Leukocyte adhesion: mechanisms and applications

Ellie Hughes Microcirculation group

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- Principles of leukocyte-endothelium interactions
 - the leukocyte recruitment cascade
 - adhesion molecules: classes, structures, involvement in recruitment
- Diseases caused by defects in adhesion molecules
- Adhesion molecules as drug targets
- Methods of imaging leukocyte adhesion (as a marker of inflammation)



Early work Cardinal signs observed by Celsus (200 AD)





"It is possible that chemical agents seep into interendothelial junctions and thereby attract pavemented leukocytes, but there is, as yet, no evidence for this concept."





Arnold, 1870's

Early work Substances can move through vascular walls





"Inflammation is the expression and consequence of a molecular alteration in the vessel walls. By it, adhesion between the vessel wall and the blood is increased."

B



Julius Cohnheim, 1982

Early work The birth of leukocyte-endothelium interactions (L/ECIs)



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Leukocyte recruitment



Leukocyte recruitment

Measuring inflammation by intravital microscopy



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Adhesion molecules

Overview













Adhesion molecules Adhesion: integrins and Ig superfamily



Adhesion molecules

Adhesion: integrin signalling pathway (inside-out signalling)



Adhesion: integrins and Ig superfamily



Adhesion: integrins and Ig superfamily



Adhesion molecules Adhesion

Adhesion and crawling: integrins and Ig superfamily



Adhesion molecules

Emigration: interactions within endothelial junctions



Adhesion molecules

Emigration: interactions within endothelial junctions



Leukocyte recruitment

Summary

Selectins	Mucin-like	Integrins	IgSF
L-selectin (CD62L)	GlyCAM-1	α4β1 (VLA-4)	ICAM-1,-2,-3
P-selectin (CD62P)	CD34	α6 β 1 (VLA-6)	VCAM-1
E-selectin (CD62E)	PSGL-1	αLβ2 (LFA-1)	LFA-2, -3
	MAdCAM-1	αMβ2 (Mac-1)	MAdCAM-1
			JAMs
			PECAM-1 (CD31)

Adhesion molecules Summary

Syndrome	Phenotype	Defect
LADI	severe, recurrent infections; delayed wound healing; no pus formation	β ₂ integrin causing diminished expression of CD18 on leukocytes (firm adhesion)
LADII	recurrent infections in early life; decreased chemotaxis; motor retardation; short stature; facial stigmata; Bombay blood group phenotype	defective fucose metabolism causing absence of SLe ^x (leukocyte rolling)
LADIII	LADI + severe bleeding tendency	mutation in kindlin 3 causing general activation defects in all β integrins
Glanzmann's thrombasthenia	bleeding tendency of varying degrees	mutated α Ilb β 3 integrin on platelets, which are unable to aggregate
Wiskott-Aldrich syndrome	raised pathogen susceptibility; eczema; small platelets; thrombocytopaenia	mutations in WASp mean monocytes cannot form podosomes and have limited phagocytotic capabilities

Adhesion molecules

Human genetic defects causing disease



Selectin antagonists

Small molecule inhibitors

•Bimosiamose (pan-selectin) - allo-/ xenograft rejection, asthma, COPD, psoriasis

Carbohydrate-based



Peptides/recombinant proteins
YSPSL (rPSGL-1) - renal I/R



Antibodies



- •Efalizumab ($\alpha I\beta 2$) psoriasis
- •Natalizumab (α4 chain) MS, Crohn's disease

Non-peptide inhibitors

- •Tirofiban ($\alpha II\beta$ 3) platelet aggregation •Valateorast ($\alpha IR1$) - orthogonal
- •Valategrast ($\alpha 4\beta 1$) asthma

Peptide mimetics

•Cilengitide ($\alpha \lor \beta$ 3) - glioblastoma (via angiogenisis)

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•Eptifibatide (\alpha II\beta3) - anti-platelets
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CAMs as drug targets Dr

Drug delivery via adhesion molecules





PET



Scintigraphy

Optical



Ultrasound



MRI

Imaging inflammation

Different modalities









Intravital microscopy









Intravital microscopy

Neutrophil migration towards a focal necrotic lesion in the liver

Intravascular routes of neutrophil chemotaxis



McDonald et al. (2010)

Imaging inflammation

Intravital microscopy



ProSense 680

- An inducible fluorescent imaging probe
- Used in arthritis and cancer models
- Shows local density of proinflammatory cell infiltrates

Imaging inflammation



Activated optical probes



Imaging inflammation Radiopharmaceuticals - scintigraphy



Imaging inflammation Radiopharmaceuticals/scintigraphy/PET







Imaging inflammation Radiopharmaceuticals - scintigraphy



Imaging inflammation Radiopharmaceuticals - scintigraphy



Imaging inflammation Radiopharmaceuticals - PET



Radiopharmaceuticals - PET



Radiopharmaceuticals - PET





Radiopharmaceuticals - PET

MRI contrast agents



Imaging inflammation MRI

Reproducibility between animal models and human patients

EAE (mouse)



Photochemically-induced thrombosis (rat)



Imaging inflammation MRI

MS



Ischaemic stroke



- L/EC interactions are mediated by a variety of adhesion molecules
 - rolling selectins
 - adhesion integrins and IgSF
 - emigration integrins and IgSF (with some cadherin involvement)
- Genetic mutations in adhesion molecules are the cause of several diseases
 - LADI, II, III
 - Glanzmann's thrombasthenia
 - Wiskott-Aldrich syndrome
- Emerging novel therapies
 - Targeting adhesion molecules
 - Adhesion molecules as drug delivery agents
- Imaging leukocyte adhesion: advantages and disadvantages of different modalities
 - Optical imaging intravital microscopy, activated optical probes
 - Radiopharmaceuticals scintigraphy, PET
 - MRI contrast agents

Learning objectives

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