Wallerian degeneration
the innate immune response of the PNS to traumatic injury

Macrophage and Schwann cell activation
&
the cytokine network of Wallerian degeneration

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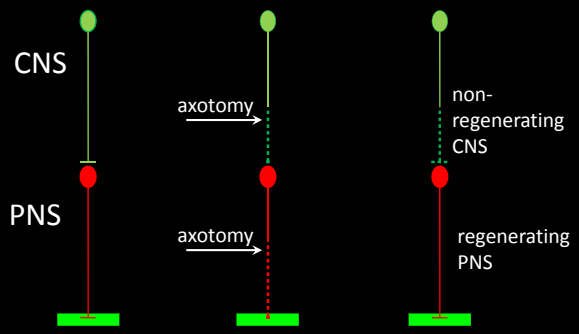
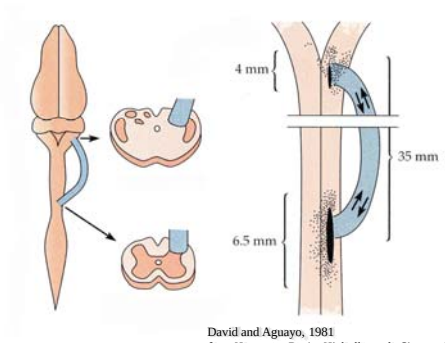
PNS is regenerating - CNS is not

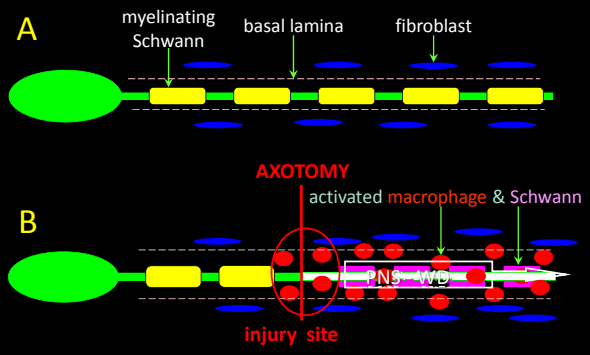


CNS neurons regenerate axons through Wallerian degenerated PNS but not CNS tissue

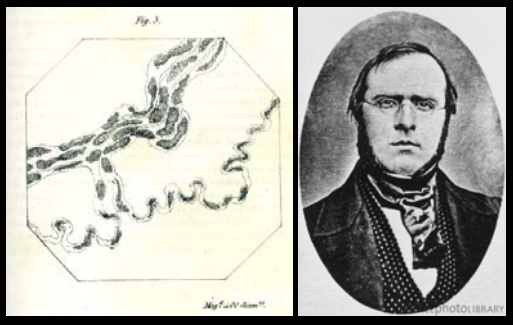


David and Aguayo, 1981
from Neuron to Brain, Nicholls et al., Sinauer 2001

PNS: (A) intact and (B) Wallerian degeneration

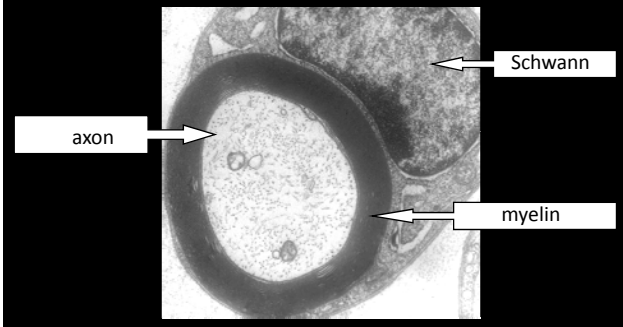


Wallerian degeneration
Augustus Waller (1850)



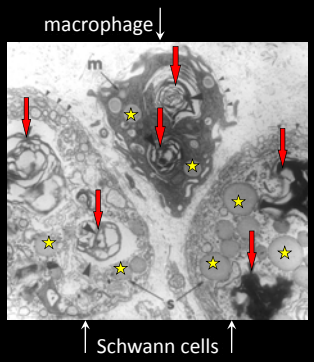
Waller, *Phil. Transact. Royal. Soc. London* 1850

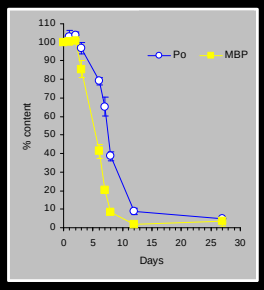
Intact PNS: myelinated axon



Reichert, Saada & Rotshenker, *J. Neurosci.* 1994

Normal PNS Wallerian degeneration





Reichert, Saada & Rotshenker, *J. Neurosci.* 1994

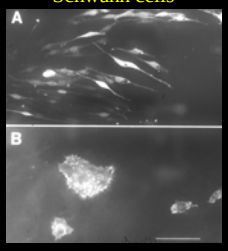
Be'eri, Reichert, Saada & Rotshenker, *Eur. J. Neurosci.* 1998

Normal Wallerian degeneration

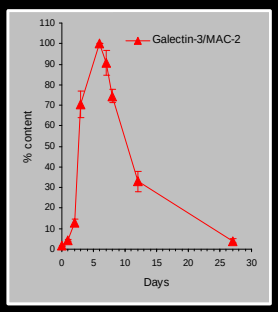
- Axons degenerate
- Schwann cells reject their myelin
- Schwann cells & fibroblasts proliferate
- Macrophages are recruited from circulation
- Macrophages & Schwann cells are activated to clear and degrade degenerated-myelin

Galectin-3/MAC-2 marks macrophage and Schwann cell activation in normal Wallerian degeneration

Schwann cells



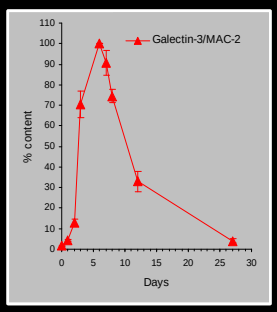
macrophages

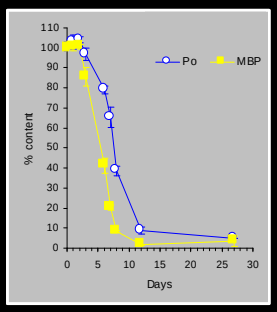


Reichert, Saada & Rotshenker, *J. Neurosci.* 1994

Be'eri, Reichert, Saada & Rotshenker, *Eur. J. Neurosci.* 1998

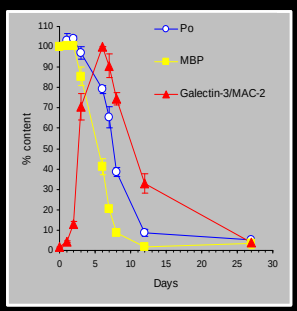
Schwann cell & macrophage activation myelin phagocytosis & degradation





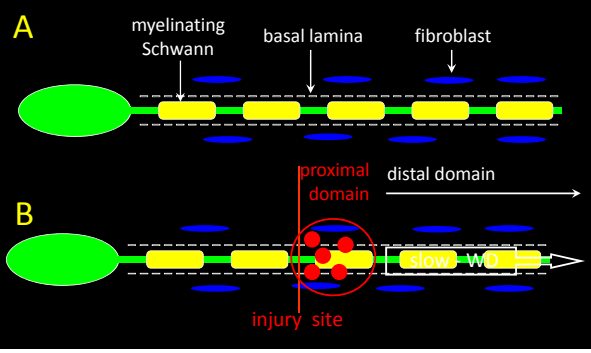
Be'eri, Reichert, Saada & Rotshenker, *Eur. J. Neurosci.* 1998

Schwann cell & macrophage activation myelin phagocytosis & degradation are orchestrated in time

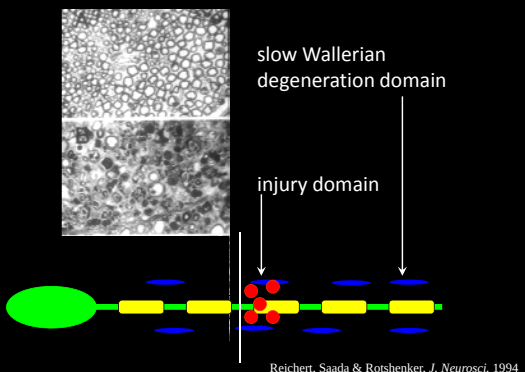


Be'eri, Reichert, Saada & Rotshenker, *Eur. J. Neurosci.* 1998

slow Wallerian degeneration (slow-WD) in mutant Wld^{s} (Ola) mice

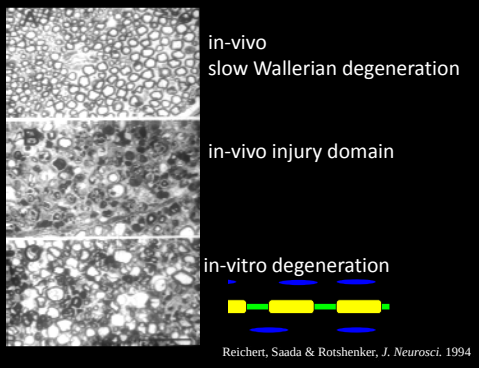


slow Wallerian degeneration in Wld^s mice



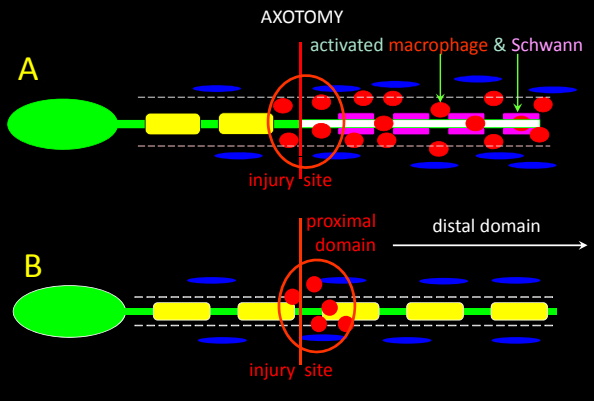
Reichert, Saada & Rotshenker, *J. Neurosci.* 1994

in-vitro degeneration in Wld^s mice



Reichert, Saada & Rotshenker, *J. Neurosci.* 1994

PNS: (A) normal-WD and (B) slow-WD



Events associated with Wallerian degeneration

normal - WD	slow - WD
• Normal regeneration	• Delayed regeneration
• NGF is upregulated	• NGF is not upregulated
• Neuropathic pain – rapid development	• Neuropathic pain – delayed development

Is there a single mechanism that underlies normal- and slow-Wallerian degeneration and the events associated with them ?

Wallerian degeneration is the innate immune response of the PNS to traumatic axonal injury

- Normal-WD results from and is a manifestation of a normal innate immune response
- slow-WD results from and is a manifestation of a deficient innate immune response

Cytokines are the mediators of inflammation

Inflammatory – initiation and progression

- TNFα – tumor necrosis factor α
- IL-1α – interleukin-1α
- IL-1β – interleukin-1β
- IL-6 – interleukin 6
- GM-CSF – granulocyte macrophage colony stimulating factor

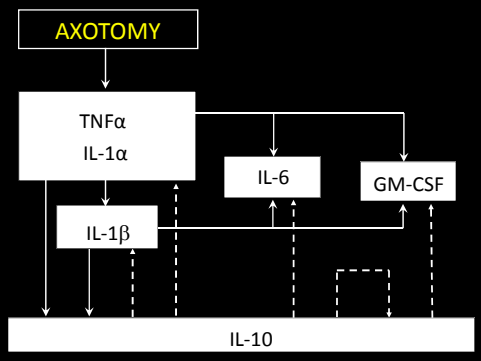
Anti-inflammatory – down regulation

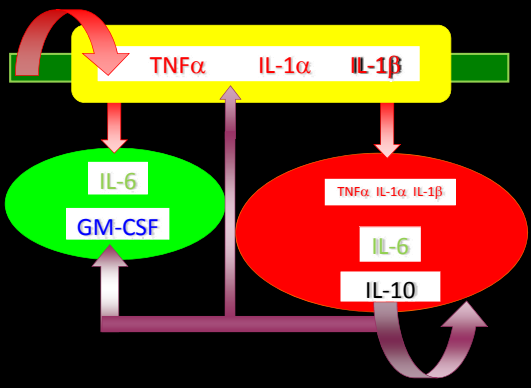
- IL-10 – interleukin-10

Cytokines in Wallerian degeneration

1. Mediators of inflammation
2. Activate Schwann cells and macrophages to clear degenerated-myelin
3. Regulate NGF production
4. Nerve growth factors
5. Regulate neuropathic pain

Cytokine Network of Wallerian degeneration



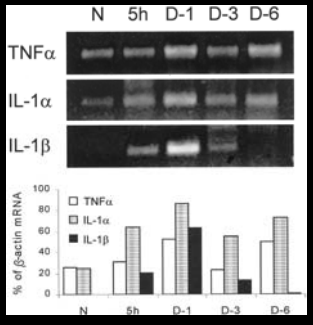


Studying cytokine production

Immune and non-immune cells synthesize and secrete cytokines that bind and activate cognate receptors on immune cells

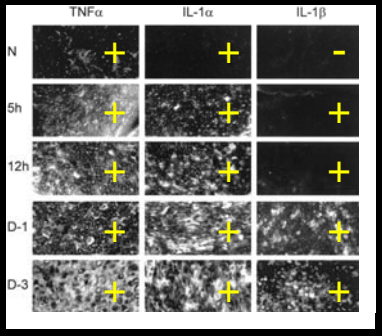
- transcription – cytokine mRNA expression
- translation – cytokine protein in cells
- secretion – cytokine protein in extracellular space
- which cell type

TNFα, IL-1α & IL-1β mRNA in normal Wallerian degeneration



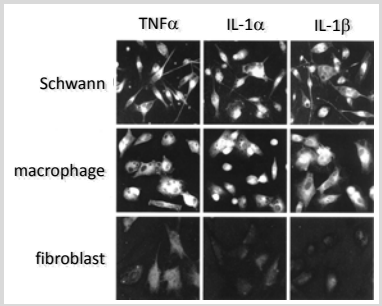
Shamash, Reichert & Rotshenker, *J. Neurosci.* 2002

IL-1α, IL-1β & TNFα proteins in normal Wallerian degeneration



Shamash, Reichert & Rotshenker, *J. Neurosci.* 2002

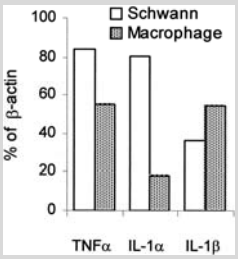
TNFα, IL-1α & IL-1β protein in nerve derived non-neuronal cells



F4/80	CR3	Gal-3	S-100
-	-	+	+
+	+	+	-
-	-	-	-

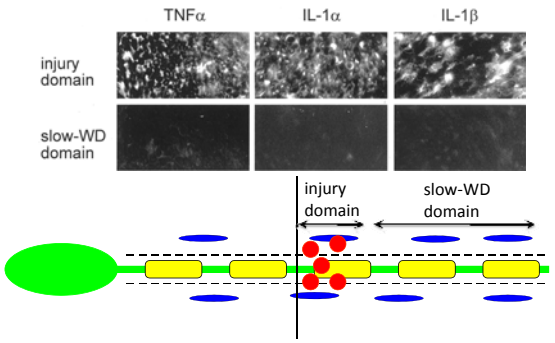
Shamash, Reichert & Rotshenker, *J. Neurosci.* 2002

IL-1α, IL-1β & TNFα mRNA in Schwann cells & macrophages



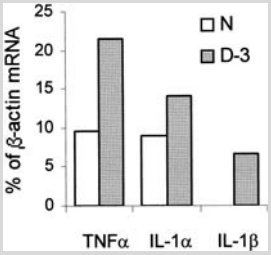
Shamash, Reichert & Rotshenker, *J. Neurosci.* 2002

IL-1α, IL-1β & TNFα protein in slow Wallerian degeneration



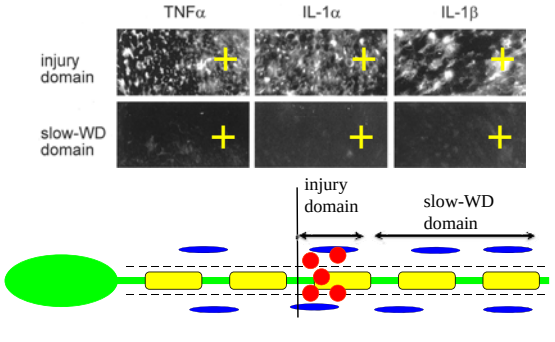
Shamash, Reichert & Rotshenker, *J. Neurosci.* 2002

IL-1α, IL-1β & TNFα mRNA in slow Wallerian degeneration



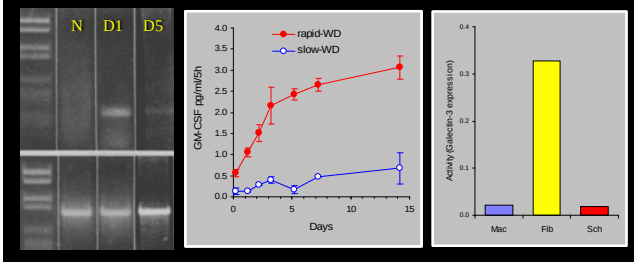
Shamash, Reichert & Rotshenker, *J. Neurosci.* 2002

IL-1α, IL-1β & TNFα in slow Wallerian degeneration



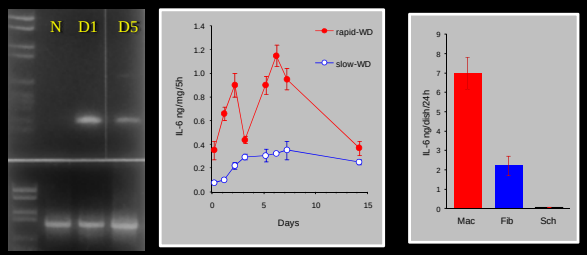
Shamash, Reichert & Rotshenker, *J. Neurosci.* 2002

GM-CSF production in normal and slow Wallerian degeneration



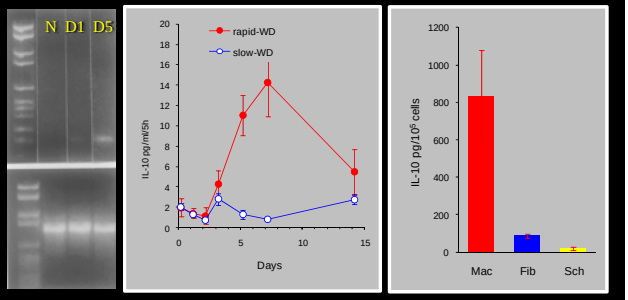
Saada, Reichert & Rotshenker, J. Cell Biology 1996
Be'eri, Reichert, Saada & Rotshenker, *Eur. J. Neurosci.* 1998

IL-6 production in normal and slow Wallerian degeneration



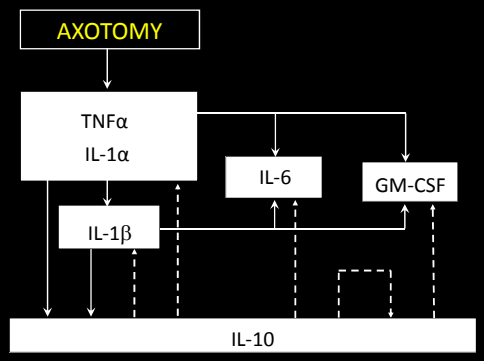
Reichert, Levitzky & Rotshenker, *Eur. J. Neurosci.* 1996

IL-10 production in normal and slow Wallerian degeneration

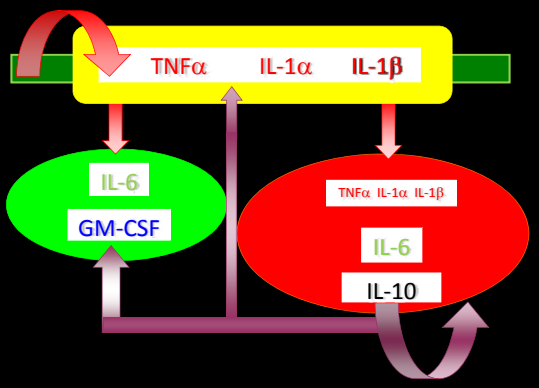


Be'eri, Reichert, Saada & Rotshenker, *Eur. J. Neurosci.* 1998

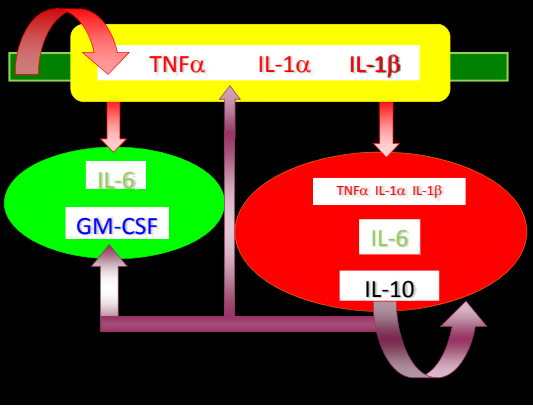
Cytokine Network of Wallerian degeneration



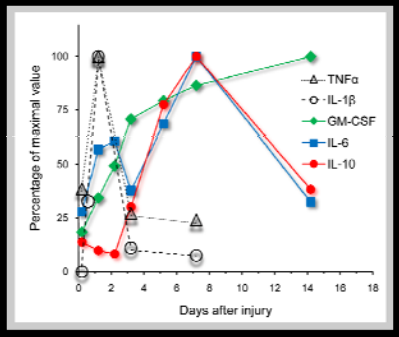
normal Wallerian degeneration



slow Wallerian degeneration

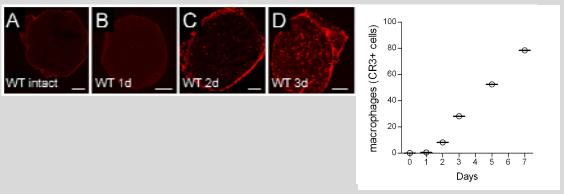


Cytokine production in normal Wallerian degeneration



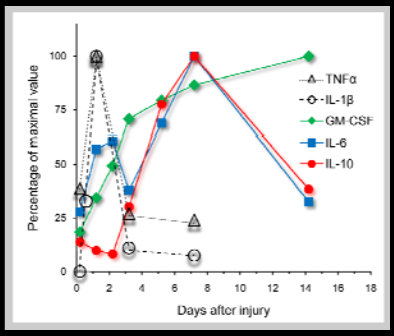
Rotshenker, *J. Neuroinflammation.* 2011

Macrophage recruitment in normal Wallerian degeneration



Gitik, Reichert & Rotshenker, *unpublished*

Cytokine production in normal Wallerian degeneration

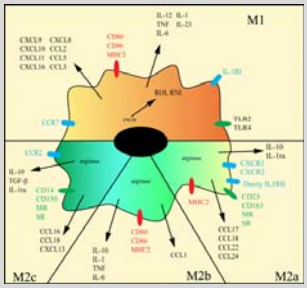


Rotshenker, *J. Neuroinflammation*. 2011

M1-type and M2-type macrophages

Ly6C/Gr1⁺

TNFα & IL-1
ROS
IL-6
phagocytosis
killing



Ly6C/Gr1⁻

IL-10
IL-6
phagocytosis
wound-healing
tissue remodeling

Benoit, Desnues & Mege, *J. Immunol*. 2008
Auffray, Sieweke, & Geissmann, *Ann. Rev. Immunol*. 2009

Are macrophages friend or foe?

M2 type – Wound healing

Anti-Inflammatory cytokines
- IL-10

Phagocytosis
- apoptotic cells
- pathogens
- neurotoxic products of neurodegeneration
- tissue debris

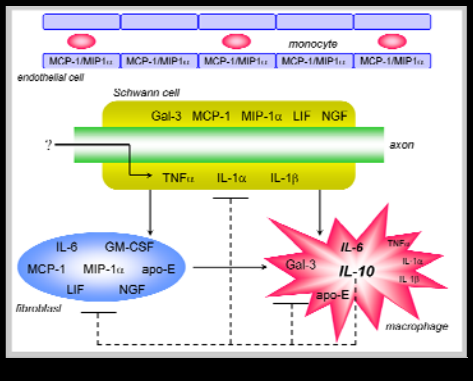
M1 type – Killer

Inflammatory cytokines
- TNFα, IL-1β

Neurotoxicity
- ROS
- PGE

Phagocytosis

Cytokine network of Wallerian degeneration



Events associated with Wallerian degeneration

normal - WD	slow - WD
• Normal regeneration	• Delayed regeneration
• NGF is upregulated	• NGF is not upregulated
• Neuropathic pain – rapid development	• Neuropathic pain – delayed development

Normal regeneration

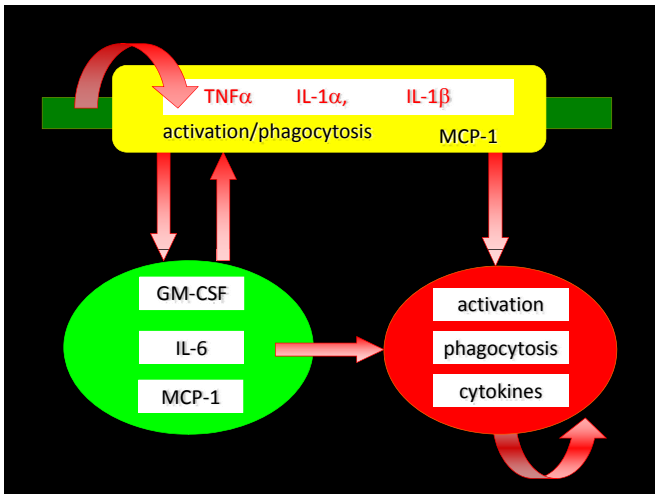
Normal removal of myelin
- macrophage recruitment and activation
- Schwann cell activation
- myelin phagocytosis

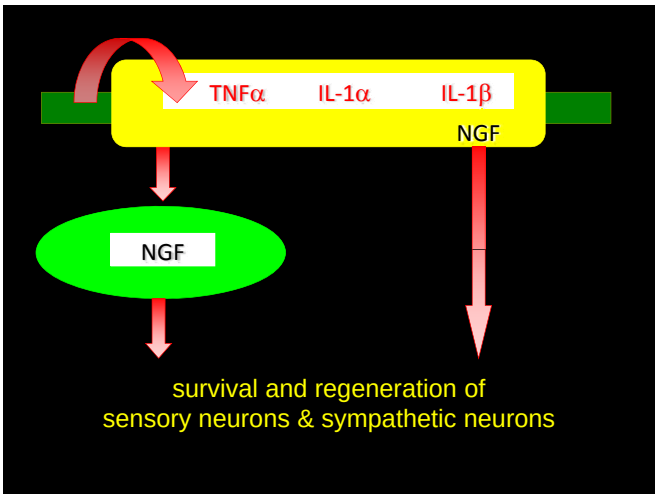
Neurotrophic factors
- NGF

Fixed attraction molecules
- Schwann cells, basal lamina, collagen

Cytokines regulate myelin removal

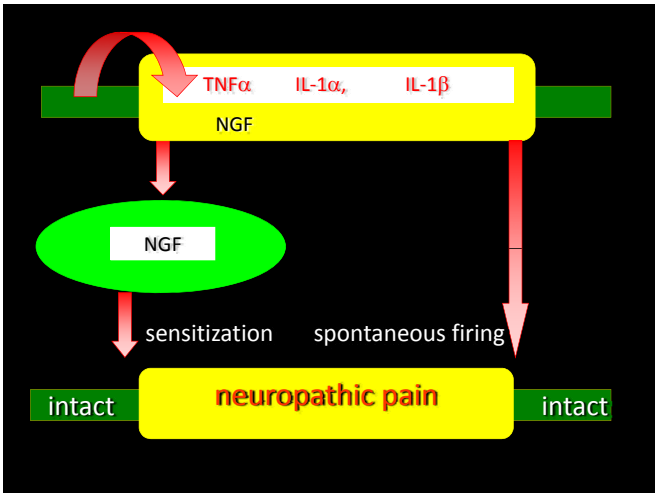
- TNFα, IL-1α and IL-1β function as chemoattractant to recruit macrophages
- TNFα, IL-1α and IL-1β up-regulate production of macrophage chemoattractants MCP-1 and MP-1
- GM-CSF activates macrophages and Schwann cells to clear degenerated-myelin by up-regulating Galectin-3/MAC-2
- GM-CSF up-regulates CR3/MAC-1
- cytokines up-regulate myelin phagocytosis





Cytokines regulate neuropathic pain

- TNFα, IL-1α and IL-1β produce spontaneous firing in sensory neurons
- TNFα, IL-1α and IL-1β up-regulate NGF production in fibroblasts and in vascular endothelial cells. In turn, NGF produces neuropathic pain by sensitizing sensory nerve endings/receptors
- IL-6 deficient mice display reduced neuropathic pain



Wallerian degeneration is the innate immune response of the PNS to traumatic axonal injury

normal - WD	slow - WD
• Normal myelin removal	• Delayed myelin removal
• Normal regeneration	• delayed regeneration
• NGF is upregulated	• NGF is not upregulated
• Neuropathic pain – normal development	• Neuropathic pain – delayed development

Normal Wallerian degeneration