

Sclérose en plaques et affections inflammatoires du système nerveux central

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Non-infectious inflammatory disorders of the CNS

- Multiple sclerosis
- Neuromyelitis optica and anti-aquaporin 4 antibody syndrome
- Post-infectious acute disseminated encephalomyelitis (ADEM)
- Auto-immune encephalitis, either paraneoplastic or not
- Encephalitis associated with systemic diseases : Sjögren syndrome, lupus erythematosus, Hashimoto encephalitis, hypereosinophilic syndrome
- Bickerstaff encephalitis (anti-GQ1b antibody syndrome)
- Neurosarcoïdosis
- Neuro-Behcet disease
- Primary CNS vasculitis
- Susac syndrome (brain, retina, cochlea)

Serum and CSF analysis

Serum :

- increased levels of immunoglobulins
- ANA, SSA, SSB, ANCA
- anti-TPO and anti-thyroglobulin antibodies
- anti-AQP4 antibody
- anti-GQ1b antibody
- anti-neuronal nucleoproteins antibody : Hu, Ri, Ma2
- anti-CV2, amphiphysin, Yo, GAD 65, NMDAR, AMPAR, LGI1 antibodies...

CSF :

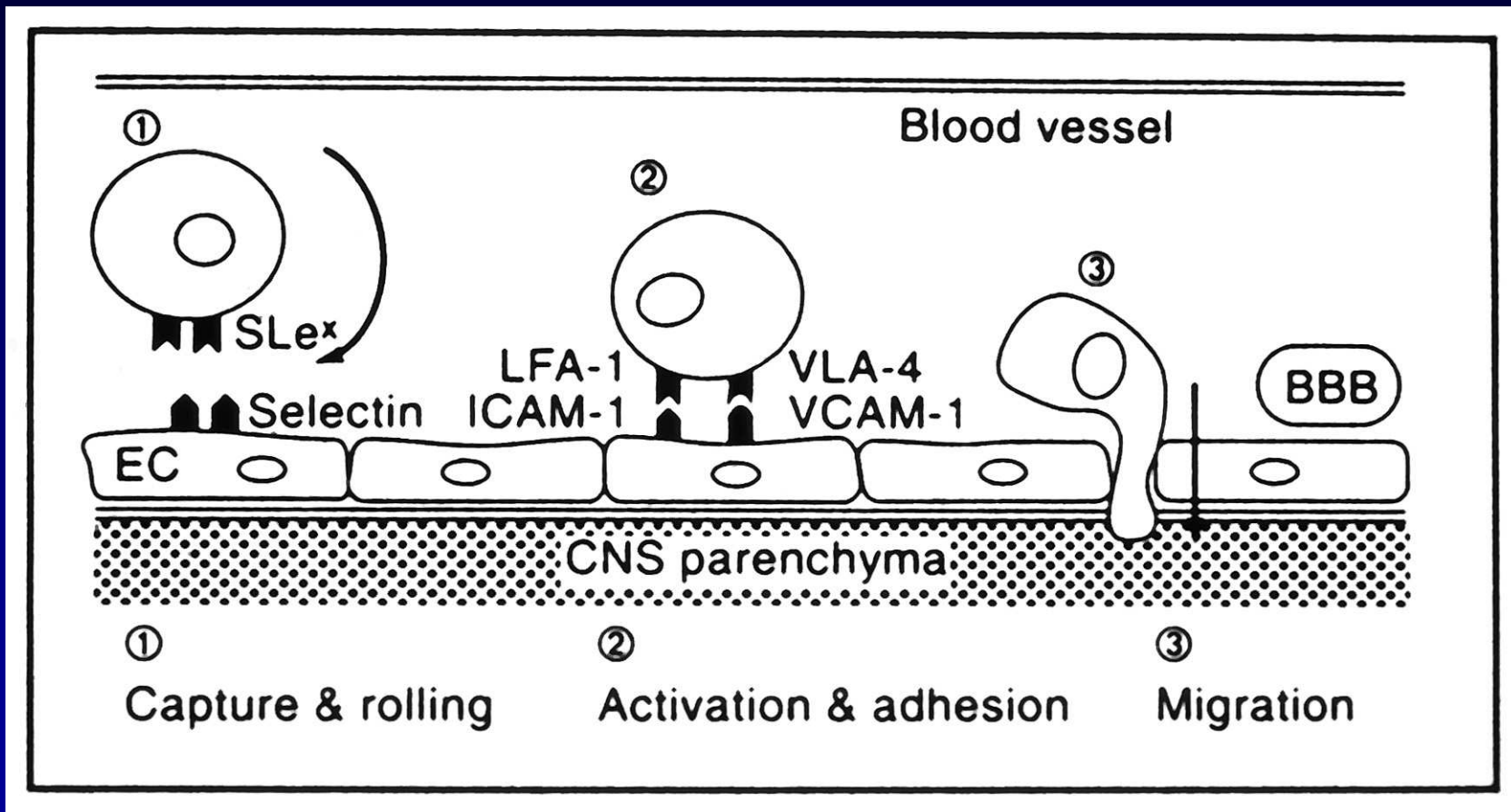
- pleocytosis $> 4/\mu\text{L}$; lymphocytes, plasma cells, eosinophils, neutrophils
- albumin quotient
- CSF-restricted oligoclonal IgG bands
- intra-thecal synthesis of specific antibodies

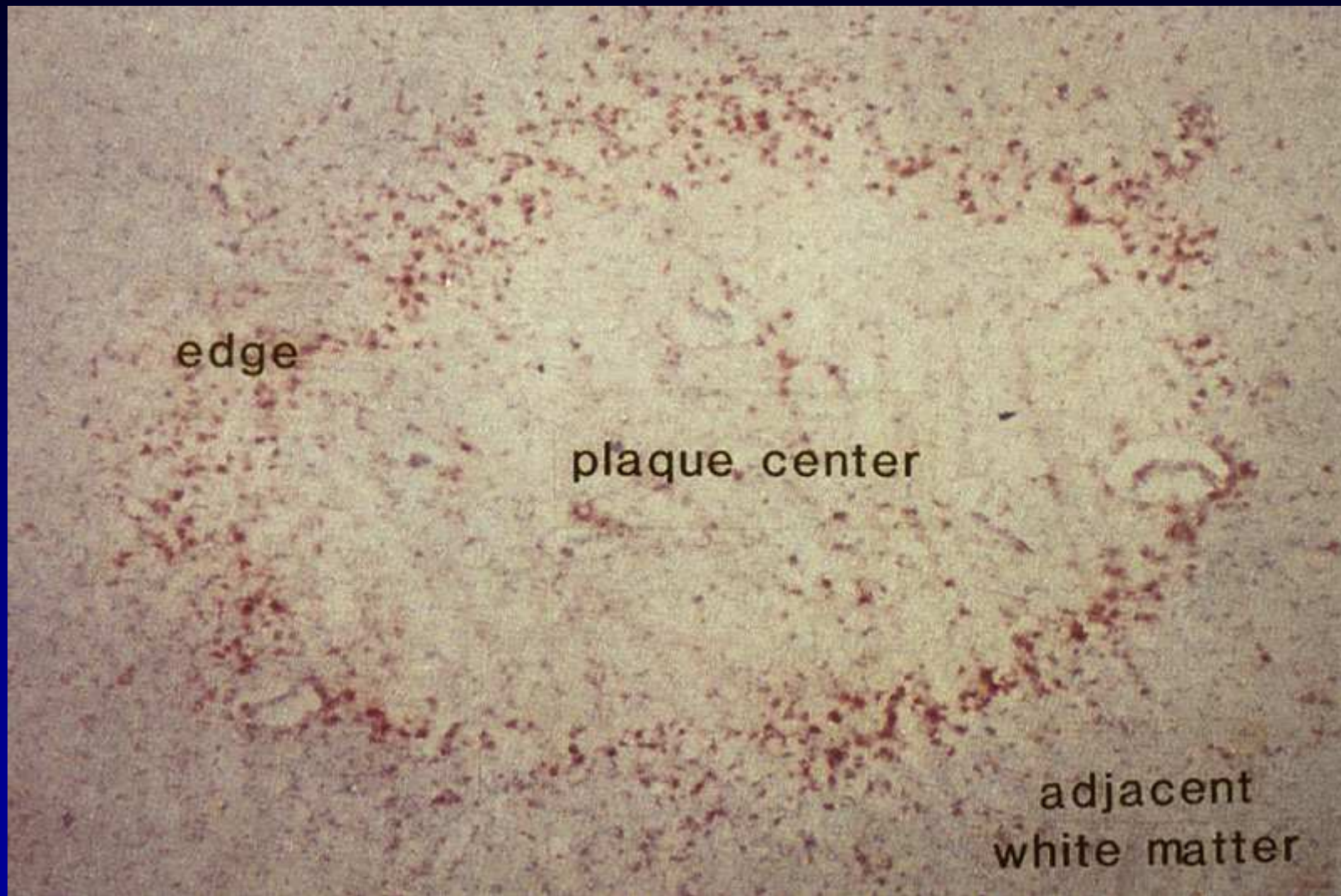
Auto-immune reactions against nervous antigens

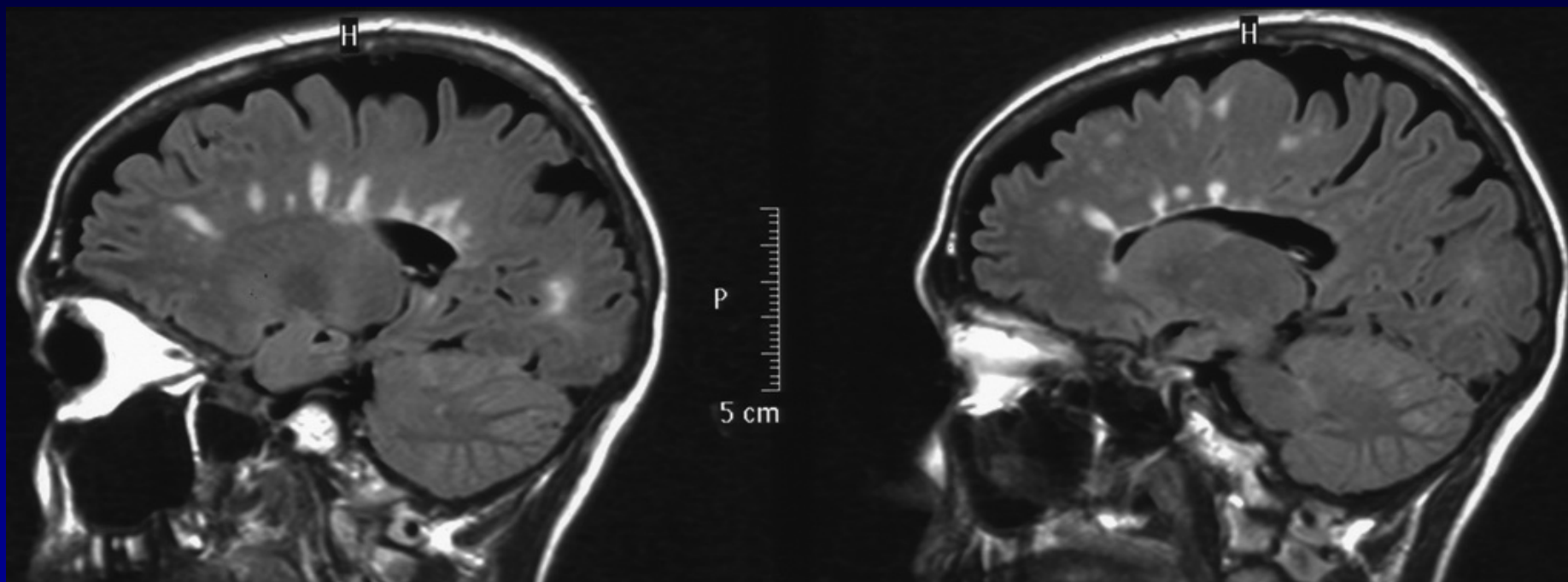
- molecular mimicry (Campylobacter jejuni in GBS and MFS)
- ectopic onco-neural antigens : paraneoplastic diseases
- tolerance breaking in the thymus
- defect in regulatory T cells
- stress-oxydized proteins with neo-epitopes
- ...

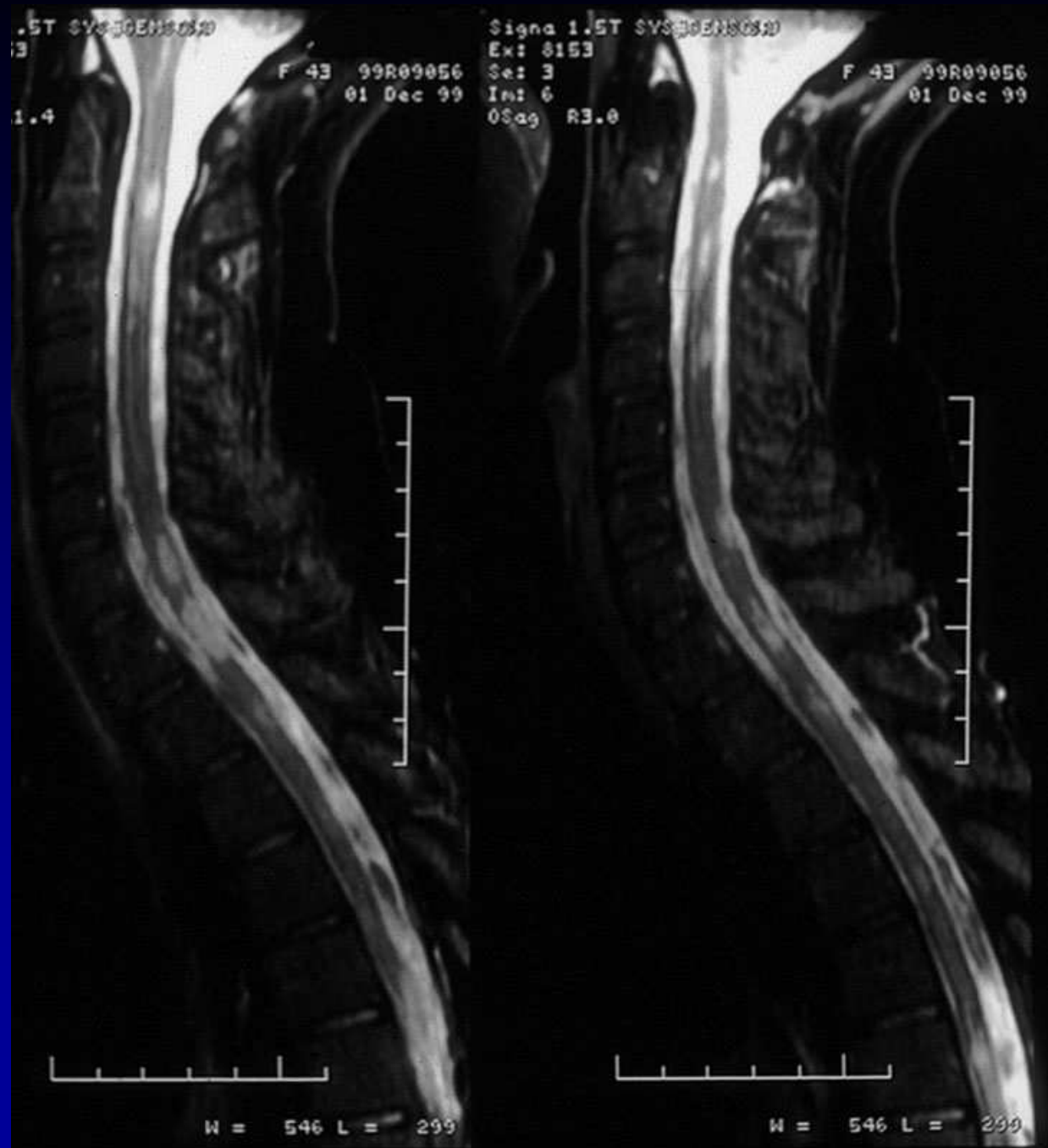
Multiple Sclerosis

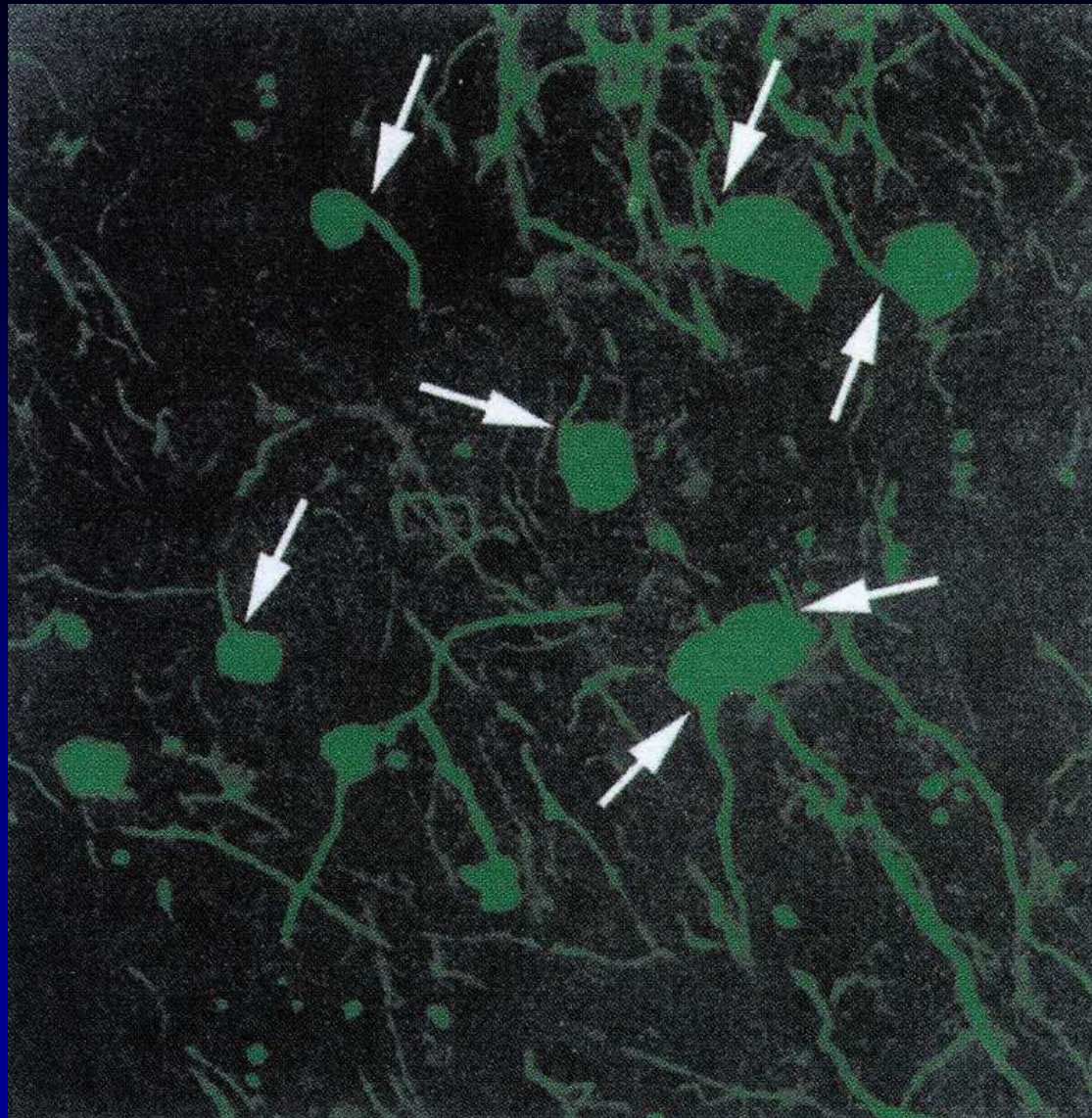
- an inflammatory and neurodegenerative disease of the CNS, associated with immune dysregulation
- a true auto-immune mechanism is not yet demonstrated



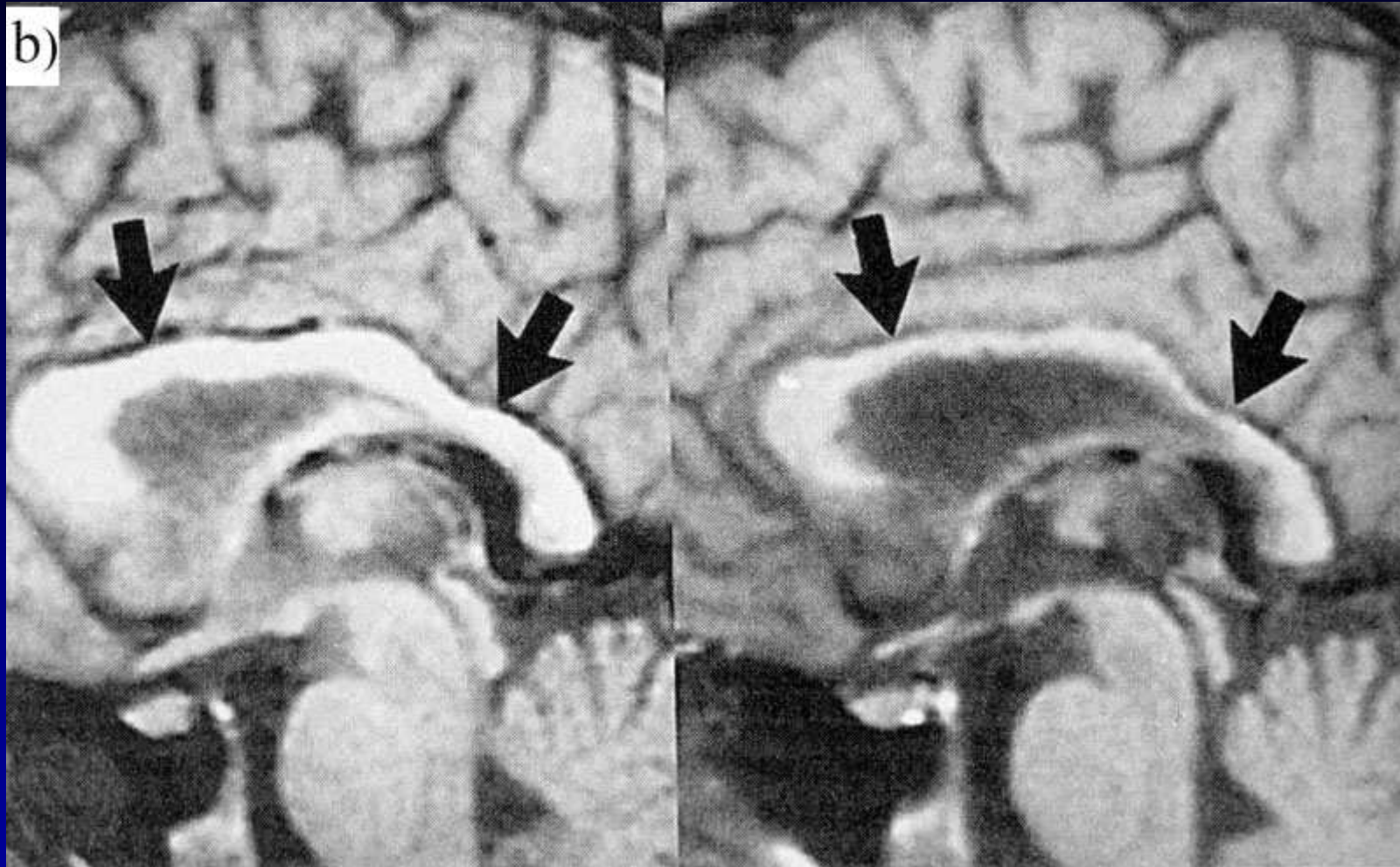


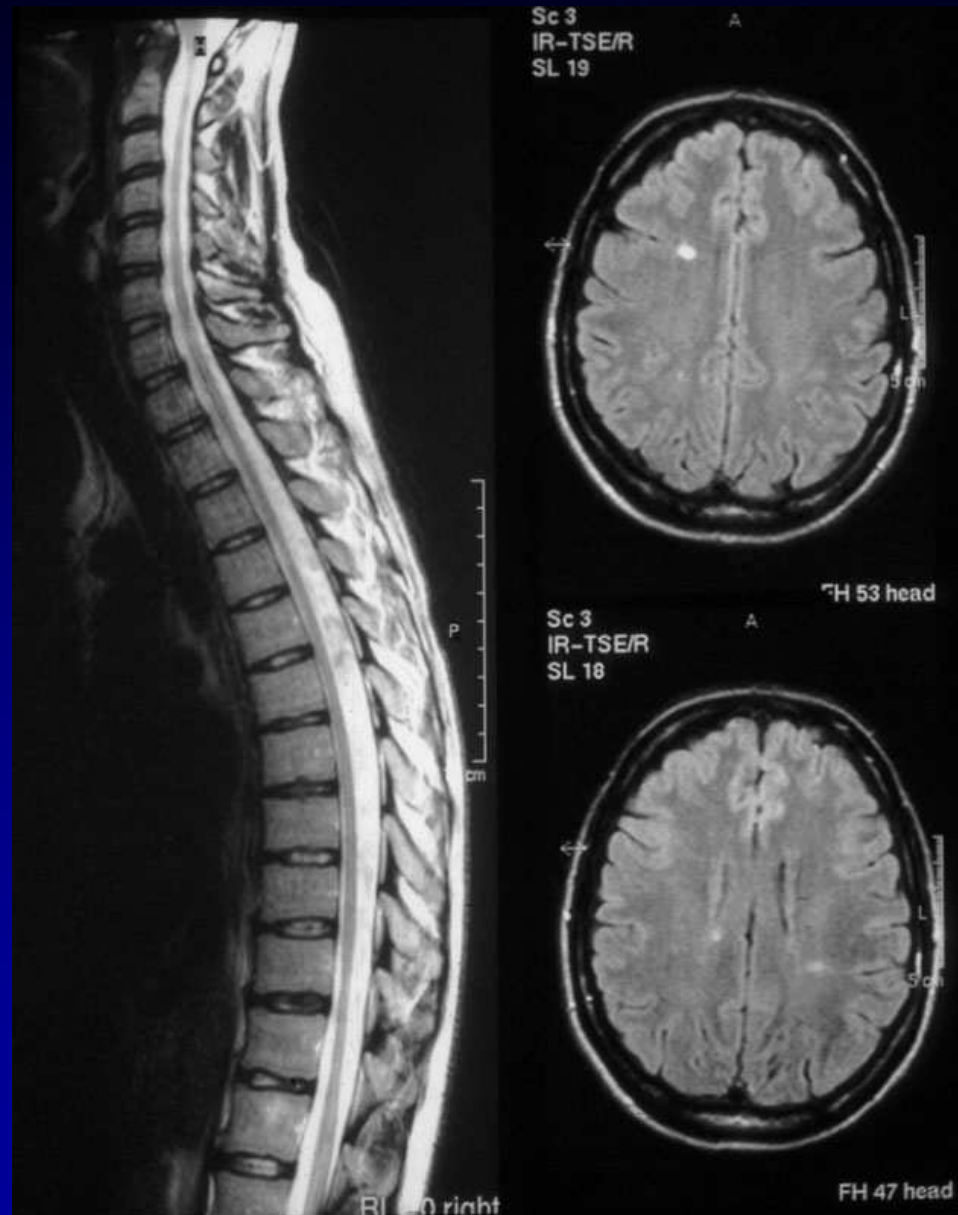


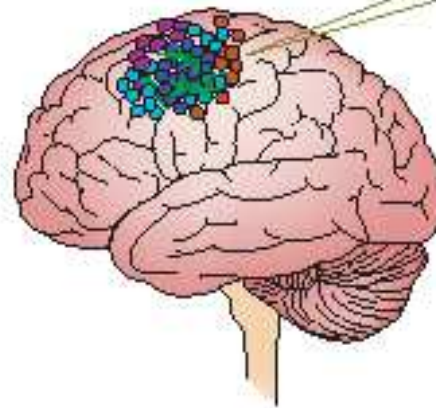
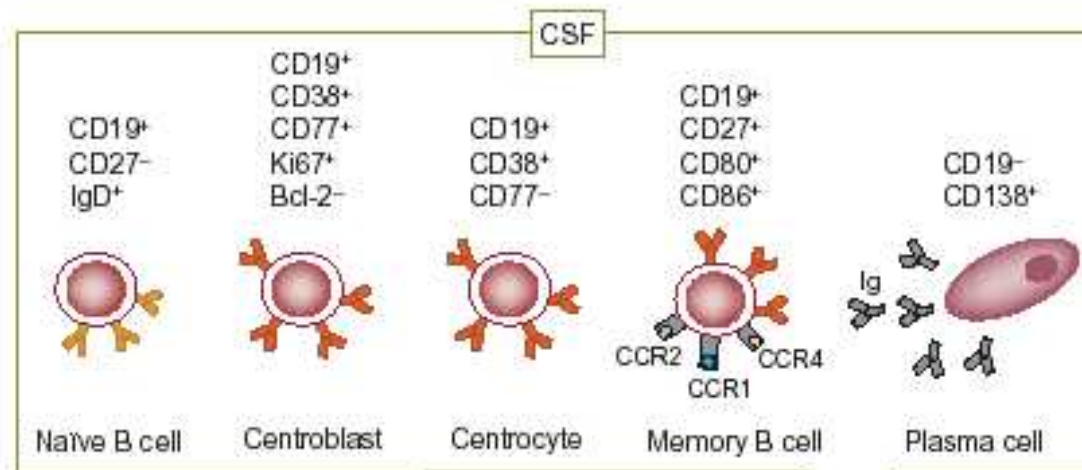


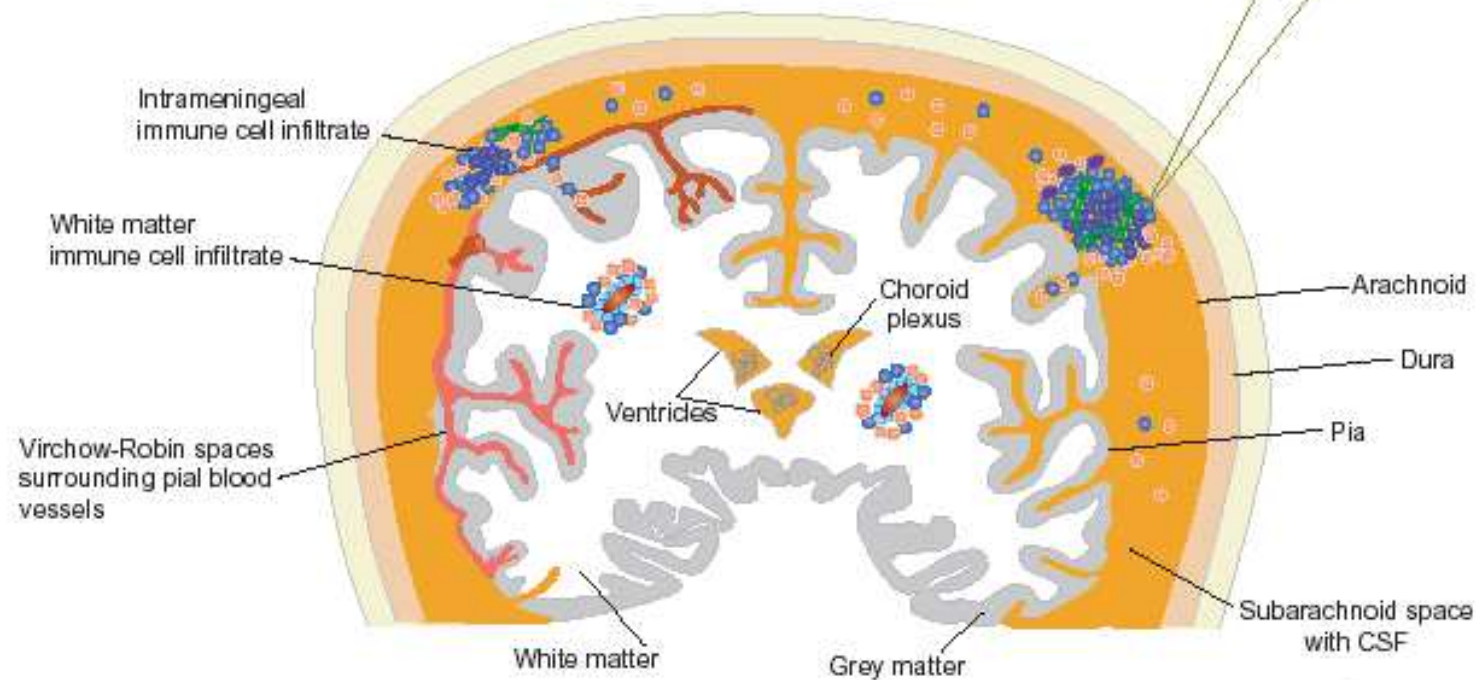
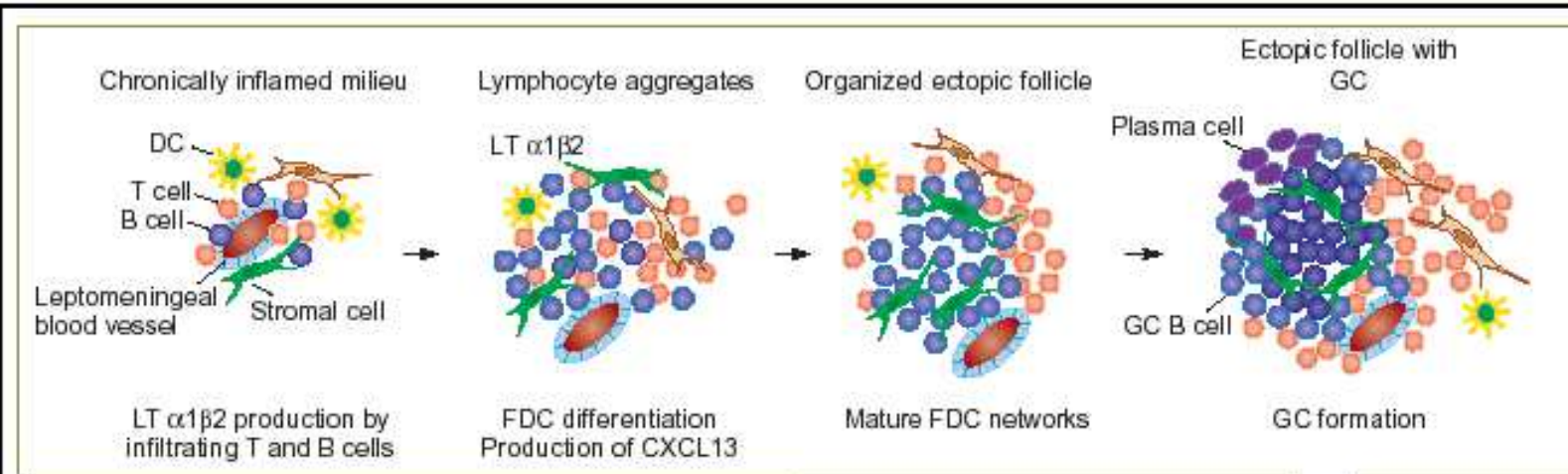


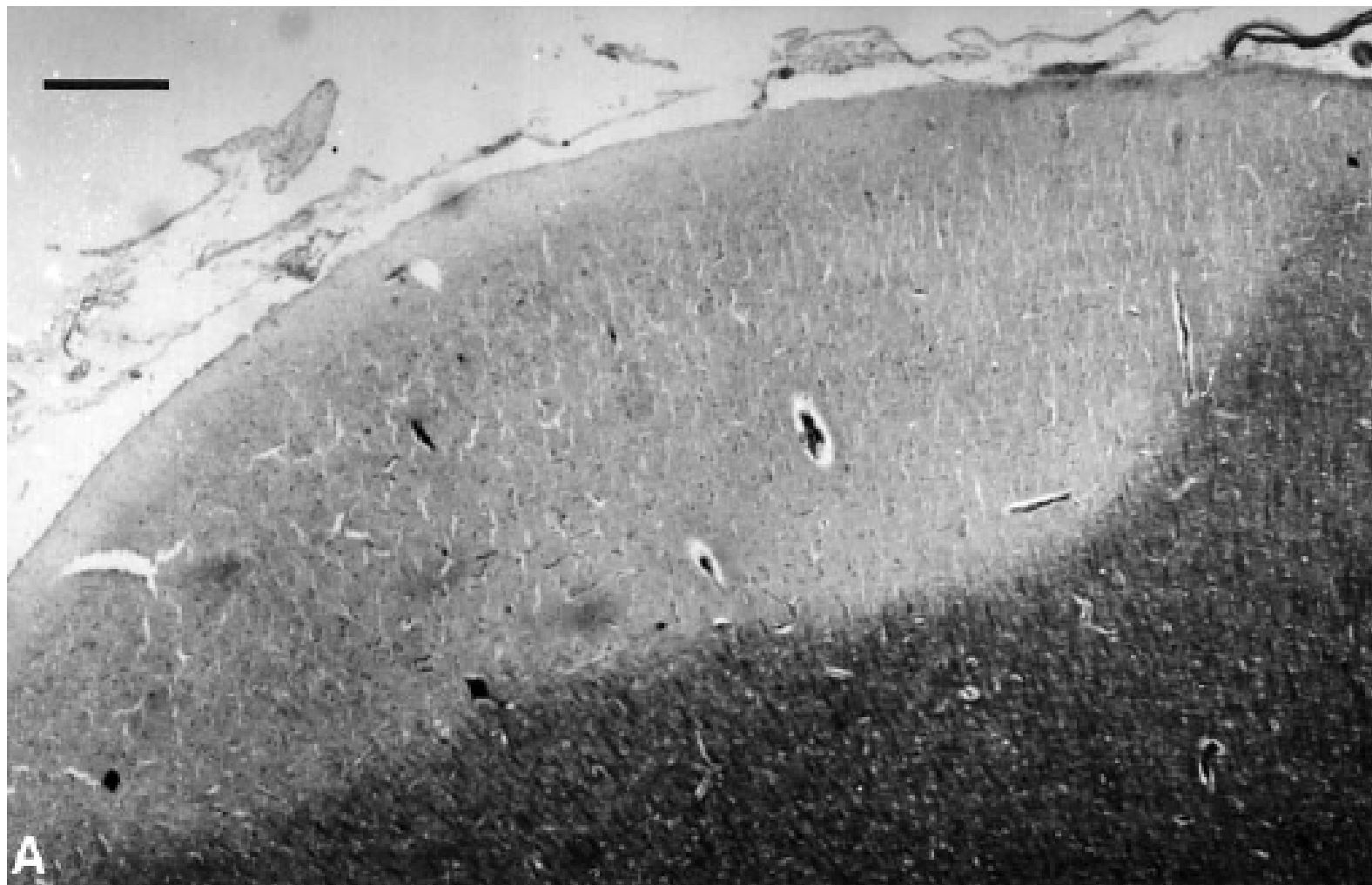
TRAPP et al, N. Engl. J. Med., 398 : 278-285, 1998











Kidd et al, Brain, 1999, 122 : 17–26

Oligoclonal IgG bands

The occurrence of CSF-restricted oligoclonal IgG bands

Inflammatory diseases of the CNS

- multiple sclerosis (95 %)
- paraneoplastic disorders of the CNS (95 %)
- more rarely (< 25 %):
 - neurolupus
 - neurosarcoïdosis
 - NeuroBehcet
 - primary CNS vasculitis
 - neuromyelitis optica

Tumoral diseases

- carcinomatous and lymphomatous meningitis
- primary lymphoma of the CNS

The occurrence of CSF-restricted oligoclonal IgG bands

Infectious CNS diseases

- viral encephalitis (SSPE, herpes simplex, HIV ...)
- viral meningitis due to enveloped viruses (herpes simplex, mumps, varicella zoster, HIV, HTLV I ...)
- late phase of pyogenic meningitis
- tuberculous meningitis
- neurobrucellosis - neuroborreliosis - neurosyphilis
- CNS Whipple's disease
- fungal meningitis (Candida, Cryptococcus, Aspergillus, ...)
- parasitic infections of the CNS (toxoplasmosis, toxocara, neurocysticercosis, schistosomiasis, ...)
- in utero infections with CNS involvement (CMV, rubella, toxoplasmosis)

CSF-restricted oligoclonal IgG bands are not observed :

- in acute disseminated encephalomyelitis (ADEM)
- in Guillain-Barré syndrome and in CIDP
- in vascular, toxic, metabolic, traumatic or psychiatric disorders
- in Susac syndrome
- in neurodegenerative diseases (Parkinson, Alzheimer, ALS)
- in mechanical radicular syndromes
- in other peripheral neuropathies

**The presence of CSF-restricted
oligoclonal IgG bands is not always
associated with a quantitatively
detectable intrathecal IgG synthesis**

Diagnostic biomarkers

In a study of 415 CIS patients (*Tintore et al, Neurology, 2008, 70 : 1079-1083*)

- **the presence of CSF-restricted oligoclonal IgG bands doubles the risk for having a second attack after a mean follow-up of 50 months, independently of baseline MRI**
- **only 4% of patients with normal MRI and no oligoclonal bands developed CDMS; but 23% did so in presence of oligoclonal bands in spite of normal MRI**
- **of the total cohort, oligoclonal bands were positive in 61% of the patients : in 31% with 0 Barkhof criteria, 69% with 1 or 2 Barkhof criteria, and 85% with 3 or 4 Barkhof criteria**

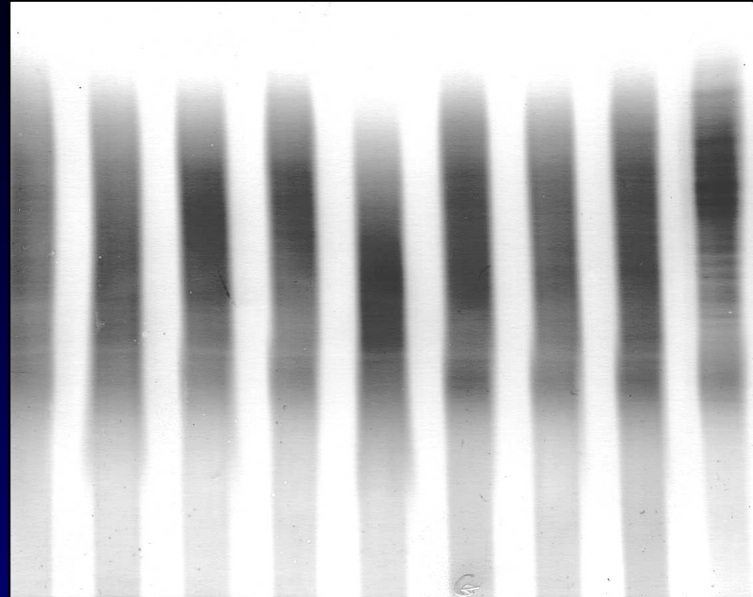
Oligoclonal free kappa chains

Oligoclonal free kappa chains

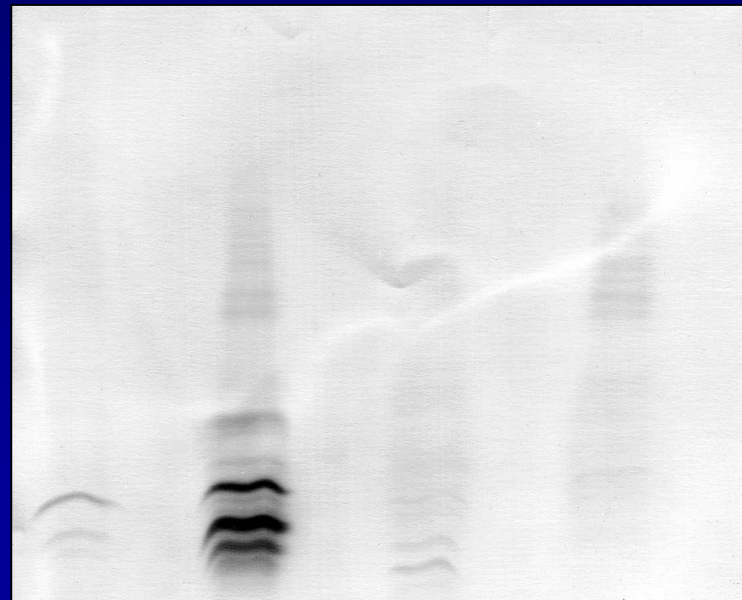
In a series of 33 patients with clinical signs and symptoms suggestive of MS, but without CSF oligoclonal IgG,

- 18 (54 %) contained oligoclonal free kappa bands
- all patients with a positive MRI according to the 4 Barkhof 's criteria (N = 6) had CSF free kappa bands
- Chi squares (positive or suggestive MRI versus aspecific or normal MRI; positive versus negative free kappa bands) with Yate 's correction : $p = 0.023$

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8.5
8.0
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pH 9.0
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Goffette et al, JNNP, 2004, 75 : 308-310

Results of initial MRI and patterns of CSF analysis

Patients N = 33	Positive MRI *	MS-suggestive MRI **	Aspecific of Normal MRI
IgG (-)/k(+) N = 15	5	6	4
IgG (1 bande)/k(+) N = 3	1	1	1 ***
IgG (1bande)/k (-) N = 1	0	0	1
IgG (-)/k (-) N = 14	0	4	10 ***
TOTAL	6	11	16

* According to the strict criteria of Barkhof et al (1997) (one spinal cord lesion can substitute for one brain lesion)

** Two or more brain lesions suggestive of MS either in the brain or in the spinal cord

*** one case with only one spinal cord lesion

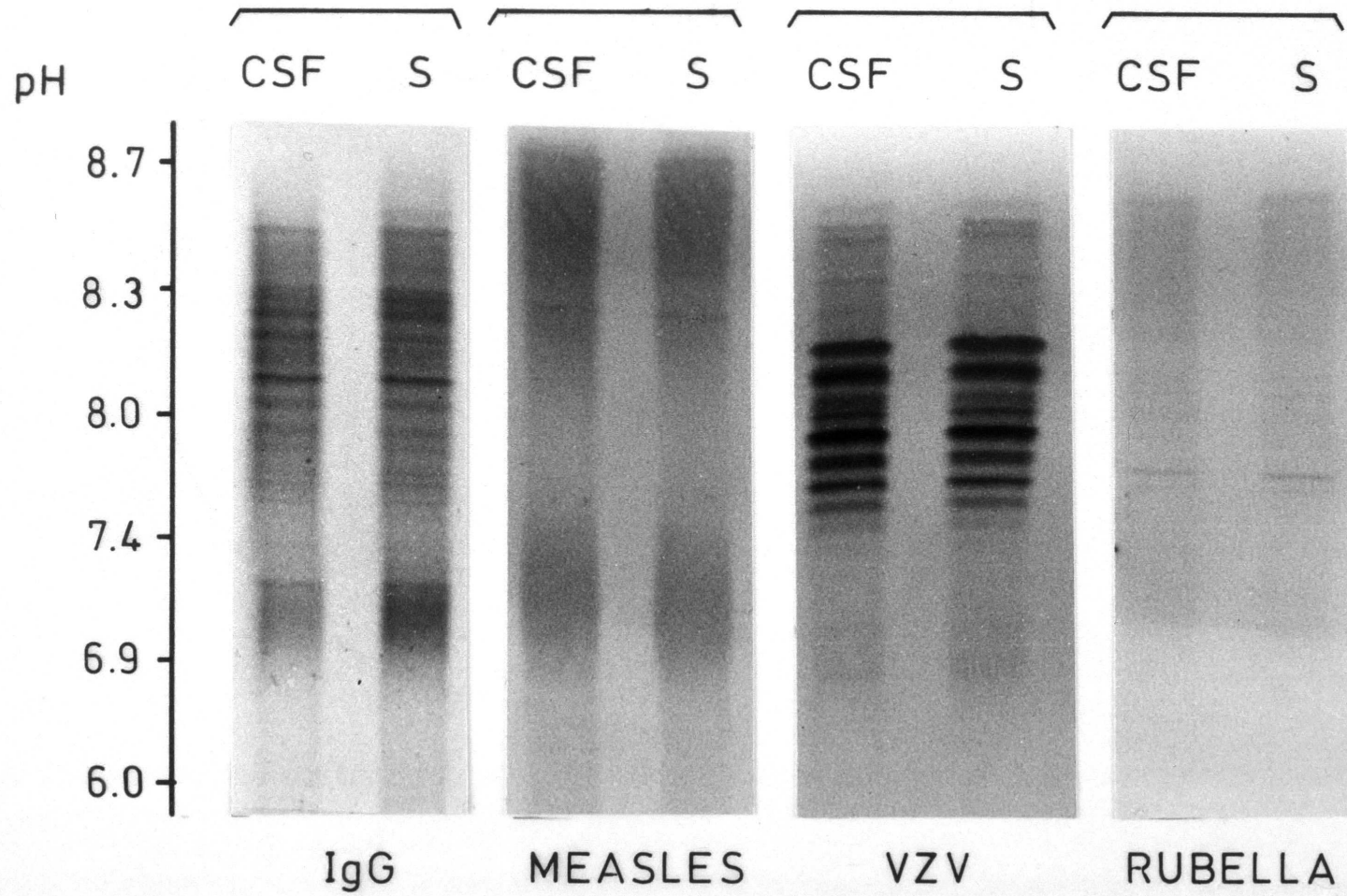
The mirror pattern

- is characterized by the presence of identical IgG bands located at the same isoelectric points and similarly immunostained in both CSF and serum
- indicates a systemic immune activation without intrathecal synthesis
- results from the passive transudation of oligoclonal IgG from blood to CSF through a normal **or** an impaired blood-CSF barrier

The mixed pattern

- is characterized by the simultaneous presence of a mirror pattern and a local synthesis of some IgG bands
- is mainly observed in systemic inflammation or in systemic infections with CNS involvement but also in some MS cases

GUILLAIN-BARRE SYNDROME



Revised diagnostic criteria for neuromyelitis optica (NMO)

Wingerchuk et al, Neurology, 2006

**Optic neuritis AND acute transverse myelitis
+ 2 criteria out 3 :**

- spinal cord MRI : lesion over more than 3 contiguous vertebral segments (longitudinally extensive transverse myelitis)
- brain MRI : lesions not meeting criteria for MS
- presence of anti-AQP4 IgG antibodies

Neuromyelitis optica (Devic disease) (1)

Onset :

- occurrence of both optic neuritis (ON) and myelitis within a few days/weeks (10 – 15 %)
 - ON followed by myelitis during the follow-up (50 %)
 - myelitis followed by ON during the follow-up (35-40 %)
-
- the first two index attacks of ON and myelitis may be separated by an interval of several months or years (median : 1 year)
 - relapsing ON or relapsing myelitis may occur before the appearance of an acute myelitis, or an acute ON, respectively
 - the presence of anti-aquaporin-4 antibodies predicts relapse and conversion to NMO



Fig 1A. T2 cervical spinal cord.



Fig 1B. Coronal T1 optic chiasm

RATIO NMO/MS

- **1 to 300 in France**
- **1 to 30 in Cuba**
- **1 to 7.5 in French West Indies**

Aquaporin 4 (AQP4)

- the predominant CNS water channel
- highly concentrated in the astrocytic foot processes abutting microvessels and pia, and in ependymal cells
- also highly expressed in the hypothalamus and periventricular regions, including the circumventricular organs and the area postrema (which lack a blood-brain barrier)

Dosage

- in the serum
- immunofluorescence assay :
 - tissue based / cell based
 - linear pattern in cerebellum and midbrain
 - HEK cells (human embryonic kidney cell line) : transfected cells expressing a transgene encoding full-length AQP4

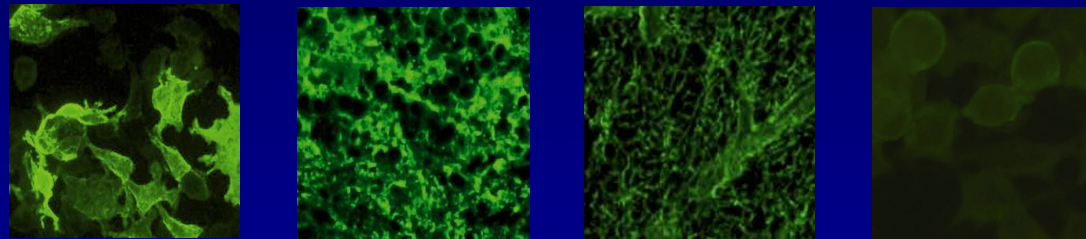


Figure 7 : Indirect immunofluorescence of the AQP 4 (Biognost)

- 1. Marker the transfected cells with AQP4 (HEK 293)*
- 2. Marker of the cerebellum (along the Virchow Robin space and the arterioles of the white matter)*
- 3. Marker of the optic nerve (linear)*
- 4. Non transfected cells : negative immunofluorescence*

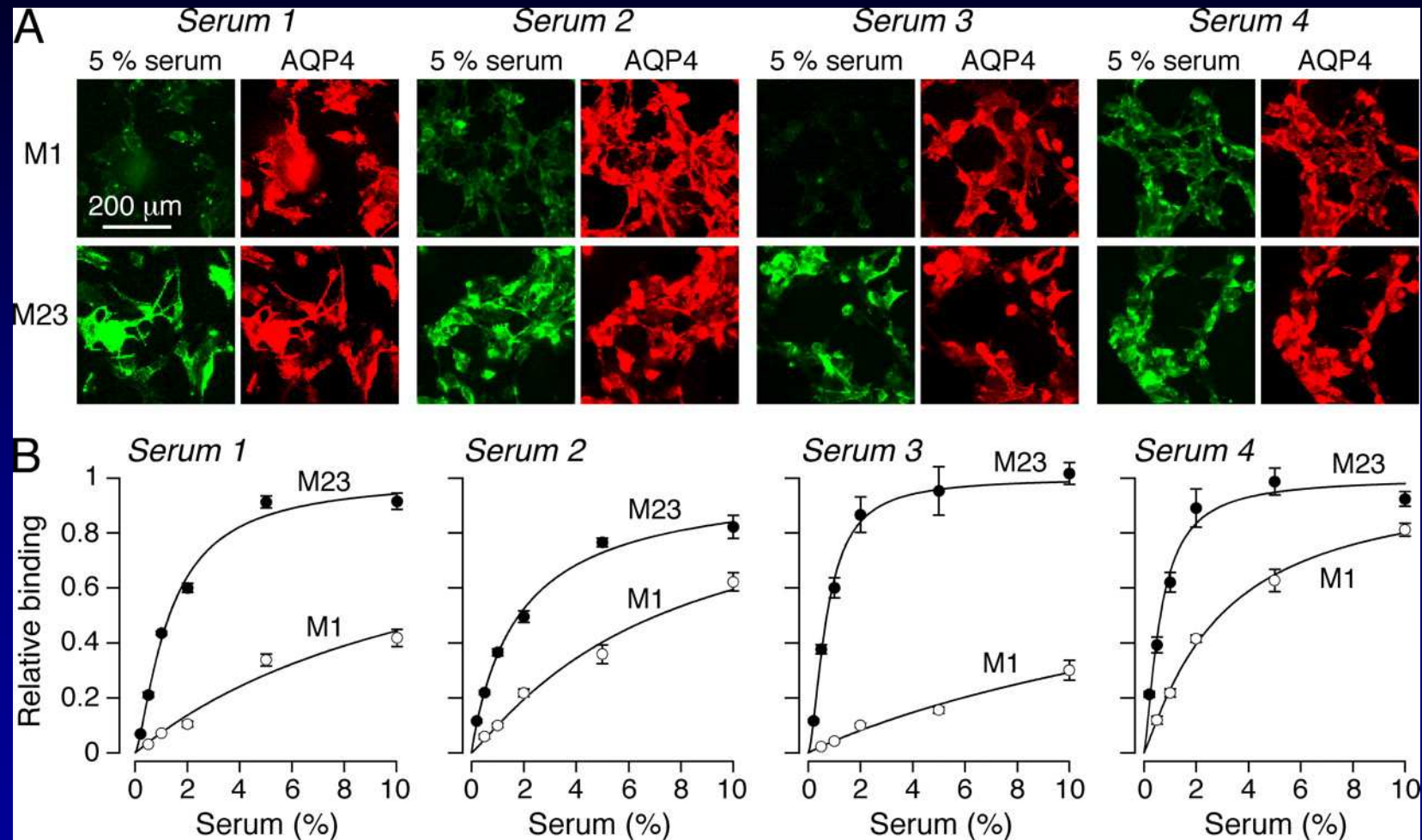
The anti-AQP4 antibody assay

- higher sensitivity than the indirect immunofluorescence test on mouse brain tissue (NMO-IgG)
- titers higher in cases with permanent complete blindness or with extended spinal cord lesions
- titers became lower after high-dose intravenous methylprednisolone
- titers remained low in relapse-free periods under immunosuppressive therapy
- titers are consistently higher in serum than in CSF
- an intrathecal synthesis of anti-AQP4 antibodies has been rarely reported

Aquaporin 4 (AQP4)

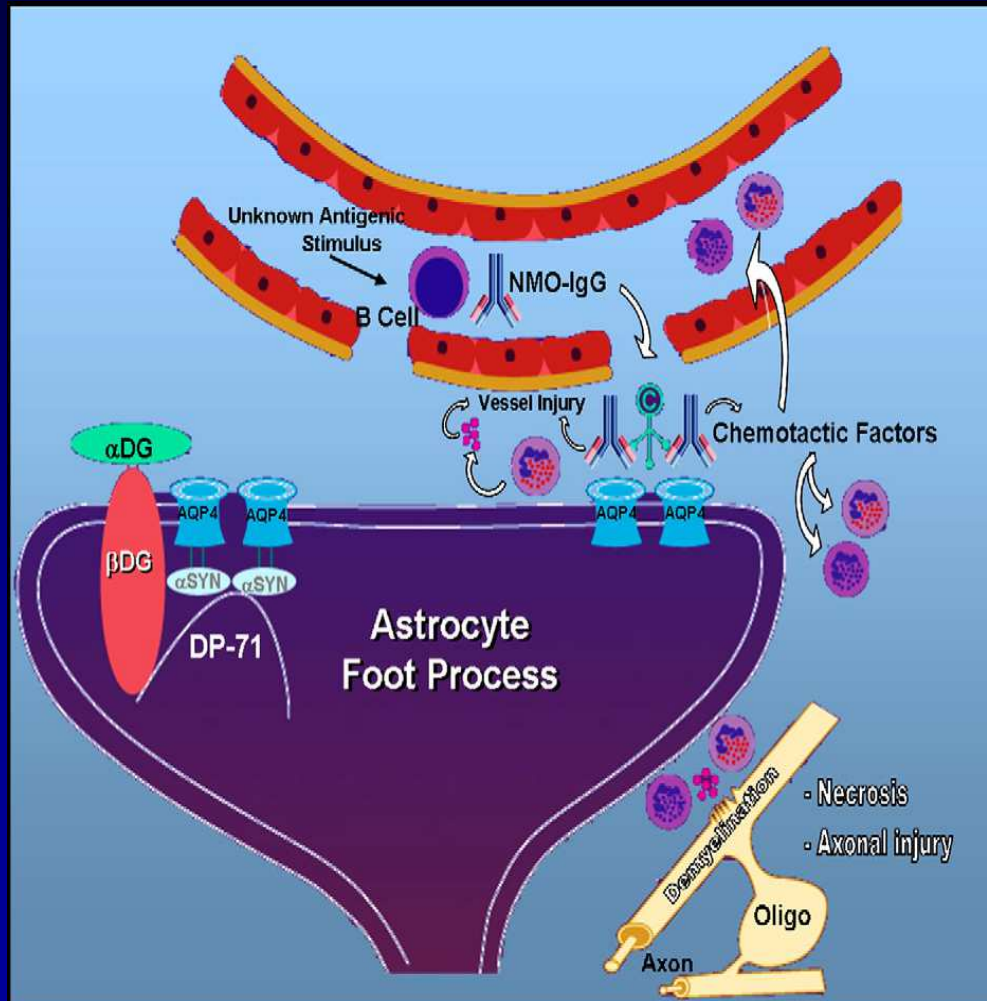
- **two isoforms :**
 - **the full length molecule : M1 (32 kDa, 323 residues)**
 - **a smaller isoform : M23 (30 kDa, 301 residues)**
- **M23 forms orthogonal arrays of particles due to tetramer-tetramer interactions**
- **M1 does not form such arrays but can co-assemble with M23 in hetero-tetramers**
- **NMO-IgG antibodies have a higher affinity for the M-23 AQP4 isoform**
- **NMO-IgM antibodies have lower affinity, lower specificity, lower sensitivity**
- **currently, the most widely used assay for NMO-IgG antibodies is performed in M1 AQP4-expressing HEK 293 cells**

Differential binding of NMO-IgG in NMO patient serum to M1 versus M23 AQP4.



Crane J M et al. *J. Biol. Chem.* 2011;286:16516-16524

Pathogenesis



- antibody-induced inflammatory reaction complement-dependent
- Activated macrophages + eosino/neutrophils (IL-8 and 17) and cytolytic cascade resulting in irreversible damage to target cell membranes
- Cytokines, proteases, oxygen/nitrogen free radicals

Vascular and parenchymal damage

Secondary demyelination

Post-infectious inflammatory neurological disorders

A] involving the central nervous system

ADEM : acute disseminated encephalomyelitis

- relapsing forms ?
- site-specific variants *
 - encephalitis (20 %), including cerebellitis and rhombencephalitis
 - myelitis (23 %)
 - encephalomyelitis (13 %)
 - encephalomyeloradiculoneuritis (27 %)
 - myeloradiculitis (17 %)

* Percentages from Marchioni et al, Neurology, 2005, 65 : 1057-1065

Post-infectious encephalomyelitis (acute disseminated encephalomyelitis, ADEM)

Triggered by :

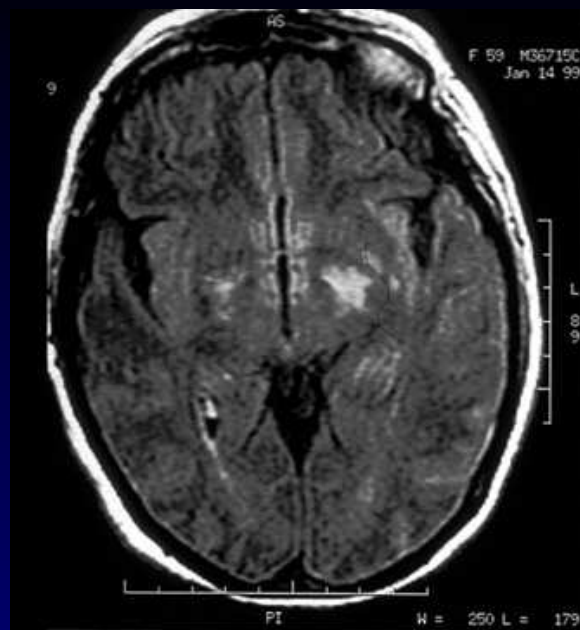
- measles, rubella, mumps viruses: very rarely now because of generalized vaccination
- varicella zoster (cerebellitis)
- respiratory syncytial virus or other respiratory viruses
- adenoviruses
- Epstein-Barr virus
- *Mycoplasma pneumoniae*

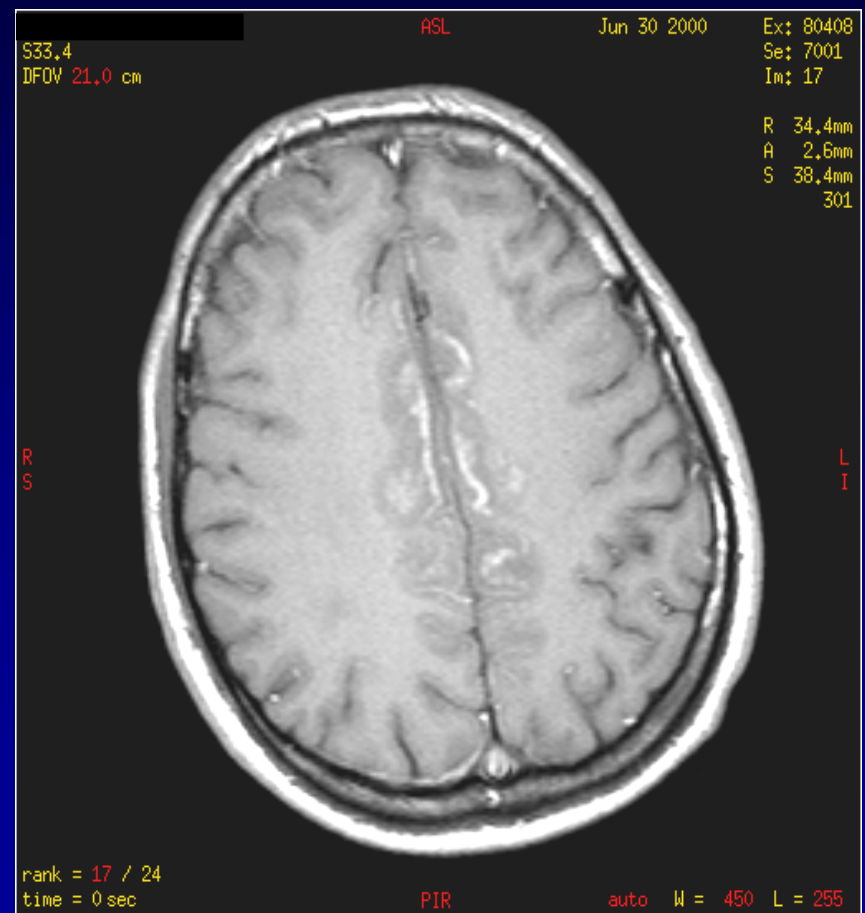
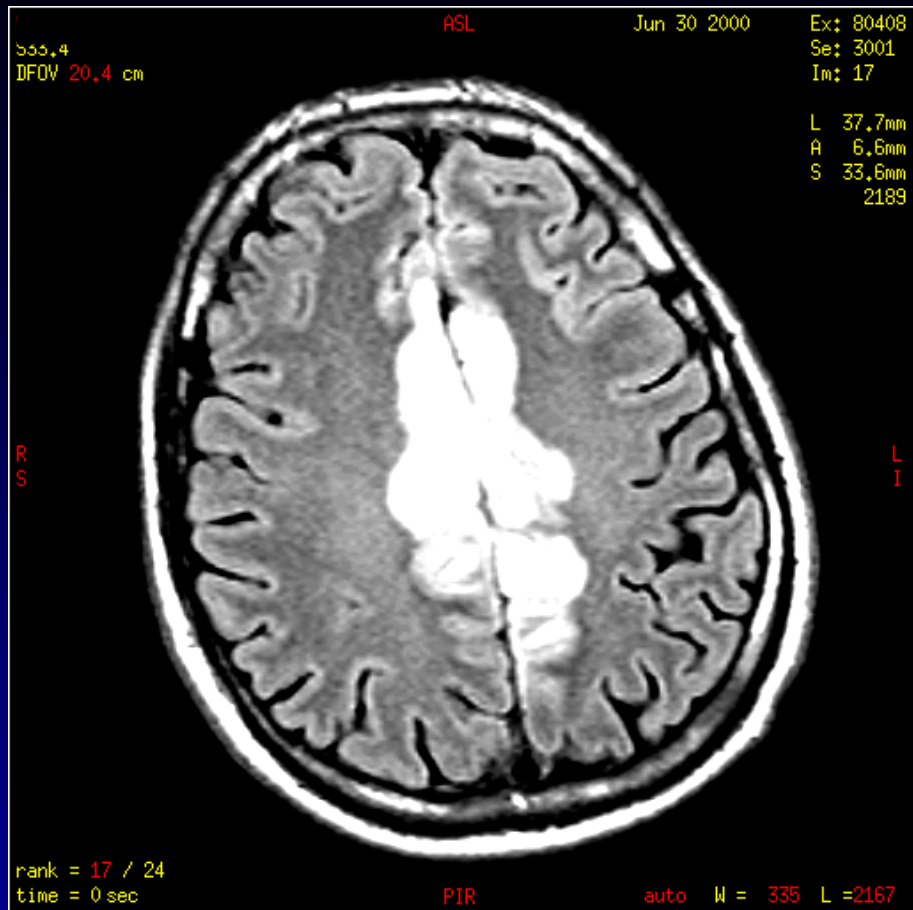
ADEM

- **mainly observed in children, adolescents, young adults**
- **acute or subacute; the hyperacute form is hemorrhagic (Hurst)**
- **sometimes restricted to the spinal cord or the cerebellum**
- **basal ganglia often involved**
- **all lesions are Gd+**
- **drowsiness, stupor, seizures, focal deficits**
- **early therapy with corticosteroids !**

The CSF in ADEM

- slight pleocytosis in most cases
- slight increase of the CSF protein content
- all PCR negative
- no oligoclonal bands, no intrathecal synthesis of antibodies





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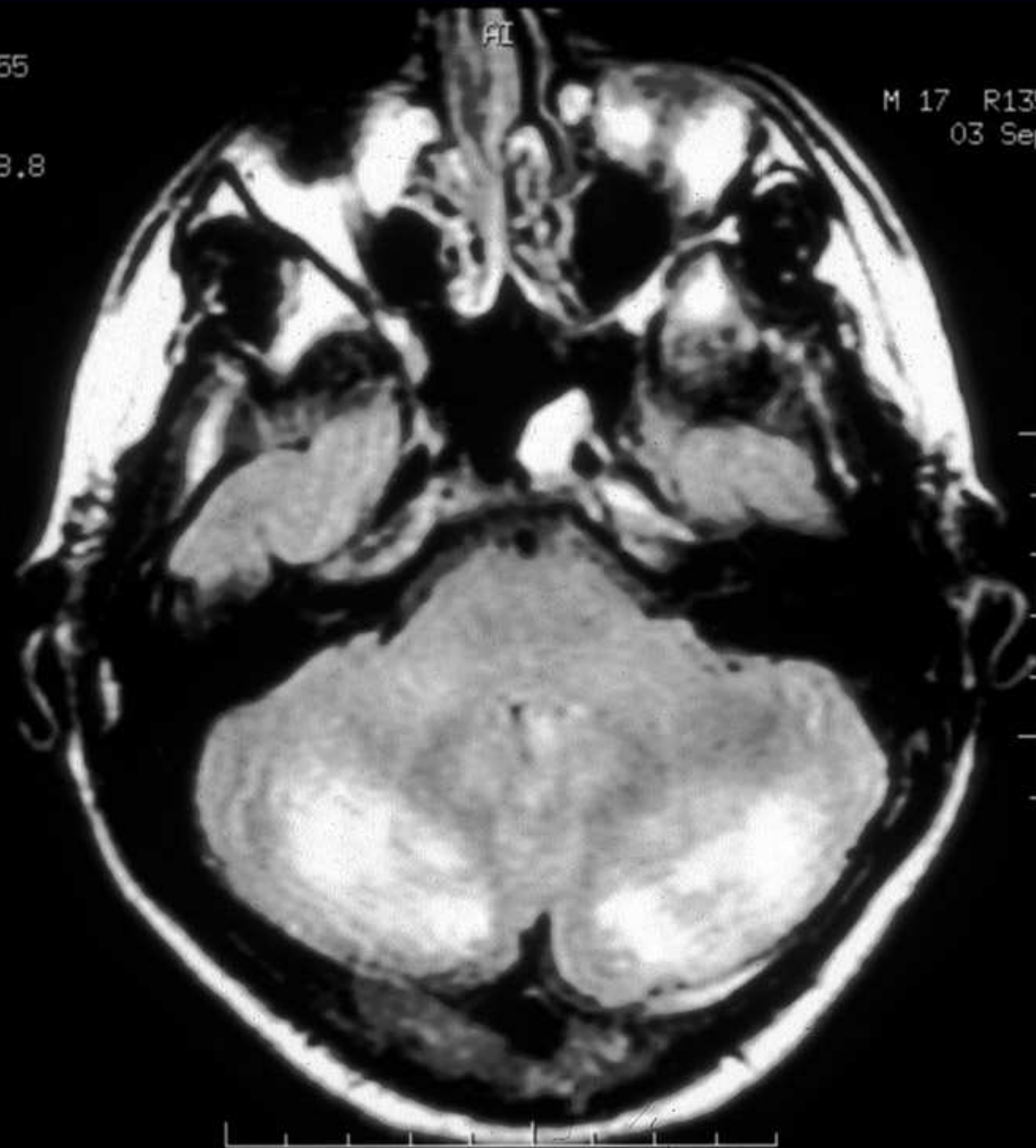
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Post-infectious inflammatory neurological disorders

B] involving the peripheral nervous system

GBS : Guillain-Barré syndrome (in two third of cases)

- rare relapsing forms
- demyelinating (80%) or axonal (20%) forms
- site-specific variants
 - The Miller Fisher syndrome : an anti-GQ1b syndrome with rare CNS involvement : the Bickerstaff encephalitis
 - Facial diplegia

Susac syndrome

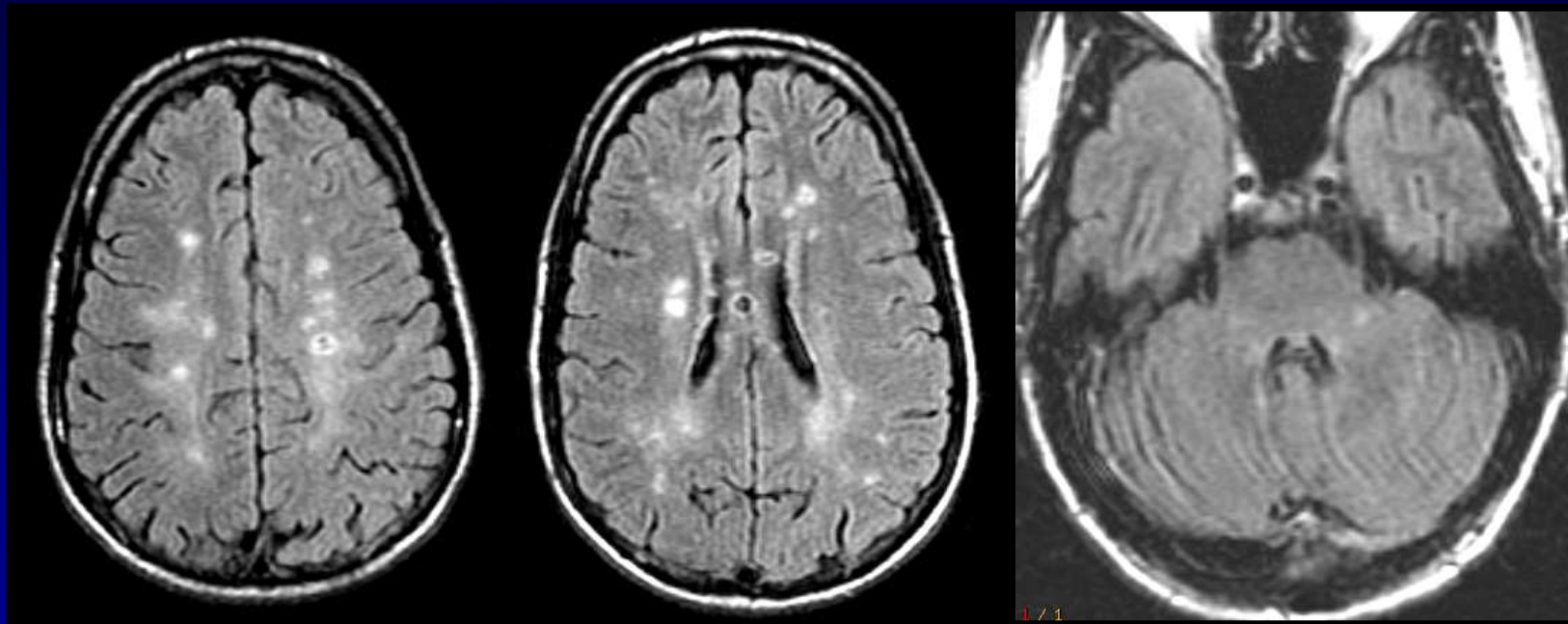
A clinical triad of

- encephalopathy
- hearing loss
- visual field defects

in young adults

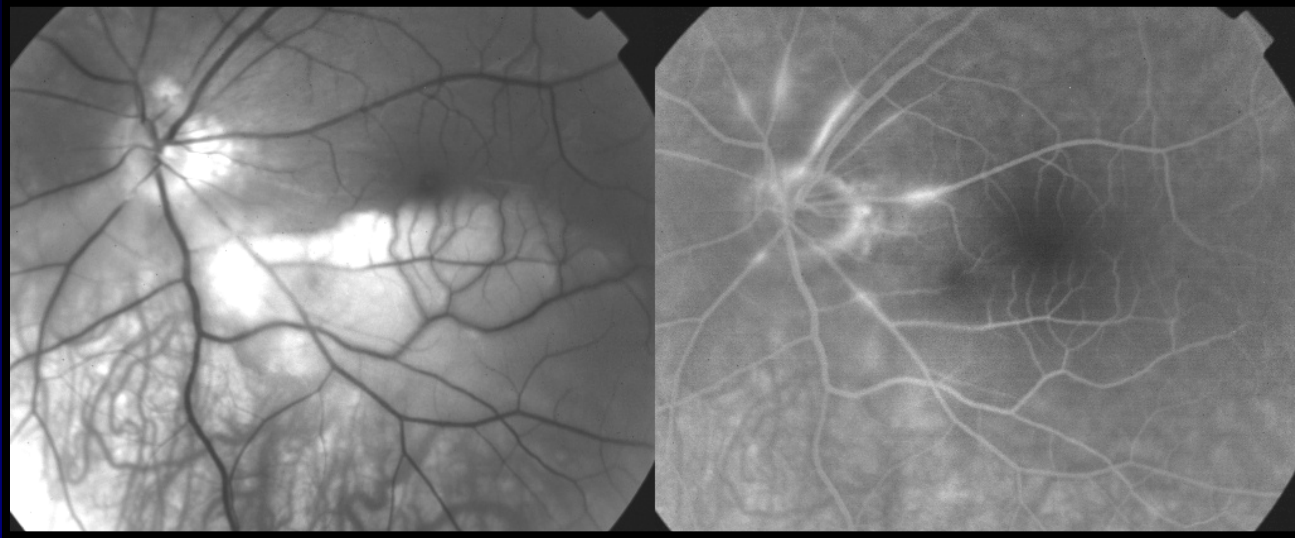
due to a microangiopathy of unknown cause affecting
selectively arterioles of the cochlea, brain and retina

Case 2 – November 2001



Case 2 : transverse FLAIR images through three levels showing diffuse hyperintense white matter lesions

Case 2 - March 2002



Left eye



Conclusions

- as a rule, we have no specific and sensitive biomarkers for most inflammatory disorders of the CNS
- immune biomarkers are relevant in NMO (anti-aquaporin 4 antibody), in paraneoplastic encephalitis, in some auto-immune encephalitis, in Bickerstaff encephalitis
- an auto-antibody may hinder another one ! Co-occurrence of NMO and LED, or Sjögren disease, or myasthenia gravis
- CSF analysis remains a cornerstone in the diagnostic process of these disorders