A new approach to understanding the pressure gradients in the brain that cause hydrocephalus

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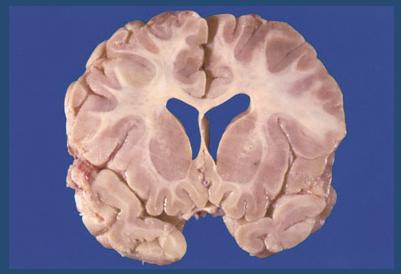
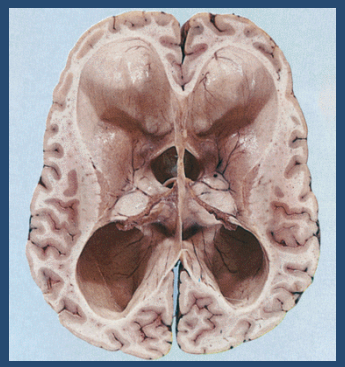
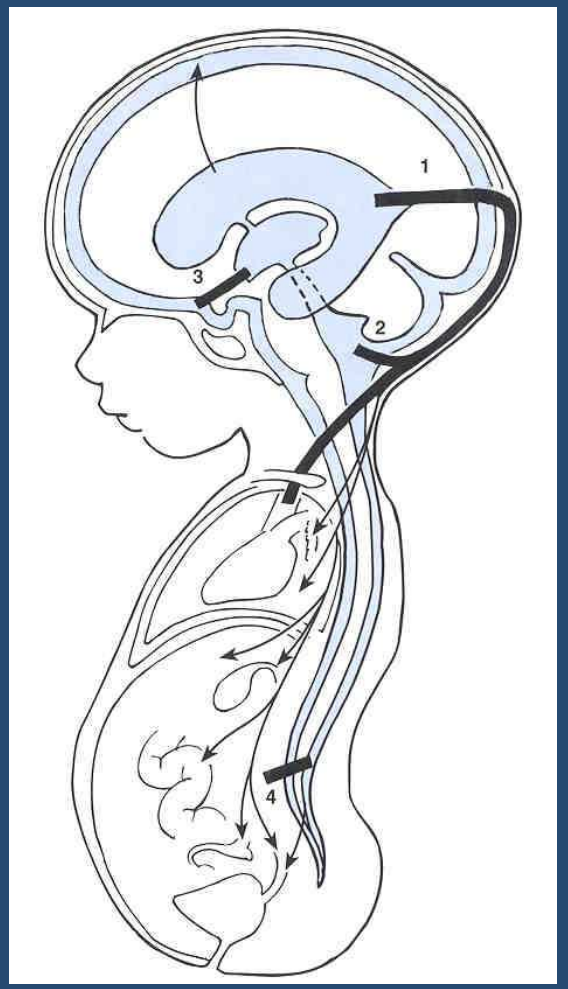
Causes of hydrocephalus

- Injury (trauma, intracranial hemorrhage etc)
- NPH and ageing
- Cytokine concepts
- Pulsatility
- 40 mutants and 9 genes have been identified in animal models. Most of the gene products are the cytokines and growth factors involved in brain development
- In humans, only one HC gene has been identified (X-linked) encoding for the cell adhesion molecule L1



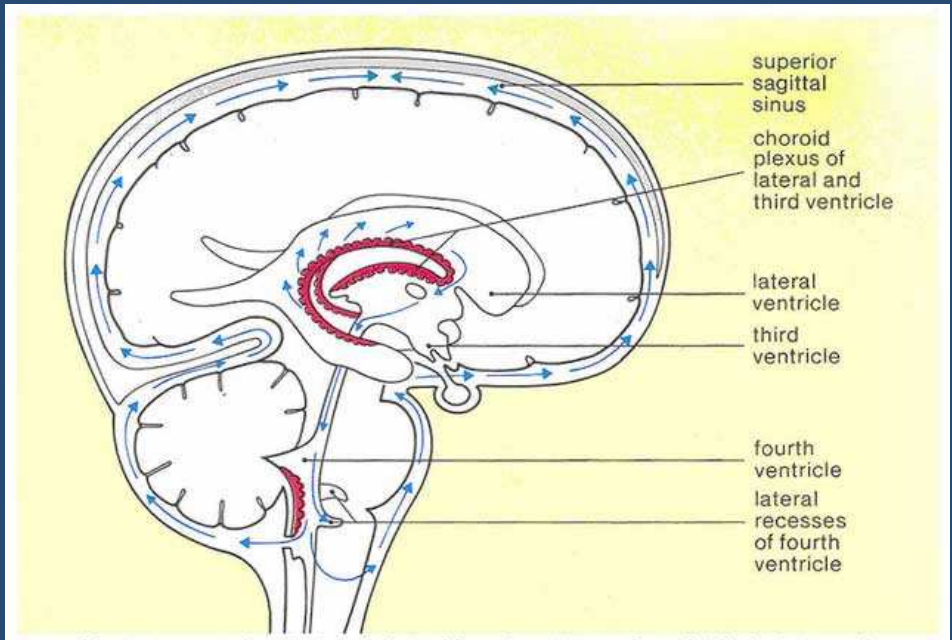




Hydrocephalus definition

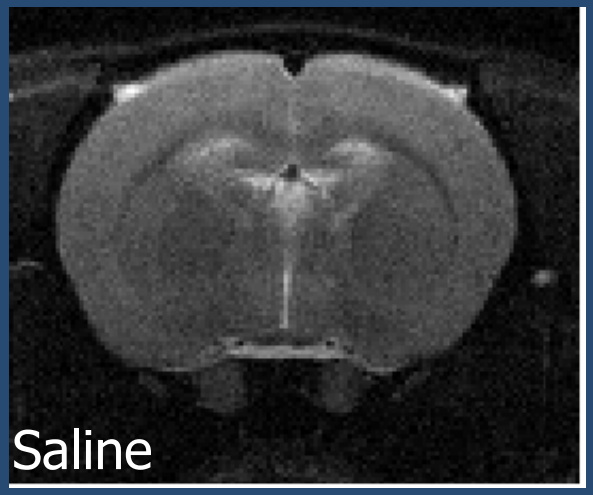
Hydrocephalus is an active distension of the ventricular system of the brain related to inadequate passage of CSF from its point of production within the ventricular system to its point of absorption into the systemic circulation

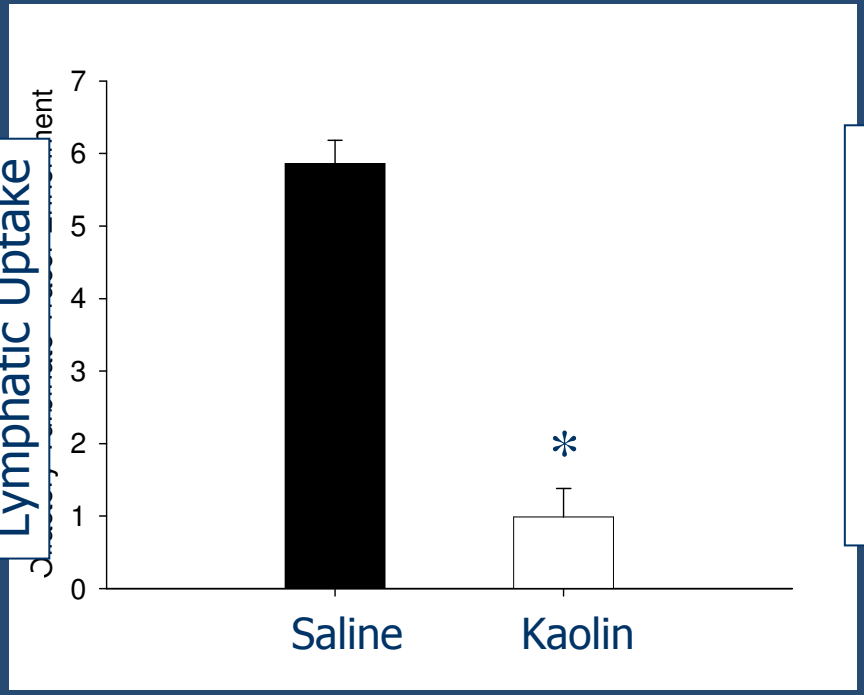
Rekate H
Semin Pediatr Neurol. 2009 Mar;16(1):9-15

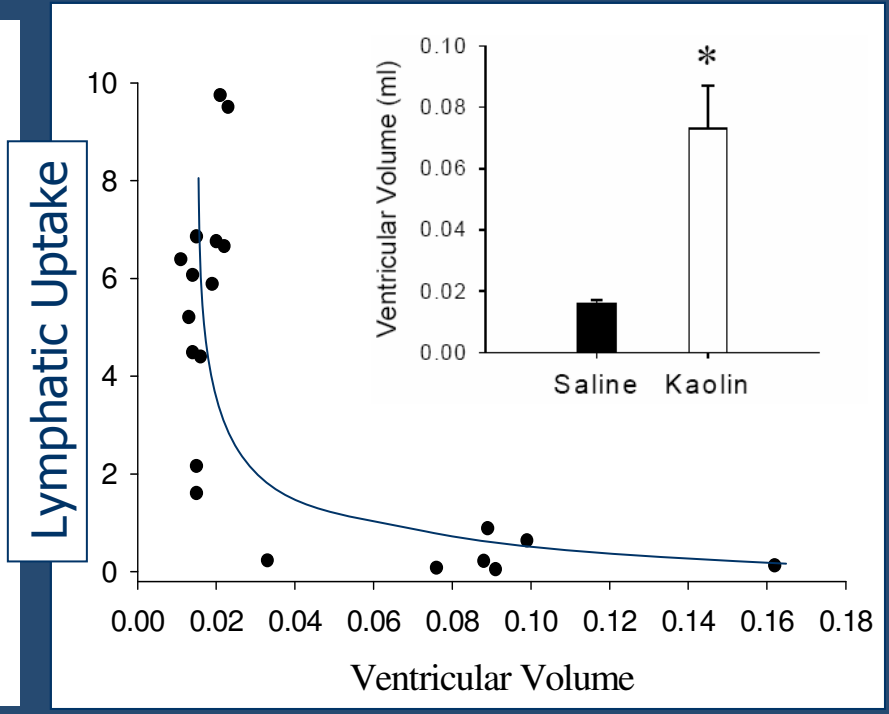


Perkin D, Rose FC, Blackwood W, Shawdon HH. Atlas of Clinical Neurology. Philadelphia: JB Lippincott, 1986. Fig 1.1.

Lymphatic CSF absorption is impaired in a rat kaolin hydrocephalus model

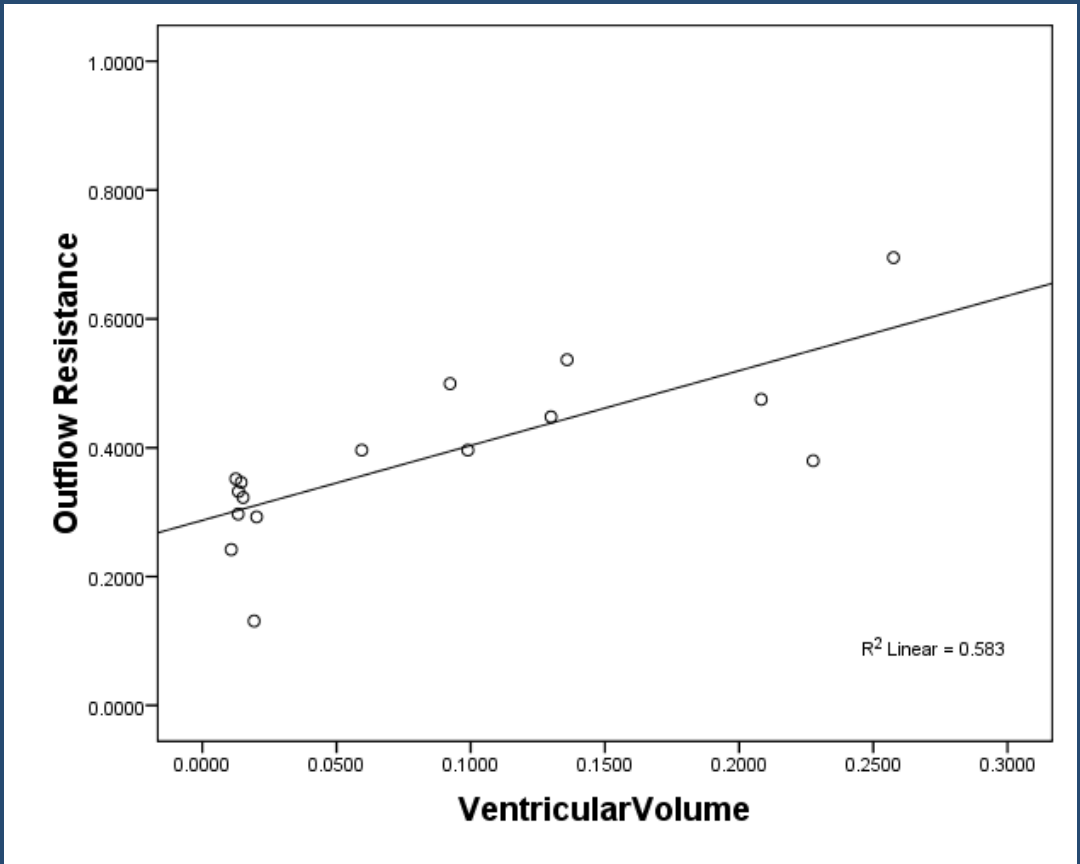






Nagra et al., 2007

CSF absorption deficit (outflow resistance) correlation with ventricular volumes

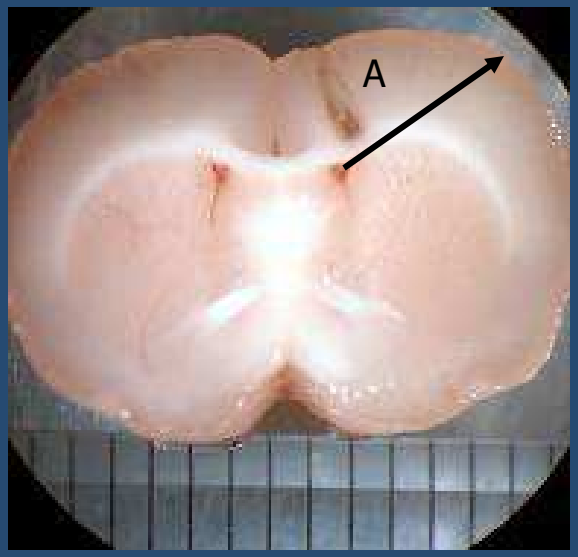


The linear regression line reveals that there is a significant correlation between these two variables, with $r^2 = 0.583$ ($p<0.0001$)

Pressure gradients that facilitate ventricular enlargement

Where are they ??? It has been very difficult to measure trans-cerebral pressure gradients

Ventricle to subarachnoid space (A)



Normal

Hydrocephalus

What is the mechanism that causes the pressure gradient ?

Levine DN. Intracranial pressure and ventricular expansion in hydrocephalus. Have we been asking the wrong question? J Neurol Sci 269: 1-11, 2008.

Classical View

- Initiating event, is an obstruction to CSF flow
- A very small gradient of pressure (< 1 mm Hg) between ventricle and subarachnoid space is sufficient to induce hydrocephalus. This small gradient is needed to overcome the increased resistance to CSF flow so that CSF absorption can balance CSF formation.
- Ventriculomegaly occurs at the expense of the brain's interstitial fluid. Interstitial fluid pressure will increase and will be 'driven' into blood capillaries. Capillary resorption of interstitial fluid reduces the transmission of ventricular pressure to the periphery.

Problems with the ‘classical’ concept

- In communicating HC, how would pressure gradients form?
- Levine’s postulated small pressure gradients have never been verified directly.
- While hydrocephalus is associated with elevations in CSF outflow resistance, the opposite is not necessarily true as elevations in CSF outflow resistance do not always correlate with hydrocephalus (pseudotumor cerebri)
- In previous studies from our group, it was clear that a lymphatic CSF absorption deficit occurred in ageing rats and yet no hydrocephalus was present in these animals
- In 3 sheep, we sealed the cribriform plate and even though that this procedure elevates intracranial pressure and CSF outflow resistance in this species, we did not observe any ventricular expansion over 3 months (unpublished observations)

It is not apparent whether an impediment to CSF drainage represents a pivotal event in hydrocephalus development or whether it is a ‘co-conspirator’ in the pathogenesis of ventricular enlargement with some other factor denoting the definitive cause

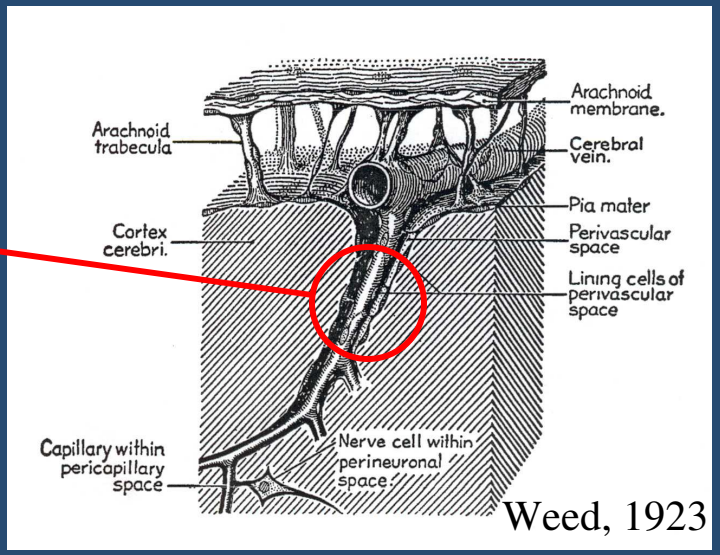
Other possibilities ?

Extracellular fluid in the brain is unusual (two compartments)

Parenchymal Interstitial fluid

CSF

CSF absorption



Weed, 1923

Two independent lines of evidence from the literature suggest that interstitial pressures may decrease

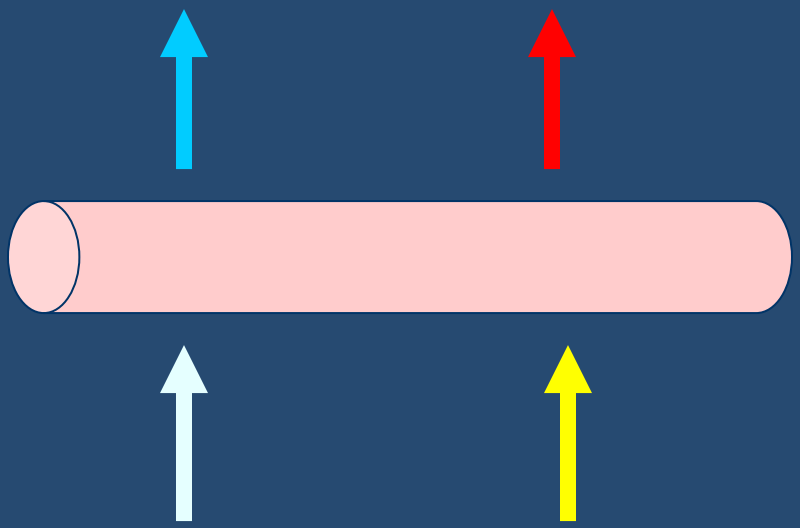
Reed's group in Norway

Pickard's group in the UK

Work of Professor Reed and colleagues: Interstitial matrix actively contributes to regulation of interstitial fluid pressure

- Lund et al., Am J Physiol 255:H1069, 1988
- Reed and Rodt, Am J Physiol 260: H1985, 1991
- Koller and Reed. J Appl Physiol 72: 53, 1992
- Reed et al., Circ Res 71: 978, 1992

Starling Forces

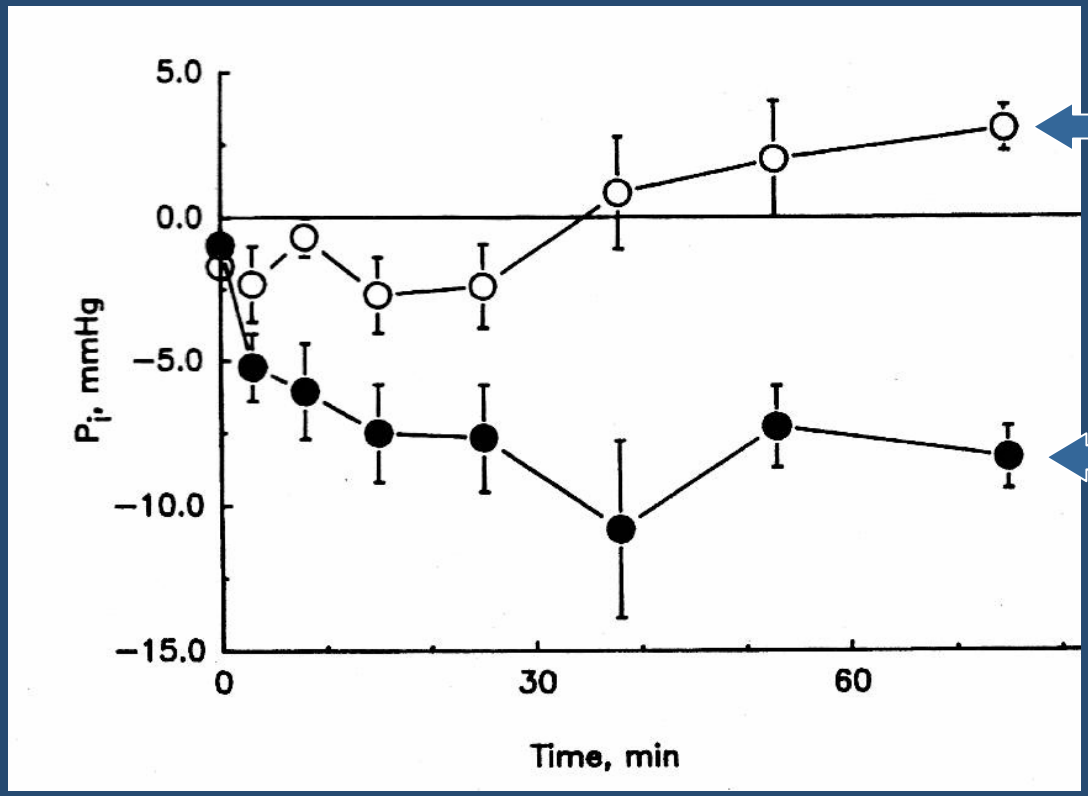


capillary

$$Q = K\,(Pmv\text{-}Pi) - \delta(\pi mv\text{-}\pi i)$$

Problem – in inflammation, the measured accumulation of interstitial water was often much greater than that estimated from calculated values based on steady-state data

Work of Professor Reed and colleagues: Interstitial matrix actively contributes to regulation of P(i)



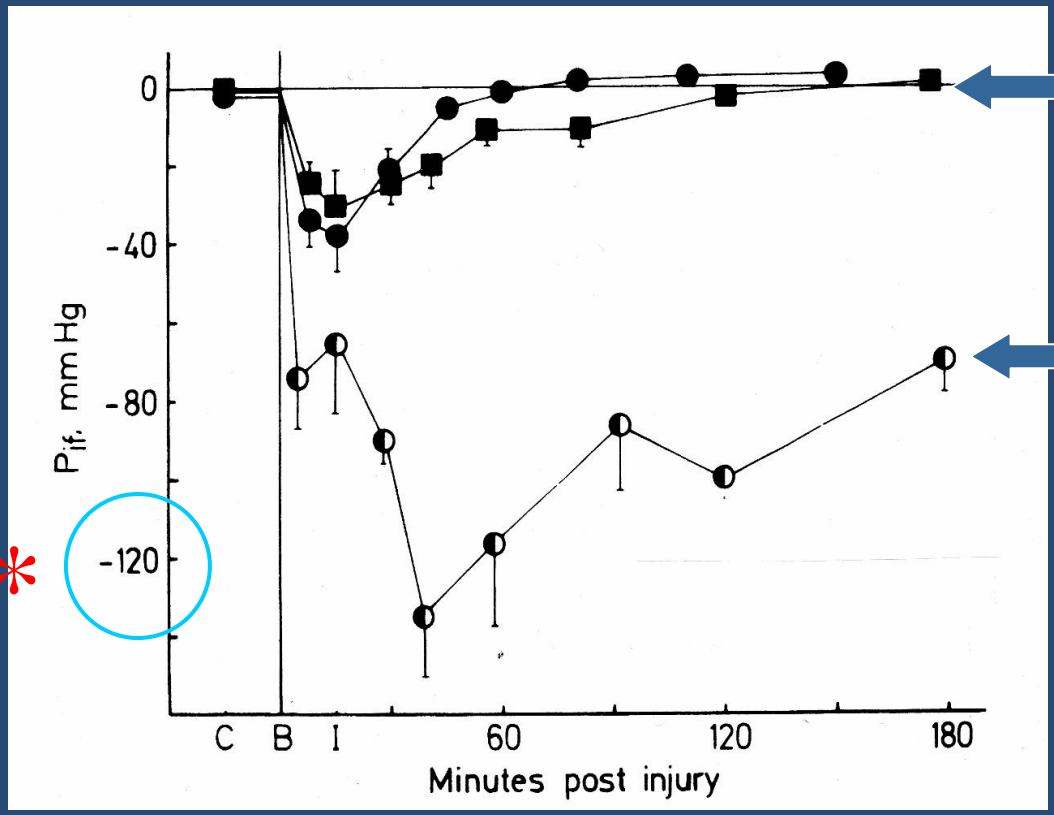
Intact circulation

Circulatory arrest

Effect of dextran anaphylaxis in P(i) in rat paw skin

Circulatory arrest limits the increased capillary fluid filtration associated with the anaphylactic reaction. This increased filtration will otherwise raise interstitial volume and thereby p(i) and cause an underestimation of a potential increased negativity of p(i).

Impact of full thickness burn injury on P(i) in rat skin



Intact circulation

Circulatory arrest

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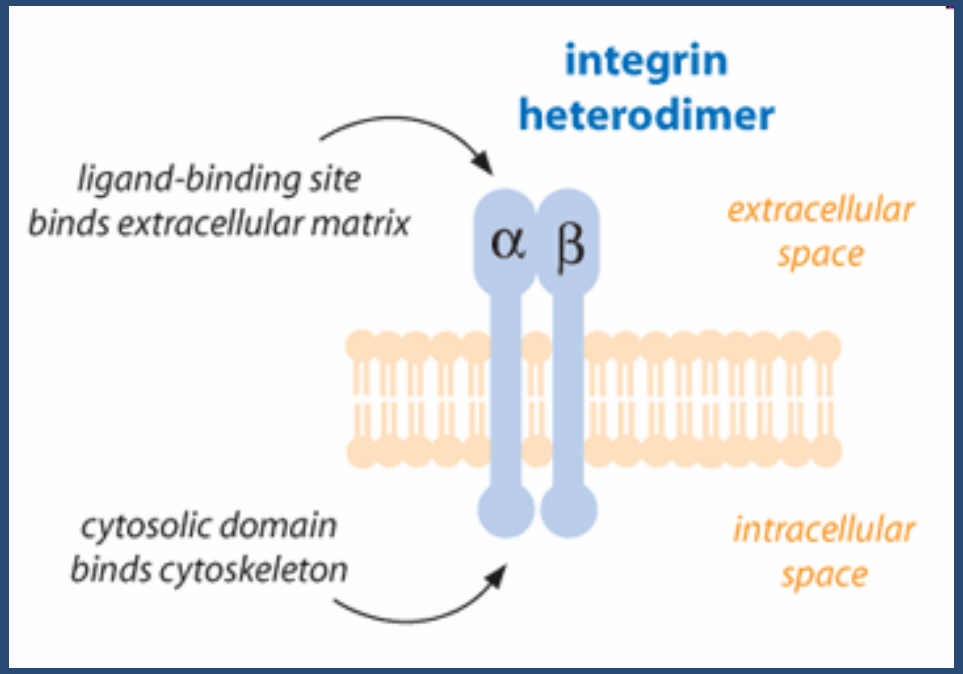
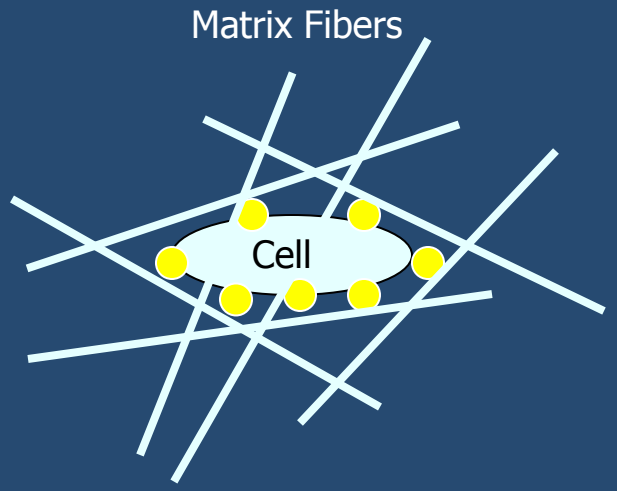
Integrin - Matrix interactions



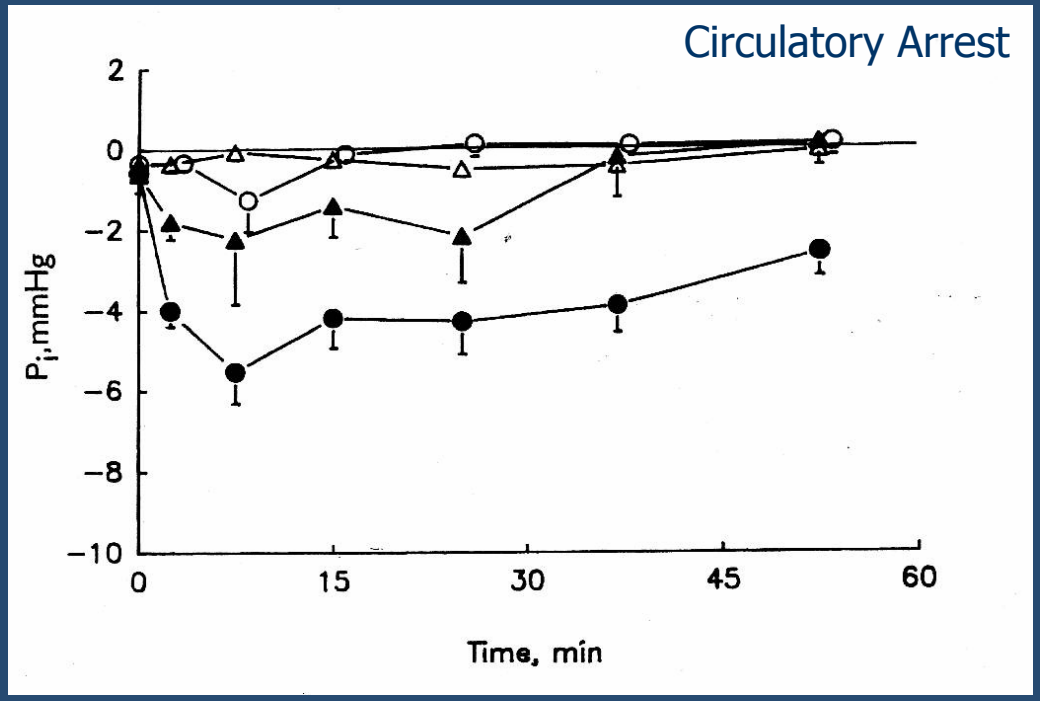


Integrin receptors

The evidence suggests that disruption of beta1 integrin-matrix interactions is the cause of the pressure effect

○ Pre-immune IgG

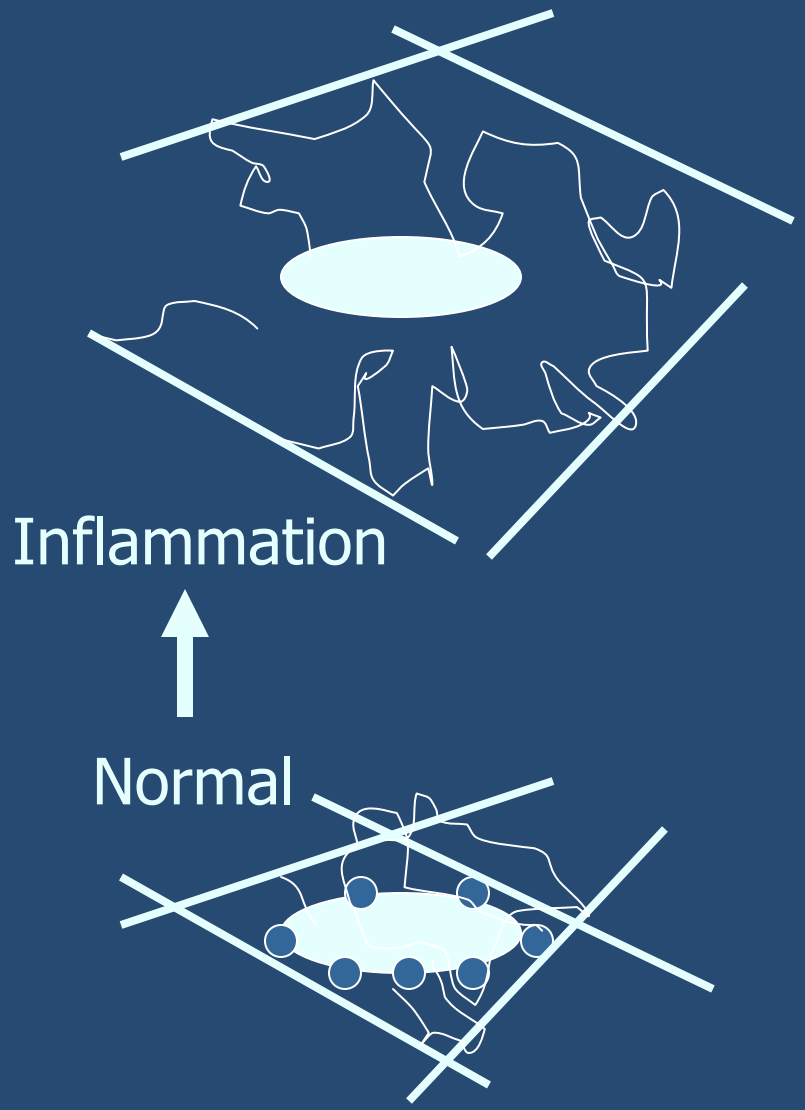
● Anti beta1 integrin



Concept:

Rubin and colleagues have proposed that fibroblasts in the skin regulate interstitial fluid pressure by exerting a tensile force on the collagen matrix, which restrains the interstitial gel from swelling.

Integrin function can be modulated by cytokines and other factors that regulate the balance between grip and release leading to compaction or tissue swelling which in turn affects interstitial fluid pressure

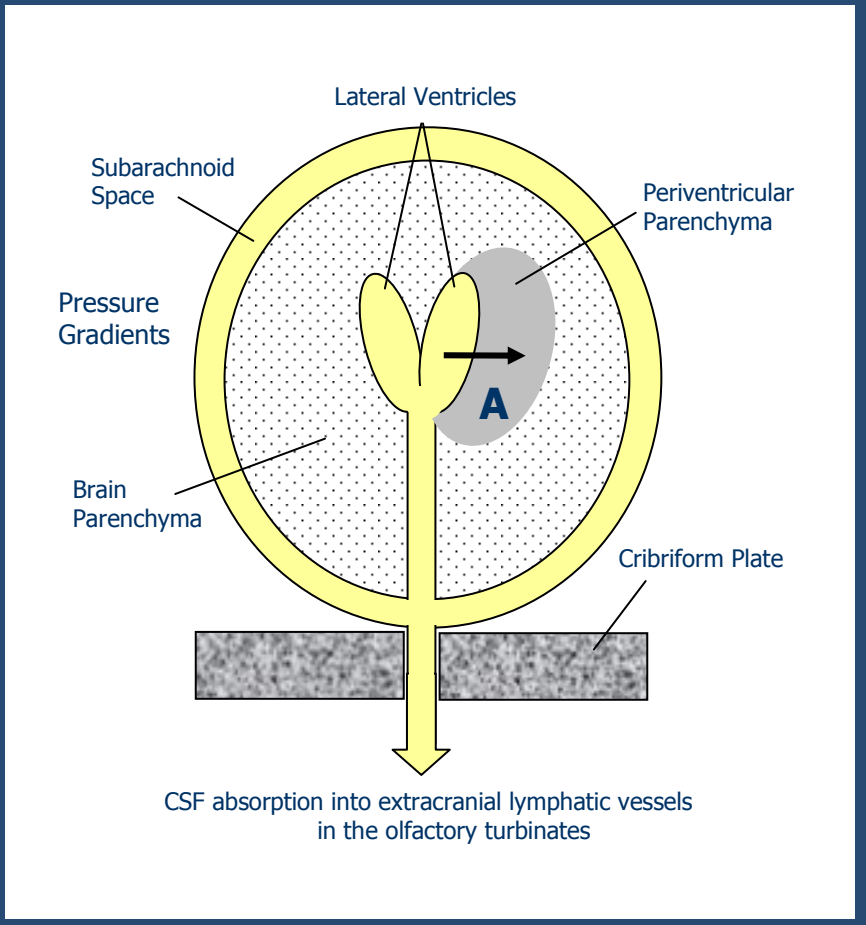


$$Q = K\,(Pmv\text{-}Pi) - \delta(\pi mv\text{-}\pi i)$$

Pickard Concept
Mathematical approach

Relative reduction in parenchymal interstitial fluid pressure in conjunction with a reduction in tissue elasticity may contribute to a trans-parenchymal pressure gradient (A) and hydrocephalus.

** Reverse flow into tissues with absorption of CSF in parenchyma



Peña A, Harris NG, Bolton MD, Czosnyka M and Pickard JD. Communicating hydrocephalus: the biomechanics of progressive ventricular enlargement revisited. *Acta Neurochir Suppl* 81: 59-63, 2002.

Can a similar phenomenon occur in the brain? Are there integrins in the brain?

B1-integrin expression in brain

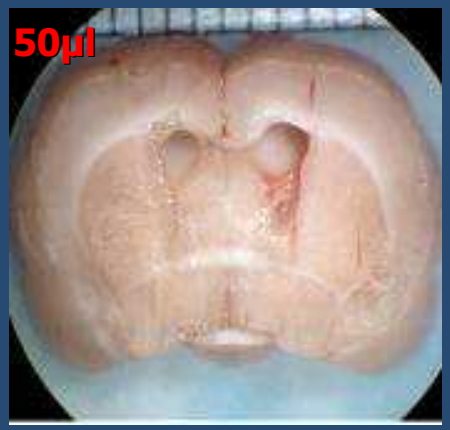
- associated with astrocytes
- blood vessel walls
- periventricular region and ependyma
- choroid plexus
- grey matter
- cerebral cortex

Do antibodies against the beta-1 integrins induce hydrocephalus ?

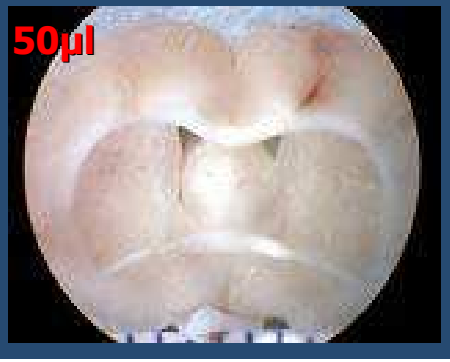
Impact of anti-integrin antibodies (assessed 2 weeks post injection): chronic experiments

Rabbit IgG isotype control

Hamster IgM isotype control

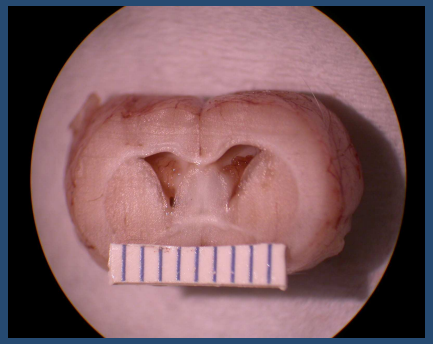


Anti-β1 integrin antibody (IgG)



Anti-β1 integrin antibody (IgM)

Anti-$\alpha_2\beta_1$ integrin antibody (IgG)



Kaolin injected

21 of 29 animals (72%) receiving antibodies to beta integrins developed hydrocephalus

No animal receiving the isotype controls developed hydrocephalus

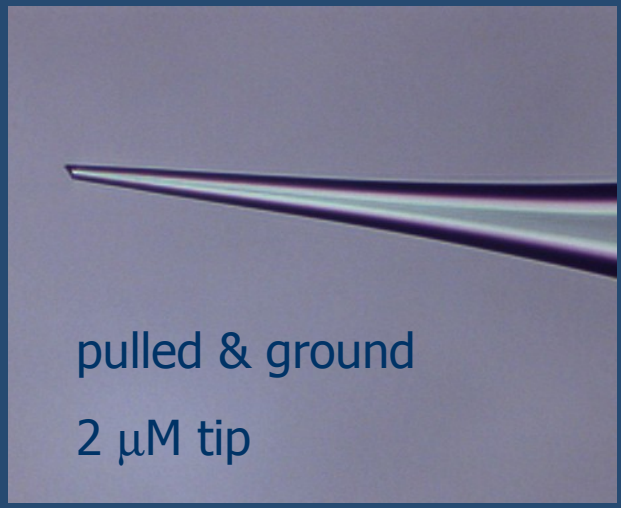
Do antibodies against the beta-1 integrins

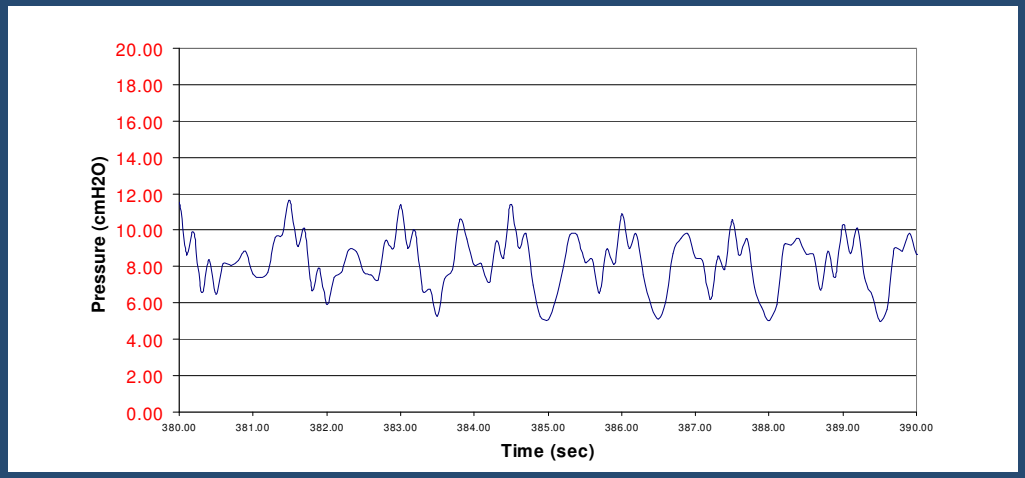
A) reduce parenchymal interstitial fluid pressure ?

B) induce ventricular to parenchymal pressure gradients ?

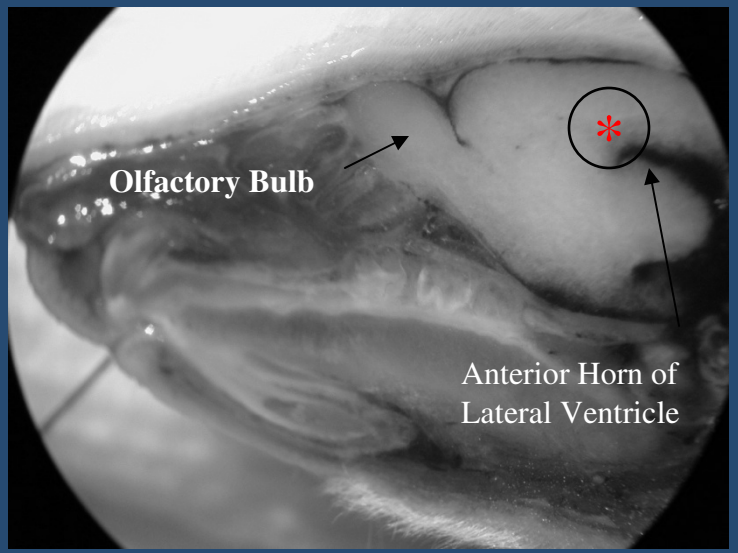
Servonull Micro-pressure System

Ability to quantify interstitial, capillary, and lymphatic pressures



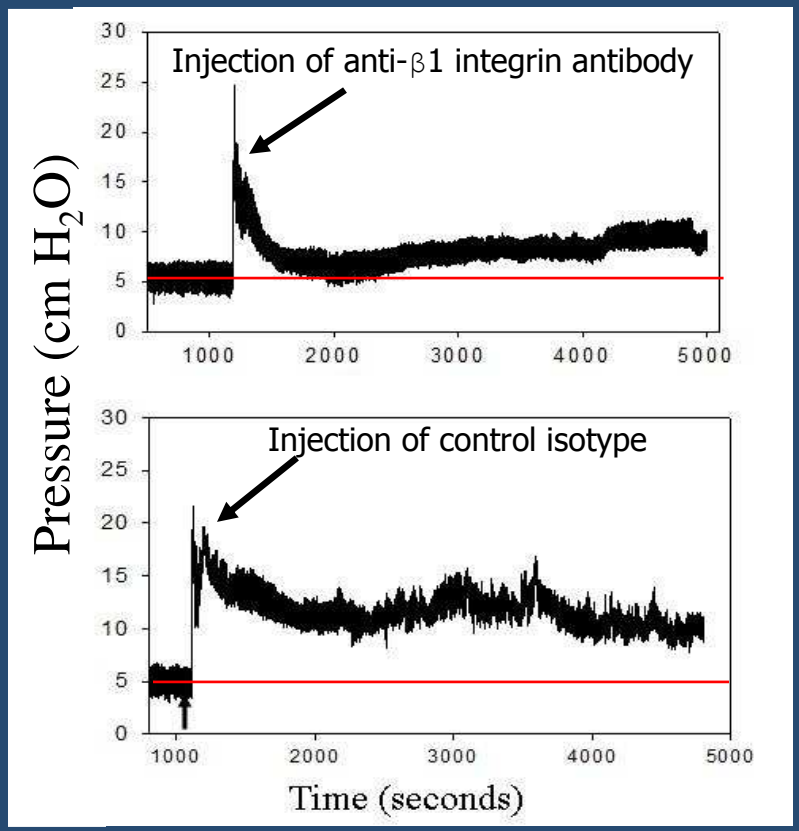


Resistance Null-Balance Feedback

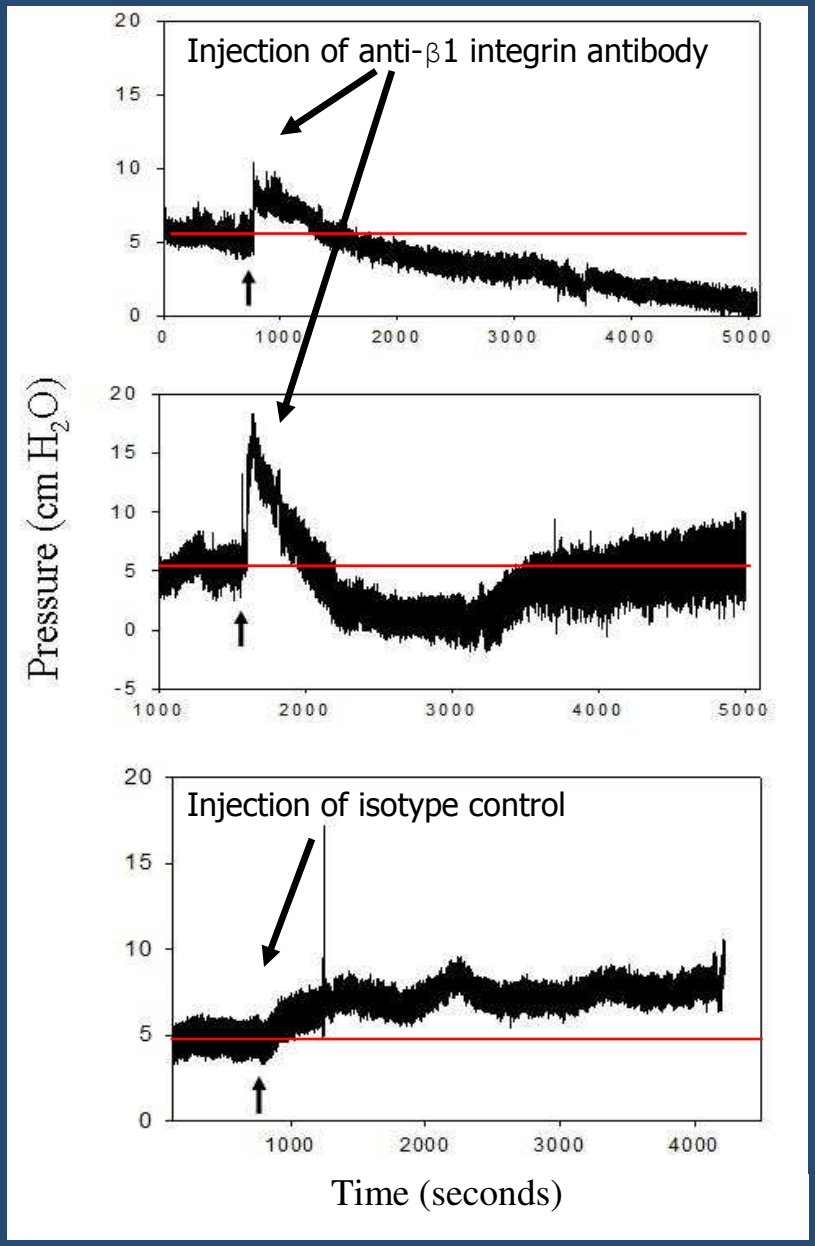


Examples of pressure traces:

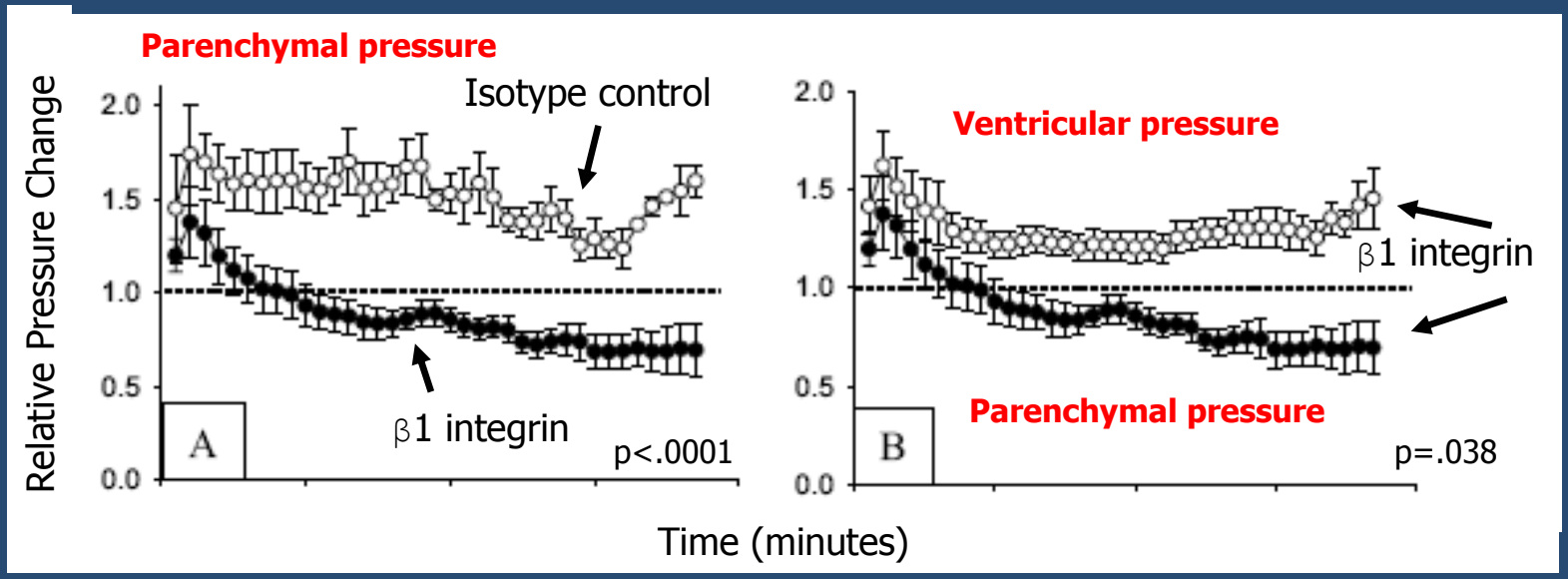
Ventricular Pressures



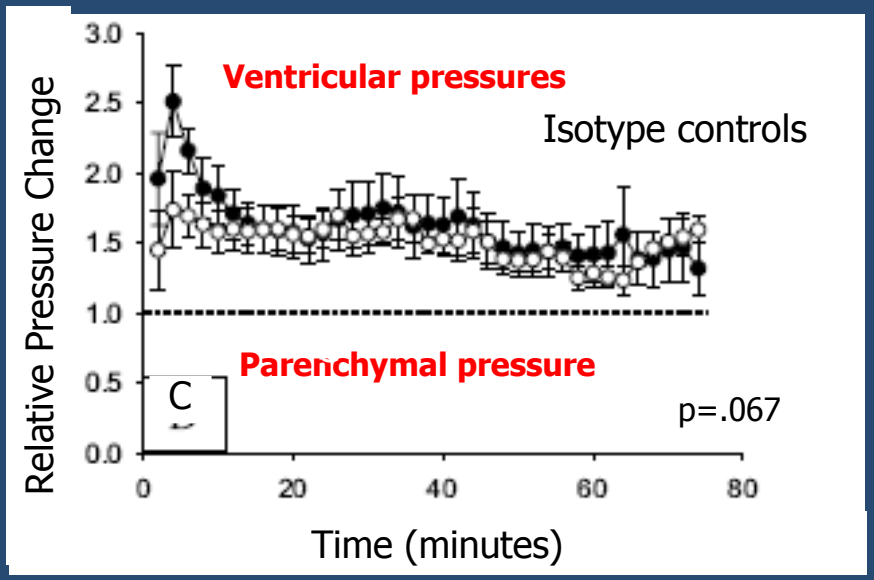
Parenchymal pressures



Impact of antibodies to β1Integrins on ventricular and parenchymal pressures



Antibodies to the beta-1 integrins can 'uncouple' ventricular and parenchymal pressures and induce pressure gradients between 4 - 5 cm H_2O.



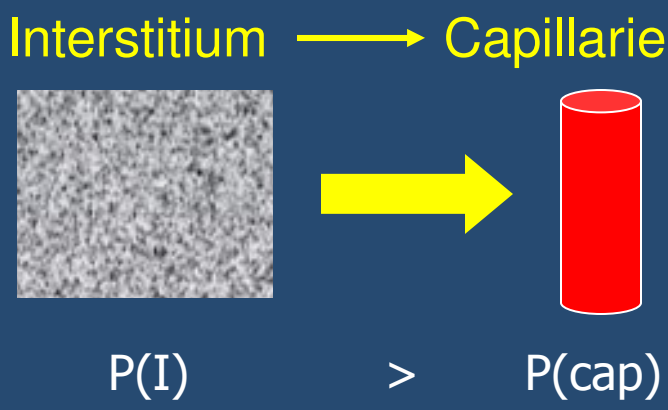
Matrix hypothesis

In order for pressure to equal in the ventricles and SAS but low in the parenchyma, some removal of fluid must occur from the tissues

Arciero J, Begg R, and Wilkie K.,
http://www.math.uwaterloo.ca/~kpwilkie/OCCAM_Hydrocephalus_Report.pdf 2009.

- Would this water removal be governed by hydrostatic or osmotic forces?
- Molecular mechanism ?

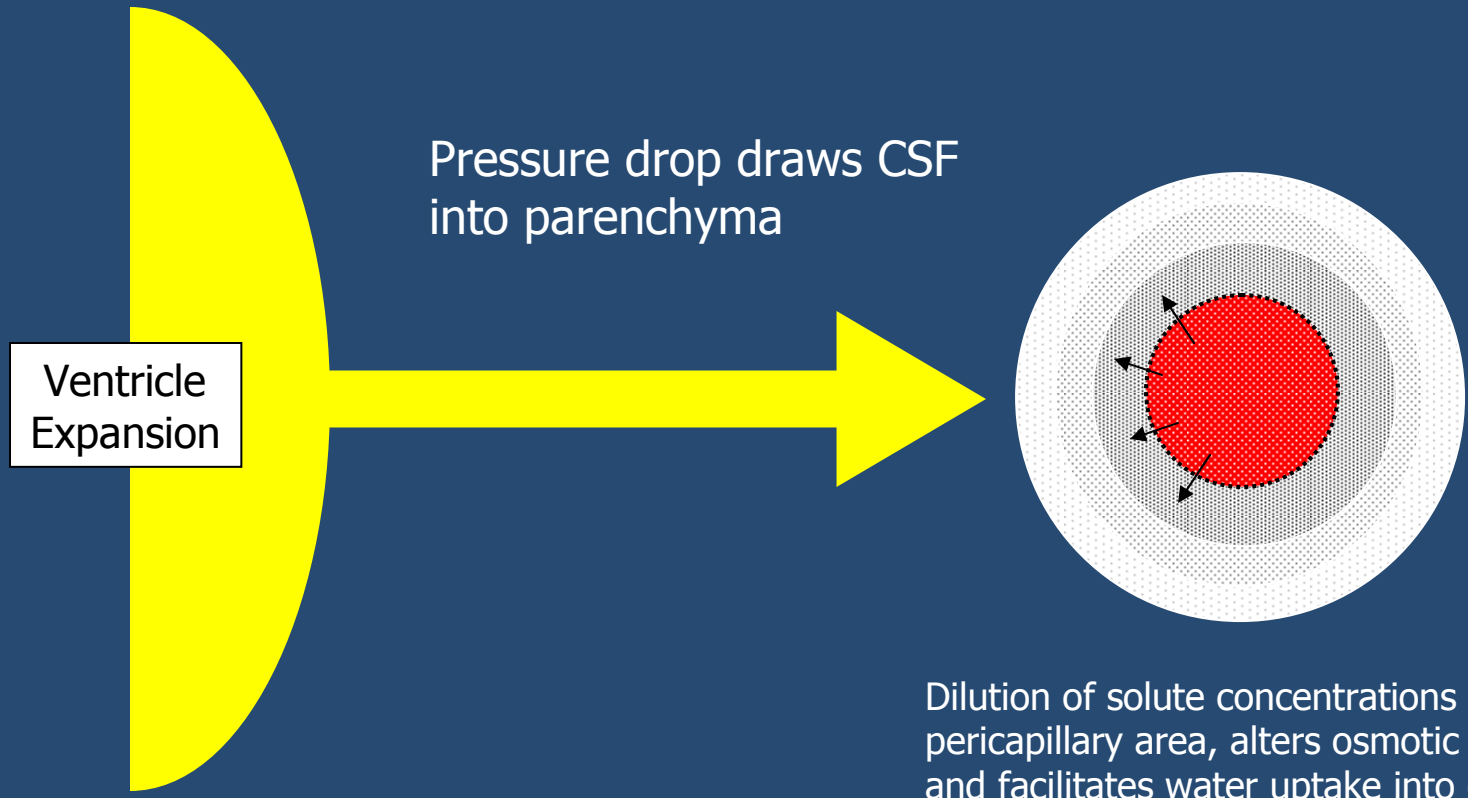
Water absorption - hydrostatic gradients



Hydrostatic pressure gradients force fluid into the capillary network

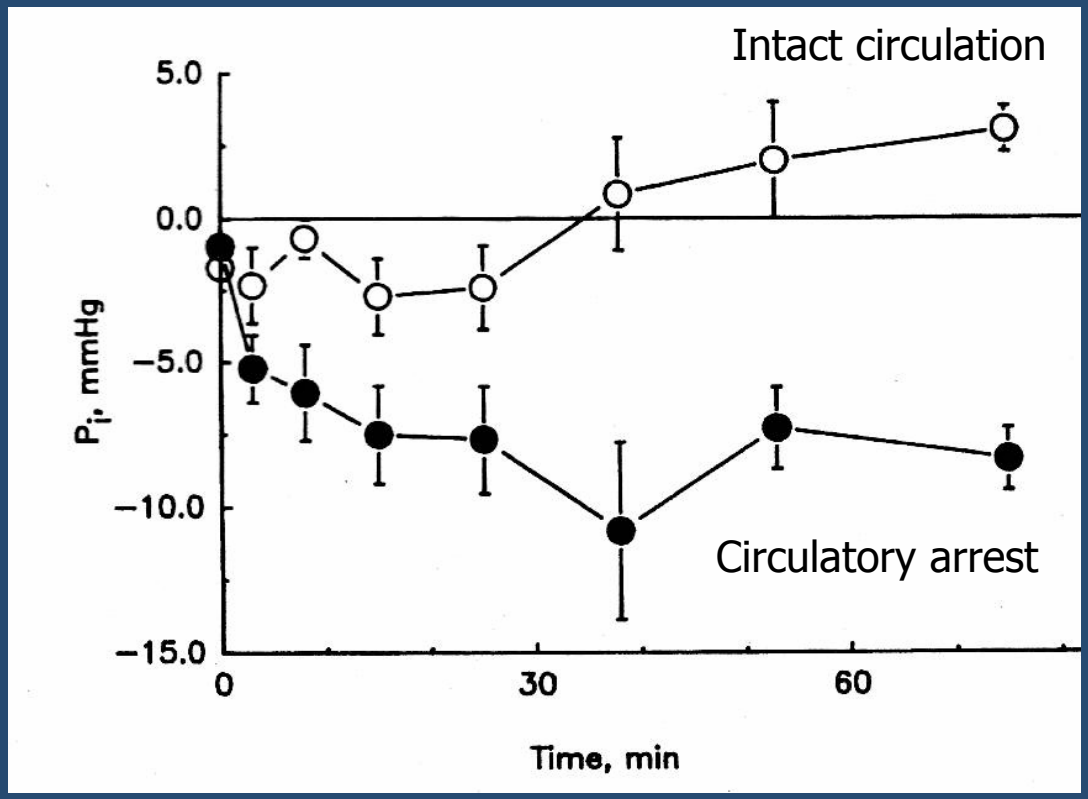
- Interstitial fluid pressure would have to greater than that in the capillary (would capillaries collapse?)
- Interstitial solutes would be held up at the capillary membrane because of the blood brain barrier (BBB). This would increase the concentration of solutes at this location and increase the pericapillary osmotic pressure, which would limit further water absorption (osmotic buffering)

In the brain parenchyma, most likely that osmotic forces control water movement through aquaporins

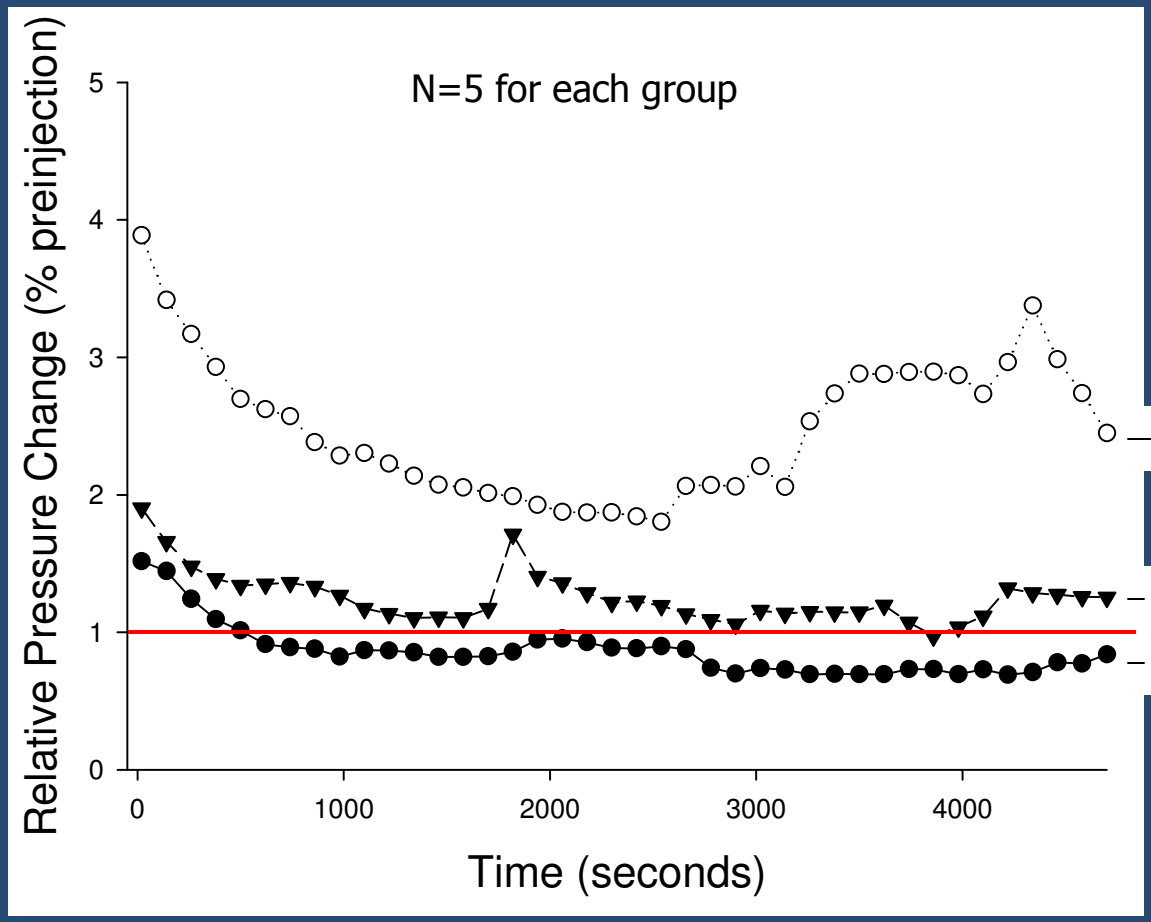


*** A concentration difference of 1.0 mOsm for an impermeable solute generates an equivalent hydrostatic driving pressure of 19.3 mmHg

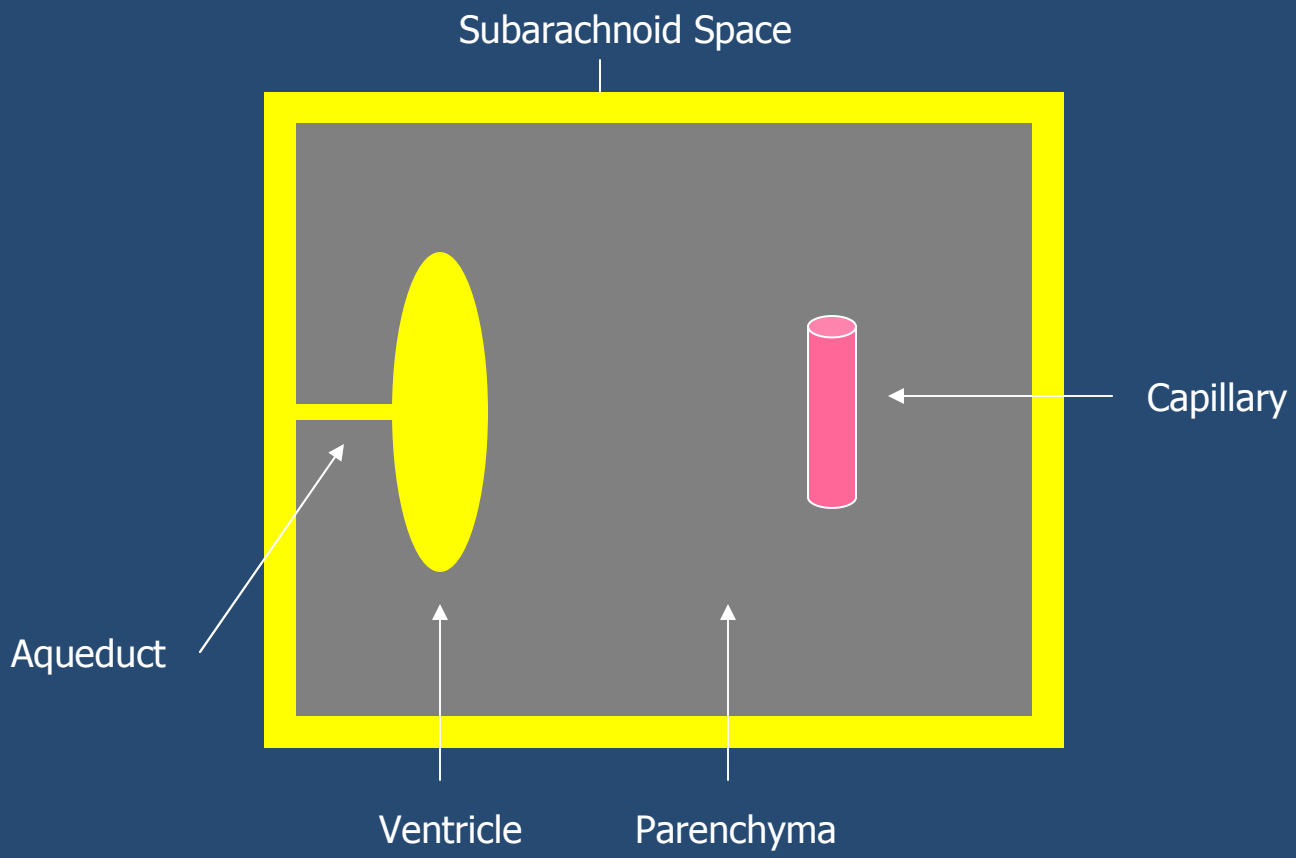
Do we have evidence that parenchymal water removal in the matrix contributes to the drop in tissue pressure ?

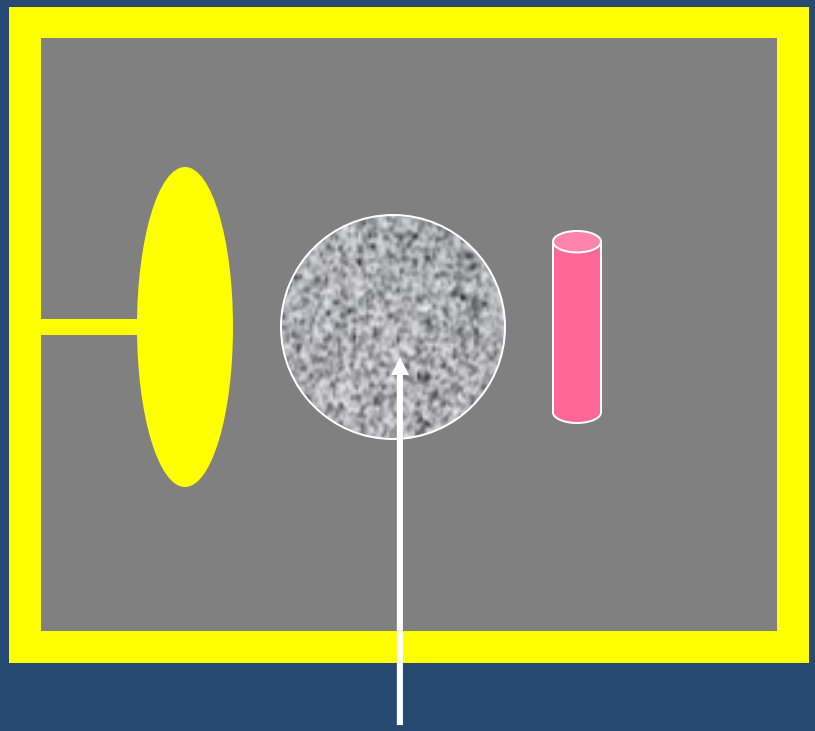


What happens to parenchymal pressures if water movement into or out of the capillaries is prevented ?

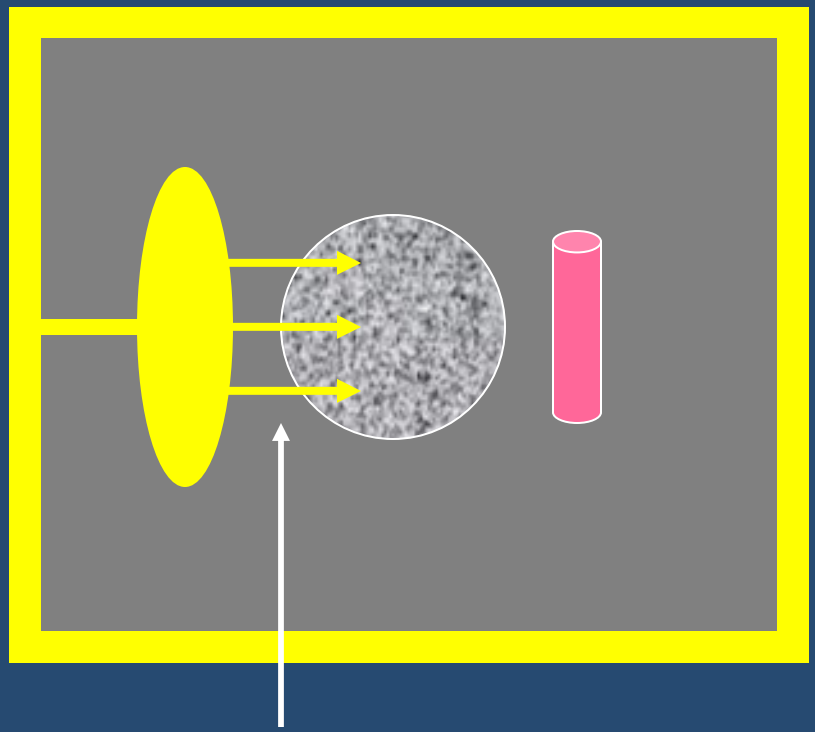


Summary of concept



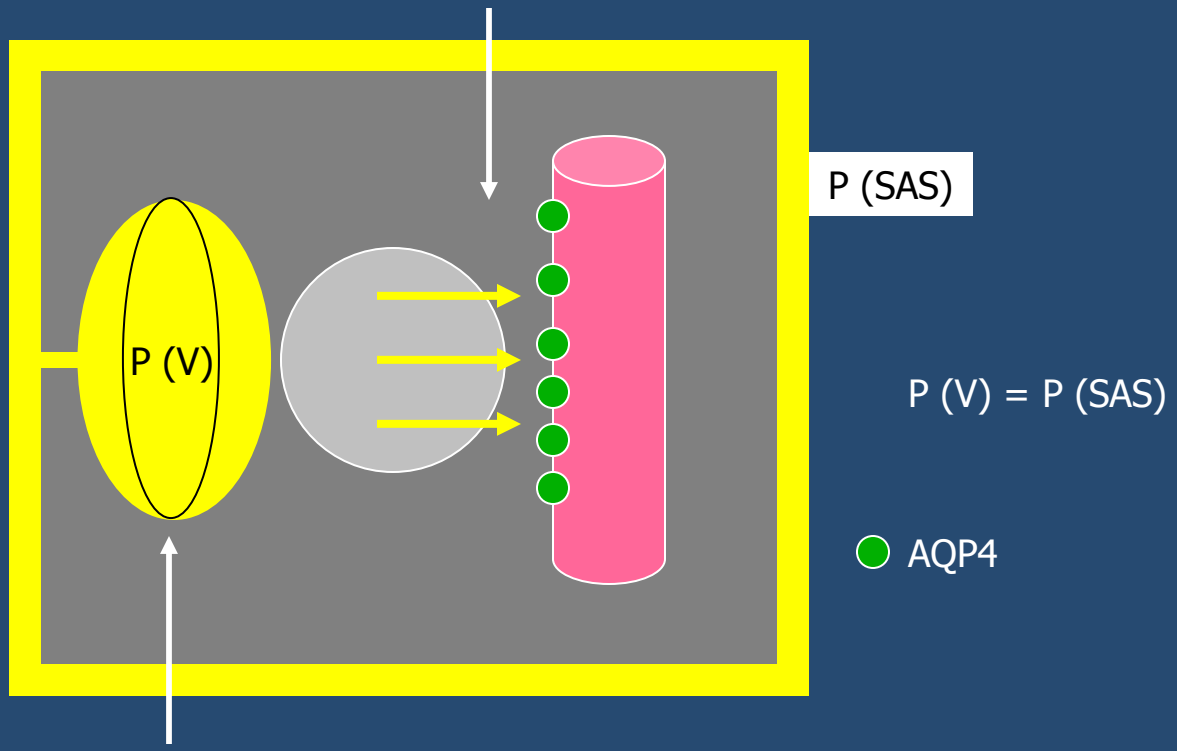


Disruption of Integrin-Matrix interactions
'tends' to lower interstitial fluid pressure



This results in CSF movement from ventricle into parenchymal tissues

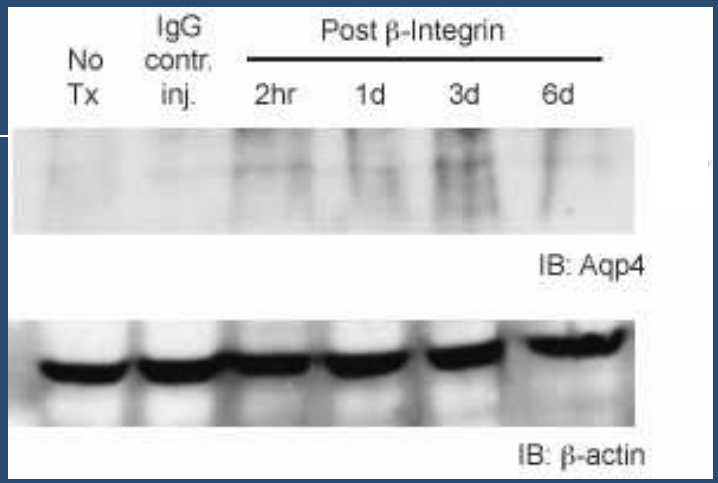
Pressure in the interstitium would increase if not for water movement into the parenchymal capillaries. This movement is controlled by an osmotic imbalance at the capillary membrane.

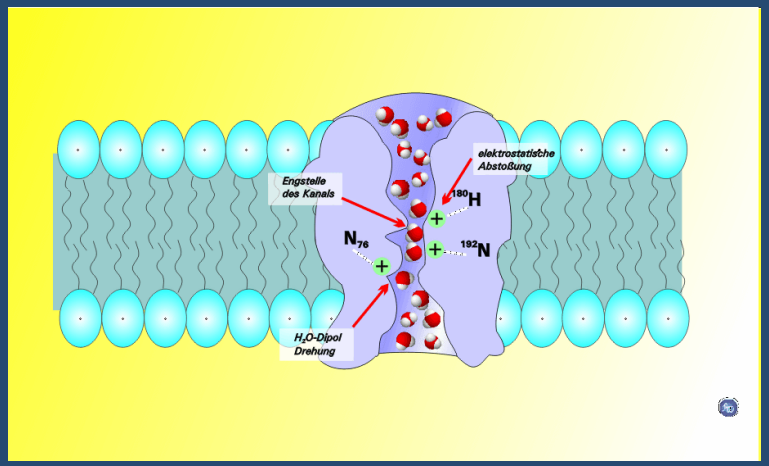


This results in the ventricular pressure being greater than that in the surrounding parenchyma and the ventricle expands

Aquaporins

Rat brain extract after injection of
Anti beta-1 integrin antibodies



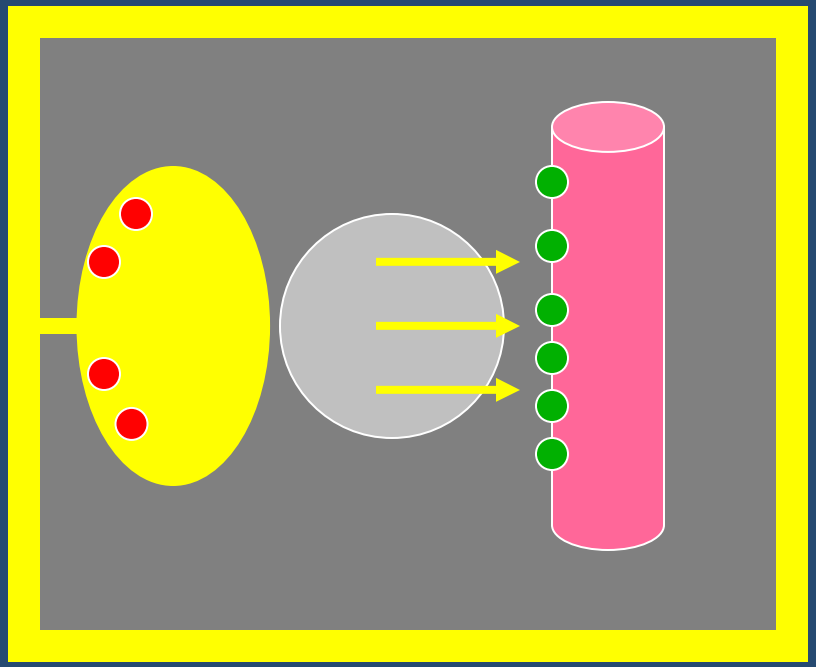


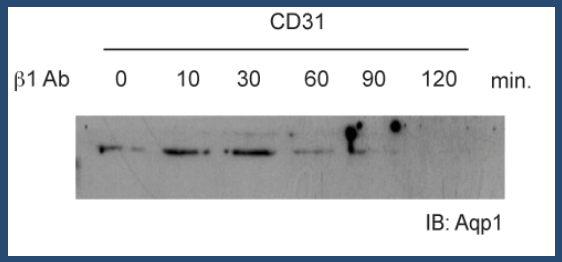
Aquaporin-4 (AQP4) is the predominant water channel in the brain and is expressed on endothelial cells and astrocytic foot processes that surround capillary endothelial cells. These proteins appear to function as bi-directional channels that facilitate water movement driven by hydrostatic or osmotic forces

Questions

- What is the temporal relationship between ventricular expansion and the pressure drop in the interstitium ? The ventricles do not expand immediately even though the pressure gradient exists !!

- Where does the fluid in the expanded ventricle come from? (AQP1 upregulation ● ● and increased CSF formation ?)

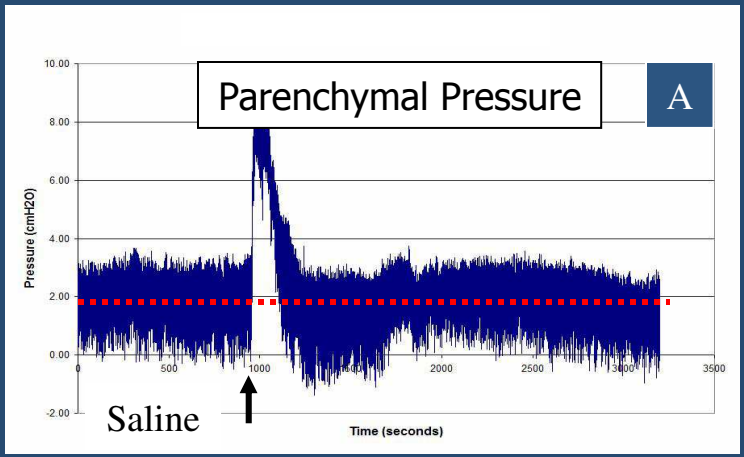


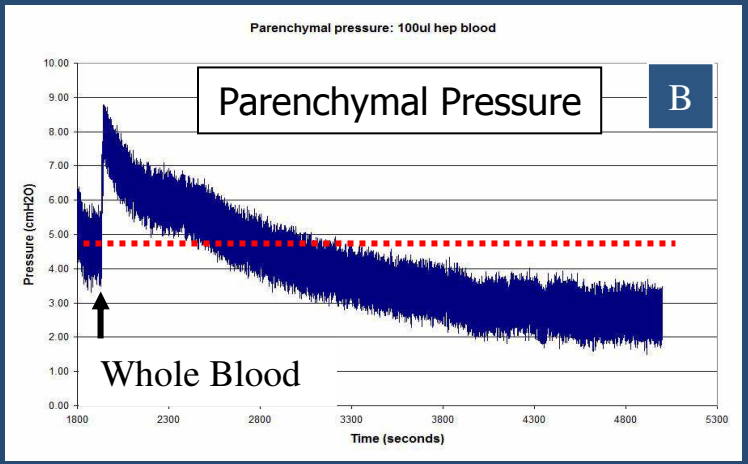


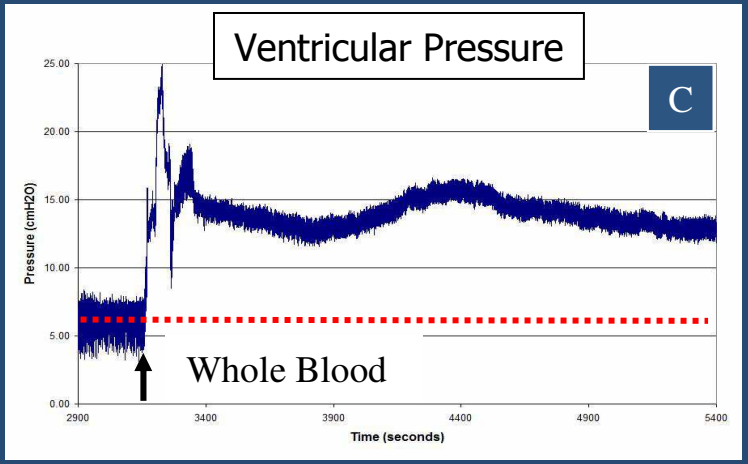
Isolated rat brain vascular endothelial cells

New fluid may have to enter the ventricle from the choroid plexus before ventricles can expand ???

Is the matrix concept relevant to human Hydrocephalus ?







Introduction of whole blood into ventricular system

Can the classical and matrix hypothesis co-exist ?

Inflammation

Cytokines

Lymphatics

CSF absorption deficit

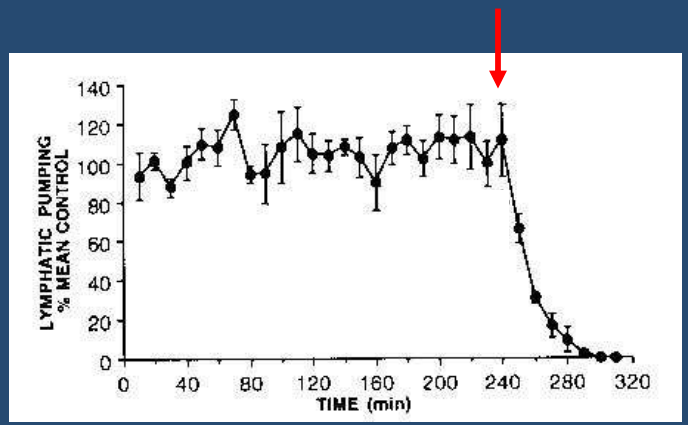
Lymphatics

Beta-1 Integrin – Matrix interactions

??

Hemorrhage

Intra-parenchymal pressure gradients and Hydrocephalus



Importance of matrix concept

Provides a new conceptual foundation for hydrocephalus research - NO obstruction to CSF flow is necessary

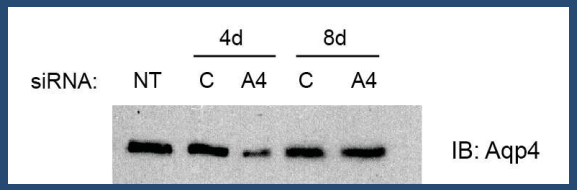
We can 'uncouple' ventricular and interstitial fluid pressure in the brain

Anti-inflammatory drug α-trinositol inhibits lowering of P(i) that was induced by anti β1 integrin antibodies.

Aquaporins can be blocked with drugs or RNAi technology

Therefore, potential may exist for pharmacological strategies to certain forms of hydrocephalus.

A



B AqB013 - inhibits AQP1 and AQP4

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