NANOTECHNOLOGIES POUR L'ADRESSAGE DE MEDICAMENTS AU NIVEAU CEREBRAL

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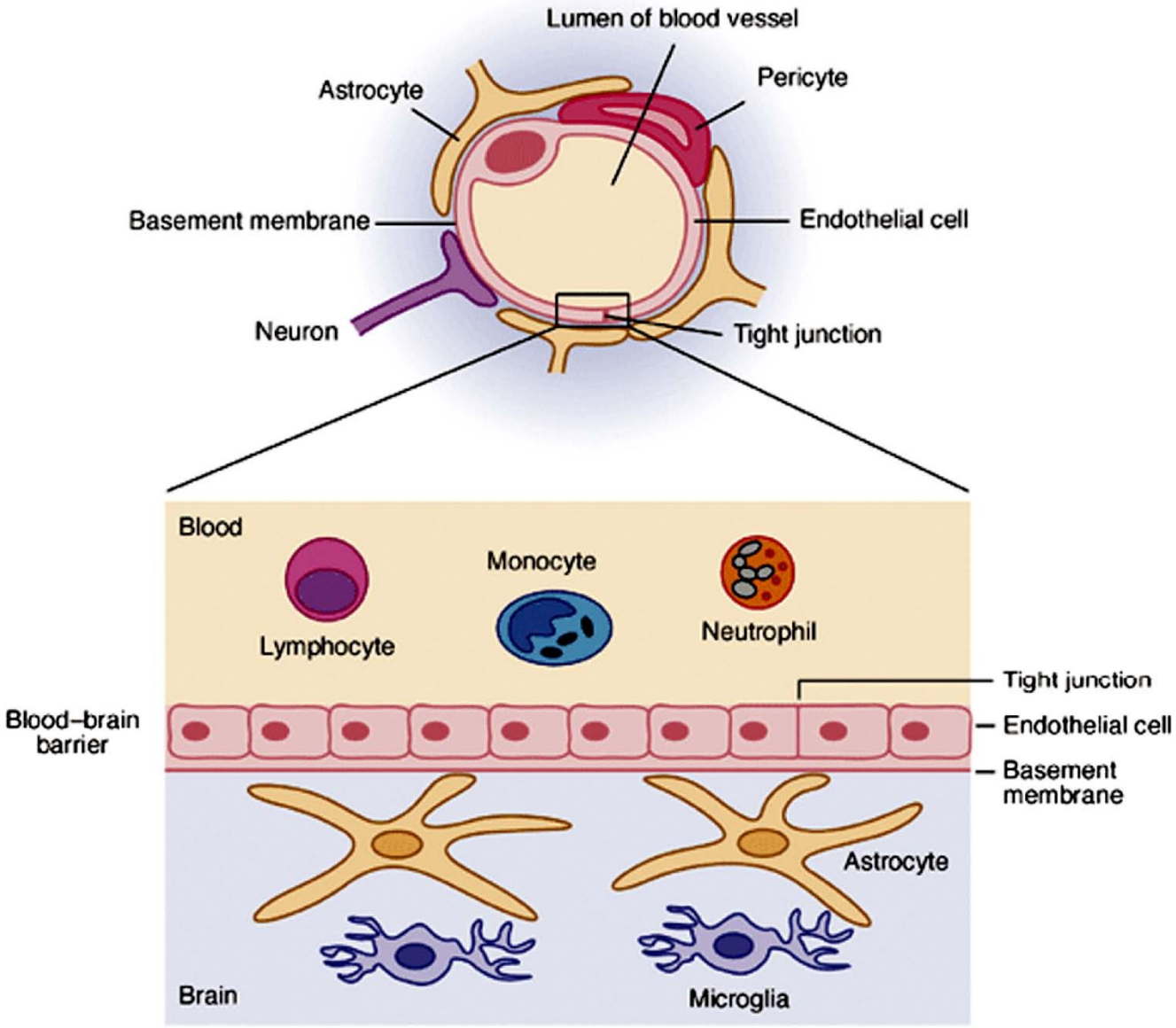
2009-2010

HISTOLOGY OF BBB



MECHANISMS OF TRANSPORT THROUGH THE BBB

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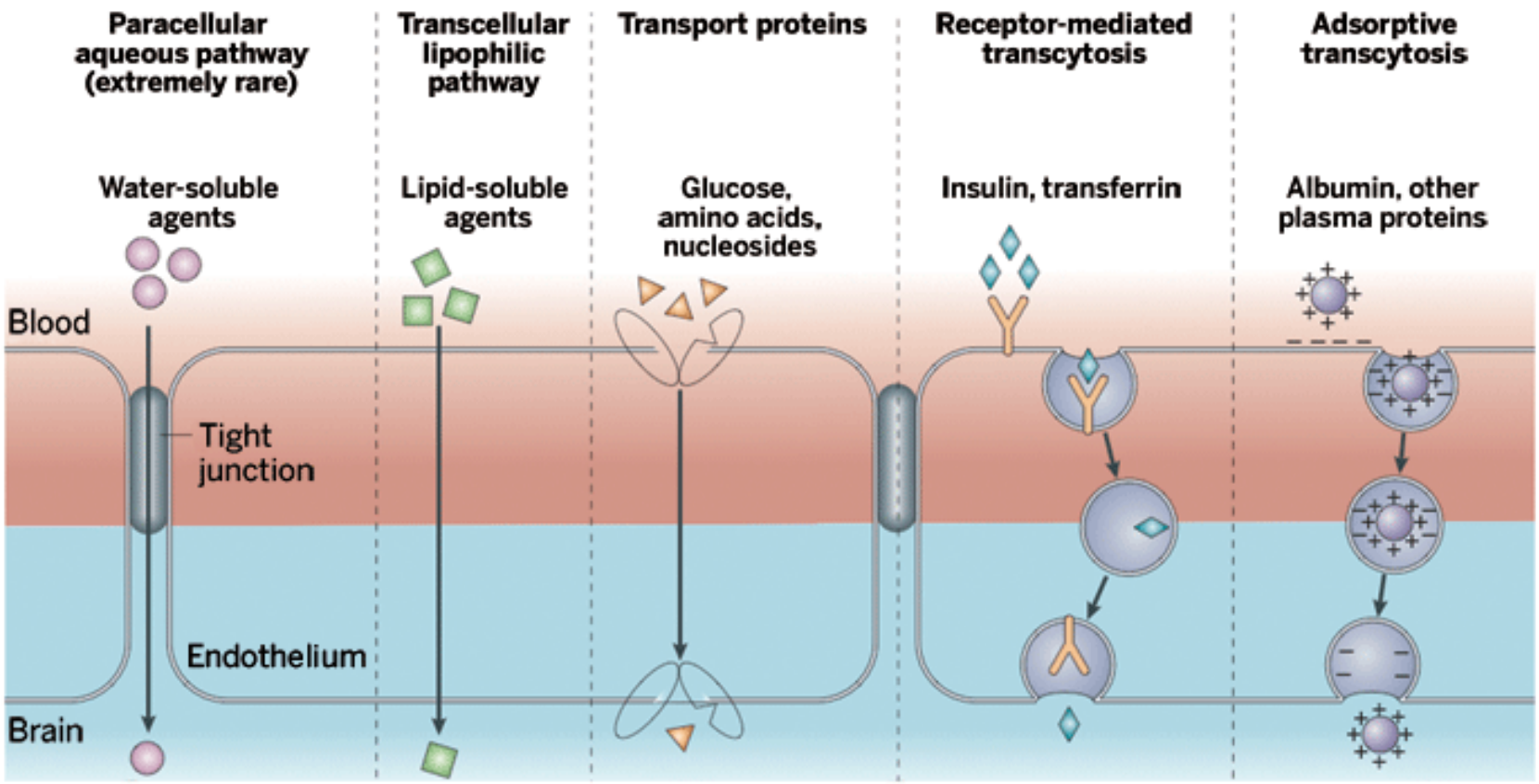
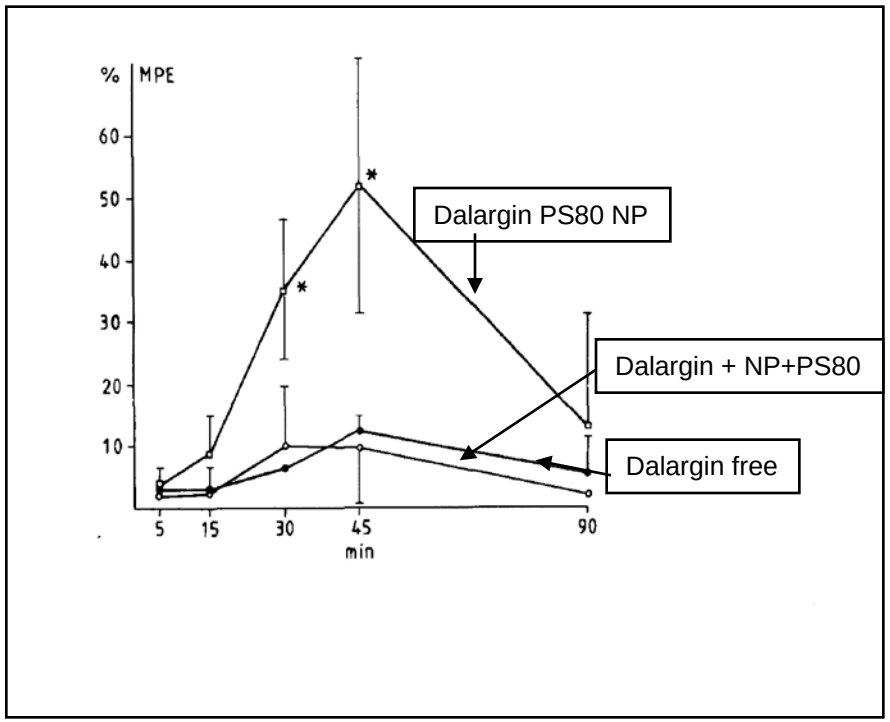


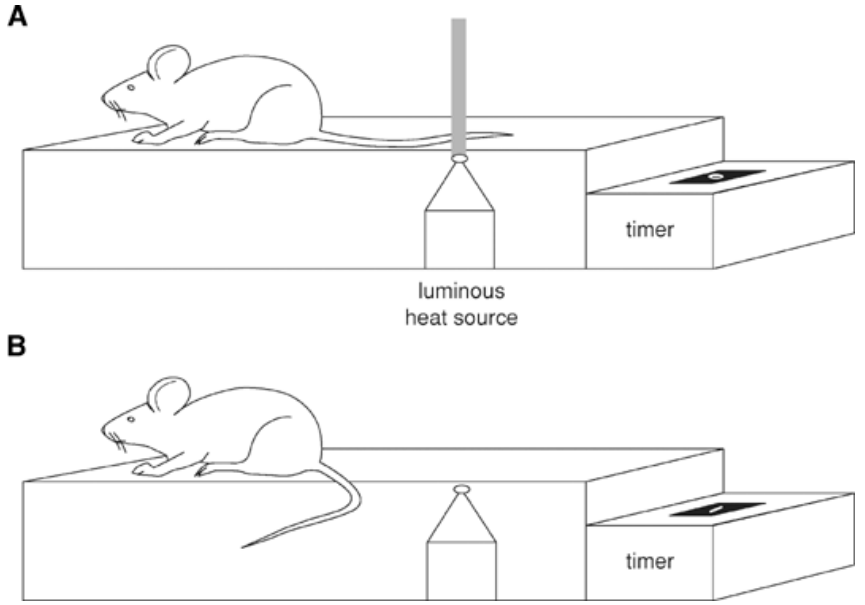
LES NANOPARTICULES PEUVENT-ELLES SERVIR DE « CARGO » POUR L'ADRESSAGE CEREBRAL?

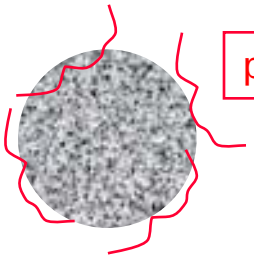
- Nanoparticules recouvertes de polysorbate
- Nanoparticules recouvertes de PEG

Analgesic effect of polysorbate 80-coated and dalargin-loaded nanoparticles (i.v. injection)

J. Kreuter et al., Brain Research, 674, 171-174, 1995
J. Kreuter et al., J. Controlled Rel., 49, 81-87, 1997







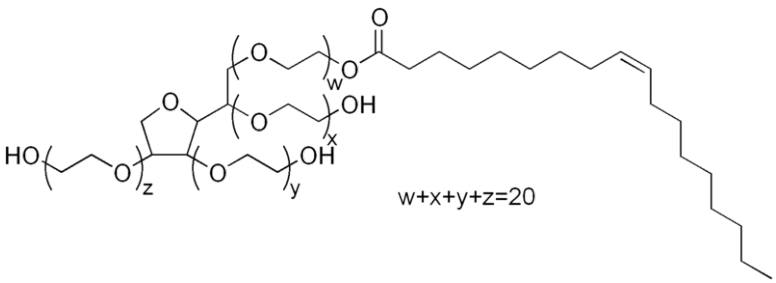
polysorbate

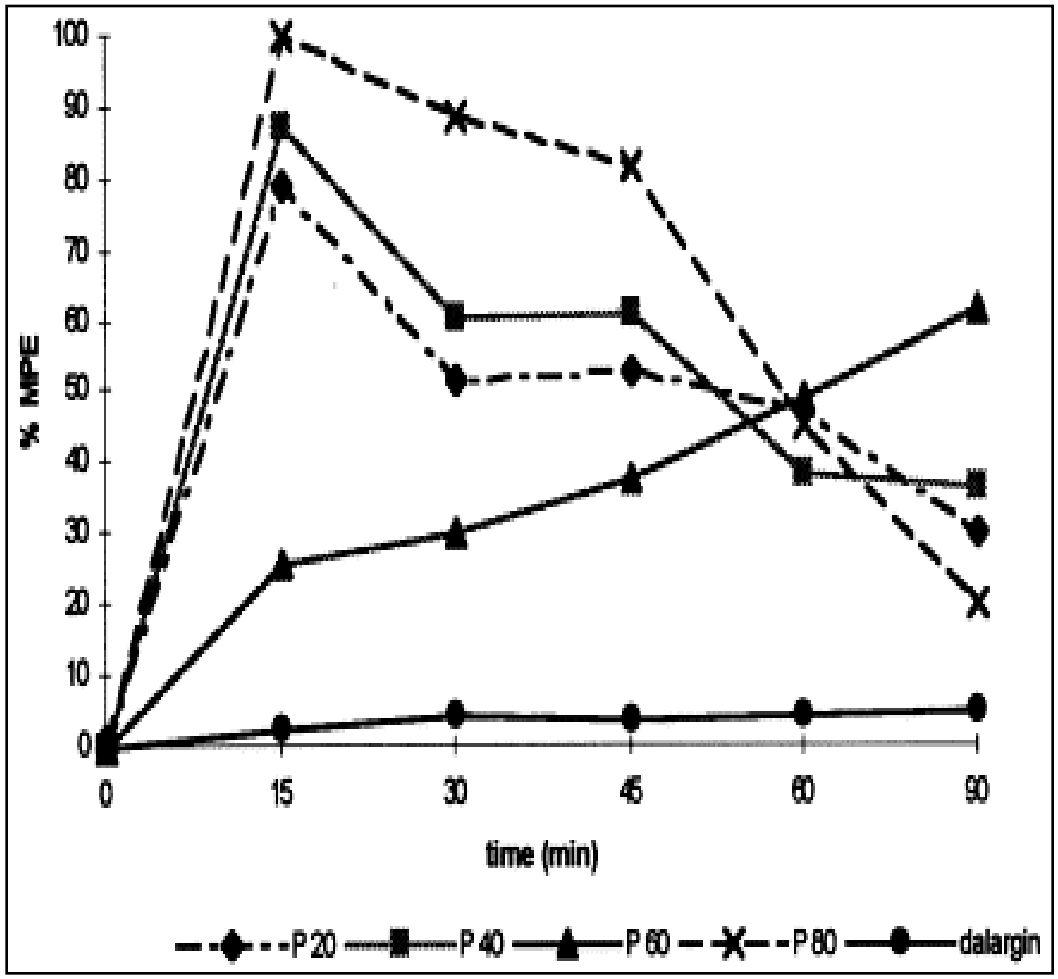
% MPE= $\frac{\text{time post drug latency- time pre drug latency}}{\text{cut off time- time pre drug latency}}$

Cut off time= 10sec before tissue damage

Analgesic effect of polysorbate 20, 40, 60 and 80-coated and dalargin-loaded nanoparticles (i.v. injection)

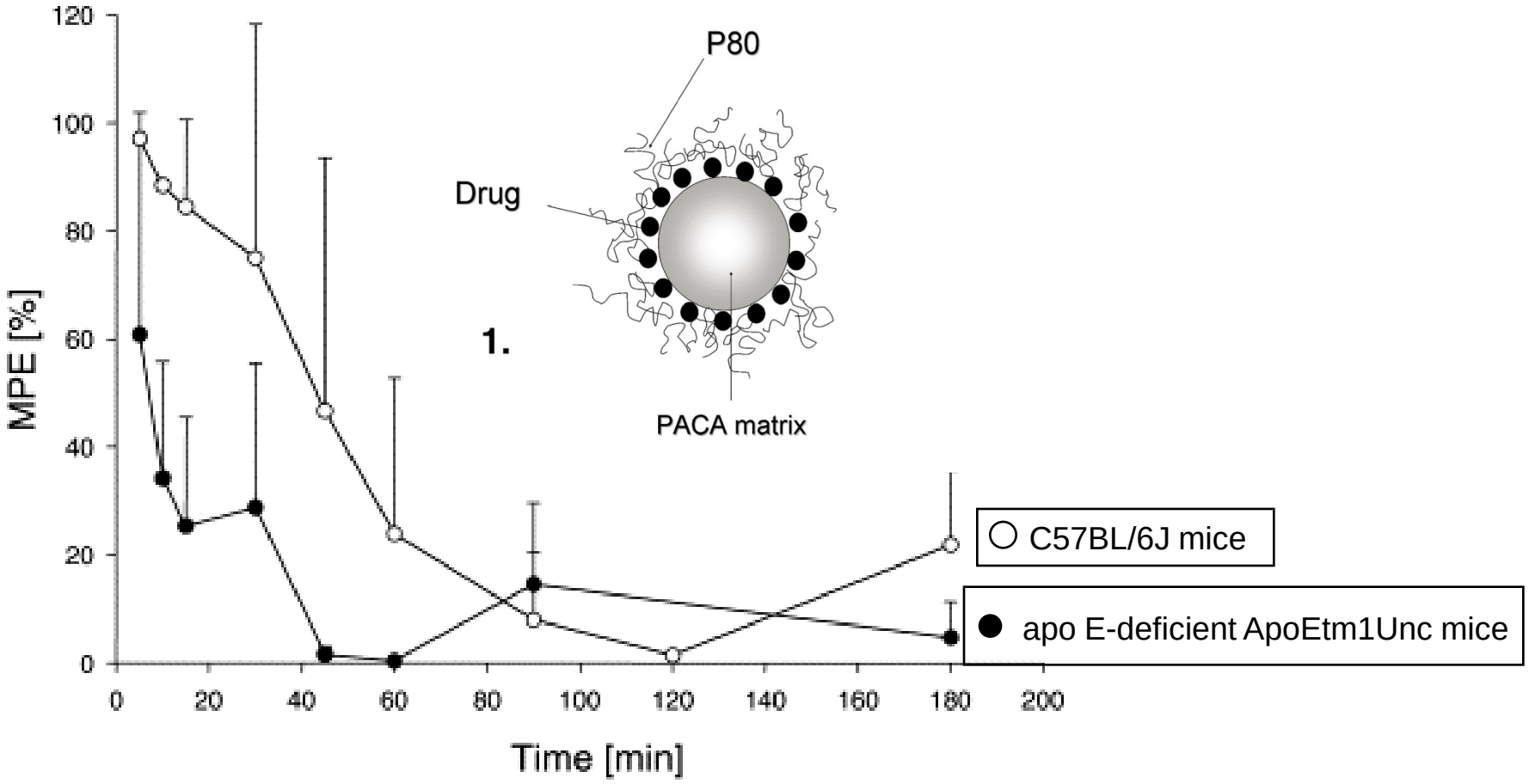
J. Kreuter et al., J. Controlled Rel., 49, 81-87, 1997





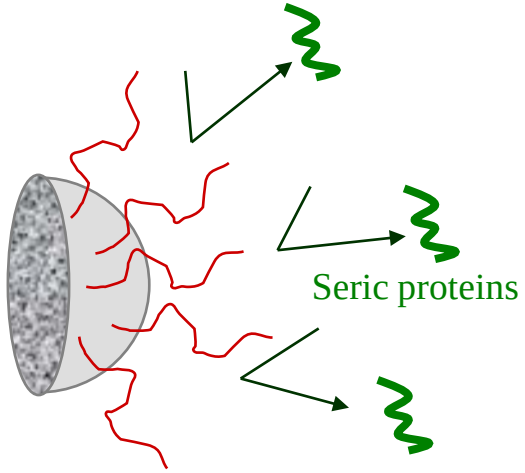
COMPARATIVE ANTINOCICEPTIVE EFFECT (TAIL FLICK TEST) ON WILD TYPE AND APOE DEFICIENT MICE

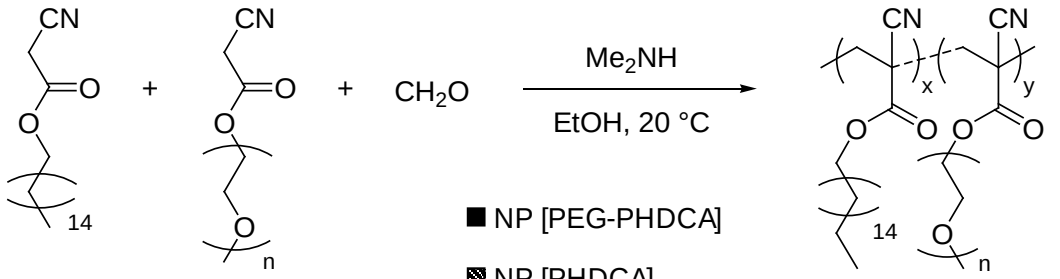
J. Kreuter, Adv. Drug Deliv. Rev., 47, 65-81, 2001



CAPTURE CEREBRALE DES NANOPARTICULES DE PEG-PACA

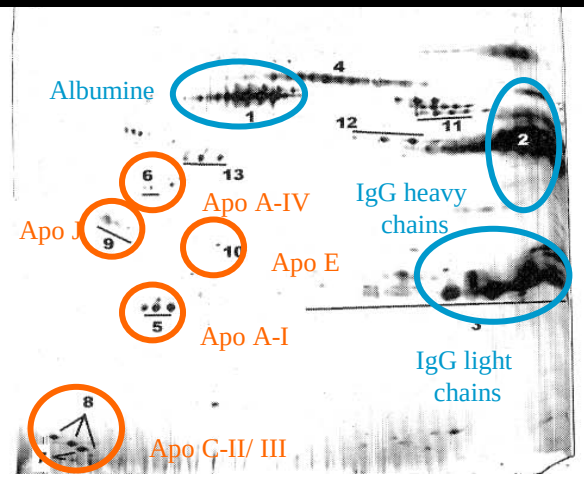
Perrachia et al., J. Control. Rel, **60**, 121-128 (1999)
Perrachia et al., Biomaterials, **20**, 1269-1275 (1999)



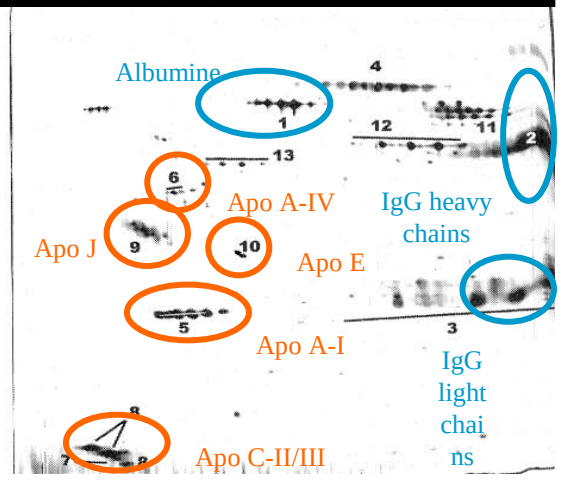


- NP [PEG-PHDCA]
- NP [PHDCA]
- NP [PHDCA]-Polox 908
- NP [PHDCA]-P80

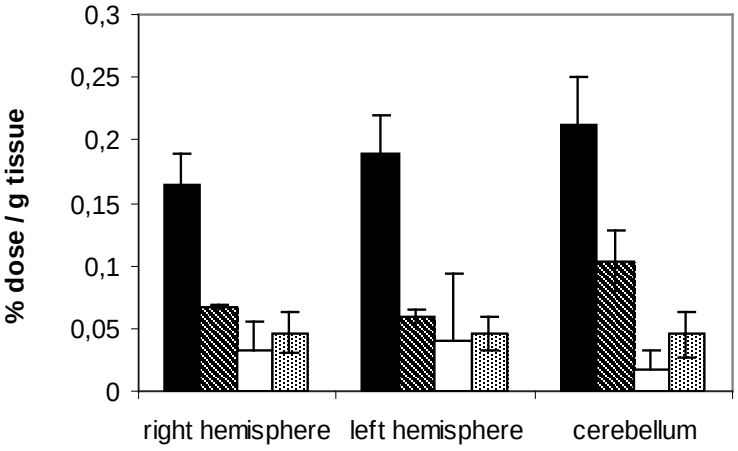
PHDCA



PEG-PHDCA

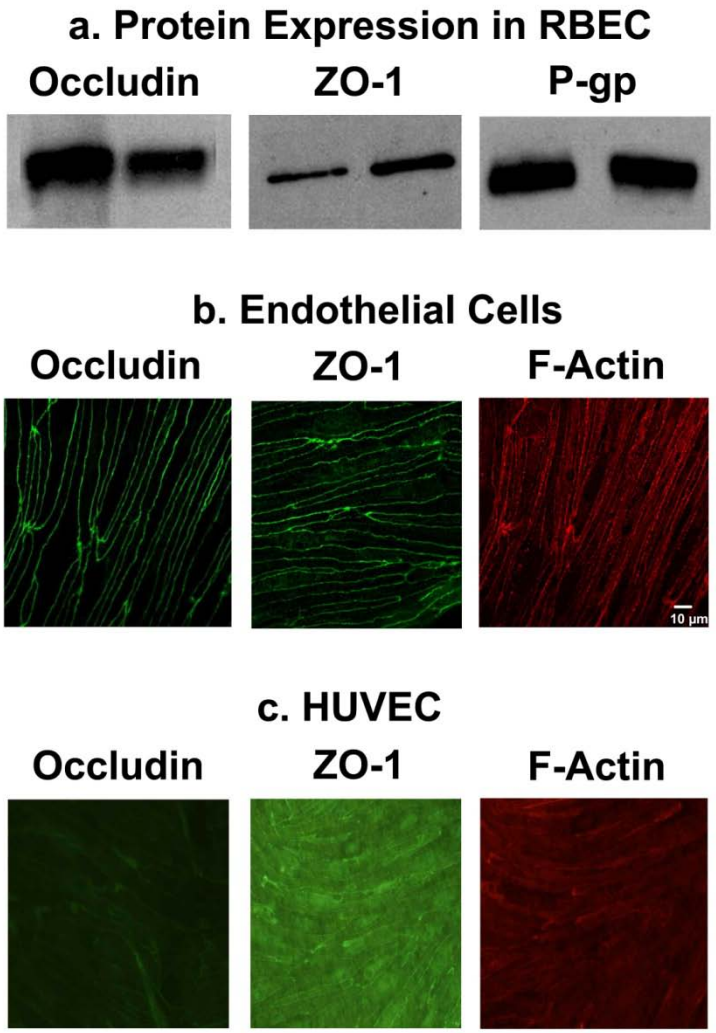


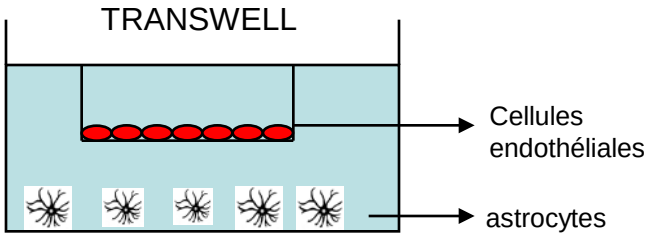
a) Mice, 1h

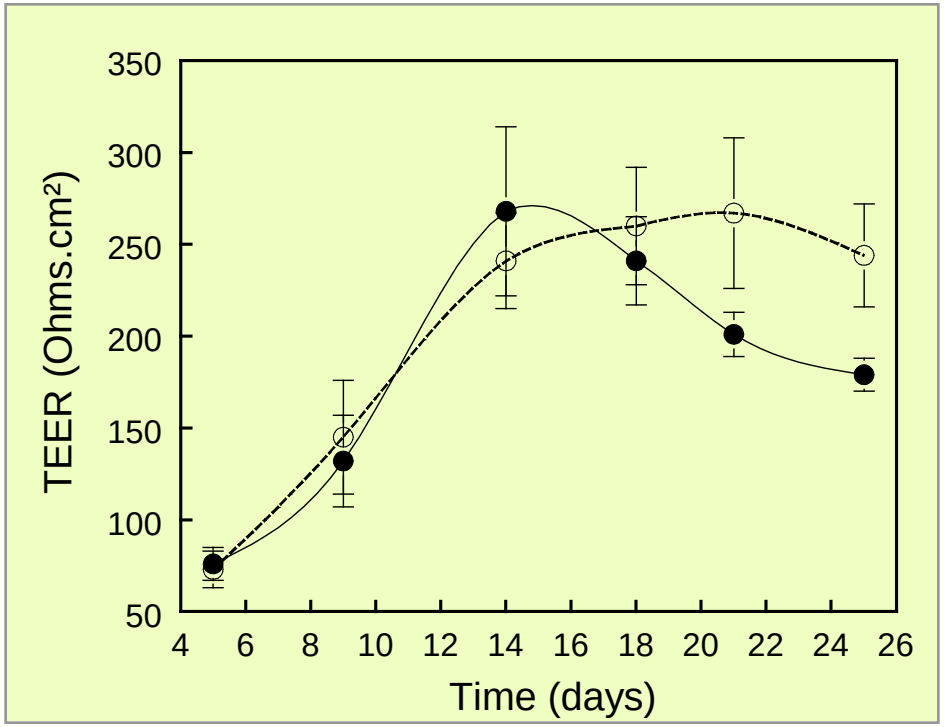


FUNCTIONALITY OF THE IN VITRO MODEL OF RAT BBB

E. Garcia-Garcia et al., CMLS, 62, 1400-1408 (2005)

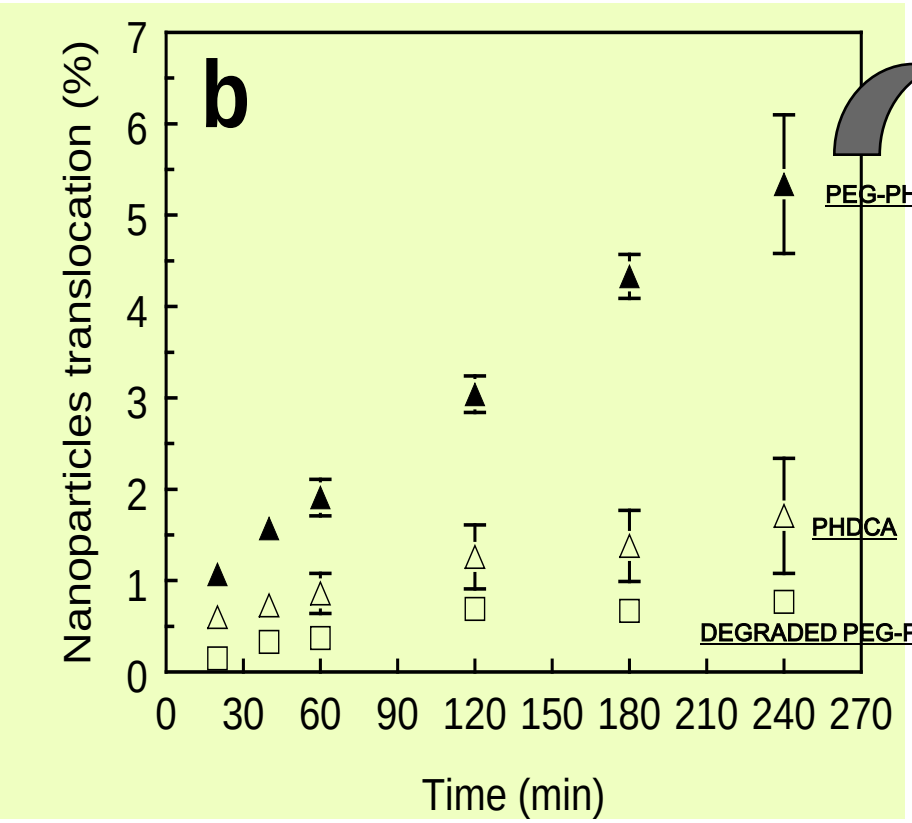


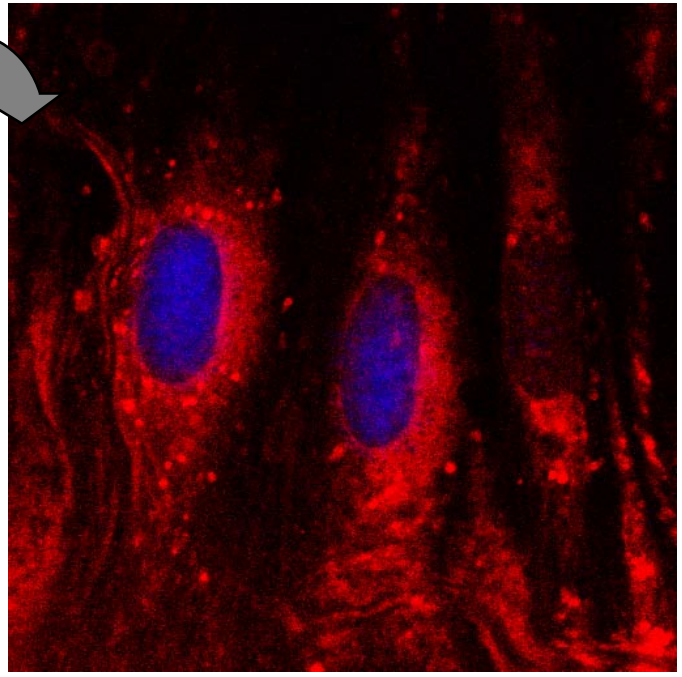


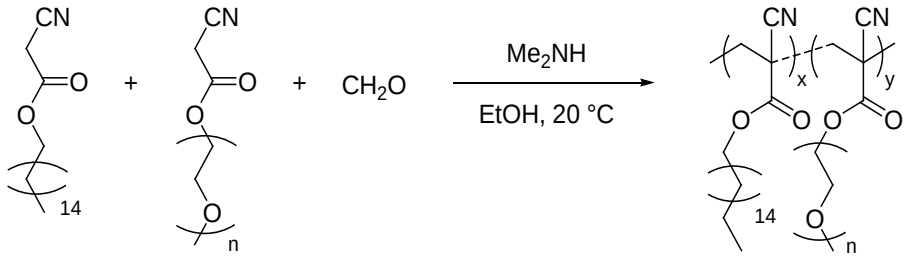


ANOPARTICLES TRANSLOCATION THROUGH EXPERIMENTAL RAT BBB

E. Garcia-Garcia et al., CMLS, 62, 1400-1408 (2005)

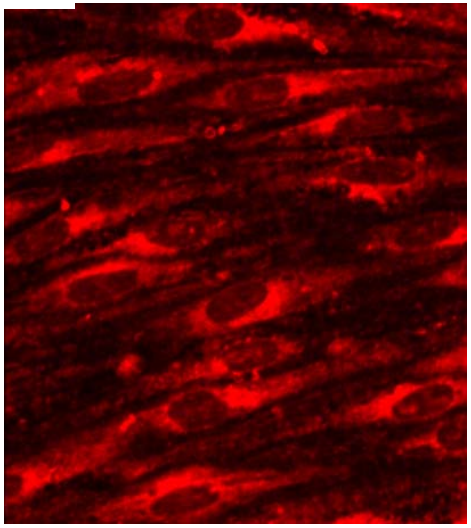


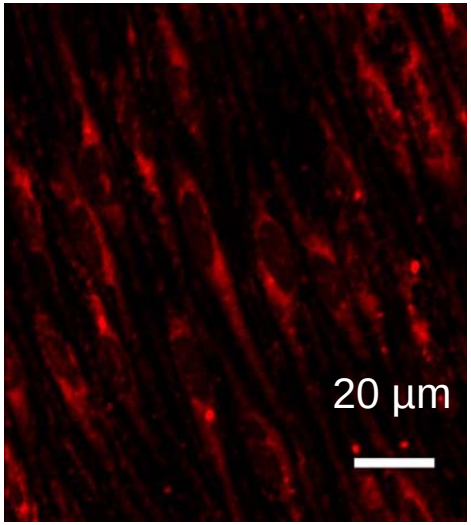


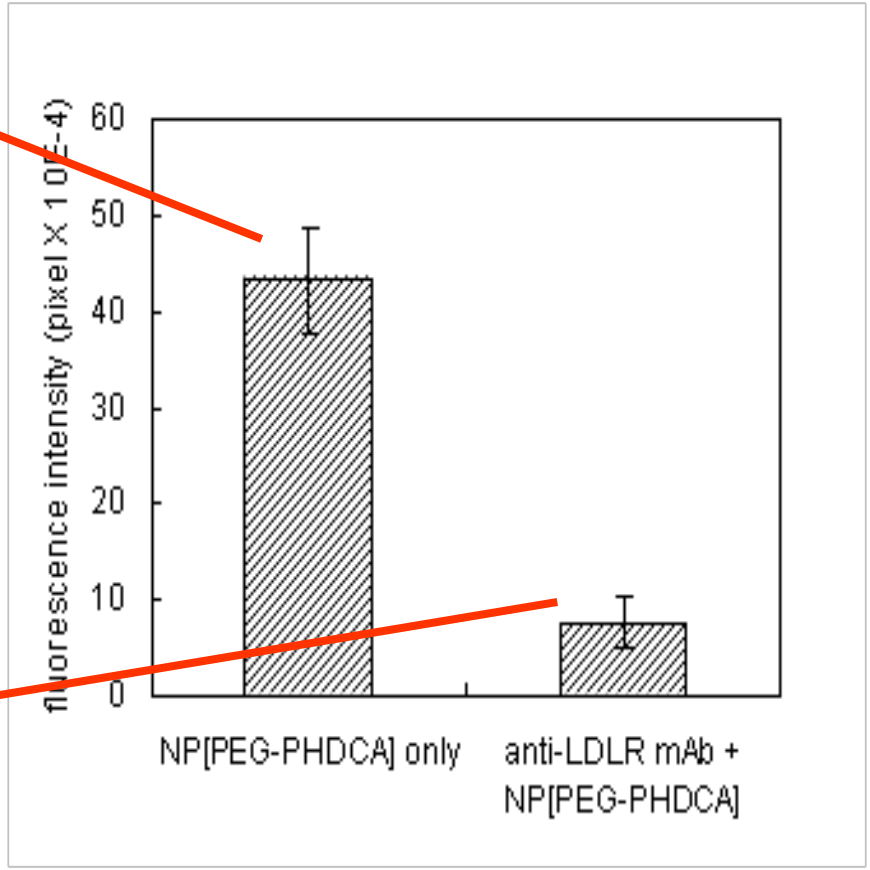


… AND LDL RECEPTOR IS INVOLVED

Kim HR et al Cell Mol Life Sci., 64, 356-364 (2007)

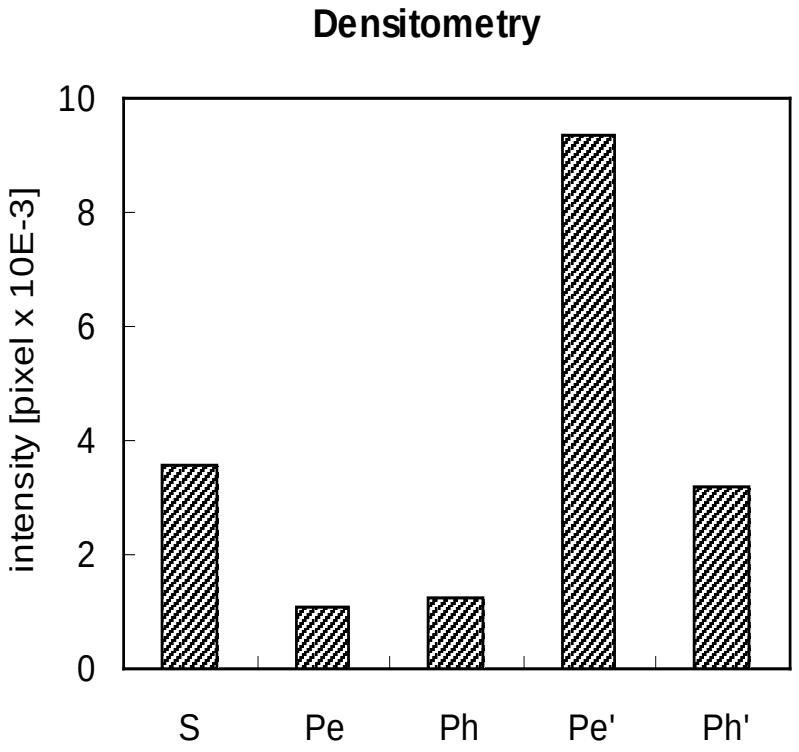


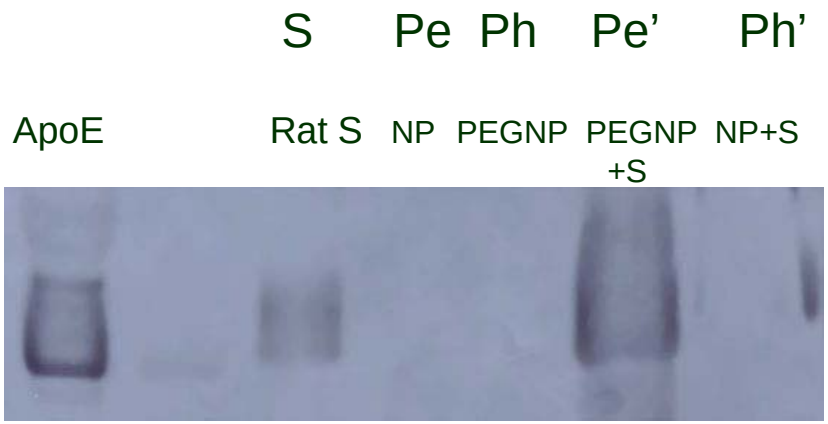


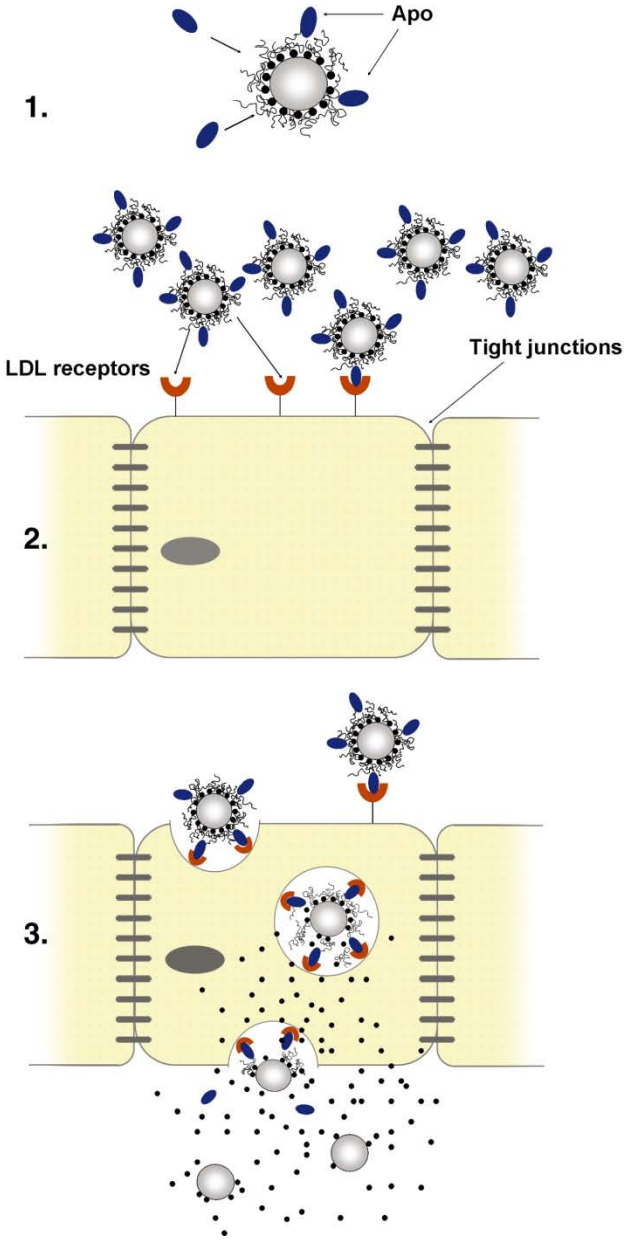


ADSORPTION OF RAT APOE ONTO PEGYLATED AND NON PEGYLATED NANOPARTICLES

Kim HR et al., Biomacromolecules. 8:793-9 (2007)





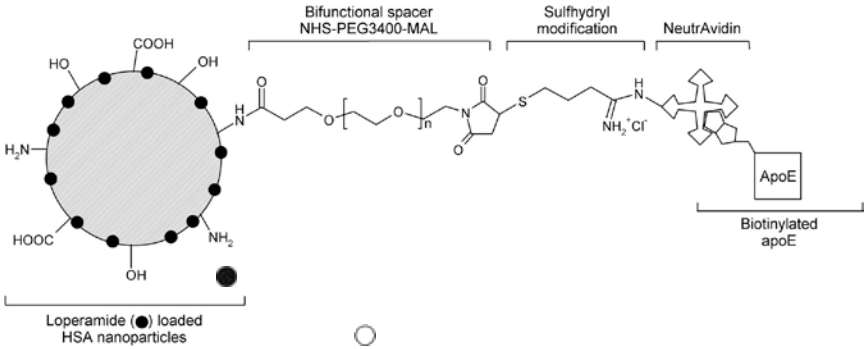


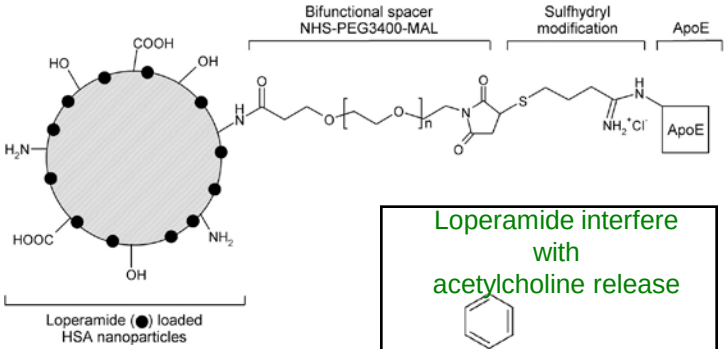
BRAIN TRANSLOCATION OF NANOPARTICLES THROUGH THE LDL RECEPTOR PATHWAY

ApoE DECORATED NANOPARTICLES FOR BRAIN DELIVERY

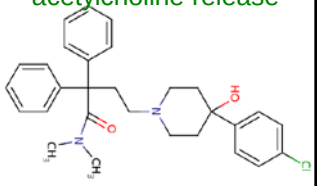
LOPERAMIDE LOADED APO E FUNCTIONALIZED HUMAN SERUM ALBUMINE NANOPARTICLES

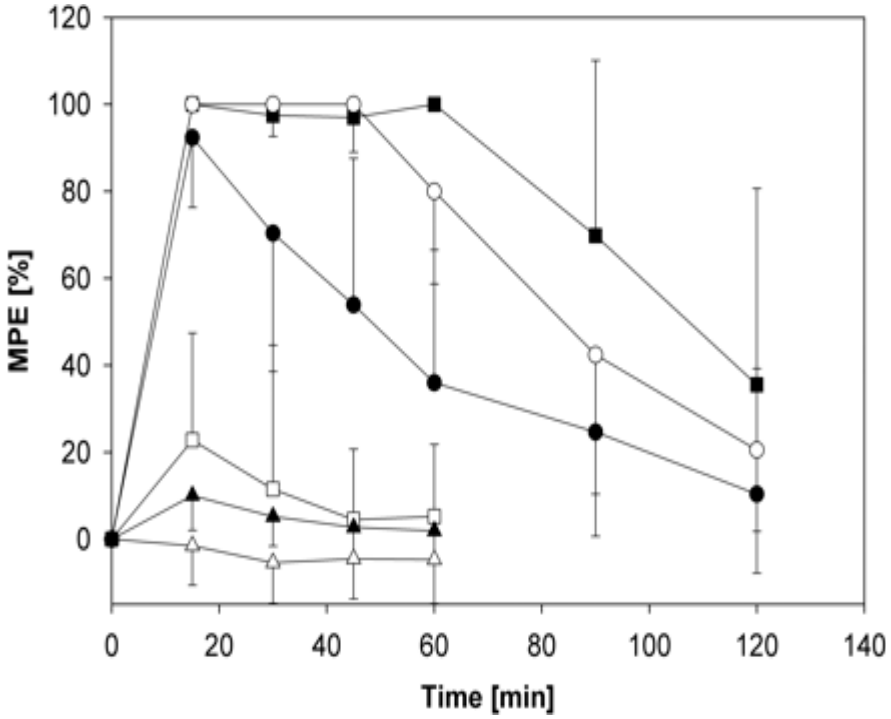
Michaelis K et al, JPET, 317, 1246-1253, 2006





Loperamide interfere with acetylcholine release

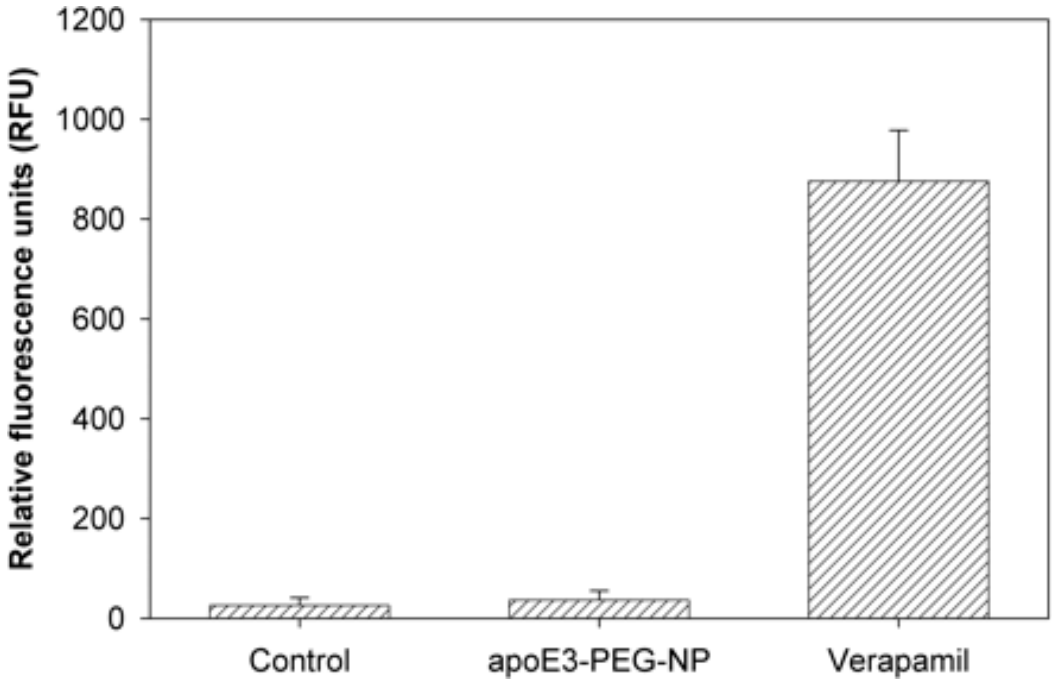




■ apolipoprotein E HSA-NP with 7.0 mg/kg loperamide; ● apolipoprotein E HSA-NP with 4.0 mg/kg loperamide; □ HSA-NP with loperamide (no apolipoprotein E attached); ▲ loperamide solution; △ apolipoprotein E HSA-NP without loperamide; and ◯ polysorbate 80-coated HSA-NP with 7.0 mg/kg loperamide.

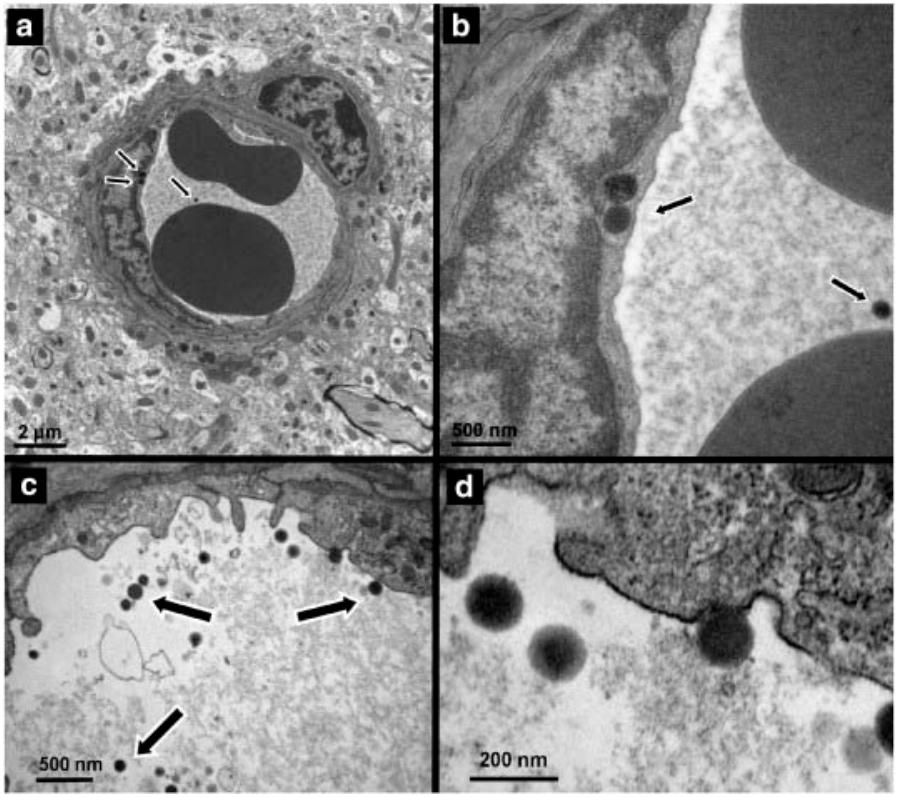
APOE NANOPARTICLES DO NOT INHIBIT PgP FUNCTION

Michaelis K et al, JPET, 317, 1246-1253, 2006

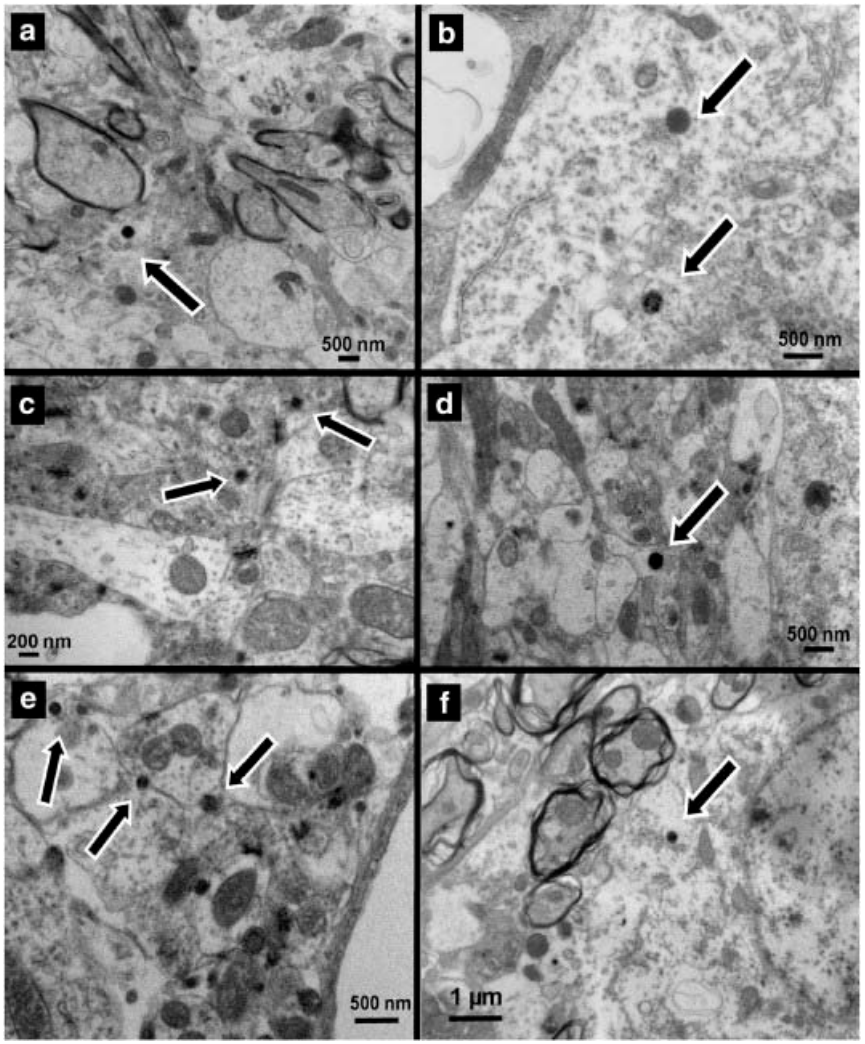


BRAIN LOCALIZATION OF APOE DECORATED ALBUMINE NANOPARTICLES

Anja Zensi et al., J Control Rel, 2009



Cortex(a, b) and hippocampus (c,d) region of SV 129 mice after the injection of Apo E-modified nanoparticles



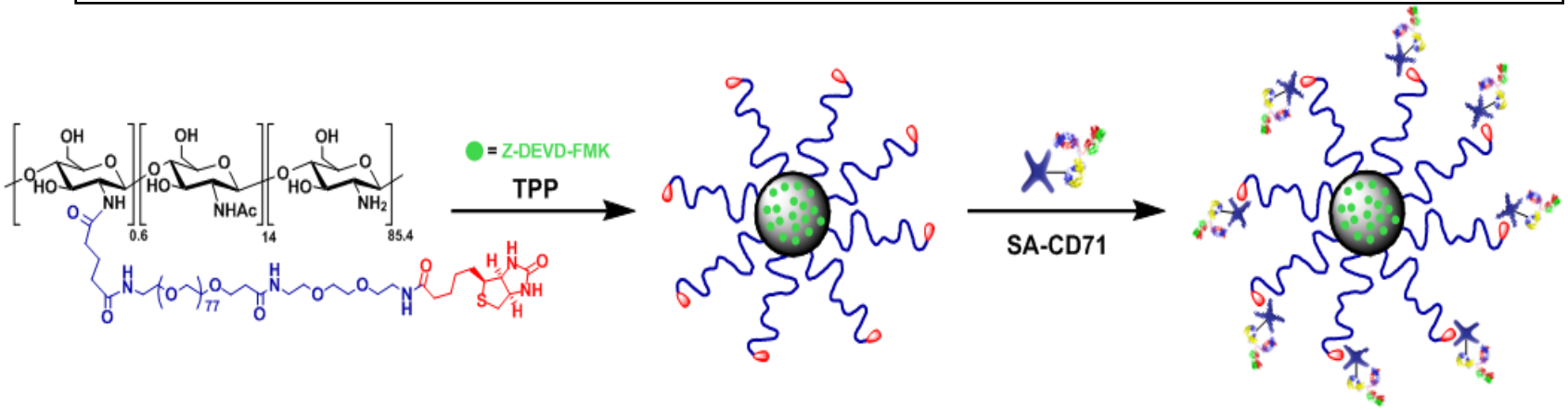
Nanoparticles could be found in all investigated brain regions

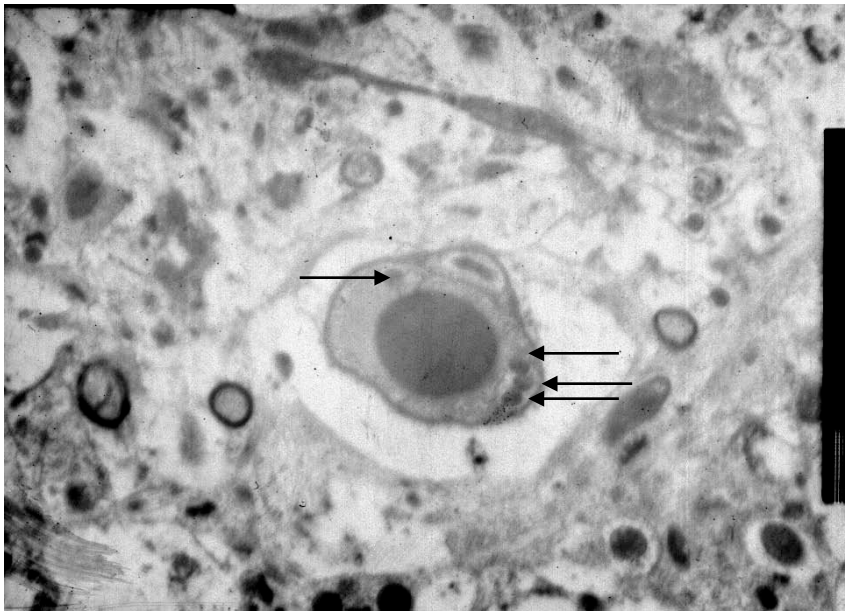
ANTITRANSFERRIN mAb DECORATED NANOPARTICLES FOR BRAIN DELIVERY

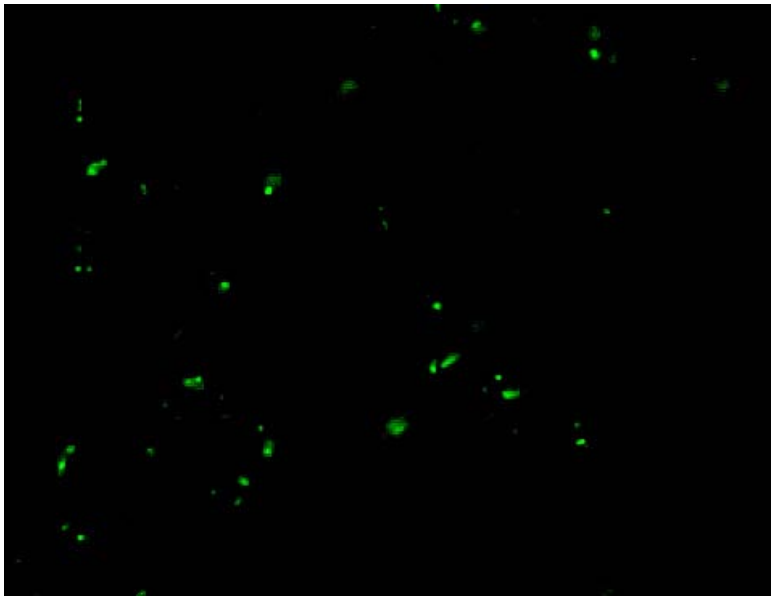
X-26 PEGylated AND ADRESSED CHITOSAN NANOPARTICLES FOR BRAIN DELIVERY OF Z-DEVD-FMK

Y. Aktaş et al. , Bioconj. Chem., 16, 1503-1511 (2005)

N-benzyloxycarbonyl-Asp(OMe)-Glu(OMe)-Val-Asp(OMe)-fluoromethyl ketone (Z-DEVD-FMK)

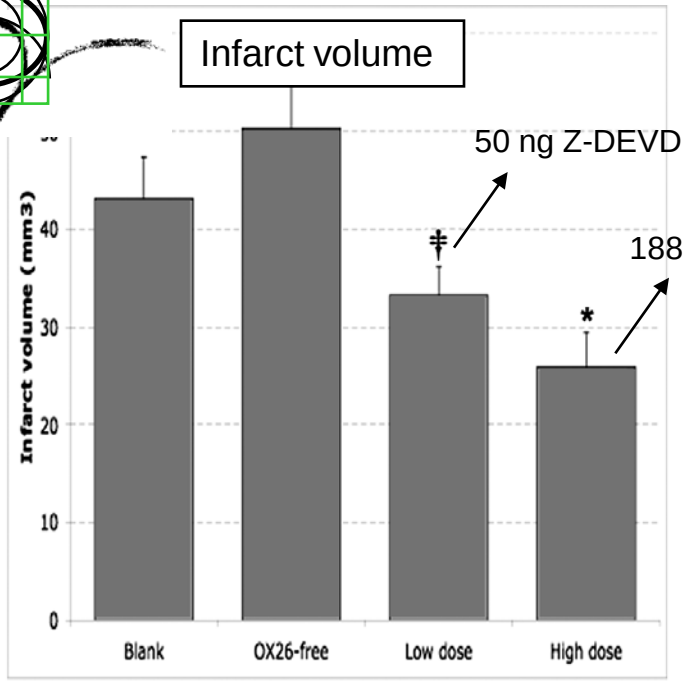




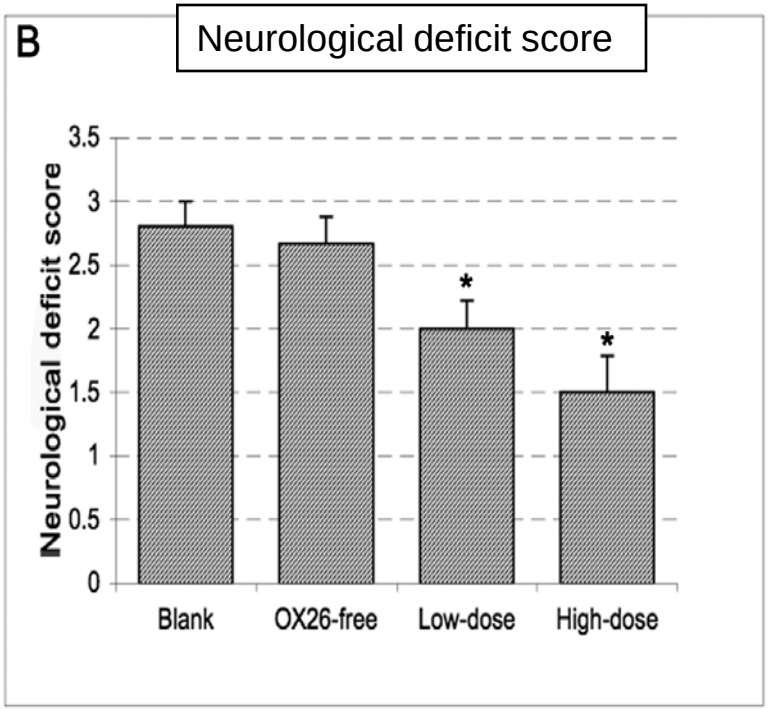


BRAIN DELIVERY BY OX 26 CHITOSAN NANOPARTICLES OF PEPTIDE CASPASE INHIBITOR ZDEVD FMK

Karatas H et al., J Neurosci., 2009



Ischemia induced by a nylon filament inserted into the common carotid artery (20 min + reperfusion 10 min)

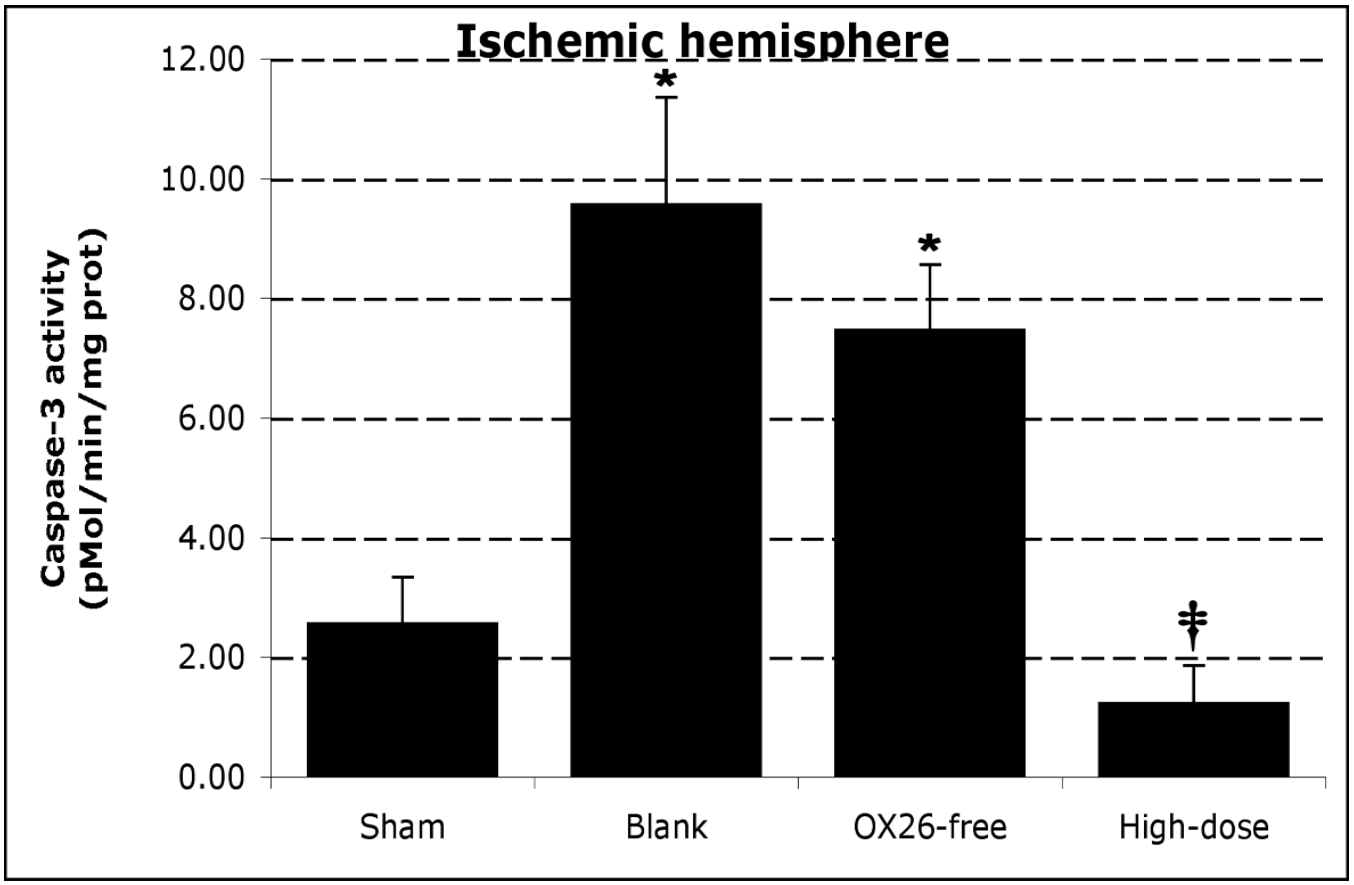


0, no observable neurological deficits (normal); 1, failure to extend left forepaw on lifting the whole body by the tail (mild); 2, circling to the contralateral side (moderate); 3, leaning to the contralateral side at rest or no spontaneous motor activity (severe).

BRAIN DELIVERY BY OX 26 CHITOSAN NANOPARTICLES OF ZDEVD FMK

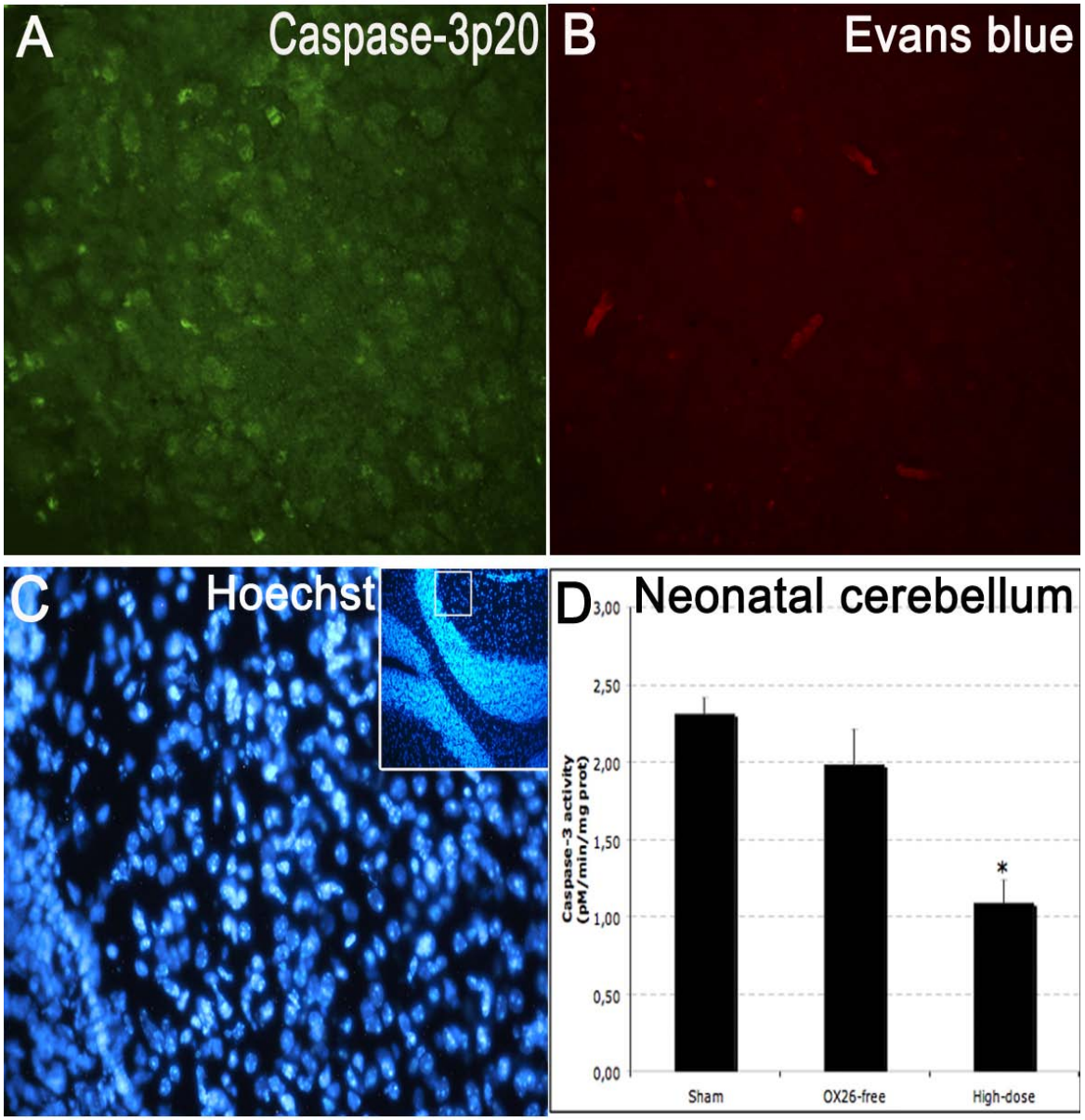
Karatas H et al., J Neurosci., 2009

Caspase-3 activity



NEONATAL MICE CASPASE ACTIVITY IN DEVELOPMENTAL APOPTOSIS

Karatas H et al., J Neurosci., 2009



A Cerebrellar cells show Caspase-3 activity

B Evans blue doesn't leak out of the capillaries showing intact BBB

C Cell nuclei staining

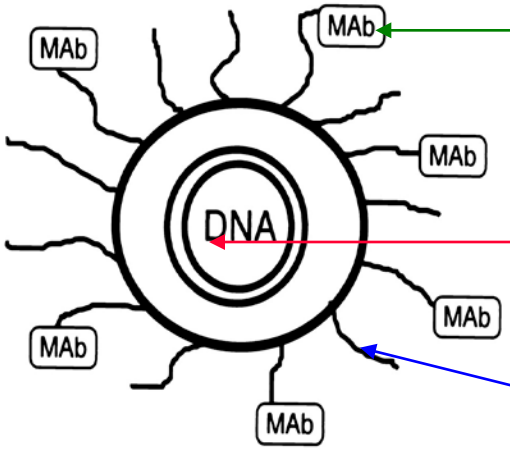
D Treatment with nanoparticles loaded with high-dose Z-DEVD-FMK and conjugated with OX26 antibody (high-dose) significantly inhibited caspase-3 activity

ANTITRANSFERRIN mAb DECORATED LIPOSOMES FOR BRAIN DELIVERY

IMMUNOLIPOSOMES FOR BRAIN TARGETING

Shi N. et.al. PNAS;98:12754-12759, 2001

A

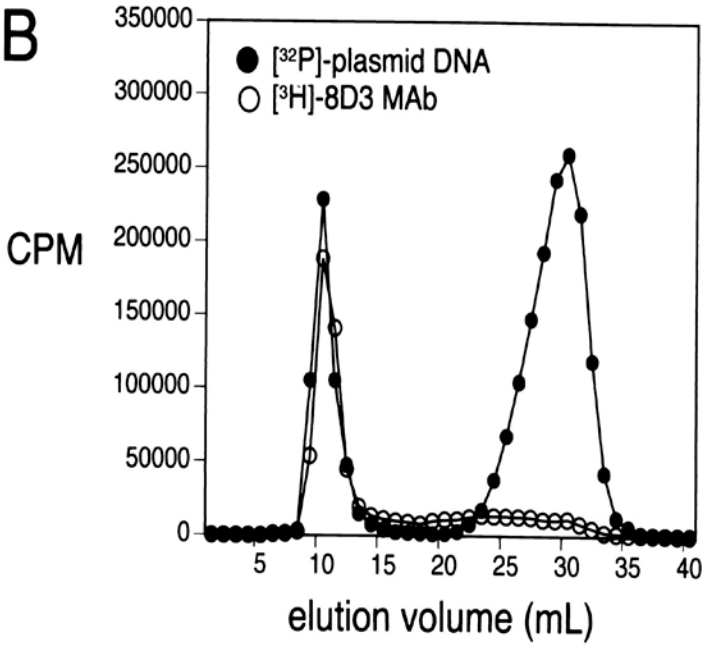


rat 8D3 mAb allow the recognition of the mouse brain Transferrin receptor

human glial fibrillary acidic protein (GFAP) promoter allow The expression in the brain tissue

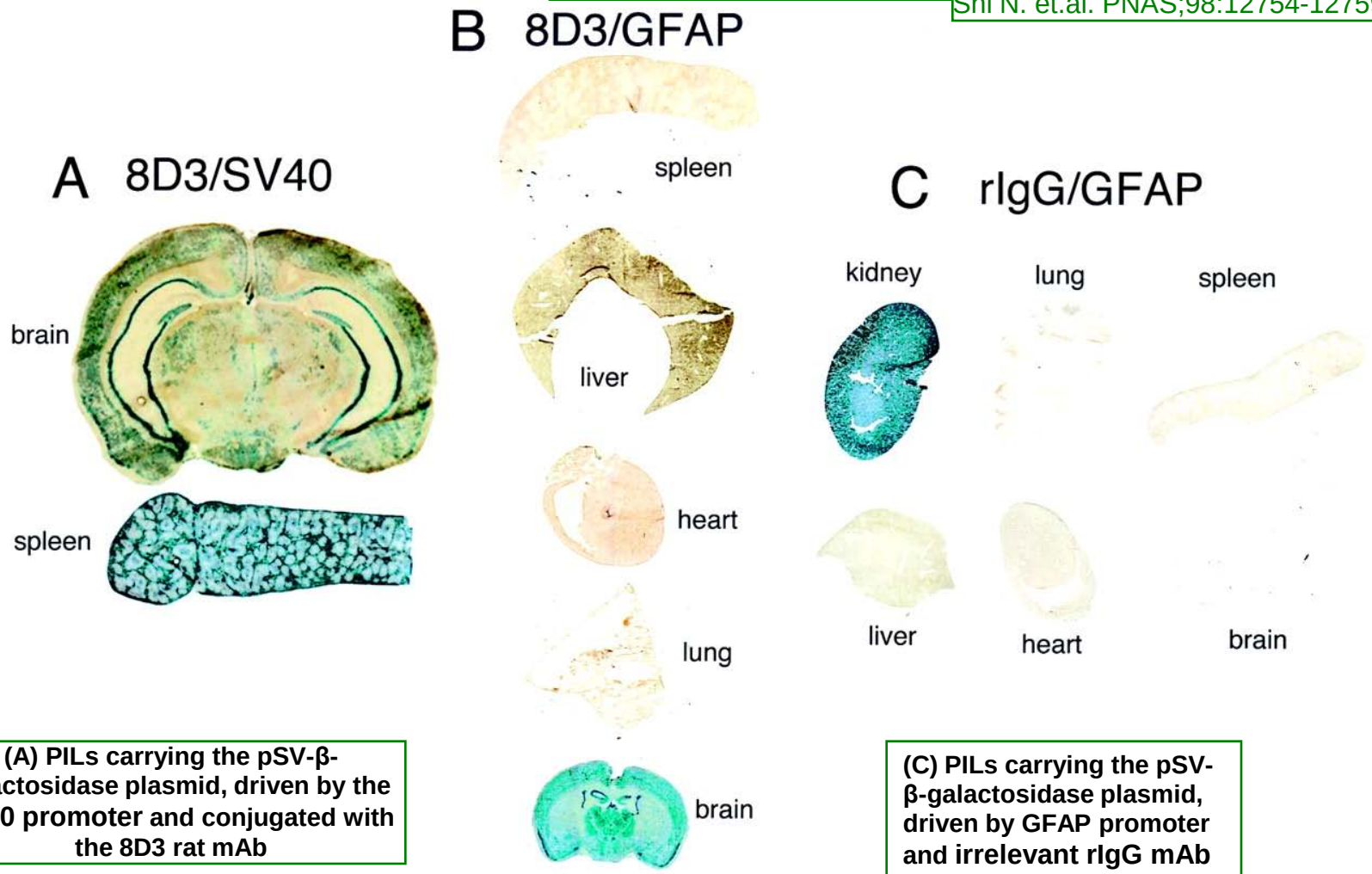
PEG allows to escape the RES

B



Galactosidase plasmid DNA with a brain-specific promoter (GFAP) encapsulated in the 8D3 mAb targeted PEGylated Immunoliposomes

Shi N. et.al. PNAS;98:12754-12759, 2001



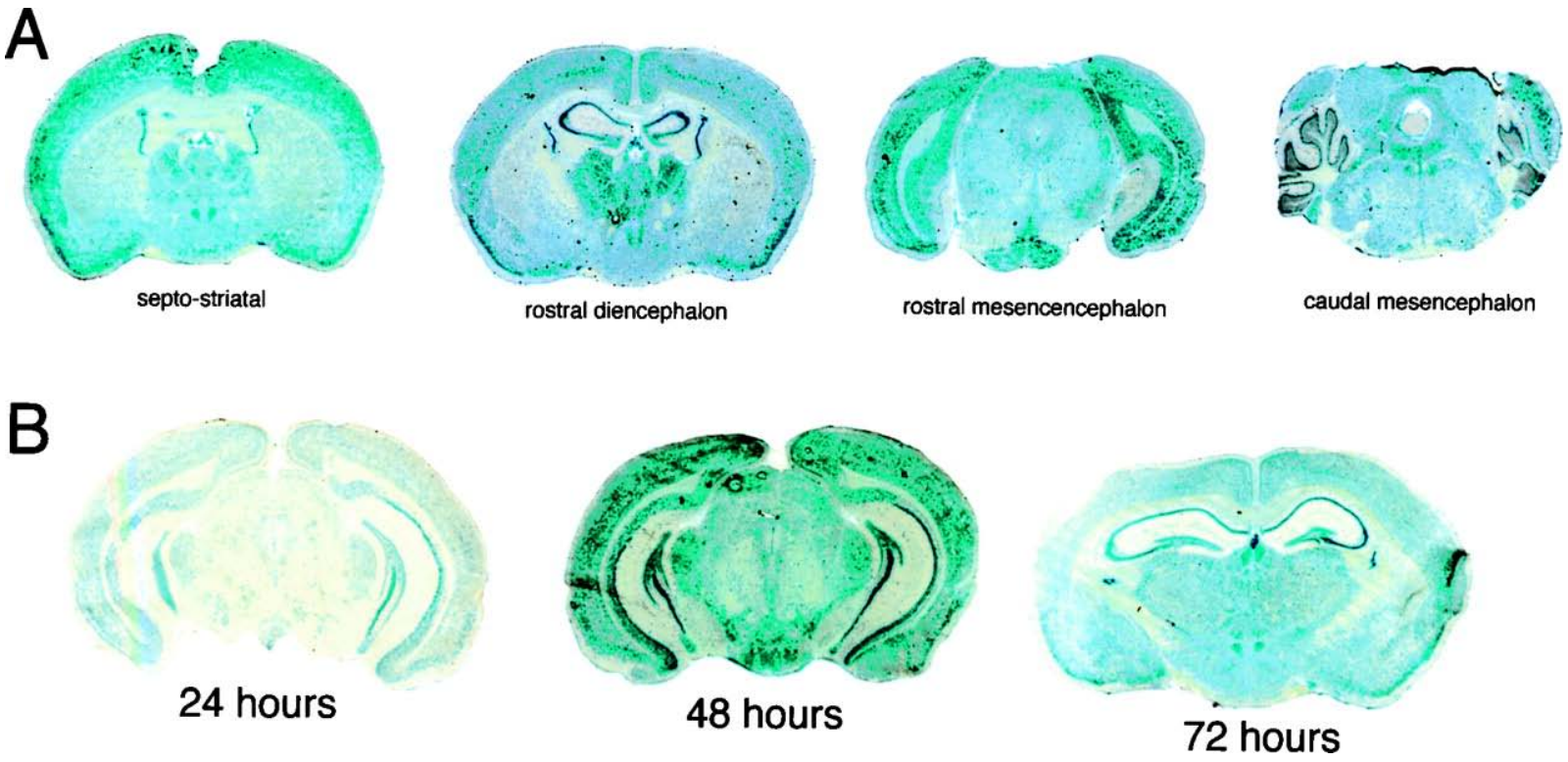
(A) PILs carrying the pSV-β-galactosidase plasmid, driven by the SV40 promoter and conjugated with the 8D3 rat mAb

(B) PILs carrying the pSV-β-galactosidase plasmid, driven by the GFAP promoter and conjugated with the 8D3 rat mAb

(C) PILs carrying the pSV-β-galactosidase plasmid, driven by GFAP promoter and irrelevant rIgG mAb

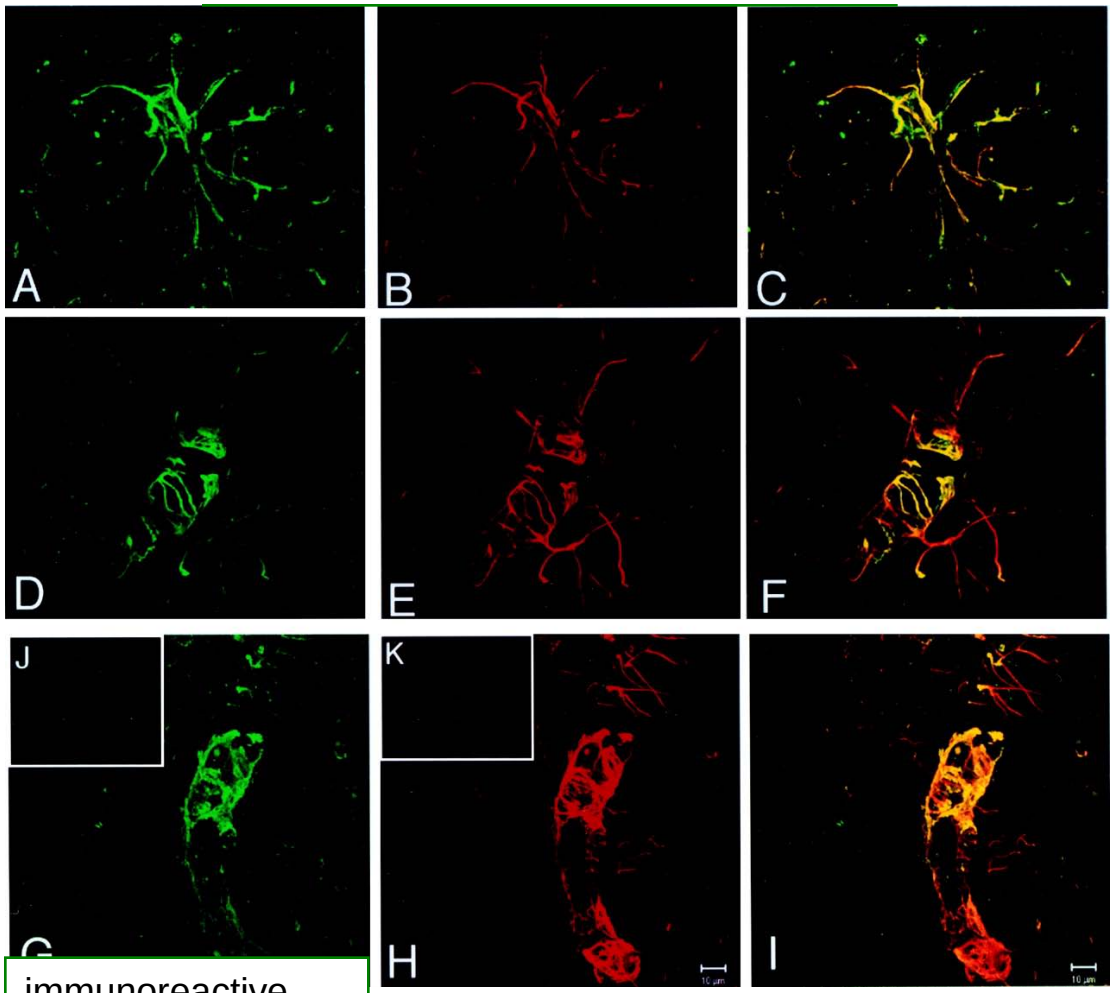
Galactosidase plasmid DNA and GFAP brain specific promotor encapsulated in the 8D3 mAb targeted PEGylated Immunoliposomes

Shi N. et.al. PNAS, 98:12754-12759, 2001



(A) β-Galactosidase histochemistry in mouse brain removed 48 h after a single i.v. injection of the GFAP/β-galactosidase plasmid encapsulated in the interior of 8D3-targeted PILs
(B) β-Galactosidase histochemistry is shown for brain removed from mice at 24, 48, and 72 h after a single i.v. injection of the GFAP/β-galactosidase plasmid encapsulated in the interior of 8D3 PILs.

Confocal microscopy of mouse brain after i.v. injection of the GFAP/β-galactosidase plasmid encapsulated in the interior of 8D3 Pegylated immunoliposomes



immunoreactive galactosidase stained with a fluorescein-labeled antibody

Immunoreactive GFAP stained with rhodamine-labeled antibody

merge

Shi N. et.al. PNAS 2001;98:12754-12759

OPIOID PEPTIDES DECORATED NANOPARTICLES FOR BRAIN DELIVERY

PLGA NANOPARTICLES FUNCTIONALIZED WITH OPIOID PEPTIDES

H_2N–Gly-l-Phe-d-Thr-Gly-l-Phe-l-Leu–X–$CONH_2$

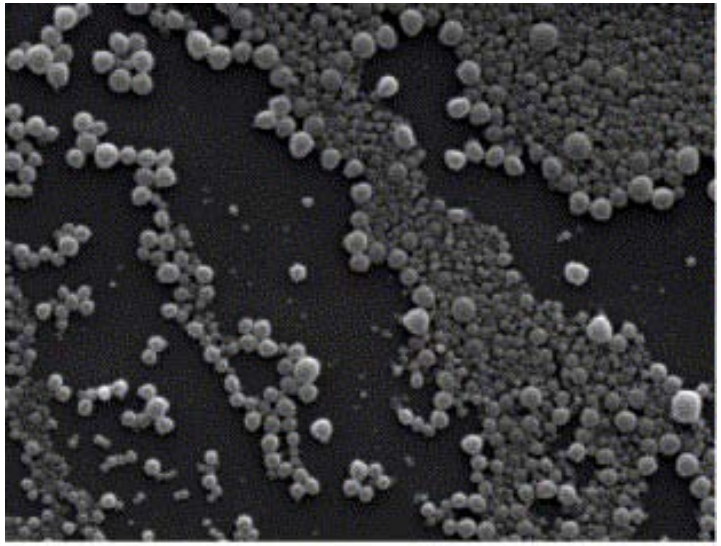
1 X = L-Ser–OH

2 X = L-Ser-*O*-β-D glucose

3 X = L-Ser-*O*-β-D galactose

4 X = L-Ser-*O*-β-D xylose

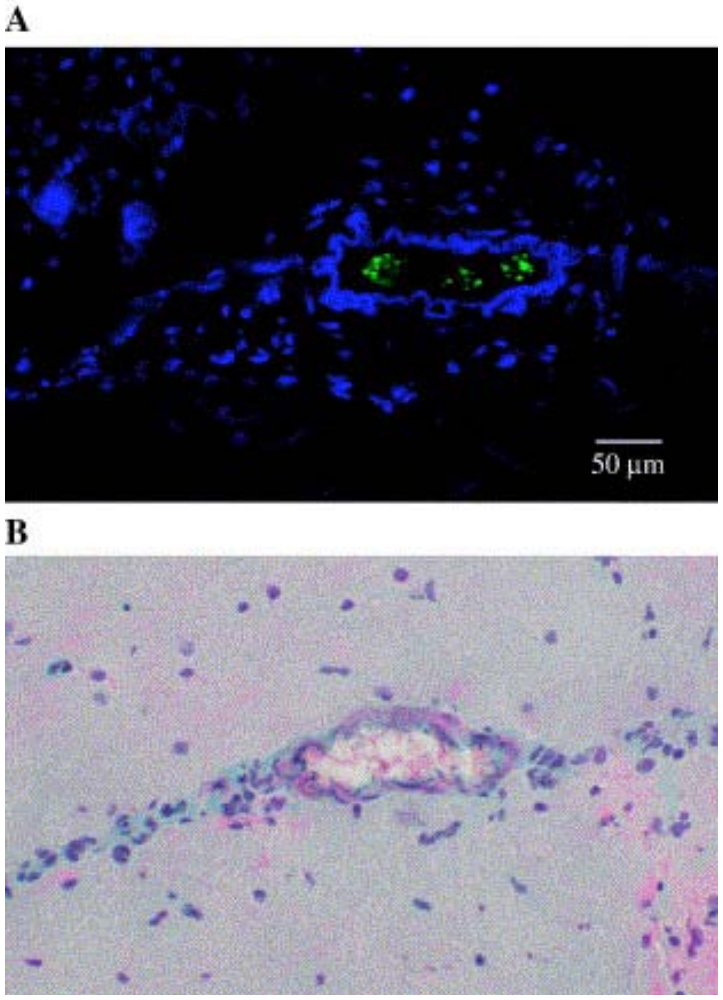
5 X = L-Ser-*O*-β-D lactose



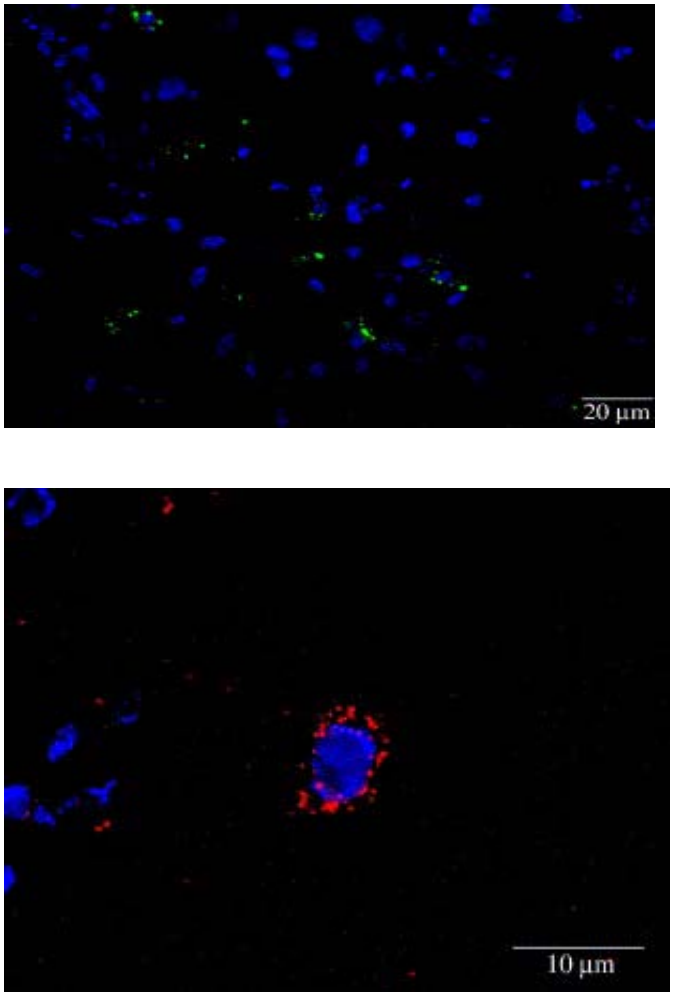
—— 1 μm.

-Synthetic opioid peptides were shown to be BBB permeable
-Permeability may be enhanced in the presence of glucosidic moieties

BRAIN TRANSLOCATION OF THE NANOPARTICLES OF PLA CONJUGATED WITH OPIOID PEPTIDES

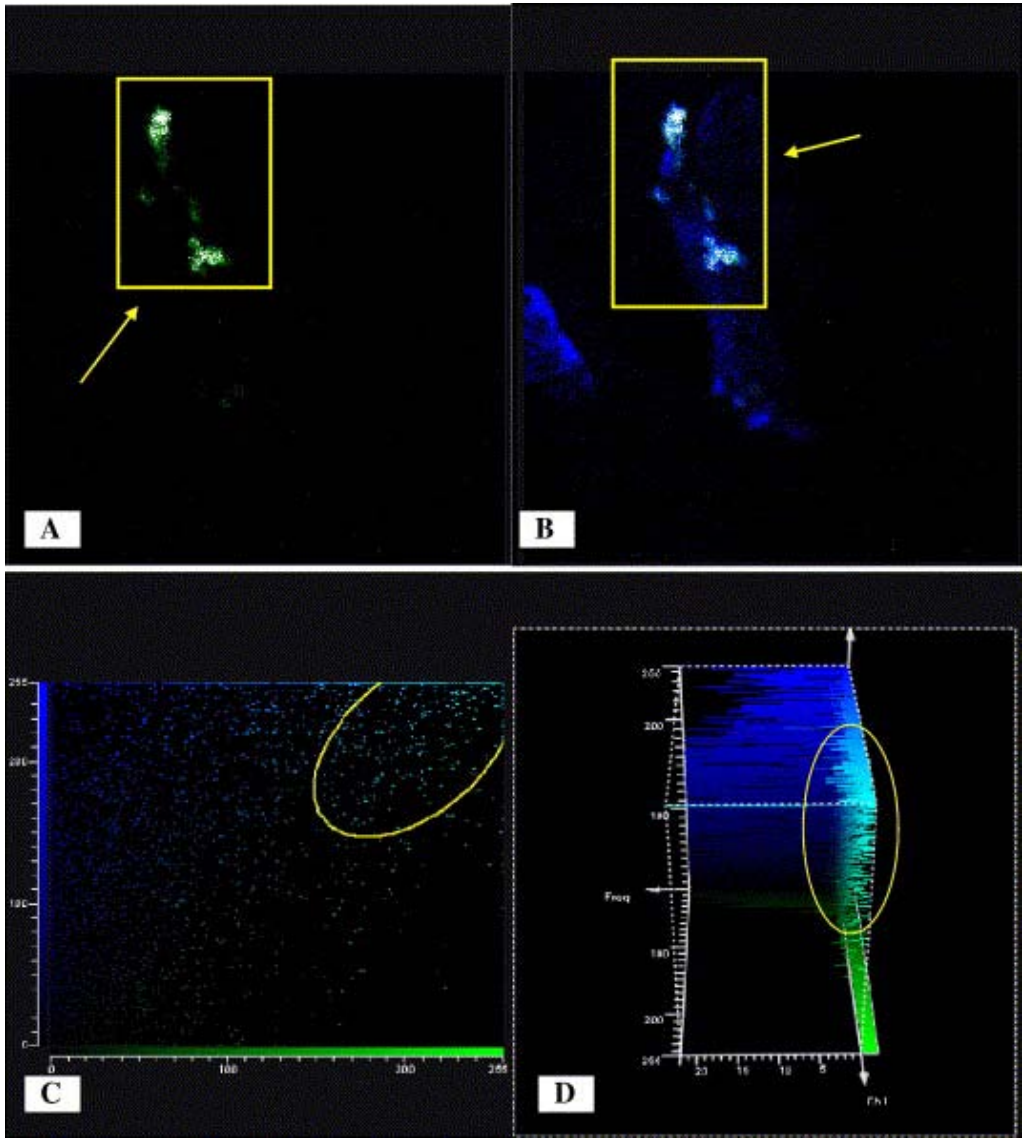


PLGA nanoparticles are located only in the cerebral vascular space



PLGA nanoparticles linked with opioid peptide translocate the brain tissue PLGA-PEPT 1 (green) and PLGA-PEPT 2 (red)

BRAIN TRANSLOCATION OF THE NANOPARTICLES OF PLA CONJUGATED WITH OPIOID PEPTIDES COLOCALIZATION

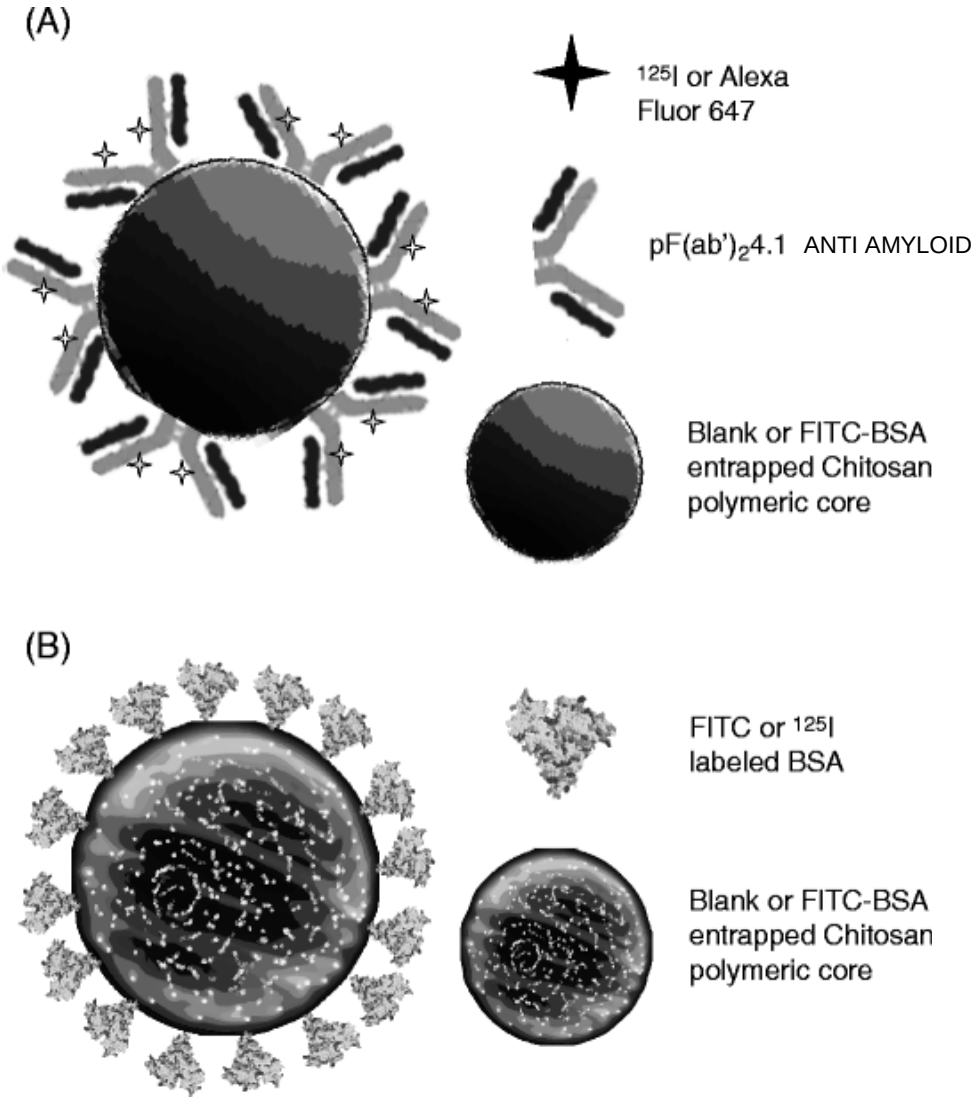


Confocal study Images 7C and 7D are referred to the intensity per cent of green and blue coloration coming respectively from fluorescent Np and DAPI-double strand DNA complexes of cerebral cells. The spots included into the yellow ellipsis in (C) and (D) are considered as the points of interaction between fluorescent Np and cells because of their same position in the thickness of the sample. These sites of interaction are also well recognizable as the white spots enclosed in the yellow squares in (A) (PLGA-peptide **1** Np in green) and (B) (cerebral cells–DAPI complexes in blue).

NANOPARTICLES FOR RECOGNITION OF AMYLOID PLAQUES

SMART NANOVEHICLES TO TARGET AMYLOID DEPOSITS

E K Agyare et al., Pharmaceutical Research, 25, 2674-2684 (2008)



A Design of the smart nano-vehicle (SNV); **B** Design of the control nano-vehicle (CNV).

SMART NANOVEHICLES TO TARGET AMYLOID DEPOSITS

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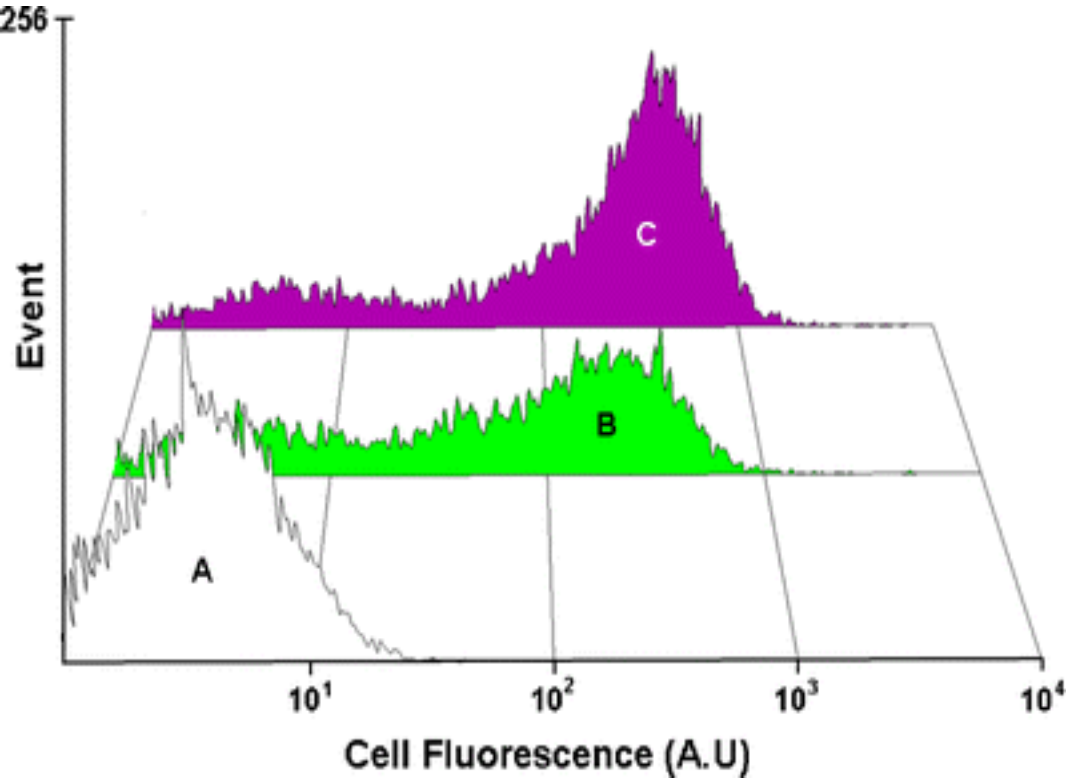
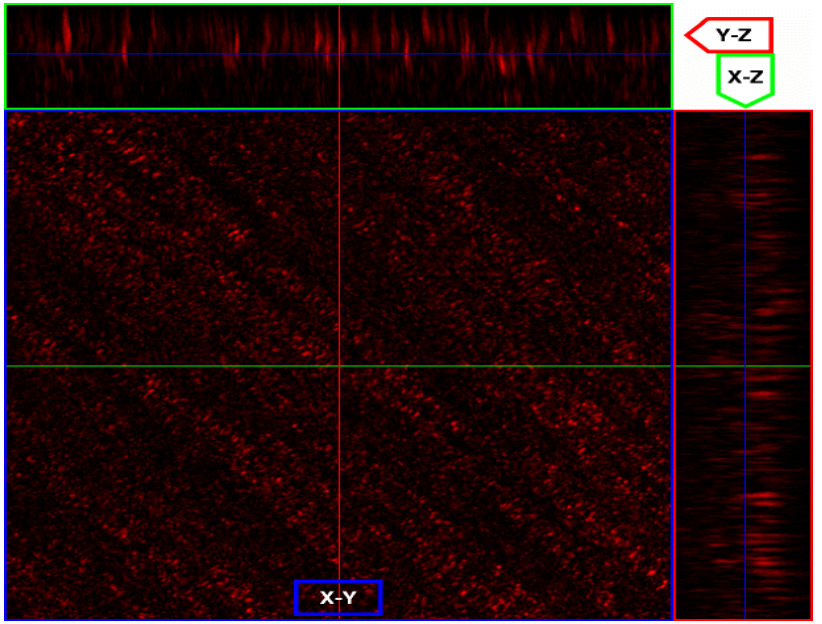


flow cytometry: **A** untreated bovine brain microvascular endothelial cells (BBMECs), **B** BBMECs treated with FITC-BSA-CNVs; and **C** BBMECs treated with FITC-BSA-SNVs.

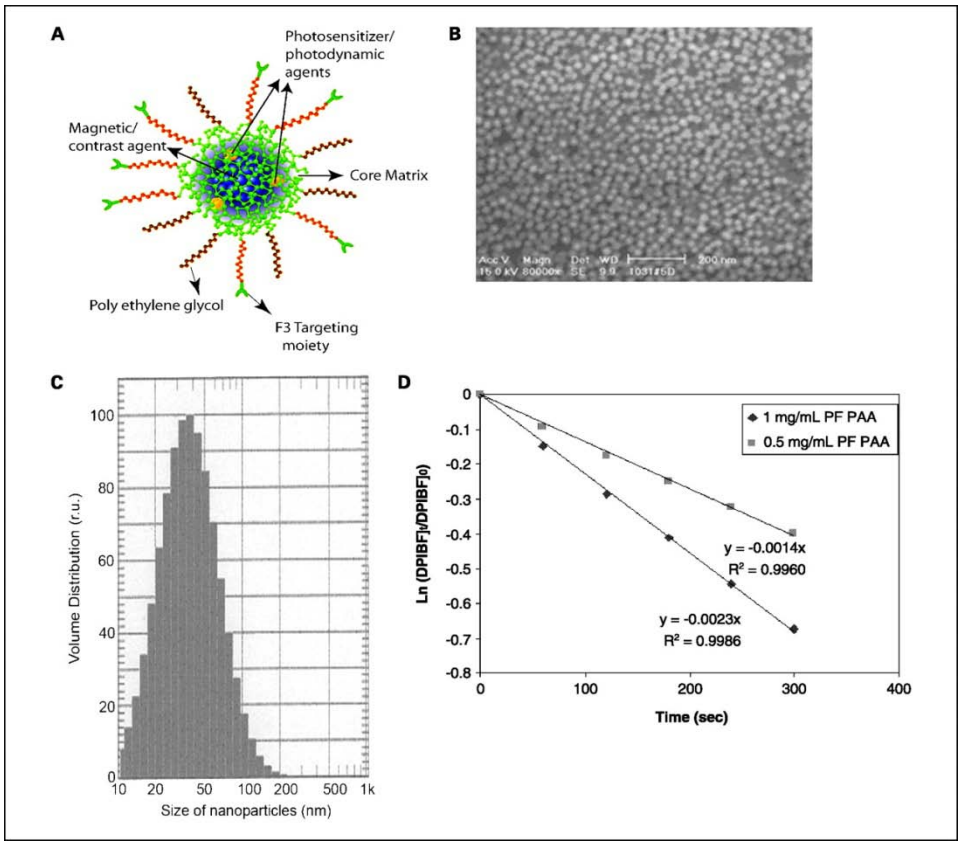


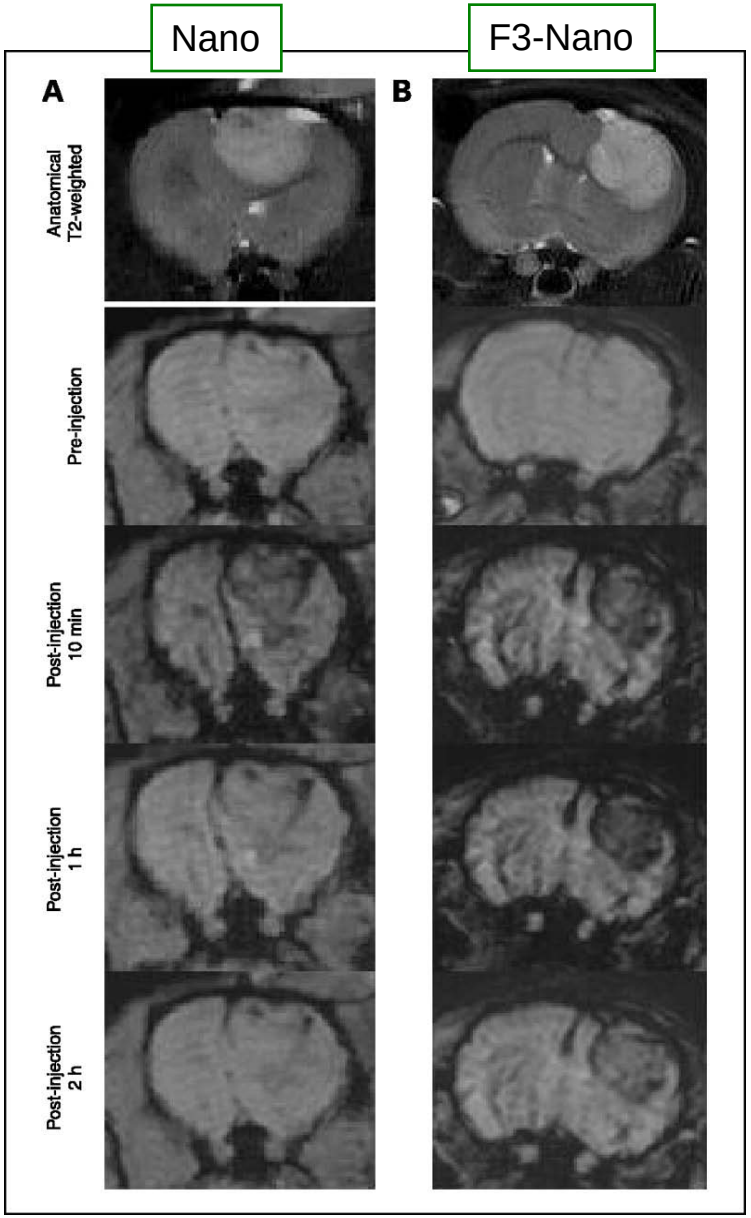
Localization of SNVs labeled with Alexa Fluor 647 in bovine brain microvascular endothelial cell (BBMEC) monolayer. Images were taken as z-stack **A** Z-stack presented in x–y plane clearly demonstrates cellular uptake of AF647-SNVs. Projection in both the x–z and y–z orthogonal planes confirms the transcytosis of AF647-SNVs across the BBMEC monolayer.

IRON OXIDE NANOPARTICLES FOR BRAIN IMAGING

BRAIN TUMOR IMAGING WITH MULTIFUNCTIONAL NANOPARTICLES

Reddy R. et al.,Clin Cancer Research, 22, 6677-6686, 2006





BRAIN DELIVERY USING NANOCARRIERS: CONCLUSIONS

- The BBB is equipped with tight junctions and is of low permeability to various drug molecules
- Control non functionalized nanotechnologies don’t diffuse spontaneously through the BBB
- Functionalization of nanocarriers using different ligands (antitransferrin mAb, ApoE, synthetic opioid peptides etc.) allows significant brain translocation
- Applications in the fields of pain treatment, neurodegenerative diseases, brain cancers, imaging etc.