

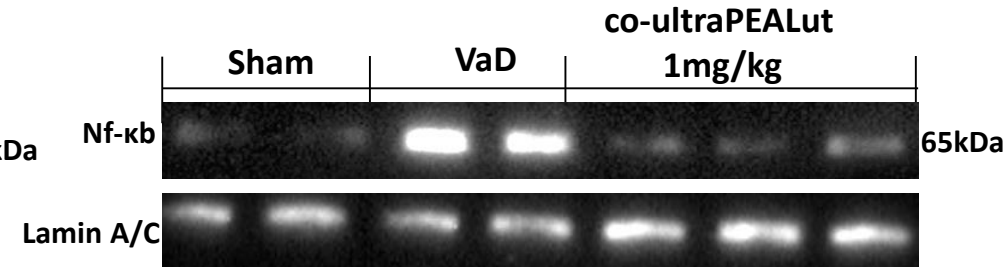
*Rosalba Siracusa[1], Daniela Impellizzeri[1], Marika Cordaro[1], Rosalia Crupi[1], Emanuela Esposito[1], Stefania Petrosino[2,3] and Salvatore Cuzzocrea[1,4]**

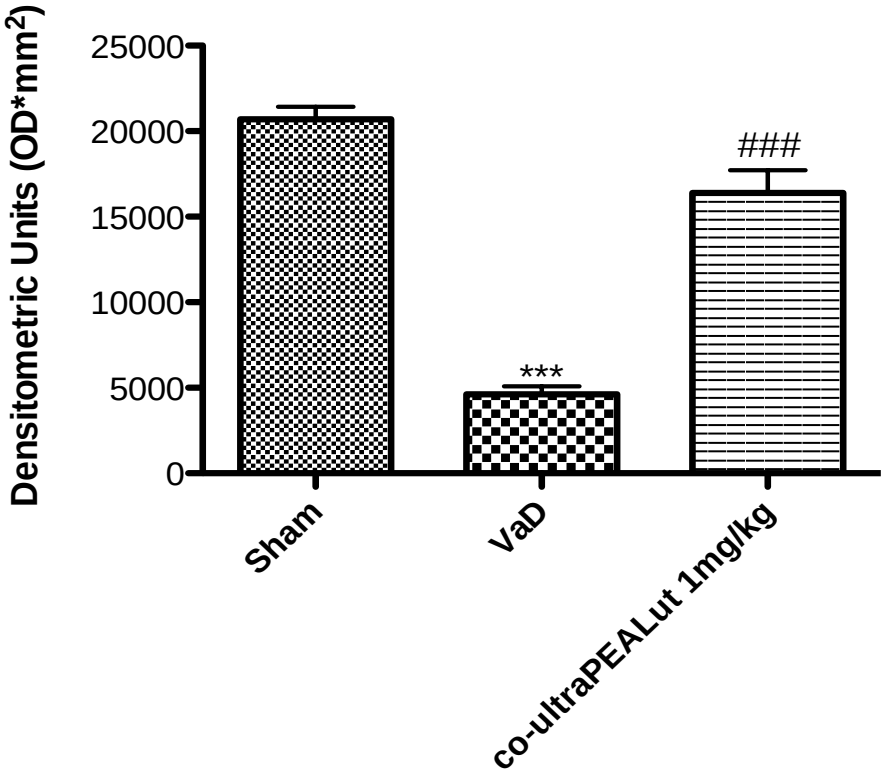
Experimental design

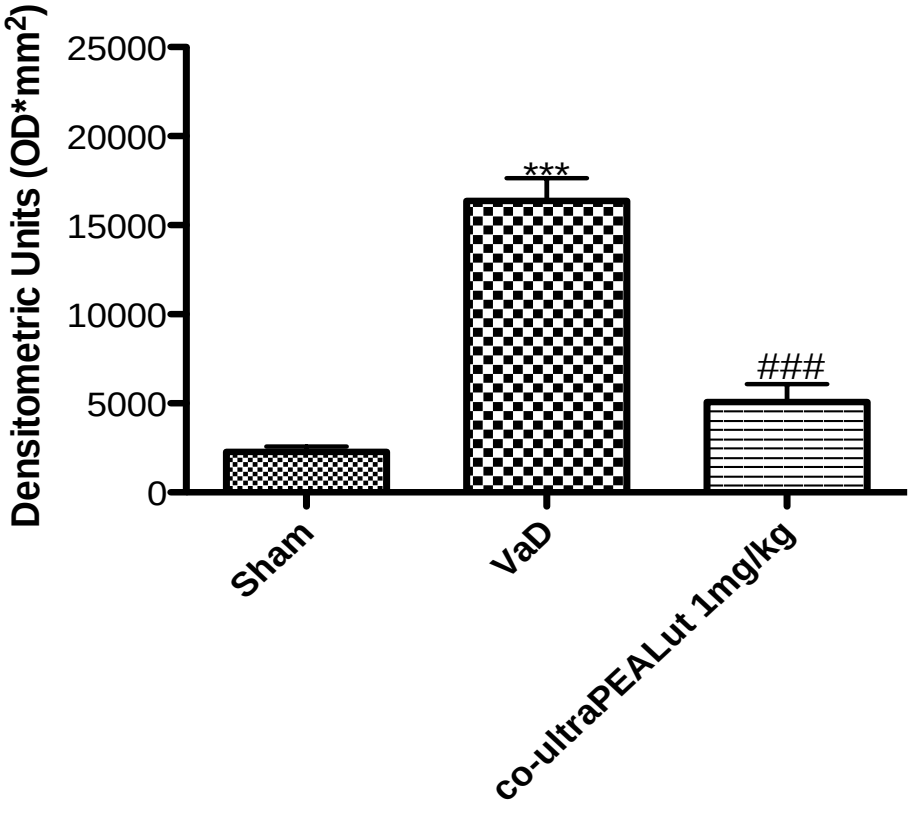
An anterior neck middle incision was made, and the bilateral carotid arteries were separated, and then a thread was passed below each carotid artery for closure. The bilateral carotid arteries were closed by ligation for 10 min, and then loosened for 10 min, and this was repeated 3 times

Inflammatory pathway

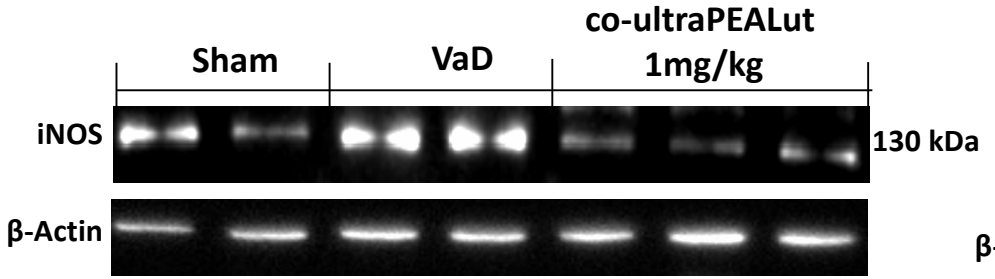


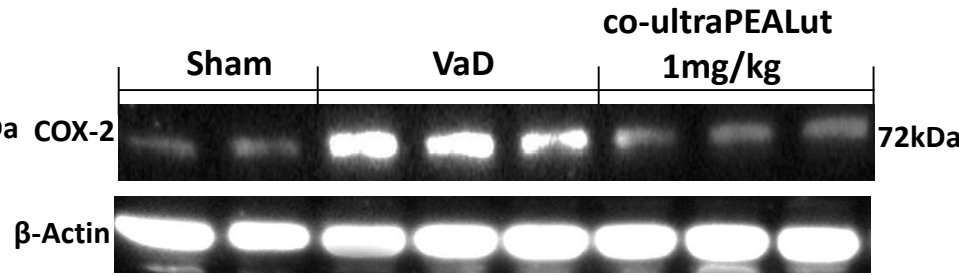


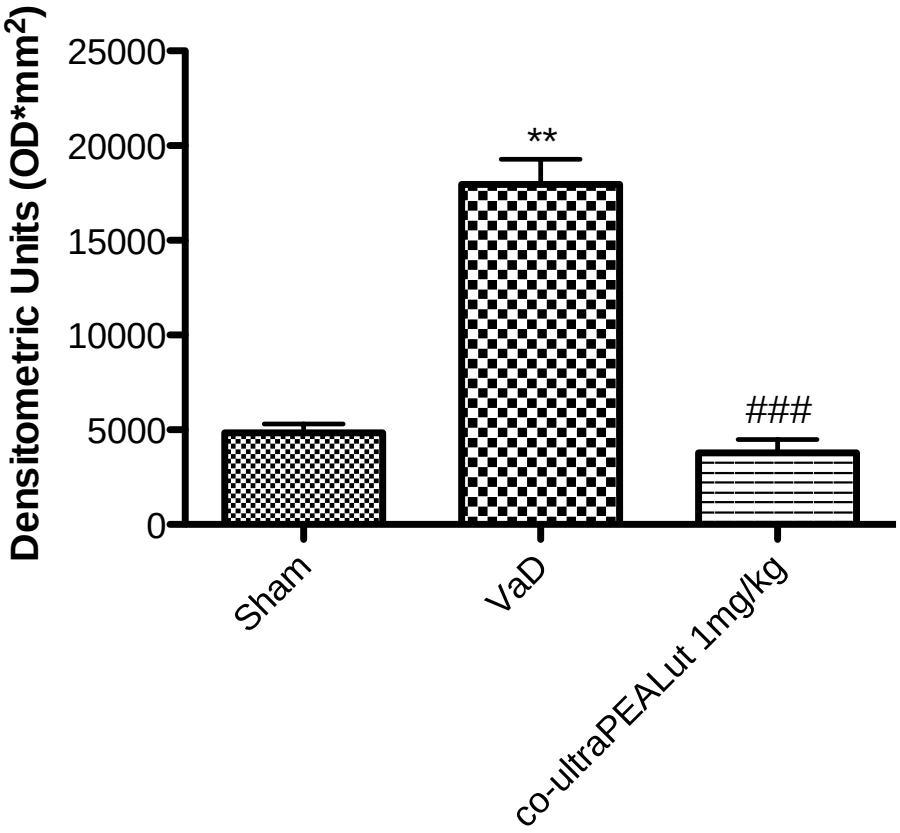


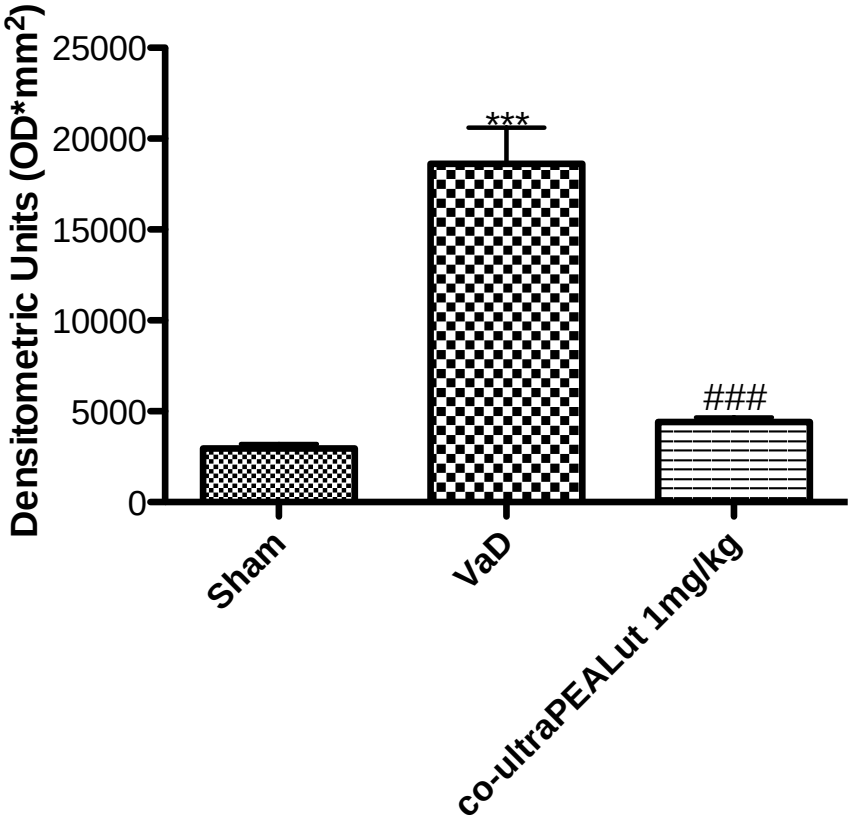


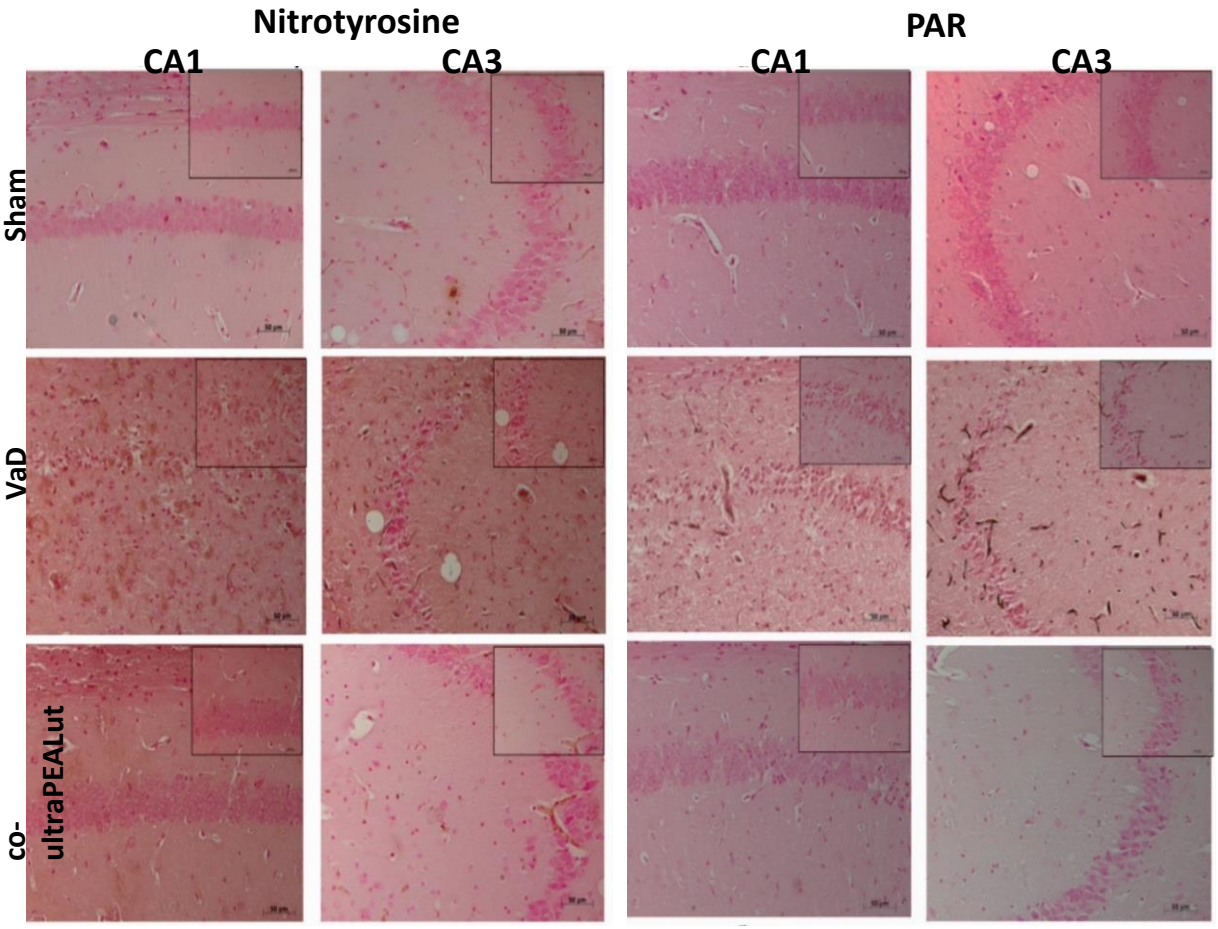
Inflammatory pathway



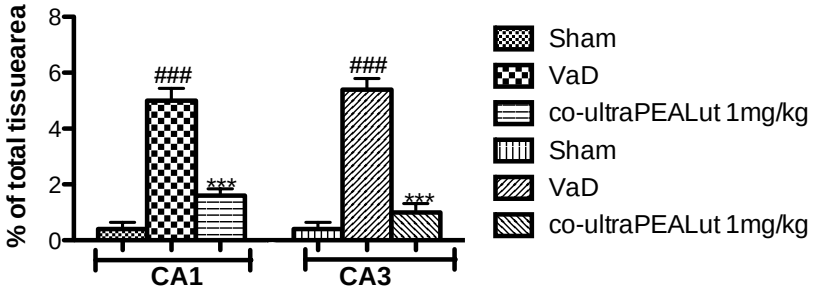


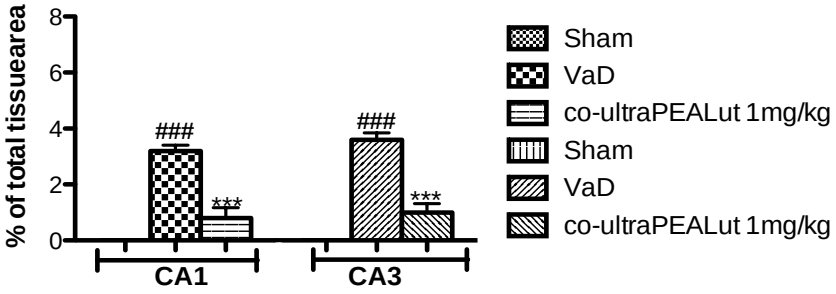


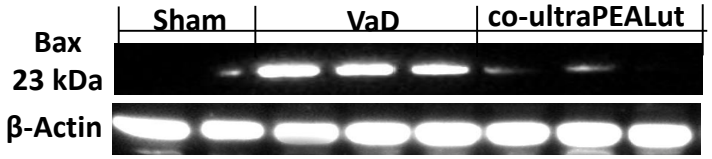


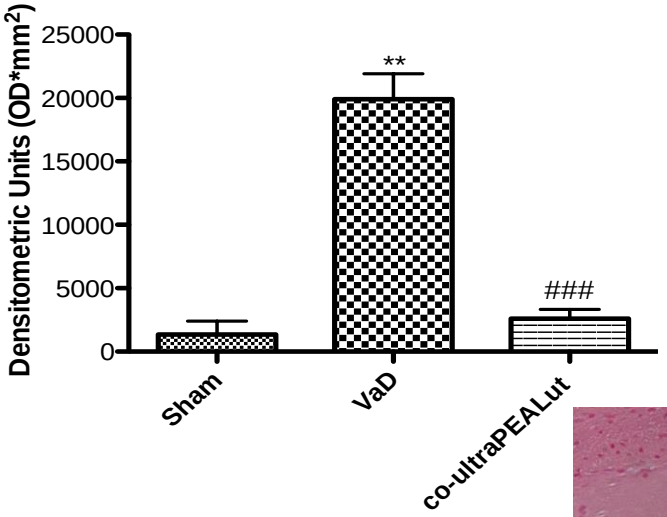


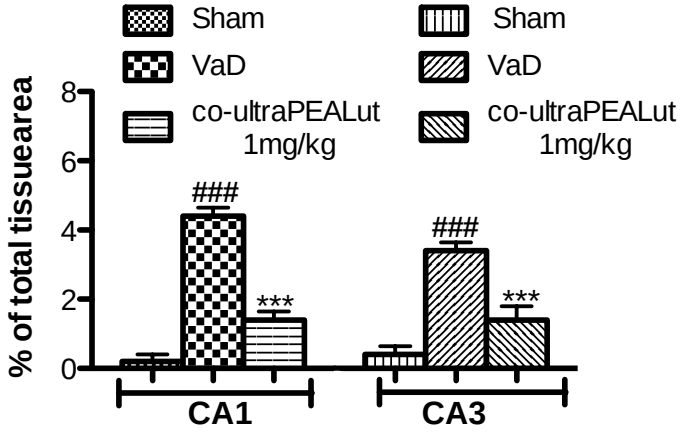
Oxidative stress



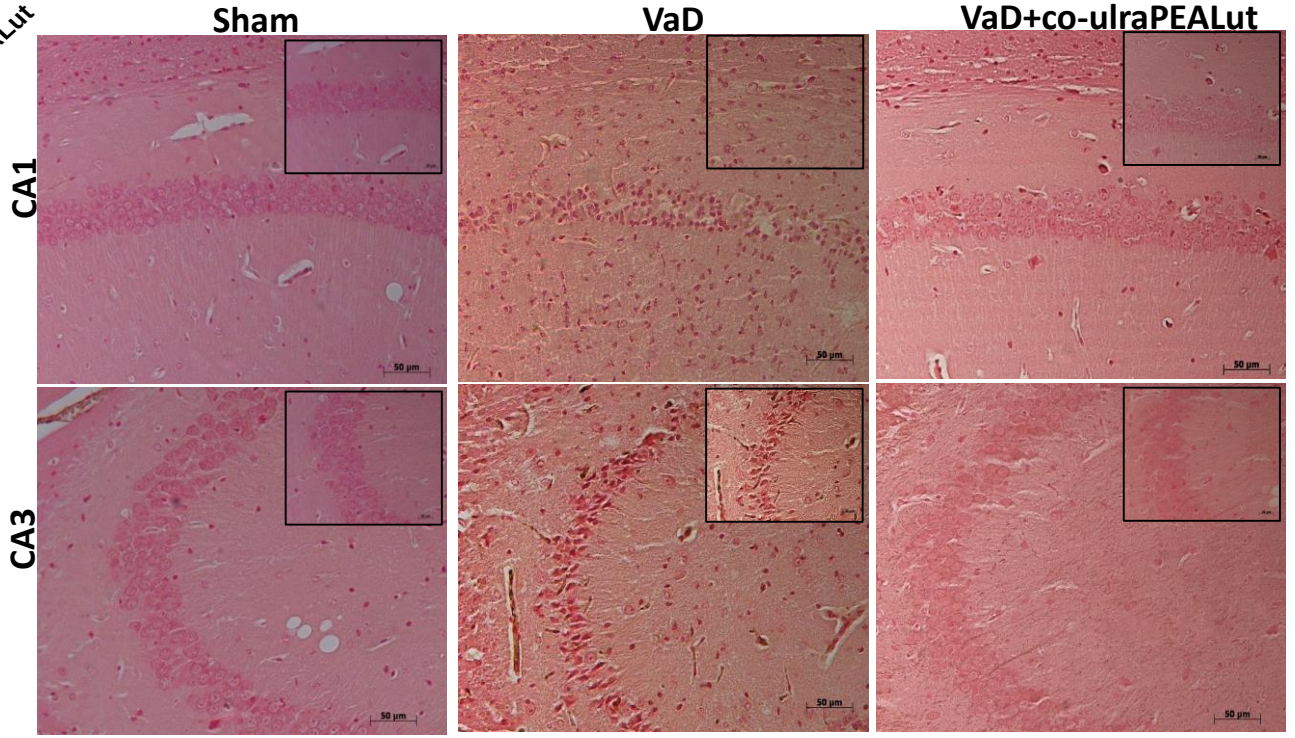


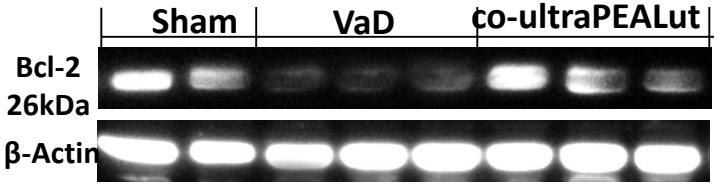


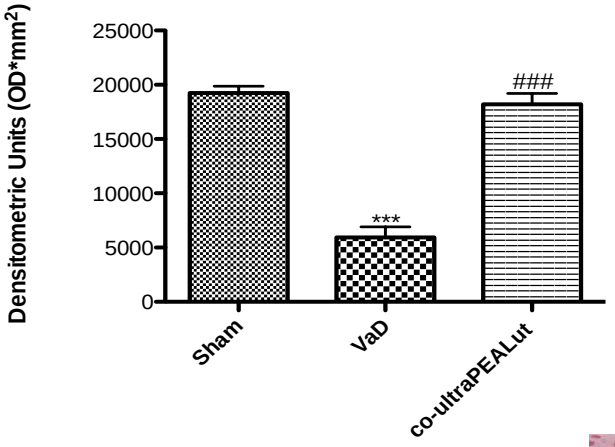


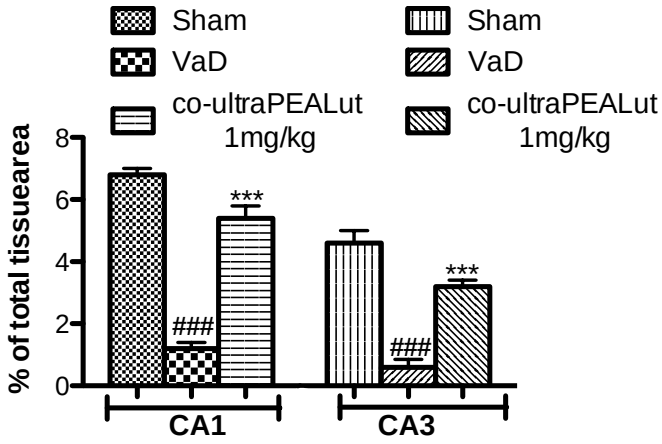


Apoptotic studies

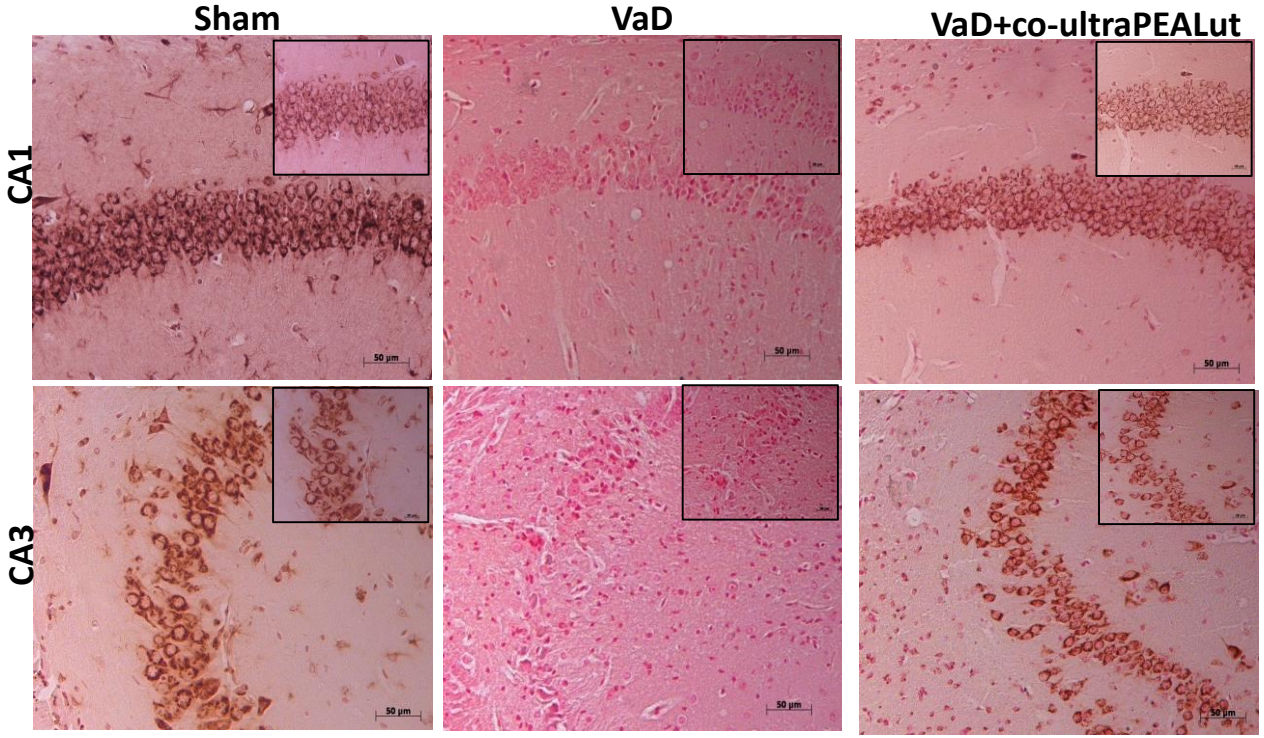




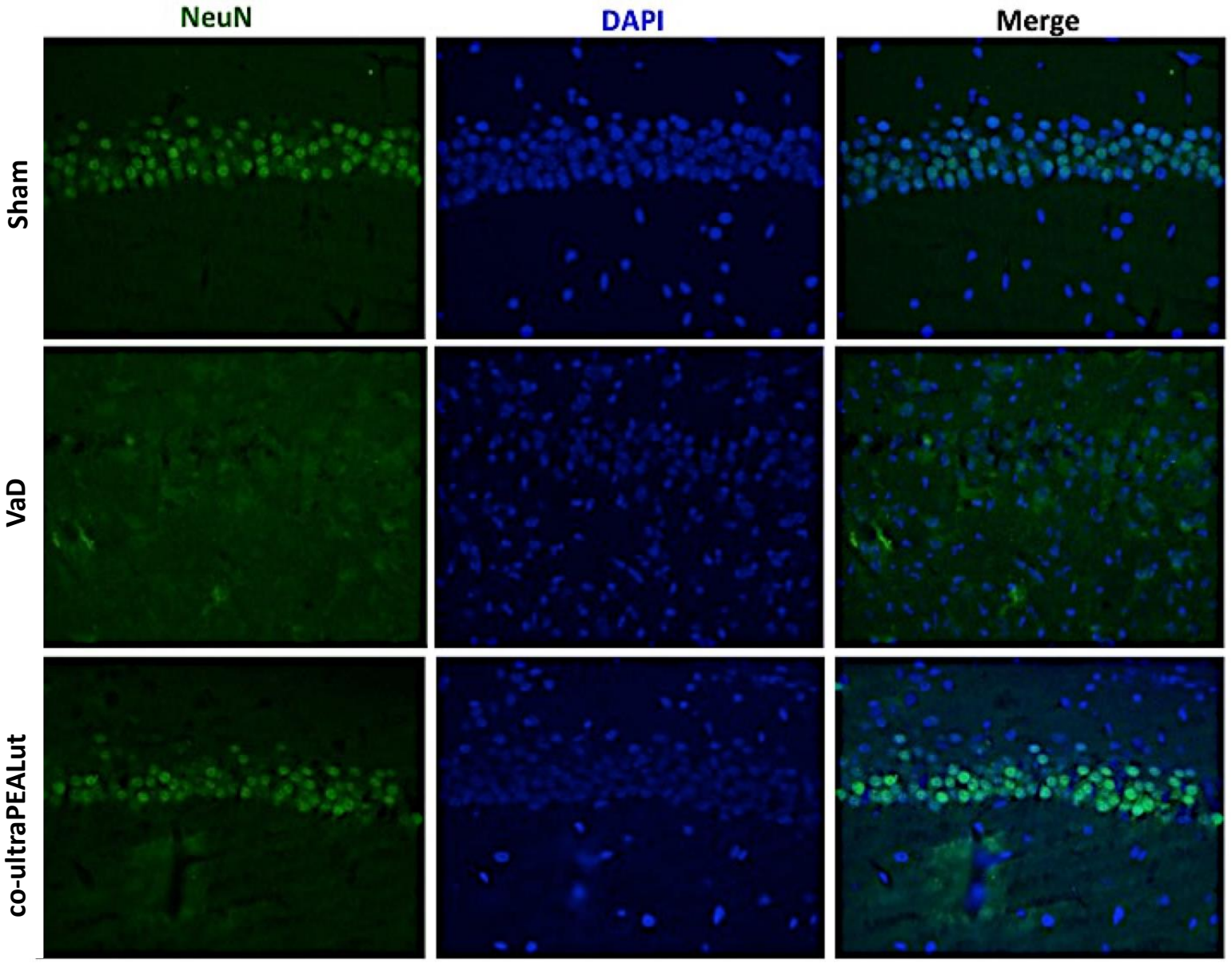




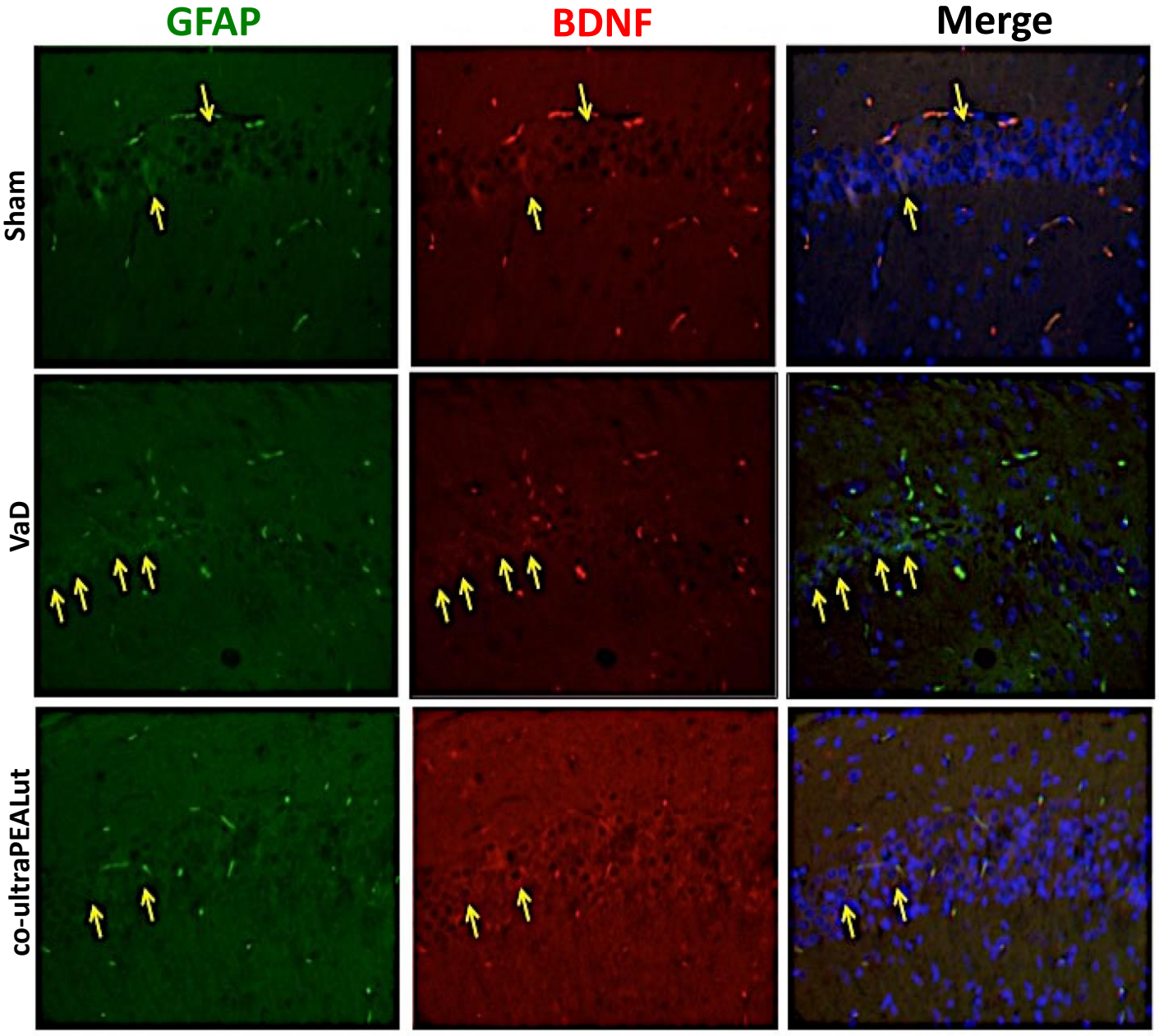
Apoptotic studies



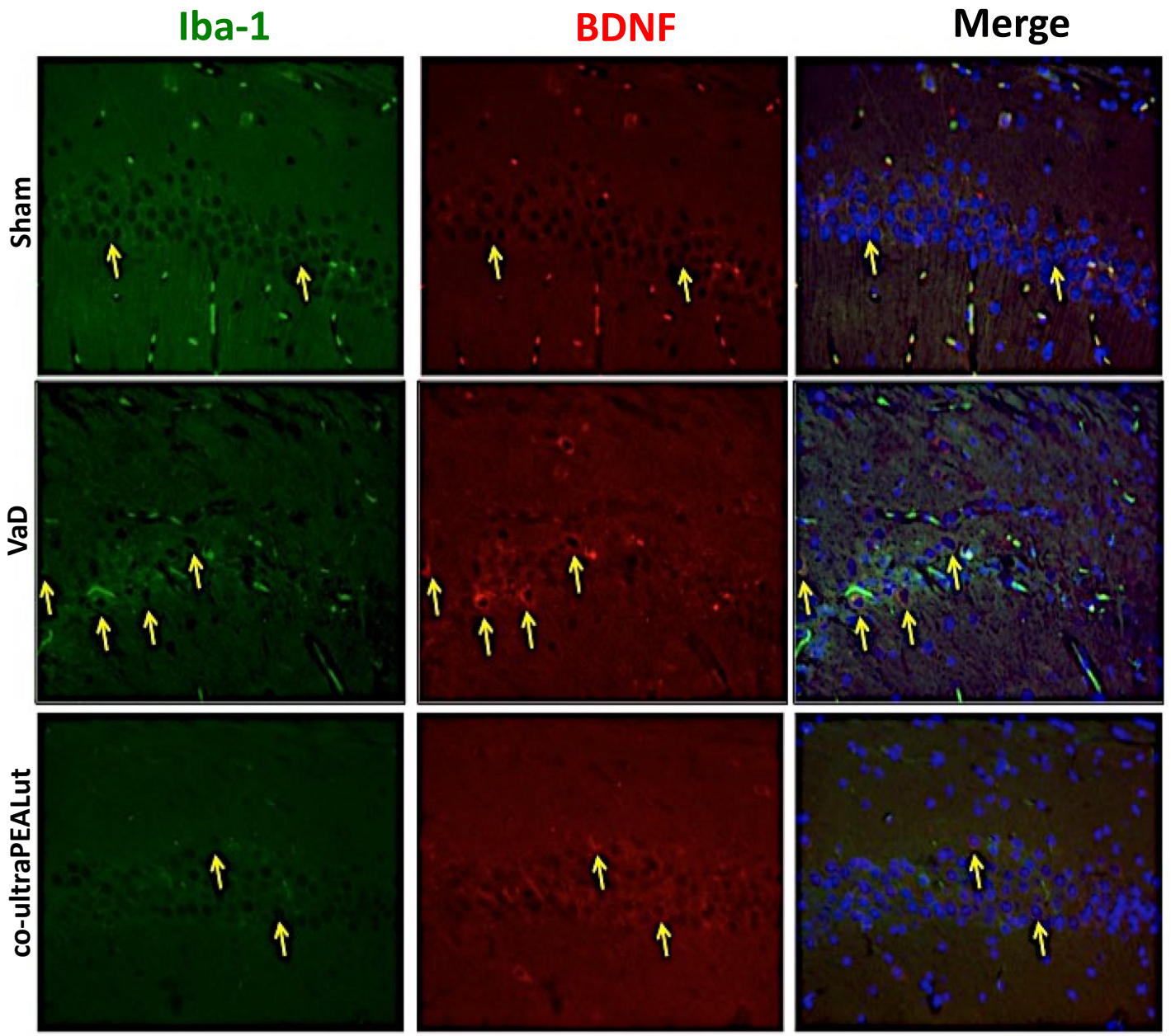
Effects of co-ultraPEALut on neuronal cells after carotid arteries occlusion



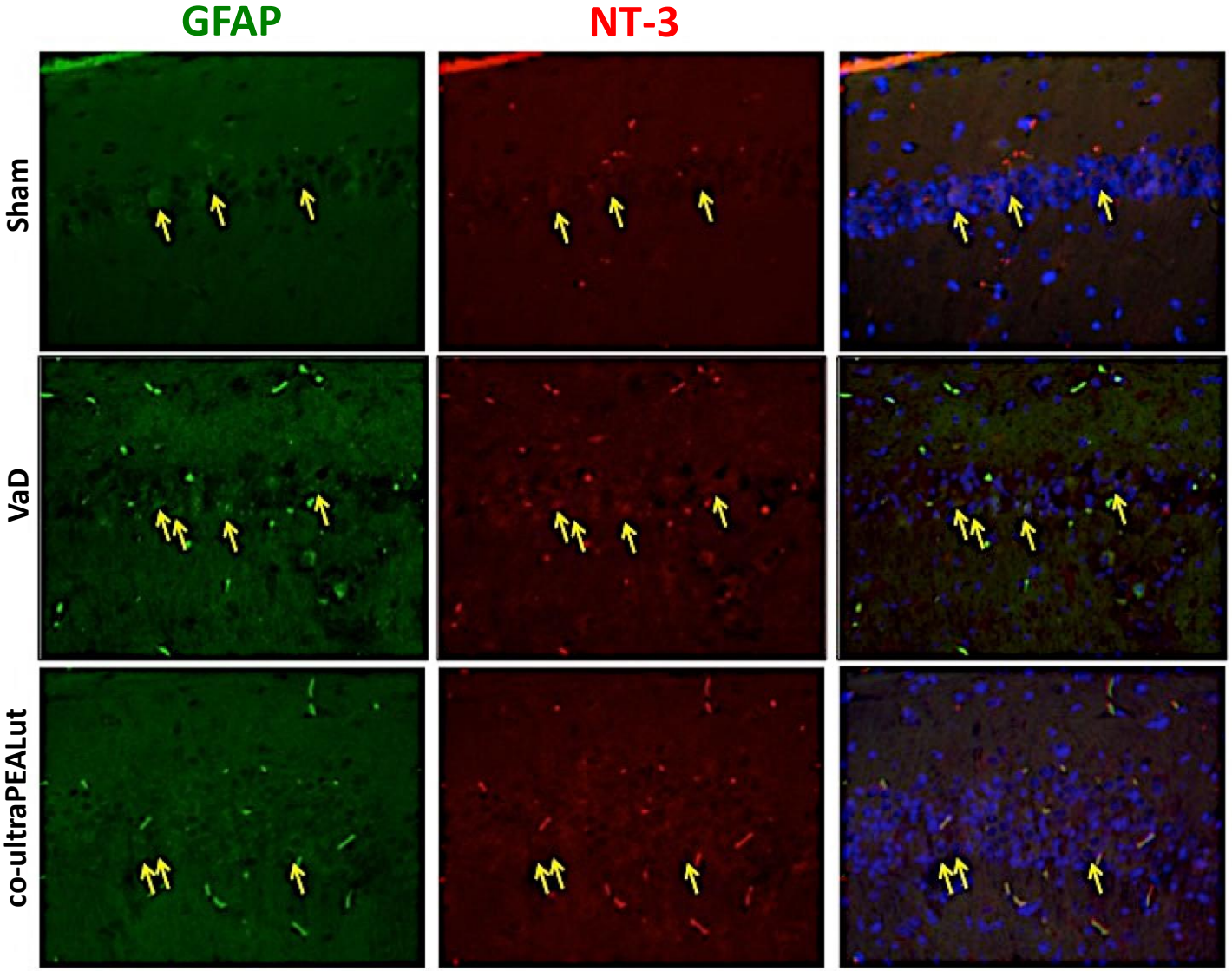
Co-localization of GFAP/brain-derived neurotrophic factor (BDNF) after carotid arteries occlusion



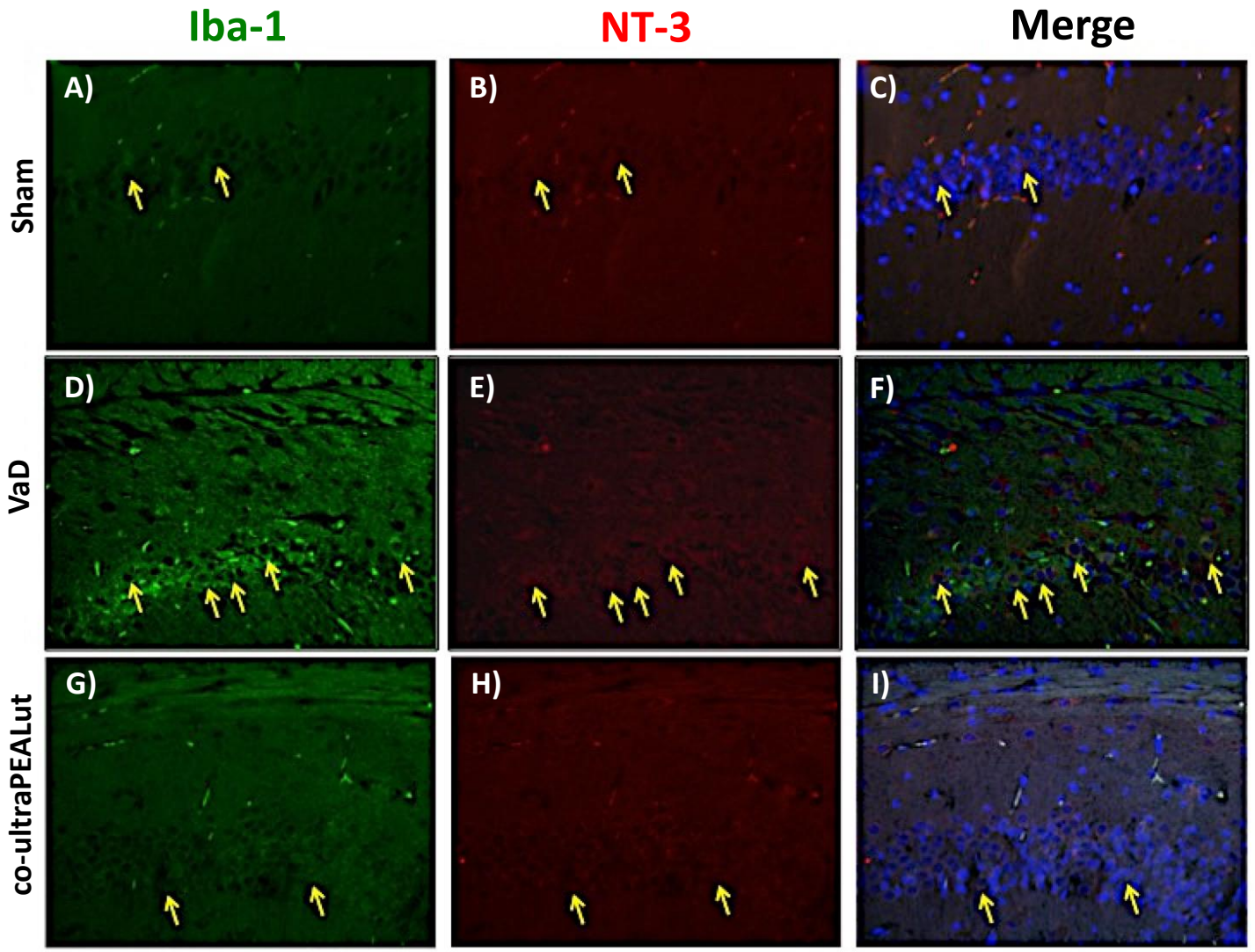
Co-localization of Iba-1/brain-derived neurotrophic factor (BDNF) after carotid arteries occlusion

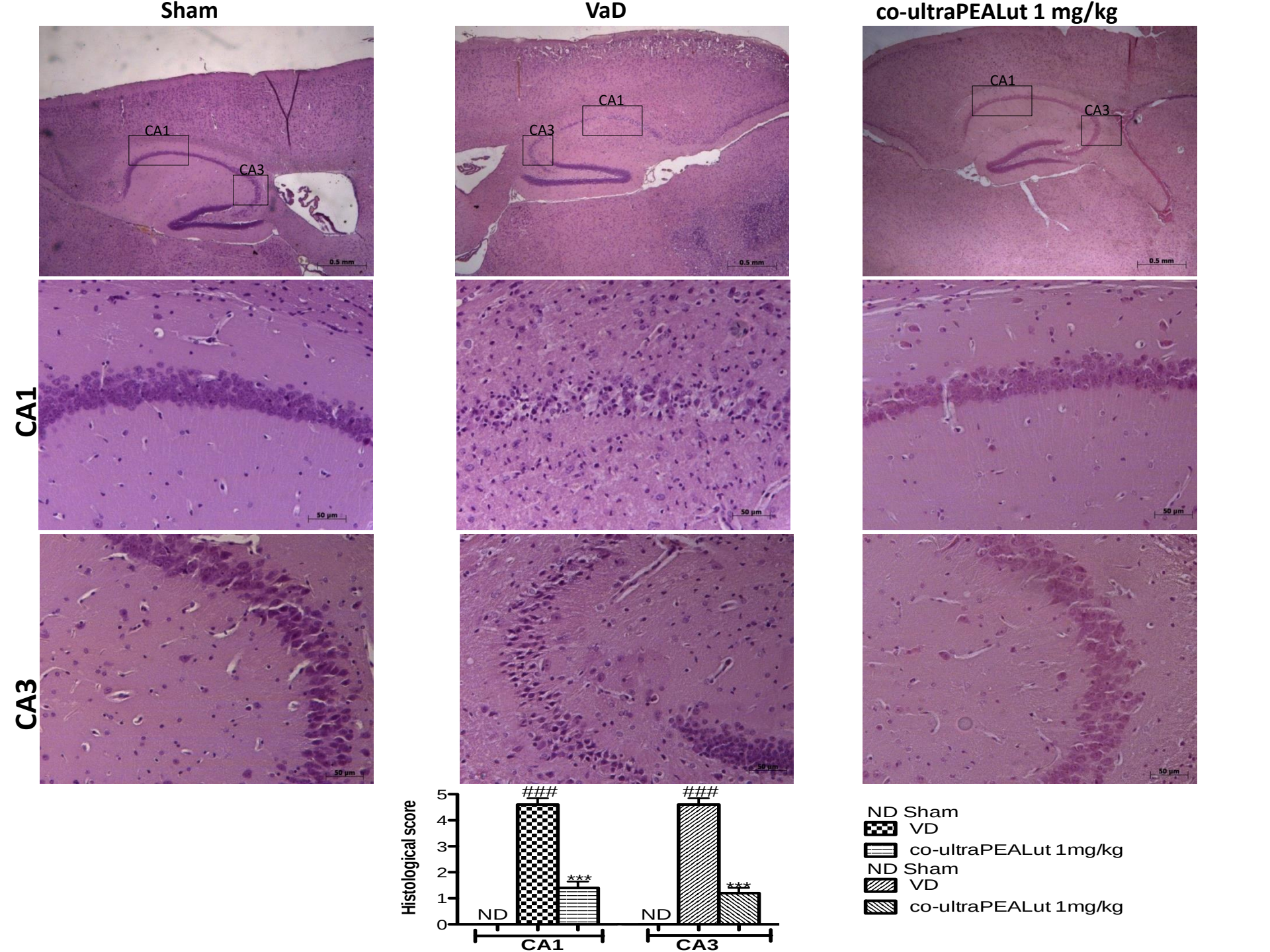


Co-localization of GFAP/NT-3 after carotid arteries ligation



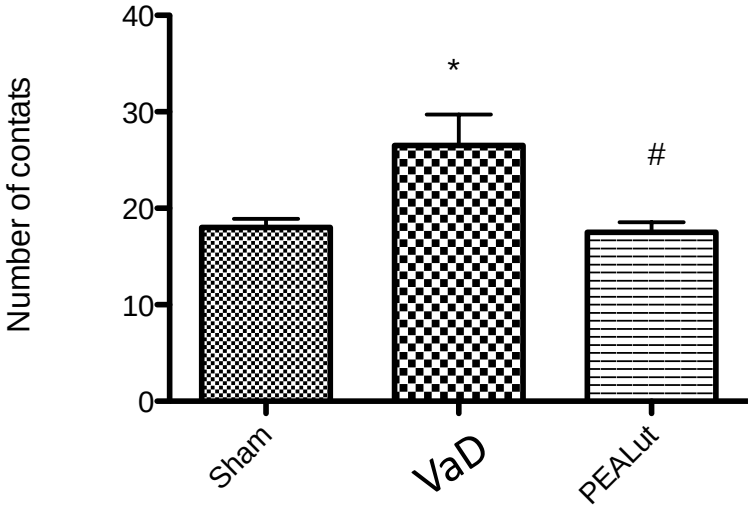
Co-localization of Iba-1/NT-3 after carotid arteries ligation

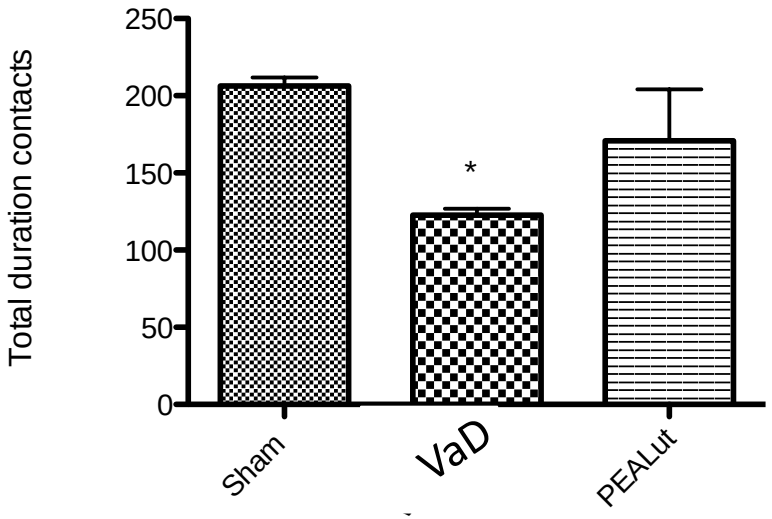



Sham
VaD
co-ultraPEALut 1 mg/kg
CA1
CA3
CA1
CA3
0.5 mm
50 µm
Histological score
###
***
ND
CA1
CA3
ND Sham
VD
co-ultraPEALut 1mg/kg

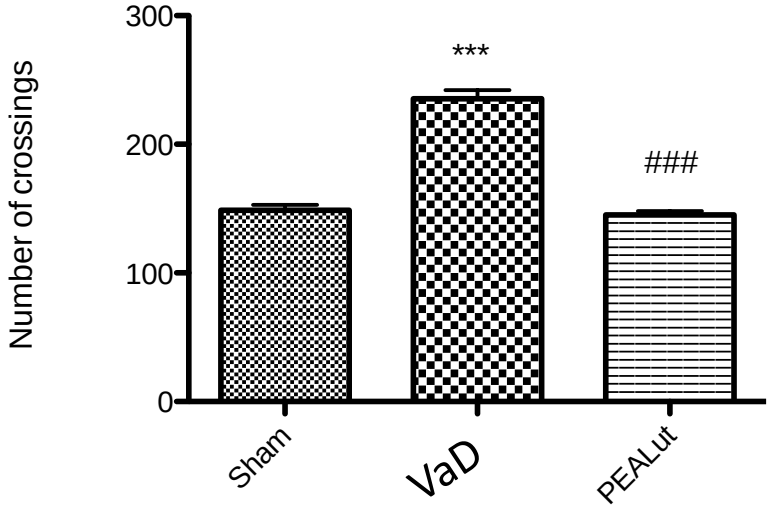
Behavioral testing

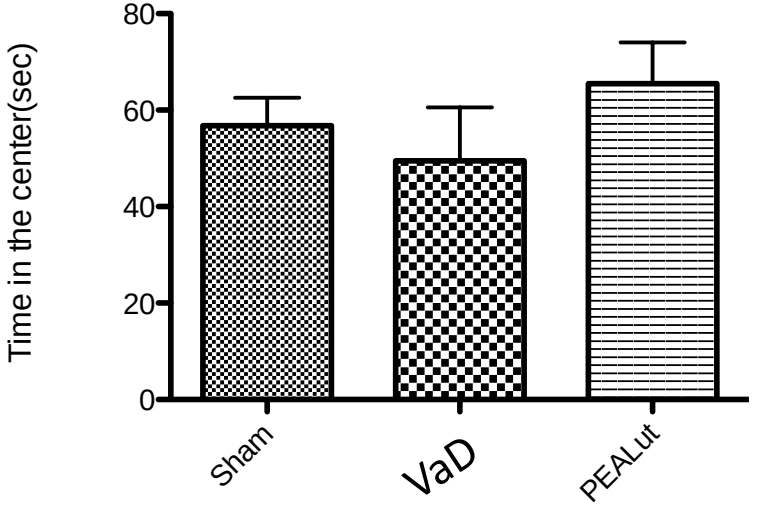
Reciprocal Social interactions



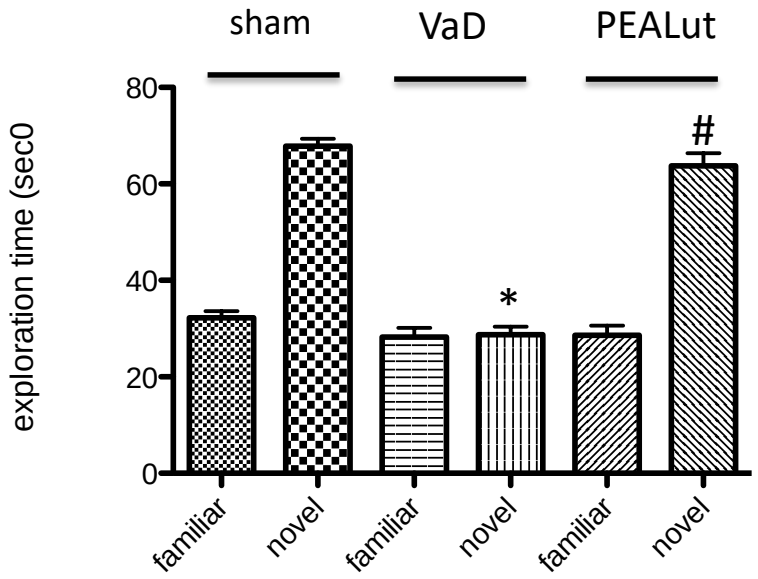


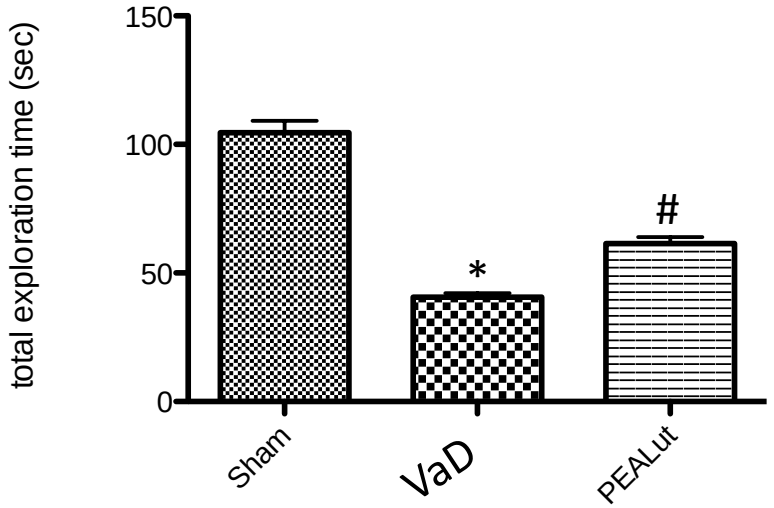
Open Field test





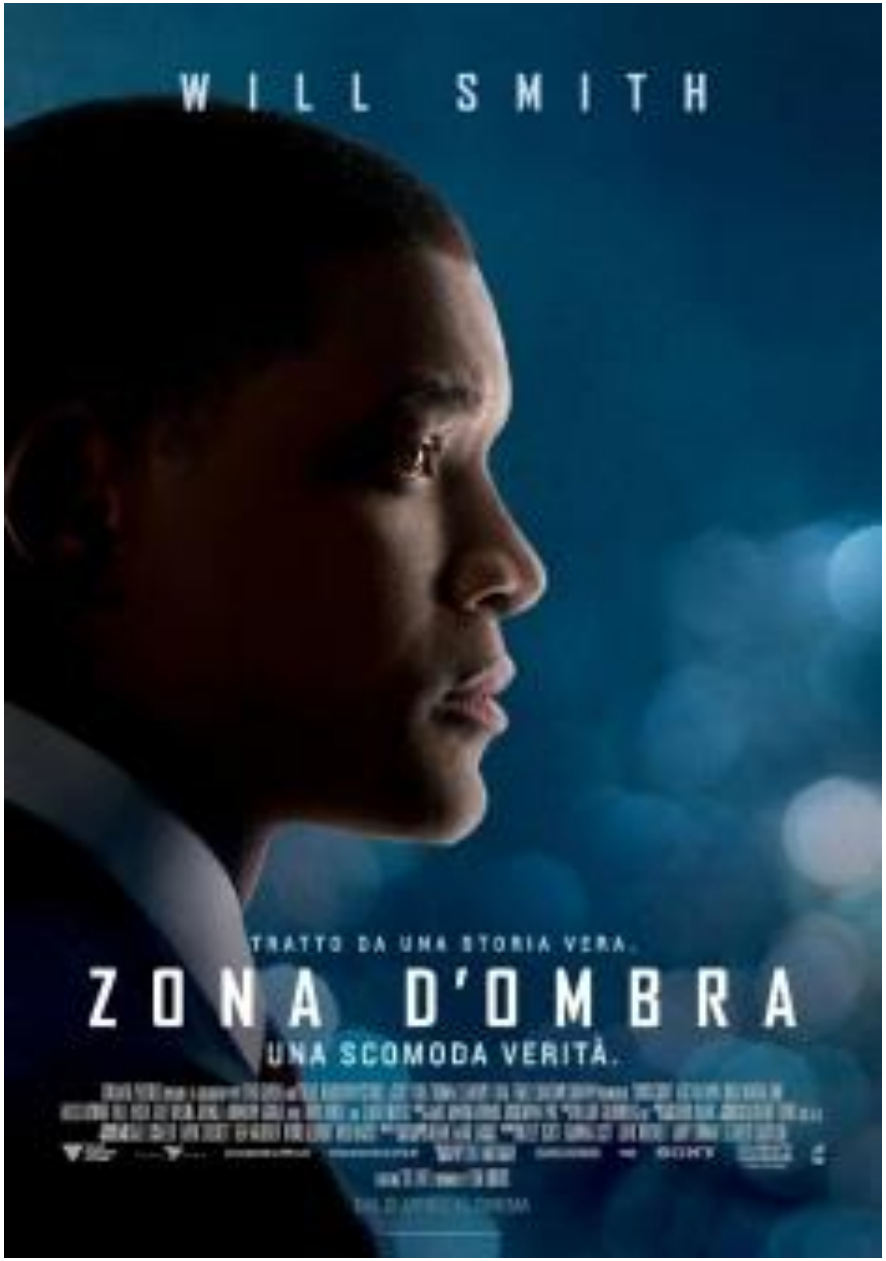
Novel objection recognition test





II° Hypothesis

Could co-ultraPEALut treatment reduces neuroinflammation associated with traumatic brain injury?


WILL SMITH
TRATTO DA UNA STORIA VERA.
ZONA D'OMBRA
UNA SCOMODA VERITÀ.

WILL SMITH
BASED ON A TRUE STORY
CONCUSSION
EVEN LEGENDS NEED A HERO
CHRISTMAS

The NEW ENGLAND JOURNAL of MEDICINE

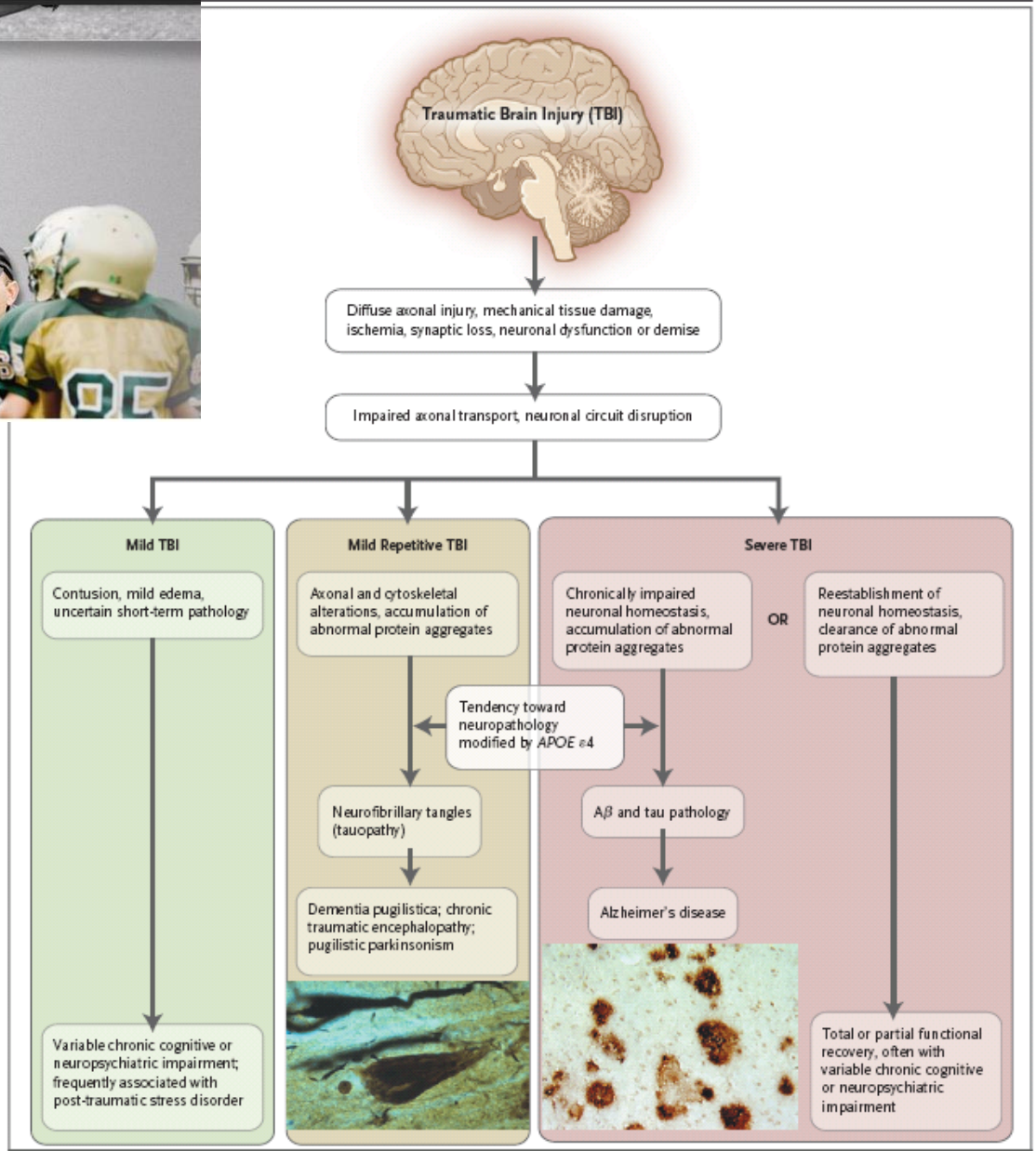
Perspective

SEPTEMBER 30, 2010

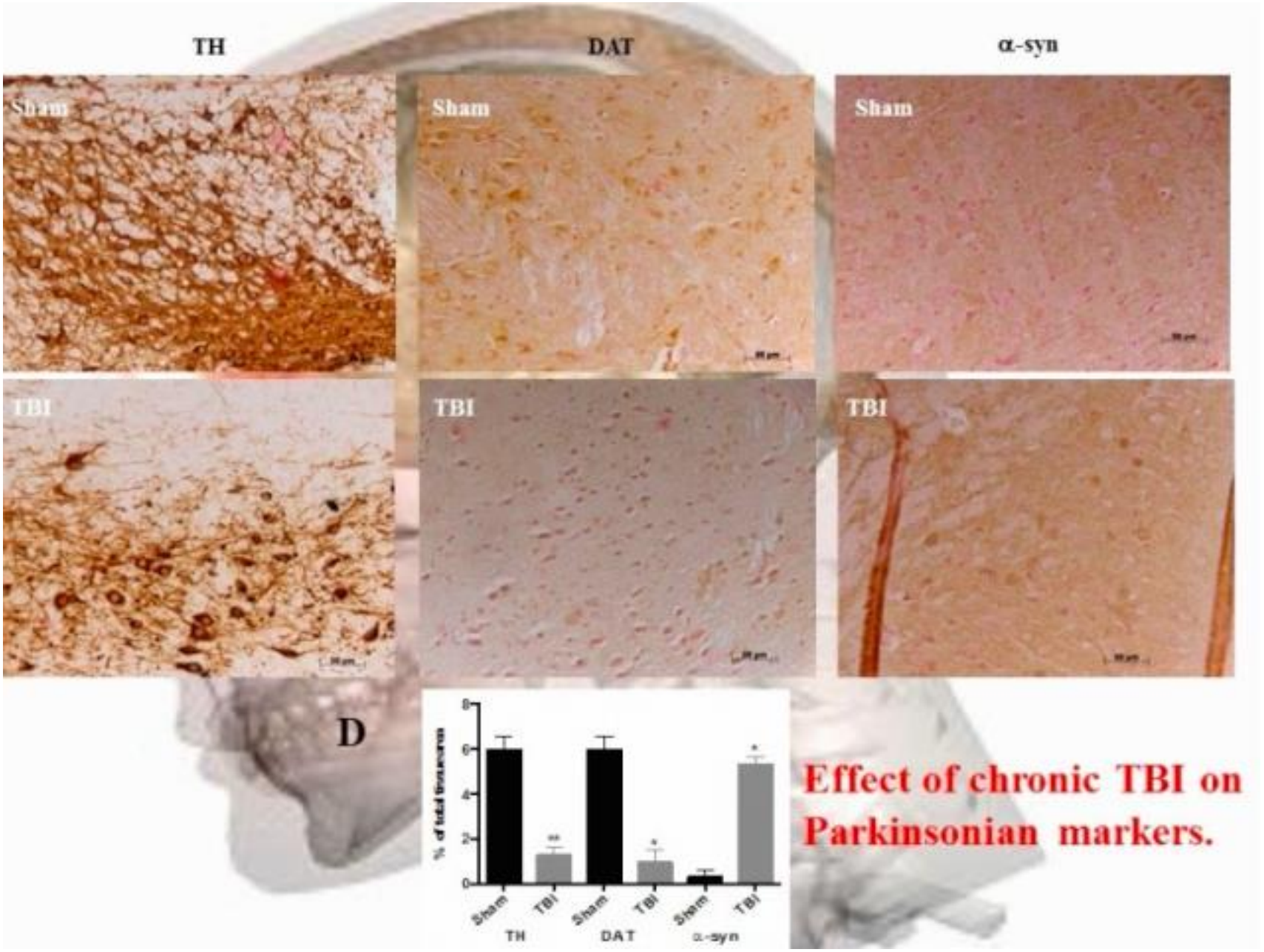
Traumatic Brain Injury — Football, Warfare, and Long-Term Effects

Steven T. DeKosky, M.D., Milos D. Ikonomovic, M.D., and Sam Gandy, M.D., Ph.D.

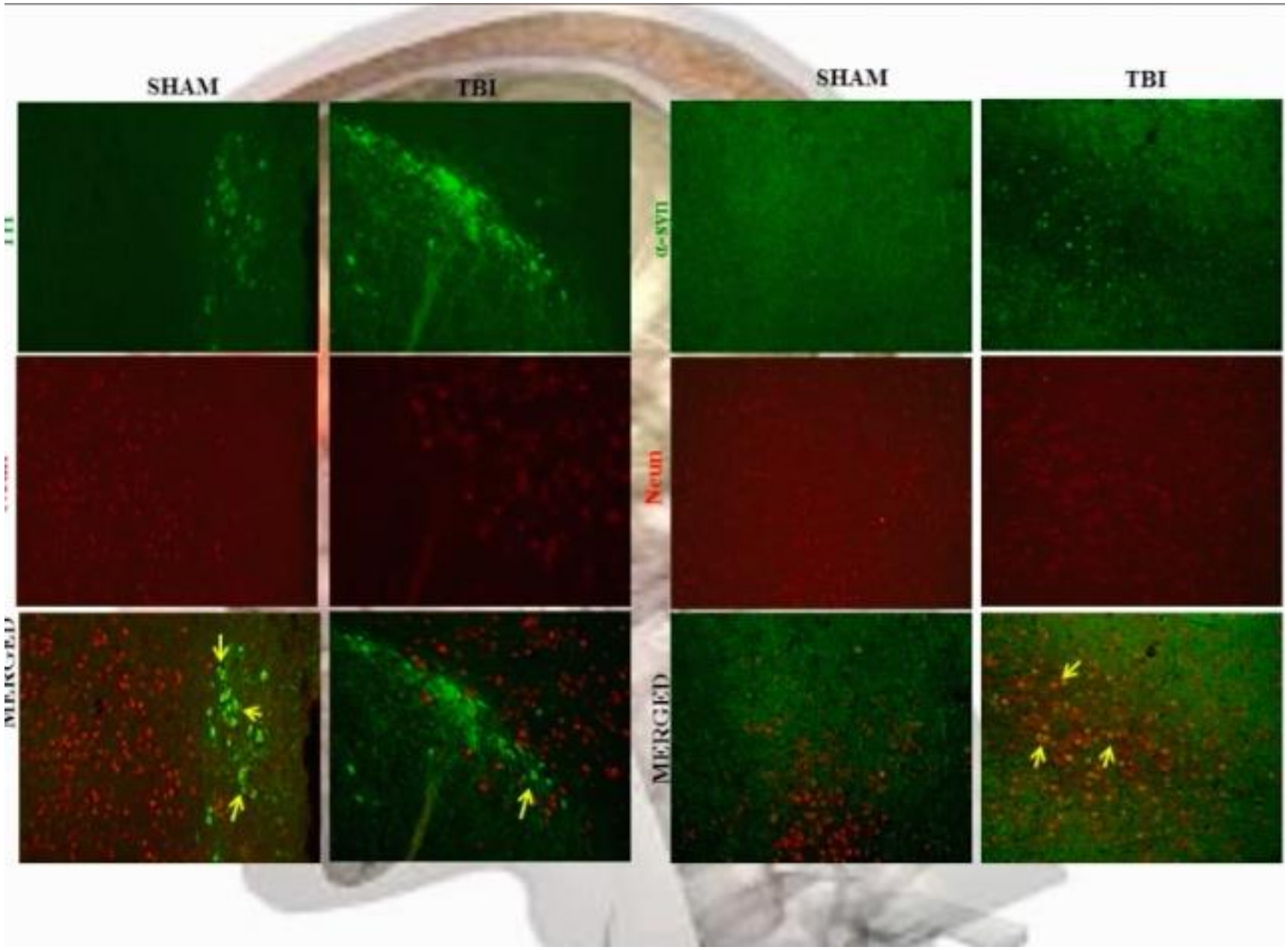
In late July, the National Football League introduced a new poster to be hung in league locker rooms, warning players of possible long-term health effects of concussions. Public awareness of the

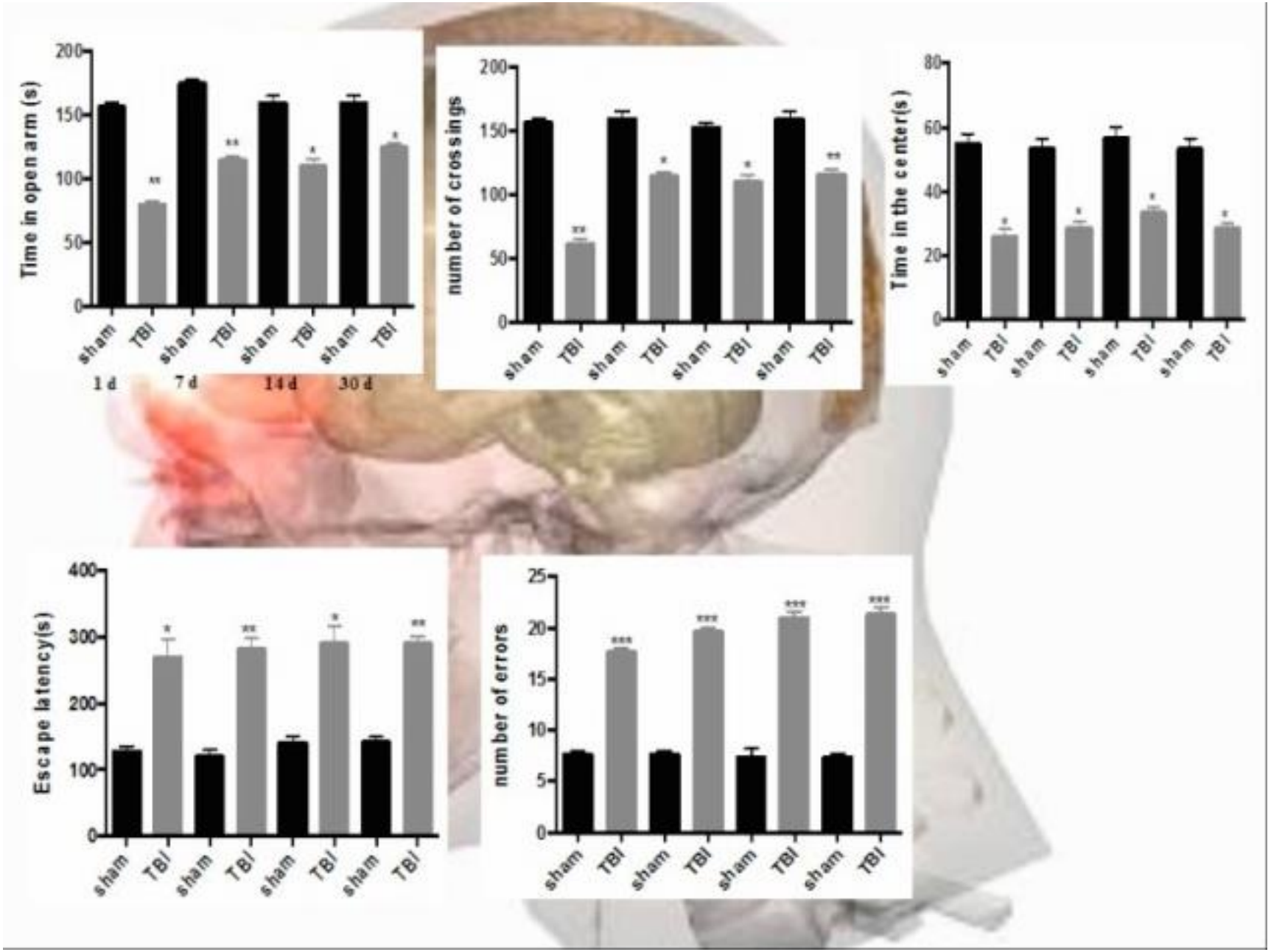


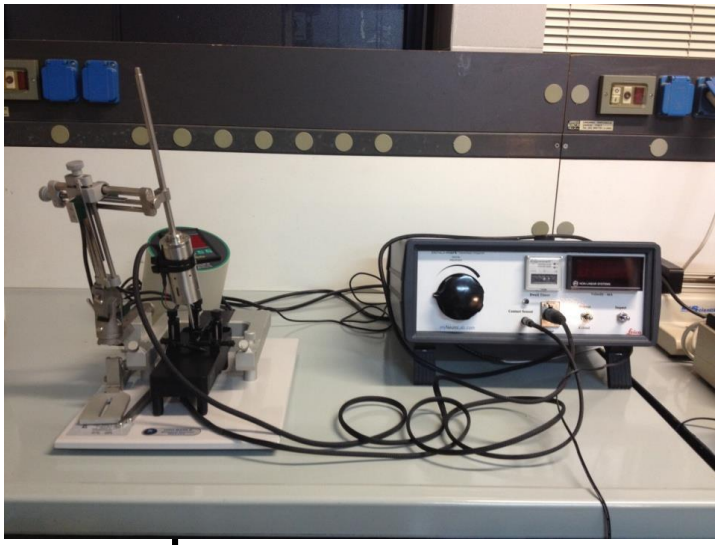
Spectrum of Pathologic Features and Outcomes of Traumatic Brain Injury (TBI).
In the left inset, Bielschowsky silver stain shows intraneuronal and extracellular neurofibrillary tangles in temporal cortex from a retired boxer with dementia pugilistica.[1] The right inset shows diffuse Aβ plaque deposits in temporal cortex from a subject who sustained severe TBI.[2]



Effect of chronic TBI on Parkinsonian markers.


SHAM
TBI
SHAM
TBI
α-SYN
Neun
MERGED
MERGED


Time in open arm (s)
200
150
100
50
0
sham
TBI
1 d
7 d
14 d
30 d
number of crossings
Time in the center(s)
80
60
40
20
Escape latency(s)
400
300
number of errors
25
20
15
10
5



Controlled Cortical Impact Model

PEA-LUT
(1 mg/kg os)

TBI

EPM

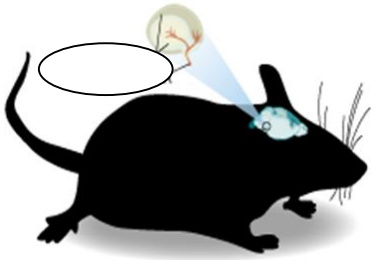
EPM

EPM

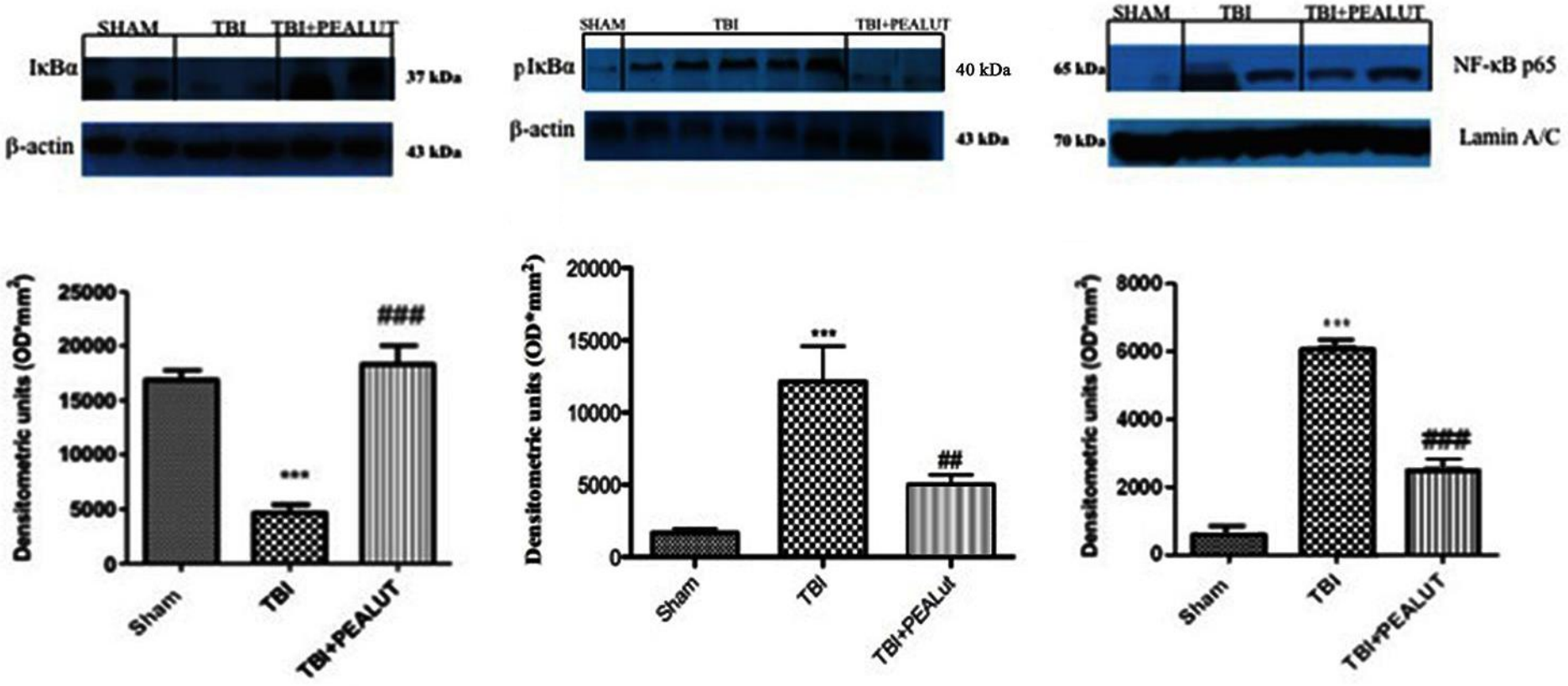
EPM

EPM
sacrifice

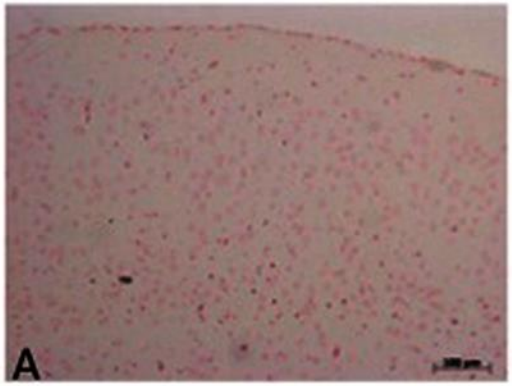
EPM

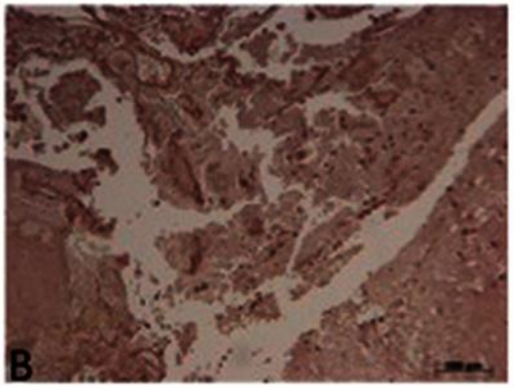


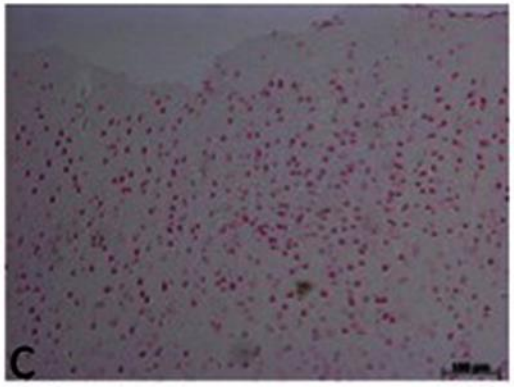
- WB
- Immunostaining
- Motor function

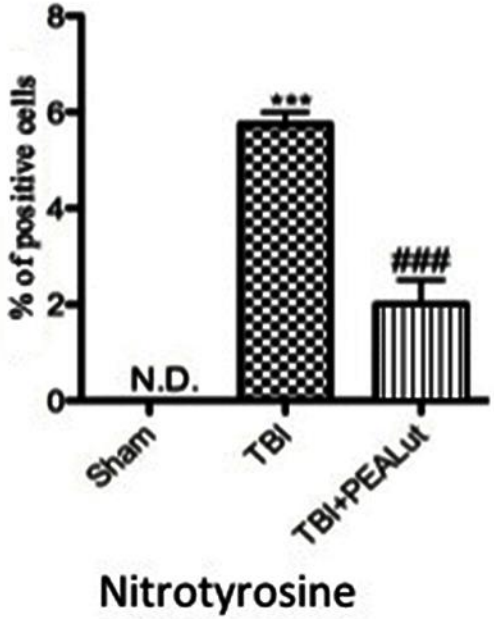


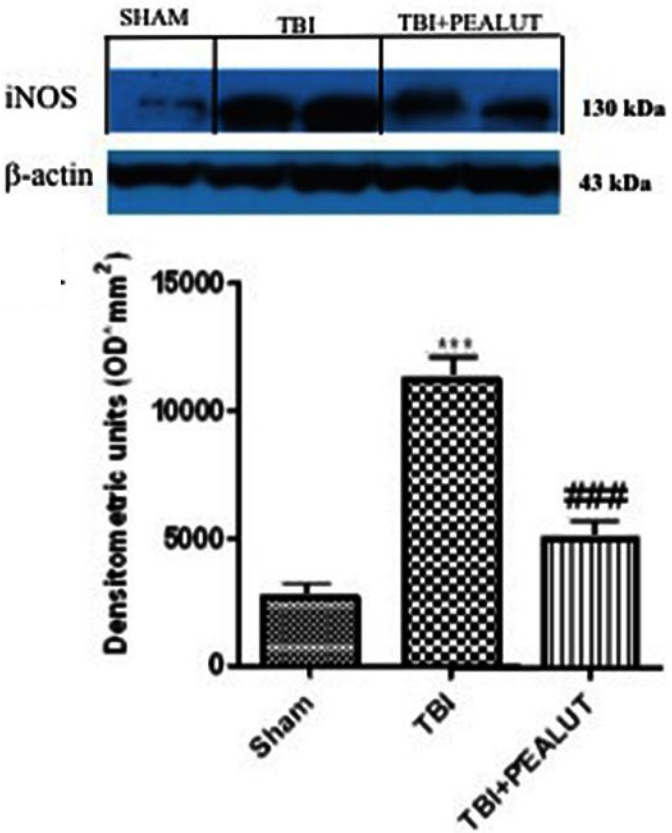
Co-ultraPEALut modulates IκB-a degradation and NFkBp65 nuclear translocation

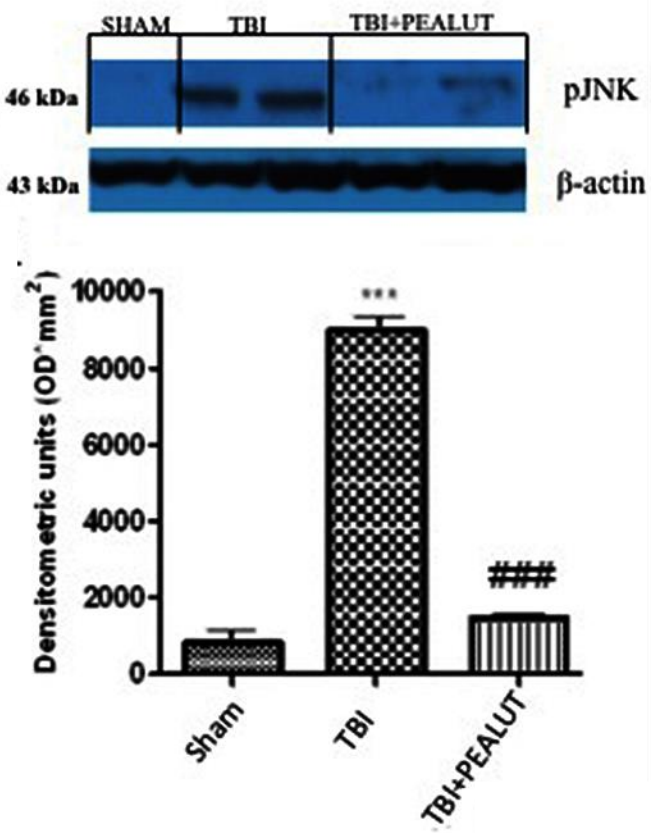

A

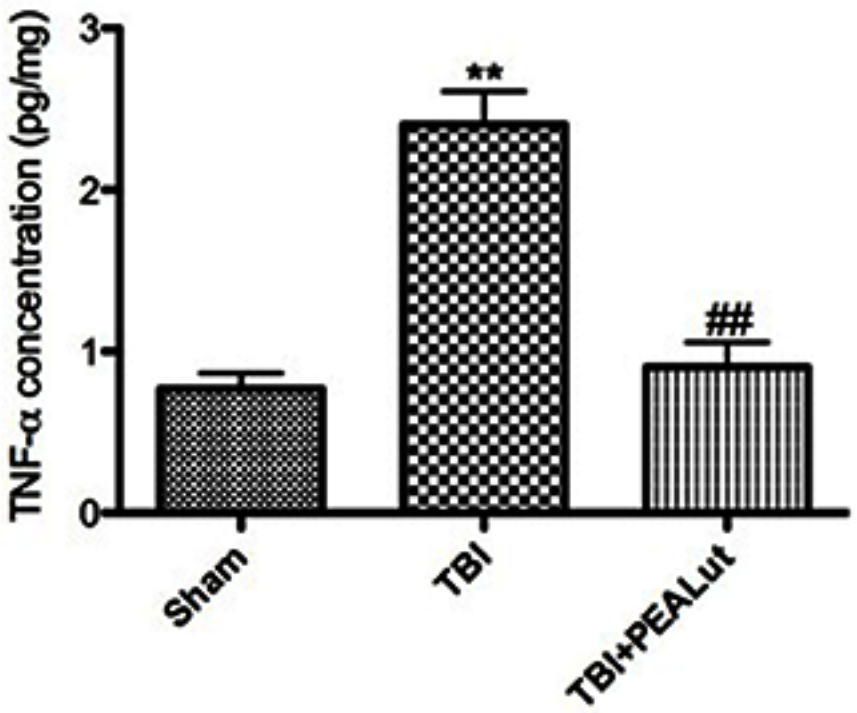

B

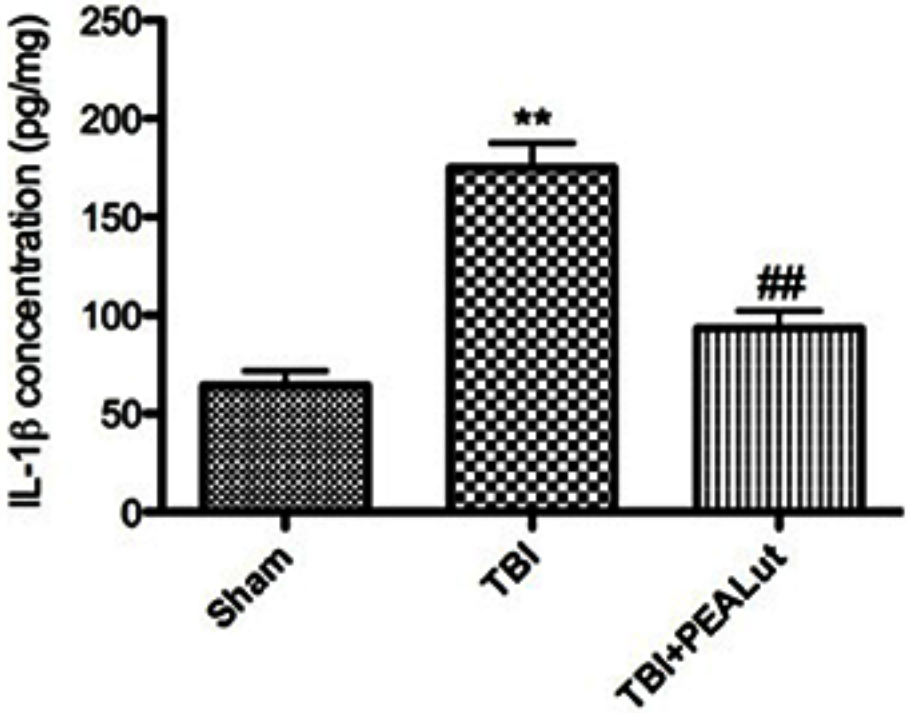

C


% of positive cells
8
6
4
2
0
N.D.
***
####
Sham
TBI
TBI+PEALut
Nitrotyrosine

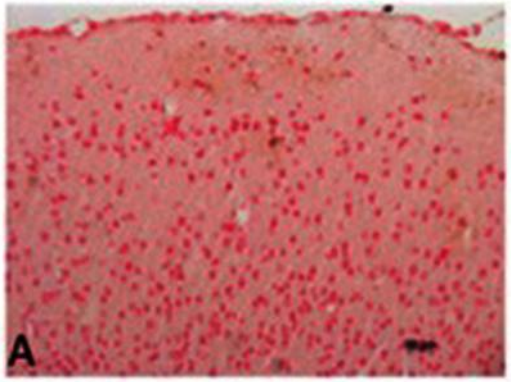

SHAM
TBI
TBI+PEALUT
iNOS
130 kDa
β-actin
43 kDa
Densitometric units (OD*mm2)
15000
10000
5000
0
***
###
Sham
TBI
TBI+PEALUT


SHAM
TBI
TBI+PEALUT
46 kDa
pJNK
43 kDa
β-actin
Densitometric units (OD*mm2)
10000
8000
6000
4000
2000
0
***
###
Sham
TBI
TBI+PEALUT





Co-ultra PEALut administration reduces TNFalfa and IL1beta expression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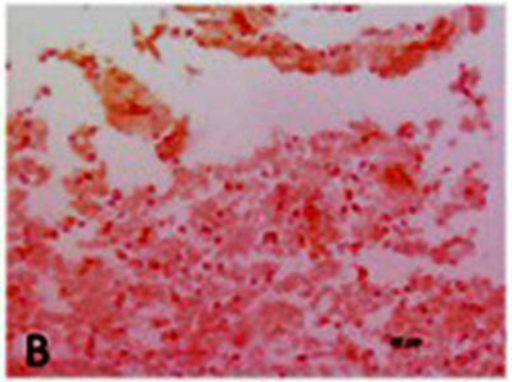
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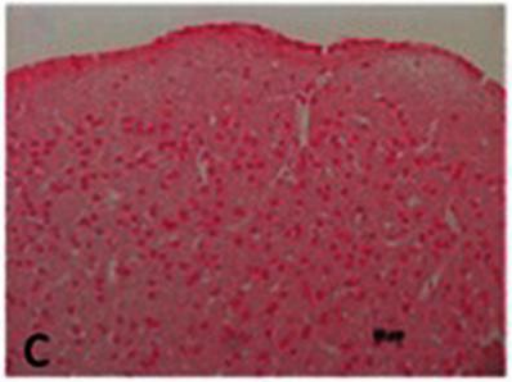
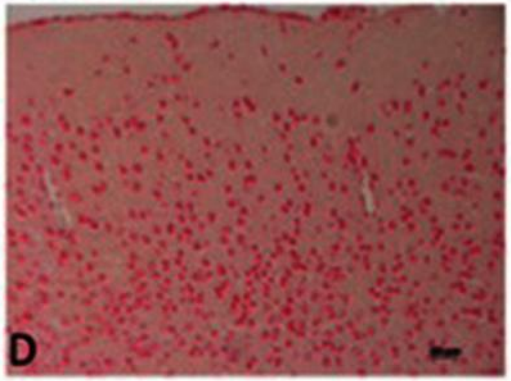
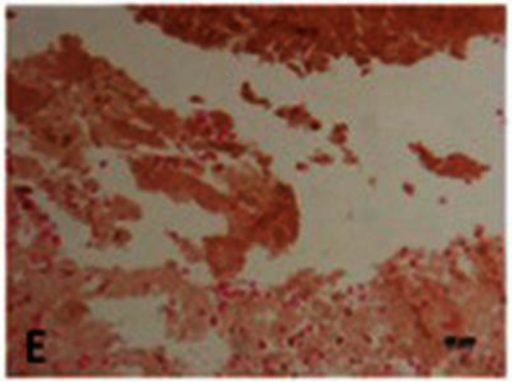
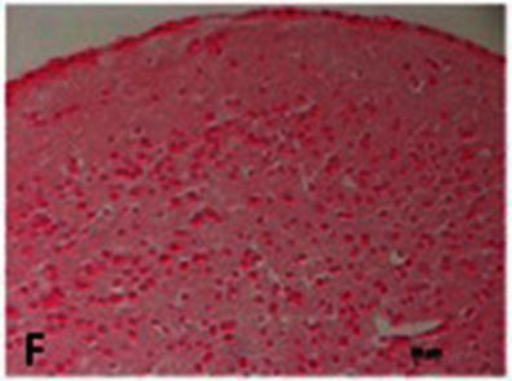

C


D


E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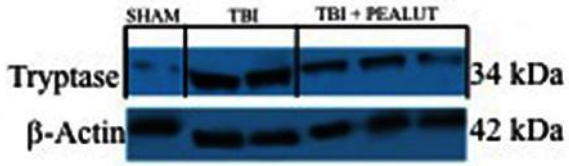
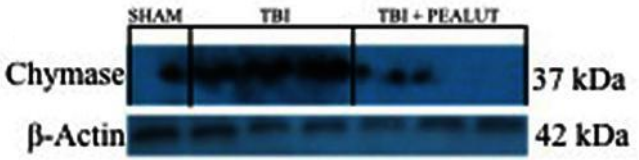
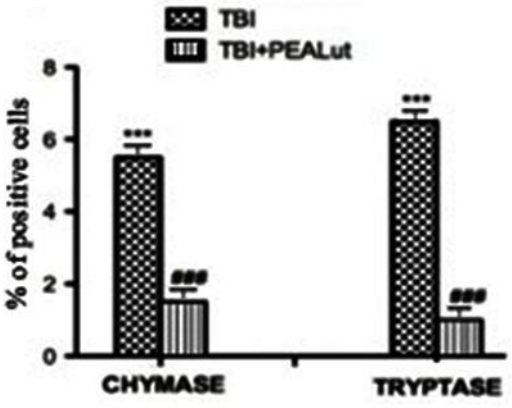
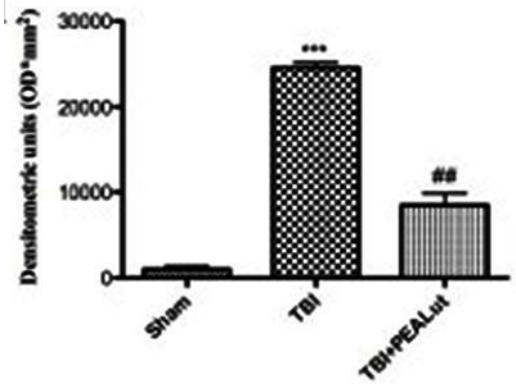
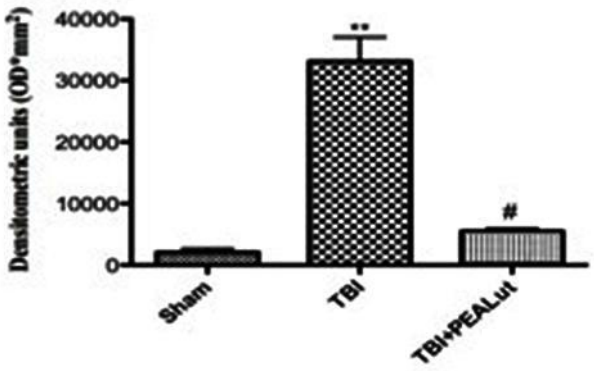
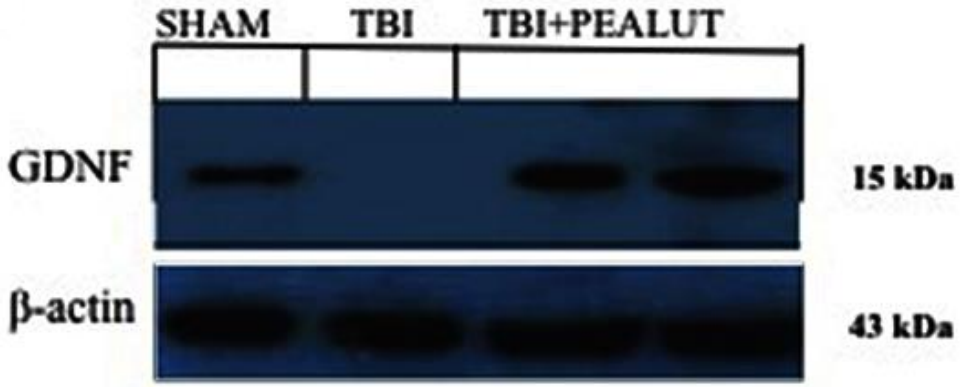

SHAM
TBI
TBI + PEALUT
Tryptase
34 kDa
β-Actin
42 kDa

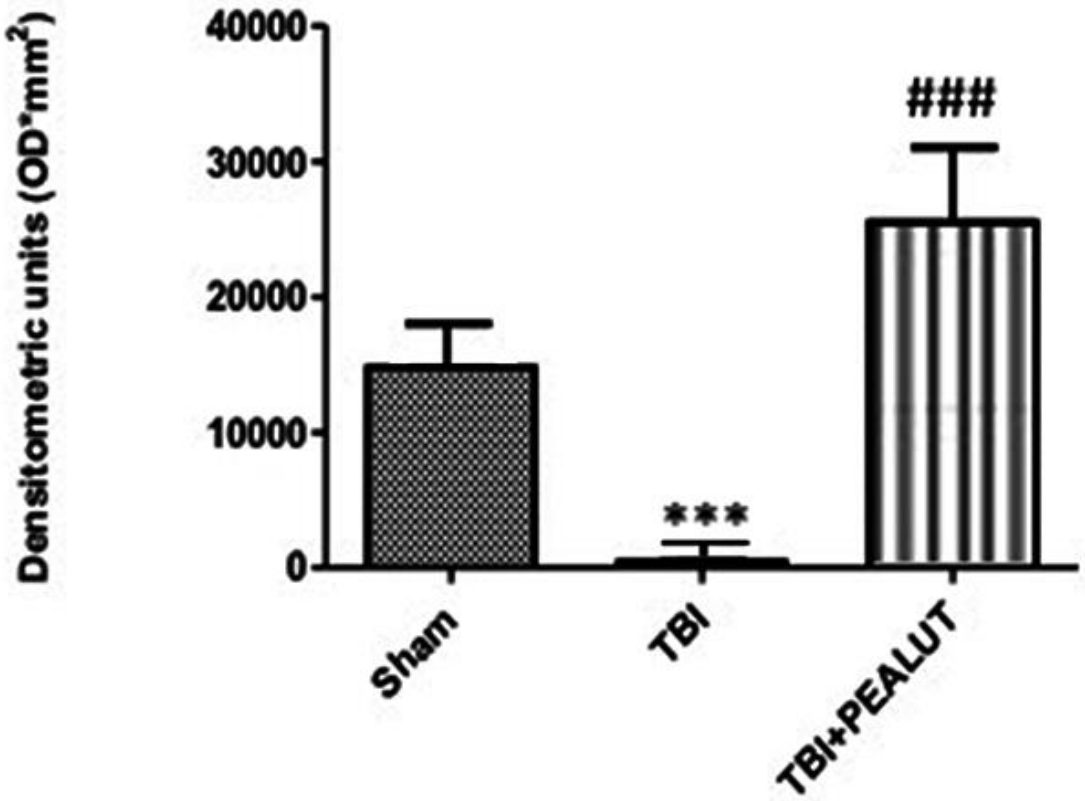

SHAM
TBI
TBI + PEALUT
Chymase
37 kDa
β-Actin
42 kDa


TBI
TBI+PEALut
% of positive cells
0
2
4
6
8
***
###
***
###
CHYMASE
TRYPTASE

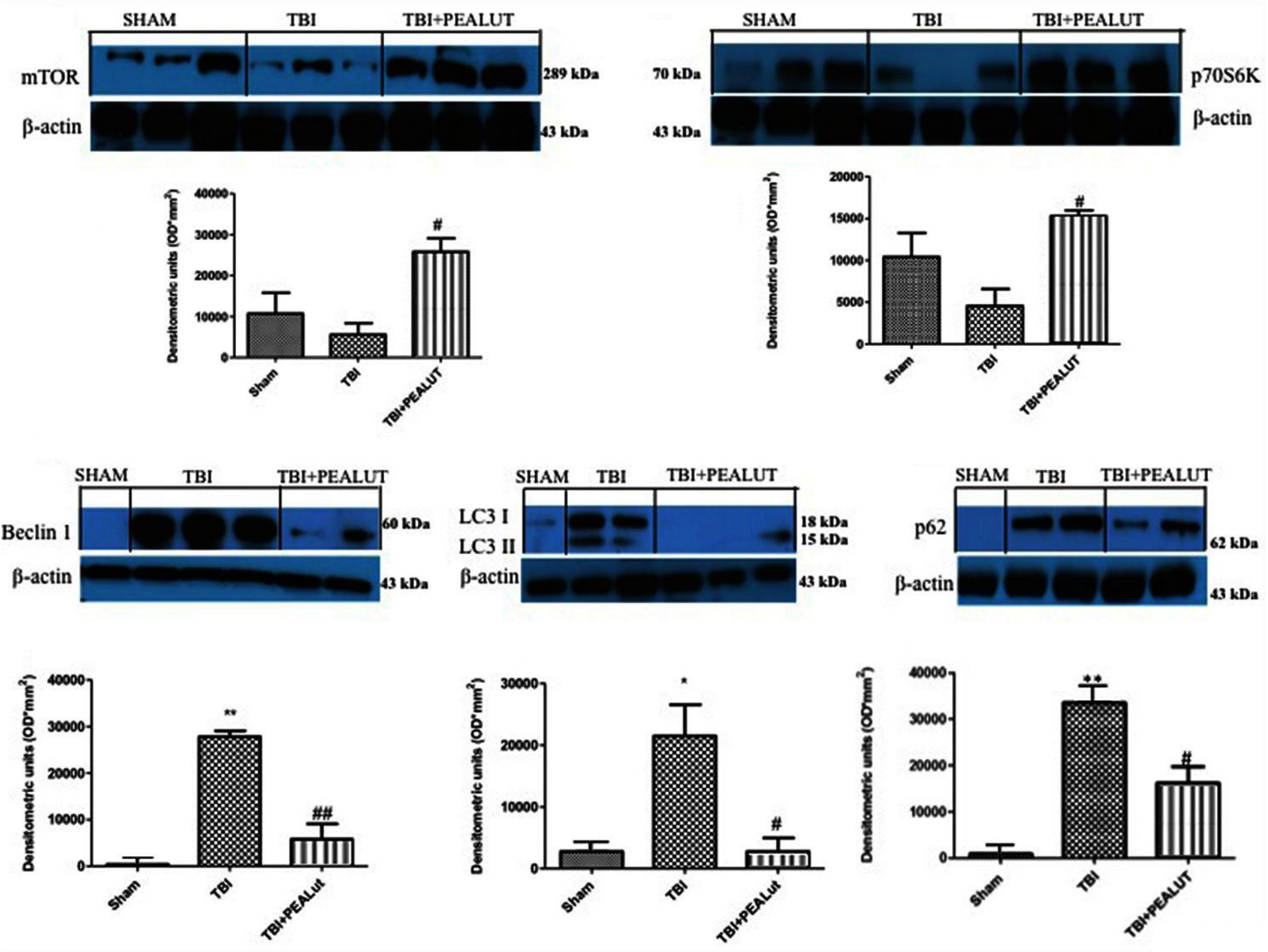

Densitometric units (OD*mm²)
0
10000
20000
30000
***
##
Sham
TBI
TBI+PEALut


Densitometric units (OD*mm²)
0
10000
20000
30000
40000
**
#
Sham
TBI
TBI+PEALut

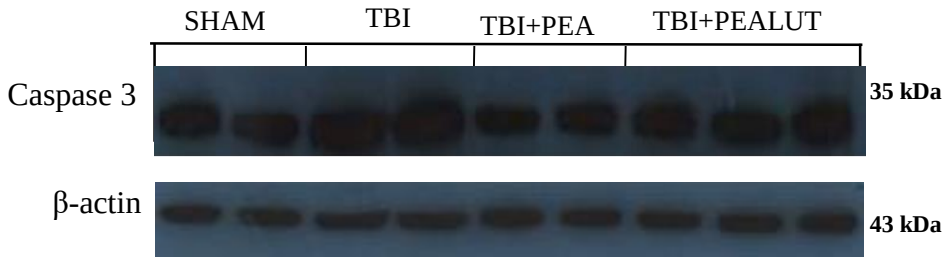

SHAM
TBI
TBI+PEALUT
GDNF
15 kDa
β-actin
43 kDa

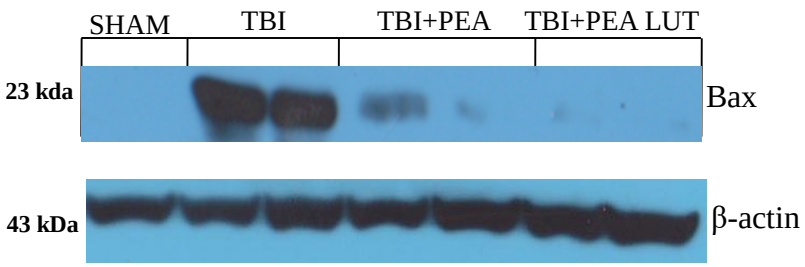

Densitometric units (OD*mm²)
40000
30000
20000
10000
0
###
***
Sham
TBI
TBI+PEALUT

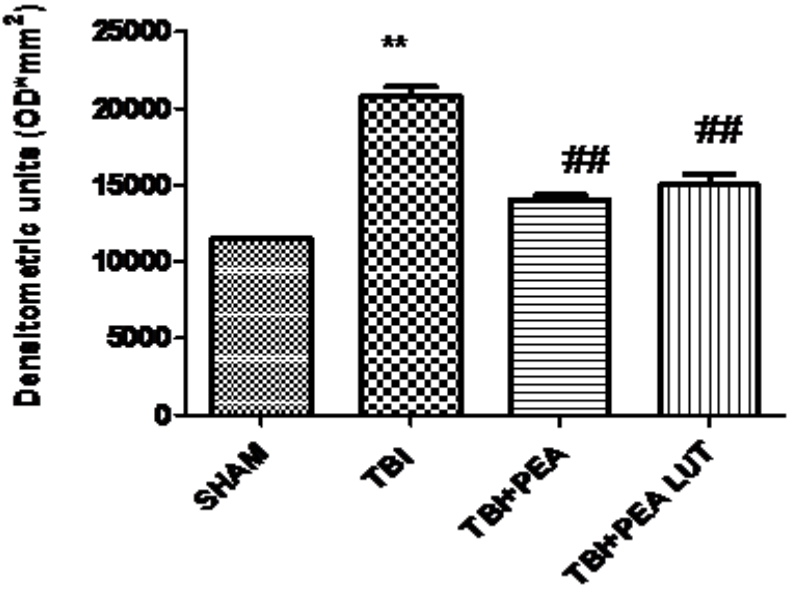
GDNF


SHAM
TBI
TBI+PEALUT
mTOR
289 kDa
β-actin
43 kDa
Densitometric units (OD*mm²)
40000
30000
20000
10000
0
#
Sham
TBI
TBI+PEALUT
70 kDa
p70S6K
43 kDa
β-actin
20000
15000
10000
5000
0
#
Sham
TBI
TBI+PEALUT
SHAM
TBI
TBI+PEALUT
Beclin 1
60 kDa
β-actin
43 kDa
LC3 I
LC3 II
18 kDa
15 kDa
β-actin
43 kDa
p62
62 kDa
β-actin
43 kDa
Densitometric units (OD*mm²)
40000
30000
20000
10000
0
**
##
Sham
TBI
TBI+PEALut
30000
20000
10000
0
*
#
Sham
TBI
TBI+PEALut
40000
30000
20000
10000
0
**
#
Sham
TBI
TBI+PEALUT

APOPTOSIS


SHAM
TBI
TBI+PEA
TBI+PEALUT
Caspase 3
35 kDa
β-actin
43 kDa


SHAM
TBI
TBI+PEA
TBI+PEA LUT
23 kda
Bax
43 kDa
β-actin


Densitometric units (OD*mm2)
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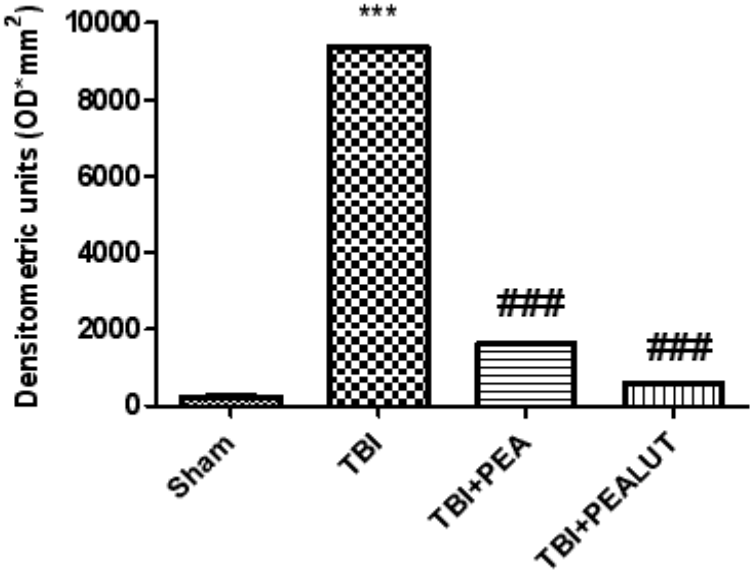
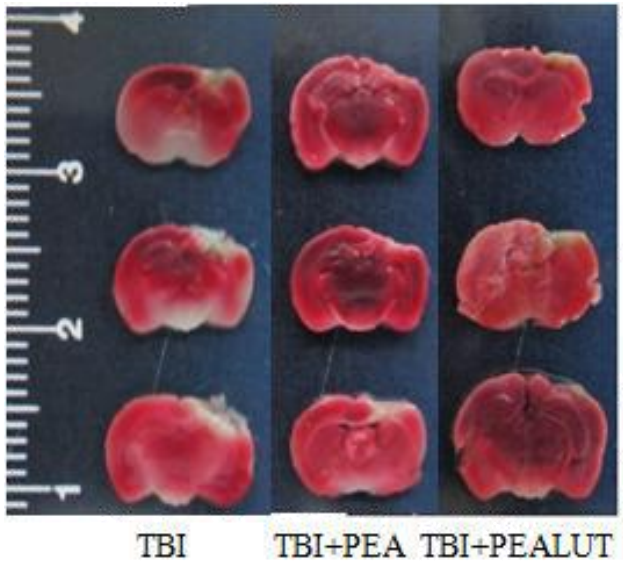
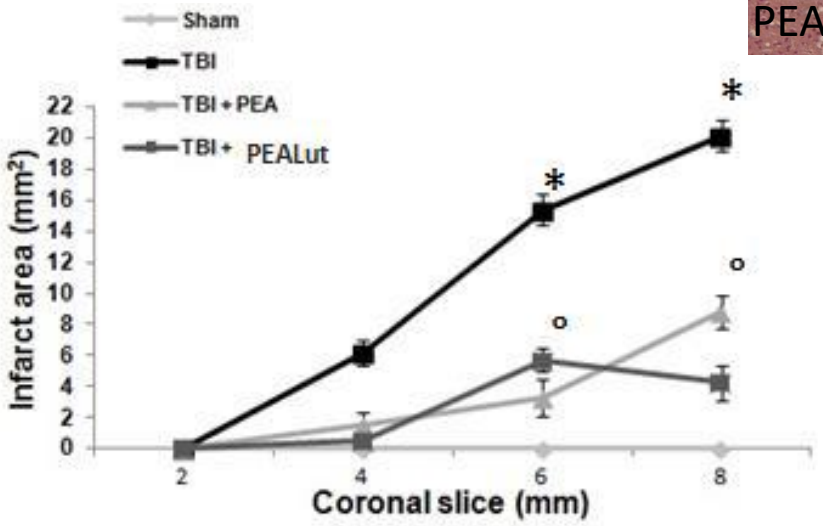
20000
15000
10000
5000
0
**
##
##
SHAM
TBI
TBI+PEA
TBI+PEA LUT

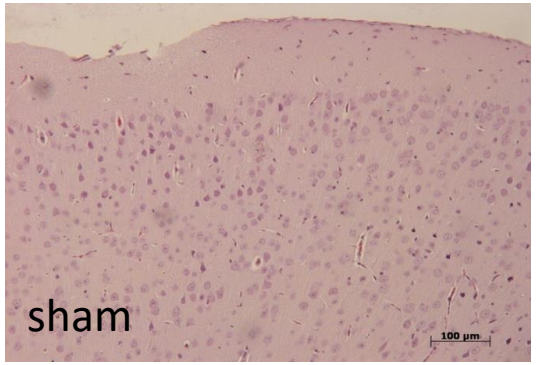

Densitometric units (OD*mm2)
10000
8000
6000
4000
2000
0
***
###
###
Sham
TBI
TBI+PEA
TBI+PEALUT

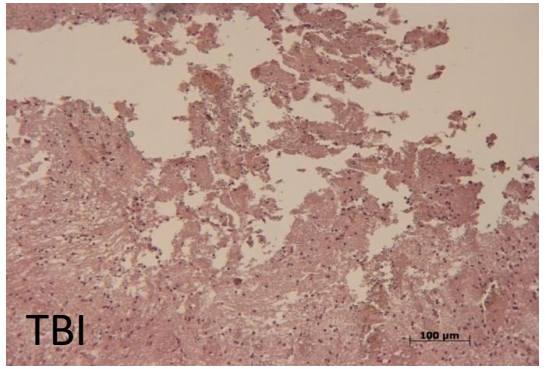
Co-ultraPEALut decreased edema and brain infarction area

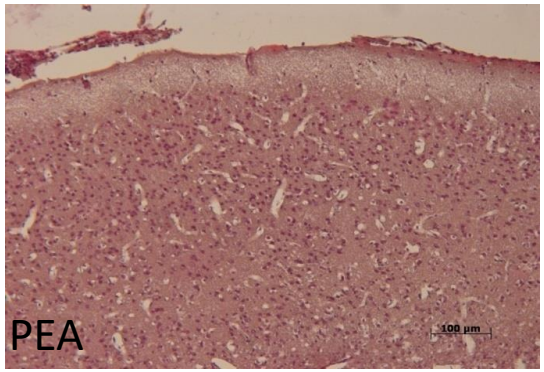


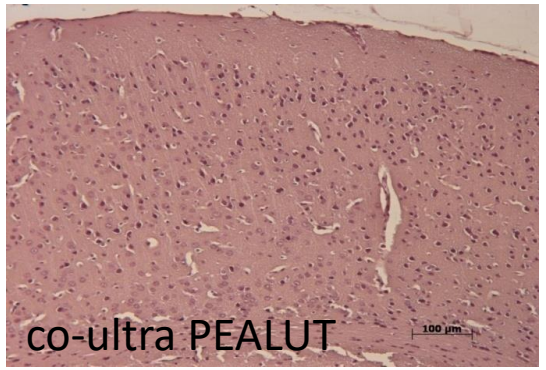


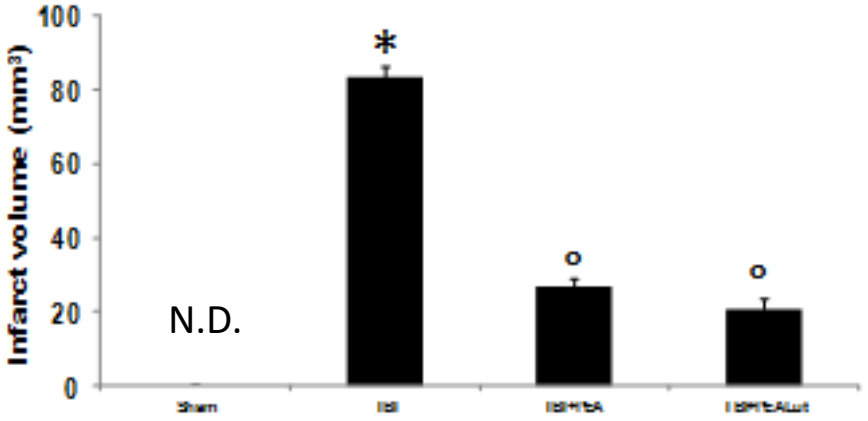
Effect of Co-ultraPEALut treatment on histological alterations of the brain tissue 24 hours after injury





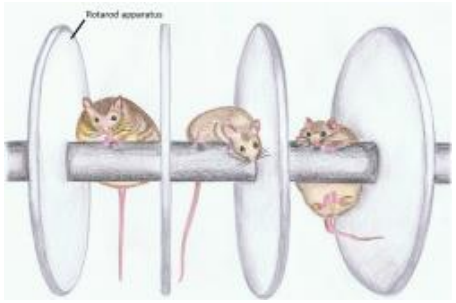






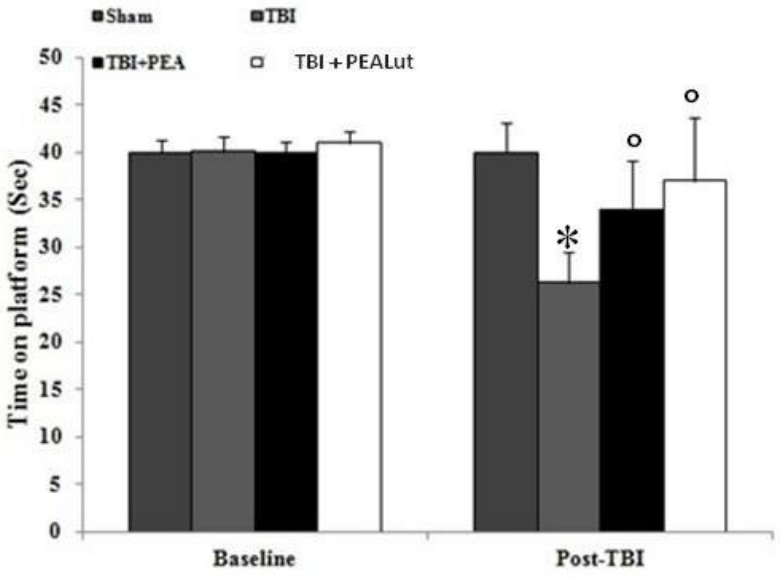
Behavioral Test

- Rotaroad test

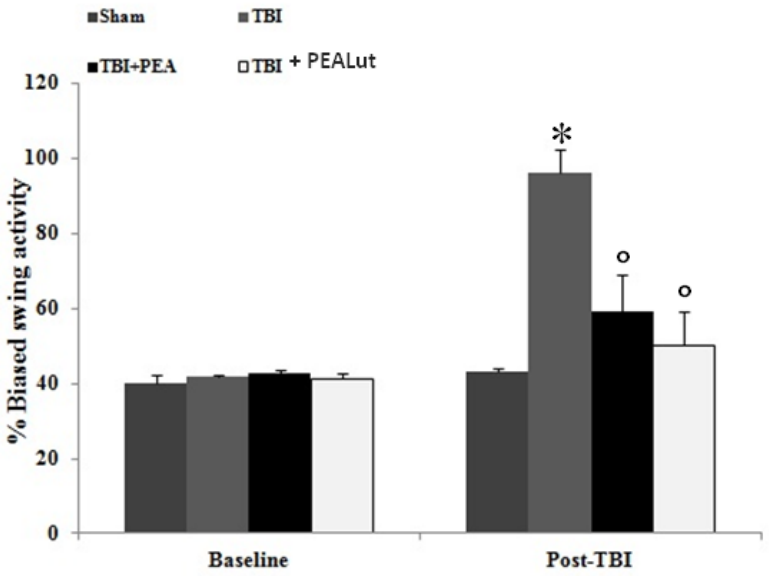


- Elevated Biased Swing Test (EBST)

ROTAROAD TEST



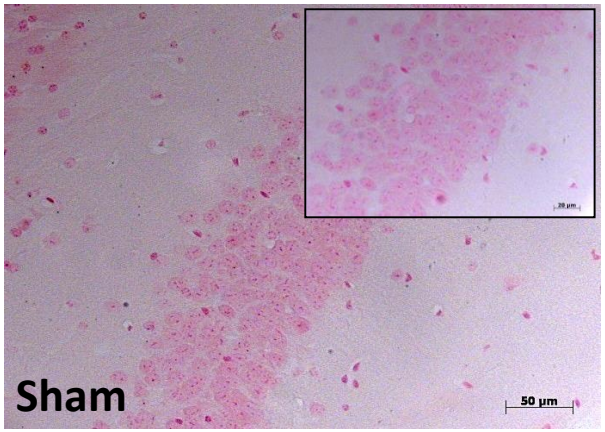
EBST

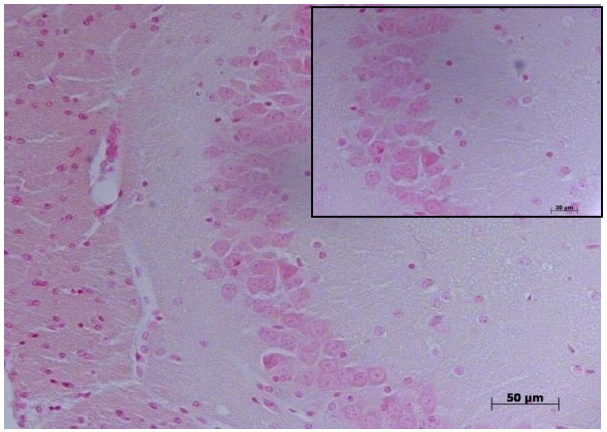


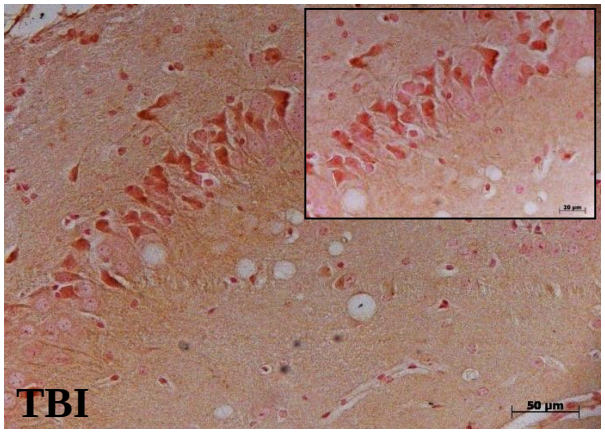
72h

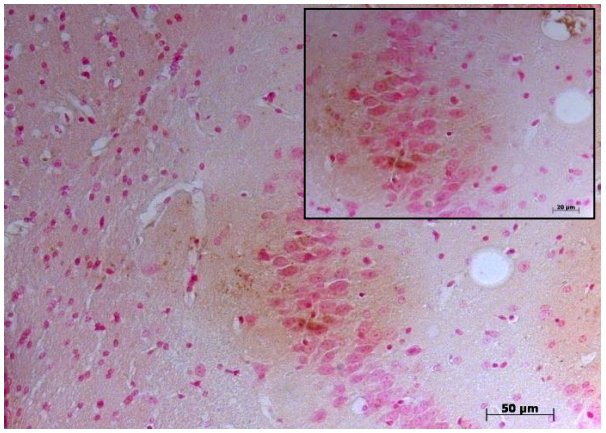
7d

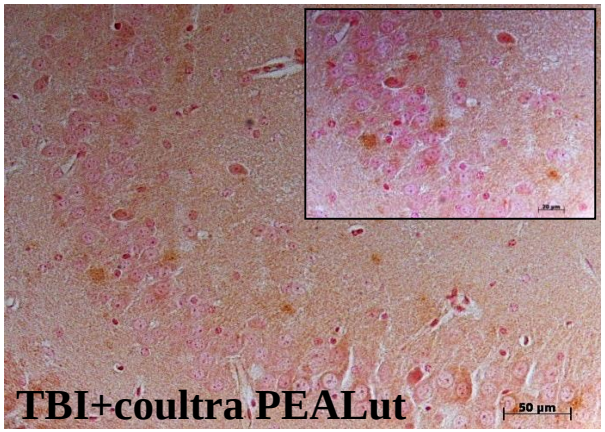
Effect of co-ultra PEALut on newly born cell

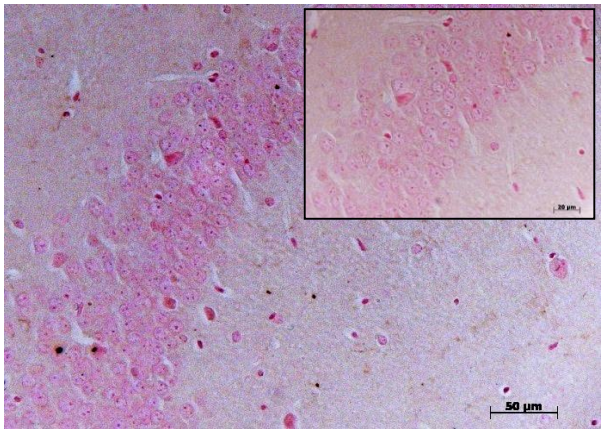




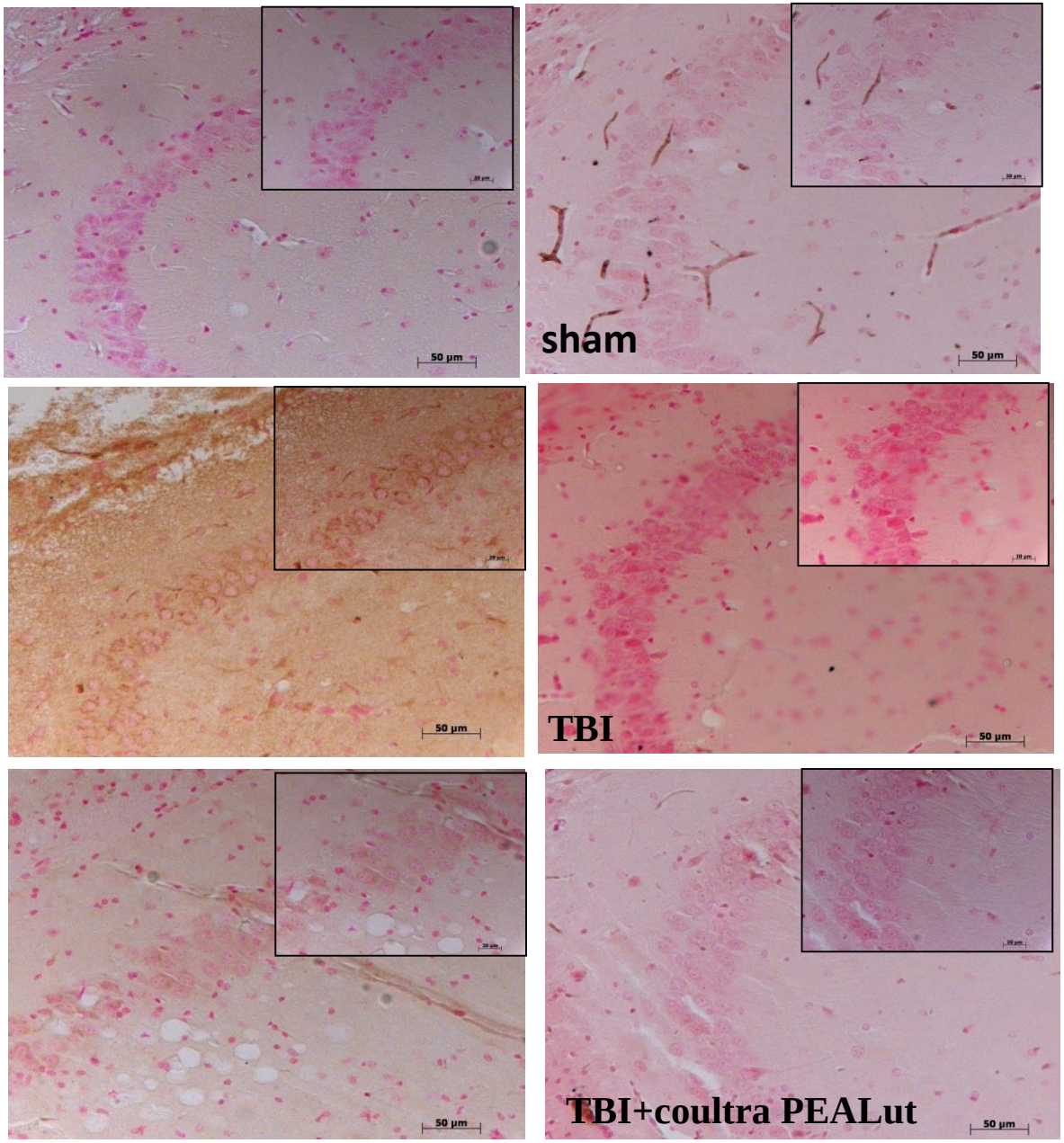




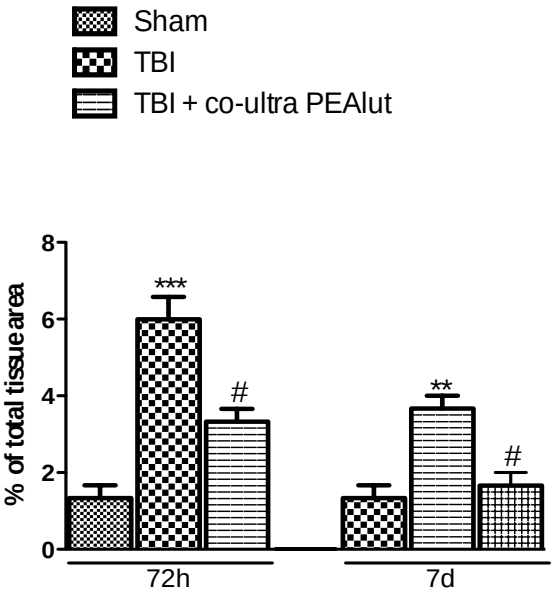




72h *7d*



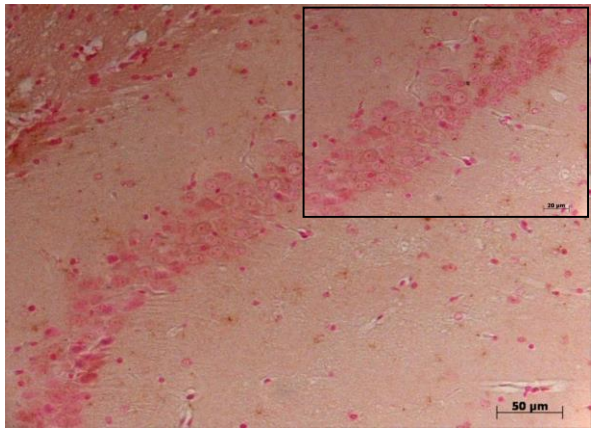
Effect of co-ultra PEALut on astroglial cell activation

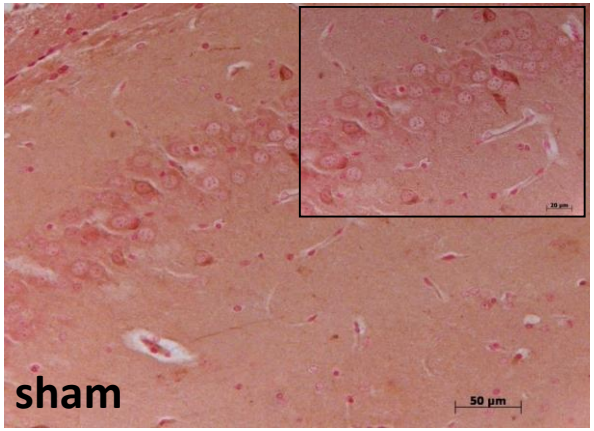


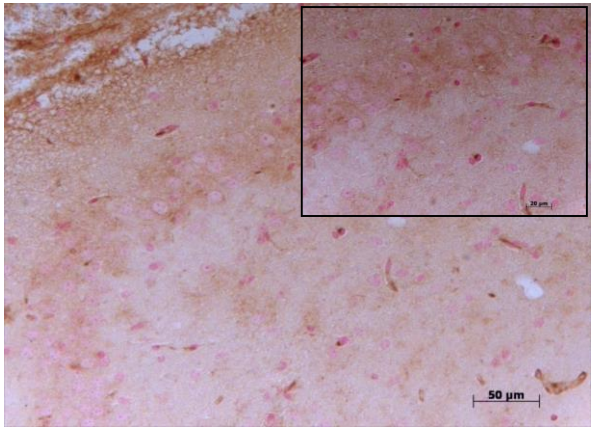
Effect of co-ultra PEALut on BDNF expression

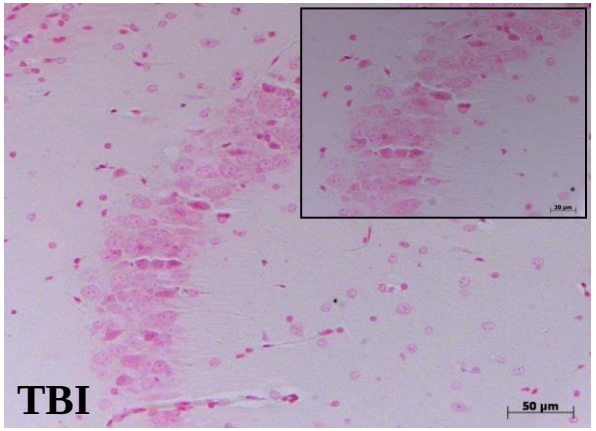
72h

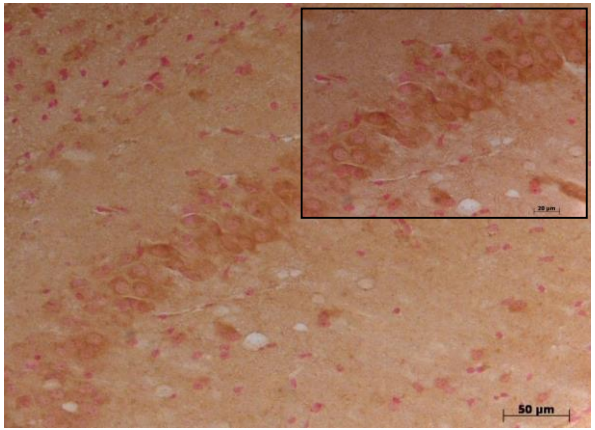
7d

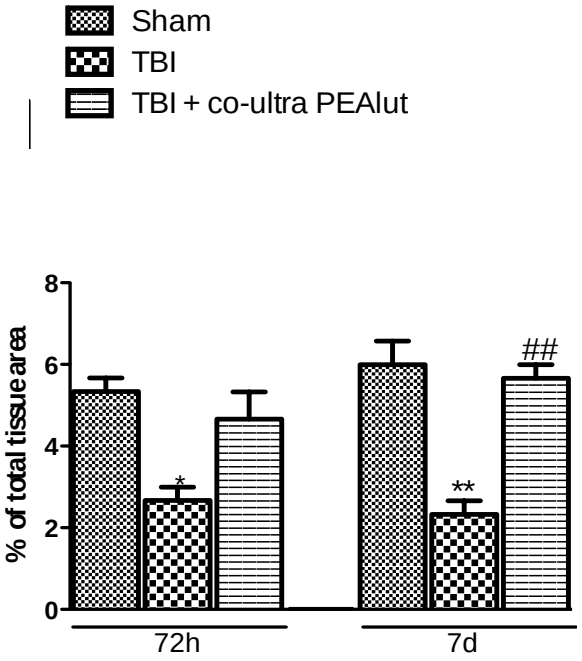












III° Hypothesis

Could co-ultraPEALut ® enhances neuronal recovery after a moderate traumatic brain injury: from mice to human?

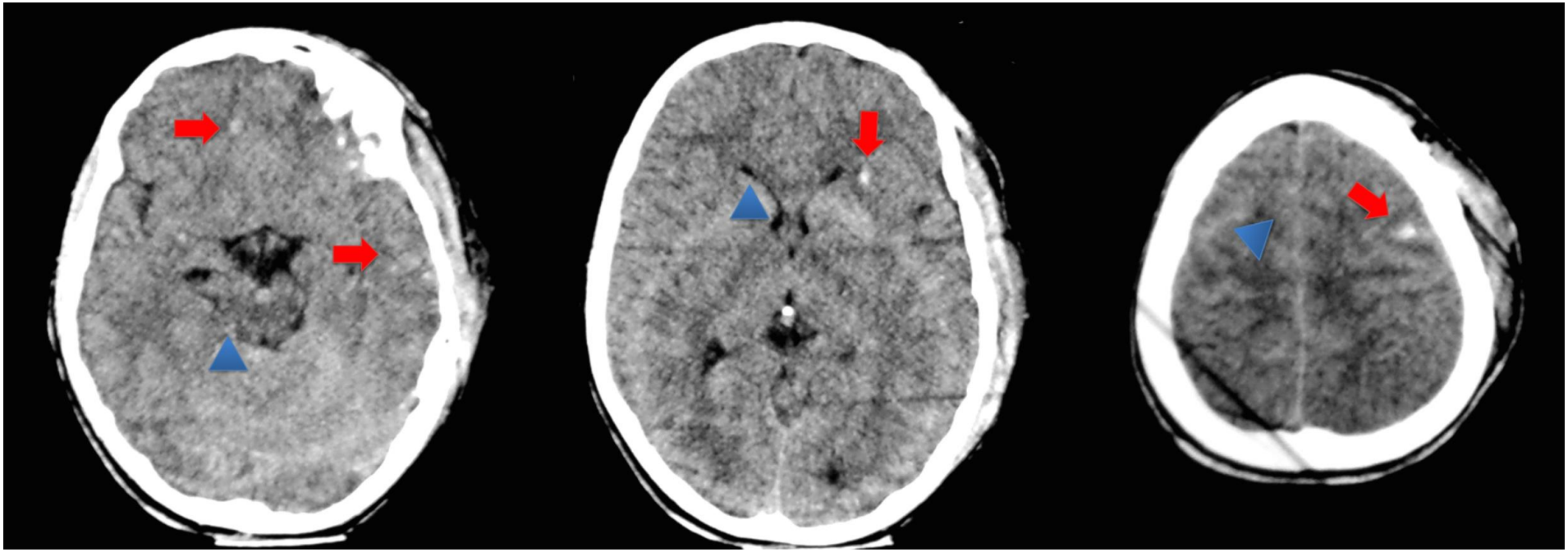
Case Report

- A 12 year-old male patient with TBI from a bicycle-car accident was admitted to inpatient rehabilitation unit.
- On arrival to the emergency department, his Glasgow Coma Scale (GOS) score was 3. He had a history of coma for 2 months in the intensive care unit.
- Neurologically, he had diffuse axonal brain injury, large frontal and temporal contusions, brain swelling, and a massive epidural hematoma on the left parietal lobe

Treatment

- Co-ultraPEALut (GLIALIA®) was administered to the patient starting the first week of admission to our department in an attempt to improve functional recovery and consciousness, and in association with intensive neurorehabilitation.
- GLIALIA® treatment was carried out for the first 10 weeks enterally as a suspension in polyethylene glycol, at a dose of 700 mg + 70 mg twice daily. This was followed by sublingual administration of microgranular GLIALIA® at the same dose until discharge

Axial computed tomography on the day of injury for acute evaluation of TBI: note the evident brain edema and hematoma in the left parietal lobe.
MRI images after surgical treatment: note the persistence of edema and emergence of bifrontal hygroma



The extent of improvement achieved by the patient after 16 weeks of rehabilitation and treatment with GLIALIA®) is represented by the difference between the area enclosed within the red circle (indicating complete absence of any degree of functional independence at baseline) and that enclosed by the blue line (indicating the degree of functional autonomy and the parameters evaluated)

FIM scores in a 13-year-old TBI patient before and after 16 weeks of treatment with GLIALIA®

FIM SCALE	Admission Time 0	Discharge 16 weeks after
ACTIVITY OF DAILY LIVING		
1) Eating	1	5
2) Grooming	1	5
3) Bathing	1	5
4) Upper body dressing	1	3
5) Lower body dressing	1	3
6) Toileting	1	3
SPHINCTER CONTROL		
7) Bladder management	1	3
8) Bowel management	1	4
TRANSFER		
9) Bed to chair transfer	1	4
10) Toilet transfer	1	3
11) Shower transfer	1	3
LOCOMOTION		
12) Walking or wheelchair	1	4
13) Stairs	1	2
TOTAL MOTOR SCORE	**13**	**47**
COMMUNICATION		
14) Comprehension	1	7
15) Expression	1	6
SOCIAL COGNITION		
16) Social Interaction	1	7
17) Problem solving	1	6
18) Memory	1	5
TOTAL COGNITIVE SCORE	**5**	**31**
TOTAL FIM SCORE	**18/126**	**78/126**

FIM and LCF scores in a 13-year-old TBI patient before and after 16 weeks of treatment with GLIALIA®

FIM Score	Time 0	16 weeks treatment with co-ultraPEALut
Total	18/126	78/126
Motor subscale	13/91	47/91
Cognitive subscale	5/35	31/35
LCF Score	1	6

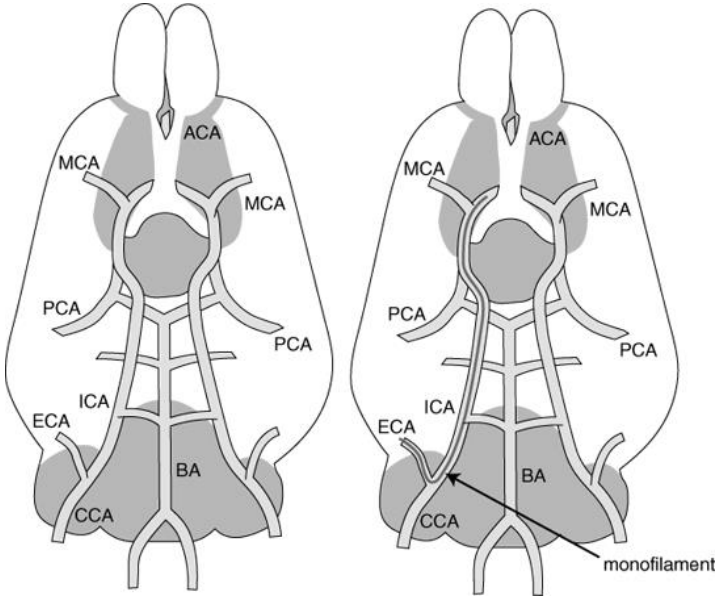
IV° Hypothesis

Transl. Stroke Res. (2016) 7:54–69
DOI 10.1007/s12975-015-0440-8

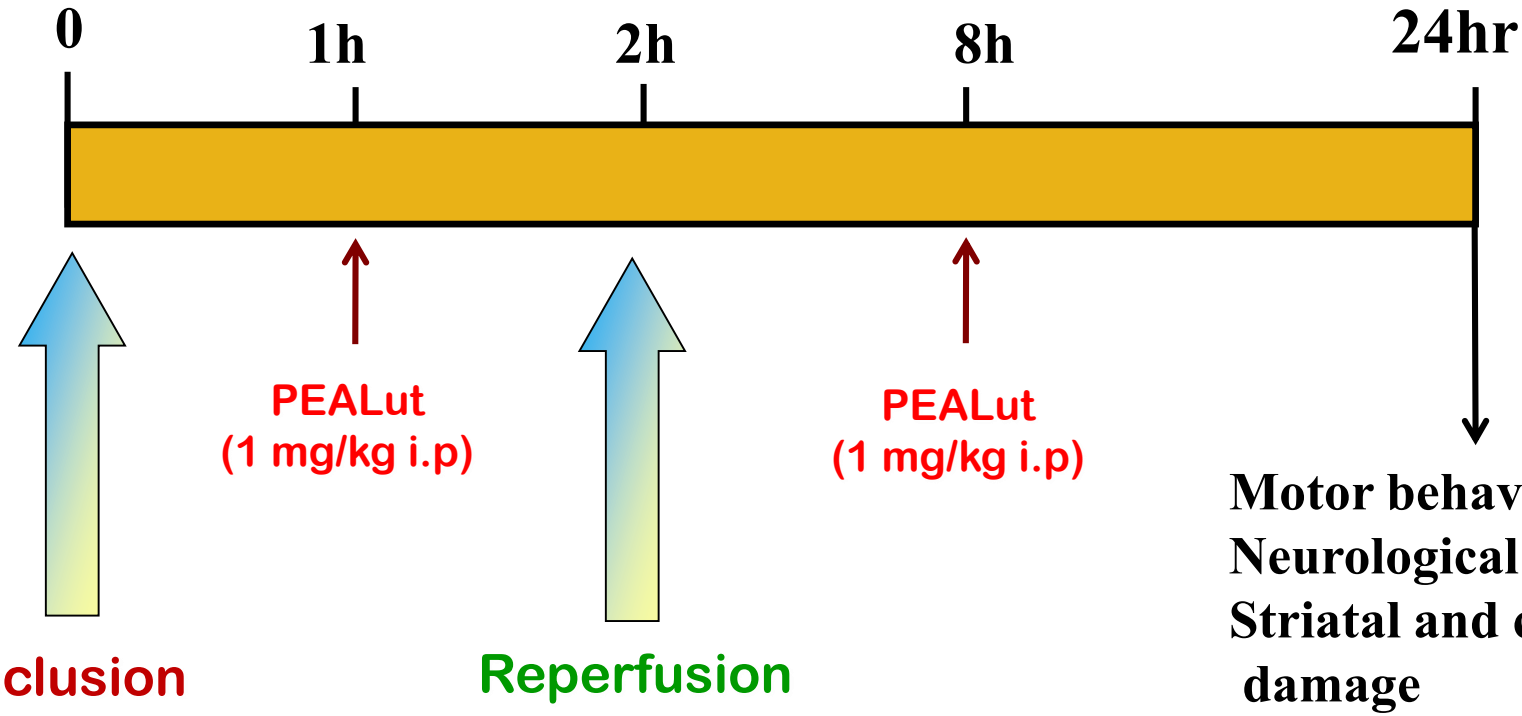
ORIGINAL ARTICLE

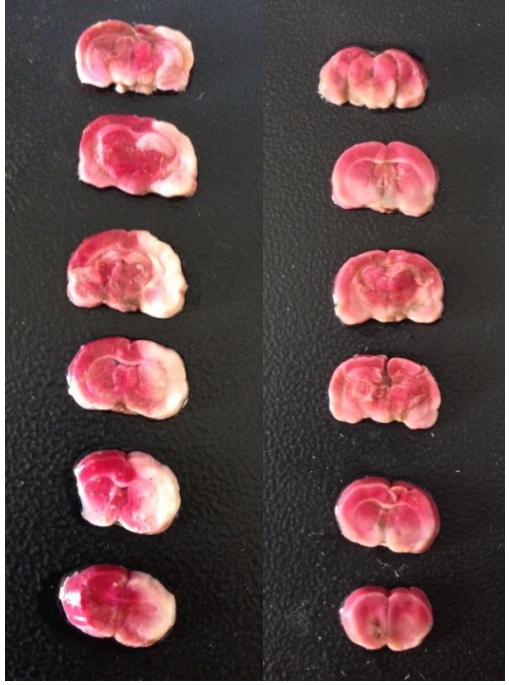
Co-ultramicronized Palmitoylethanolamide/Luteolin in the Treatment of Cerebral Ischemia: from Rodent to Man

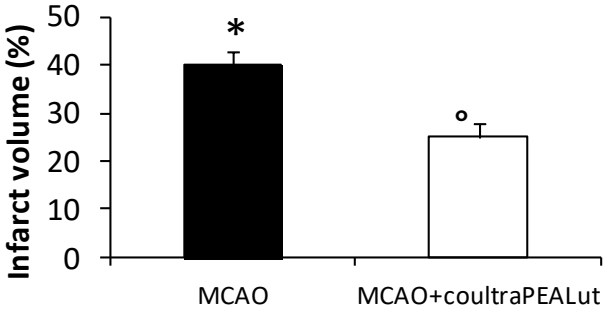
Carlo Caltagirone[1] · Carlo Cisari[2] · Carlo Schievano[4] · Rosanna Di Paola[3] · Marika Cordaro[3] · Giuseppe Bruschetta[3] · Emanuela Esposito[3] · Salvatore Cuzzocrea[3] · Stroke Study Group

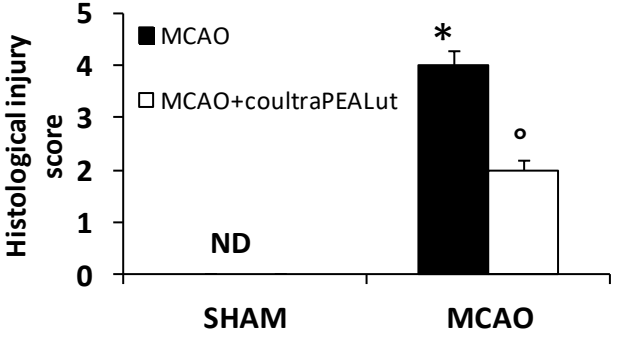


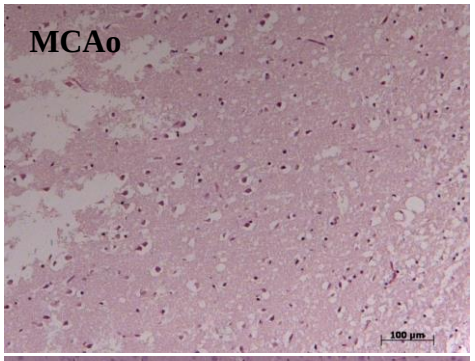
Experimental model of middle cerebral artery occlusion middle (MCAo) in rats

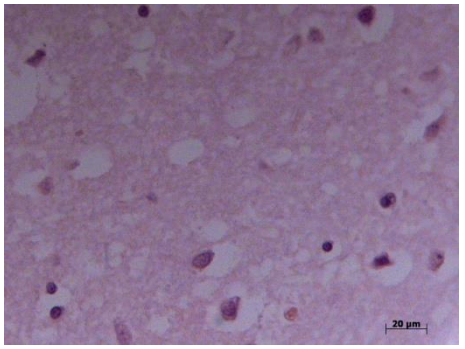


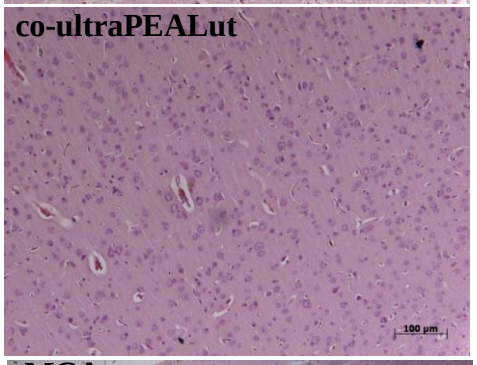



Infarct volume (%)
50
40
30
20
10
0
*
°
MCAO
MCAO+coultraPEALut


Histological injury score
5
4
3
2
1
0
MCAO
MCAO+coultraPEALut
*
°
ND
SHAM
MCAO

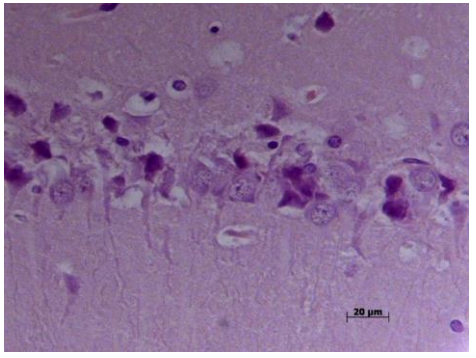

MCAo
100 µm


20 µm

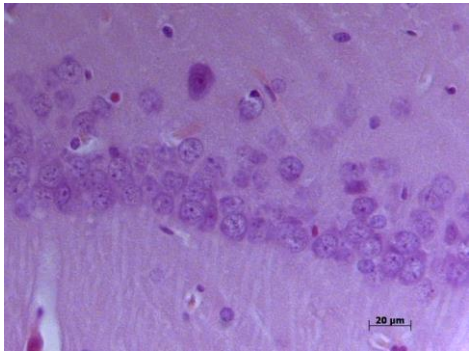

co-ultraPEALut
100 µm

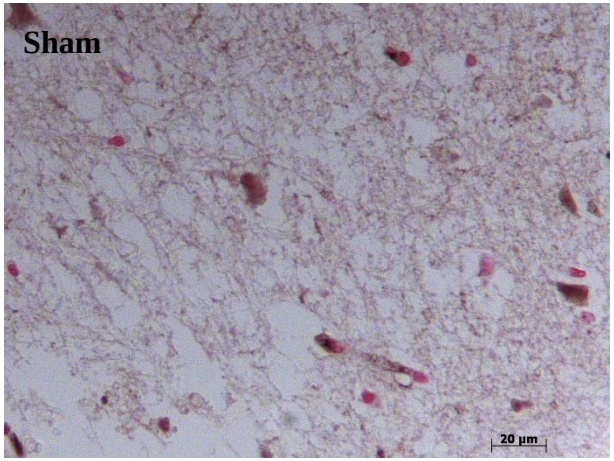
20 µm

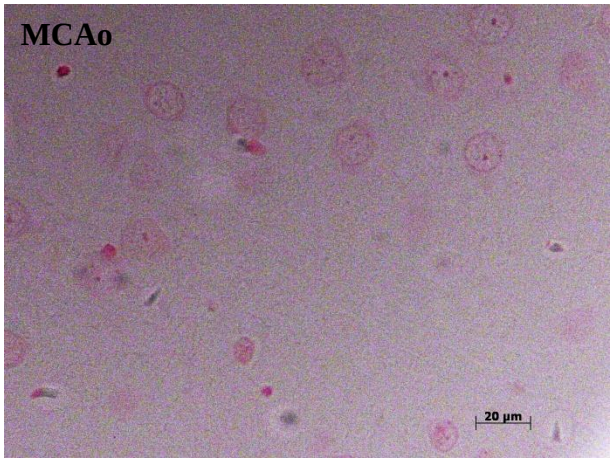
MCAo
0.5 mm

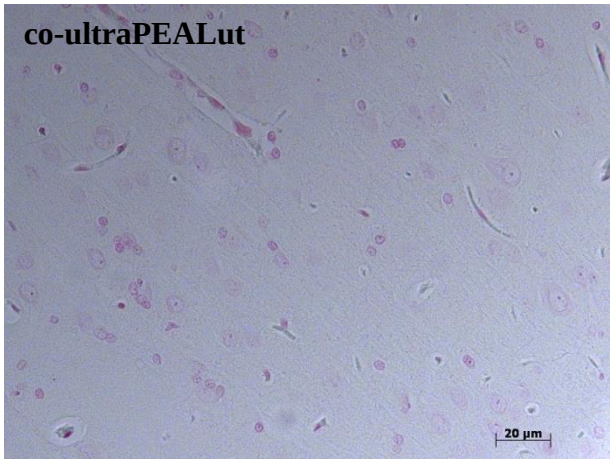

20 µm

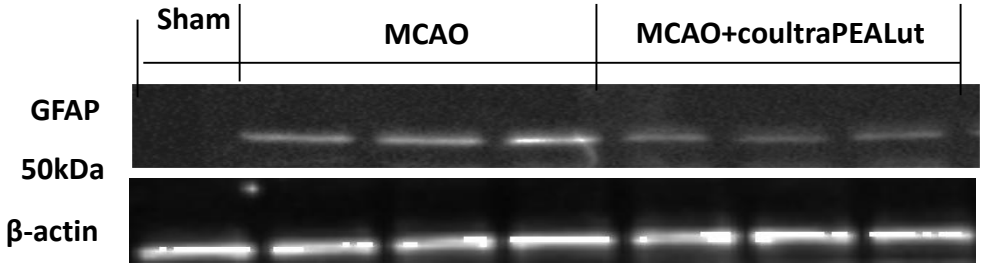
co-ultraPEALut
0.5 mm

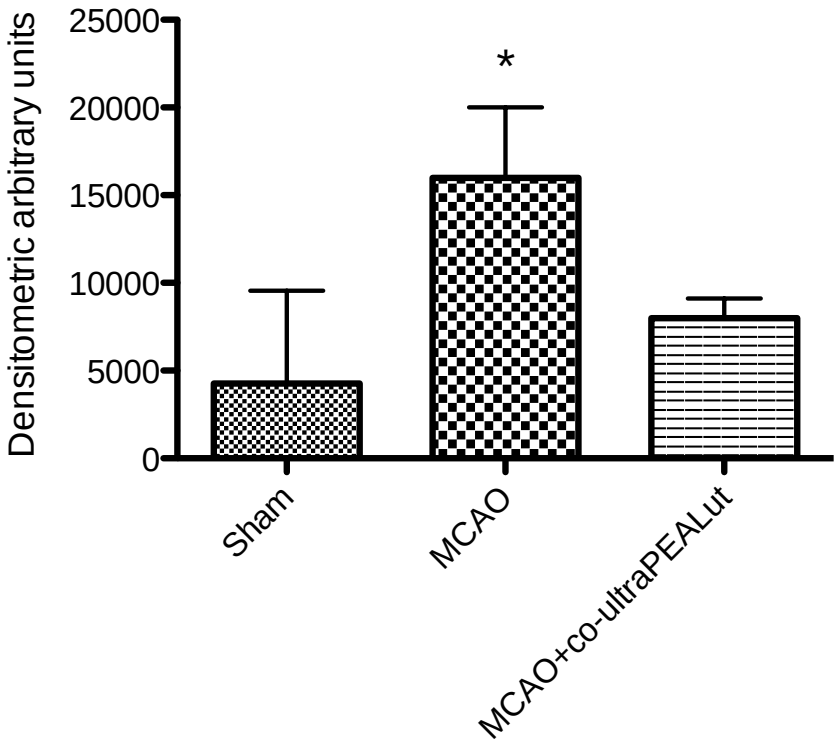

20 µm


Sham
20 µm

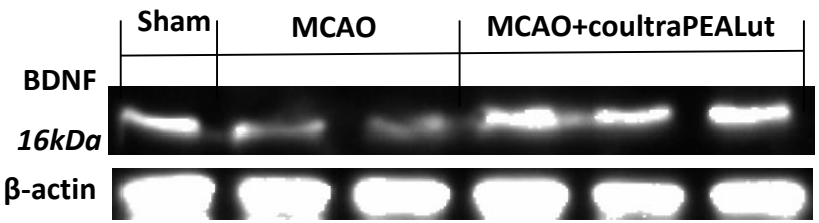

MCAo
20 µm

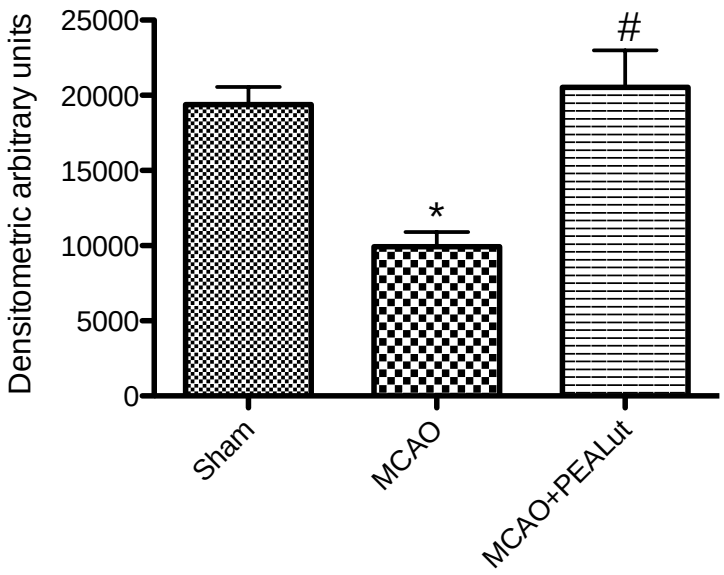

co-ultraPEALut
20 µm

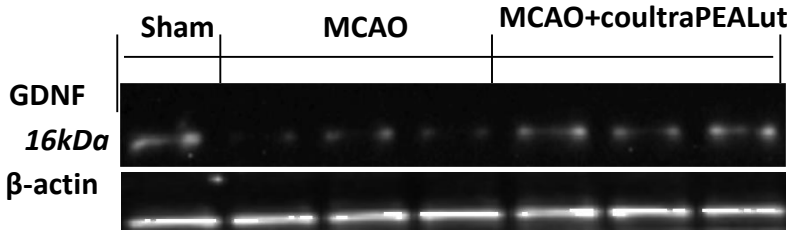

Sham
MCAO
MCAO+coultraPEALut
GFAP
50kDa
β-actin

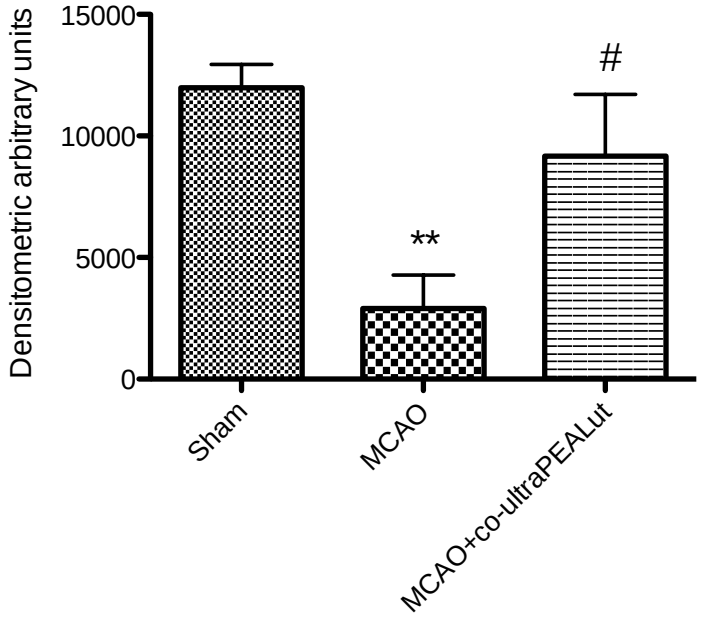

Densitometric arbitrary units
25000
20000
15000
10000
5000
0
*
Sham
MCAO
MCAO+co-ultraPEALut

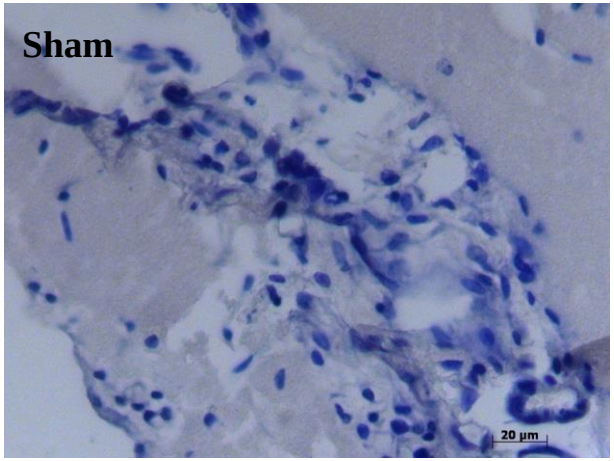
Neurotrophic Factor

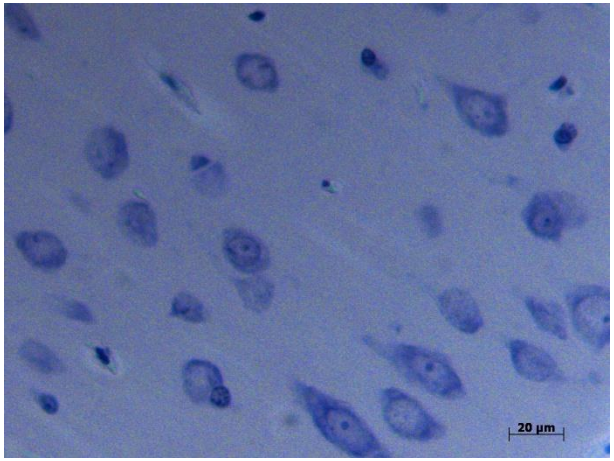


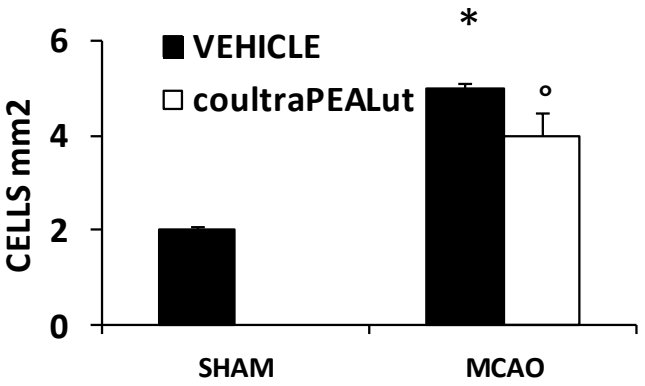


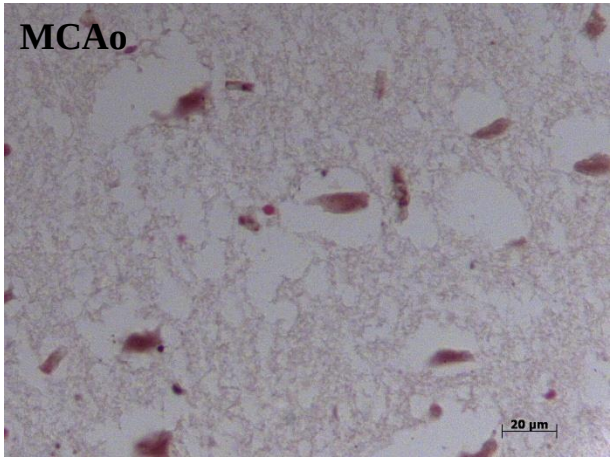




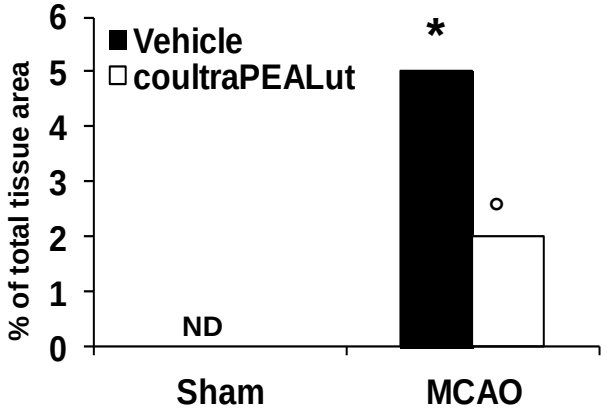

Sham
20 µm

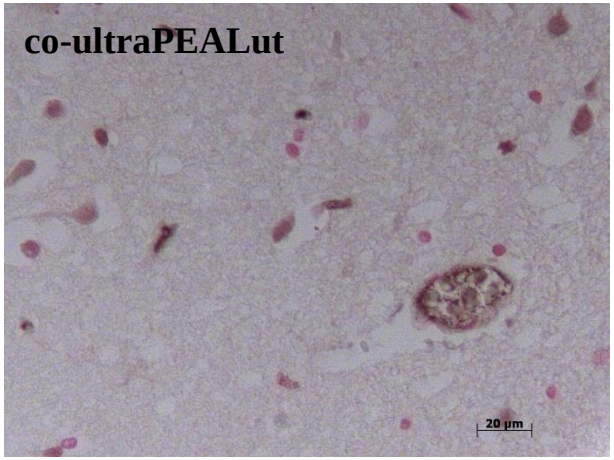

20 µm

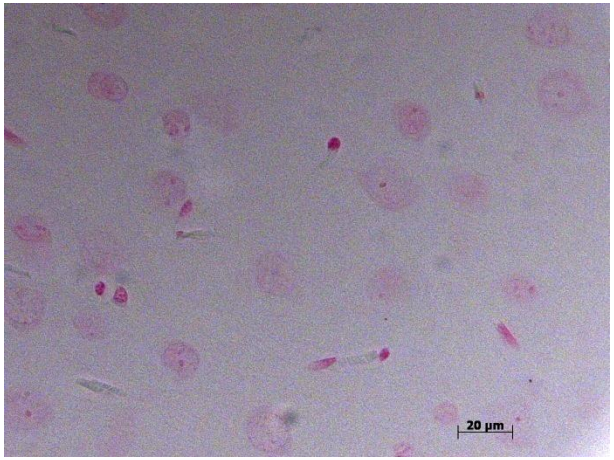

CELLS mm2
6
4
2
0
VEHICLE
coultraPEALut
*
°
SHAM
MCAO

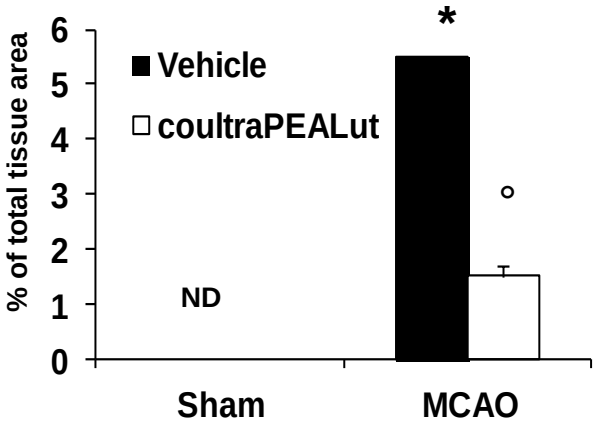

MCAo
20 µm

20 µm

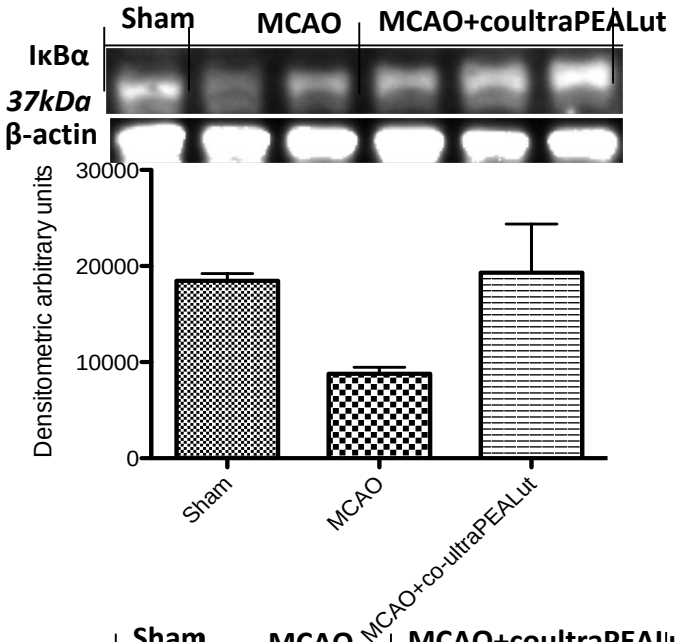

% of total tissue area
6
5
4
3
2
1
0
Vehicle
coultraPEALut
*
°
ND
Sham
MCAO

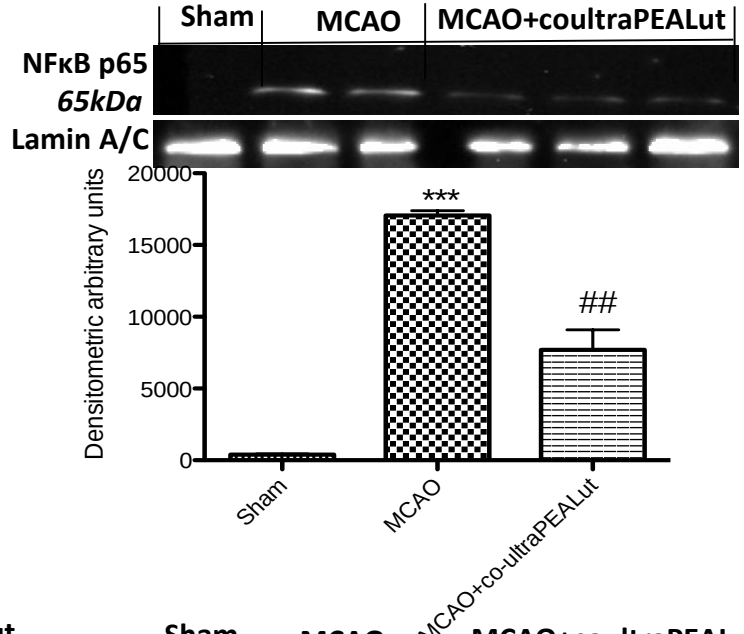

co-ultraPEALut
20 µm

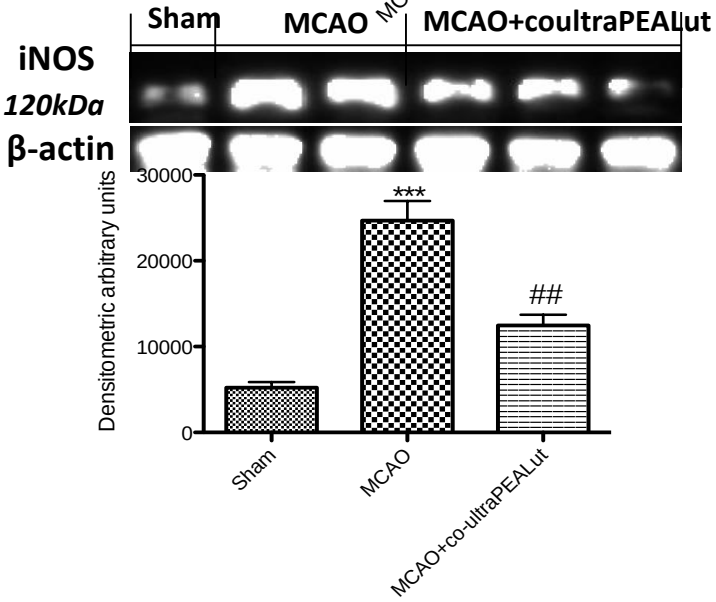

20 µm

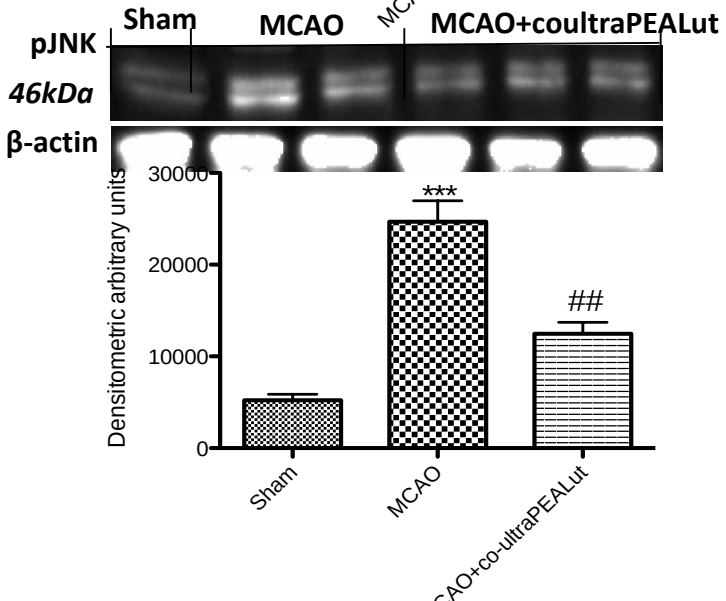

% of total tissue area
6
5
4
3
2
1
0
Vehicle
coultraPEALut
*
°
ND
Sham
MCAO

Inflammatory Studies

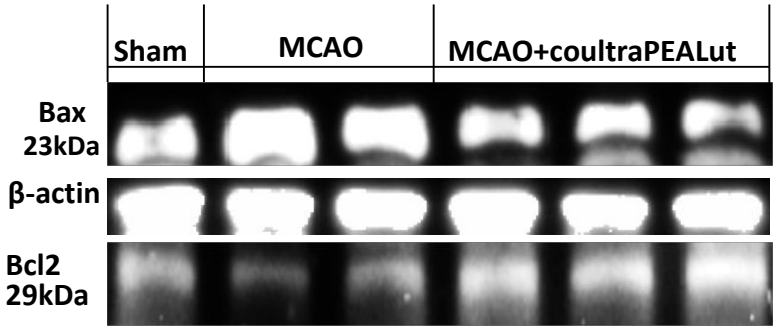


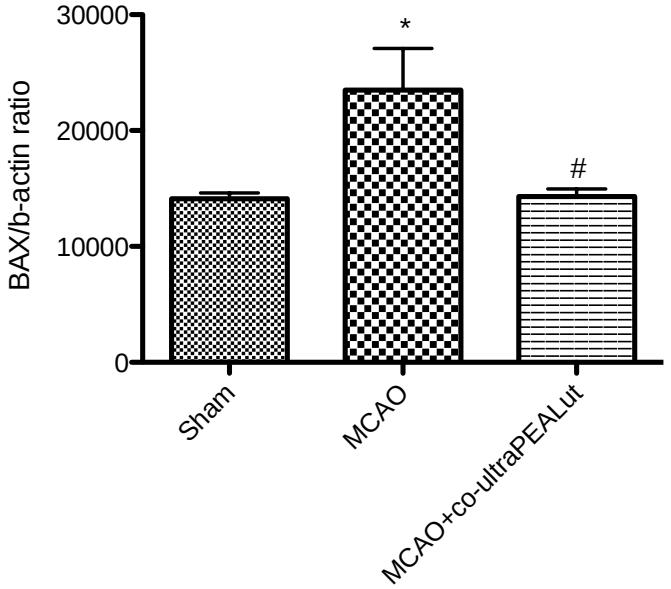


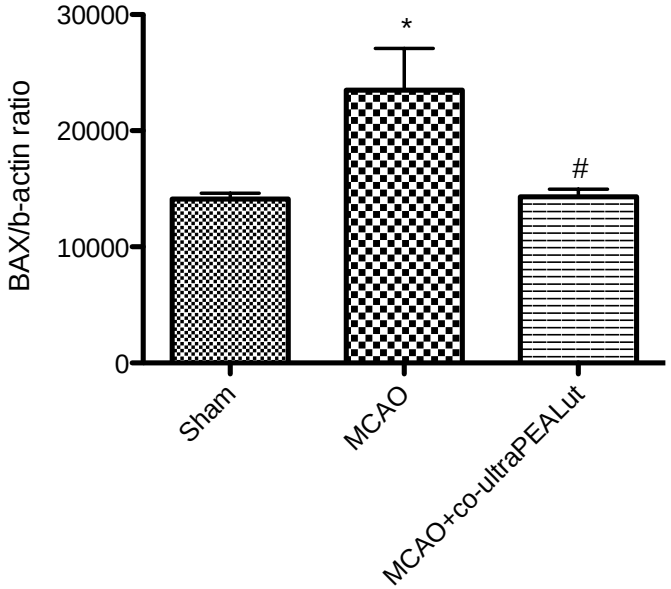


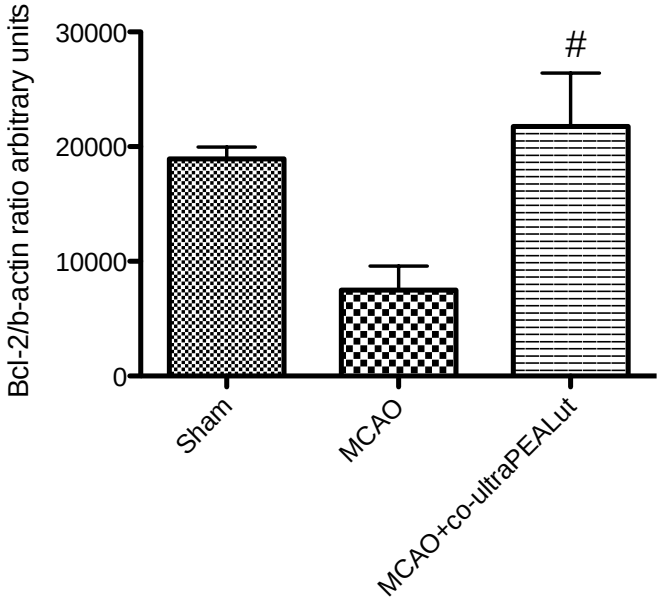


Apoptotic Studies









PROGETTO Glialia®

IN RIABILITAZIONE POST-ICTALE

Disegno di Studio

Obiettivi: valutare l'effetto del trattamento con Glialia® in pazienti affetti da ictus ischemico di primo evento in fase clinica stabilizzata e sottoposti a terapia riabilitativa.

Criteri di inclusione: Pazienti con ictus-ischemico di primo evento (lesione unilaterale) stabilizzato e sottoposti a terapia riabilitativa.

Trattamento: Glialia (700mg+70mg) microgranuli da assumere per via sublinguale ogni 12 ore in associazione alle terapie farmacologiche usualmente somministrate.

Tempo di trattamento: 60 giorni.

Valutazione	Scala	Time-points		
		Baseline (T0)	*Mese 1 (T30)*	*Mese 2 (T60)*
Gravità Neurologica	Canadian Neurological Scale (CNS)	√	√	
Compromissione cognitiva	Mini Mental State Examination (MMSE)	√	√	
Spasticità degli arti	Ashworth Scale (AS)	√	√	
Dolore	Numeric Rating Scale (NRS)	√	√	
Autonomia funzionale	Barthel Index (BI)	√	√	√

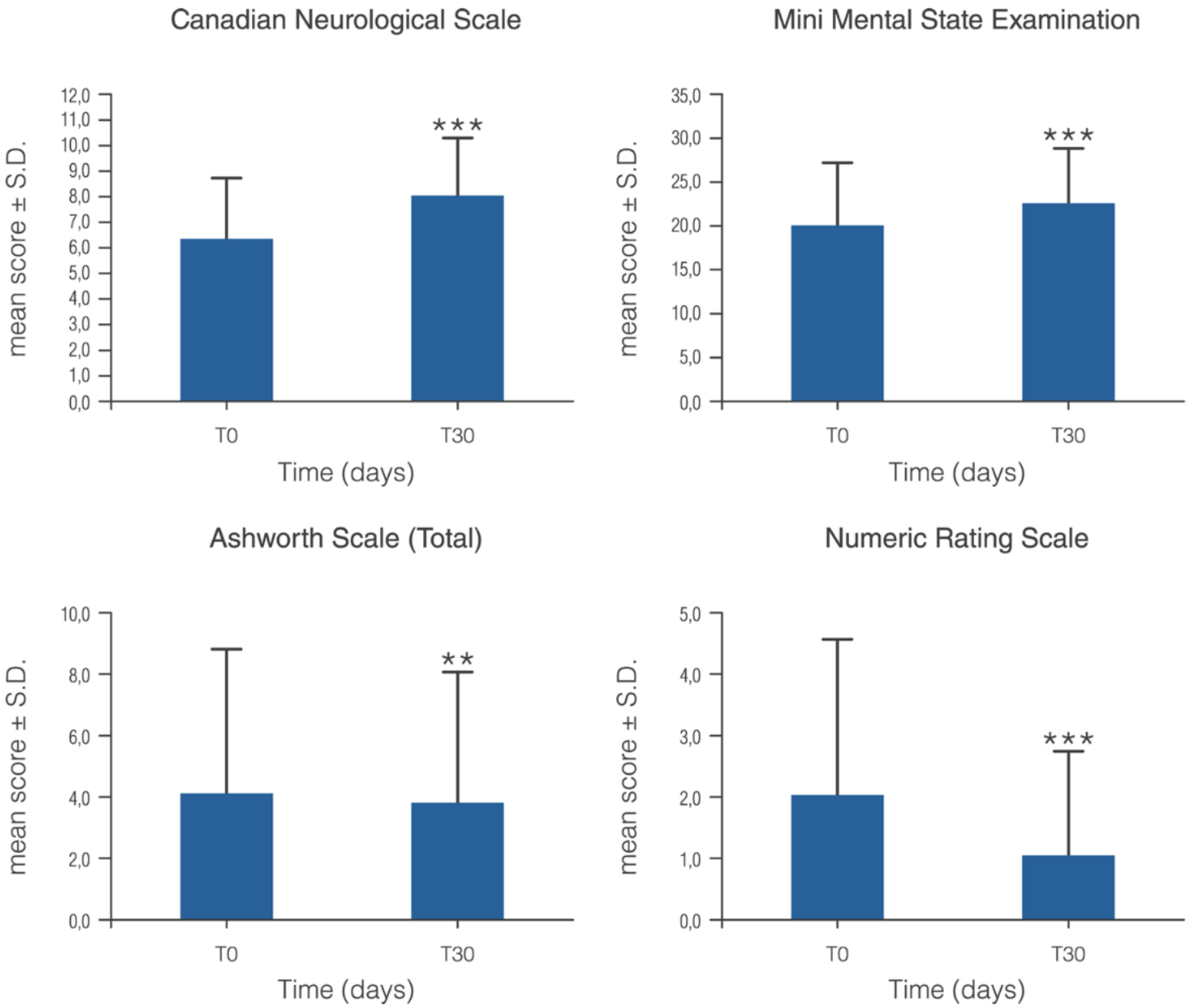
Procedure statistiche: Analisi della varianza (ANOVA) a misure ripetute tra i time-points dello Studio;

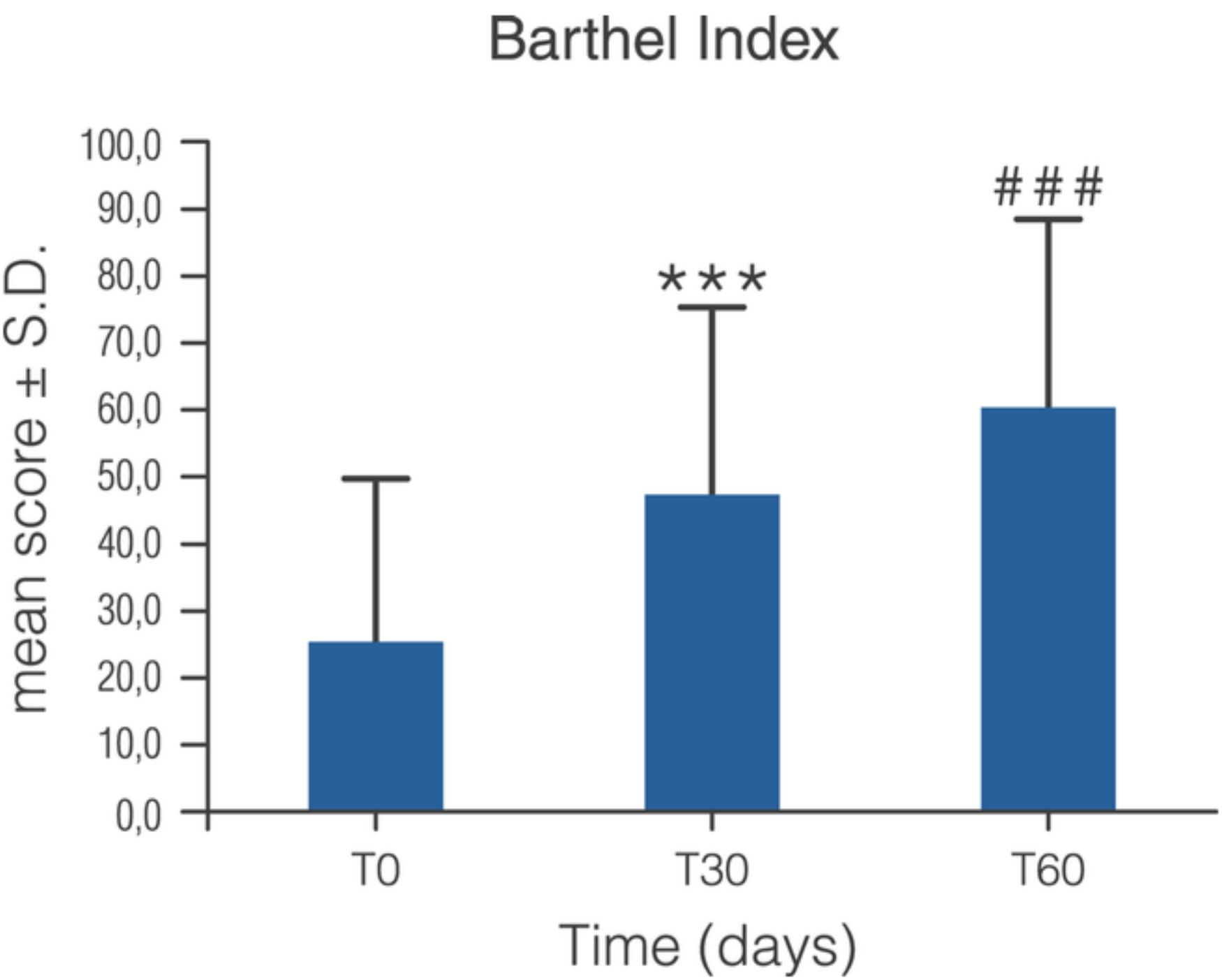
Per CNS, AS, BI confronto con le medie riportate in letteratura (T-test)[1,2].

1 *Fornasari P., Montanari S. Utilizzo del protocollo di minima per l'ictus cerebrale (PMIC) per la valutazione dell'outcome e del percorso riabilitativo di un gruppo di pazienti ricoverati presso l'U.O. di medicina riabilitativa di Cesenatico. Eur Med Phys. 2008; 44 (Suppl.1 to No.3*).

2 *Cisari C . Direttore S.C. Medicina Fisica e Riabilitativa dell'AOU "Maggiore della Carità" Novara . Studio di fattori prognostici di outcome di stroke acuto, riabilitazione ospedaliera e territoriale per ottimizzare il percorso clinico-assistenziale*.

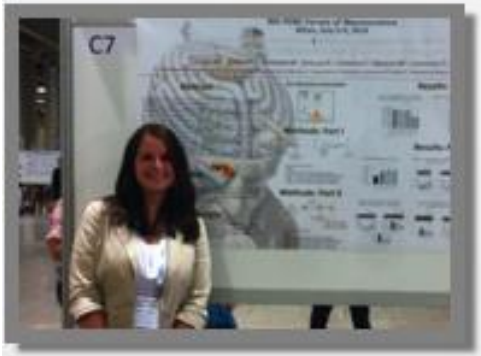
Neurological scoring for stroke patients treated with Glialia®

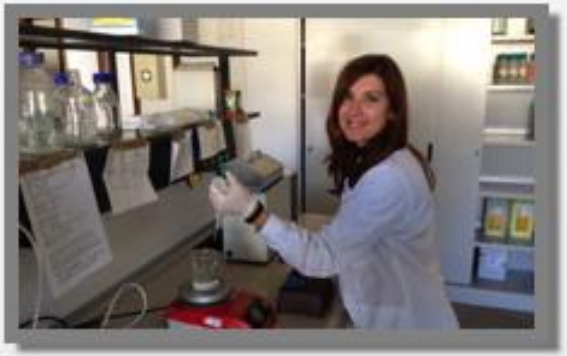



Barthel Index
100,0
90,0
80,0
70,0
60,0
50,0
40,0
30,0
20,0
10,0
0,0
mean score ± S.D.
***
###
T0
T30
T60
Time (days)

Take home message…

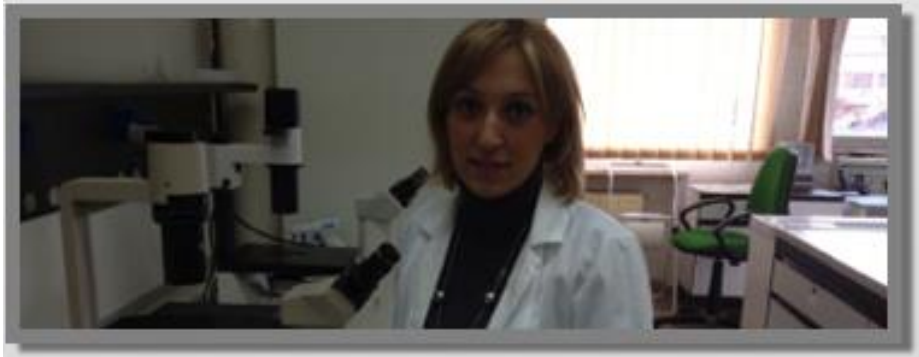
The results obtained support the view that PEALut exerts a significant neuroprotective effect being a useful novel therapeutic strategy in neuroinflammatory diseases

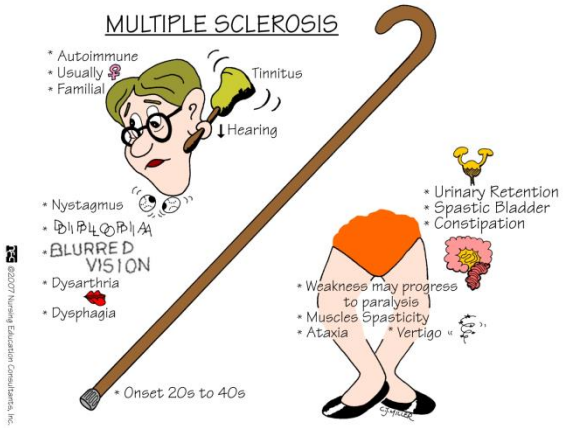

C7

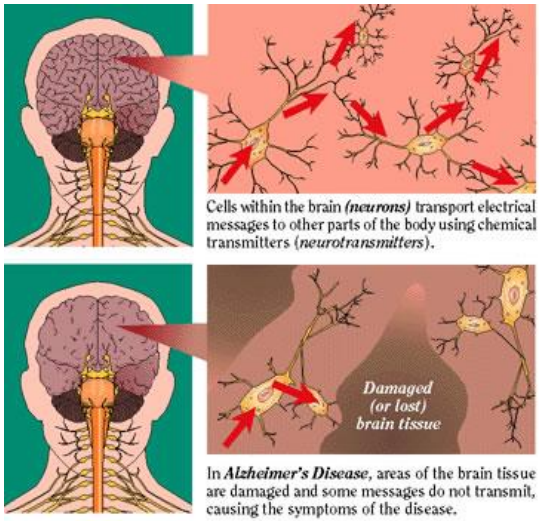


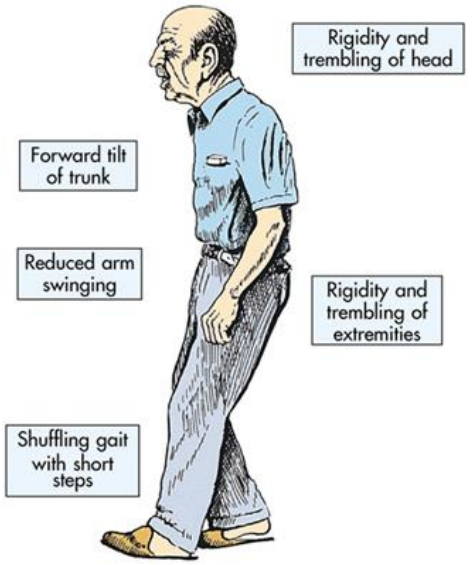
P7

BOSTON

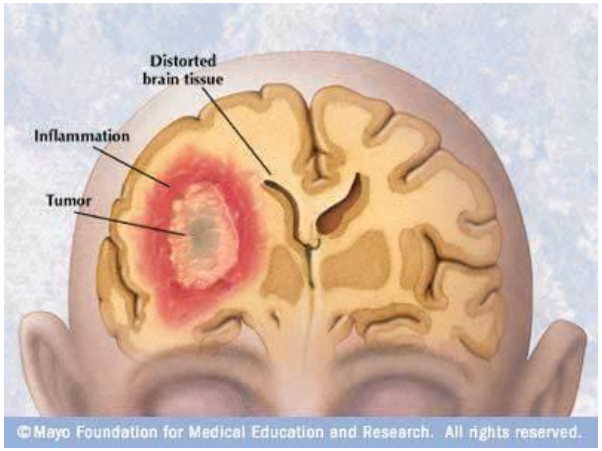


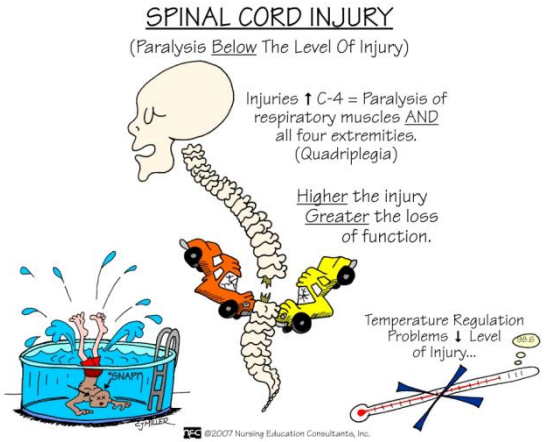


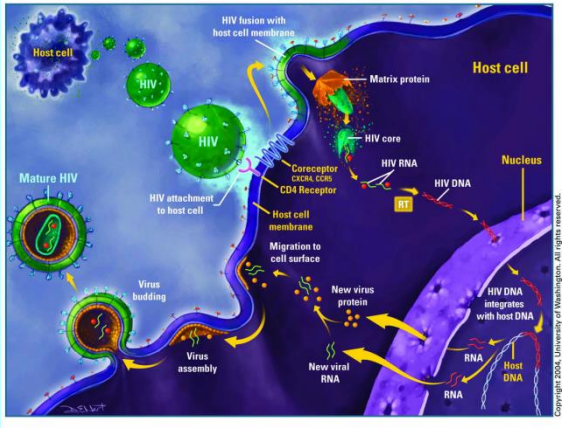




Neuroinflammation







PHYSIOLOGY

[55,57,62,63] INNATE IMMUNITY
Host defence
(control of bacterial,
viral, parasitic infection)

[57, 62] ACQUIRED (ADAPTIVE) IMMUNITY

[60] HAIR FOLLICLE CYCLING

[60] BONE REMODELING

[57] PREGNANCY

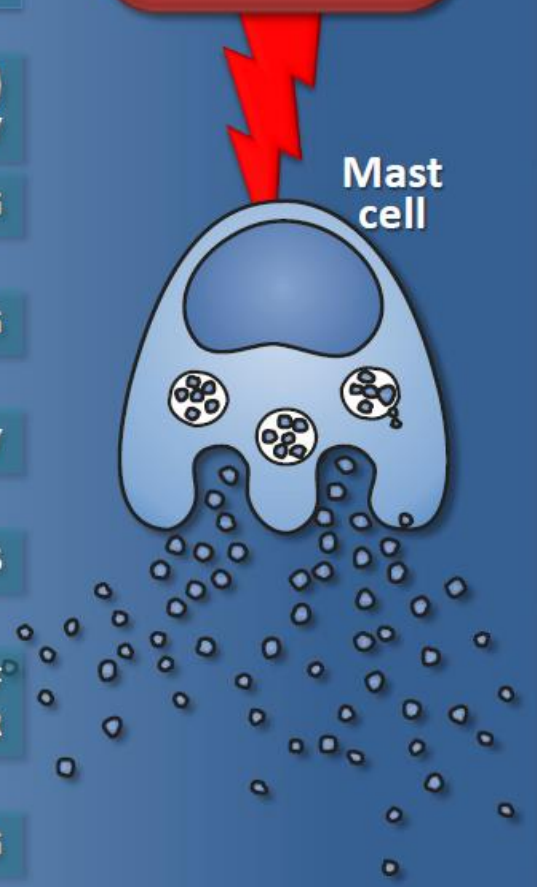
[65] INTESTINAL HOMEOSTASIS

[71] MODULATION OF ANXIETY-LIKE BEHAVIOR

[58] WOUND HEALING

[58, 61, 63] ANGIOGENESIS

Neurogenic
Immunogenic
Physical
Chemical
Microbial
STIMULI



DISEASE

ALLERGIC DISEASES[56]
- Asthma[56,74,76]
- Allergic rhinitis[56]
- Atopic derm.[56,70,72,74-76]
- Allergic contact derm.[72]
- Food allergy[60,65]
- Allergic eye d.[56]

NEURO-INFLAMM. DISEASES[59,74]
- Periodontitis[76]
- OA & rheumatic d.[66,74-76]
- Nonallergic GI d.[65,67,74-76]
- Inflam. kidney d.[69]
- Interstitial cystitis[74]
- Migraines[74-76]
- Cerebral ischemia and hemorrhage[73]

AUTOIMMUNE DISEASES
- Psoriasis[68,70]
- Autoimmune kidney d.[69]
- Bullous pemphigoid[70,72]
- Multiple sclerosis[74,75]

PERIPHERAL NOCICEPTOR SENSITIZATION (PAIN)[64,66]

FASEB J. 2012 Aug;26(8):3103-17. doi: 10.1096/fj.11-197194. Epub 2012 Apr 19.

Microglia and mast cells: two tracks on the road to neuroinflammation.

Skaper SD, Giusti P, Facci L.

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