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THE ENGINE OF THE NEW NEW ZEALAND

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
Wednesday, 23 October.

Julie Collins-Emerson

mEpiLab

OIE

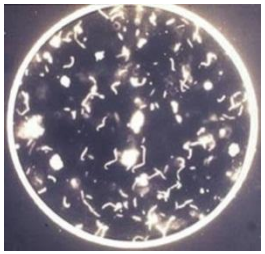
OIE Collaborating Centre for Veterinary Epidemiology and Public Health

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

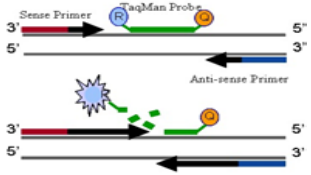
Commonly-used diagnostic tests in NZ

- Microscopic agglutination test (MAT) (serology)



by Becca Chandler

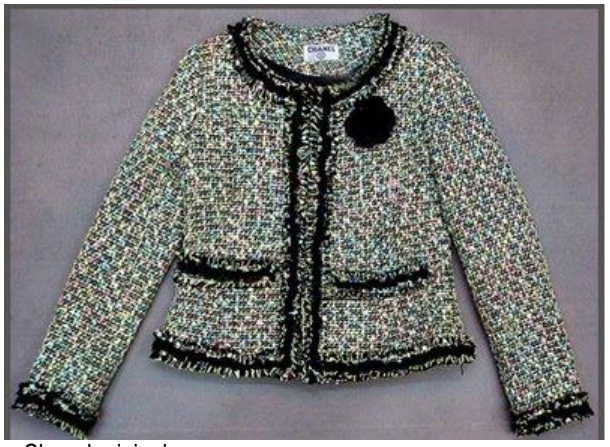
- Polymerase Chain Reaction (PCR)
  – chemical reaction that amplifies minute quantities of DNA



- Bacterial culture

- Darkfield microscopy (DFM) (not generally used in commercial labs, but we use it for research purposes.

Serology- what is it based on?



Chanel original

- Serology is based on bacterial cell surface antigens - immunological

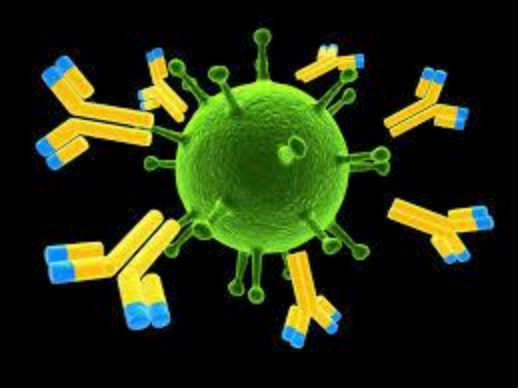
Think of a jacket...

•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.

Chanel knock-off

•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.



Model: antibodies attaching to bacterial cell-surface antigens

Serology

- 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

PCR – not too predicatable
Culture – not reliable
DFM - unreliable

PCR- generally reliable – minute quantities DNA needed & serovar info missing usually
Culture - possible but contamination issues, very slow
DFM – not used in Vet/human Path labs usually, can be sensitive

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.

Lepto colonising organs

Lepto shed, sometime intermittently in urine

Urine

Blood

serum

Lepto circulating in blood stream

0 7 14 21 28 35 42 49 56

Days post infection

rising titres

falling titres

PCR – good , not necessarily serovar-specific
DFM - variable sensitivity
Culture – reasonably good but very slow
MAT – insensitive
Disadvantage – narrow window of opportunity

MAT serology – more sensitive, serovar info
Culture -not suitable
PCR – not so sensitive in blood now

MAT serology – can indicate past exposure.
Low titres may be confused with vaccination titres. Can have "silent shedders" i.e. MAT negative carriers

Diagnostic Test Choice: Summary

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- No single test meets all diagnostic needs
- Different tests more suitable for diff. 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.