ANIMAL MODELS IN THE STUDY OF HUMAN SPIROCHETAL INFECTIONS

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HUMAN SPIROCHETAL DISEASES

LYME DISEASE - *B. burgdorferi, B. afzelii, B. garinii*

SYPHILIS AND OTHER TREPONEMATOSES - *T. pallidum* subsp. *pallidum*, *pertenue*, *endemicum*, *T. carateum.*

LEPTOSPIROSIS - >250 Pathogenic *Leptospira* serovars (8 species).

RELAPSING FEVERS- *B. recurrentis* (louse-borne); *B. hermsii*, *B. turicatae, B. parkeri,* several additional species (tick-borne)

PERIODONTAL DISEASE – *T. denticola, T. pectinovorum, T. socranskii,* and *T. vincentii.*

IDEAL CRITERIA IN CHOOSING AN ANIMAL MODEL

- Economically feasible.
- Relatively easy to handle.
- Sites of infection are anatomically similar or identical to those of the human disease.
- Induces a host response identical or closely similar to the human infection when utilizing the same portal(s) of entry.
- Results in genotypic and phenotypic properties of the etiologic agent identical or similar to human infection isolates during host-parasite interaction.
- Permits the recovery of relatively large numbers of organisms from the animal host for propagation and/or biological study.
- Inbred (for immunogenetic studies).
- Immunologically deficient (for selected studies).

ANIMAL MODELS VS HUMAN INFECTIONS: FACTORS THAT MAY INFLUENCE SIMILARITIES AND DIFFERENCES IN PATHOGENESIS

- Strain, serotype, culture state of pathogen.
- Dosage.
- Age of animal.
- Sex of the animal.
- Portal of entry (Site of inoculation).
- Anatomical structure.
- Immunologic state.
- Genotype.

LYME DISEASE

Key Clinical Features

- EM, lymphocytoma, and ACA (late).
- Lymphadenopathy.
- Arthritis.
- Myocarditis and pericarditis.
- CNS and peripheral neuropathies.
- Chronicity.

ANIMAL MODELS OF LYME DISEASE

1) **Mouse (Inbred C3H & C3H Scid) - Infant and weanling.**
   - Skin, ear, kidney, bladder, brain, and spleen infection by most strains; EM and clinical neuroborreliosis absent; evidence for tissue tropism; organisms in ear tissue demonstrable by PCR and infectivity studies.
   - Synovitis, arthritis and carditis; more severe in young weanling and Scid mice; characterisitic histopathological response.
   - Chronic persistence of skin infection.

   Mouse Permits studies of:
   - Mechanisms of pathogenicity and pathogenesis following infection, including tick attachment, initiation of infection, gene and protein expression and regulation, chronicity, and acute synovitis, arthritis, and carditis.
   - Vaccinogen safety and efficacy, including immune and histopathological responses following vaccination and challenge.
   - Immune mechanisms of protection.
   - Tissue tropism among Bb strains.

2) **Rabbit (Outbred New Zealand White) - Adult**
   - Skin, popliteal lymph node, joint, and spinal cord infection; evidence for tissue tropism; arthritis, carditis, and clinical neuroborreliosis absent.
   - EM at site of inoculation; characteristic histopathological response; morphologically intact organisms detectable in infected skin.
   - In vivo (peritoneal) implant chambers result in host-adapted Bb.
   - Clearance rather than persistence in 3 months followed by complete "infection-derived immunity" to challenge by needle inoculation or skin implantation with relatively large inocula.
   - Absence of a cellular response to intradermal challenge of infection-derived immune animals.

   Rabbit (New Zealand White) permits studies of :
   - Mechanisms of pathogenicity and pathogenesis, including tick attachment, initiation of infection, gene and protein expression and regulation.
   - Molecular and cellular basis for EM development.
   - Immune mechanisms of clearance and protection against challenge of infection-derived immune animals.
   - Vaccinogen safety and efficacy, including immune and histopathological responses following vaccination and challenge.

3) **Dog (Beagle) - Age 6 weeks**

- Infected tick-feeding.
- Persistent infection of skin, cervical lymph nodes, and pericardium.
- Moderate to chronic progressive synovitis; lameness; characteristic histopathological response.

Dog (Beagle) permits studies of :

- Mechanisms of pathogenicity and pathogenesis, including tick attachment, initiation of infection, histo- and immunopathological response, gene and protein expression and regulation, and synovitis.
- Vaccine safety and efficacy including immune and histopathological response to vaccination and challenge.
- Immune mechanisms of protection

4) **Rat (Inbred LEW/N) Infant and weanling.**

- Ear, skin, spleen, and brain infection.
- Acute exudative clinical arthritis in tibiotarsal joints, myocarditis, and vasculitis ; characteristic histopathological response.
- Persistence of infection (chronicity).
- Immune and histopathogical response to infection, vaccination, and to challenge.
- In vivo (peritoneal) chamber implants (4 to 6 week Sprague-Dawley outbred) result in host-adapted Bb.

Rat (Lew/N and Sprague Dawley) permit studies of:

- Mechanisms of pathogenicity and pathogenesis of infection, including initiation of infection, histo- and immunopathological responses, gene and protein expression and regulation, and induction of arthritis, myocarditis, and vasculitis.
- Vaccinogen safety and efficacy including immune and histopathological response to vaccination and challenge.
- Immune mechanisms of protection.
- Tissue tropism among Bb strains.

5) **Rhesus Monkey - Adult.**

- Consistent skin infection and EM at the inoculation site; characteristic histopathological response.
- Chronically persistent joint inflammation, myocarditis and conjunctivitis; clinical arthritis and neuroborreliosis with characteristic histopathological responses; evidence for tissue tropism.

Rhesus Monkey permits studies of:

- Mechanisms of pathogenicity and pathogenesis following infection, including tick attachment, initiation of infection, histo-and immunopathological response, gene and protein expression and regulation, chronic arthritis, myocarditis, and neuroborreliosis.
- Molecular and cellular basis for EM development.
- Vaccinogen safety and efficacy, including immune and histopathological responses following vaccination and challenge.
- Immune mechanisms of protection.
- Tissue tropism among Bb strains.

6) **Hamster (Outbred Golden Syrian and Inbred LHS)**

- Adult and immunologically competent.
- Susceptible to infection by hind paw (foot pad) injection.
- Persistence of infection in ear, skin, heart, and bladder (chronicity).
- EM and neuroborreliosis absent.
- Persistent mild synovitis to severe clinical arthritis in all paws; characteristic histopathological response.
- Severe destructive arthritis following whole organism or OspA vaccination and challenge; characteristic histopathological response.

Hamster (Golden Syrian and LSH) permits studies of:

- Mechanisms of pathogenicity and pathogenesis responsible for arthritis and chronicity.
- Vaccinogen safety and efficacy including immune and histopathological response following vaccination and challenge.
- Immune mechanisms of protection

7) **Gerbil (Outbred) - Young adult.**

- EM and neuroborreliosis absent.
- Persistence of localized and disseminated infection in skin, joints, and spleen (chronicity).
- Characteristic histopathological response in the skin and most of the infected viscera

Gerbil permits studies of:

- Mechanisms of pathogenicity and pathogenesis, including histo- and immunopathogological response, gene and protein expression and regulation, and chronicity.
- Vaccinogen safety and efficacy, including immune and histopathological responses following vaccination and challenge.
- Immune mechanisms of protection.
- Tissue tropism among Bb strains.

8) **Guinea Pig (Outbred Hartley) Young adults (< 2 months).**

- Infection of the bladder and knee joints; occasional infection of the heart, bladder, spleen, and spinal cord; EM, ear infection, clinical arthritis, and clinical neuroborreliosis absent; minimal histopathological responses in the bladder and knee joints.
- Chronic persistence of infection in knee joints, and clearance in other tissues.

ANIMAL MODELS OF SYPHILIS

1) Rabbit (Outbred New Zealand White or Dutch)

Acquired Syphilis

- Papules that progress to ulceration (chancre) following intradermal inoculation.
- Orchitis following intratesticular inoculation.
- Popliteal and inguinal node lymphadenopathy by both routes of inoculation.
- Characteristic histopathological and serological responses.
- Healing of lesions and latency (persistence of spirochetes in nodes, spleen , liver for life of animal); minimal to no histopathological changes; correlative reversal of nontreponemal antibody to nonreactive.
- Development of infection-derived immunity.
- Absence of secondary and late manifestations.

Congenital syphilis

- controversial.

Rabbit (New Zealand White or Dutch) Acquired Permits:

- In vivo strain propagation, source of treponemes for study, and maintenance.
- Studies of mechanisms of pathogenicity and pathogenesis including attachment, initiation of infection, histo- and immunopathological response, latency, gene and protein expression and regulation.
- Vaccinogen safety and efficacy; homologous protection and cross-protection against subspecies and strains.
- Immune and histopathological responses to vaccination and challenge following vaccination.
- Immune mechanisms of homologous and cross-protection.
- Tissue tropism.

2) Mouse (Outbred Swiss-Webster) - Weanling infant and adult.

Acquired Syphilis

- No skin lesions; absence of late manifestations.
- Skin, spleen, nodes, and brain infection; persistence for at least 3 months; absence of a histopathological response (latency).
- Treponemal but not nontreponemal antibody response.

Congenital

- Not susceptible.

Mouse Permits studies of:

- Vaccinogen efficacy of native *T. pallidum* subsp. *pallidum* molecules which exist in low amounts (Tromps).
- Humoral immune mechanisms of protection.
- Hamster (Outbred and Inbred)
- Intradermal lesions rare and atypical.
- Popliteal and inguinal node lymphadenopathy.

NON-VENEREAL TREPONEMATOSES

Key Clinical Features

Yaws (Frambesia)

Primary

- Hyperkeratotic papilloma (mother yaw).
- Regional lymphadenopathy.

Secondary

- Crops of papillomatous lesions (daughter yaws).
- Generalized lymphadenopathy.
- Hyperkeratosis of palms and soles.
- Long bone osteoperiostitis.

Latency with Relapses.

Late

- Benign gummas of skin and bones of the face.
- Hyperkeratosis of palms and soles.
- Cartilage destruction.
- Bone deformities.

Endemic Syphilis (Bejel, Dichuchwa)

Primary

- Rare (Papule or shallow ulcer).
- Regional lymphadenopathy.

Secondary

- Ulcerative mucous patches on the oropharygeal muscosa.
- Long bone osteoperiostitis.
- Generalized lymphadenopathy.

Latency

Late

- Benign gummata of nasopharynx, skin, and bone.
- Bone destruction.

Pinta

Primary

- Pigmented papules, single or multiple.
- Regional lymphadenopathy.

Secondary

- Generalized hyperpigmented papules (pintids).

Late

- Depigmented lesions (achromia).
- Hyperkeratosis of the palms and soles.

ANIMAL MODELS OF NON-VENEREAL TREPONEMATOSES

Yaws and Endemic Syphilis

1) **Rabbit (Outbred New Zealand White or Dutch) - Adult.**
   - Typical papules and ulceration or macular lesions with no induration (Intradermal).
   - Granular periorchitis by most yaws treponeme strains and some endemic syphilis strains (Intratesticular).
   - Popliteal and inguinal lymphadenopathy.
   - Characteristic histopathological and serological response.
   - Healing of lesions and latency.
   - Absence of secondary and late manifestations.

2) **Hamster (Outbred Golden Syrian, Inbred LSH, CB) -Adult. Intradermal in groin area.**

Yaws (Golden Syrian and CB)
- Strain variability.
- Skin lesions absent, inguinal lymphadenopathy.
- Chronic skin lesions, inguinal lymphadenopathy.
- Chronic skin lesions, absence of lymphadenopathy.
- Characteristic histopathological and serological response when lesions and/or lymphadenopathy present.
- Healing and latency.
- Absence of secondary and late manifestations.

Endemic Syphilis (Golden Syrian and LSH)
- Chronic skin lesions, inguinal lymphadenopathy (all strains tested).
- Characteristic histopathological and serological response.
- Healing and latency.
- Absence of secondary and late manifestations.

Rabbit and Hamster Permit (forYaws and Endemic Syphilis):
- In vivo propagation, source of treponemes for study, and maintenance in rabbits (only some strains of yaws treponeme).
- Studies of mechanisms of pathogenicity and pathogenesis, including attachment, initiation of infection, histo-and immunopathological response, latency, gene and protein expression and regulation.
- Vaccinogen safety and efficacy; homologous protection and cross-protection among subspecies and strains.
- Immune and histopathological responses to vaccination and challenge following vaccination
- immune mechanisms of homologous and cross- protection.
- Tissue tropism.

Pinta

1) **Chimpanzee**
   - Scarification and intradermal.
   - Pintids (2 of 6 animals).
   - No serological response.

LEPTOSPIROSIS

Key Clinical Features (Serovar Dependent)

Anicteric

- Fever followed by afebrile period.
- Rash, uveitis, hepatitis, renal infection, meningitis.

Icteric (Weil Syndrome)

- Vascular collapse and hemorrhage.
- Renal failure and hepatic dysfunction.
- Jaundice.

ANIMAL MODELS OF LEPTOSPIROSIS

1) **Hamster (Golden Syrian) and Guinea Pig (Hartley) - Young adult or weanling. Intraperitoneal inoculation.**
   - Liver and renal infection, jaundice, and petechial hemorrhage; characteristic histopathological response.
   - High lethality rate - dose dependent.
   - Chronicity with small inoculum.

2) **Calf and Steer -Young adult and adult. Conjunctival inoculation of *L. interrogans* serovar hardjo.**
   - Conjunctivitis and renal infection; characteristic histopathological response; persistence and shedding for at least 8 months (chronicity).

Hamster, Guinea Pig, Calf, and Steer Permit studies of:

- Mechanisms of pathogenicity and pathogenesis following infection, including histo-and immunopathological response, gene and protein expression and regulation, chronicity.
- Vaccinogen safety and efficacy including immune and histopathological responses following vaccination and challenge.
- Immune mechanism(s) of protection.

RELAPSING FEVERS

Key Clinical Features

- Multiple alternating afebrile and febrile periods as a result of antigenic variation.
- Macular or petechial rash.
- Splenomegaly and hepatomegaly.
- Occasional meningitis and encephalitis.
- Latency (probable persistence of organisms in CNS, bone marrow, spleen, and liver during remissions).

ANIMAL MODELS OF RELAPSING FEVERS

1) **Mouse (Inbred Balb/c and C3H, Scid) - Any age, but weanling best.**

***hermsii* (Balb/c and C3H)**
- recurrent bouts of spirochetemia and clearing; multiphasic variation of VMPs (Vlp and Vsp) as a result of gene rearrangement; progeny of a single organism may give rise to 40-50 antigenically distinct VMPs.

***turicatae*, serotype A (Scid)**
- CNS invasion, myocarditis, uveitis, and cranial nerve disorder;

***turicatae*, serotype B (Scid)**
- severe arthritis, myocarditis, uveitis, and cranial nerve disorder. Vsp homology to *B. burgdorferi* OspC.

turicatae, serotype A and B differ only in the size of a single expressed Vsp protein, homologous to *B. burgdorferi* OspC.

***B. hermsii* Immunocompetent Mouse Model Permits studies of:**
- Mechanisms of pathogenicity and pathogenesisis including:
- Tick attachment, initiation of infection, gene and protein expression and regulation.
- Mechanism by which Vmp antigenic variation is selected by the host.
- Role of OM proteins.

***B. turicatae* Scid Mouse Model Permits studies of:**
- Mechanisms by which borreliae enter the CNS and joints; role of the Vsp-OspC family of expressed proteins.

PERIODONTAL DISEASE

Key Clinical Features

- Inflammation of the supporting tissues around and beneath the gums (periodontal membrane, gingiva (gums), subgingiva, and alveolar bone (tooth socket).
- Tooth loss, destruction of periodontal ligaments and supporting alveolar bone.

ANIMAL MODELS OF PERIODONTAL DISEASE

1) **Mouse (Balb/c and ICR) -Young adult. Subcutaneous or intraperitoneal injection of: *T. denticola*, *T. pectinovorum*, *T. socranskii*, and *T. vincentii*.**
   - Local skin abscesses and necrosis, spreading exudative hemorrhagic lesions; requires 10^9- 10^{10} organisms.

   Mouse Permits studies of:
   - Virulence factors associated with periodontitis (???).
   - Mechanisms of pathogenicity and pathogenesis (applicability to periodontitis ???)

2) **Baboon (5 to 30 years of age) and Dog (Beagle)**
   - Large oral cavity and oral structure identical to humans; different premolar shape and structure; different dental arch shape (baboons).
   - Naturally occurring periodontitis increasing in severity with age (similar to humans).
   - Experimentally induced in dogs with corticosteroid treatment.
   - Ligatures, and/or surgically produced periodontal defects.

   Baboon and Dog (Beagle) Permits studies of:
   - Definitive etiology of periodontitis (Treponemal???).
   - Mechanisms of pathogenicity, pathogenesis, and repair.