

La leptospirose humaine et animale:

Prévention vaccinale

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15^{ème} colloque sur le contrôle Epidémiologique des Maladies Infectieuses
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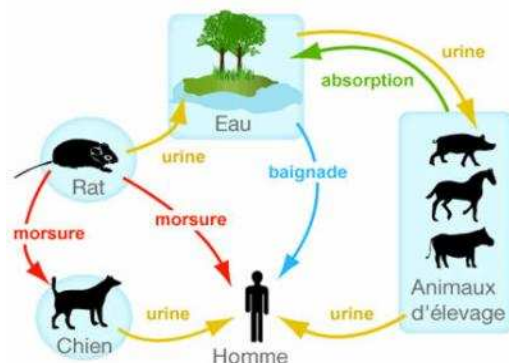
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La leptospirose:



Ordre des Spirochaetales
 Classification compliquée
 Leptospira: 24 sérogroupes,
 230 sérovars
 Plusieurs espèces pathogènes



- Zoonose la plus répandue dans le monde, forme endémique ou épidémique

- Problème de santé publique: 500 000 cas sévères par an, mortalité 10%

- Maladie professionnelle et de loisir, transmise par l'eau

- Prévalence: régionale associée à un ou plusieurs hôtes réservoirs

- Homme: sérogruppe icterohaemorrhagiae

- Chien: sérogroupes icterohaemorrhagiae, canicola, australis, sejroe, grippothyphosa, ...



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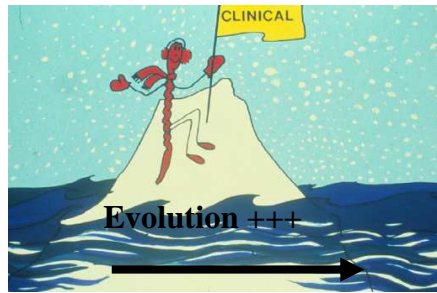


Clinical Diagnostics

Clinical diverse :

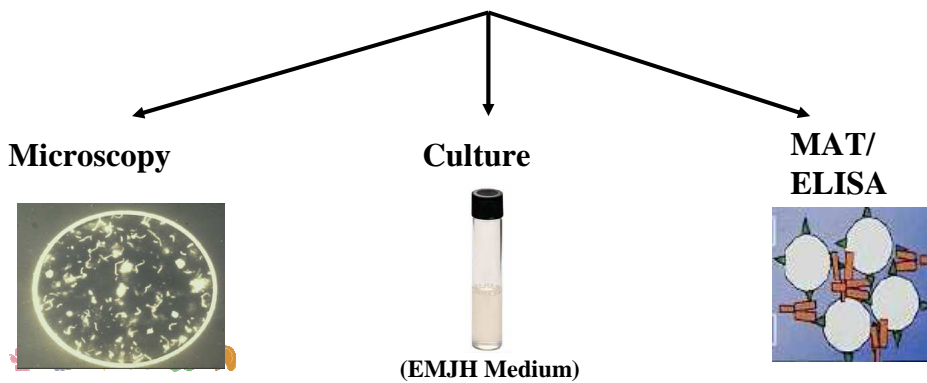
- Flu form
- Fever
- Headache
- Myalgia

- Liver and kidney form

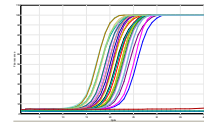


Chronic

Bacteriological diagnosis



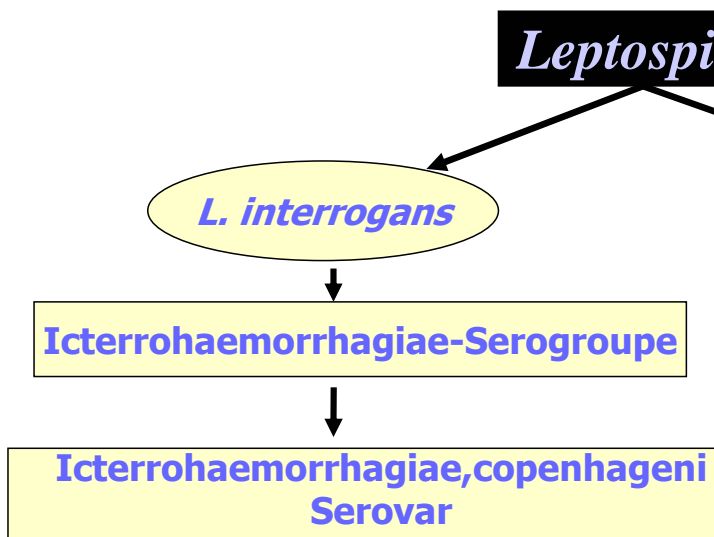
Molecular diagnosis



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Leptospira species



Genomic classification

TABLE 2. Genomespecies of *Leptospira* and distribution of serogroups^a

Species	Serogroups ^b
<i>L. interrogans</i>	Icterohaemorrhagiae, Canicola, Pomona, Australis, Autumnalis, Pyrogenes, Grippityphosa, Djasiman, Hebdomadis, Sejroe, Bataviae, Ranarum, Louisiana, Mini, Sarmin
<i>L. noguchii</i>	Panama, Autumnalis, Pyrogenes, Louisiana, Bataviae, Tarassovi, Australis, Shermani, Djasiman, Pomona
<i>L. santarosai</i>	Shermani, Hebdomadis, Tarassovi, Pyrogenes, Autumnalis, Bataviae, Mini, Grippityphosa, Sejroe, Pomona, Javanica, Sarmin, Cynopteri
<i>L. meyeri</i>	Ranarum, Semarang, Sejroe, Mini, Javanica
<i>L. wolbachii</i>	Codice
<i>L. biflexa</i> ^c	Semarang, Andamana
<i>L. fainei</i>	Hurstbridge
<i>L. borgpetersenii</i>	Javanica, Ballum, Hebdomadis, Sejroe, Tarassovi, Mini, Celledoni, Pyrogenes, Bataviae, Australis, Autumnalis
<i>L. kirschneri</i>	Grippityphosa, Autumnalis, Cynopteri, Hebdomadis, Australis, Pomona, Djasiman, Canicola, Icterohaemorrhagiae, Bataviae,
<i>L. weilii</i>	Celledoni, Icterohaemorrhagiae, Sarmin, Javanica, Mini, Tarassovi, Hebdomadis, Pyrogenes, Manhao, Sejroe
<i>L. inadai</i>	Lyme, Shermani, Icterohaemorrhagiae, Tarassovi, Manhao, Canicola, Panama, Javanica
<i>L. parva</i> ^c	Turneria
<i>L. alexanderi</i>	Manhao, Hebdomadis, Javanica, Mini

**A total of 24 serogroups identified,
...divided into over 230 serovars**



Leptospirosis

1. A multifaceted disease
2. Epidemiology
3. Diagnostic
4. Treatment and Prevention



1. Leptospirosis: a disease with zoonotic implications

Health & Fitness y DT
TUESDAY, OCTOBER 10, 2000

The New York Times

Deadly Infection Re-emerges As People Get Adventurous

By ALICIA AULT

After falling out of her raft on a white-water expedition this spring in the Costa Rican rain forest, Dr. Nicole MacLaren was so grateful she was alive that it did not occur to her that her near-drowning might lead to something else — a potentially fatal bacterial infection called leptospirosis.

But four days after returning to Park City, Utah, Dr. MacLaren, 37, developed the first signs of infection: a fever that soared to 103.5 degrees and drenching sweats that alternated with chills so severe they felt almost like a seizure. Dr. MacLaren, a veterinary ophthalmologist, had a splitting headache, and her eyes and muscles ached. In the emergency room, her kidney and liver were found to be inflamed.

The effect on the kidney and the discovery that she also had meningitis suggested that she had leptospirosis, but her joint aches made dengue fever, a mosquito-borne illness that causes severe joint pain, a suspect as well. Malaria had been ruled out by a blood test. All three illnesses are found in the Costa Rican rain forests.

She was treated with intravenous antibiotics on the assumption that she had an



The *Leptospira interrogans*, cause of a disease transmitted from animals to humans.

Annabella Chang, Microbiology Department, Monash University, Australia



American participants in a race in Malaysian Borneo contracted leptospirosis while swimming in the Segama River.

Don Campbell



1. Epidemiology in Humans

Indirect transmission

- A water story



Participants in the Iron Horse Triathlon begin the swimming portion of the event in Lake Springfield in this June 21 photo.



Triathlon Illinois, USA,
1998
98/ 834 swimmers



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2. Leptospirosis Worldwide

Table 1 Annual incidence of leptospirosis worldwide^a

Countries with the highest incidence			Countries for which no data are available, probably endemic	Other countries	
Rank	Country	Annual incidence per million population		Country	Annual incidence per million population
1	Seychelles	432.1	India	Belarus	3.4
2	Trinidad and Tobago	120.4	Malaysia	Bulgaria	3.7
3	Barbados	100.3	Bangladesh	Chile	1.6
4	Jamaica	78	Vietnam	Colombia	1.6
5	Costa Rica	67.2	Laos	Czech Republic	1.8
6	Sri Lanka	54	Nepal	France	3.9
7	Thailand	48.9	Cambodia	Germany	0.7
8	El Salvador	35.8	Indonesia	Greece	3
9	New Zealand	26	Myanmar	Honduras	3.1
10	Uruguay	25	China	Hungary	3.1
11	Cuba	24.7	Iran	Ireland	2.2
12	Nicaragua	23.3	Suriname	Italy	0.7
13	Croatia	17.3	Haiti	Lithuania	2.2
14	Russia	17.2	Peru	Mexico	1
15	Ukraine	15.3		Netherlands	1.9
16	Dominican Republic	13.8		Panama	1.3
17	Brazil	12.8		Paraguay	1.9
18	Ecuador	11.6		Serbia and Montenegro	1.5
19	Argentina	9.5		Singapore	2
20	Romania	9.4		South Korea	2.8
21	Australia	8.9		Spain	0.3
22	Portugal	6.8		UK	0.6
23	Denmark	6		USA	0.1
24	Latvia	5.6		Venezuela	3.8
25	Slovenia	5.4			
26	Philippines	4.8			
27	Slovakia	4.4			
28	Taiwan	4.1			

Pappas G et al. (2008) - The globalization of leptospirosis - worldwide incidence trends (Int J Inf Dis)



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2. Epidemiology in Humans Indirect and Direct transmission

- Activités professionnelles à risque:**

% du nombre de cas sur 15 ans

Activités professionnelles	%
Agriculture ou élevage	54.7
Egout ou voirie	14.5
BTP	13
Boucherie ou abattoirs	5.4
Forestiers	4.8
Jardinier	1.9

Rapport du groupe de travail du Conseil Supérieur d'hygiène et de Sécurité, 18 mars 2005



METRO.co.uk
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NEWS
Pet rat owner died after being bitten
Thursday, September 27, 2007

A man died after being bitten by a pet rat, an inquest heard.
Matthew Wareing was scratched and bitten on the right hand by his children's rat Roger in July.
Within 10 days his arm had swollen and antibiotics prescribed by his doctor failed to improve his condition.



2. Human Leptospirosis : exemple in France

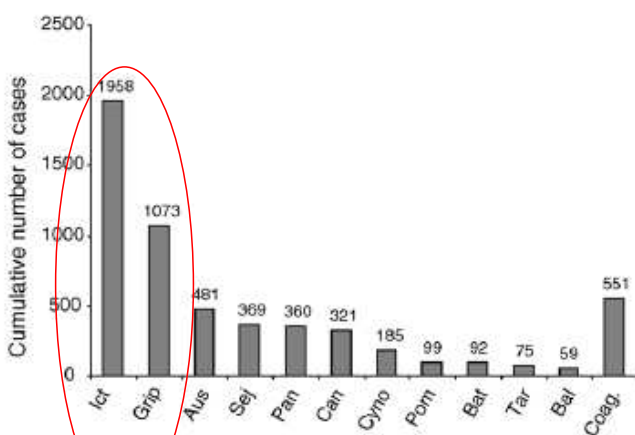
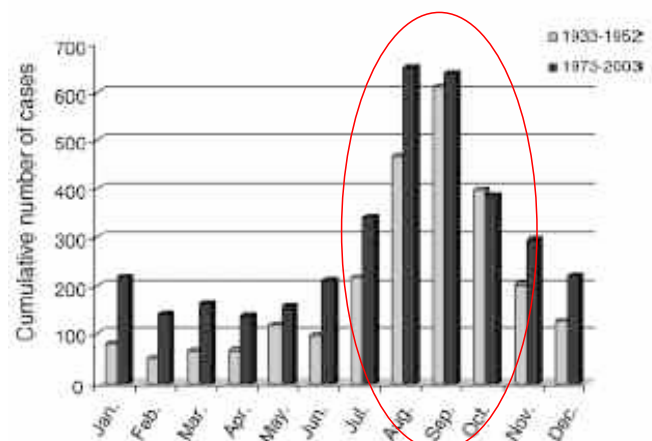


Figure 3 Serogroups involved in human leptospirosis from 1985 to 2003 in France. Ict: Icterohaemorrhagiae; Grip: Grippityphosa; Aus: Australis; Sej: Sejroe; Pan: Panama; Can: Canicola; Cyno: Cynopteri; Pom: Pomona; Bat: Bataviae; Tar: Tarassovi; Bal: Ballum; Coag: Co-agglutinins.



High seasonality

Main serovars



2. Epidemiology on dogs

Literature prevalence study 2005-07

- Key findings and questions:
 - Relatively high prevalence of serovars other than **Ictero** and **Canicola** are observed across Europe.
 - **Grippotyphosa** is among those serovars but other serovars are of equal or higher importance :
 - from serogroup australis (bratislava, australis)
 - from group sejroe (hardjo, saxkoebing, sejro)
 - ...
 - Europe ≠ North-America (Pomona?)
 - What about clinical relevance?



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2. Evolution of serogroup prevalence on dog in France (July to December 2007)

Pr A. Kodjo

- 184 samples, 120 seropositive
 - **Ictero** : 60% (73/120)
 - **Canicola** : 54,1% (65/120)
 - Autumnalis : 28,3 (34/120)
 - **Australis** : 23,3% (28/120) ↗
 - **Sejroe** : 16,6% (20/120) ↗
 - Grippo : 13,3% (16/120)
 - Pyrogenes : 8,3% (10/120)
 - Bataviae : 3,3% (4/120)
 - Panama : 3,3% (4/120)



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3. Diagnostic



3. Laboratory diagnosis Which test?

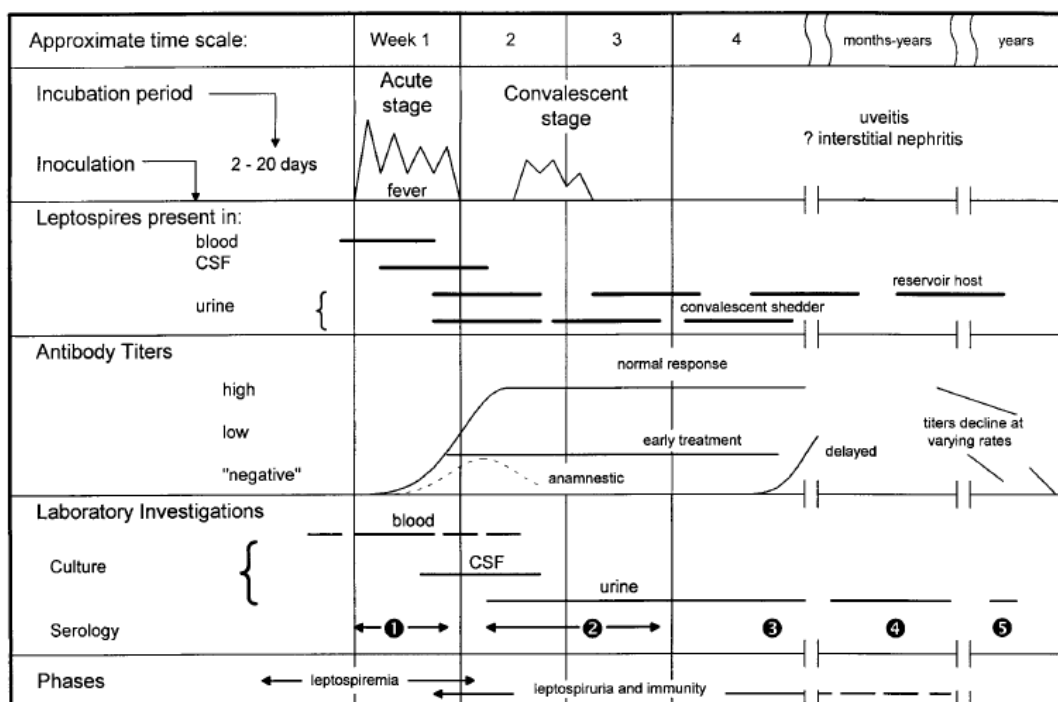


FIG. 2. Biphasic nature of leptospirosis and relevant investigations at different stages of disease. Specimens 1 and 2 for serology are acute-phase specimens, 3 is a convalescent-phase sample which may facilitate detection of a delayed immune response, and 4 and 5 are follow-up samples which can provide epidemiological information, such as the presumptive infecting serogroup. (Adapted from reference 586a with permission of the publisher.)



3. Leptospirosis

Laboratory diagnosis



Laboratory blood testing parameters

	Birnbaum & al., 1998 (USA) (36 dogs)	Goldstein & al., 2006 (USA) (55 dogs)	Geisen & al., 2007(Germany) (42 dogs)	Hugonnard & al., 2009 (France) (37 dogs)
Creatinin	83%	93%	57%	78%
Glycosuria	9%	30%	NR	64%
↗ PAL	56%	57%	81%	49%
↗ ALAT	33%	32%	74%	26%
Anemia	33%	53%	45%	NR
Thrombo- cytopenia	NR%	30%	53%	NR

Hematological anomalies related to bleeding syndrome (vasculatitis)



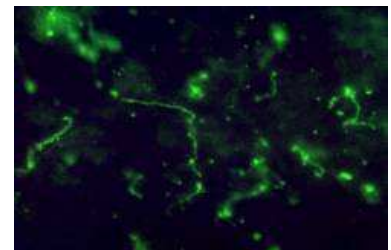
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3. Leptospirosis

Serology and direct testing

- Serologic testing
 - MAT (Microscopic Agglutination Test): considered as the golden standard
 - ELISAs (IgM / IgG)
 - Indirect fluorescent antibody tests
- To be considered
 - MAT: good sensitivity and allows for the detection of group-specific antibodies
 - ELISA: detect specific IgM with a rather high sensitivity (as a sign of current or recent leptospirosis). These tests do not distinguish group-specific antibodies
 - Abs need 3 – 10 d to reach detectable level (IgM → IgG)
 - Need to demonstrate 4-fold titer increase over 2 w
 - Titer raise can be impacted by treatment



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3. Leptospirosis

Serology and direct testing

- Direct testing
 - Immunofluorescence (blood & CFS) by dark-field or phase-contrast microscopy
 - PCR (serum & urine, LCR): high sensitivity. Usually a generic PCR which do not distinguish different groups/serovar
 - → Use of VNTR-PCR (Variable Number Tandem Repeats) in specialized labs (Institut Pasteur, Paris)
 - Culture: “gold standard”, but difficult and many false negative due to sample treatment



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3. Key learnings

About Epidemiology and clinical diagnosis

- Epidemiology and serovar involvement not always well-known
- Leptospirosis is often underdiagnosed
 - In animals (various clinical signs)
 - In Humans (flu-like syndrom)
- Clinical signs are extremely variable: not only hepatic and renal.
 - Frequent haemorrhagic gastroenteritis
 - Ocular signs
 - Babesiosis like symptoms / dog



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4. Treatment & Prevention

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4. Canine Leptospirosis Treatment

- Supportive treatment + antibiotherapy
- **Penicillins** (Ampicillin and Amoxicillin)
Tetracyclins (Doxycycline)
- Diuresis monitoring: use furosemid if
 - Diuresis $< 0.25\text{ml/kg/h}$
 - Correct hydration
- Liquid therapy
- Antivomitives
- Pain killers
 - Corticosteroids or non-steroid
 - NB: with use of morphinics, mortality rate may be reduced by around 50%)



Siamu - Physiology ENVL



4. Leptospirosis

Sanitary prevention



- Environment control :
 - Cleaning
 - Disinfection
- Control of rodents in kennel
- Infected animals isolation



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La prévention par la vaccination

Sociétés	IMAXIO	MERIAL
Espèce cible	Homme	Chien
Valences	<i>L. interrogans icterohaemorrhagiae</i>	<i>L. interrogans icterohaemorrhagiae</i> <i>L. interrogans canicola</i>
Efficacité	<ul style="list-style-type: none"> •Taux de séroconversion de: 93% en IgM après la 2em injection 97% en IgG un mois après le 1^{er} rappel à 6 mois 100% en IgG après le 2em rappel •Aucun cas de Leptospirose rapporté dans la population vaccinée 	<ul style="list-style-type: none"> •Réduction: <ul style="list-style-type: none"> -de la mortalité -des signes cliniques -de l'excrétion bactérienne •Durée d'immunité: 14 mois
AMM	France ATU / Italie	<ul style="list-style-type: none"> •Europe •Zone Internationale
Produits	SPIROLEPT 	EURICAN L EURICAN LR EURICAN CHPPi2 EURICAN CHP 



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4. Leptospirosis vaccination

- Commercially available vaccines rely on inactivated (adjuvanted) whole cell or membrane preparations of pathogenic leptospire

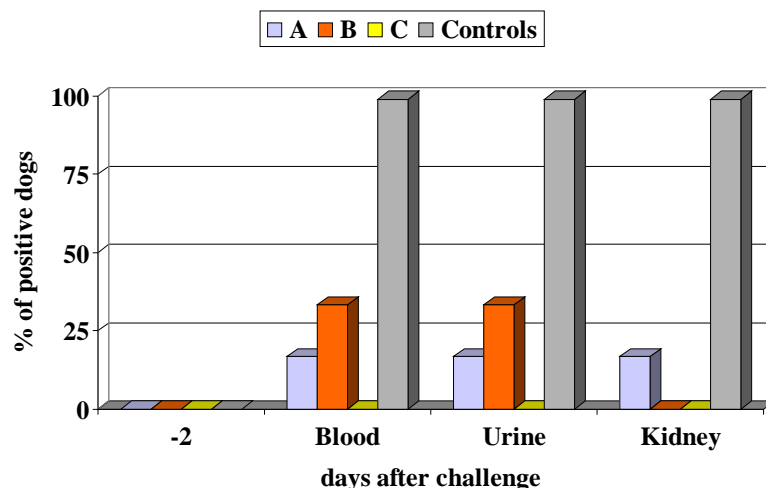


- In Europe, *L. icterohaemorrhagiae* and *L. canicola* bacterins are the most frequent bacterin preparations available
- New serovars to be included in the future: but clinical and epidemiological relevance to be carefully considered



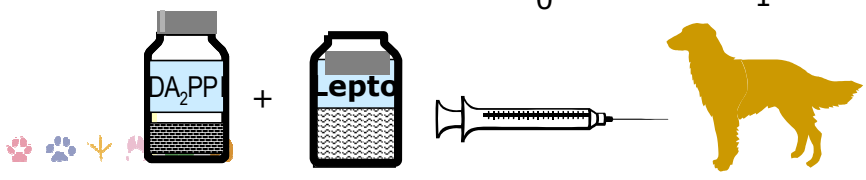
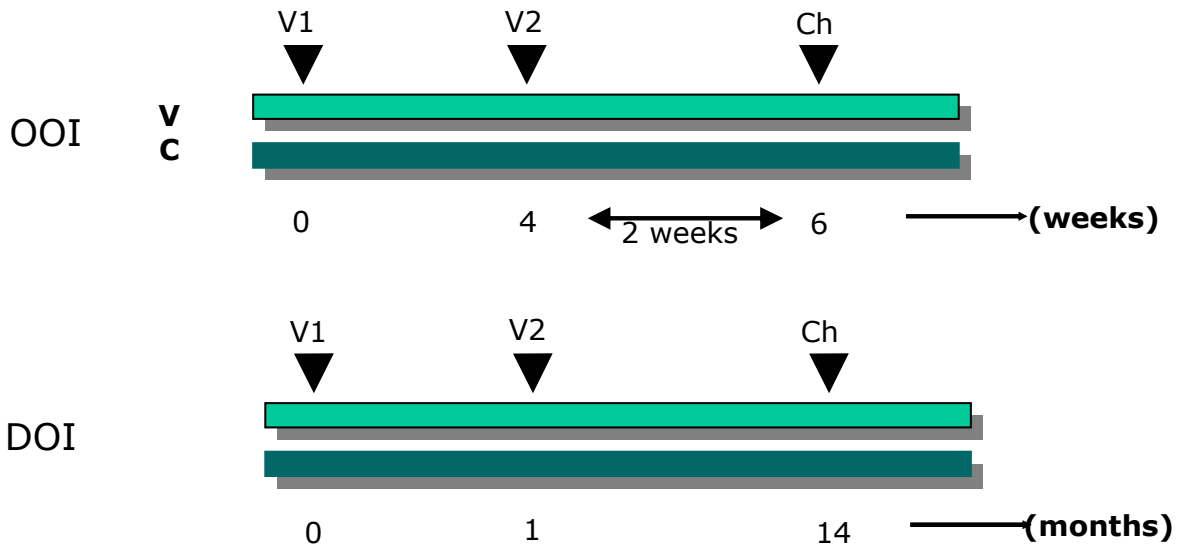
4. Leptospirosis vaccination

- All vaccines are not equivalent
 - The challenge induced only mild clinical signs of leptospirosis.(no mortality was observed)
 - Even under these mild conditions, some vaccines failed to protect against infection and renal carrier state



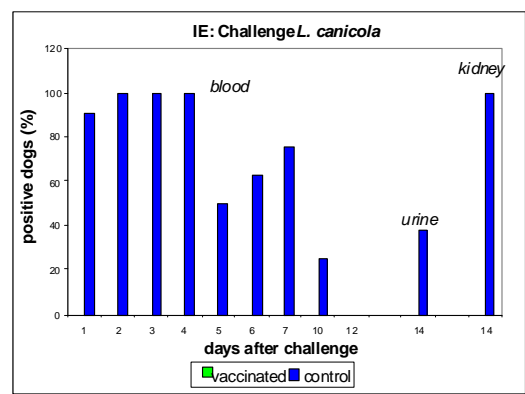
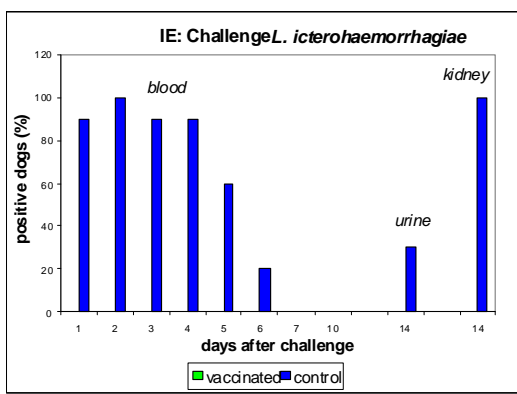
4. Eurican Lepto Protection data

- Onset and Duration of immunity studies



4. Eurican lepto Short term protection

OOI



- High mortality rate observed in controls (50-60%) None in the vaccinated group

→Leptodog provided complete clinical and bacteriological protection against severe challenges with Li and Lc as soon as 2 weeks after vaccination

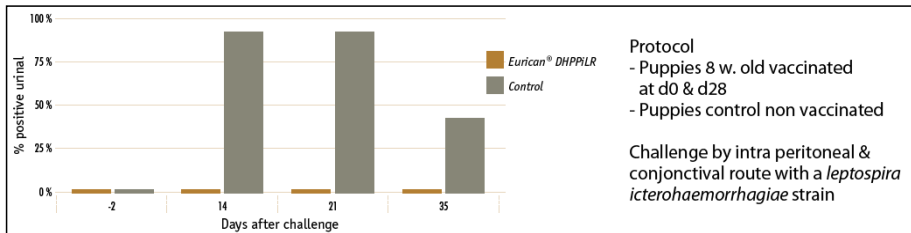
Minke J. et al. (2009, Vet Microb)- OOI & DOI against clinical disease & renal carriage in dogs provided by a bivalent leptospirosis vaccine



4. Eurican lepto Long term protection

- Results of the challenge performed 14 months after the primary course of two injections
 - Li and Lc challenges induced mortality in respectively 3/10 and 1/8 of control dogs, whereas no mortality occurred in the vaccinates
 - No renal carriage / urine shedding in vaccinated dogs

DOI



→ Leptodog provided significant protection against clinical signs and renal carrier stage after challenge with Li and Lc

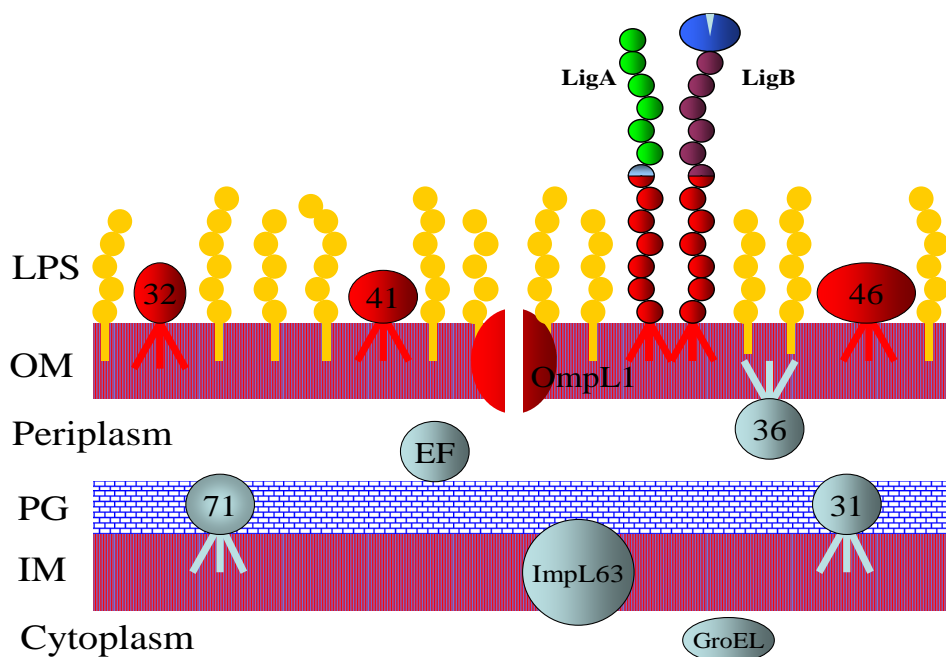
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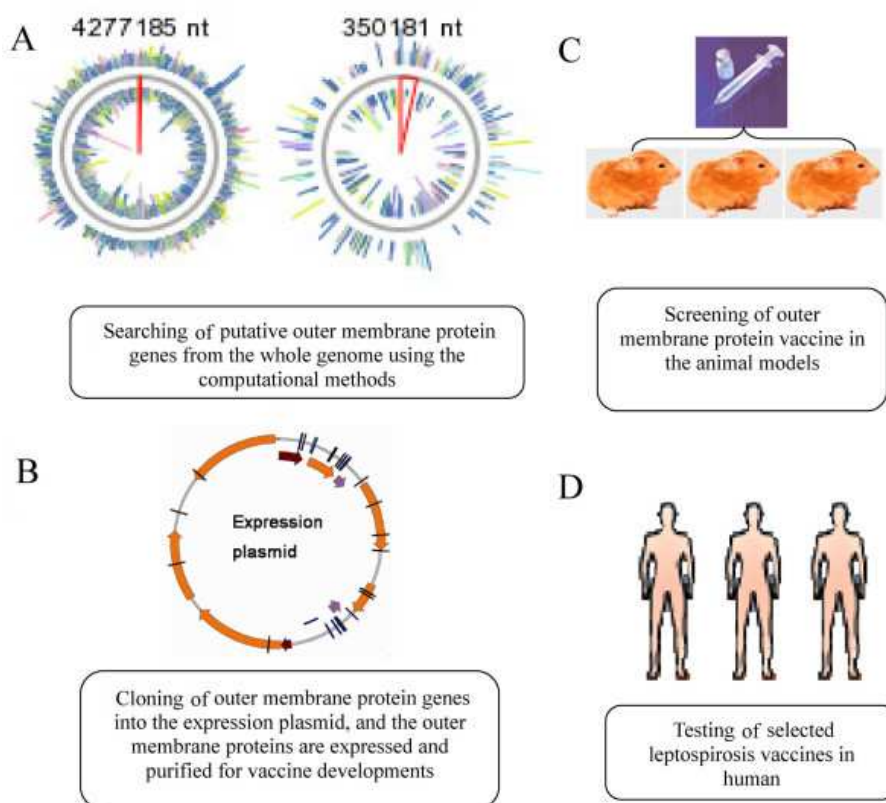
Structure of leptospira membrane



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Large-scale screening of the leptospirosis vaccines



Z. Wang et al, 2007



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Leptospirosis vaccines candidates

Recombinant protein vaccines: Search of pan-lepto antigen (s)

- Recombinant OMP vaccines (OmpL1, Lig, LAg42, Loa22, Lk73.5)
- Recombinant lipoprotein vaccines (LipL41, LipL32, LipL45..., GLPs)
- Recombinant virulence factor vaccines (Hsp58, Fla, hemolysin SphH, ChpK)

DNA vaccines

LPS vaccines

Inactivated vaccines

Z. Wang et al, 2007



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**Any
question ?**

