Fibrinogen in Neurological Diseases: Mechanisms, Imaging, Therapeutics

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Disclosures:
Named Inventor: US 8980836, 8877195, 7807644, 8569242
Brain Vasculature

3D-Immuno-labeling in Cleared Brain

Pamela Rios
Victoria Rafalski
Blood-Brain Barrier Disruption

Multiple Sclerosis Animal Model

Blood Vessels

Blood Leakage

Merge

Pamela Rios, Maya Ellisman
Blood coagulation factors
Fibrinogen
"If one looks carefully at freshly altered parts of the white matter ... one perceives already with the naked eye a red point or line in the middle of each individual focus,.. the lumen of a small vessel engorged with blood ... All this leads us to search for the primary cause of the disease in an alteration of individual vessels and their ramifications; All vessels running inside the foci, but also those which traverse the immediately surrounding but still intact parenchyma are in a state characteristic of chronic inflammation."
Fibrinogen is abundantly deposited in neurological diseases

Human Brain
Multiple sclerosis (MS), Traumatic brain injury (TBI)
Alzheimers disease (AD)
Fibrinogen is abundantly deposited in neurological diseases

Control - Human

Human Active MS lesion

Fibrinogen

Mark Petersen
Fibrinogen is abundantly deposited in neurological diseases
Fibrinogen

Multiple Sclerosis
EAE
Animal Model
Fibrinogen

Multiple Sclerosis
EAE
Animal Model

Pamela Rios
Maya Ellisman
Fibrinogen is converted to inflammatory fibrin.

Fibrinogen to fibrin conversion:
- Fibrinogen
- Thrombin
- Coagulation
- Fibrin
- Inflammation
Fibrin is a “Druggable” Target

Fibrin γ chain

CD11b/CD18
(Mac-1, Complement receptor 3, αMβ2)

Microglia, Macrophages

Inflammation

αIIbβ3

Platelets

Coagulation

γ377-395

γ400-411
Fibrin is required for nervous system disease

PNS
• Nerve regeneration
  
  Fibrin depletion enhances nerve remyelination
  Akassoglou et al., 2002, Neuron, 33:861-875

CNS
• Multiple Sclerosis
  
  Fibrin depletion decreases inflammation and delays the onset of demyelination
  Akassoglou et al., 2004, Proc Natl Acad Sci USA, 101:6698-6703
  
  Fibrin depletion suppresses relapsing paralysis
  
  Fibrin depletion protects from axonal damage

• Brain Trauma
  
  Fibrin depletion reduces astrocyte scar formation
  Schachtrup et al., 2010, J Neurosci, 30:5843-54

• Alzheimer Disease
  
  Fibrin depletion reduces neuroinflammation and cognitive impairment
  Cortes-Canteli et al., 2010, Neuron. 66:695-709.
Blood vessel

Nervous system

Fibrinogen

Multiple sclerosis, Traumatic brain injury, Alzheimer's disease
Blood vessel

Fibrinogen

Microglia

CD11b/CD18 (CR3, Mac-1)


Multiple sclerosis, Traumatic brain injury, Alzheimer's disease
Dynamic Interaction between Microglia-Vasculature


Dynamic Interaction between Microglia-Vasculature


Davalos et al., 2012, Nat Commun, 3:1227
Vascular damage is a robust inducer of microglia responses

- Microglia
- Blood vessel
- Laser ablation

Victoria Rafalski
Fibrinogen is sufficient to induce rapid microglial responses in vivo

Fibrinogen is sufficient to induce rapid microglial responses in vivo

Fibrinogen is sufficient to induce rapid microglial responses in vivo

Fibrinogen is specific among plasma proteins to induce microglial activation

Fibrinogen-Induced Encephalomyelitis (FIE)

Ryu et al., 2015, Nat Commun: 6:8164

- Novel experimental setting to study vascular-driven neuroinflammation
- Fibrin-induced, microglia-driven

Demyelination

CCR2<sup>RFP</sup>

T-cells
Microglia cluster around blood vessels at sites of active BBB disruption

Microglia cluster around blood vessels at sites of active BBB disruption

Fibrin / CD11b signaling is required for microglial perivascular clustering and axonal damage

Coagulation activity in the CNS as a sensitive imaging biomarker

- **Thrombin ACPP** (Activatable Cell Penetrating Peptide): thrombin specific sequence links two oppositely charged peptides

- *Thrombin* activity releases polycation with cy-5 cargo that is uptaken by cells

- Clinical applications for ACPPs due to dual labeling with fluorescence and Gd.

- MMP ACPPs used in cancer detection and removal

In collaboration with Michael Whitney, Roger Tsien, UCSD

Coagulation activity in the CNS as a sensitive imaging biomarker

Whole Spinal Cord Imaging

EAE

Healthy

Coagulation activity correlates with demyelinated lesions

Thrombin Probe / Myelin

Coagulation activity detected early in EAE

Thrombin Probe

Fibrin

Microglia

Fibrin in early MS lesions

Human MS

Fibrin/Iba-1

Fibrin deposition during the course of MS

Human MS

- Proteomic analysis of chronic active plaques reveals activation of the coagulation cascade
- BBB opens 4 weeks prior to demyelination in marmoset EAE
- Impaired fibrinolysis in MS lesions

Excessive/persistent fibrin deposition in MS lesions

Coagulation Cascade Biomarkers for MS?

**EPIC MS COHORT**
N=582

**Genetics**
- Genome Side Association Studies
- High resolution HLA-A, B, C, DR, DQ genotypes
- KIR genotypes
- MS-validated risk allele genotypes and weighted log-additive genetic burden

**Therapies**

**MRI**

**Disability progression**

**in collaboration with the UCSF Multiple Sclerosis Center**
Multiple sclerosis, Traumatic brain injury, Alzheimer's disease

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