Leptospirosis diagnostic tests and typing.

A presentation at:

“Leptospirosis - a global disease but a local phenomenon”

Rural Women New Zealand and the Farmers Leptospirosis Action Group (FLAG)
Landcorp, Level 2, 15 Allen Street, Wellington
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Diagnostic sample types for various tests

- **Culture:** blood, urine, CSF, organ tissue (e.g. kidney)

- **MAT:** serum

- **PCR:** blood/serum, urine, tissues

- **DFM:** blood, urine, tissue slurries
Commonly-used diagnostic tests in NZ

- Microscopic agglutination test (MAT) (serology)

- Polymerase Chain Reaction (PCR)
  – chemical reaction that amplifies minute quantities of DNA

- Bacterial culture

- Darkfield microscopy (DFM) (not generally used in commercial labs, but we use it for research purposes.)
Serology - what is it based on?

• Serology is based on bacterial cell surface antigens - immunological

Think of a jacket...

• Genetically closely related *Lepto* with the almost identical jackets - look the same to the immune system (e.g. Hardjobovis and Balcanica. Same DNA species, almost identical jackets – serologically indistinguishable.

• Two genetically very different bacteria can “wear” the same jacket (e.g. Hardjobovis and Hardjoprajitno). Different DNA species, same jacket – hence, two disparate classification systems for *Leptospira*.

• Either similar species or different species of *Lepto* with similar looking jackets – same serogroup and get cross-reactivity.
• These factors are what lead to some of the vagaries of the serology testing.
Why use serology- what does it tell us?

- Not as informative in acute stages of disease when the body is learning to recognise the antigen

- Screening for exposure to disease

- Often it tells us the serovar in NZ - epidemiologically useful!

- ...but some animals are “silent carriers” – no titres but they do carry Lepto

- Can’t always distinguish between vaccination and exposure titres
PCR: is DNA-based

- Theoretically can amplify 1 gene copy
- Organism does not have to be alive
- **Routine** Vet. Path Lab. methods identify it only as *Lepto* (not the strain)
Bacterial Culture

• **Pros:**
  - Concrete evidence of the presence of live *Lepto*
  - MAT and DNA analysis can both be performed on cultures

• **Cons:**
  - exceedingly slow
  - also often contamination problems, therefore low sensitivity is an issue
Darkfield Microscopy

• Takes time, practice and skill

• Not easy to see if concentration is low

• Easier if alive – not so easy to identify if dead.
Course of the disease – choice of test

- **Lepto colonising organs**
- **Urine**
- **Blood**
- **serum**

**PCR**
- Not too predictable
- Good, not necessarily serovar-specific
- Variable in blood
- Not so sensitive in blood now

**Culture**
- Reasonably good but very slow
- Not suitable
- Not reliable

**DFM**
- Not used in Vet/human Path labs usually, can be sensitive
- Not suitable
- Unreliable

**Lepto shed, sometime intermittently in urine**

**Days post infection**
- 0
- 7
- 14
- 21 rising titres
- 28
- 35
- 42
- 49
- 56 falling titres

**MAT**
- More sensitive, serovar info
- Can indicate past exposure
- Narrow window of opportunity
- MAT serology – can indicate past exposure.
- Low titres may be confused with vaccination titres. Can have “silent shedders” i.e. MAT negative carriers

**Disadvantage**
- Narrow window of opportunity

**PCR**
- Generally reliable – minute quantities DNA needed
- & serovar info missing usually
- PCR and culture may detect “silent shedders”.
- PCR & culture success depends on whether the organism has been cleared from the body – can be serovar – dependent.
Diagnostic Test Choice: Summary

• No single test meets all diagnostic needs

• Different tests more suitable for different sample types at various stages of the disease

• All have benefits and drawbacks

• The limitations of each test, and the choice of test at the stage of disease at which the patient presents, means many cases can go undiagnosed.