Analysis of CSF in the diagnosis of Multiple Sclerosis

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Multiple Sclerosis

- MS is a chronic disease of the central nervous system.
- Progressive deterioration of various functions of CNS.
- MS refers to the scars (plaques/lesions) which form in the white matter of the brain over time.
- Demyelinating disease
  - Myelin sheath around the axons of nerve cells becomes damaged
  - Re-myelination by oligodendrocytes decreases progressively
- Damage disrupts cell to cell communication resulting in various neurological symptoms including:
  - physical, mental and sometimes psychiatric problems
Multiple Sclerosis

- MS can occur in isolated attacks (relapsing forms) or progressive forms.
- Relapsing forms- acute attack, disappearing symptoms, some permanent neurological damage.
- Progressive forms- Symptoms worsen over a long period of time as opposed to acute attacks.
- Onset 20-40yrs.
- Twice as common in females.
- Effects 2-2.5 million people globally. Apprx 6000 in Ireland.
MS aetiology

- Cause is unknown, underlying mechanism - autoimmune destruction of myelin & failure/loss of oligodendrocytes.
- Some proposed causes include the interaction between an individual's genetics, environmental factors and infections.
- No known cure
- Treatments attempt to improve function after an attack and seek to prevent further attacks.
- However medications are poorly tolerated, adverse side effects occur:
  - Tachycardia
  - Fatigue
  - Low grade fever
  - Chills
Signs & Symptoms

- Almost any neurological sign or symptom can be a feature of MS.
- Some symptoms include:
  - Loss of sensitivity
  - Tingling, pins and needles, numbness
  - Muscle weakness/spasms
  - Very pronounced reflexes
  - Difficulties with coordination and balance
  - Difficulties with speech or swallowing
  - Visual problems
  - Lehermittes sign- an electrical sensation that runs down the back when bending the neck.
  - Uhthoffs phenomenon- worsening of symptoms due to exposure to higher than usual temperatures.
Three main characteristics of MS

1. Inflammation
2. Destruction of myelin sheath
3. Formation of lesions in white matter
Inflammation

- **The blood brain barrier** - a capillary system, prevents entry of T-cells into the CNS. It may become permeable to these types of cells secondary to an infection by a virus or bacteria.
- T-cells gain entry to the brain via disruptions in the blood brain barrier.
- Recognises myelin as foreign and attacks it.
- Attack of myelin starts an inflammatory process
- Cytokines and antibodies (IgG) are produced
- Other immune cells are recruited to the site
- Further breakdown of the blood brain barrier, progression of disease.
Destruction of the Myelin sheath

- Oligodendrocytes are the cells responsible for creating and maintaining the fatty layer known as the myelin sheath.
- ODs are targets of immune attack in MS, gradual loss.
- Thinning/complete loss of myelin sheath and as the disease progresses the breakdown of the axons of neurons.
- Reduced remyelination & repeated attack means a scar like plaque is built.
Formation of Lesions

- The lesions most commonly effect the cells of the white matter.
- Optic nerve, brain stem, spinal cord, basal ganglia.
- White matter cells carry signals between grey matter areas - where processing is done.
- The rest of the body, the peripheral nervous system is rarely effected.
Diagnosis of MS

- Typically diagnosed based on the presenting signs and symptoms with supporting medical imaging and laboratory testing.

- Most commonly used diagnostics tools:
  - Neuroimaging- MRI to detect lesions in CNS
  - Evoked potentials- is an electrical potential recorded from the nervous system following presentation of a stimulus
  - CSF analysis for the presence of Oligoclonal bands
CSF/Serum analysis

- CSF analysis for the diagnosis of MS is considered the gold standard with a sensitivity and specificity of >95%
- A paired CSF and serum sample are required.
- Must be collected within two weeks of each other.
- Half life of IgG in serum- 23 days.
- We are looking for the presence/absence of protein bands in the CSF/serum.
CSF Isoelectric focusing

- CSF and serum samples are ran simultaneously by means of isoelectric focusing.
- It is the most sensitive method for the detection of OBS in CSF/serum.
- Principle:
  - The separation of proteins in paired patient serum and CSF using agarose gel electrophoresis followed by passive transfer onto a nitrocellulose membrane. The separated IgG’s are then detected directly by horseradish peroxidase labelled anti-human antibodies.
  - The patterns on the gel are then visualised and interpreted.
Once T-cells gain entry to CNS, become auto-reactive or begin to secrete cytokines.

Cytokines and other immune cells including B-cells are recruited to the site of inflammation.

Intrathecal oligoclonal IgG is then produced by B-cells.

When analysing CSF in the lab, intrathecally produced IgG is the protein of interest.

OBS can be produced in the CSF in many other disease states.

It is the pattern of OBS in comparison with serum OBS which determines a MS diagnosis.
OBS interpretation

- Five Patterns

**Oligoclonal Band pattern**

- **Normal**
- **Infection/inflammation**
- **Monoclonal protein**
- **CSF pattern.**
- **extra CSF bands.**

Also seen in MS
Thanks Girls....

- Questions?