DISTINCT IMMUNE INJURIES FROM ENVIRONMENTAL EXPOSURES TO WATER-DAMAGED BUILDINGS

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CAUSATION

WHAT IS "CHRONIC INFLAMMATION?"

- Two types of inflammation
  - **Acute inflammation** is the body's more common **rapid** response to trauma, microbial invasion or noxious compounds
  - **Chronic inflammation** is less common and is defined as **slow**, **long-term** inflammation lasting several months to years

ETIOLOGY OF **CHRONIC** INFLAMMATION

- Persistent infection
- **Persistent exposure to irritant**
- **Autoimmunity**
- **Auto-inflammatory**
- **Inflammatory and biochemical inducers**
- **Hypersensitivity to noxious stimuli**

CIRS
DIAGNOSIS

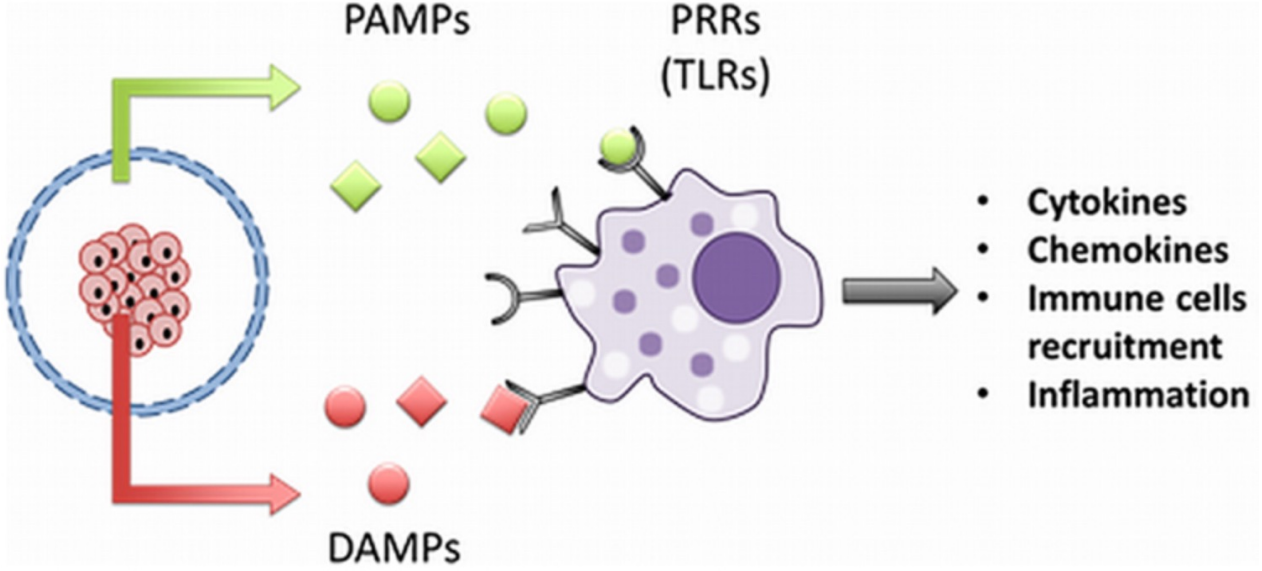
- **Shoemaker**
- **GAO**
- **McMahon Criteria**

OBJECTIVE TESTS

1	VISUAL CONTRAST SENSITIVITY TESTING
2	HUMAN LEUKOCYTE ANTIGEN TESTING
3	MARCoNS
4	COMPLEMENT C4a
5	TRANSFORMING GROWTH FACTOR-BETA 1
6	MATRIX METALLOPROTEINASE-9
7	ANTI-DIURETIC HORMONE/OSMOLALITY
8	ADRENOCORTICOTROPIC HORMONE/CORTISOL
9	MELANOCYTE STIMULATING HORMONE
10	ANTICARDIOLIPIN ANTIBODY or ANTIGLIADIN ANTIBODIES

FUNGAL SURFACE AND INNATE IMMUNE RECOGNITION OF FILAMENTOUS FUNGI

- Detection of infectious agents through **pattern recognition receptors**
- Distinct composition of fungal cell wall makes it target for immune cell
  - Molecular patterns
  - TLR2 and TLR4



FUNGAL RECEPTORS

Table 1 | Pattern recognition receptors involved in detection of fungal molecules.

Receptor	Fungal molecules	Fungal species
TLR4	O-linked mannans	*C. albicans*
	Mannans	*Saccharomyces* spp.
	Rhamnomannans	*P. boydii*
	Phospholipomannans	*C. albicans*
	?	*A. fumigatus*
TLR2/TLR6	Phospholipomannans	*C. albicans*
	Glucuronoxylomannans	*C. neoformans*
TLR2/TLR1	Glucuronoxylomannans	*C. neoformans*
TLR2	α1,4-glucans	*P. boydii*
	?	*A. fumigatus*
MR	N-linked mannans	*C. albicans*
	Mannans	*P. carinii*
	Mannoproteins	*C. neoformans*
DC-SIGN	Galactomannans	*A. fumigatus*
	Mannans	*C. albicans*
Dectin-1	β1,3-glucans	*A. fumigatus*
		C. albicans
		Saccharomyces spp.
Dectin-2	α-mannans	*C. albicans*
Mincle	Polysaccharides containing α-mannosyl residues?	*C. albicans*
		Malassezia spp.
CD14	Mannans	*Saccharomyces* spp.
	?	*A. fumigatus*
	α1,4-glucans	*P. boydii*
NLRP3 inflammasome	β1,3-glucans	*C. albicans*
		S. cerevisiae
		A. fumigatus

The table summarizes PRRs that have been identified in recognition of fungal molecules, but in many cases the molecules responsible for activation are unknown. Activation of the NLRP3 inflammasome by β-glucans and C. albicans is dependent on Dectin-1. In this sense NLRP3 does not recognize fungal molecules directly but instead it senses intracellular signals generated by activation of PRRs, like Dectin-1. The question mark indicates that the fungal molecules recognized by the indicated receptor are unknown.

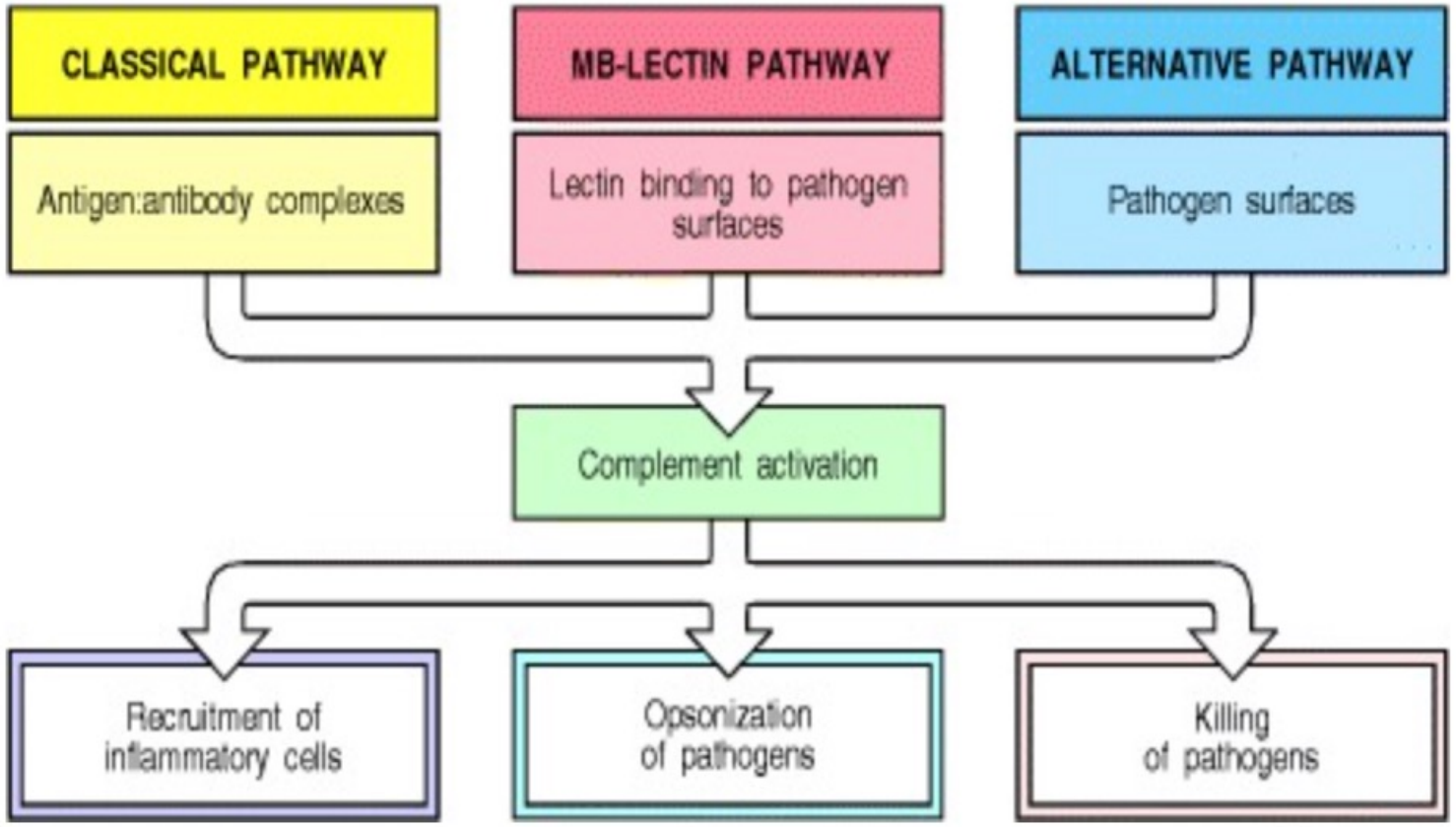
THE ROLE OF FUNGAL PROTEINASES IN PATHOPHYSIOLOGY OF STACHYBOYTRES CHARTARUM

Host	Exposure	Result
Human tracheal epithelial cells	Spore extracts	↑Il-6, IL-8, TNF-a
	Protease inhibitor	Completely abolished cytokine production
Rat pups	Extracts of trichothecenes or proteinases	↑Neutrophils, IL1-B, TNF-a

COMPLEMENT ACTIVATION

- Complement system is made up of **plasma proteins** that react with each other to fight infection
- Three ways to activate complement system:
  - Classical pathway
  - MB-Lectin Pathway
  - Alternate Pathway

COMPLEMENT PATHWAYS



COMPLEMENT ACTIVATION IN A MODEL OF CHRONIC FATIGUE SYNDROME (CFS)

- Chronic fatigue syndrome is a clinical diagnosis that needs biological markers to validate individual cases for diagnostic and medicolegal purposes.
- This study evaluated the effect of an exercise challenge and an allergy challenge on symptoms and biological markers.

COMPLEMENT ACTIVATION IN A MODEL OF CFS (CONT.)

- Chief finding of study was exercise challenge induced significant increases of C4A but not C3A or C5A **only** in the CFS group (6 hours after challenge)
  - In most subjects, only elevated complement split products are consistently found of C3A, C4A, and C5A

LAB	CFS with allergy	CFS without allergy	Allergy without CFS
Eosinophilic cationic protein	↑	↑	↑
C-reactive protein	–	–	–
Autoantibodies	↑	↑	?
C4a	↑	↑	–

COMPLEMENT SPLIT PRODUCTS

TABLE I. Levels of complement (C) components (±SE) from patients seen in NJMRC adult clinic for chronic fatigue and healthy controls

C component	n =	Chronic fatigue	n =	Control subjects
C4a (ng/mL)	83	2202.1 ± 278.4*	31	1121.1 ± 98.6
C3a (ng/mL)	77	887.7 ± 102.7†	31	488.3 ± 32.8
C5a (ng/mL)	66	12.9 ± 0.5*	32	6.3 ± 0.4
SC5b-9 (μg/mL)	74	154.2 ± 13.9*	26	91.8 ± 42.4
Bb (μg/mL)	83	0.91 ± 0.03*	26	0.46 ± 0.03

*Significant relative to control patients, at $P < .001$.
†Significant relative to control patients, at $P < .005$.

EXERCISE CHALLENGE RESULTS

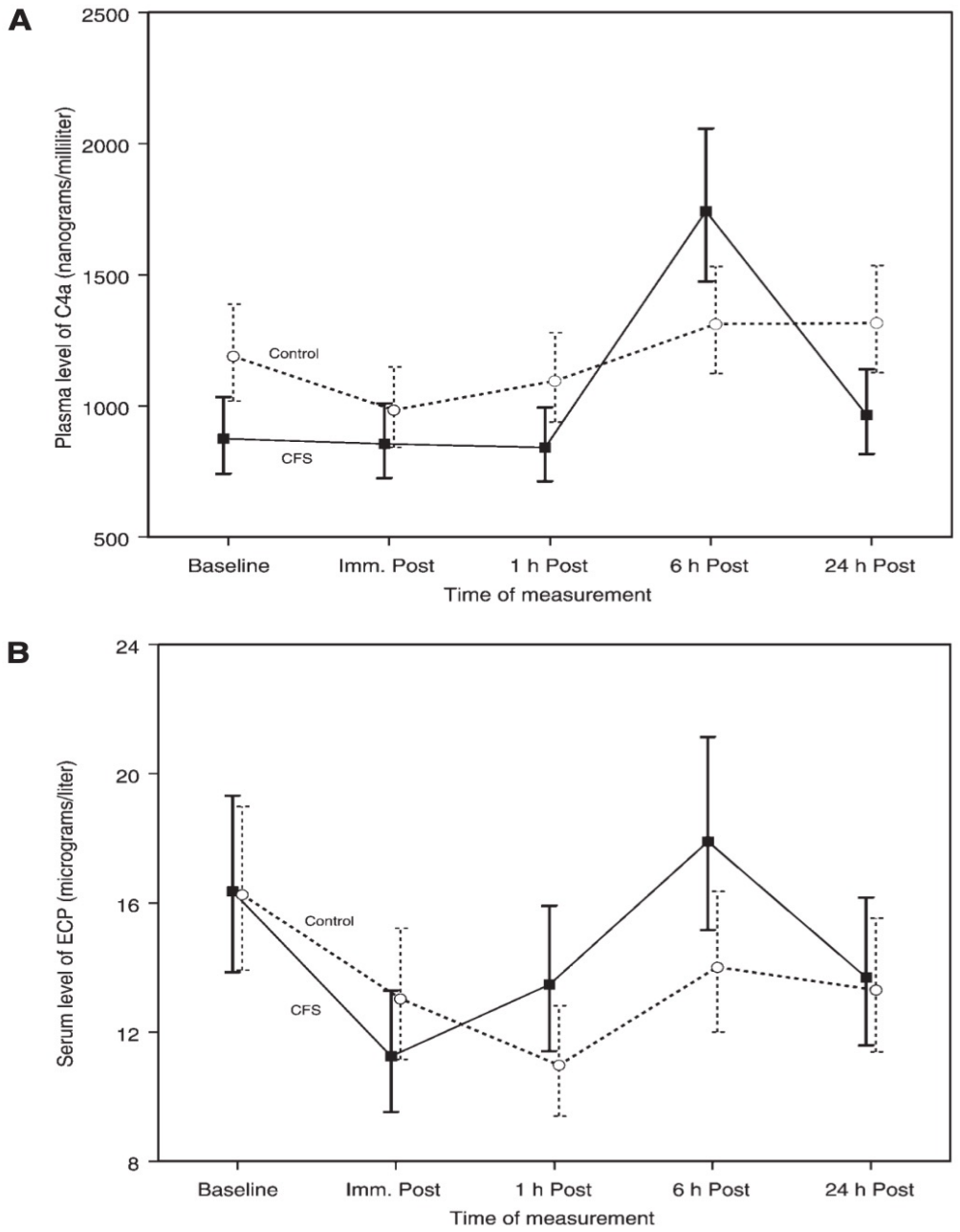


FIG 1. A and **B**, Estimates of mean C4a and ECP levels before and after exercise challenge for CFS and control populations, with standard error bars. Data were analyzed on the log scale and then inverted back for presentation, resulting in a longer +se bars than –se bars.

COMPLEMENT ACTIVATION IN TEAR FLUID DURING OCCUPATIONAL MOLD CHALLENGE

- Complement pathways present in eye tissues and fluid
- **Mold exposure causes complement activation in the eye more frequently than eosinophilia**

A	Exposure	Removal
Exposure	↑tear fluid C3a ↑Rare eosinophilia	↓ C3a and symptoms during sick leave ↓ eosinophilia
Control	No change	No change

COMPLEMENT SYSTEM IN LUNG DISEASE

- Complement activation can exacerbate lung injuries or conditions – which include:
  - **Asthma**
  - **pulmonary artery hypertension**
  - **idiopathic pulmonary fibrosis**
- Lung may provide a local source of complement proteins

COMPLEMENT PROTEINS AND THE LUNGS

Lung Cell Type	Complement Proteins
Alveolar type II epithelial cells	C2, C3, C4, C5, factor B
Bronchilolar epithelial cells	C3
Alveolar macrophages	C5a

Lung Conditions	Complements
Increased pulmonary arterial pressure	↑plasma C3 and C4a
Asthma	↑C3a and ↑C5a in bronchoalveolar lavage fluid in response to allergen challenge in patients with asthma C3a may regulate recruitment and activation of Th2 cells

MODEL OF IMMUNE COMPLEX-MEDIATED LUNG INJURY

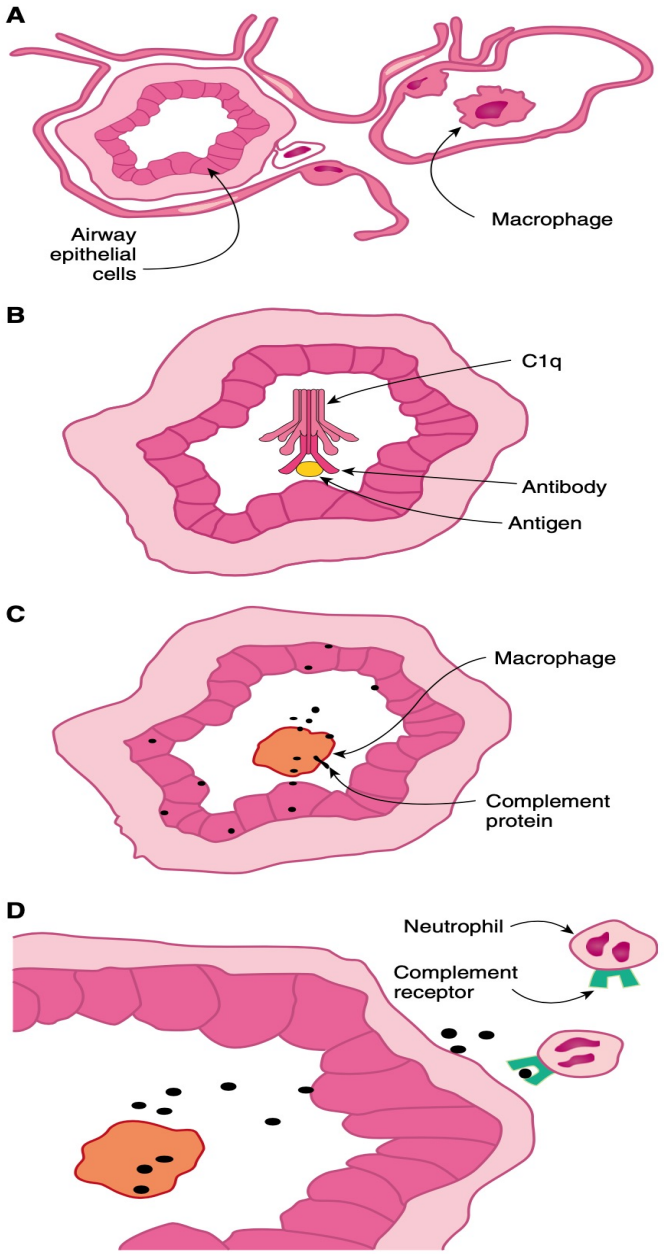


Figure 2. Model of immune complex–mediated lung injury. (*A*) Normal airways. (*B*) Injury to the lung can be mediated by immune complex–mediated injury in the lung epithelium. Antigen inhaled or present in the lung is bound by its antibody. C1q binds to the Fc portion of the antibody and activates complement cascade. (*C*) Lung injury can also initiate inflammation by complement activation through local synthesis of complement proteins from alveolar macrophages or airway epithelium. (*D*) Lung inflammation is exacerbated by complement proteins that act as chemoattractants for neutrophils.

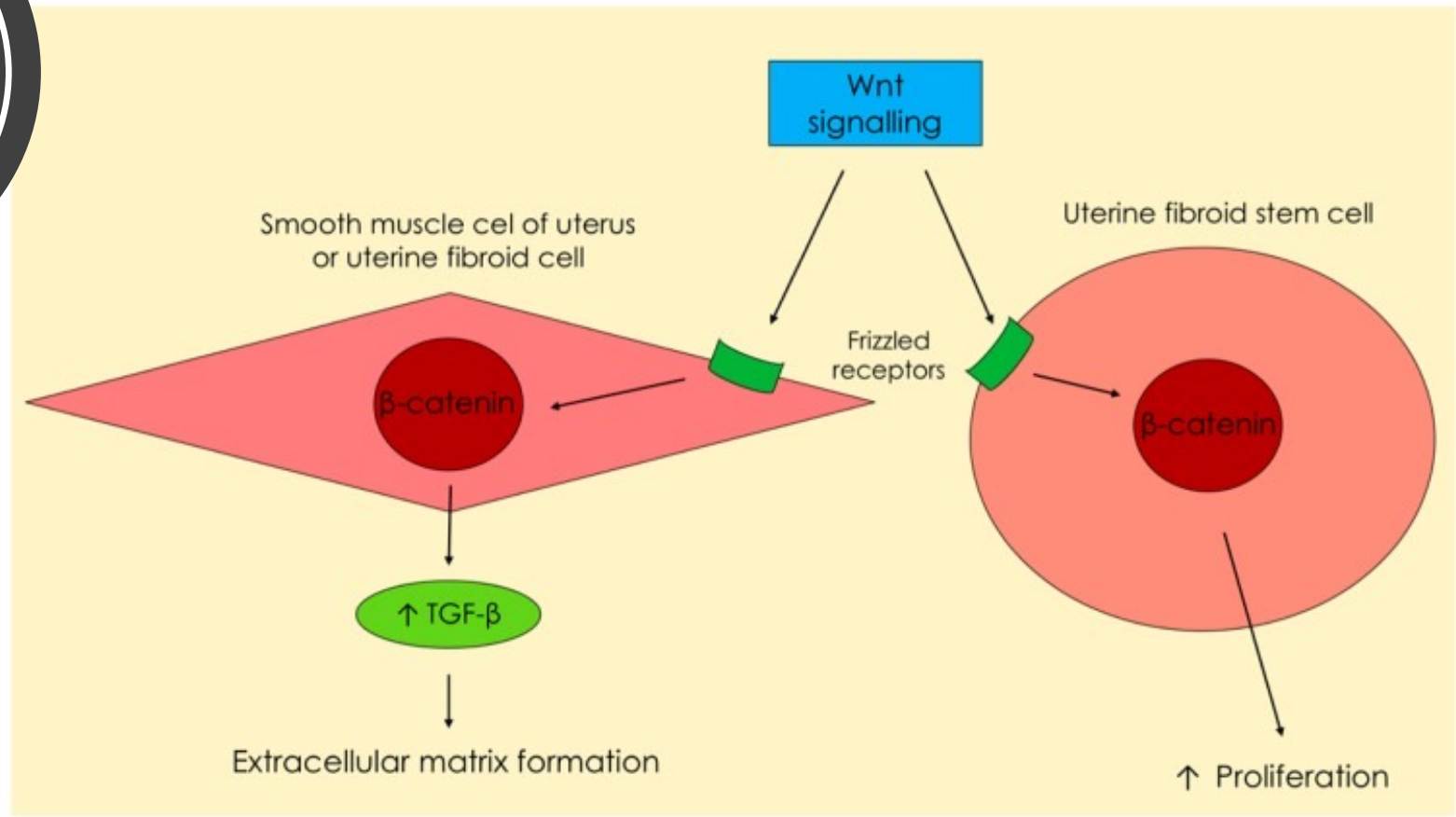
PATHOGENESIS BY TGF-**B** IN DIFFERENT PARTS OF THE BODY

- Varying magnitudes of **TGF-β expression** has been shown to induce malignant responses throughout the body
  - **Uterine fibroid**
  - **Lung monocytes and granulomas**
  - **Breast cancer**
  - **Pyogenic granuloma**
  - **High secretory immunoglobulin A (IgA)**
  - **Airway remodeling in asthma**

UTERINE FIBROID

- Uterine fibroids (UF) are benign tumors of the female genital tract made of the smooth muscle of the uterus
- Dependent on steroid hormone and selected growth factors
  - TGF-**β** is considered a **key** factor in pathophysiology of UF's

WNT/ TGFB SIGNALING PATHWAY IN UF'S

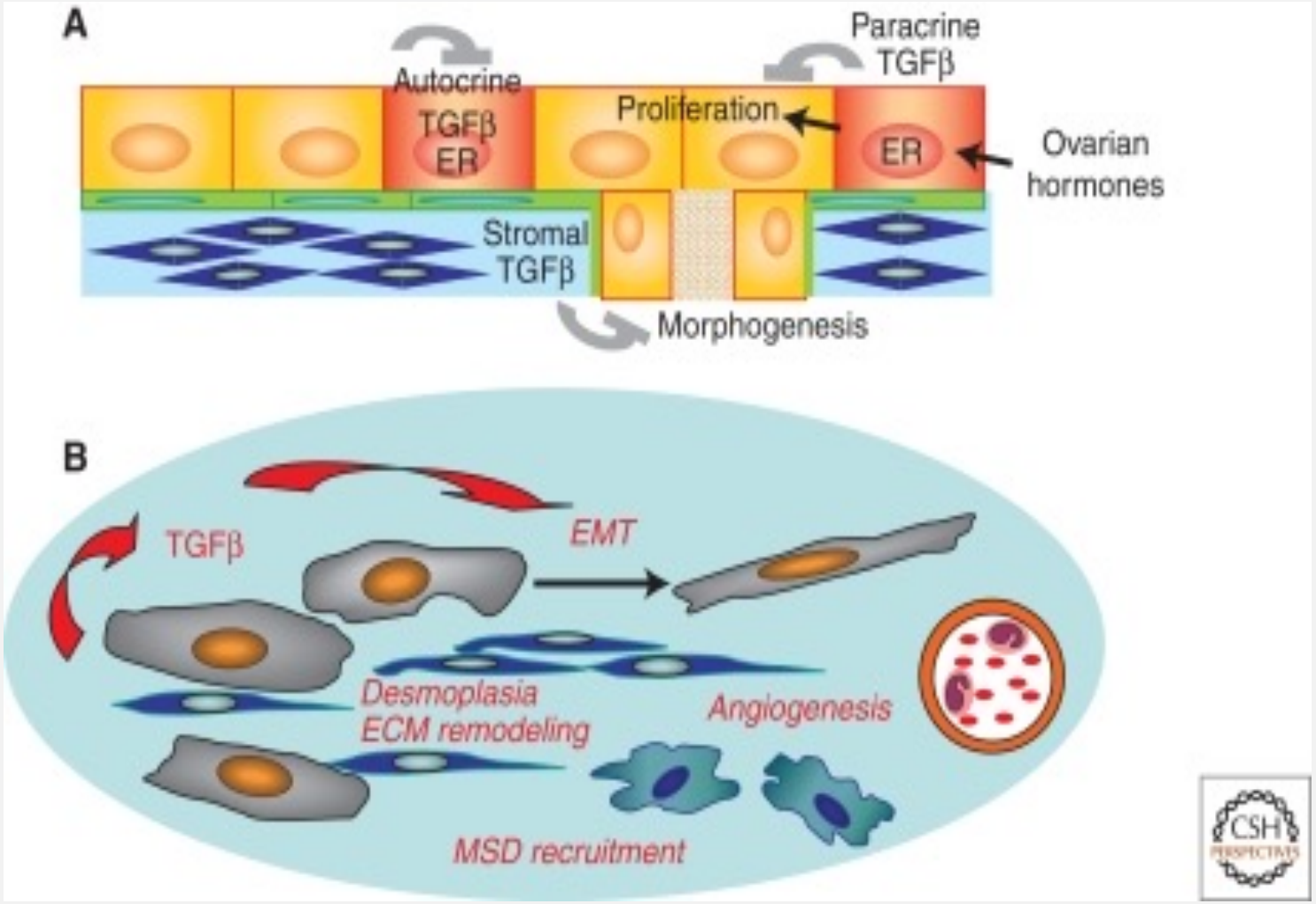


ROLE OF TGF-**B** IN MAMMARY DEVELOPMENT AND BREAST CANCER

- TGF-**β** is important for regulatory functions of mammary cells
- Limited by its nature as a latent complex, as it remains dormant until activated by a cellular response/signal
  - Studies in mice show TGF-**β**1 activity is actively regulated by estrogen and progesterone
- **Loss of TGF-β regulation mechanisms induces deregulation** of cellular interactions and phenotypes which lead to invasive disease

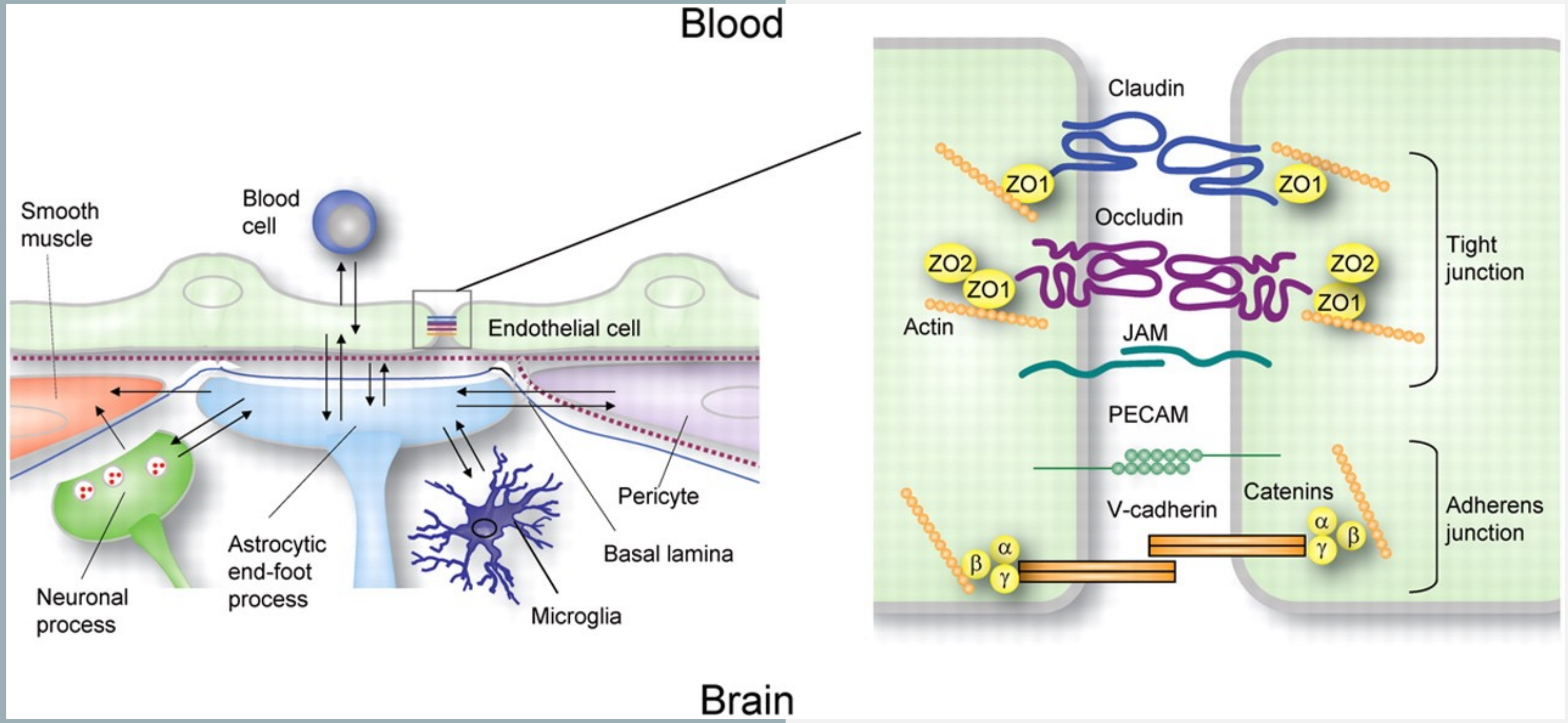
COMPARISON OF TGF-**B** IN NORMAL MAMMARY AND BREAST CANCER CELLS

- Mammary epithelial cells (Fig. A) are sensitive to TGF-**β** which is highly regulated in normal tissue
- Breast cancer cells are nonresponsive to the cytotoxic action of TGF-**β**



MMP-9

- MMP is a family of over 23 enzymes capable of modifying the ECM
- Roles
  - Angiogenesis
  - Ovulation
  - Wound Healing
- Uncontrolled regulation
  - Tissue Injury
  - Inflammation


Blood
Smooth muscle
Blood cell
Endothelial cell
Pericyte
Basal lamina
Astrocytic end-foot process
Neuronal process
Microglia
Brain
Claudin
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ZO1
Occludin
ZO2
ZO1
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Actin
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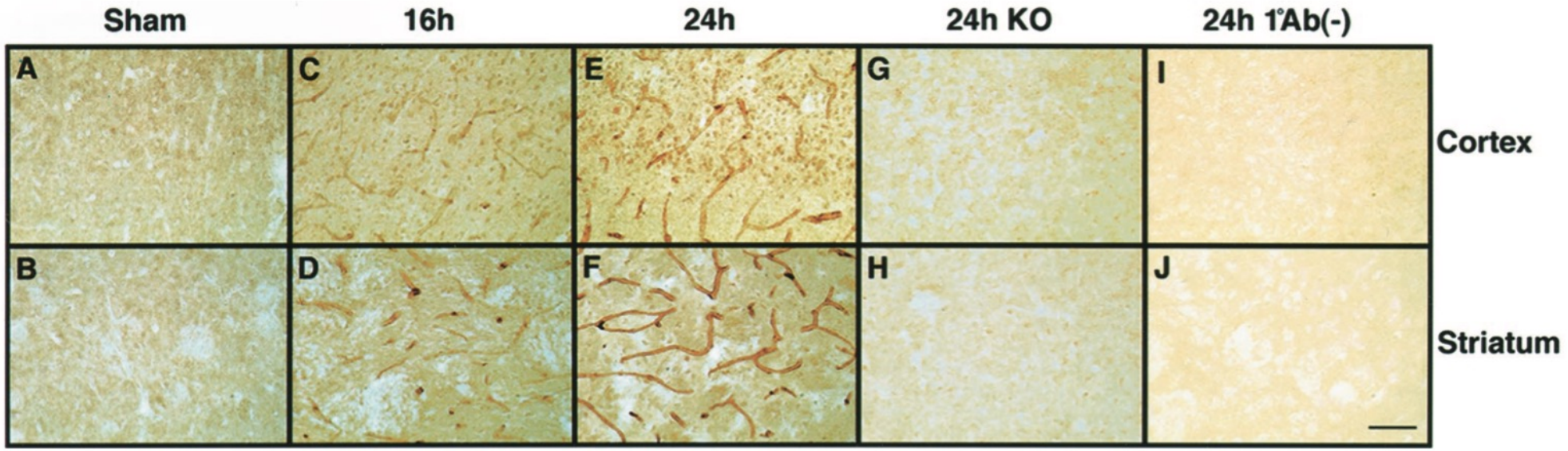
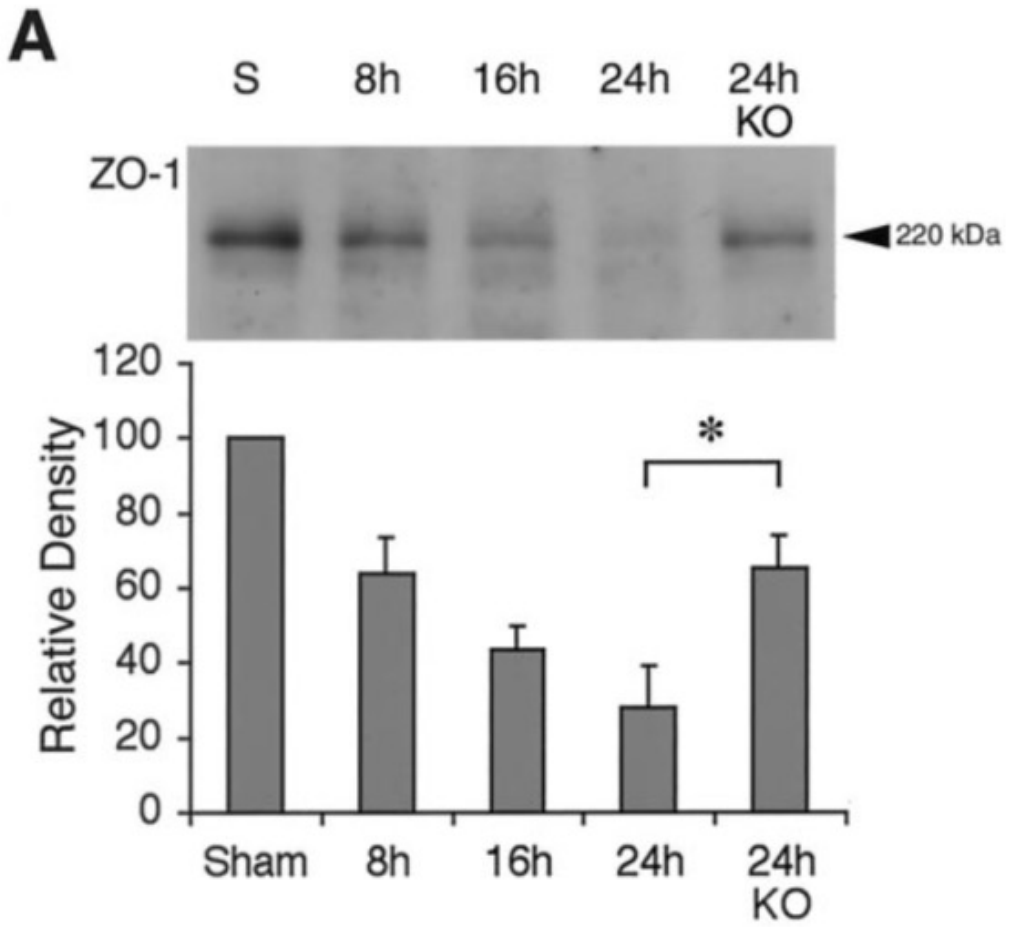


Figure 3. Immunohistochemistry with anti-MMP-9. *A, B*, Sham-operated brains showed no MMP-9 expression. *C–F*, MMP-9 staining increased in wild-type mouse brain after transient focal ischemia. Diffuse MMP-9 immunoreactivity was observed within both the cortical and striatal areas of ischemic hemispheres. Immunoreactive MMP-9 mainly appeared in endothelial cells. In addition, MMP-9 expression was also detectable in parenchymal cells of ischemic regions. *G, H*, No immunoreactive cells were observed in ischemic brains of knock-out mice (*KO*). *I, J*, Negative controls incubated without primary antibody showed no staining. Scale bar, 50 μm.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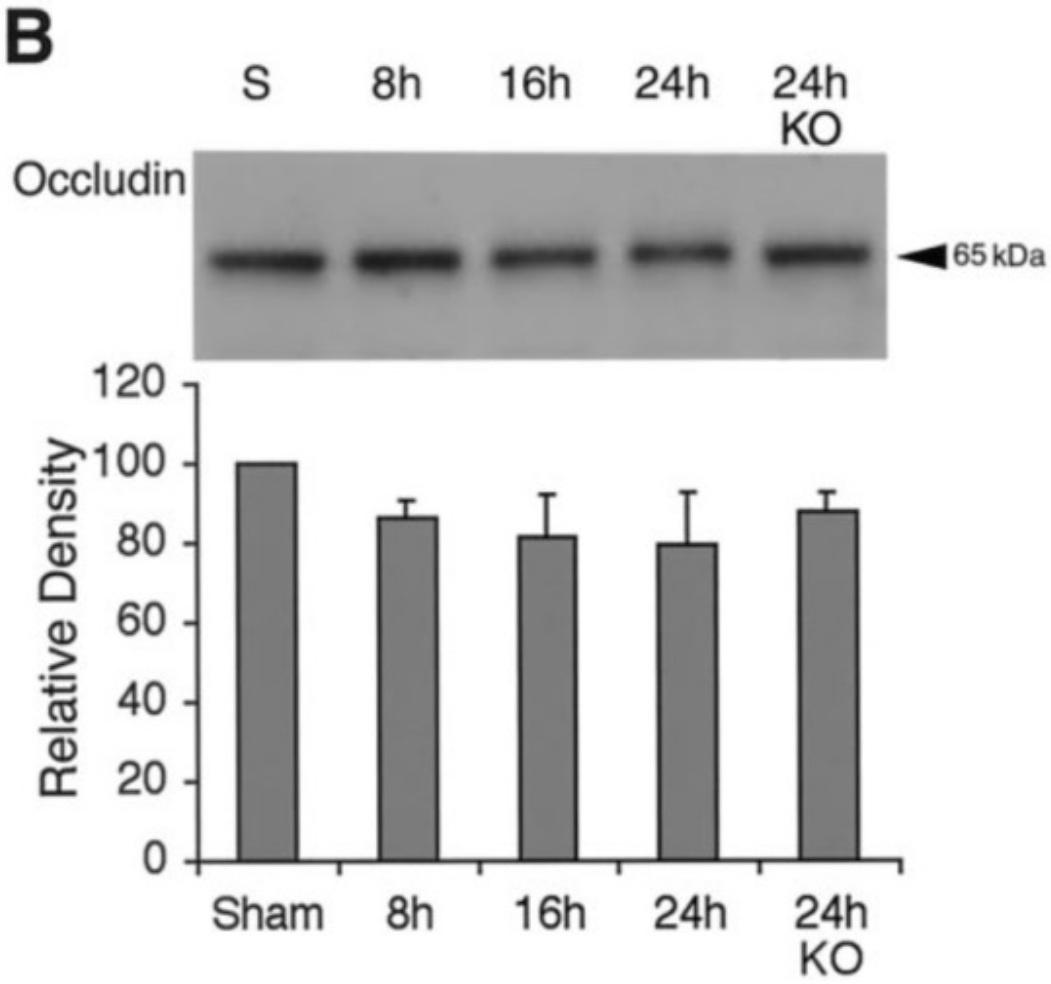
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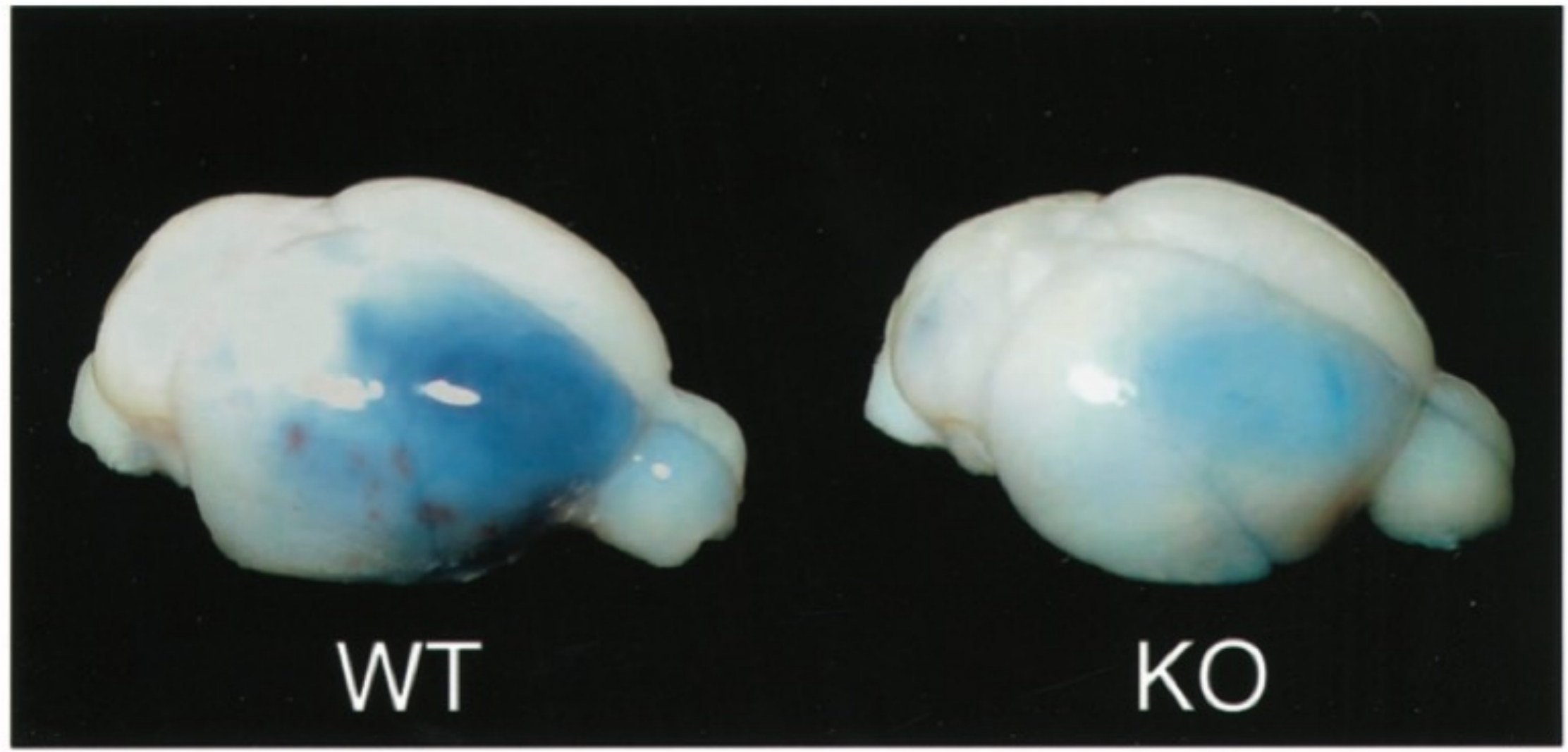
WT
KO

SUMMARY

- Water-damaged Building causes Chronic Inflammation
- Chronic Inflammation defined by proteomic elevations
- Plasma proteomics link to illness supported by basic and clinical research

ESTABLISH CAUSATION

THANK YOU

- RITCHIE SHOEMAKER, M.D.
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