Yersinia, Pasteurella and Francisella

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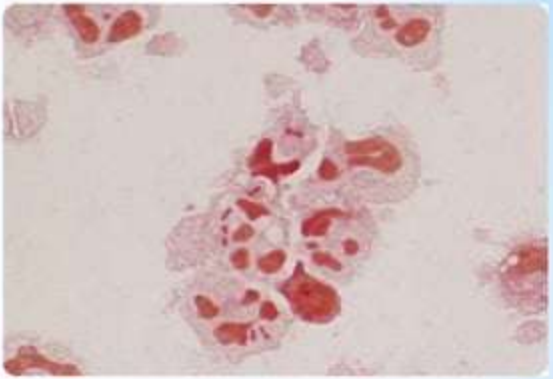
* Tribe Yersinieae comprises Genus Yersinia
* 3 well established pathogens
  * Yersinia pestis - Plague
  * Yersinia pseudotuberculosis - Yersiniosis
  * Yersinia enterocolitica - Yersiniosis

*Yersinia

*Yersinia pestis

* Isolated by Alexander Yersin - 1894 - Hong Kong
* Causes Plague - systemic zoonosis - arthropod vector - rat flea
* GN oval coccobacillus
* Bipolar staining and pleomorphism
* Non motile, capsulated


gettyimages




Bipolar staining: Dark stained **bipolar** ends in Wright's stain (blood from plague victim)

*Epidemiology of Plague

* Plague pandemics
  * 1st - AD 541 - Roman Emperor Justinian
  * 2nd - 14th century - killed 1/3 of Europe
  * 3rd - 1894 - HongKong
* Timeline of plague in India
  * 1896 - 1918 - HK pandemic entered India
  * 1918-1967 - declined, occasional cases
  * 1967-1994 - no cases
  * 1994 (Surat, Guj) - pneumonic plague
  * 2002 - Shimla
  * 2004 - Uttarkashi, Uttaranchal

*Epidemiological factors

* Reservoir - Wild rodents
* Source of infection - infected wild rodents, rat fleas and cases of pneumonic plague
* Vector - Rat flea (Xenopsylla cheopis)
* Plague cycles-
  * Domestic cycle
  * Wild or sylvatic cycle
* Mode of transmission -
  * Bite of infected rat flea
  * Direct contact with tissues of infected rodent
  * Droplet inhalation - from cases
* Blocked flea
* Cheopis index
* Seasonality - North India - (Sept - May), South - all year

*Virulence factors of Y.pestis

* Fraction1 F1 antigen
* Phospholipase D
* Surface proteases
* pH 6 antigen
* LPS
* Pigmentation
* Low calcium respond plasmid
* Siderophore

*Human plague

* 3 clinical forms
  * Bubonic plague (MC)
  * Pneumonic plague
  * Septicemic plague

* Transmitted - infected rat flea bite
* Bacilli pass thru local lymphatics and reach local lymph nodes
* IP is 2-7 days
* C/F - sudden onset fever, malaise, headache, painful lymphadenitis
* Buboes - regional LN tense, tender called buboes. MC - inguinal, crural, axillary, etc
* Bubonic plague - cannot spread
* Complications - dissemination, pneumonia and meningitis

*Bubonic plague

*Pneumonic plague

* Primary pneumonic plague - inhalation droplets
* IP - short, 1-3 days
* CF - suddent onset fever, headache, productive cough, hemoptysis, dyspnea, chest pain
* Highly infectious and fatal
* Bioterrorism - aerosolised Y.pestis

* Primary - Rare, accidental lab infections
* Secondary – due to spread of bubonic or pneumonic plague
* IP – 2-7 days
* Massive involvement of blood vessels – skin hemorrhages – gangrene – BLACK DEATH

* Septicemic plague

*Laboratory Diagnosis

* Specimen collection
  * Bubonic plague - pus, fluid aspirated from buboes
  * Pneumonic plague - sputum, blood
  * Septicemic plague - blood, splenic aspirate(PM)
* Transport medium - Cary-Blair
* Direct Microscopy -
  * Gram stain - PC and GN oval coccobacilli with rounded ends, capsulated
  * Wayson stain / Meth blue- bipolar/safety pin

* Culture
* Aerobic, facultative anaerobic, non fastidious
* Optimum temp - 27 deg C, Capsule - 37deg C
* Blood agar - NH, dark brown pigmented
* Mac - NLF
* Nutrient broth - granular turbidity
* NB with oil or ghee - stalactites like growth
* CIN agar - Cefsulodin, Irgasan, Novobiocin - sputum sample

* Biochemicals
  * Sugars - glucose, mannitol, maltose - acid, no gas
  * Catalase positive, oxidase negative
  * MR positive, VP negative
  * Indole, Urease, Citrate - negative
* Animal inoculation
  * Using 2 guinea pigs / white rats
* Molecular methods
  * PCR

*Treatment

* Gentamicin – current recommendation
* Doxycycline and Chloramphenicol - also effective
* Beta lactams and macrolides – not recommended

Prevention of plague

- Control of cases
- Control of fleas - DDT, BHC
- Control of rodents
- Chemoprophylaxis - Doxy, tetracycline - DOC
- Vaccine - only for prevention of anticipated outbreak
  - Formalin killed vaccine - Sokhey's modification of original Haffkine vaccine
    - s/c, 2 doses, 4 weeks apart, booster> 6 months
    - CI - infants<6 months
    - Protection - 6 months
    - No protection against - pneumonic plague
  - Live attenuated vaccine - strain EV76
  - Subunit recombinant F1 vaccine under trial

*Yersiniosis

* Zoonotic infection –
* Y.enterocolitica, Y.pseudotuberculosis
* Pigs and other wild and domestic animals - host
* Human infection - raw pork, milk, etc
* Abdominal pain, ocassional diarrhea
* Y.enterocolitica - northern Europe, America
* Y.pseudotuberculosis - Finland

* Common virulence factors
* Invasin
* Yersinia adhesin A
* Ail protein

* Y.enterocolitica
  * Myf antigen
  * pH6 antigen
* Y.pseudotuberculosis
  * Super antigen

*Clinical manifestations

* Self limited GE
* Intestinal complications – older children, pseudoappendicitis
* Septicemia – adults,
* Post infective phenomena
  * Reactive arthritis
  * Erythema nodosum
  * Graves' disease
  * Super Ag – mitogen – Kawasaki's disease

*Laboratory diagnosis

* Culture isolation
* From blood - BHI broth
* From LN aspirate - BA, MAC, NA
* From feces, food, soil - DCA, MAC, CIN Agar(dark red, bull's eye colony)
* Incubation - at 25 deg C and 37 deg C
* Cold enrichment

* Biochemical tests
* Differential motility - motile at 22degC (not at 37degC)
* Cold enrichment - growth improves at 4deg C
* Urease positive
* Sugar fermentation
  * Sucrose, cellobiose, sorbitol - Y.enterocolitica
  * Rhamnose, salicin, melibiose - Y.pseudotuberculosis
  * VP, Ornithine decarboxylase - only Y.enterocolitica

* Serology
  * Ab detection - agglutination, ELISA
* Treatment
  * Diarrhea, self limiting
  * Fluoroquinolone
  * 3rd gen cephalosporin

*Pasteurella

*Pasteurella

* Harbour respiratory tracts of many animals
* *Pasteurella multocida* - MC organism in dog/cat bite wounds
* *P.haemolytica* and *P.pneumotropica* rarely infect humans
* *P.aviseptica* - chicken cholera bacillus

* Clinical findings
* Following animal bite - area - red, swollen, painful - variable regional LN and low grade fever
* Pasteurella - commensal in human respiratory tract
* Infection following trauma / Sx leading to bacteremia
* Meningitis, appendicitis, Chronic respiratory infection

*Laboratory diagnosis

* Direct Microscopy - non motile GNCB with a bipolar staining
* Culture - aerobes or facultative anaerobes
  * Grow readily on ordinary media
  * Resemble Yersiniae but,
  * Oxidase - positive
  * Indole - positive
  * MAC - No growth
* Treatment - Penicillin G for *Pasteurella multocida* infections

*Francisella

* F.tularensis - causative agent of Tularemia - plague-like disease of rodents and other small animals
* Epidemiology
* Source - persists in contaminated environments, insects, animal carriers
* Transmission -
  * Blood sucking insects- ticks
  * Contact with wild or domestic animals
  * Ingestion of contaminated water or food
  * Inhalation of infective aerosols

* Francisella

* 4 subspecies
* subsp.tularensis – most virulent, North America

Clinical Manifestations

- Tularemia has various clinical syndromes
  - Ulceroglandular tularemia - MC form
  - Pulmonary tularemia - aerosol - Lab worker
  - Oropharyngeal tularemia - undercooked meat, membranous pharyngitis with cervical lymphadenopathy
  - Oculoglandular tularemia - purulent conjunctivitis
  - Typhoid-like illness
- Bioterrorism - Category A agent of bioterrorism

*Laboratory diagnosis

* Culture - highly fastidious
  * BCG agar - Blood cysteine glucose agar
  * CHAB agar - cysteine heart agar with 9% heated sheep blood
* Sample - ulcer scraping, LN biopsy, gastric washing, sputum, blood - 37deg C - 2-4 days
  * Colonies - blue gray, round, smooth, slightly mucoid
  * BSL III - mandatory

*Identification

* *F.tularensis* - small, GNCB, bipolar appearance, non motile, capsulated
* Weakly catalase positive, oxidase negative, H2S positive
* Antibody detection - Agglutination tests and ELISA tests
* Molecular - PCR assay - specific genes encoding OMP
* Treatment - Gentamicin - DOC- 5mg/kg x 7-10 days

*Thank you!!!