Queen Mary
University of London

Barts and The London
School of Medicine and Dentistry

Omega-3 Fatty Acids for the Treatment of Traumatic Spinal Cord and Brain Injuries

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NEUROTRAUMA

Brain injury and spinal cord injury

Traumatic brain injury is the leading cause of mortality and morbidity worldwide under the age of 45
Present trends indicate an increasing impact on the older population
Significant risk of developing neurodegeneration

TIMELINE OF THE CARE PATHWAY AFTER AN INJURY

Minutes to hours

Hours to weeks

Weeks to months

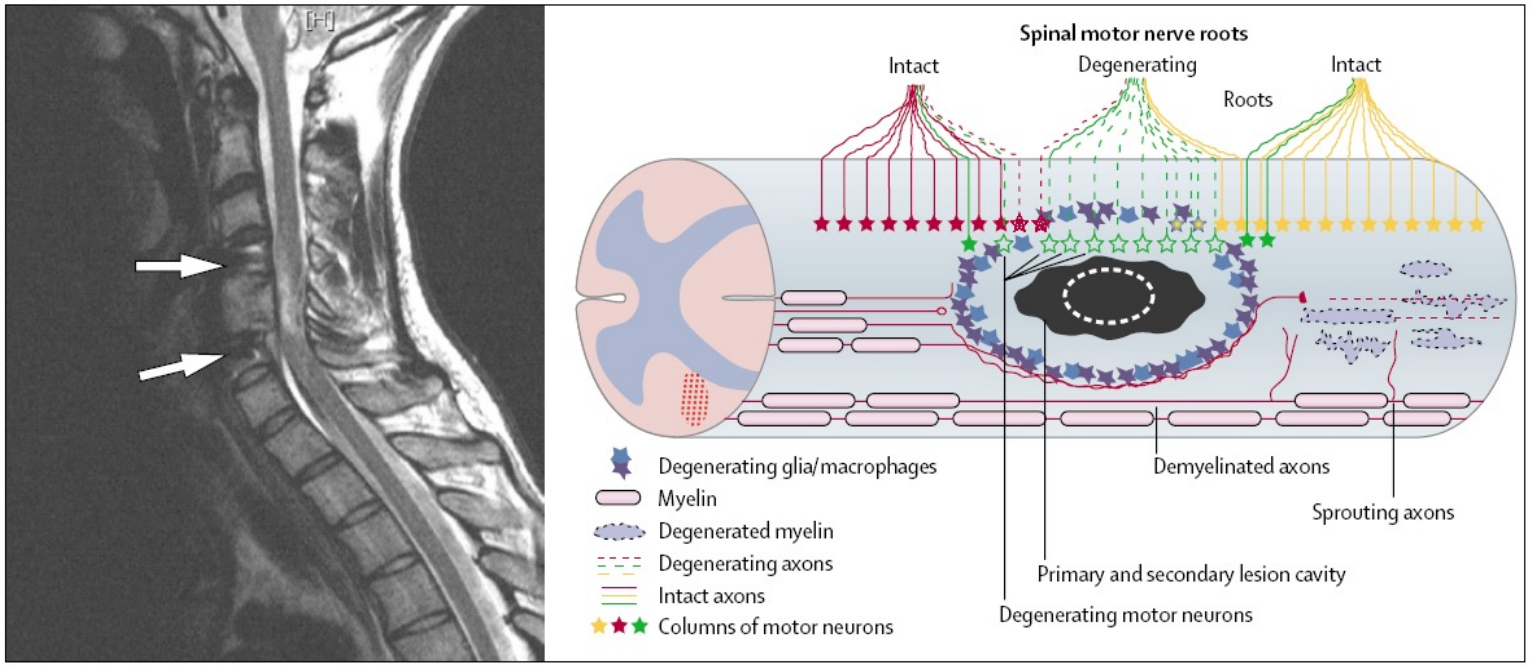
NEUROPROTECTION

Advanced Trauma Management

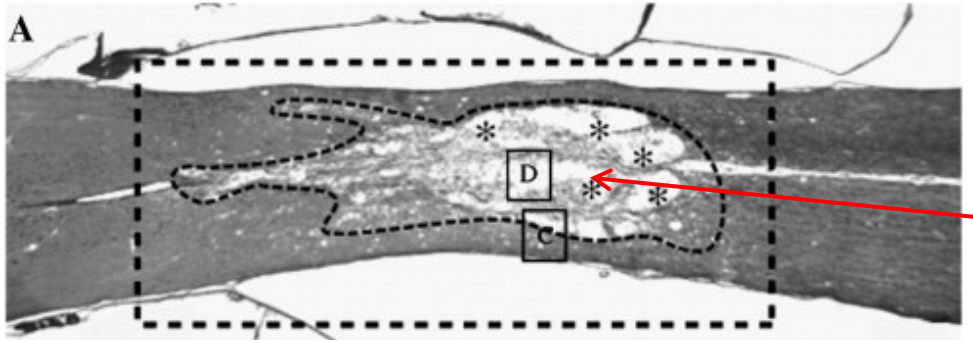
NEUROREPAIR

Neurorehabilitation

Example of cervical C5 sensorimotor complete tetraplegia



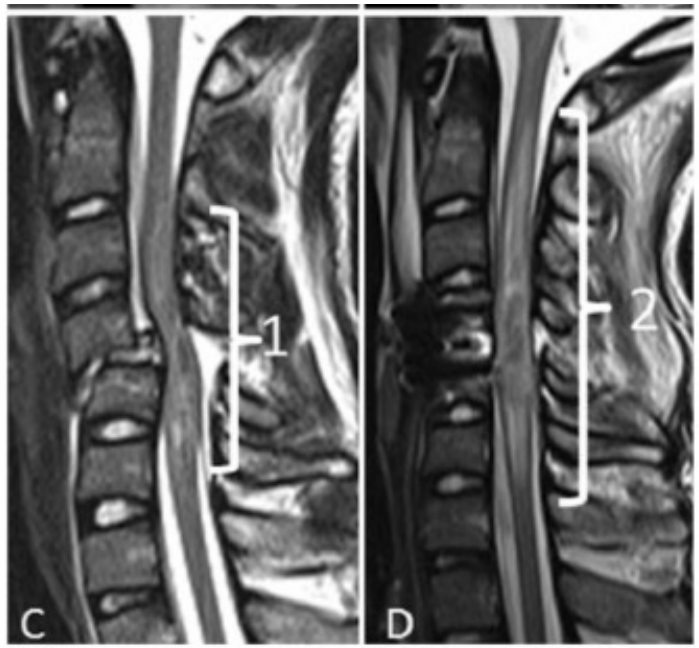
(Dietz and Curt, 2006)



Spinal cord injury lesion evolution

Fast expansion of the injury zone due to secondary injury

Intramedullary lesion expansion on magnetic resonance imaging in patients with motor complete cervical spinal cord injury



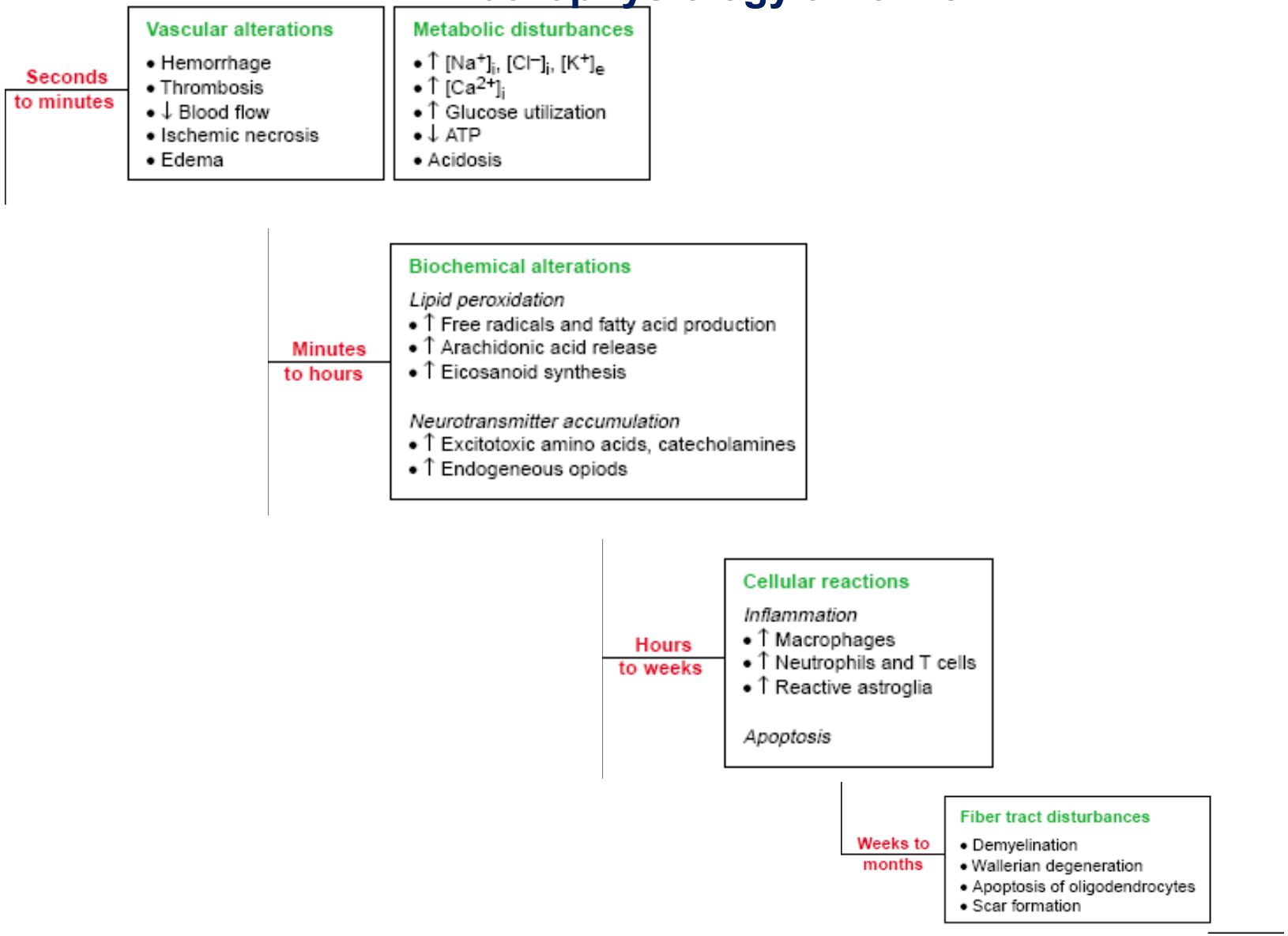
(Aarabi et al, 2012)

Expansion injury zone: 69.2 to 102.8 mm in 37 hours
Expansion rate: 0.91 mm/hour

17-year old boy, cervical spine injury during motor vehicle collision; **tetraplegia**

CONSEQUENCES OF SPINAL CORD INJURY

Pathophysiology timeline



Bareyre & Schwab 2003

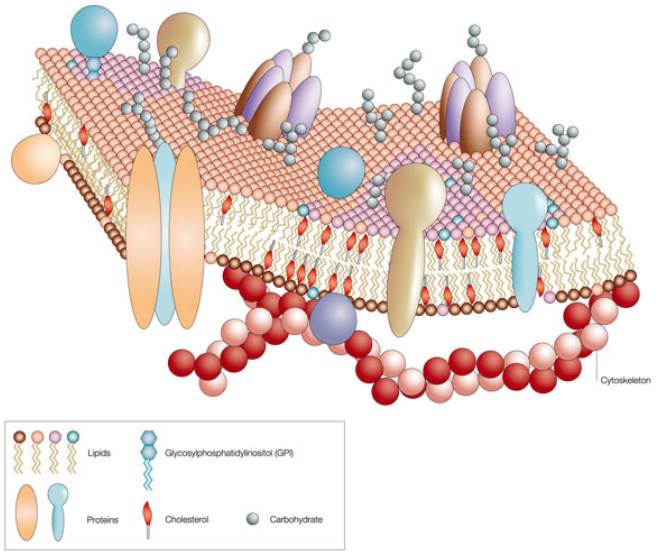
Important questions for successful clinical translation in traumatic brain and spinal cord injury

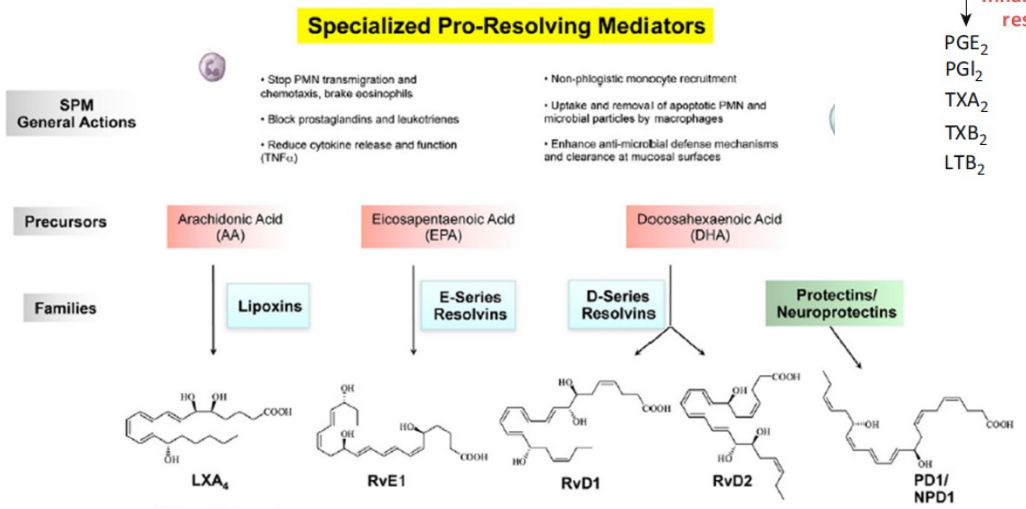
- Multiple models of injury, injury levels and injury severity
- Several species
- Dose range and regime of administration
- Stable preparation and formulation for human use
- Therapeutic window
- Impact on all the key processes linked to secondary injury

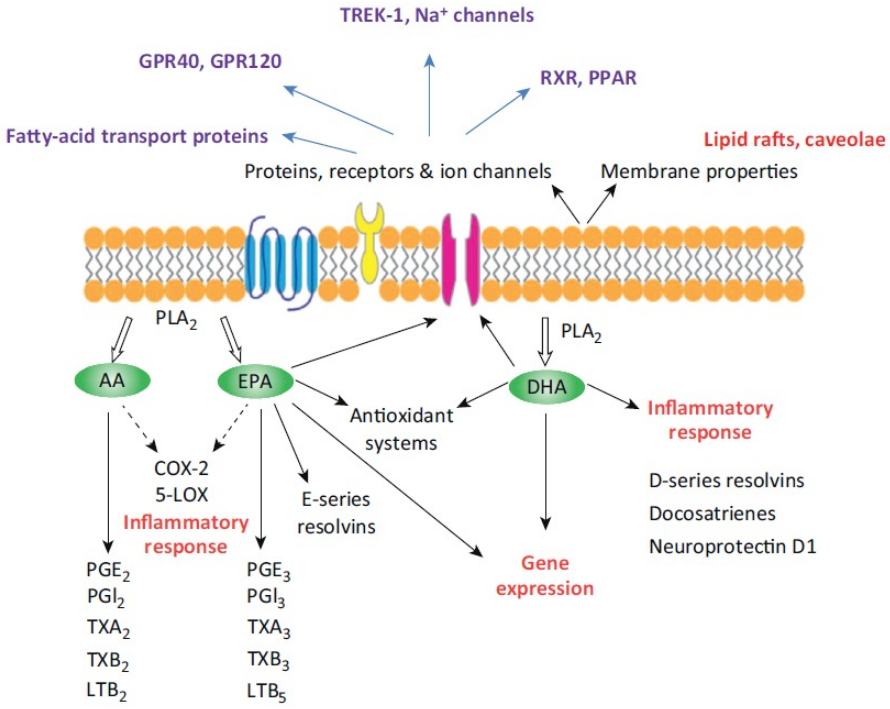
Docosahexaenoic acid (DHA) for neuroprotection in traumatic spinal cord injury

Docosahexaenoic acid – an omega-3 fatty acid with multiple cellular targets and active metabolites

- Targets: ion channels (dual pore mechano-activated background potassium channels - TREK-1), voltage-gated sodium channels, retinoid receptors (RXR), peroxisome proliferator-activated receptors (PPAR), GPCRs…
- Active metabolites (resolvins, protectins…)







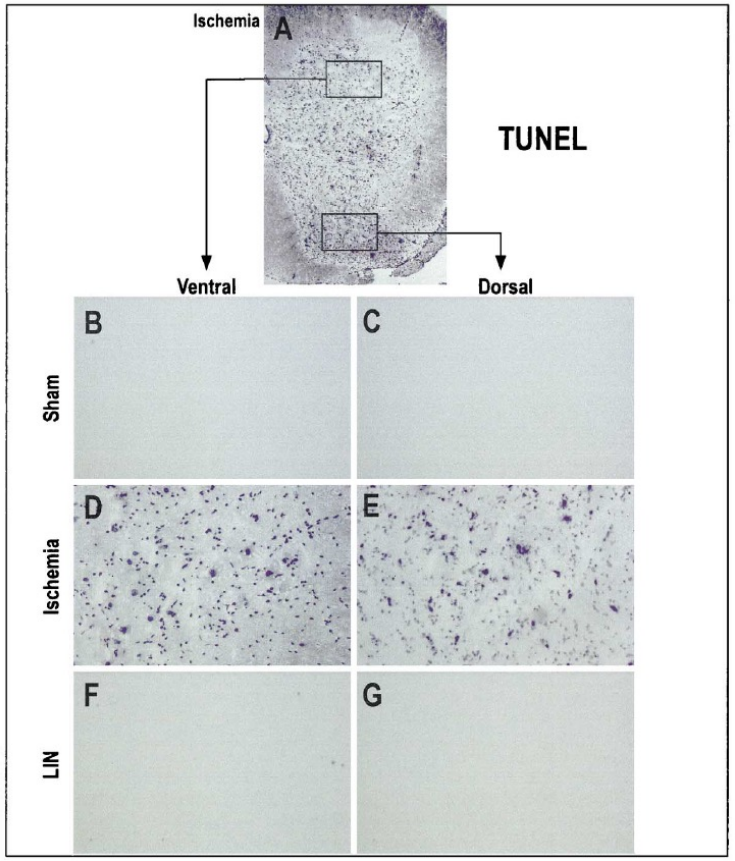
(Michael-Titus and Priestley, 2013)

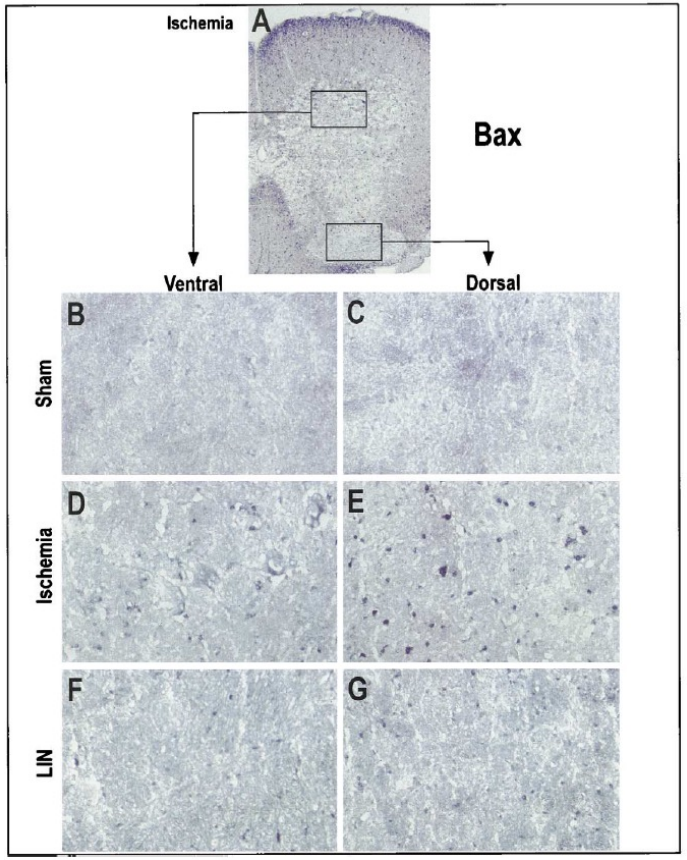
HOW IT ALL BEGAN…

Linolenic acid prevents neuronal cell death and paraplegia after transient spinal cord ischemia in rats

Loïc Lang-Lazdunski, MD, PhD,[a,b] Nicolas Blondeau, PhD,[b] Gisèle Jarretou, BS,[b] Michel Lazdunski, PhD, DSc, and Catherine Heurteaux, PhD,[b] *Clamart and Valbonne, France*

(J Vasc Surg 2003;38:564-75.)



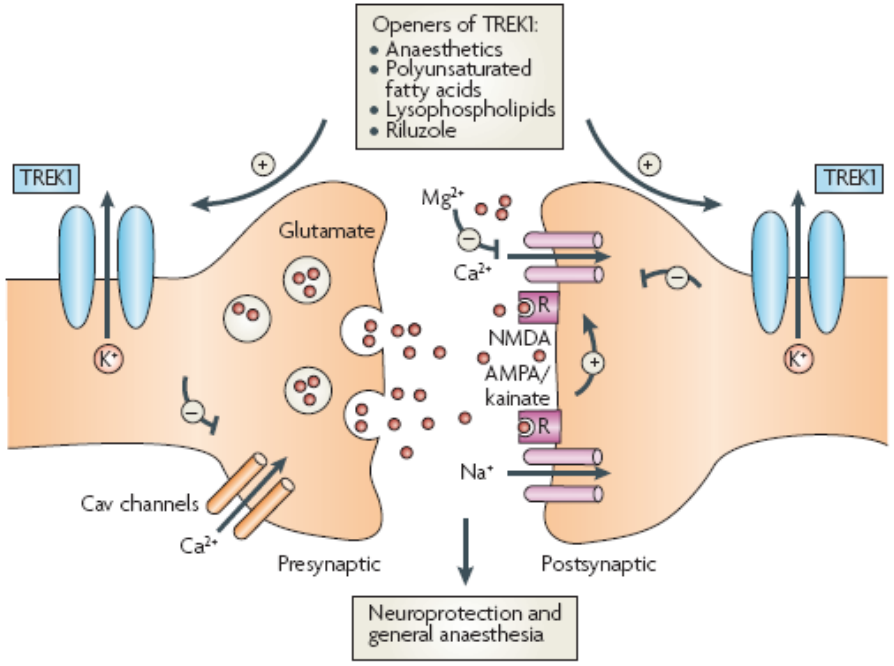


Cross-clamping of aortic arch and left subclavian artery – 14 min

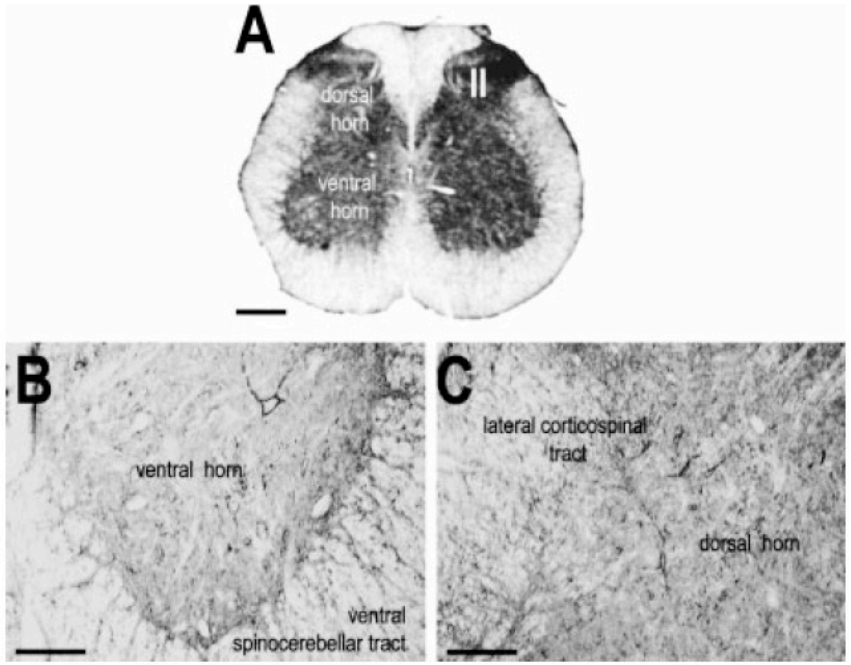
Linolenic acid 250 nmol/kg, i.v.
At onset of reperfusion

Linolenic acid is a ligand for the TREK-1 channel

DHA targets the TREK-1 potassium channel – a key modulator of the depolarization threshold



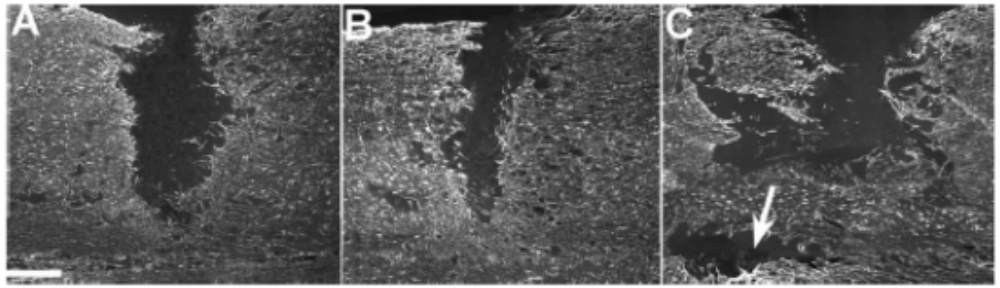
(Honore 2007)



(Hervieu et al., 2001)

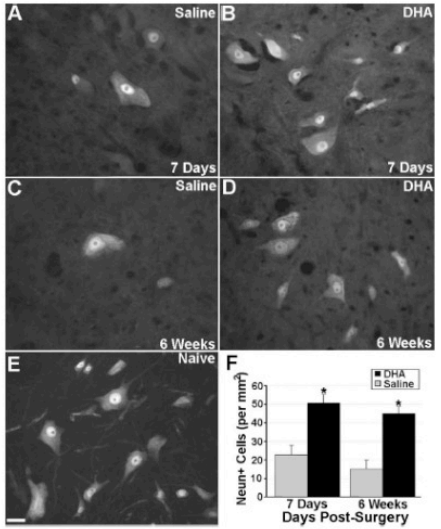
DHA induces improved neurological outcome and tissue protection in hemisection SCI

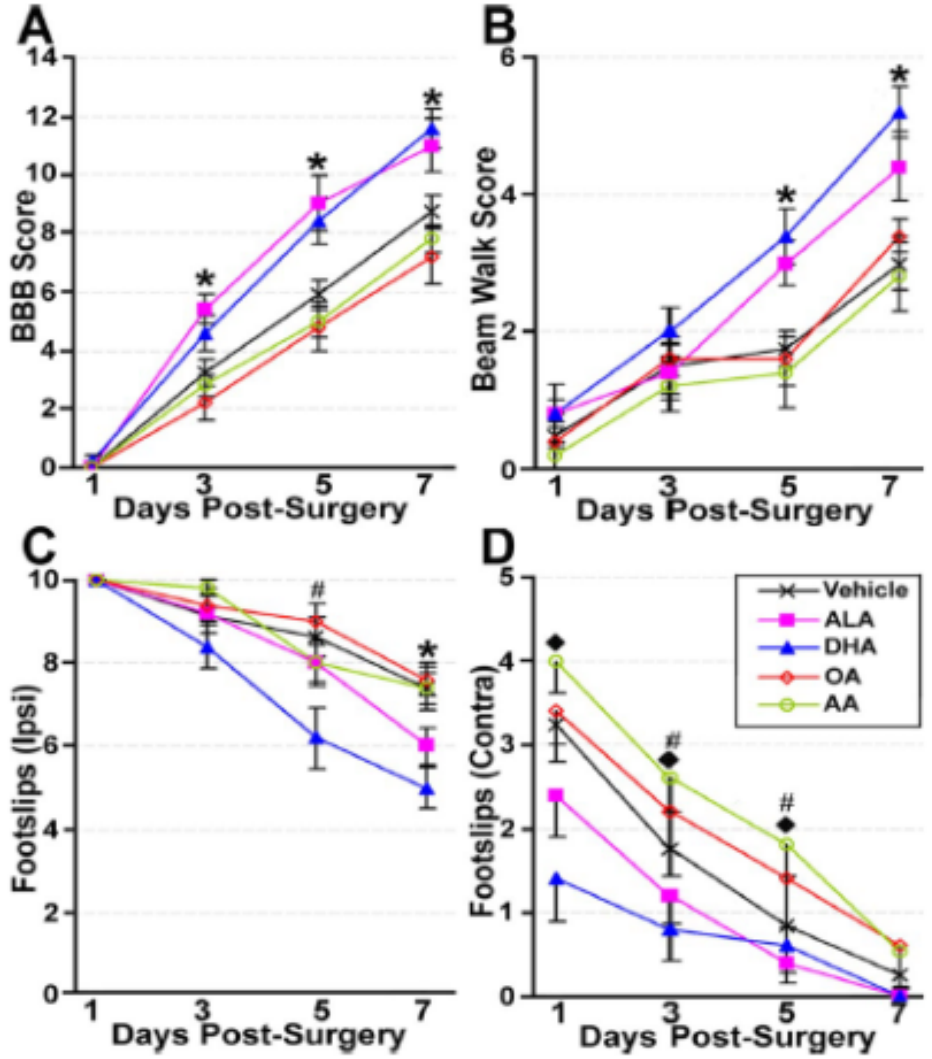
Rat - Hemisection injury - Thoracic



Vehicle DHA AA

Fatty acids: 250 nmol/kg i.v. 30 min post-SCI

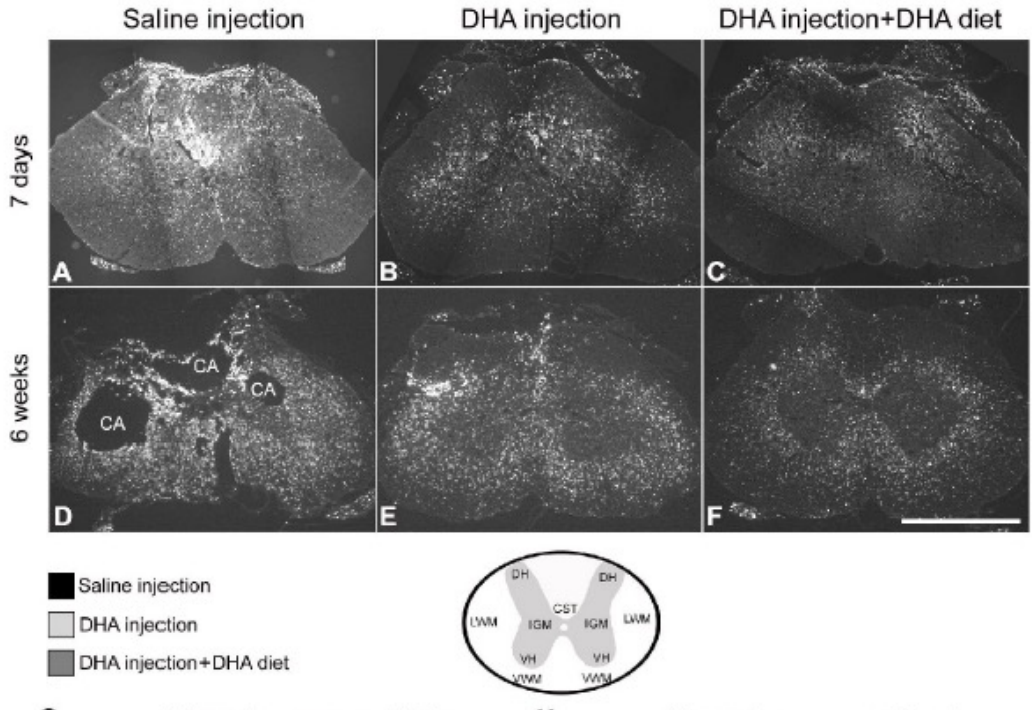


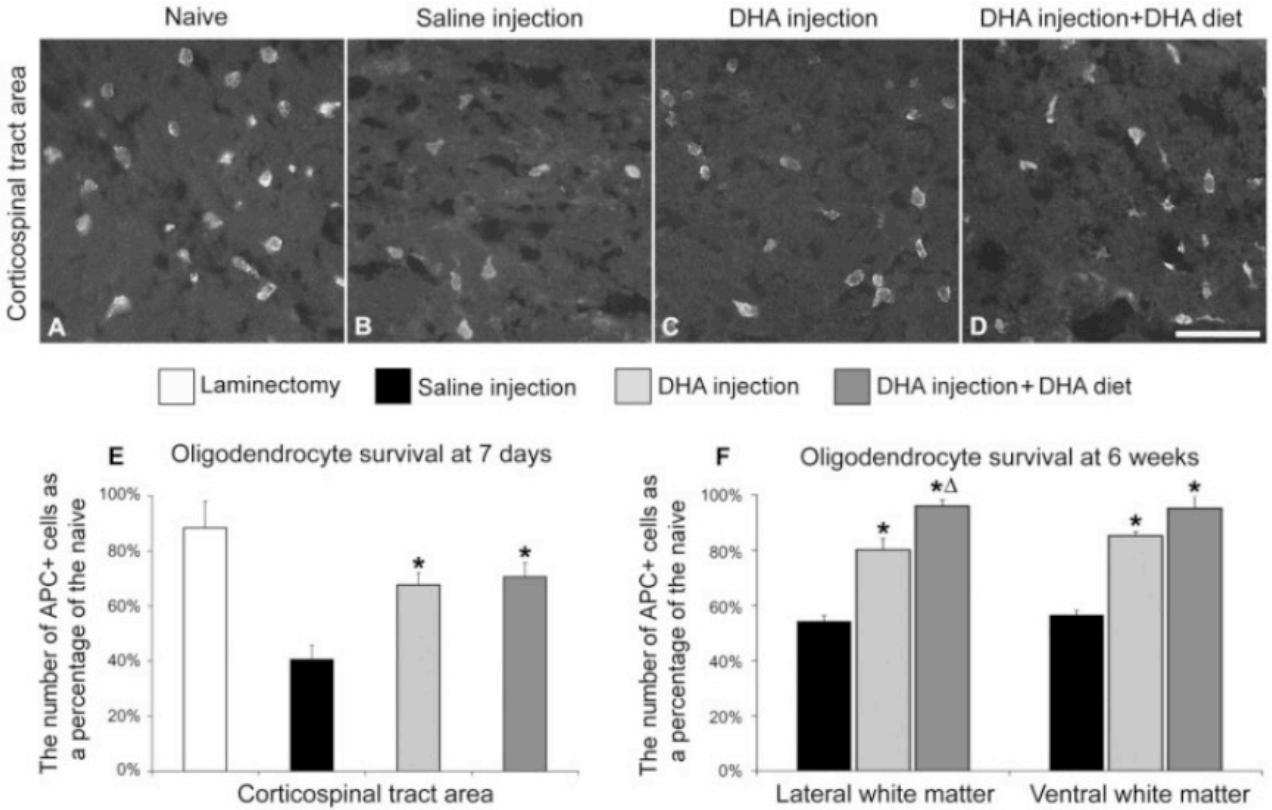


(King et al, J Neurosci, 2006)

DHA induced improved neurological outcome and tissue protection in compression SCI

Rat - Compression injury - Thoracic





ED1 (microglia/macrophages)

(Huang et al, Brain, 2007)

DHA 250 nmol/kg i.v. 30 min post-SCI

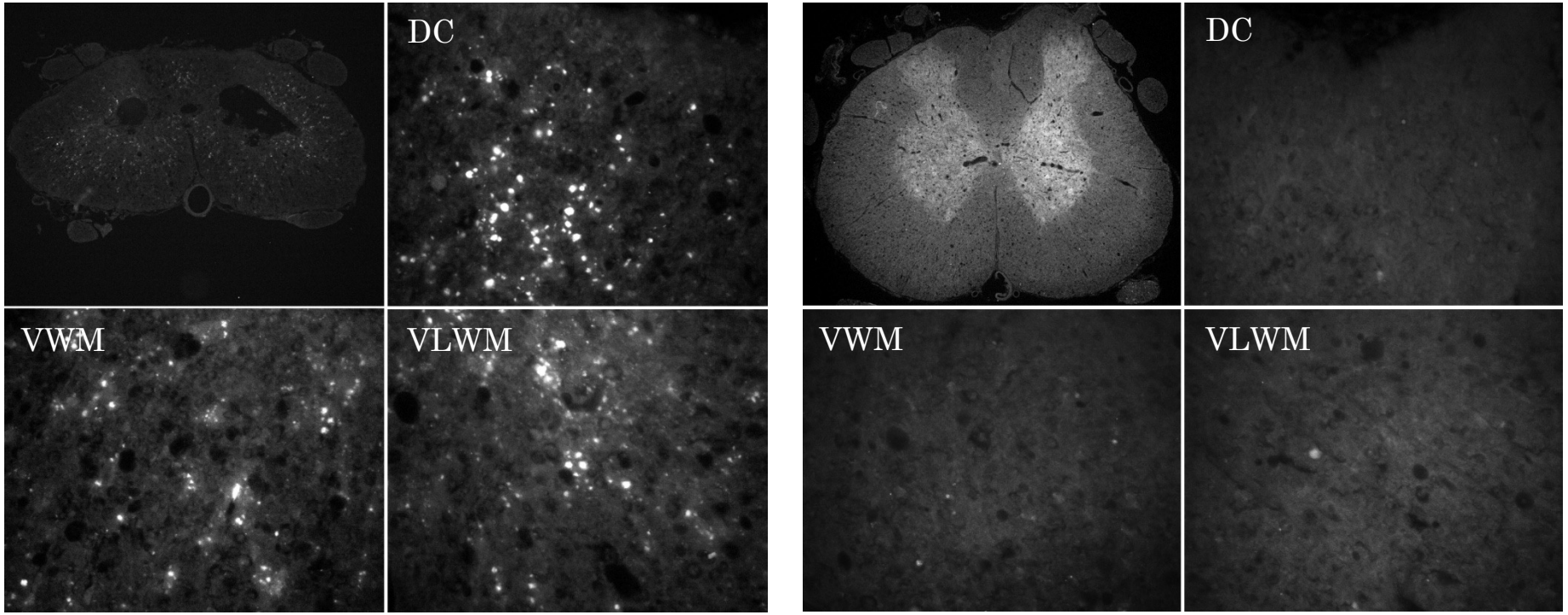
with or without an DHA-enriched diet (approx. 300-400 mg/kg/day) for 6 weeks

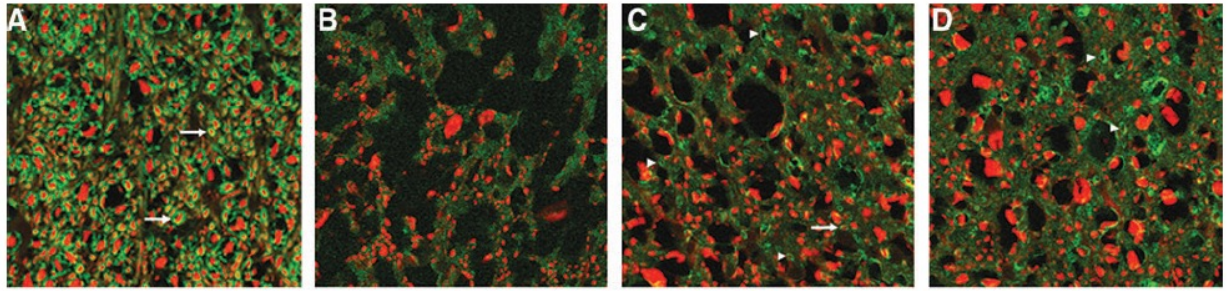
DHA EFFECT ON AXONAL INTEGRITY

β-amyloid precursor protein accumulation

Saline-treated animal

DHA-treated animal

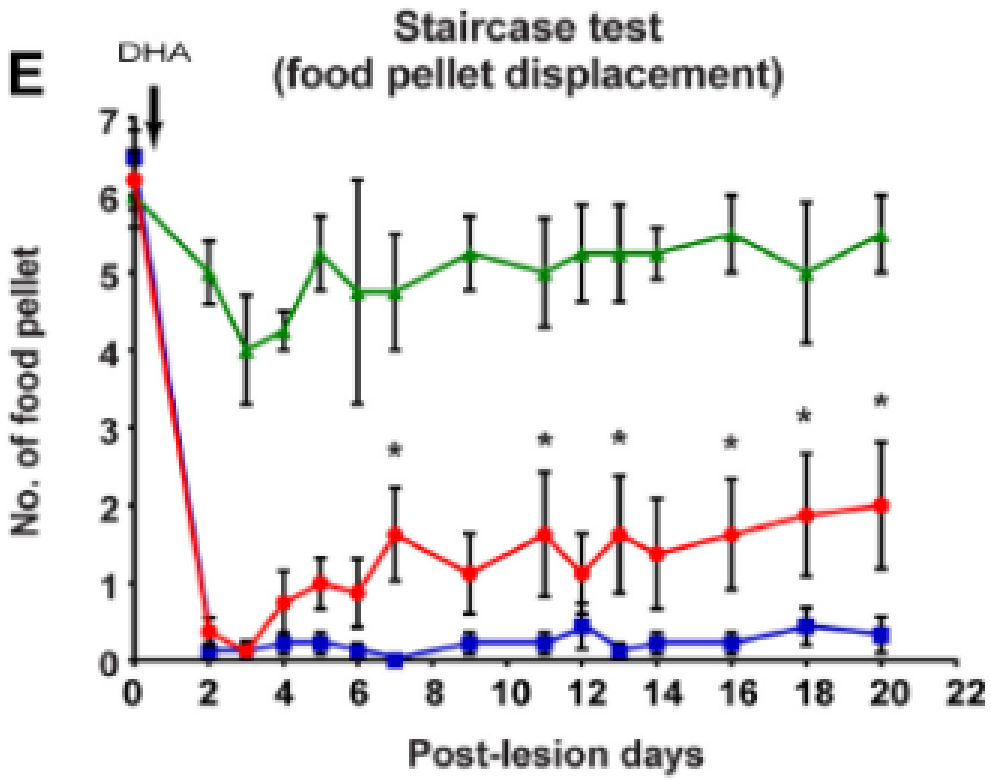


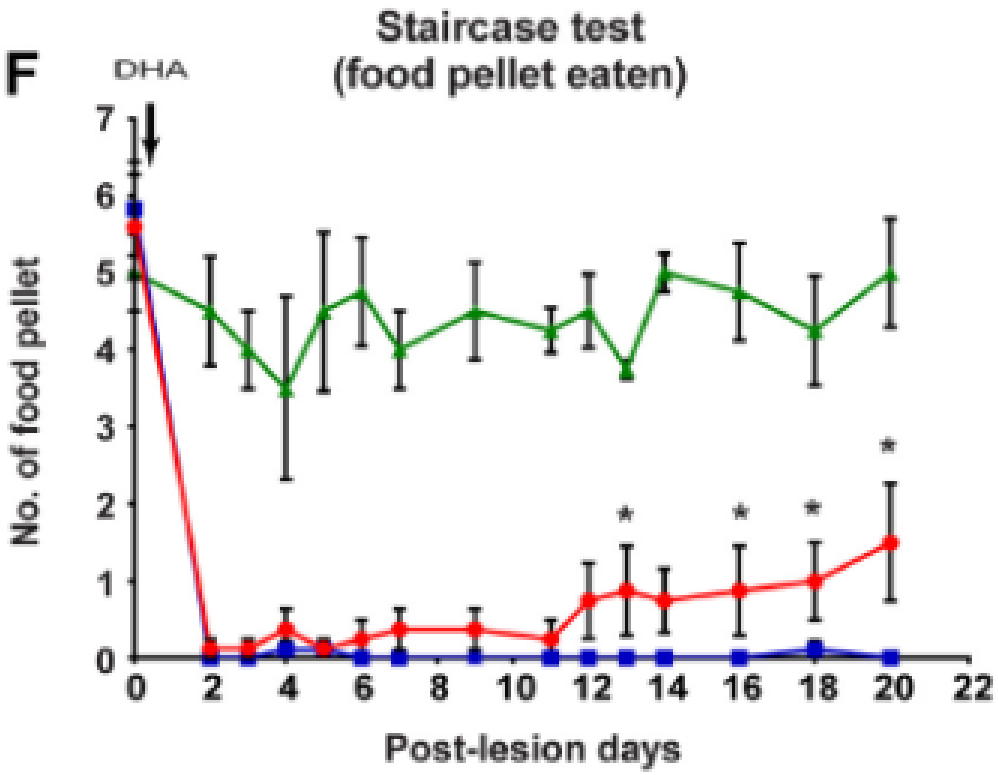


MBP / NF staining

(Ward et al 2010)

DHA in rat cervical hemisection injury

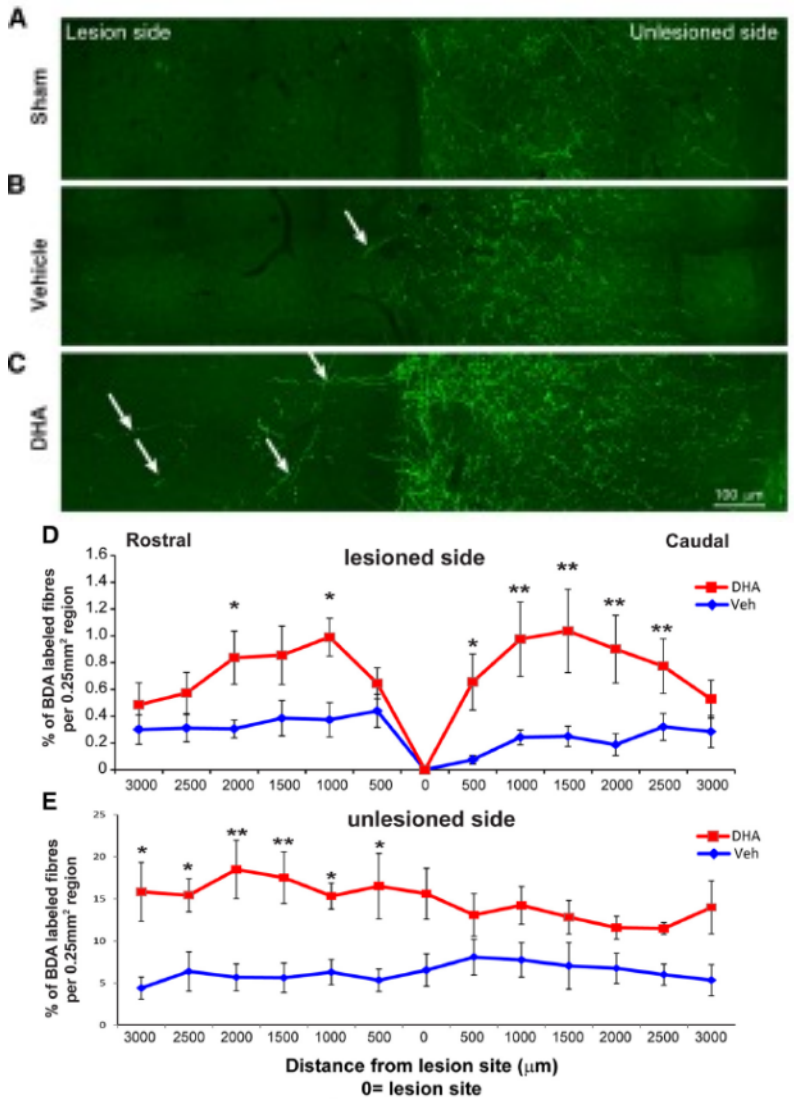


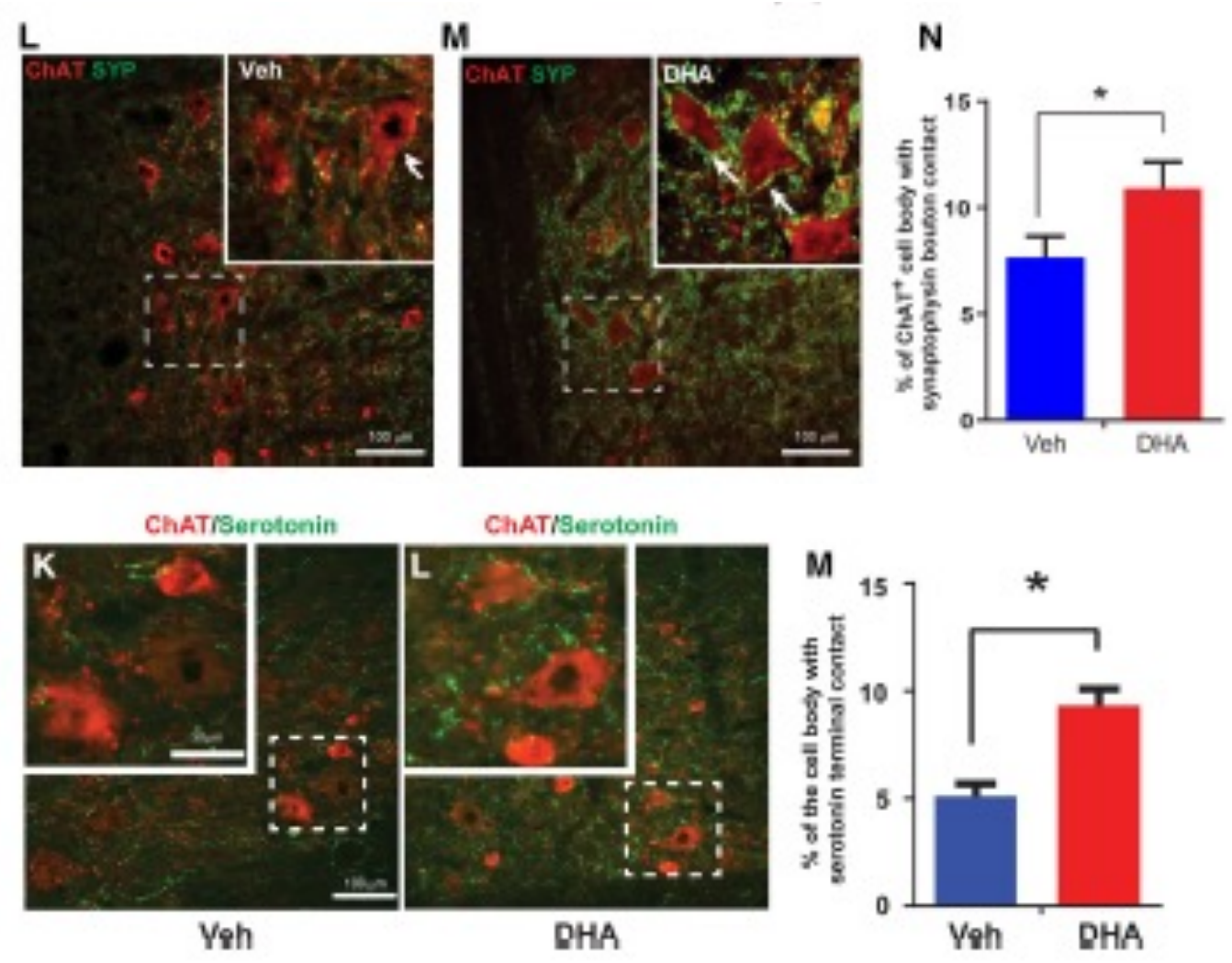


Liu et al, J Neurosci, 2015

DHA in rat cervical hemisection injury

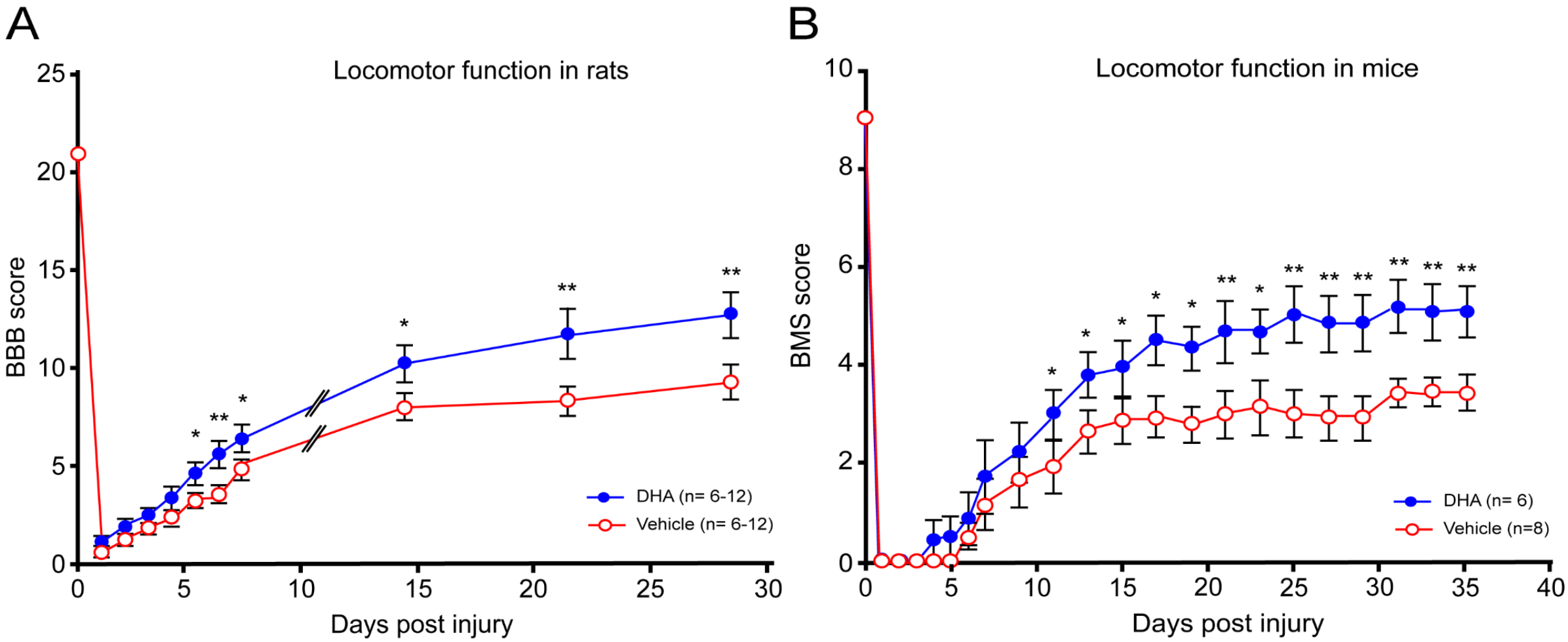
Increased sprouting and increased synaptic contacts on motor neurons





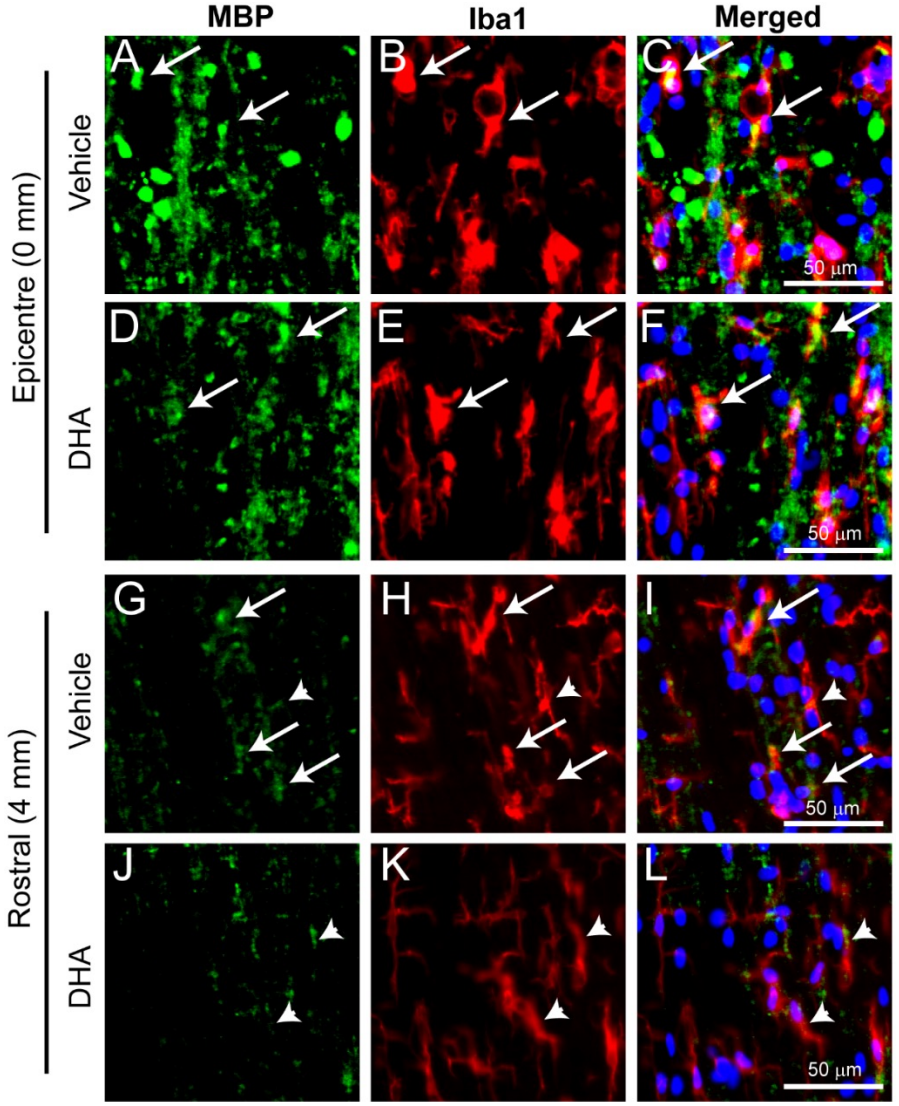
Liu et al, J Neurosci, 2015

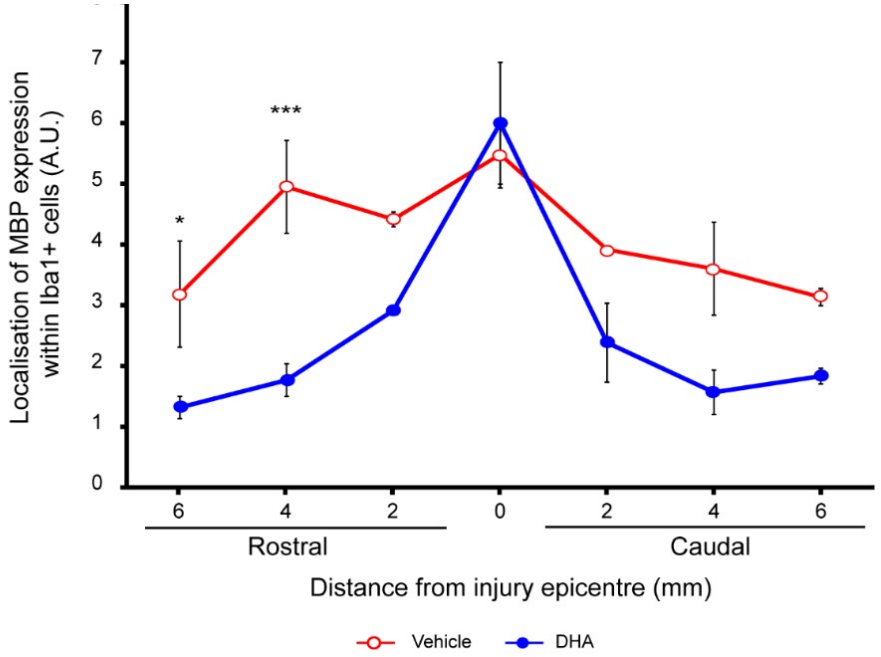
DHA in thoracic contusion injury



(Yip et al., Hum Mol Gen, 2019)

DHA effect of myelin phagocytosis by microglia/macrophages





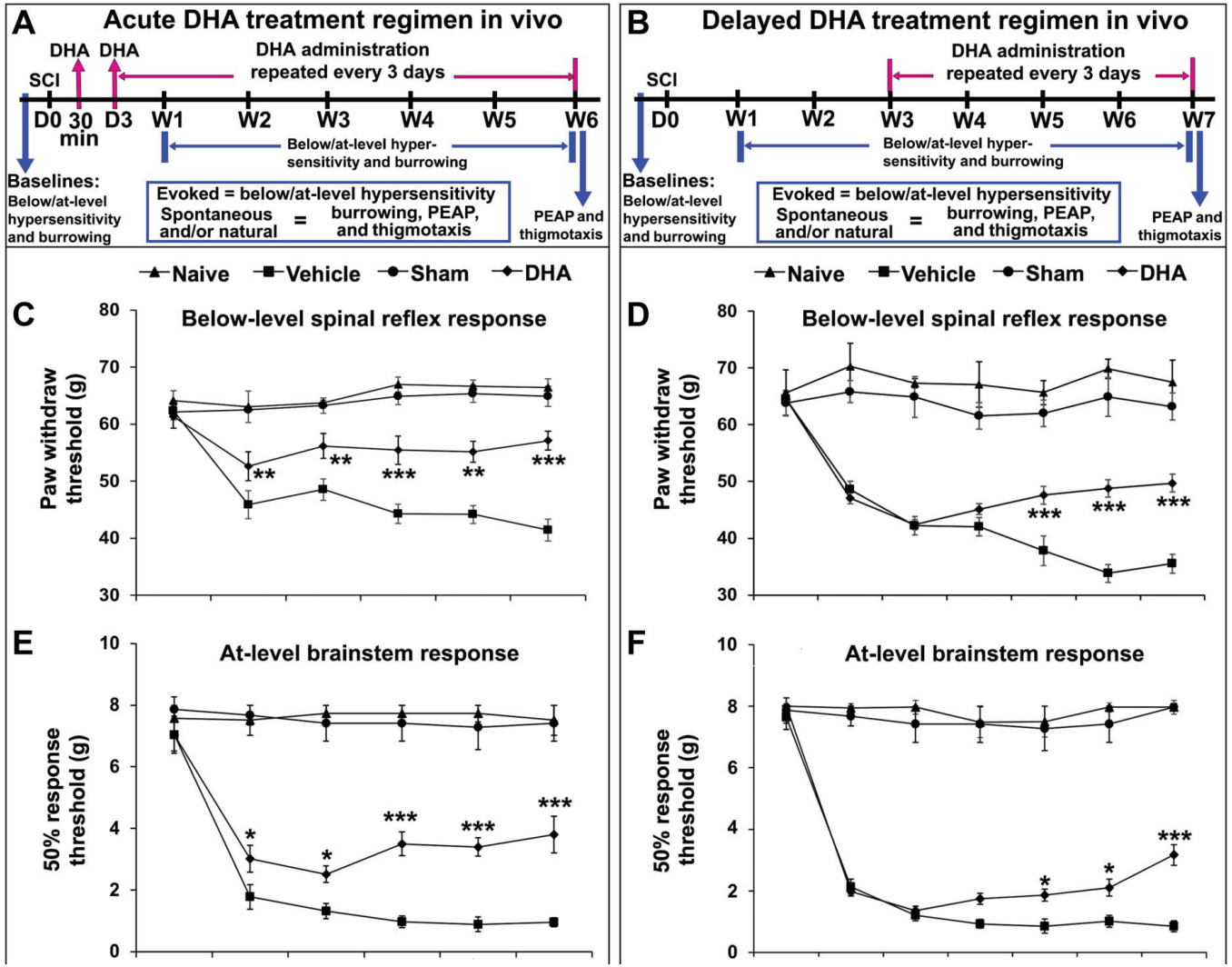
(Yip et al., Hum Mol Gen, 2019)

Impact of DHA on spinal cord injury complications

PAIN

DHA and neuropathic pain after contusion injury in the rat

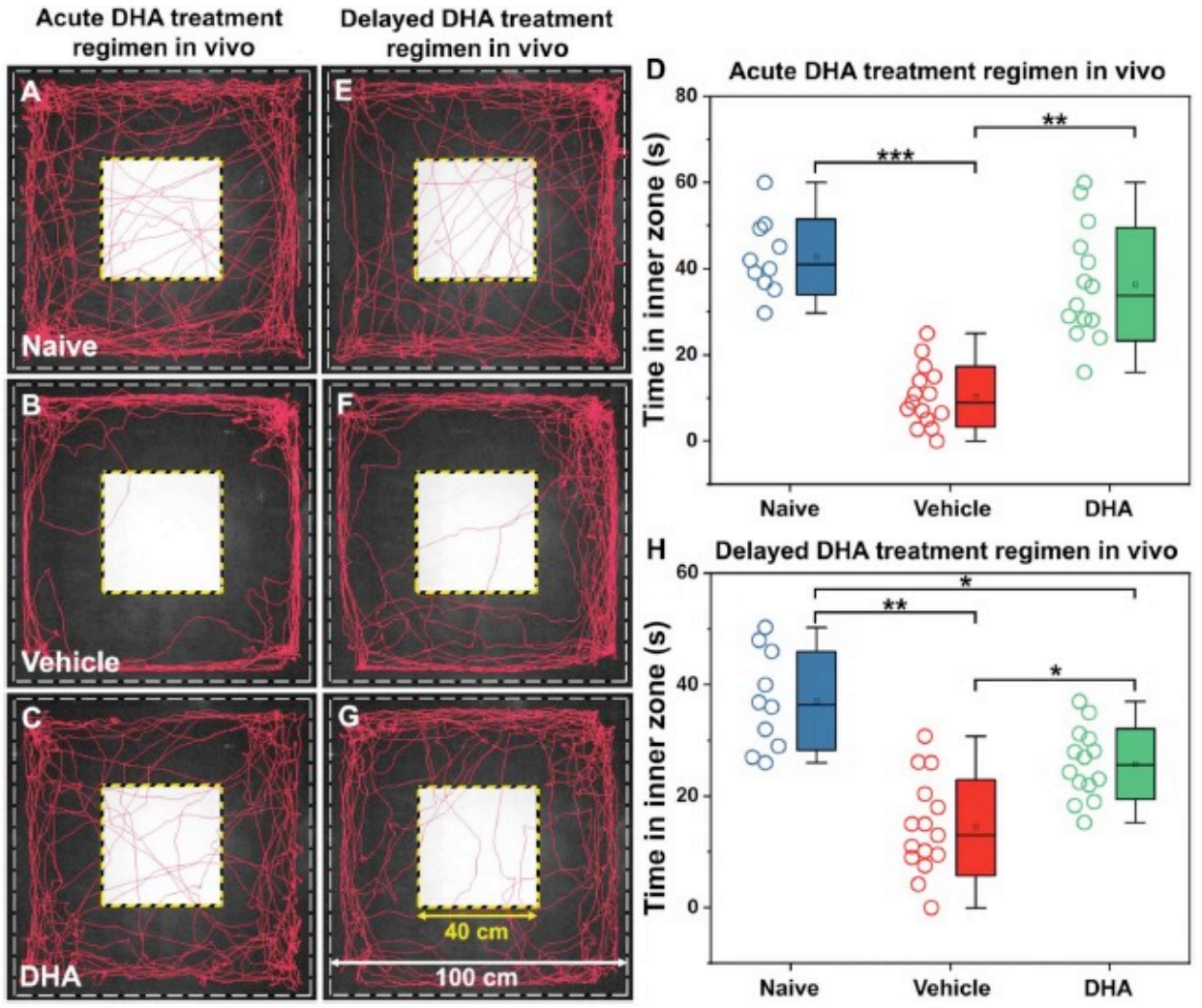
Delayed administration still has efficacy



(Georgieva et al., Pain, 2019)

DHA and neuropathic pain

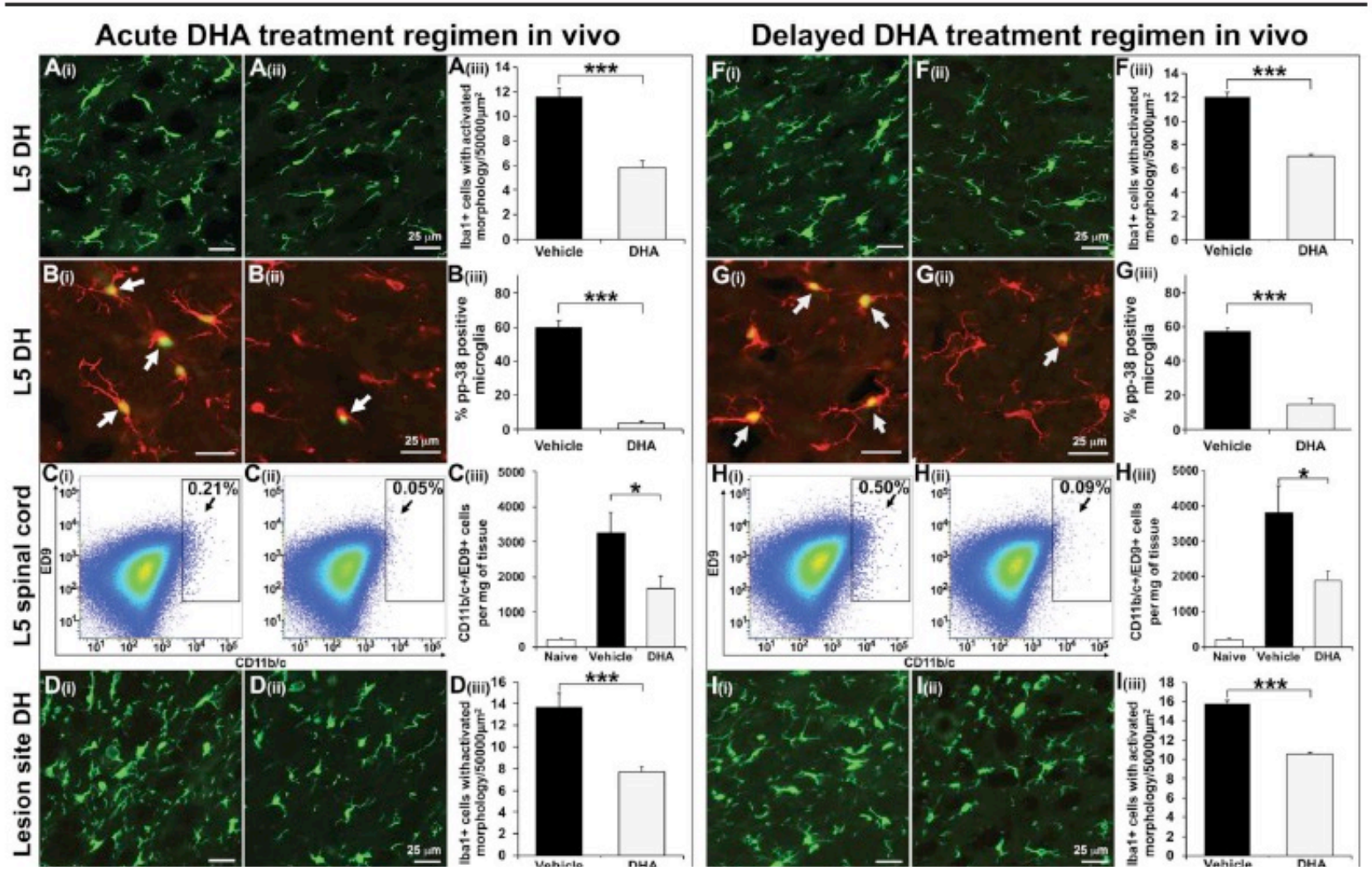
Treatment changes the altered exploration behaviour post-injury



(Georgieva et al., Pain, 2019)

DHA and neuropathic pain

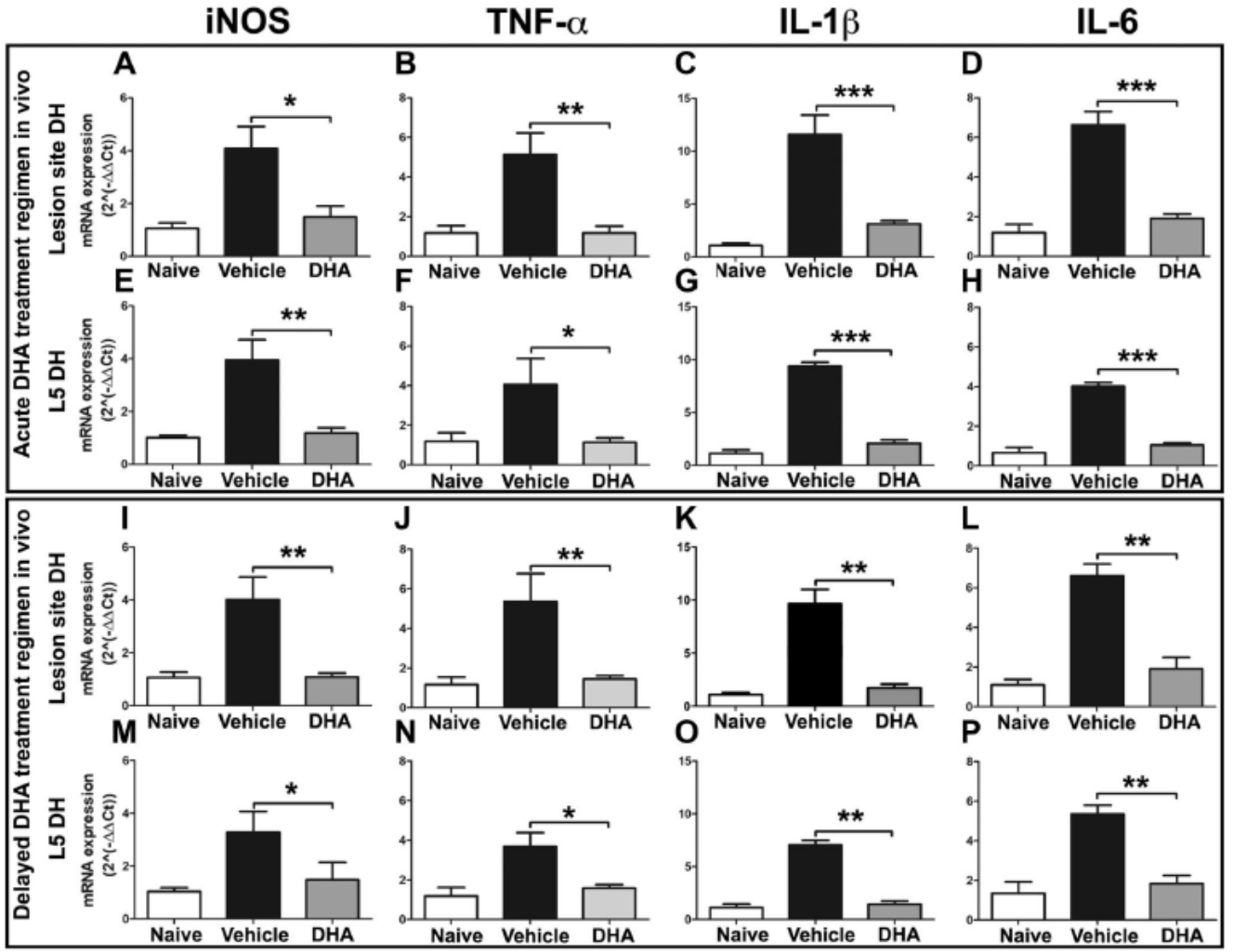
Reduction in the microglia/macrophage response



(Georgieva et al., Pain, 2019)

DHA and neuropathic pain

Reduction in expression of pro-inflammatory cytokines



Week 6 post-injury

(Georgieva et al., Pain, 2019)

DHA and neuropathic pain

Comparison with anti-neuropathic pain medication

Table 2

Behavioural outcome measures comparing pregabalin-treated animals with other experimental groups: Part A—at-level mechanical hypersensitivity, below-level mechanical hypersensitivity, and burrowing outcome measures.

Behavioural outcome measures	Treatment regimen	Behavioural assessment time points							
		Week 3 after surgery (mean ± SE)				Week 6 after surgery (mean ± SE)			
		Naive	DHA	Vehicle	Pregabalin	Naive	DHA	Vehicle	Pregabalin
At-level mechanical hypersensitivity	Acute	7.8 ± 0.2*	2.5 ± 0.2	1.3 ± 0.1†	3.2 ± 0.1	7.5 ± 0.5*	3.9 ± 0.6	1.1 ± 0.1†	5.1 ± 0.5
	Delayed	n/a				7.9 ± 0.1*	2.2 ± 0.3	1.0 ± 0.2†	4.2 ± 0.4
Below-level mechanical hypersensitivity	Acute	63.5 ± 1.3*	55.5 ± 2.1	48.2 ± 1.8†	58.0 ± 2.1	66.8 ± 1.5*	57.2 ± 1.3	41.1 ± 1.6†	56.6 ± 2.5
	Delayed	n/a				68.9 ± 1.9*	47.9 ± 1.6	33.4 ± 1.4	53.0 ± 3.0
Burrowing behaviour	Acute	1980.7 ± 199.5‡	1524.7 ± 71.2	797.9 ± 135.4†	1379.5 ± 138.8	2173.0 ± 128.2	1816.8 ± 85.5	876.3 ± 78.4†	1548.2 ± 61.3
	Delayed	n/a				1888.2 ± 43.1§	1371.3 ± 93.3	897.4 ± 114.9†	1600.5 ± 66.8

Units are 50% response threshold (g), paw withdrawal threshold (g), and gravel displaced (g) for at-level mechanical hypersensitivity, below-level mechanical hypersensitivity, and burrowing behaviour respectively. One-way ANOVA followed by Tukey–Kramer post hoc multicomparison adjustment was used for statistical analysis.

* $P < 0.05$ vs DHA, vehicle, and pregabalin.

† $P < 0.05$ vs naive, DHA, and pregabalin.

‡ $P < 0.05$ vs pregabalin.

§ $P < 0.05$ vs DHA and pregabalin. Sham data are not included, as they are similar to naive.

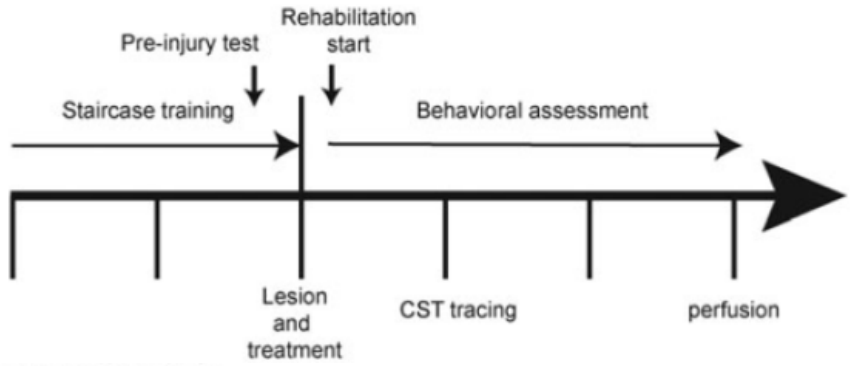
ANOVA, analysis of variance; DHA, docosahexaenoic acid.

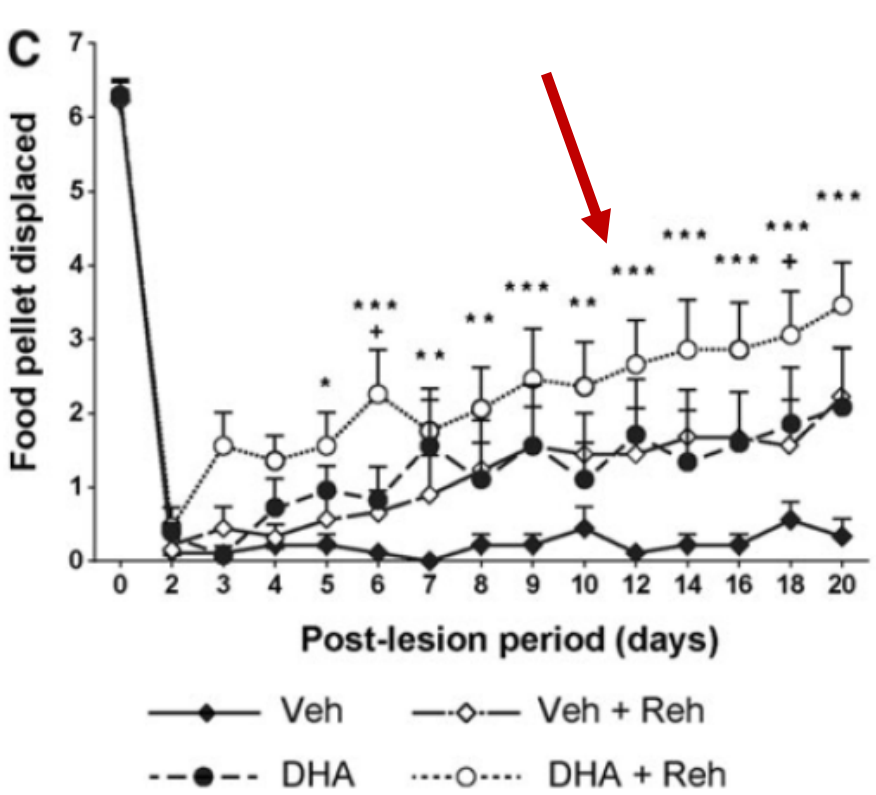
Note efficacy vs. pregabalin

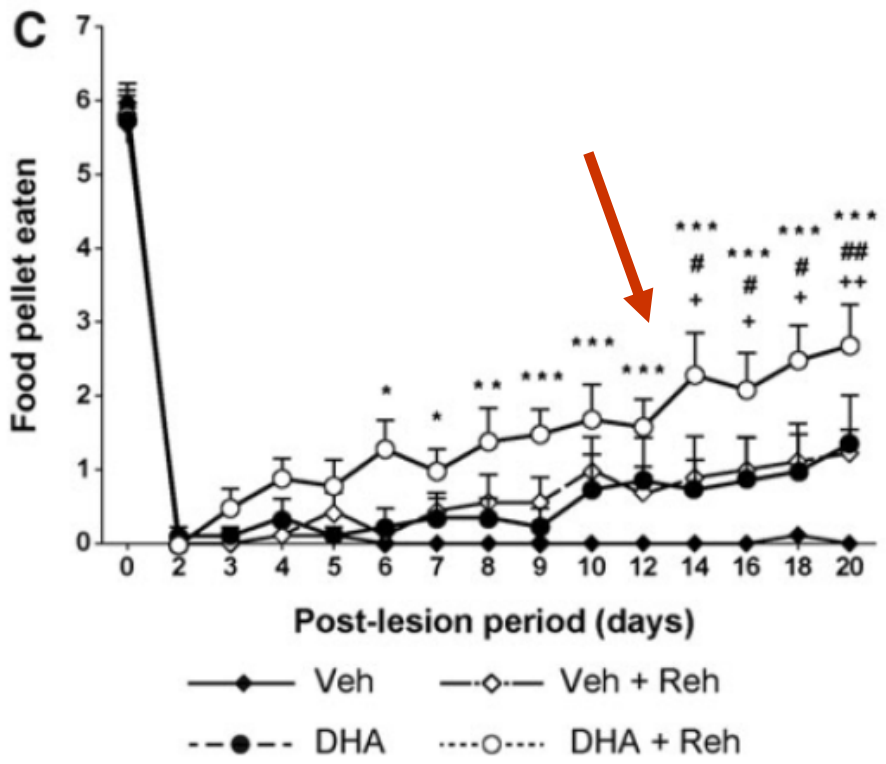
(Georgieva et al., Pain, 2019)

DHA synergises with rehabilitation

Rat cervical injury



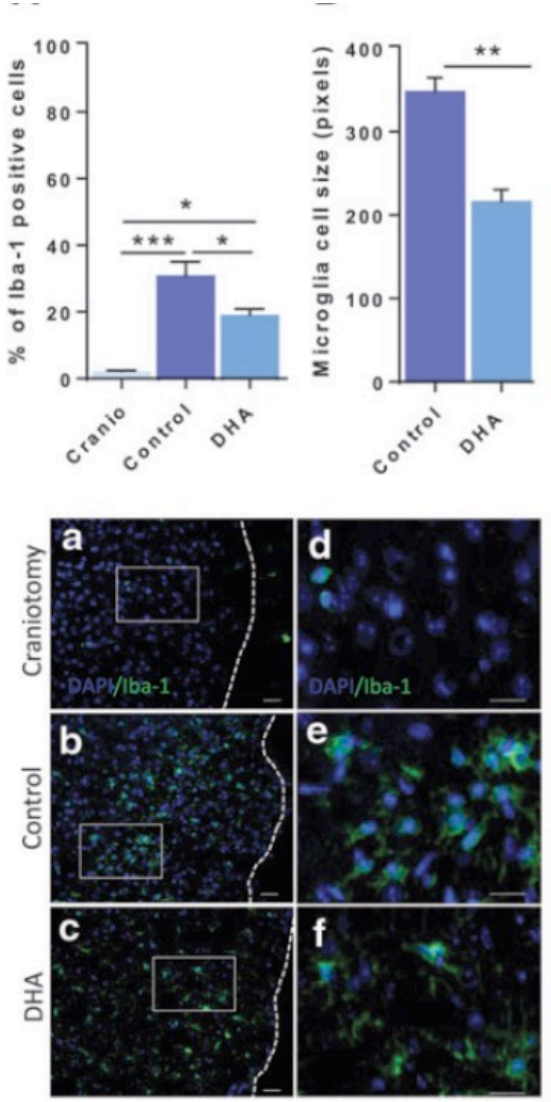


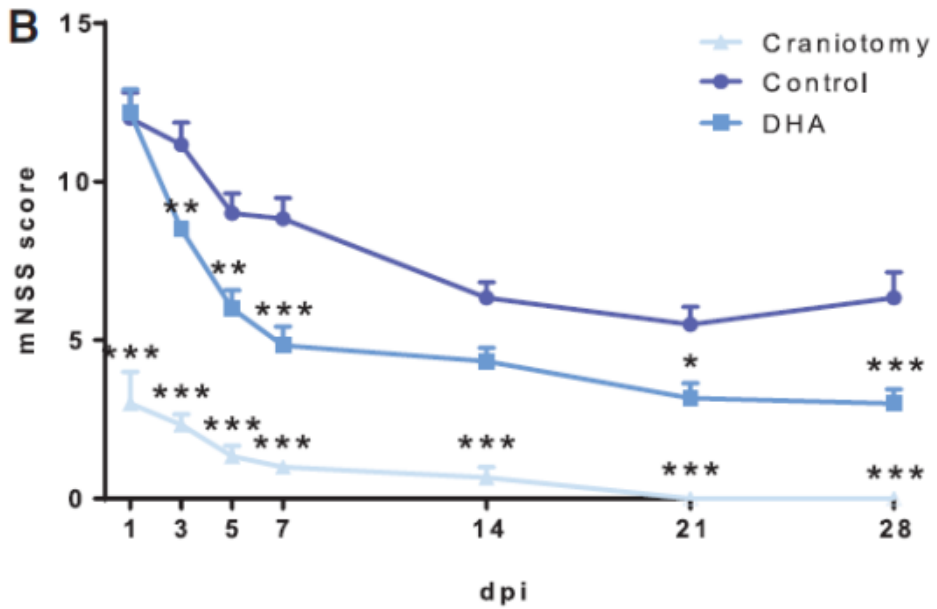


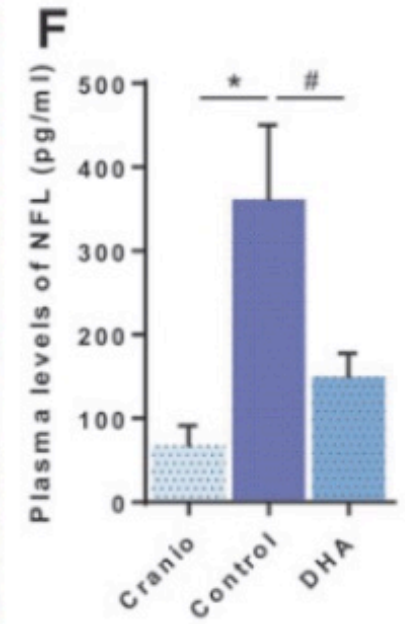
(Liu et al., J Neurotrauma, 2017)

Docosahexaenoic acid (DHA) for acute neuroprotection in traumatic brain injury

DHA in the controlled cortical impact (CCI) model in mice



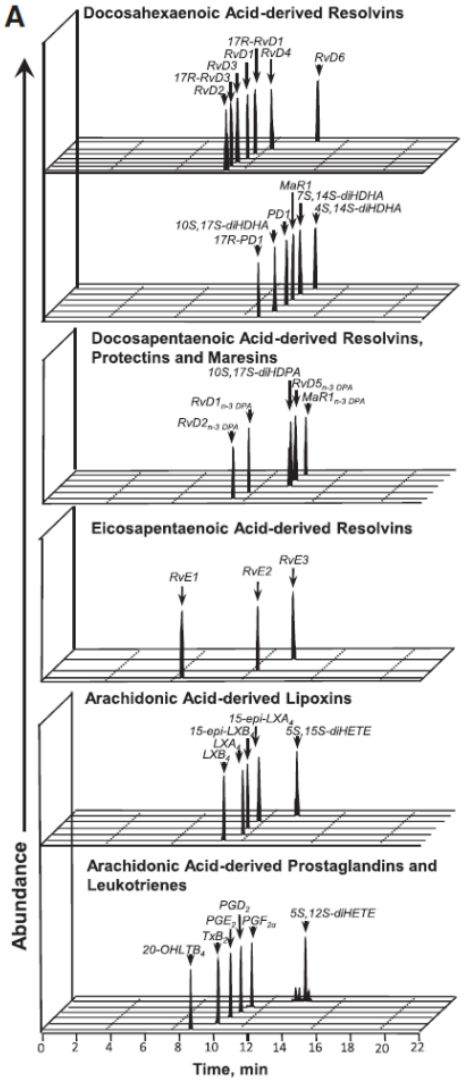


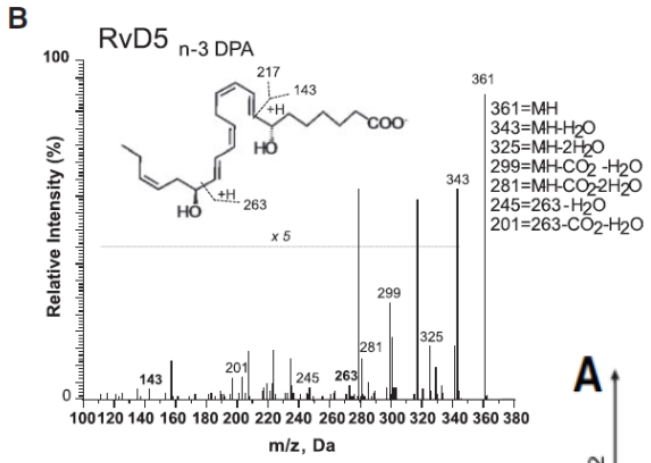


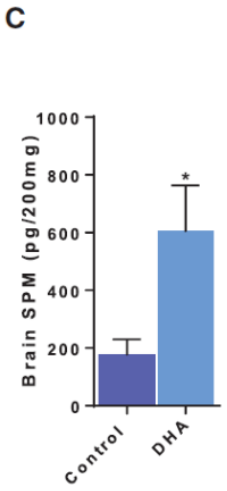
DHA 500 nmol/kg, i.v. 30 min post-injury

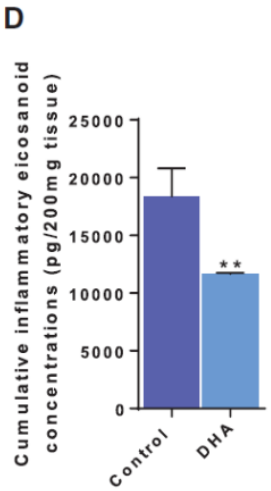
(Thau-Zuchman et al., J Neurotrauma, 2019)

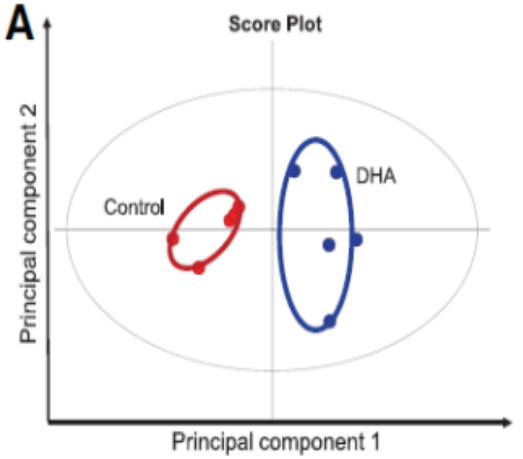
DHA and pro-resolving mediators after brain injury

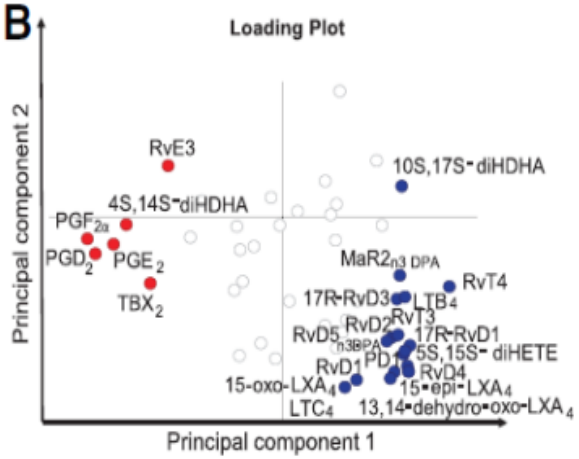






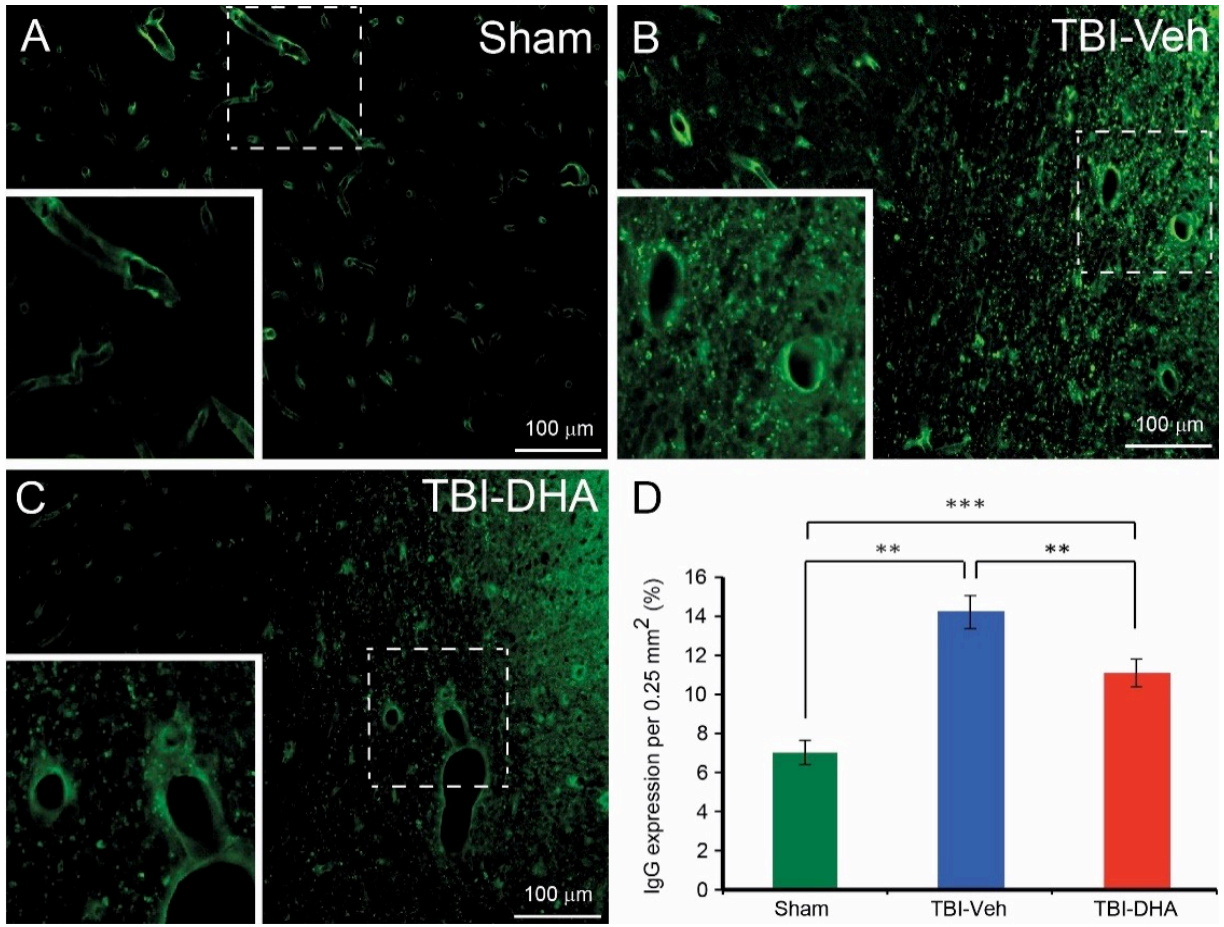


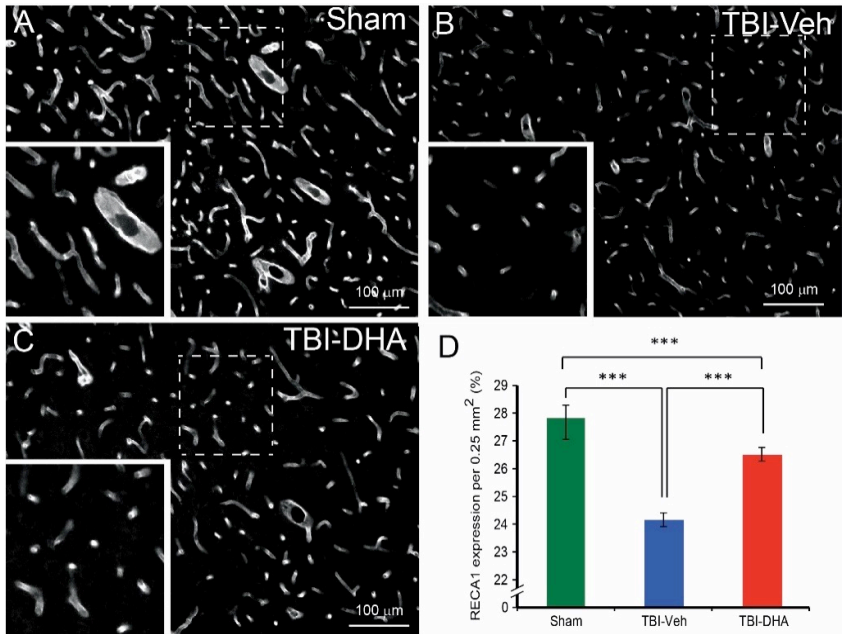


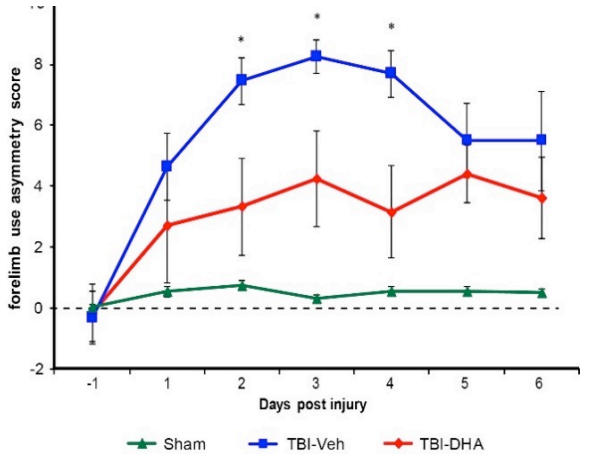


(Thau-Zuchman et al., J Neurotrauma, 2019)

DHA in the controlled cortical impact model (CCI) model in rats







DHA 500 nmol/kg, i.v. 30 min post-injury

(Liu et al., IJMS, 2020)

OMEGA-3 FATTY ACIDS IN NEUROLOGICAL INJURY

doi:10.1093/brain/awm223 Brain (2007), 130, 3004–301

A combination of intravenous and dietary docosahexaenoic acid significantly improves outcome after spinal cord injury

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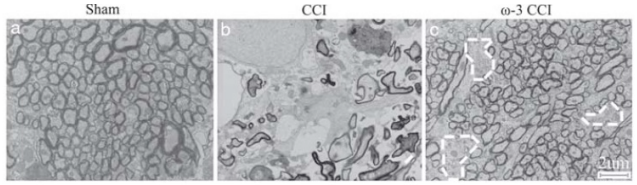
E-

ORIGINAL ARTICLE

Omega-3 polyunsaturated fatty acid supplementation improves neurologic recovery and attenuates white matter injury after experimental traumatic brain injury

Hongjian Pu[1], Yanling Guo[1], Wenting Zhang[1], Lanting Huang[1], Guohua Wang[1], Anthony K Liou[2,3], Jia Zhang[1], Pengyue Zhang[1], Rehana K Leak[4], Yun Wang[1], Jun Chen[1,2,3] and Yanqin Gao[1,2]



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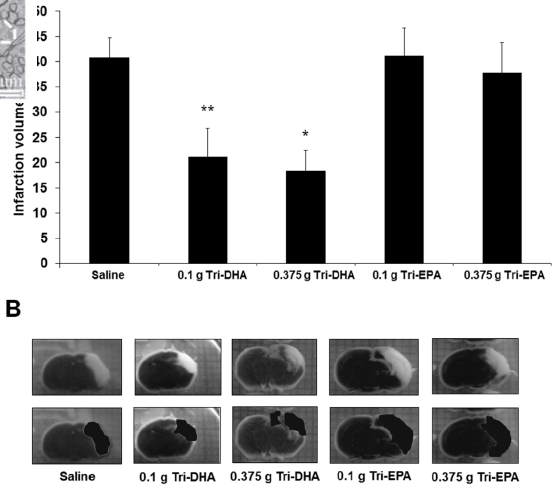
N-3 Fatty Acid Rich Triglyceride Emulsions Are Neuroprotective after Cerebral Hypoxic-Ischemic Injury in Neonatal Mice

Jill J. Williams[1¤], Korapat Mayurasakorn[1¤], Susan J. Vannucci[2], Christopher Mastropietro[3], Nicolas G. Bazan[4], Vadim S. Ten[5], Richard J. Deckelbaum[1*]

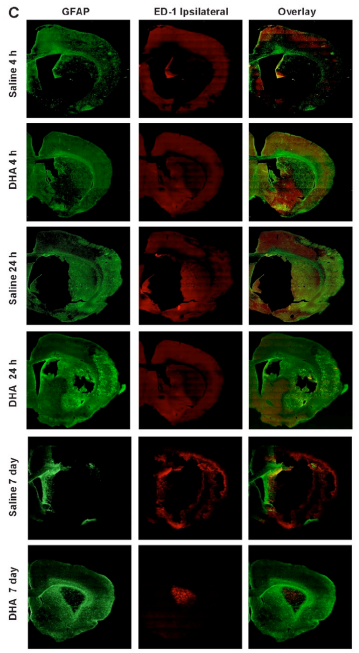


Inflammation (© 2018)
DOI: 10.1007/s10753-018-0765-z

ORIGINAL ARTICLE

Docosahexaenoic Acid (DHA) Provides Neuroprotection in Traumatic Brain Injury Models *via* Activating Nrf2-ARE Signaling

Wei Zhu[1,3] · Yueyia Ding[2] · Wei Kong,[1] Tuo Li,[1] and Hongguang Chen[1]

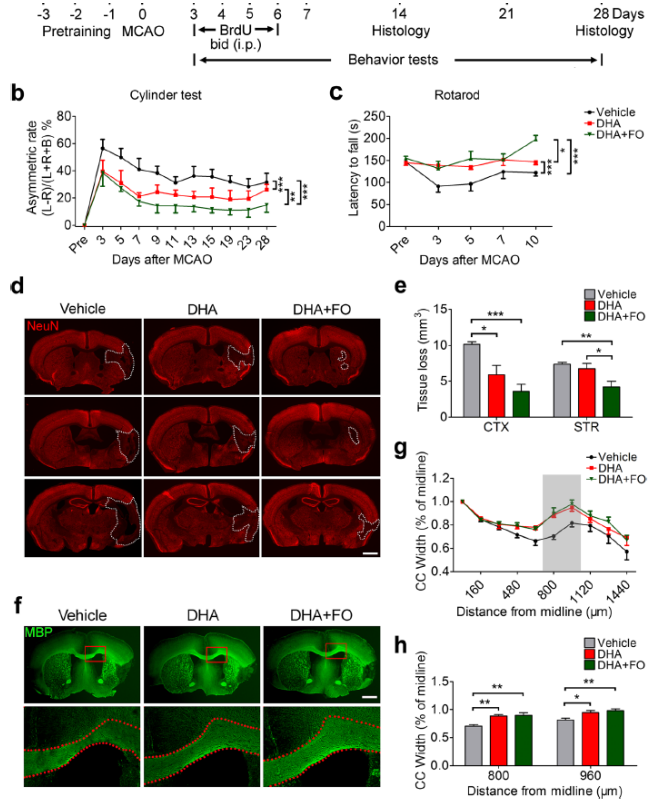


Transl. Stroke Res. (2016) 7:548–561
DOI 10.1007/s12975-016-0502-6

ORIGINAL ARTICLE

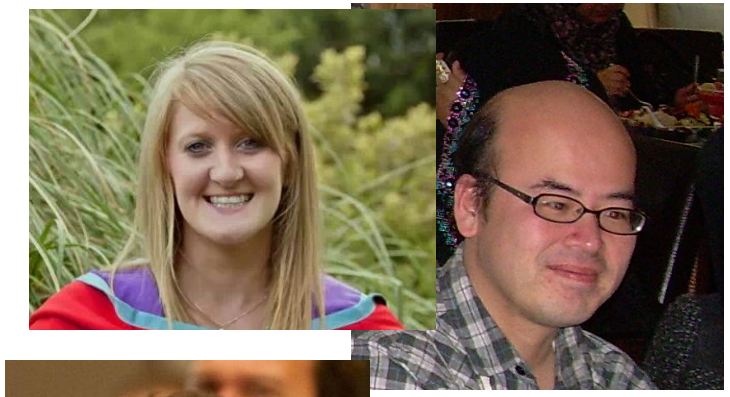
A Post-stroke Therapeutic Regimen with Omega-3 Polyunsaturated Fatty Acids that Promotes White Matter Integrity and Beneficial Microglial Responses after Cerebral Ischemia

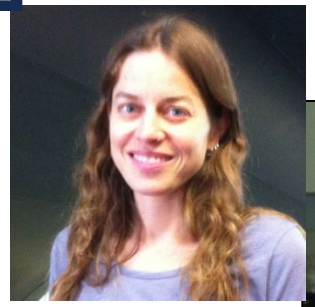
Xiaoyan Jiang[1,2] · Hongjian Pu[2] · Xiaoming Hu[1,2,3] · Zhishuo Wei[2] · Dandan Hong[2] · Wenting Zhang[1] · Yanqin Gao[1,2] · Jun Chen[1,2,3] · Yejie Shi[2,3]

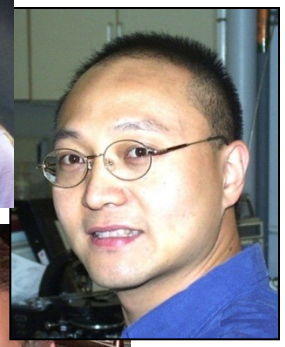


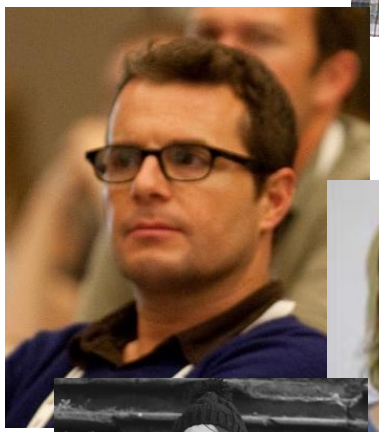
THANK YOU !

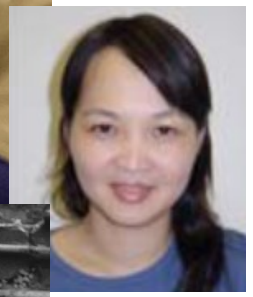
Prof John Priestley

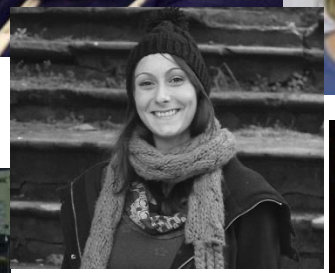












Ping Yip
Patrick Pallier
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SPINAL RESEARCH

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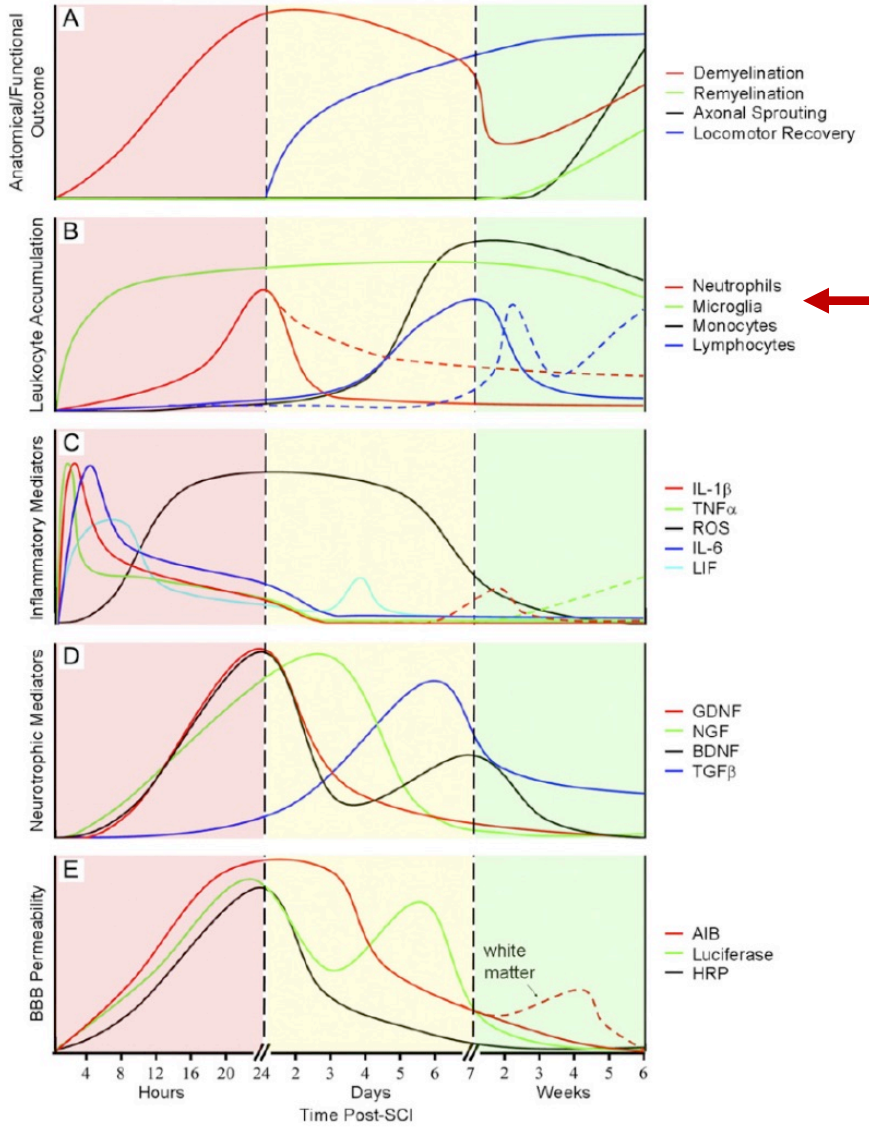
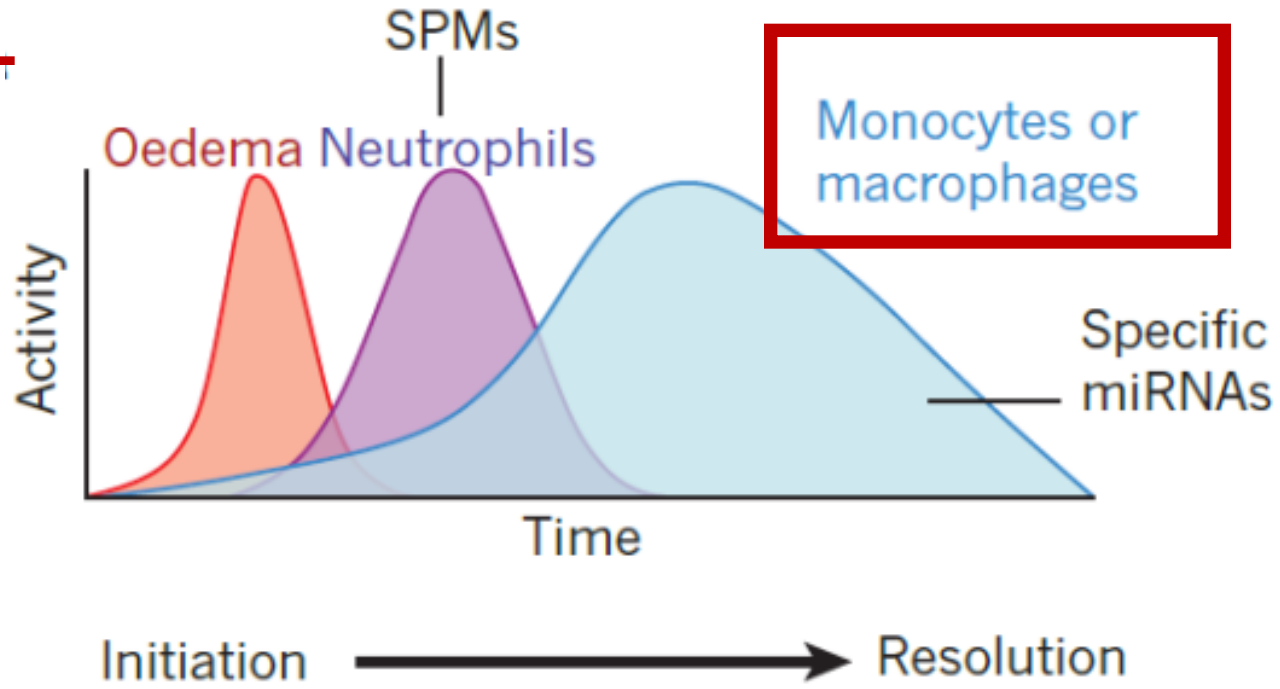
CDMRP
Department of Defense

DHA-derived lipid mediators in SCI

The omega-3 metabolome and the resolution of inflammation

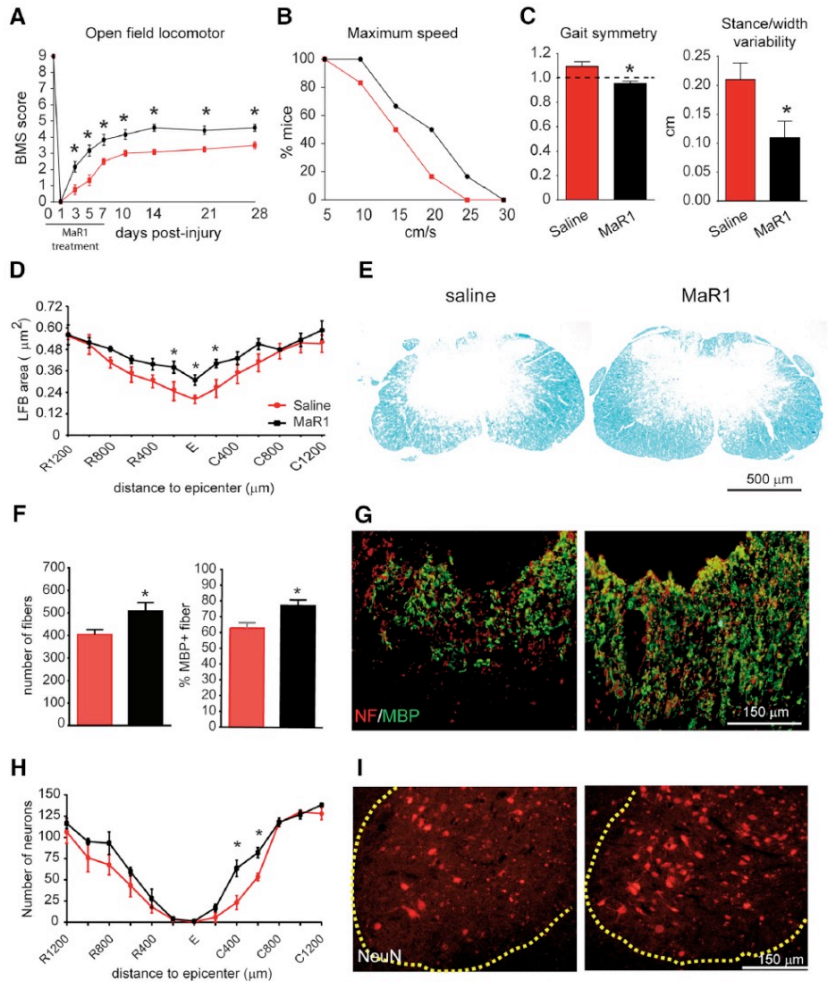


(Donnelly and Popovich, 2008)



(Serhan, Nature, 2014)

Maresin 1 – improvement in functional outcome and tissue protection



Mouse T10 contusion SCI 1 μg Mar1 i.v. ; one hour after SCI and then once a day until day 7

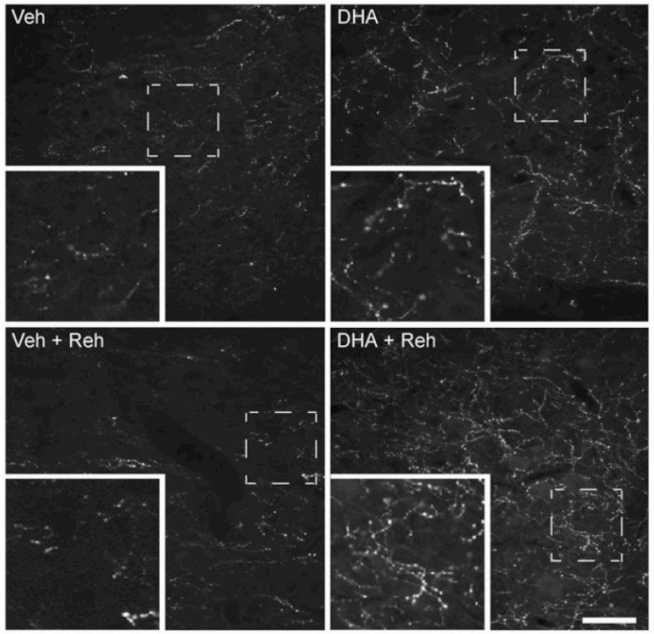
(Francos-Quijorna et al., 2017)

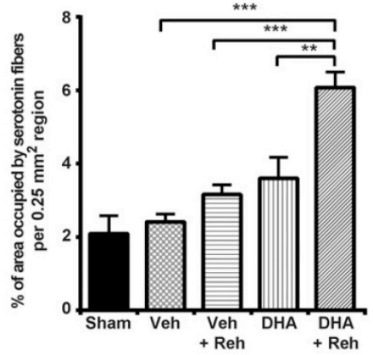
DHA synergises with rehabilitation

Serotonin fibers and synaptic markers

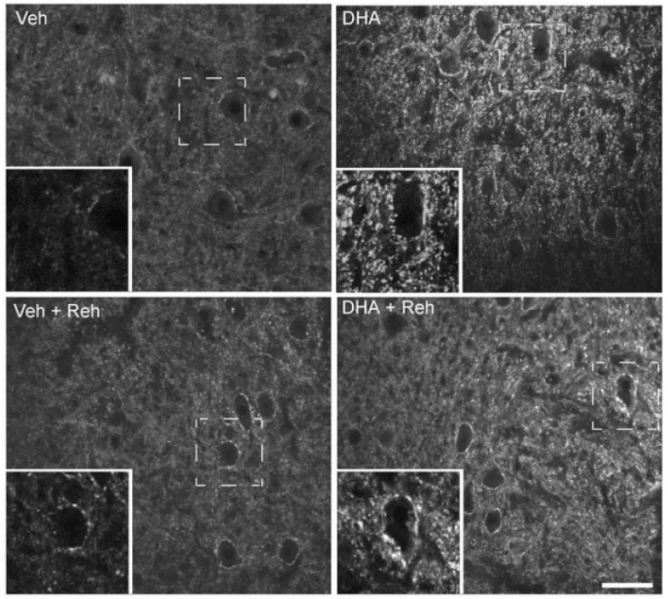
(Liu et al., J Neurotrauma, 2017)

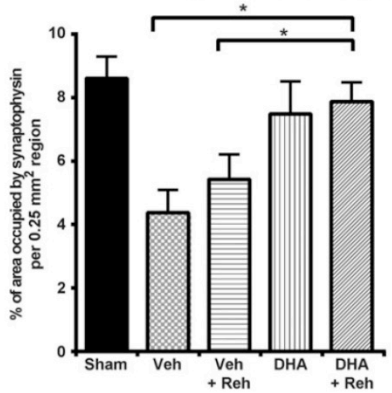
5-HT



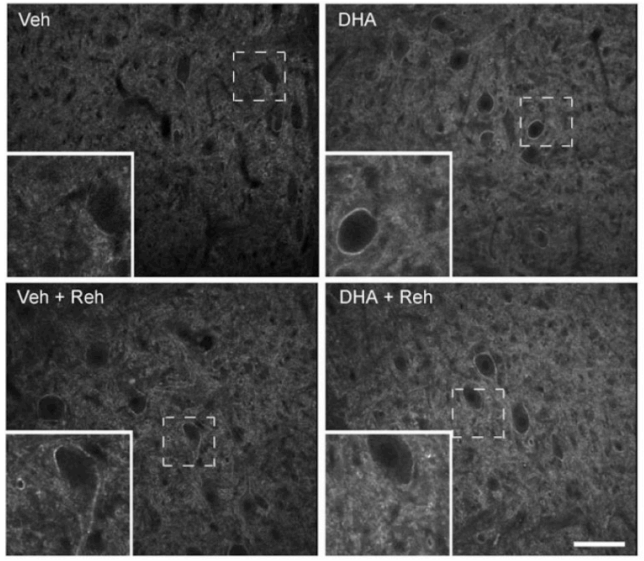


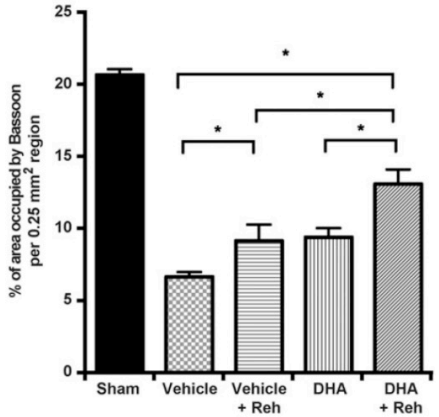
Synaptophysin





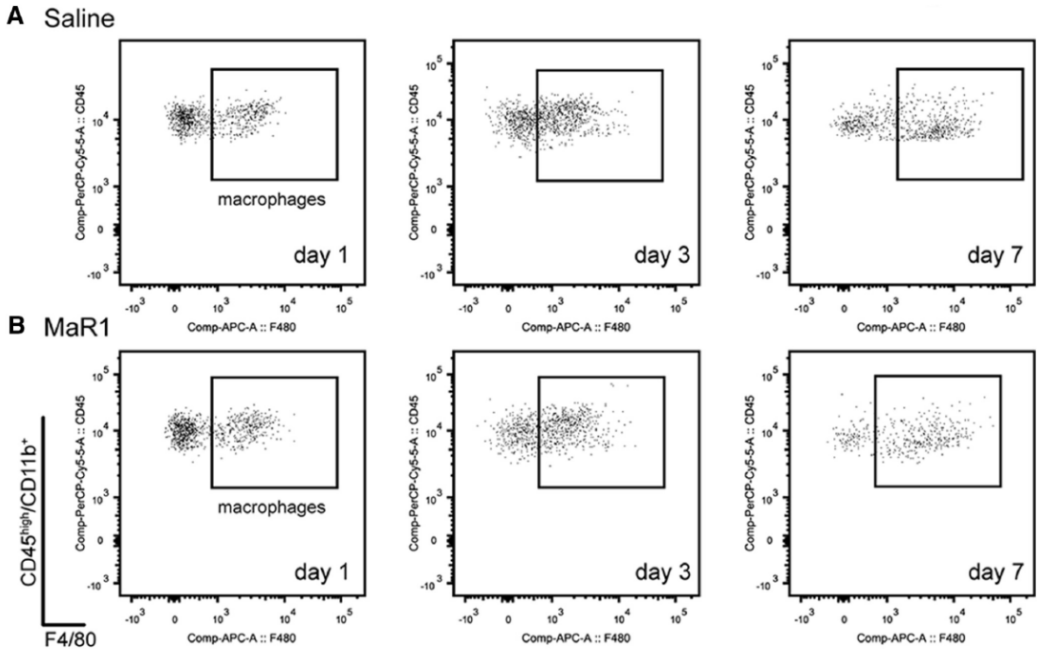
Bassoon

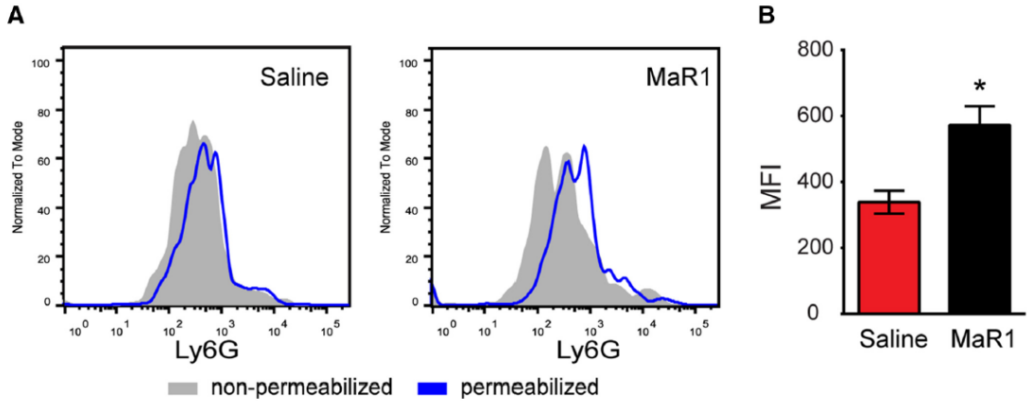




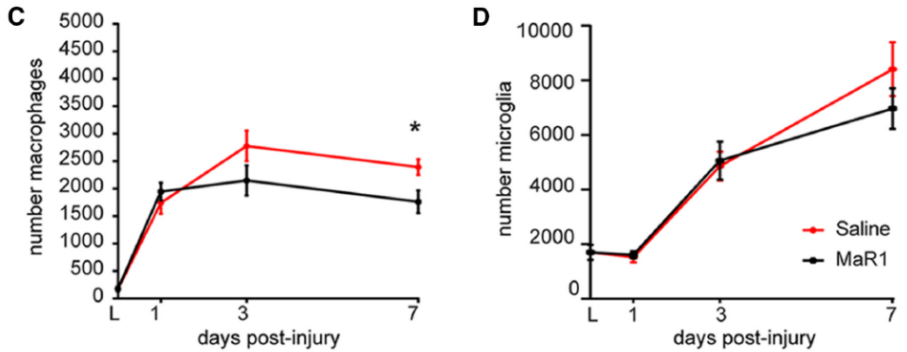
Maresin 1 – anti-inflammatory effect in SCI

Accelerated resolution of the inflammatory response





Maresin 1 increases the clearance of neutrophils



1 µg Mar1; one hour after SCI and then once a day until day 7

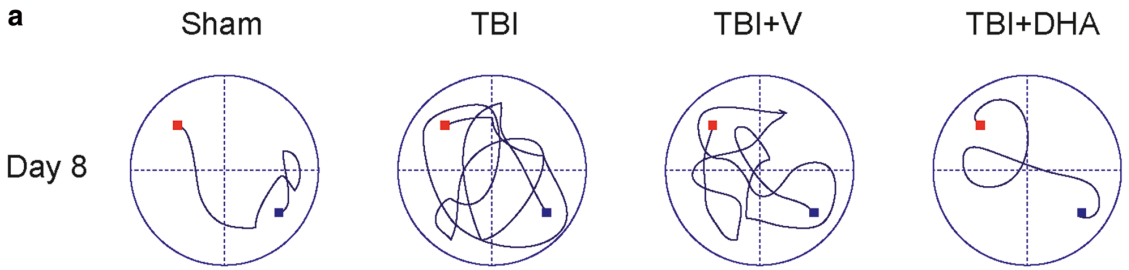
(Francos-Quijorna et al., 2017)

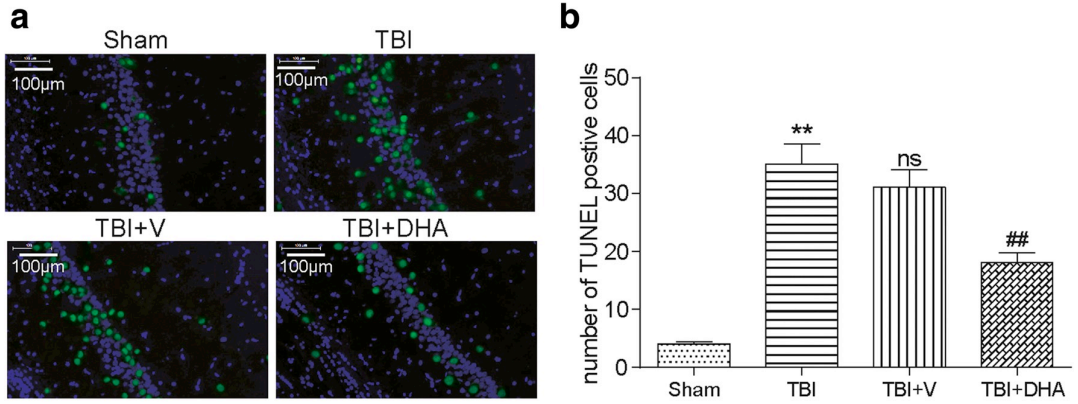
DHA IN NEUROLOGICAL INJURY

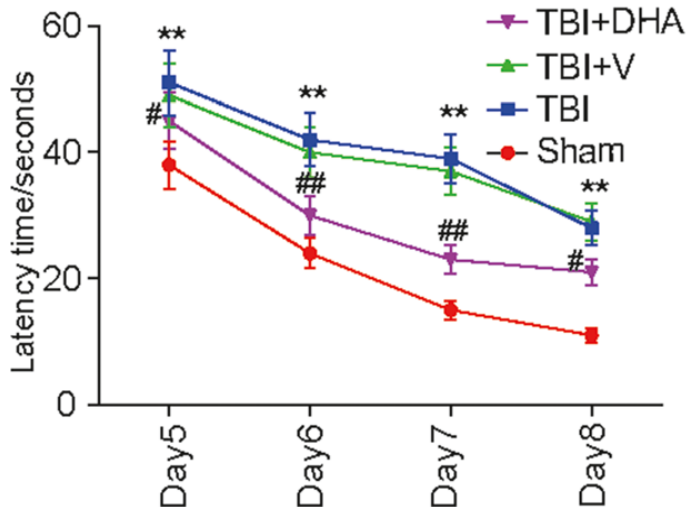
Processes targeted successfully

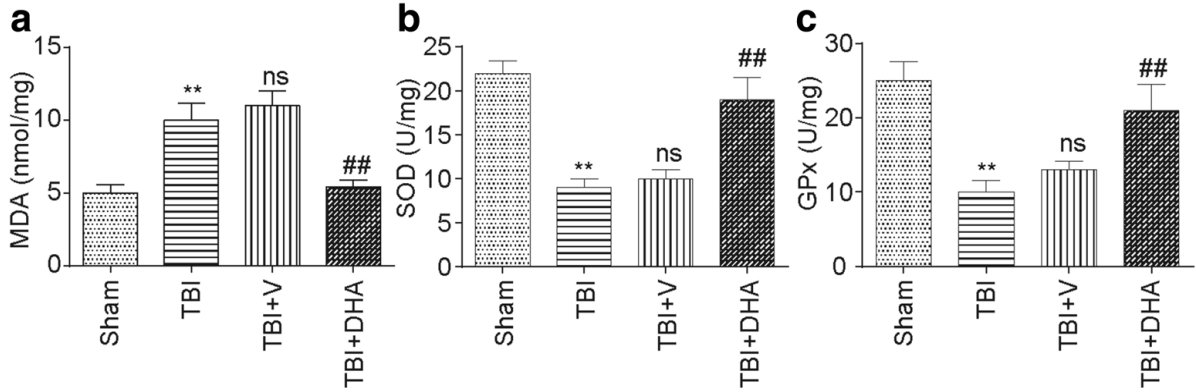
- Neuronal loss
- Oligodendrocyte loss
- Demyelination
- Astrogliosis
- Microglia and macrophage reaction to injury
- Vascular damage
- Protein and lipid oxidation
- Axonal sprouting

DHA in the fluid percussion injury (FPI) model in rats





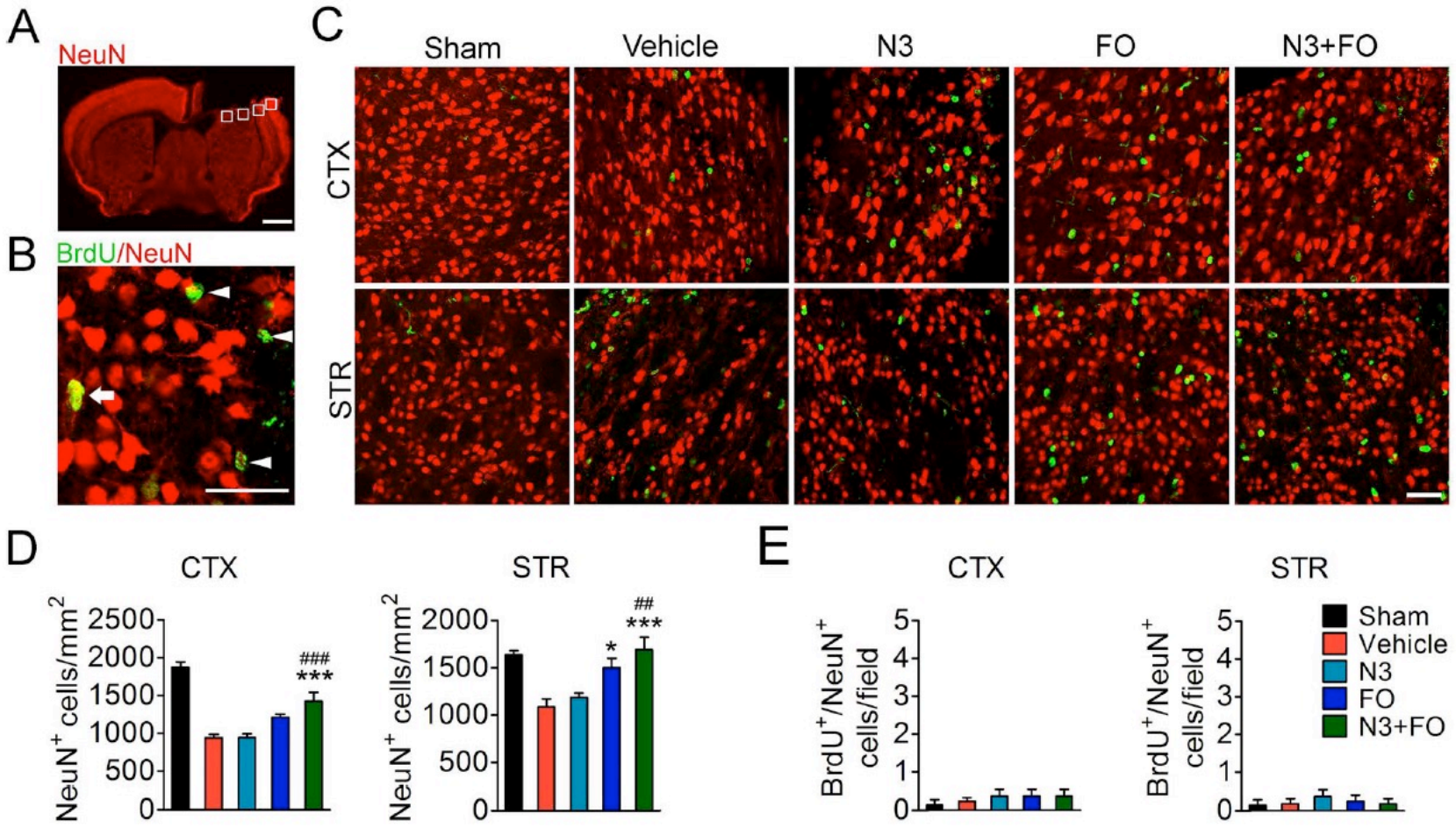




DHA oral gavage: 555 mg/kg/day- starting 30 min after the injury

(Zhu et al., Inflammation, 2018)

Omega-3 fatty acids in TBI - acute injection and oral supplementation



(Pu et al., 2017)

7 mg EPA and 3 mg DHA/kg, i.p. 2h after TBI and repeated for 14 days

DHA and EPA oil; up to 4% in diet, started 1 dat after TBI and continued for 35 days

Fat-1 Transgenic Mice with Constitutive High Levels of Omega-3 FA

- Express the *Caenorhabditis elegans* fat-1 gene encoding a n-3 fatty acid desaturase

n-6 FA: CH_3-CH_2-CH_2-CH_2-CH_2-CH =CH- … -COOH

↓ ***n-3 desaturase***

n-3 FA: CH_3-CH_2-CH =CH-CH_2-CH =CH- … -COOH

Collaboration with Dr. J. Kang (Harvard Medical School)

<u>3 groups:</u>

1) WT normal diet
2) WT omega-6 diet
3) Fat-1 omega-6 diet

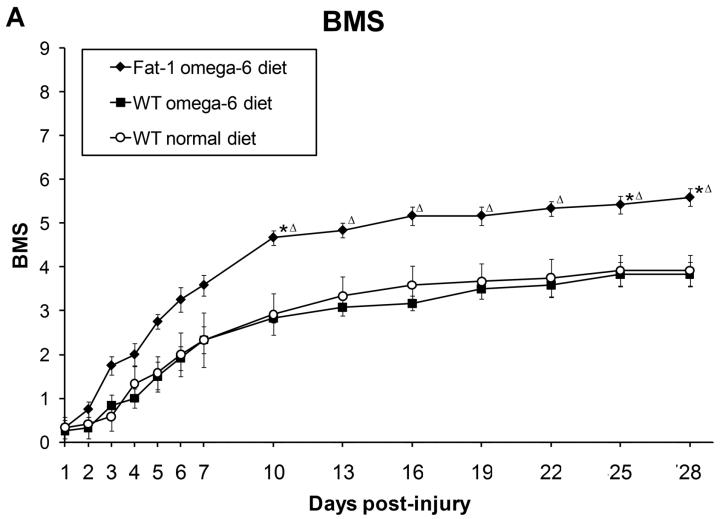
Table 1 **Fatty-acid ratios in WT and *fat*-1 mice**

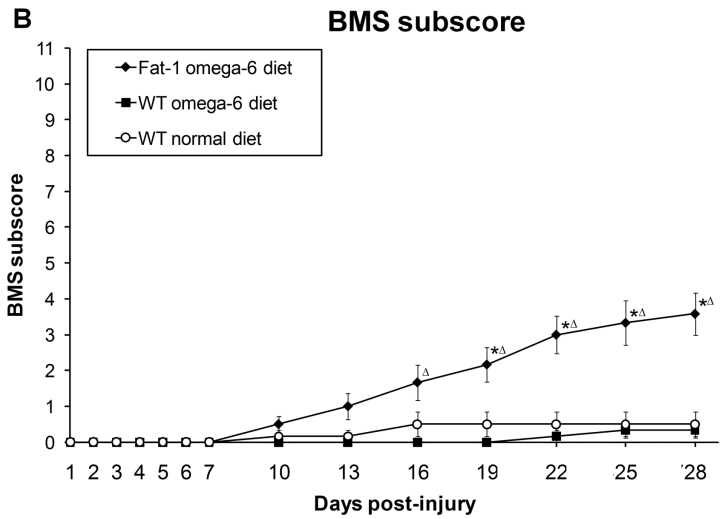
	n-6/n-3*		AA/(EPA+DPA+DHA)	
	WT	*TM*	*WT*	*TM*
Muscle	49.0	0.7	11.3	0.4
Milk†	32.7	5.7	15.7	2.5
Erythrocyte	46.6	2.9	27.0	1.6
Heart	22.8	1.8	14.3	0.9
Brain	3.9	0.8	3.6	0.7
Liver	26.0	2.5	12.5	0.9
Kidney	16.5	1.7	11.9	1.2
Lung	32.3	2.2	19.8	1.2
Spleen	23.8	2.4	17.3	1.5

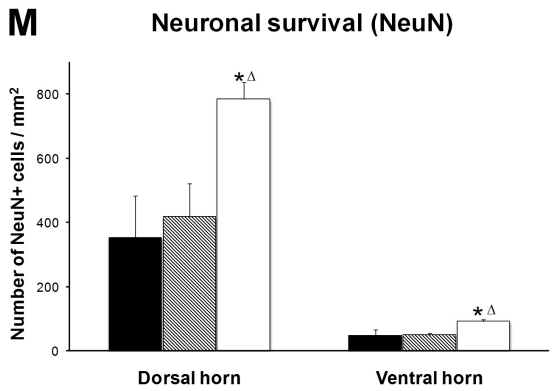
Outcome of compression SCI in *Fat-1* Transgenic Mice

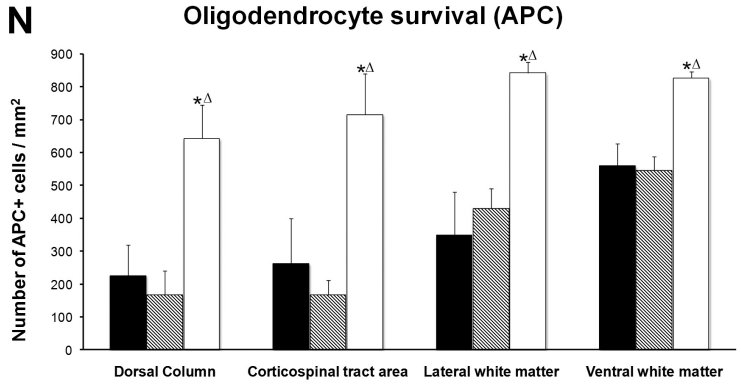
3 groups:

1) WT normal diet
2) WT omega-6 diet
3) Fat-1 omega-6 diet





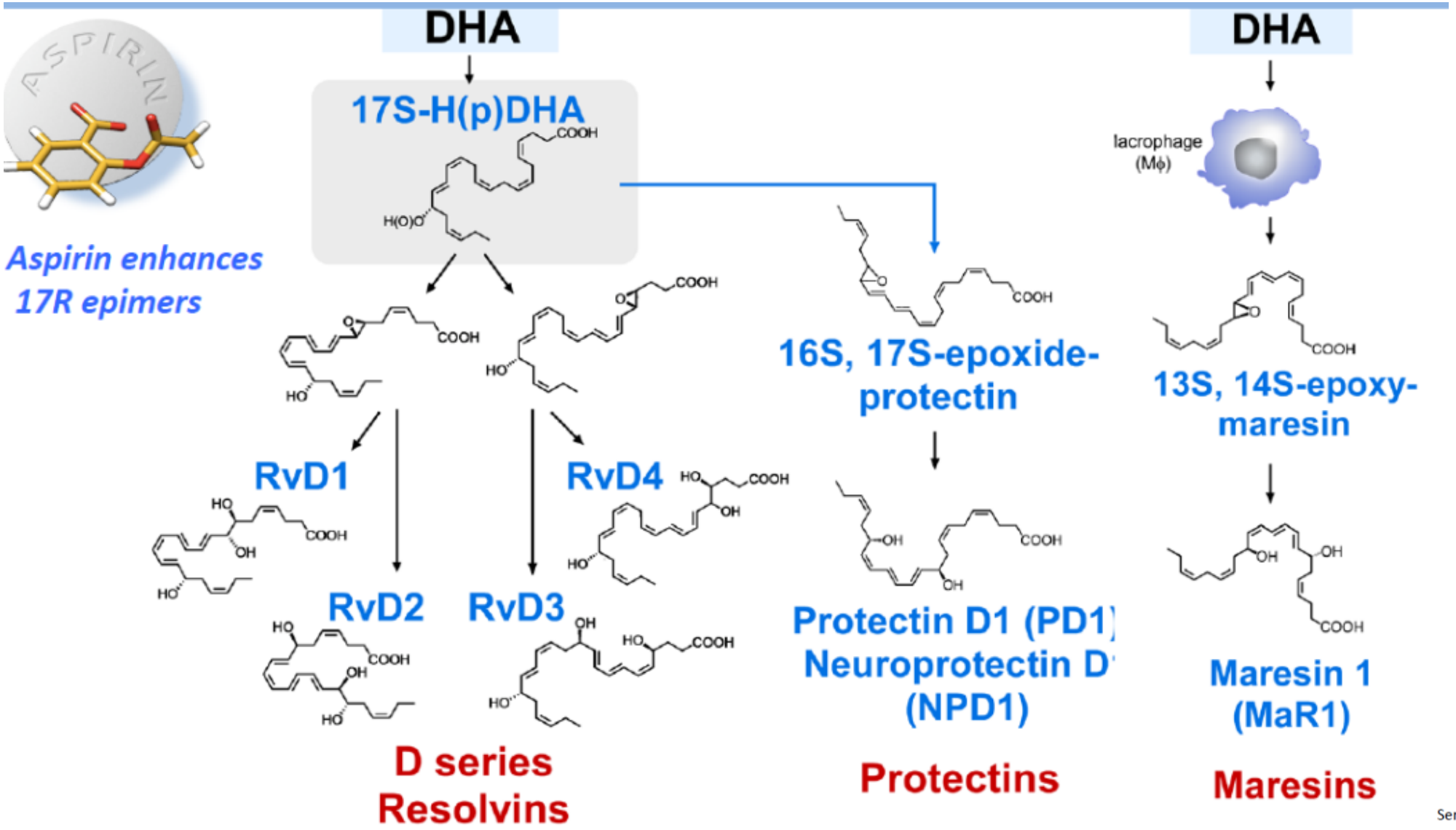




■ WT normal diet ■ WT omega-6 diet □ *Fat-1* omega-6 diet

Note the much improved outcome of the fat-1 mice

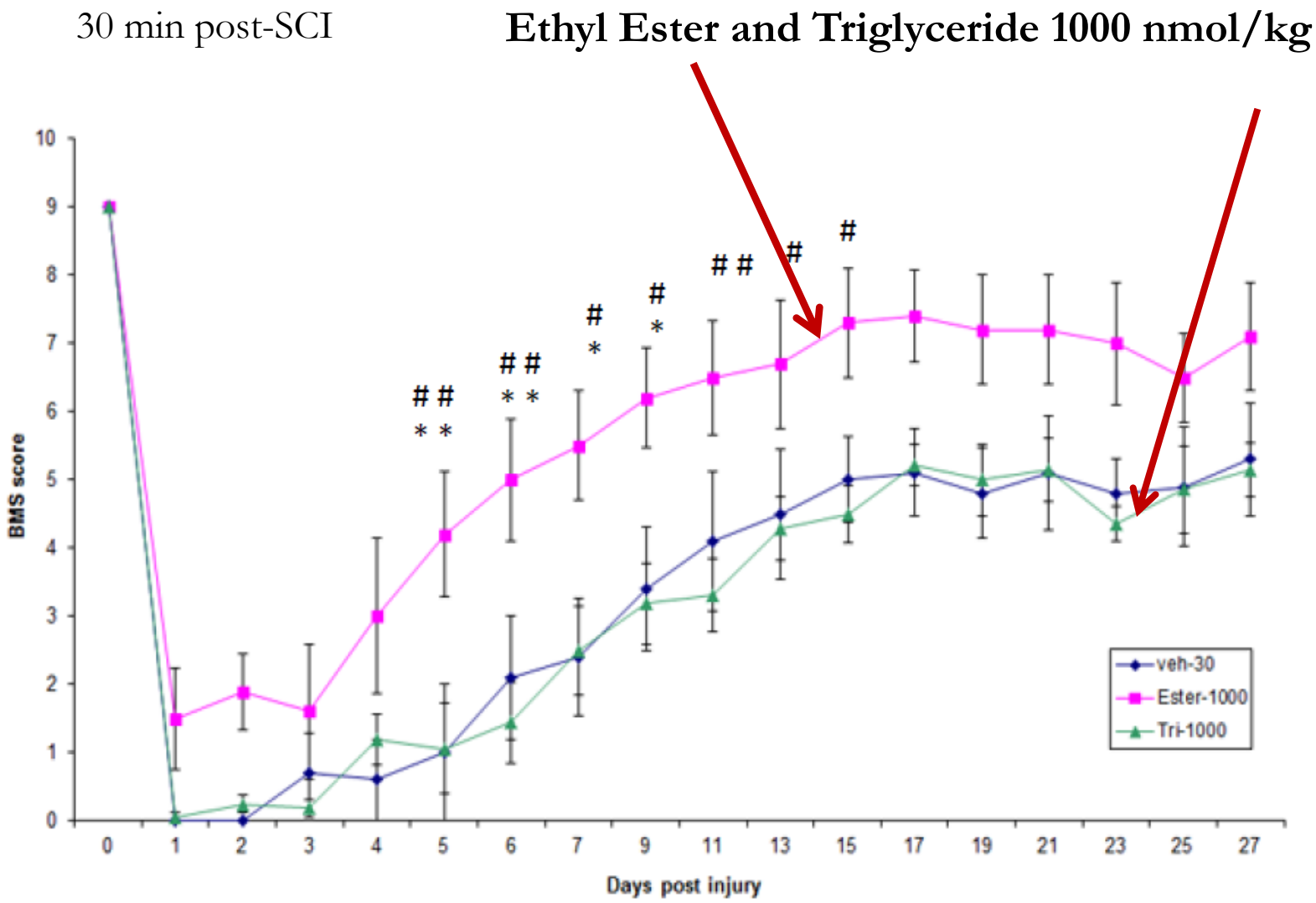
DHA-DERIVED LIPID MEDIATORS



Serhan CN. *Nature* 2014;510:92-101

DHA in mouse thoracic contusion injury

Efficacy of different esterified forms of DHA



P. Yip, unpublished

WHY ARE WE INTERESTED IN CLINICAL TRANSLATION IN NEUROTRAUMA?

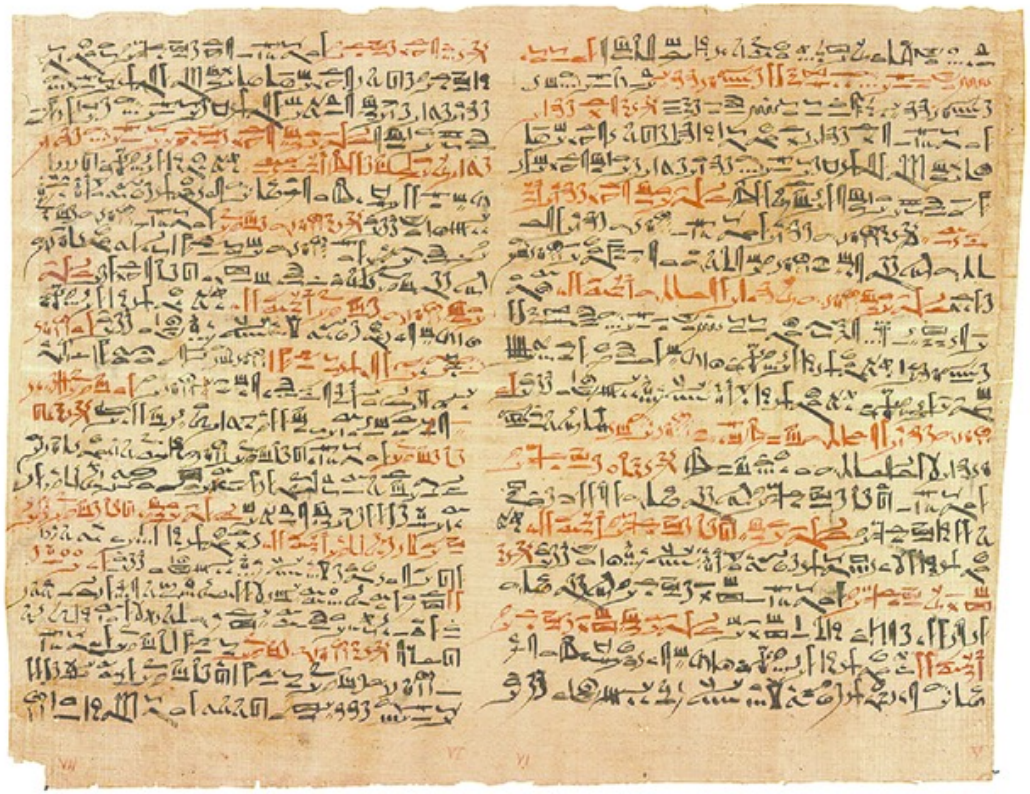
Level One Trauma Centre

Royal London Hospital - >2,000 trauma patients/year

Neurotrauma in a context of polytrauma

Intervention in the "golden hour"

The Edwin Smith papyrus



Egyptian physician Imhotep, 3rd dynasty

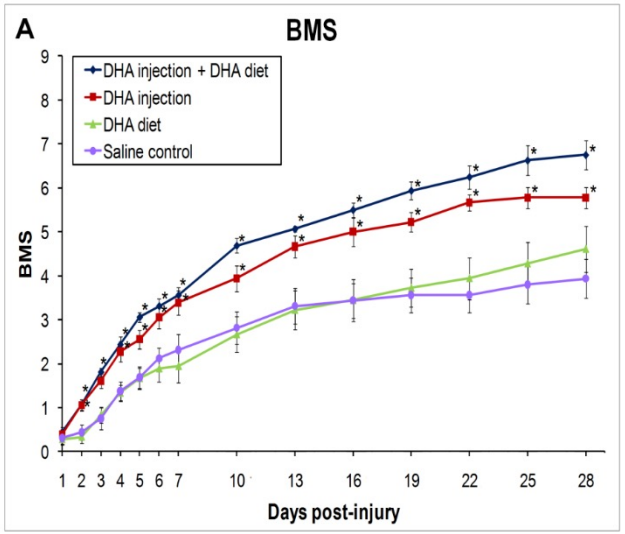
Examination If you should examine a man having a crushed vertebra in the back of neck and you find him with one vertebra fallen into its counterpart, and now he is stuporous and he does not speak. It is his fall head downward which caused a vertebra to crush into its counterpart and you find he is unaware of both his arms and his legs because of it.

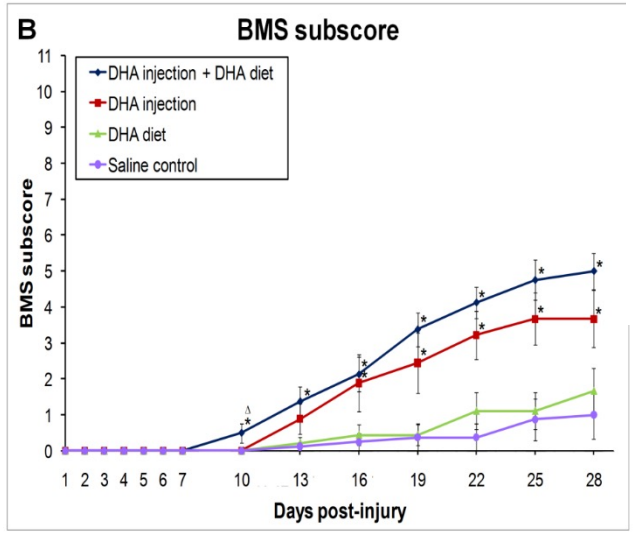
Diagnosis Then you are to say about him: "One who has a crushed vertebra in the back of his neck, and he is unaware of both his arms and legs, and is stuporous (this is) a medical condition that cannot be healed".

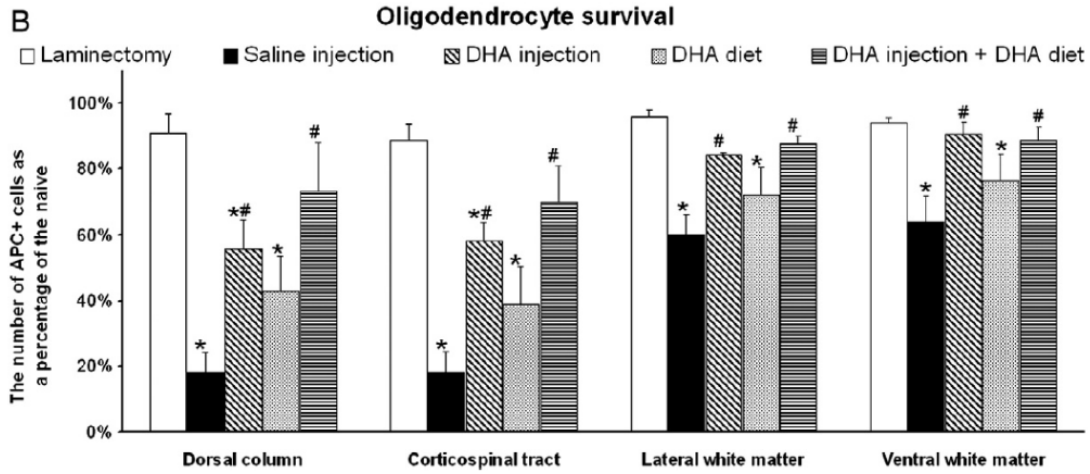
"(This is) a medical condition that cannot be healed."

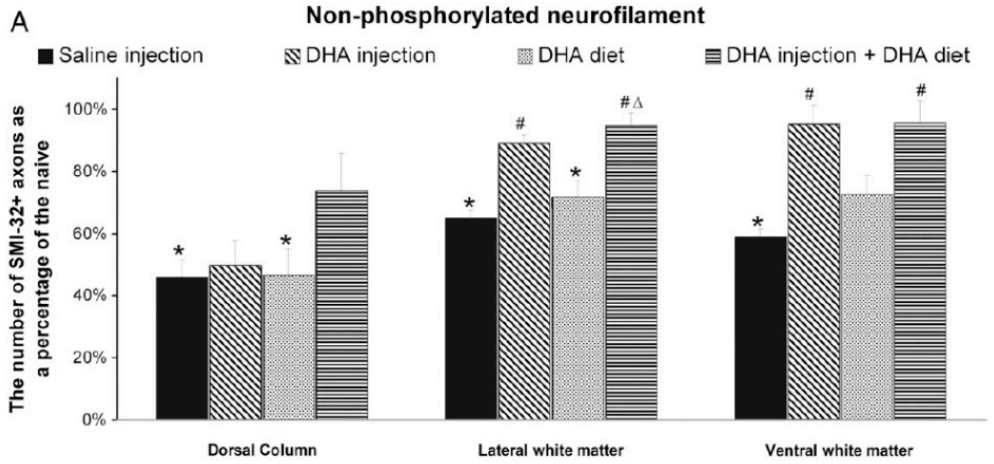
Confirmation of DHA effect in a second species

DHA in mouse compression SCI







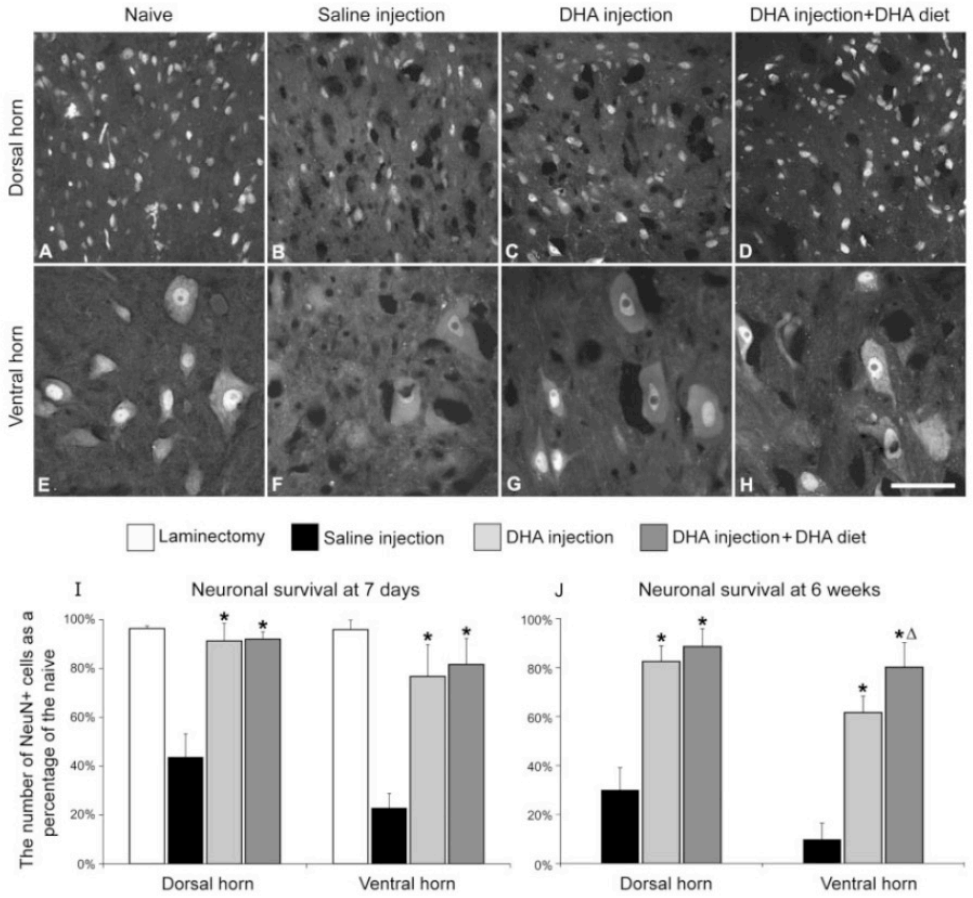


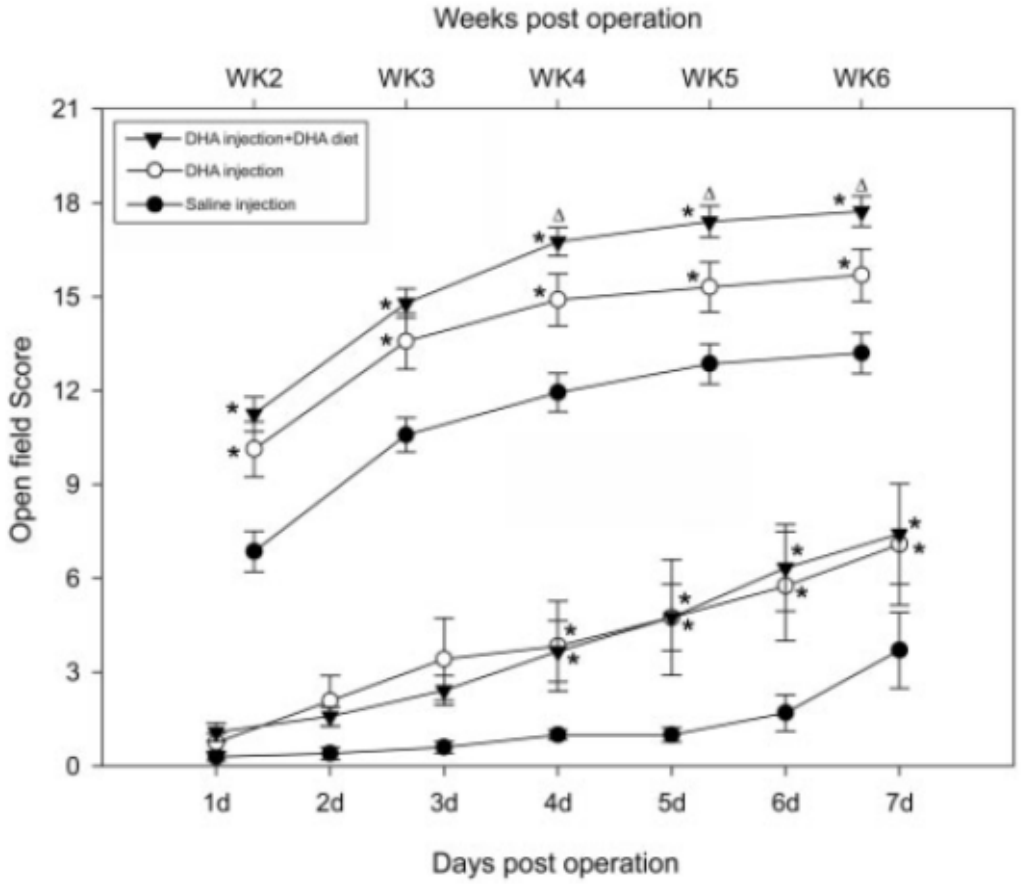
DHA 500 nmol/kg, i.v. 30 min after injury ; dietary DHA 400 mg/kg/day for 28 days

(Lim et al , Exp Neurol, 2013))

DHA induces improved neurological outcome and tissue protection in compression SCI

Rat - Compression injury - Thoracic



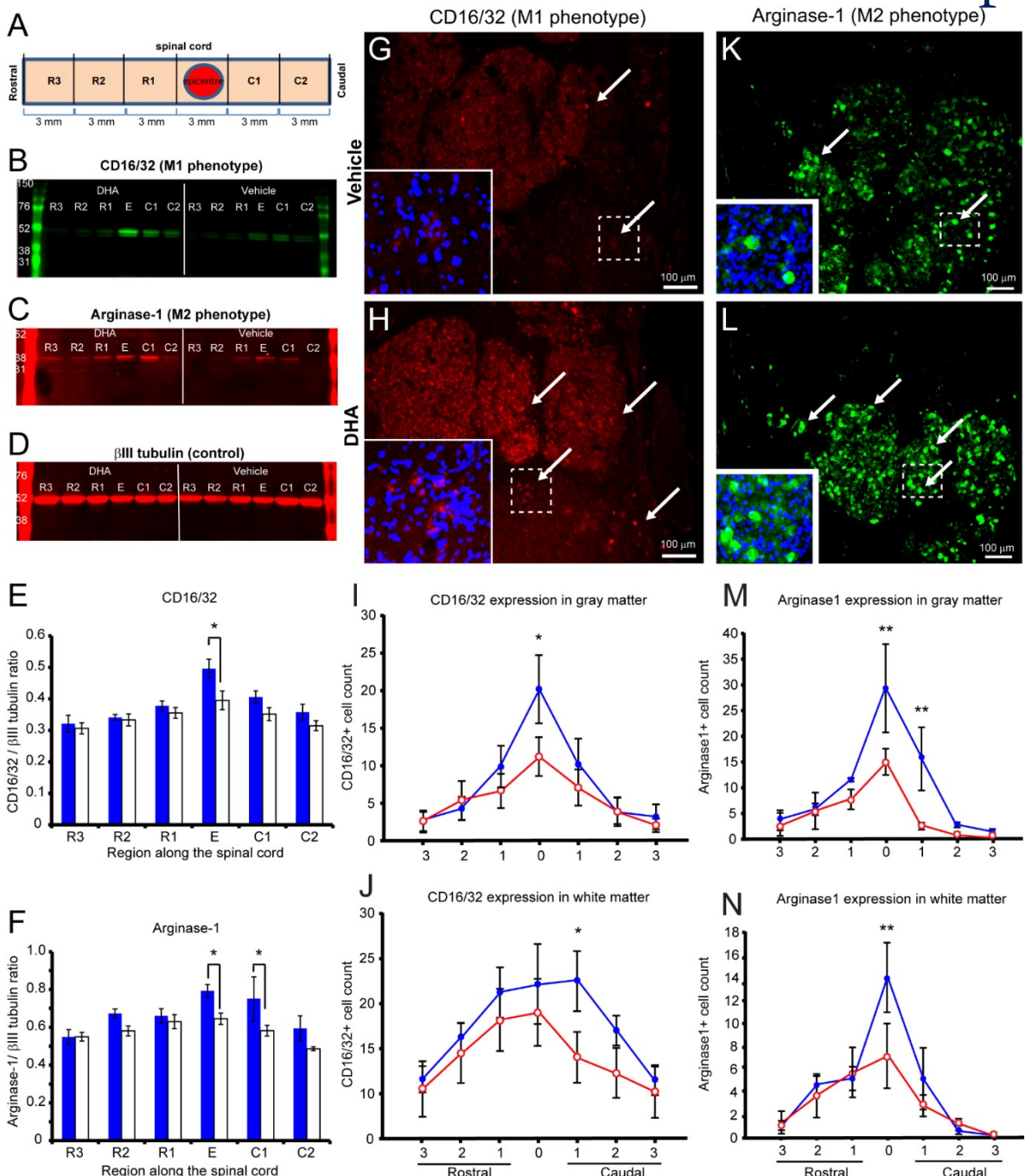


DHA 250 nmol/kg i.v. 30 min post-SCI

with or without an DHA-enriched diet (approx. 300-400 mg/kg/day)

(Huang et al, Brain, 2007)

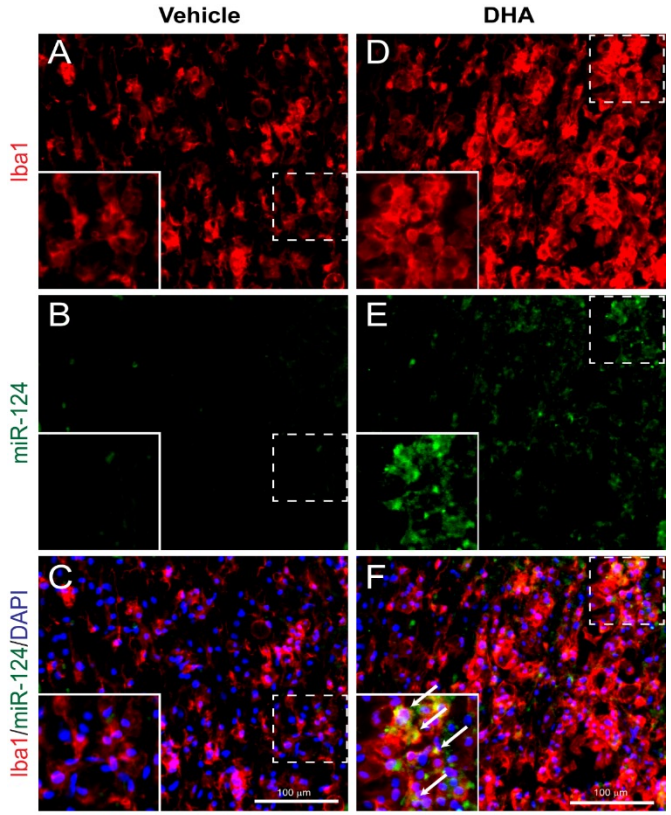
DHA effect on M1 versus M2 macrophages

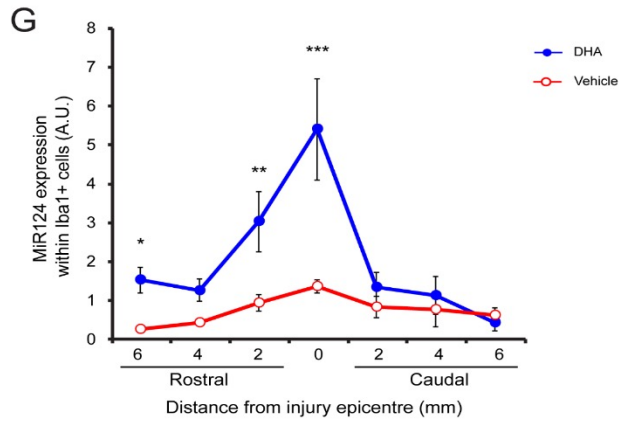


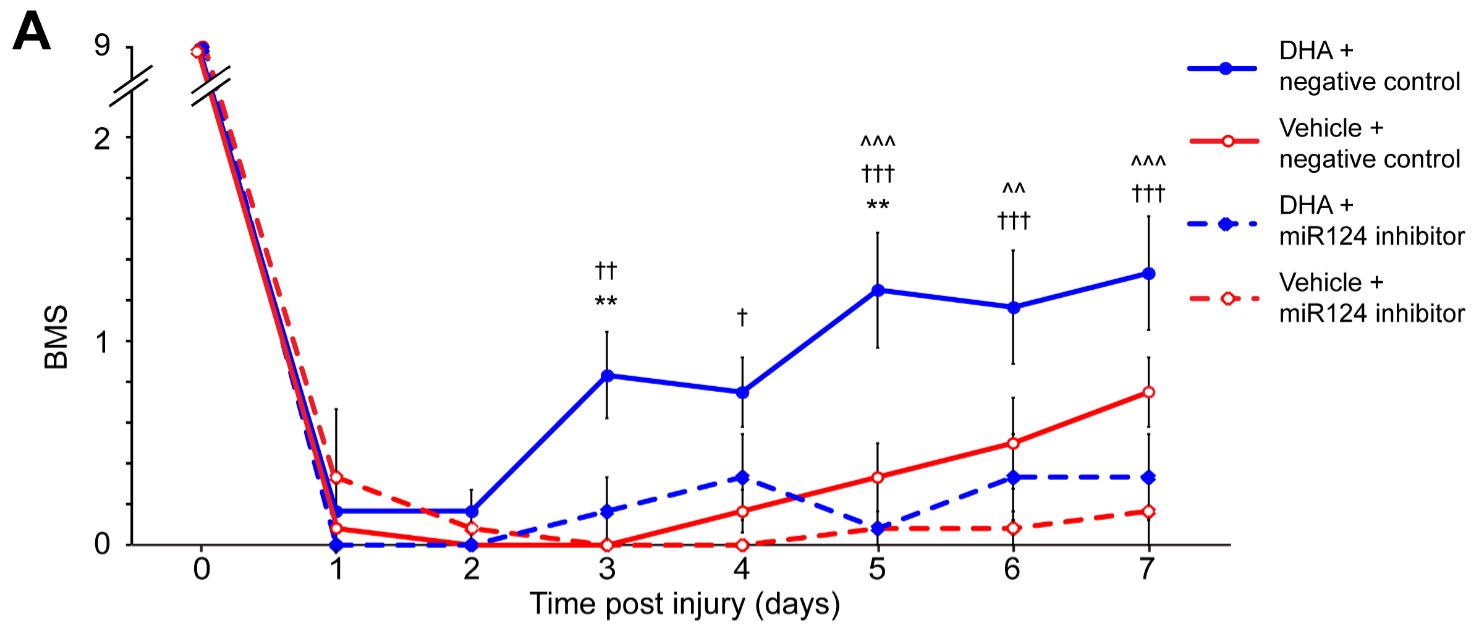
(Yip et al., Hum Mol Gen, 2019)

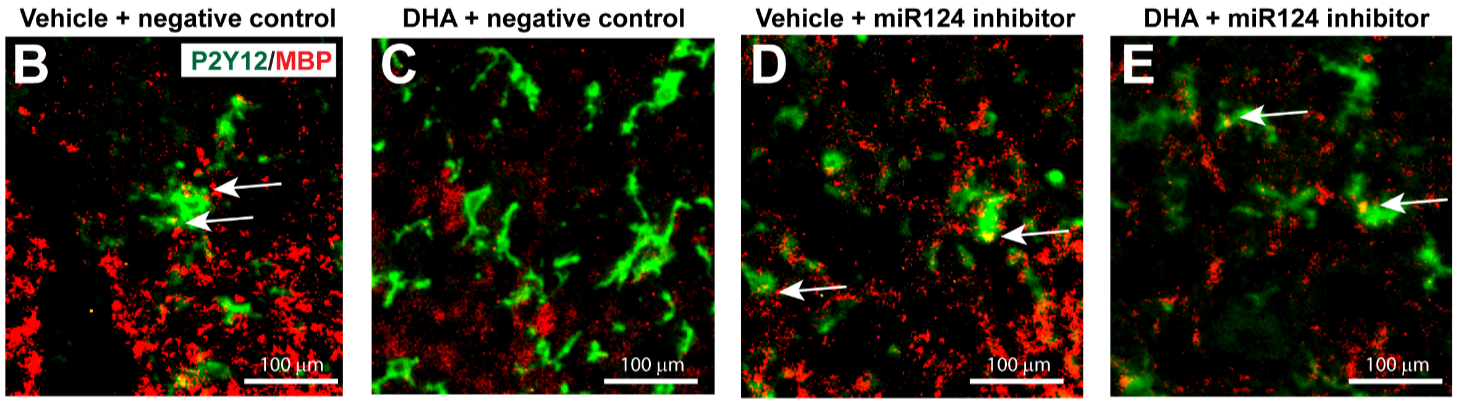
DHA and myelin phagocytosis in microglia

Role of miR-124





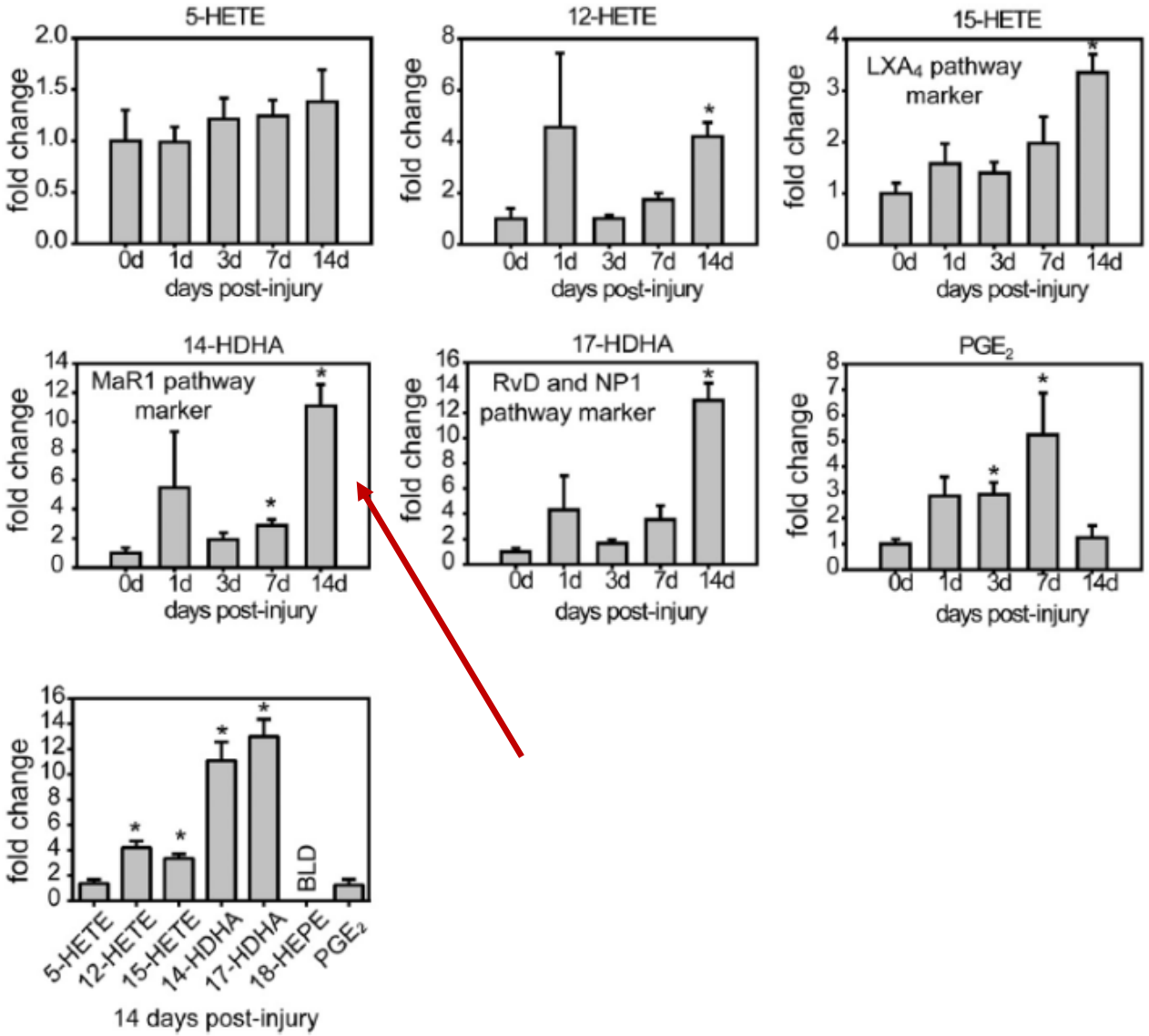




(Yip et al., Hum Mol Gen, 2019)

Maresin 1 and SCI – kinetics formation lipid mediators

Mouse T10 contusion SCI Spinal cord lysate



(Francos-Quijorna et al., 2017)

Omega-3 fatty acids, neuroplasticity and brain connectivity

The restoration of connectivity post-neurotrauma

Behavioral/Cognitive

Dietary Omega-3 Fatty Acids Modulate Large-Scale Systems Organization in the Rhesus Macaque Brain

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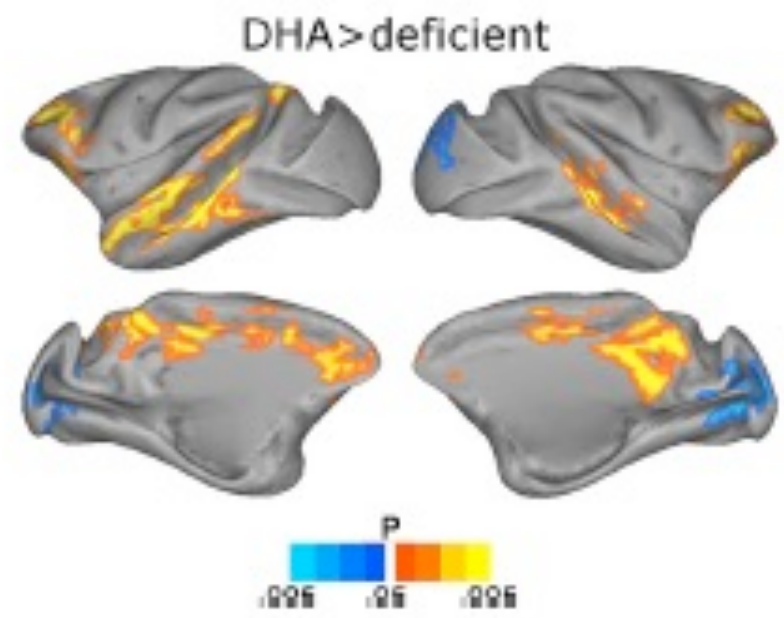
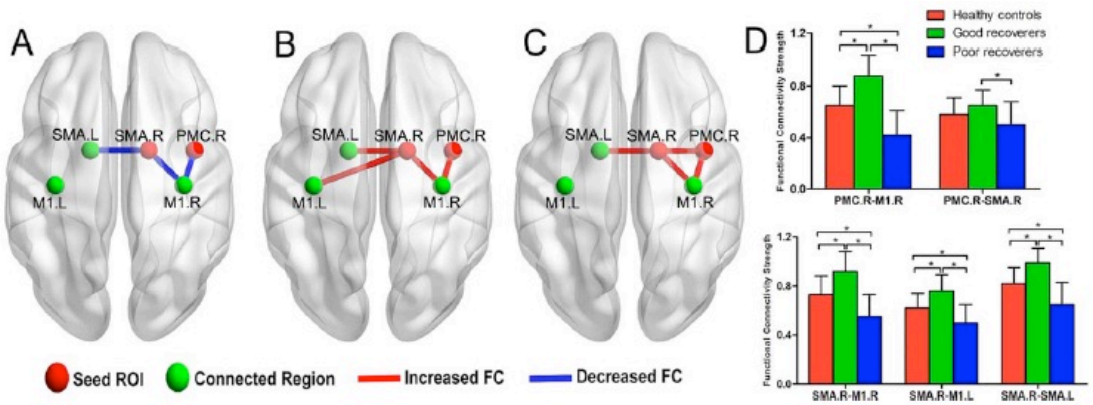


◆ Human Brain Mapping 37:2195–2209 (2016) ◆

Motor Recovery at 6 Months After Admission Is Related to Structural and Functional Reorganization of the Spine and Brain in Patients With Spinal Cord Injury

Jingming Hou,[1] Zimin Xiang,[2,3] Rubing Yan,[1] Ming Zhao,[4] Yongtao Wu,[1] Jianfeng Zhong,[2] Lei Guo,[2] Haitao Li,[4] Jian Wang,[4] Jixiang Wu,[1] Tiansheng Sun,[2]* and Hongliang Liu[1]*



Imaging of DHA effect in rat contusion injury

Reduction in vivo of a marker of microglia activation – Translocator Protein (TSPO)

Eur J Nucl Med Mol Imaging
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ORIGINAL ARTICLE

In vivo PET imaging of the neuroinflammatory response in rat spinal cord injury using the TSPO tracer [^{18}F]GE-180 and effect of docosahexaenoic acid

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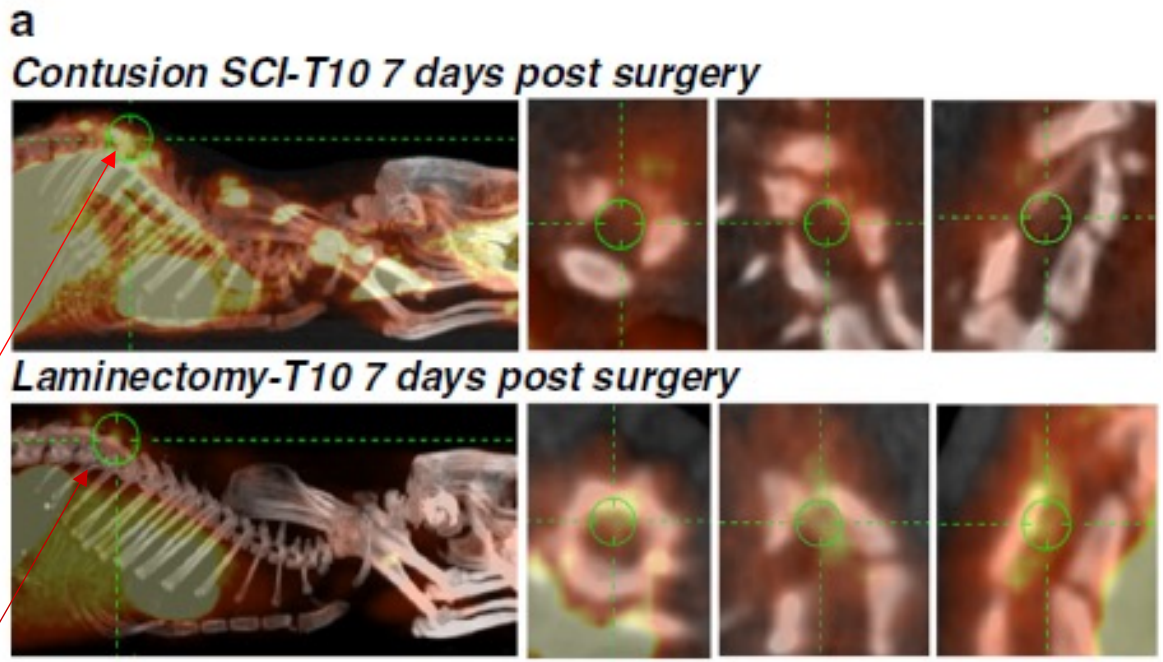
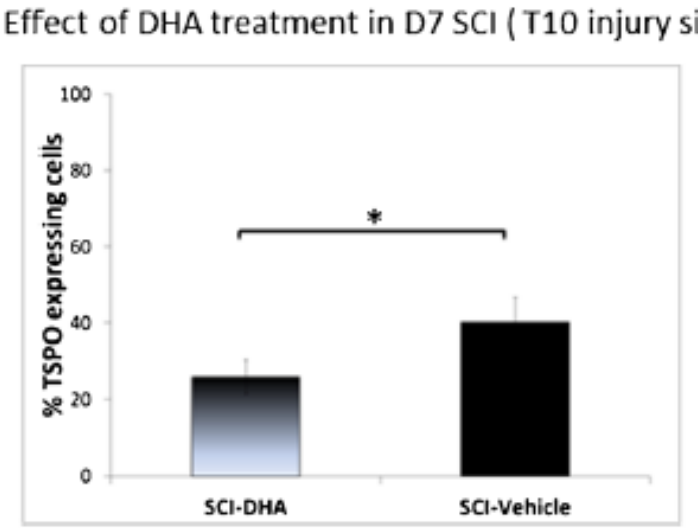
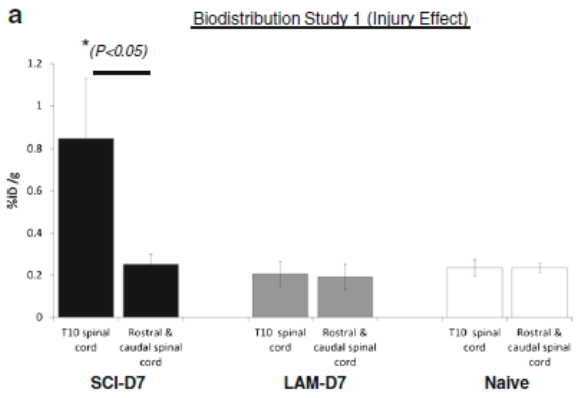
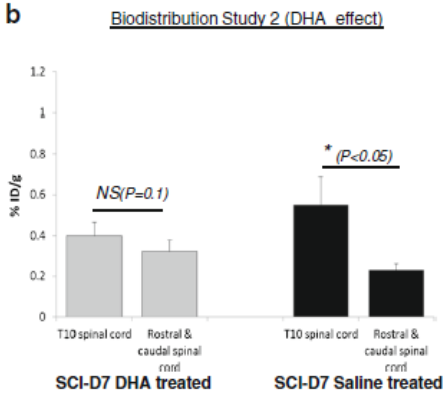






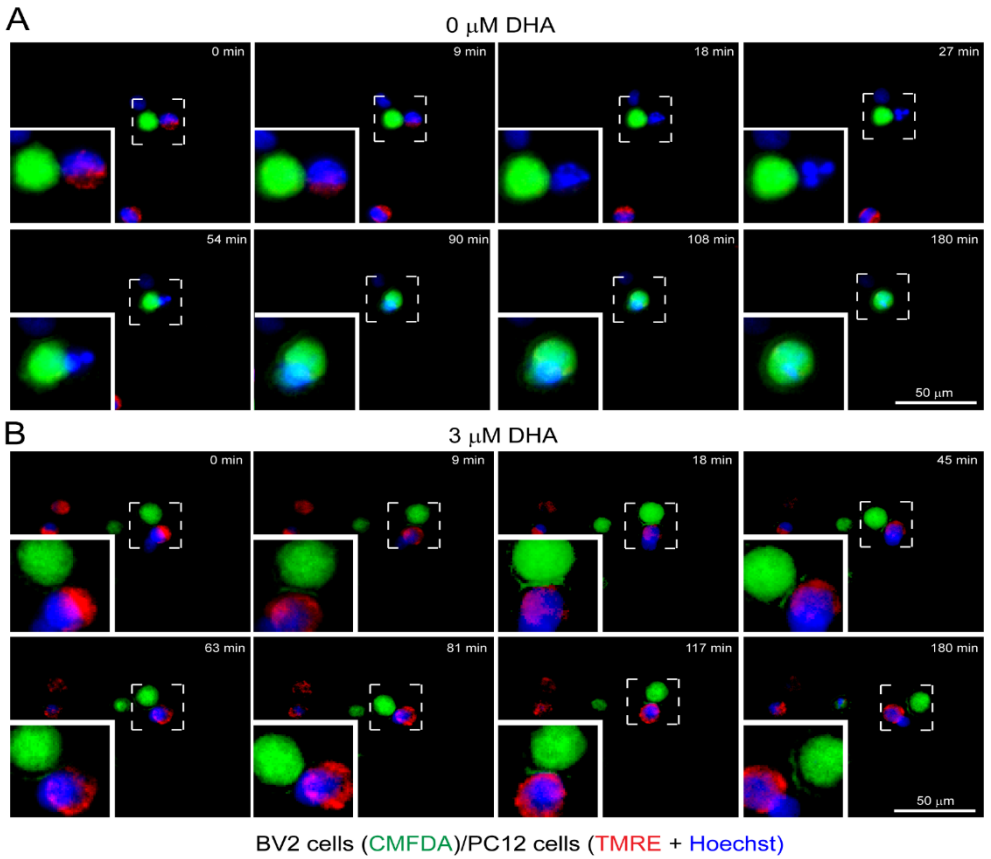


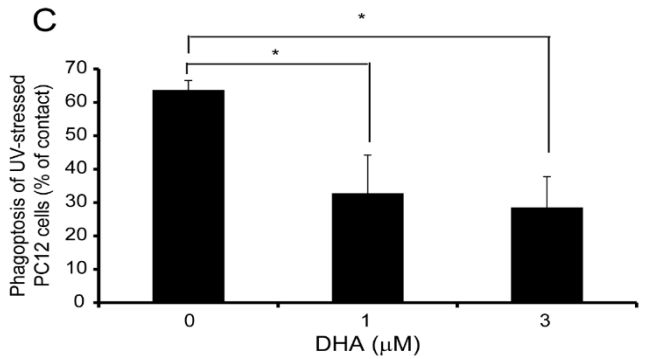
The marker for TSPO concentrates at the injury site, and DHA reduces the signal

Translational challenges in nervous system injury

- Repeated clinical translation disappointments
- Concern over limited predictive value of experimental data obtained in lower species
- Need for patient stratification
- Understand how to decrease the long-term consequences
- ***Cognitive Function*** *(attention, memory)*
- ***Motor function*** *(extremity weakness, impaired coordination and balance, paralysis)*
- ***Sensation*** *(hearing, vision, impaired perception and touch)*
- ***Behaviour*** *(emotional regulation, depression, anxiety, aggression, impairment in behavioral control, personality changes).*

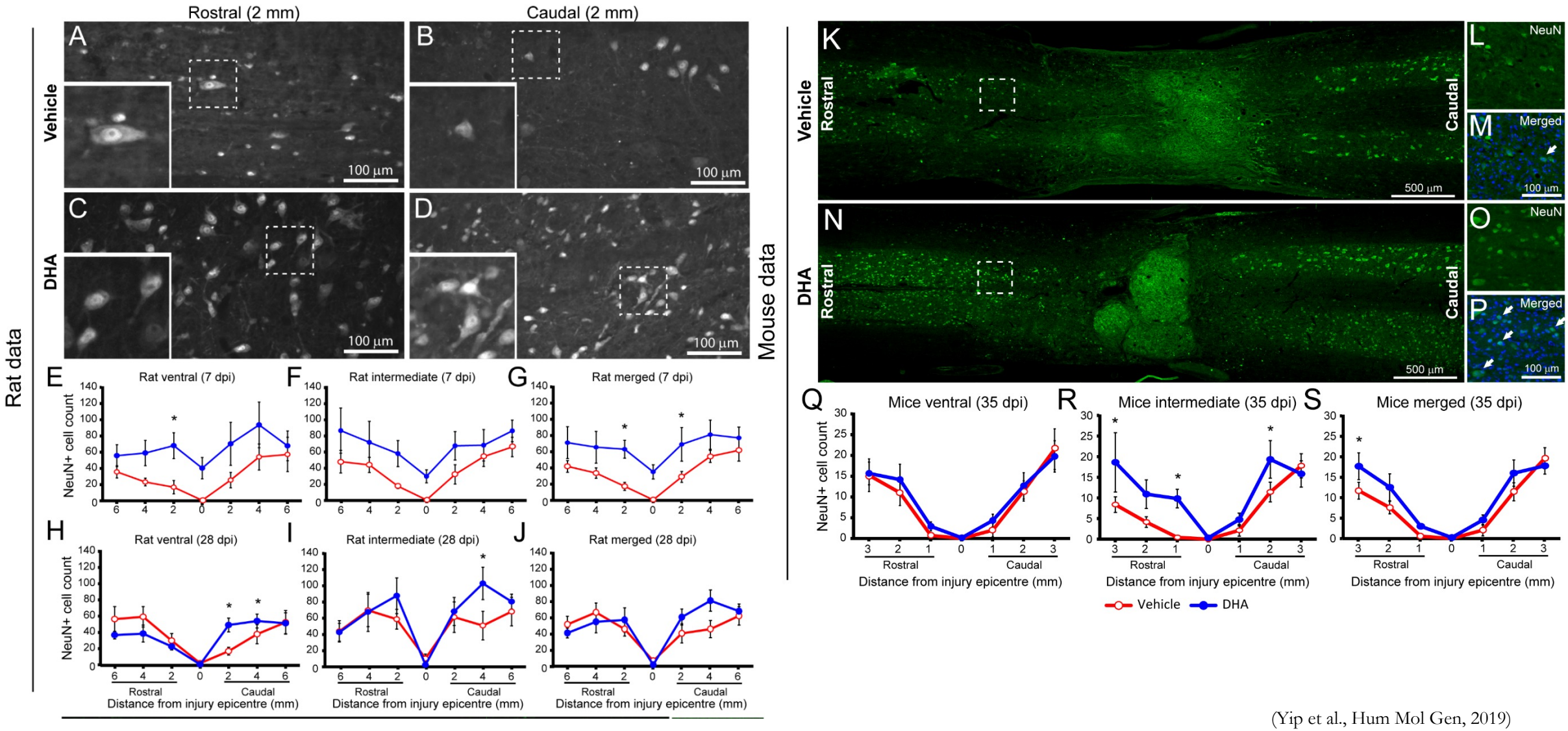
DHA reduces phagoptosis of stressed neurones





(Yip et al., Hum Mol Gen, 2019)

DHA reduces neuronal loss in contusion SCI



(Yip et al., Hum Mol Gen, 2019)