

Acute Bacterial Meningitis (aka Pyogenic Meningitis)

Meningitis: infection of the brain & spinal cord with involvement of the subarachnoid space.

- acute purulent infection within subarachnoid space
- characterized by elevated polymorphonuclear cells in CSF
- *Streptococcus pneumoniae* ⇒ most common cause (50%)
- *Meningococcus* (25%), *Streptococcus agalactiae* (15%), *Listeria* (10%), *Haemophilus influenzae* (<10%)
- Common agents of neonatal meningitis include: *Streptococcus agalactiae*, *E. coli*, *Klebsiella*, *Listeria monocytogenes*
- In elderly (>60 yrs): *Strep. agalactiae*, *Listeria monocytogenes*

- Pathogenesis:** transmitted from person-to-person through droplets/aerosols from active cases or nasopharyngeal carriers.
- Organisms gain access to meninges by several routes:
 - hematogenous spread: (most common route) entry into subarachnoid space is gained through the choroid plexus or through other blood vessels of the brain.
 - Direct spread from infected site present close to meninges – otitis media, mastoiditis, sinusitis, etc.
 - Anatomical defect in CNS: which occurs as a result of surgery, trauma, congenital defects.

Predisposing Factors:

- **Age:** Neonates have the highest prevalence due to:
 - immature immune system
 - acquiring the colonized organisms from mother's birth canal
 - increased permeability of blood-brain-barriers
- **Vaccination:** decreased incidence of *H. influenzae* meningitis following implementation of Hib vaccination.
- **Factors that promote infection at primary site:**
 - respiratory tract is the primary portal of entry for many agents of meningitis, the factors that predispose to respiratory infection can also increase the likelihood of meningitis – alcoholism, diabetes, immunosuppression, splenectomy.
- **Presence of CSF Shunts**
- **Breach in the Blood-Brain-Barrier [BBB]**

Clinical Manifestations:

- incubation period = 2-10 days
- Important symptoms: fever, vomiting, intense headache, altered consciousness, photophobia.
- Signs of Meningism (meningeal irritation):**
 - **Nuchal Rigidity (Stiff Neck):** present when the neck resists passive flexion
 - **Kernig's Sign:** severe stiffness of hamstring causes an inability to straighten the leg when the hip is flexed at 90°
 - **Brodzinski's Sign:** when the neck is passively flexed, it results in spontaneous flexion of the hips & knees
 - **In Infants:** slower onset of disease
 - neck stiffness may not be present
 - fever, irritability, bulging fontanelle
 - **Complications:** disease may involve brain parenchyma leading to meningoencephalitis – decreased consciousness, seizures, raised ICP, stroke.

- **Organ Specific Finding:** purpuric rashes seen in meningococcal meningitis.

Laboratory Diagnosis:

- **CSF Collection:** obtained by lumbar puncture; it is divided into 3 sterile containers – one each for cell count, biochemical analysis & bacteriological examination.
- **CSF Transport:** CSF should be examined immediately
 - CSF should never be refrigerated as delicate pathogens such as *H. influenzae*, pneumococci, meningococci may die. [it may be kept in an incubator at 37°C].
- for molecular diagnosis, CSF can be kept in freezer.

Other Specimens:

- Blood culture – collected in automated bottles such as Bact/ALERT
- For suspected meningococcal meningitis:
 - nasopharyngeal swabs, pus, scrapings from rashes
 - carried in transport (Stuart's) media
 - inoculated onto selective media (Thayer Martin medium or New York City medium) to suppress the growth of normal flora.
- In acute bacterial meningitis:
 - CSF usually contains >1000 leukocytes/ μ L & predominantly neutrophils (90-95%) [In *Listeria* meningitis, there is increased lymphocyte in CSF]
 - total protein content is elevated & glucose level is markedly diminished or even absent.
 - CSF pressure is highly elevated.

CSF Microscopy:

- **Heaped Smears:** as the bacterial load in CSF may be very low, to increase the sensitivity, several drops of CSF should be placed at the same spot on the slide, each drop being allowed to air dry before the next is added.
- **Centrifugation:** Alternatively, CSF can be centrifuged & the deposit is examined for gram staining
- Direct Antigen Detection:** after centrifugation of CSF, the supernatant is used for antigen detection.
- Latex agglutination test is performed using latex beads coated with anti-capsular antibodies
- From Urine:** Antigen detection in urine is useful for pneumococcal meningitis; immunochromatographic test (ICT) is used to detect C-polysaccharide antigen of *S. pneumoniae* in urine.

- Culture:** ideal media – chocolate agar, blood agar, MacConkey agar.
- **Enriching:** ∵ bacterial load is very low, a part of the CSF can be inoculated into enrichment media at bedside or BHI broth in the laboratory.
- **Culture plates** are incubated at 37°C; preferably in candle jar (provides 5% CO_2) for 48 hours.
- **Identification** of the organism by automated systems such as MALDI-TOF or VITEK or by conventional biochemical tests.
- **Antimicrobial Susceptibility Test** must be done to initiate definitive treatment; carried out by disk diffusion or by automated MIC-based methods such as VITEK.
- Sensitivity of culture drops rapidly in case of prior antimicrobial therapy or delay in processing.
- Serology:** antibodies to capsular antigens of meningococci can be detected in patient's serum by ELISA.

Molecular Methods: highly sensitive, detects even few bacteria in CSF with less turnaround time than culture

- Multiplex PCR & multiplex rt-PCR
- BioFire FilmArray (automated nested multiplex PCR)
- Common genes targeted include:
 - *Meningococcus*: *ctrA* (capsule transport gene) & *sodC* [Cu-zn superoxide dismutase gene]
 - *Pneumococci*: *lytA* (autolysin gene), *ply* (pneumolysin), *psaA* (pneumococcus surface antigen A).
 - *H. influenzae*: conserved capsular gene *bexA*

Treatment: mortality is very high (20% for pneumococci)

Empirical Therapy:

- **Adult:** IV cefotaxime or ceftriaxone & Vancomycin [if *Listeria* is suspected, IV ampicillin can be added]
- **For Neonates:** IV ampicillin + gentamycin
- IV dexamethasone is added to reduce ICP.

Definitive Therapy: alteration of empirical therapy based on antimicrobial susceptibility pattern.

Identification of Agent Based on CSF Gram Stain:

Appearance in CSF gram stain	Suggestive Agent
• Gram +ve diplococci, flame or lancet-shaped with clear halo (capsulated)	→ <i>Streptococcus pneumoniae</i>
• Gram -ve diplococci, capsulated, lens or half-moon shaped	→ <i>Neisseria meningitidis</i>
• Pleomorphic gram -ve coccobacilli, capsulated	→ <i>Haemophilus influenzae</i>
• Gram -ve bacilli, arranged singly	→ <i>E. coli</i> or others
• Gram +ve cocci in short chains	→ <i>Streptococcus agalactiae</i>
• Gram +ve short bacilli, often confused with diphtheroids	→ <i>Listeria monocytogenes</i>

Cytological & Biochemical Parameters:

Characteristic	Normal Individual	Pyogenic Meningitis	Tuberculous Meningitis	Viral Meningitis
CSF pressure (mm of H_2O)	Normal (50-150)	>180 (highly elevated)	Moderately elevated	Slightly elevated / Normal
TLC (per mm^3)	0-5	100-10,000	10-500	25-500
Predominant Cell Type	Lymphocytes	Neutrophils	Lymphocytes	Lymphocytes
Glucose (mg/dl)	40-70	<40 (decreased/absent)	20-40	Normal
Total proteins (mg/dl)	15-45	>45 (usually >250; markedly increased)	100-500 (moderate to markedly increased)	20-80 (normal or slightly increased)

Chronic Bacterial Meningitis

- **CSF findings:** Lymphocytes (100/ μ L), elevated protein level & normal/slightly low glucose level
- **Meningopolyneuritis:** radicular pain with CSF pleocytosis, without meningeal or encephalitic signs (**Bannwarth's syndrome**)
- **Treatment:** IV ceftriaxone for 14-28 days (alternatively, IV cefotaxime or IV penicillin G)
- **Neurosyphilis (*Treponema pallidum*):** tertiary form of syphilis (STD)
- **Manifestations:** meningeal syphilis, meningovascular syphilis (vasculitis of arteries leading to embolic stroke)
 - **general paresis** of insane: defects in personality, affect, reflexes (hyperactive), eye (Argyll Robertson pupils), sensorium (illusions, delusions, hallucinations), intellect (recent memory loss), speech.
 - **Tabes dorsalis:** features of demyelination of posterior columns such as ataxia, foot drop, paraesthesia, bladder disturbances & impotence
- **Laboratory Diagnosis:** CSF shows lymphocytes (>5/ μ L) & increased protein level (>45 mg/dL)
- **Treatment:** aqueous crystalline or procaine penicillin G is given for 10-14 days
- consider re-treatment if non-treponemal titres in CSF do not decrease by 4 folds within 2 years of completion of Tx.
- in patients with penicillin allergy, desensitization to penicillin has to be done, following which penicillin is administered.

Clinical Manifestations:

- persistent headache & neck or back pain / stiffness
- **Hydrocephalus:** accumulation of CSF within the ventricles causes increased ICP
- **Cranial neuropathies:** leads to facial weakness, diminished vision, papilledema, optic atrophy, hearing loss
- **Myelopathy or Radiculopathy:** leads to arm or leg weakness or numbness, urinary retention / incontinence
- **Changes in personality:** altered mental status – drowsiness, inattention, disorientation, memory loss

Agents of Chronic Meningitis:

- **Tuberculous Meningitis:** TBM results from the hematogenous spread of primary or post-primary pulmonary TB.
- **Neuroborreliosis (Lyme Disease):** (*Borrelia burgdorferi*, transmitted by tick bite)
 - Presentation: cutaneous lesions, arthritis, meningitis, subtle encephalitic signs, cranial neuropathies, radiculoneuropathy.

Agents of Pyogenic Meningitis

• **Pneumococcal Meningitis:** *Streptococcus pneumoniae* (or pneumococcus)

→ principle virulence factors: capsular polysaccharide, C-carbohydrate antigens, pneumolysin, autolysin.

• **Meningococcal Meningitis:** *Neisseria meningitidis* (meningococcus) → capsulated, gram -ve, diplococci.

→ Virulence factors: capsular polysaccharide is the principle virulence factor which protects *Neisseria* from complement-mediated phagocytosis

- outer membrane proteins (pore proteins present beneath the capsule)
- LPS & endotoxins

• IgA proteases, transferrin binding proteins.

Group A: leading cause of epidemic meningitis worldwide

Group B & C: currently major serogroups causing invasive disease worldwide

→ Age: common in early childhood (3 months - 5 years) with a 2nd peak in adolescents (15-25 years).

Clinical Manifestations:

• Rash (non-blanching; petechial or purpuric)

• Septicemia (leading to increased vascular permeability & intravascular thrombosis)

• **Waterhouse-Friedrichsen Syndrome:** severe form of fulminant meningococemia with large purpuric rashes (purpura fulminans), shock, DIC, bilateral adrenal haemorrhage & multiorgan failure.

• Pyogenic meningitis (3-5 years of age): fever, vomiting, headache, neck stiffness.

• Chronic meningococemia (repeated episodes of petechial rashes, fever, arthritic, splenomegaly)

• Post meningococcal Reactive Disease (arthritis, rash, iritis, pericarditis, polyserositis, fever)

Treatment: Drug of choice - 3rd generation cephalosporins given for > 7 days (alternative: penicillin)

→ Symptomatic treatment: aggressive fluid resuscitation for shock, measures to decrease ICP should be instituted.

Vaccine Prophylaxis: bivalent/quadrivalent

• Schedule: administered as 2 doses, 2-3 months apart to children of 3-18 months or as a single dose to older children or adults

• Indications: - contacts of patients during outbreaks

- splenic dysfunction - terminal complement component deficiency - taking eculizumab therapy - laboratory staff at risk - international travellers.

• Not given below 3 years (meningococcal capsular vaccine is T-cell independent antigen & is poorly immunogenic to children)

• Conjugated meningococcal capsular vaccine is available which can be given to young children.

Vaccine for Group B (menB vaccine): Recombinant vaccine for group B meningococcus has been licensed.

• Schedule: 2 doses given 1m one month apart

• Indication: 16-25 years of age

• **Group B Streptococcal (*S. agalactiae*) Meningitis:**

(major cause of neonatal sepsis & meningitis)

→ Treatment: penicillin for 14 days (shorter courses may lead to relapse)

• **Listeriosis:** *Listeria monocytogenes* (food-borne)

→ common in neonates & in the elderly

<i>Neisseria meningitidis</i>	<i>Neisseria gonorrhoeae</i>
→ capsulated	→ non-capsulated
→ lens-shaped / half moon-shaped (diplococci)	→ kidney-shaped (diplococci)
→ ferments glucose & maltose	→ ferments only glucose
→ rarely have plasmids	→ usually possess plasmids, coding for drug resistance genes
→ exist in both intra & extracellular forms	→ predominantly intracellular form
→ Habitat: nasopharynx	→ Habitat: genital tract (rarely pharynx)

Group-B Streptococcus Disease in Neonates:

Characteristics	Early-onset Disease	Late-onset Disease
Age of onset	0-6 days of birth	7-90 days of birth
Increased risk following obstetric complications	Prematurity & prolonged labor	Not associated
Mode of transmission to the baby	During or before birth from the colonized maternal genital tract	Contact with a colonized mother & nursing personnel
Clinical manifestations	Pneumonia, RDS, bacteraemia, meningitis	Meningitis & bacteraemia
Common serotypes	Ia, III, V, II, Ib	III
Case fatality rate	4-7%	2-8%

Neonatal Listeriosis:

Early Onset	Late Onset
• occurs < 5 days of birth • acquired from maternal genital flora • associated with obstetric complications like premature delivery & low birth weight • most common form is neonatal sepsis • Granulomatosis infantiseptica: occurs rarely, characterized by milium micro-abscesses & granulomas, mostly in skin, liver & spleen • Mortality rate is up to 50% • does not cause nosocomial outbreaks	• occurs > 5 days of birth • acquired from environment • Not associated • Most common form is neonatal meningitis • Not seen • Mortality rate < 10% • Nosocomial outbreaks are seen.

Viral Meningitis

Definition: inflammation of subarachnoid space due to viral etiology

Etiology: Enteroviruses → most common cause

• Herpes virus • Arbovirus • Mumps virus • HIV

• LCM (Lymphocytic choriomeningitis virus)

Epidemiology: can affect people of any age

→ Higher risk groups - children < 5 years of age

- elderly

- immune compromised individuals

Transmission: close contact with infected patients is necessary for transmission to set in

Clinical Manifestations:

- fever
- headache (frontal/retro-orbital)
- stiff neck (milder than bacterial meningitis)
- photophobia
- sleepiness or trouble in waking up from sleep
- nausea, vomiting
- irritability
- lack of appetite
- lethargy
- [no seizures/focal neurological signs/altered consciousness/confusion]

Laboratory Diagnosis:

• **CSF analysis:**

→ normal/slightly elevated protein level (20-80 mg/dL)

→ normal glucose level

→ mildly elevated CSF pressure

→ cell count ⇒ 25-500/ μ L (in some viral meningitis, several thousands/ μ L may be seen)

→ **Pleocytosis:** lymphocytes are predominant cell type, although neutrophils may predominate in first 48 hours of illness of some viral meningitis.

• **Molecular Methods:** Gold Standard Method

→ amplification of viral RNA/DNA from CSF by PCR

→ PCR of throat washings or stool samples for enterovirus infections

→ Multiplex PCR & multiplex real-time PCR for simultaneous detection of common agents of viral meningitis

→ BioFire Film Array

• **Viral Culture:** sensitivity of CSF cultures for diagnosis of viral meningitis is generally poor.

• **Antibody Detection:** important for the detection of less prevalent arboviruses [West Nile virus]

• **Oligoclonal gamma globulin bands:** may be detected in CSF in many viral infections.

Treatment: primarily symptomatic (use of analgesics, antipyretics, antiemetics, fluid & electrolyte replacement)

→ Oral/i.v. acyclovir may benefit patients

Agents Causing Viral Meningitis:

Non-polio enteroviruses ⇒ Most common cause

→ Picornaviridae
 Rhinoviruses: cause common cold
 Enterovirus → transmitted by feco-oral route but does not have any intestinal manifestations.

→ Polioviruses

↓ cause

Myelitis

→ other agents: Coxsackieviruses, echoviruses, parechoviruses

• Enterovirus 71 - cause aseptic meningitis.

{ Enterovirus 68 : pneumonia
 Enterovirus 70 : acute haemorrhagic conjunctivitis
 Enterovirus 72 : Hepatitis A }

Clinical Manifestation of Coxsackieviruses:

→ Coxsackieviruses spread through an infected person's nasal & throat secretions, fluid from blisters/scrabs

• **Aseptic Meningitis:** caused by all group B & some group A coxsackieviruses.

• **Herpangina:** severe febrile vesicular pharyngitis caused by certain group A viruses

• **Hand-Foot & Mouth Disease:** characterized by oral & pharyngeal ulcerations & vesicular rashes of the palms & soles which heal without crusting.

• **Pleurodynia:** fever & abrupt onset of stabbing chest pain.

• **Myositis & Pericarditis:** by coxsackie group B.

• **Acute haemorrhagic conjunctivitis:** self-limiting sub-conjunctival haemorrhage.

• **Pneumonia:** may be caused by coxsackievirus B4 & B5.

• **Generalised Disease of infants**

• **Pancreatitis**

Group A Coxsackievirus	Group B Coxsackievirus
Suckling mouse intracerebral inoculation	
• Flaccid paralysis	• Spastic paralysis
• Generalised myositis	• Focal myositis
Manifestations	
• Aseptic meningitis	→ More organ involvement seen:
• Herpangina (vesicular pharyngitis)	• aseptic meningitis
• Hand-foot & mouth disease	• pleurodynia
• acute haemorrhagic conjunctivitis	• Myocarditis, pericarditis
	• Pancreatitis
	• Pneumonia
	• Generalised disease of infants

Laboratory Diagnosis:

→ Specimen collection (throat swabs, stool, CSF)

→ Isolation of virus by intracerebral inoculation into suckling mice.

→ PCR → serology

Echoviruses: enteric cytopathogenic human orphan viruses

→ transmitted by feco-oral route

→ associated with aseptic meningitis, encephalitis, rash, common cold, ocular disease.

HSV Meningitis: Adults are more commonly affected than children

→ history of genital herpes is an important clue

→ HSV typically produces a chronic recurrent lymphocytic meningitis called **Mollaret Meningitis**.

→ other neurological manifestations:

• HSV encephalitis • Bell's palsy • Autonomic system involvement (sacral region)

• transverse myelitis (symmetric paralysis of lower extremities)

• Guillain-Barré Syndrome

• Peripheral Nervous system involvement

→ Other Herpesviruses:

• **VZV meningitis:** suspected in the presence of concurrent chickenpox or shingles (can occur in patients without h/o rash)

• **EBV meningitis:** h/o associated infectious mononucleosis & presence of atypical lymphocytes in CSF or peripheral blood.

LCM Virus Meningitis: affects people with h/o contact with rodent droppings or urine; associated rash, pulmonary infiltrates, alopecia, parotitis, orchitis, myopericarditis.

Rabies

Definition: rapidly progressive, acute infectious disease of the CNS transmitted from a rabid animal.

Viral Morphology: Rhabdoviridae family

↳ Bullet-shaped (width = 75 nm, length = 180 nm)

→ lipid envelope in which 10 nm long peplomers or spikes (glycoprotein-G) are embedded

→ envelope is lined internally by a layer of matrix protein.

→ Nucleocapsid: helical symmetry & comprises a single-stranded, negative-sense RNA

→ Antigens: Glycoprotein-G, nucleocapsid

Street Viruses: freshly isolated strains in the laboratory

→ mimic the wild viruses

→ long & variable incubation periods & produce intracytoplasmic inclusion bodies.

Fixed Viruses: When street viruses are propagated in rabbits by serial brain-to-brain passage, they lose certain properties & become fixed strains

• do not produce inclusion bodies

• do not multiply in extraneural tissues

• do not infect salivary gland

• multiply rapidly, incubation period is shortened to 4-6 days ∴ these strains are best used for vaccination.

Pathogenesis:

• **Bite Transmission:** following a deep bite or scratch from an infected rabid animal (rabid dogs, bats, jackals, foxes, raccoons, skunks)

• **Non-bite exposure:** direct contact with saliva of infected animals

• Inhalation of virus-containing aerosols

• cornea or other organ transplantation.

• **Spread of the Virus:**

→ virus starts replicating locally at the site of inoculation in muscle or in connective tissue

→ virus binds to nicotinic acetylcholine receptors present at neuromuscular junctions

→ virus spreads centripetally along the peripheral motor nerves via retrograde fast axonal transport

→ it reaches dorsal root ganglia of the spinal cord & then ascends upwards to CNS

→ it rapidly disseminates to various parts of CNS (mainly hippocampus & cerebellum)

→ from CNS, virus spreads centrifugally along sensory & autonomic nerves to various tissues such as salivary glands, pancreas, kidney, heart, retina, cornea.

→ rabies virus is shed in the saliva of rabid animals which acts as the source of infection to other animals (viral shedding occurs in human saliva but human-to-human transmission is not yet documented)

→ **Negri bodies** (intracytoplasmic eosinophilic inclusions) composed of rabies virus proteins & viral RNA, in the brain parenchyma is an important pathological finding of rabies.

Clinical Manifestations: incubation period = 20-90 days (it is directly related to the distance for the virus to travel from the site of inoculation to CNS)

• **Prodromal Phase:** 2-10 days

↳ non-specific symptoms such as fever, malaise, anorexia, nausea, vomiting, photophobia, sore throat, paraesthesia, pain, pruritis around the wound site.

• **Acute Neurologic Phase:** may be either encephalitis type (80%) or paralytic type (20%)

Encephalitic / Furious Rabies: 2-7 days

• **Hyperexcitability:** anxiety, agitation, hyperactivity, hallucinations

• **Lucid interval:** period of hyperexcitability is typically followed by complete lucidity that becomes shorter as the disease progresses

• **Autonomic (Sympathetic) Dysfunction:**

↑ lacrimation, ↑ salivation (leading to foaming of the mouth), ↑ perspiration, gooseflesh, cardiac arrhythmias, peristalsis.

• **Hydrophobia or Aerophobia:** act of swallowing precipitates an involuntary, painful spasm of the respiratory, laryngeal & pharyngeal muscles due to dysfunction of infected brainstem neurons.

Paralytic / Dumb Rabies: occurs in people who are partially vaccinated or infected with bat rabies virus

→ flaccid paralysis in the bitten limb progresses to quadriplegia with facial paralysis

→ hydrophobia absent.

• **Coma & Death:** patient develops coma that eventually leads to death within 14 days

→ patients with paralytic rabies may survive longer up to 30 days

→ But death is almost certain.

Laboratory Diagnosis:

• **Antigen detection** - from hair follicles at nape of the neck & from corneal smear by direct immunofluorescence test (direct-IF) a.k.a. direct fluorescent antibody (DFA) test.

"Gold standard"

• **Viral Isolation by:**

Mouse inoculation: intracerebral inoculation into suckling mice can cause encephalitis & death - brain biopsies of the inoculated animal shows presence of Negri bodies & rabies antigen.

Cell lines inoculation: mouse neuroblastoma & BHK cell lines (Baby Hamster Kidney)

• **Antibody Detection:** from serum & CSF

- serum antibodies appear late & can also be present after vaccination

- CSF antibodies appear early & are produced only in rabies-infected individuals.

→ **Tests** - Mouse neutralisation Test

- Rapid fluorescent focus Inhibition Test

- Fluorescent antibody virus Neutralisation Test

- Indirect fluorescence Assay.

• **Viral RNA detection:** by RT-PCR

• **Negri Body Detection:** useful to confirm postmortem diagnosis of rabies

→ intracytoplasmic eosinophilic inclusion with basophilic inner granules

→ commonly observed in Purkinje cells of cerebellum & in pyramidal neurons of hippocampus.

→ **Stains:** H&E, Sellers stain

→ **Immunohistochemistry:** peroxidase labeled specific antibodies are used to detect viral inclusions in formalin-fixed tissues

Treatment: no specific Tx; symptomatic Tx may prolong life

→ patient should be isolated in a quiet room, protected from external stimuli like noise, water, light, cold air which can precipitate spasms

→ Sedatives & anti-anxiety drugs (eg. morphine)

→ hydration & urination should be properly maintained.

Prognosis: mortality in rabies ≈ 100%; it is preventable by administration of post-exposure therapy during early incubation period.

→ prevented by post-exposure prophylaxis (PEP) & pre-exposure prophylaxis (PrEP)

PEP (For those not received PEP/PrEP previously): - local wound care + rabies vaccine + rabies immunoglobulin (RIG).

• For Category I exposure: only wound care

• For Category II exposure: wound care + rabies vaccine

• For Category III exposure: wound care + rabies vaccine + RIG.

Category	Type of Exposure	Recommended Prophylaxis
Category I (no risk)	- touching/feeding of animal - licks on intact skin	no treatment needed if history is reliable
category II (minor risk)	minor scratches or abrasions without bleeding or nibbling of uncovered skin	- wound management - rabies vaccine - observe the dog for 10 days
category III (major risk)	- single/multiple transdermal bites with oozing of blood - licks on broken skin or mucous membrane - direct contact with bats or wild animals	- wound management - rabies vaccine - rabies immunoglobulin - observe the dog for 10 days

Local Wound Care:

→ **Physical Cleansing:** wash thoroughly with soap & water for 15 minutes.

→ **Chemical inactivation:** use antiseptics such as povidone iodine or alcohol.

→ **Suturing:** it causes local tissue damage which helps the virus to spread ∴ suturing should be avoided

→ debridement of devitalized tissues, tetanus prophylaxis, antibiotic treatment to prevent secondary bacterial infection

Rabies Vaccine: cell-line derived non-neural vaccines are recommended;

→ 3 vaccines are available:

i) purified chick embryo cell (PCEC) vaccine (prepared from chicken fibroblast)

ii) purified Vero cell (PVC) vaccine

iii) Human diploid cell (HDC) vaccine (derived from human embryonic lung fibroblast)

→ Neural vaccines derived from brain of infected animals are encephalitogenic & ∴ no longer used

→ **Route of administration:** i.m. or i.d. (in the deltoid/anterolateral aspect of thigh for children < 2 years) **Never given in gluteal region**

→ **Dose:** ID dose = 0.1 mL

IM dose = entire vial (irrespective of vial size)

→ **ID PEP regimen:** 2-site ID vaccine on days 0, 3, 7.

→ **IM PEP regimen:** Total 4 doses • 1-site IM on days 0, 3, 7 & fourth dose between days 14 or 28 or

• 2-site IM on day 0 & 1-site IM on days 7 & 21.

Rabies Immunoglobulin (RIG): provides passive immunization by neutralizing the virus.

→ should be administered only once, as soon as possible, not beyond day 7 after first dose of vaccine.

→ available in two forms - human RIG (hRIG) & equine RIG (eRIG).

→ maximum dose = 20 IU hRIG / 40 IU eRIG per kg body weight (no minimum dose)

→ it should be infiltrated as much as possible into the wound

PEP [for those previously vaccinated - PEP/PrEP]: RIG not required

→ local wound care & vaccination consisting of any one of the following:

• 1-site ID on days 0 & 3.

• 1-site IM on days 0 & 3.

• 4-site ID on day 0 only. (left & right deltoid, thigh or supracapular)

PEP:

For individuals at higher occupational risk: laboratory staff handling the virus & infected material, clinicians attending to human rabies cases, veterinarians, animal handlers & travellers to endemic areas.

PEP Regimen: Two schedules are available:

- 2 site ID given on days 0 & 7
- 1 site IM given on days 0 & 7
- Boosters: not required periodically.

Epidemiology: rabies is an enzootic & epizootic disease

- rabies is endemic in >150 countries
- all age groups are affected; children aged 5-15 years are at a greater risk.

Rabies in Dogs:

- incubation period = 3-8 weeks

Clinical Manifestations: 2 types:

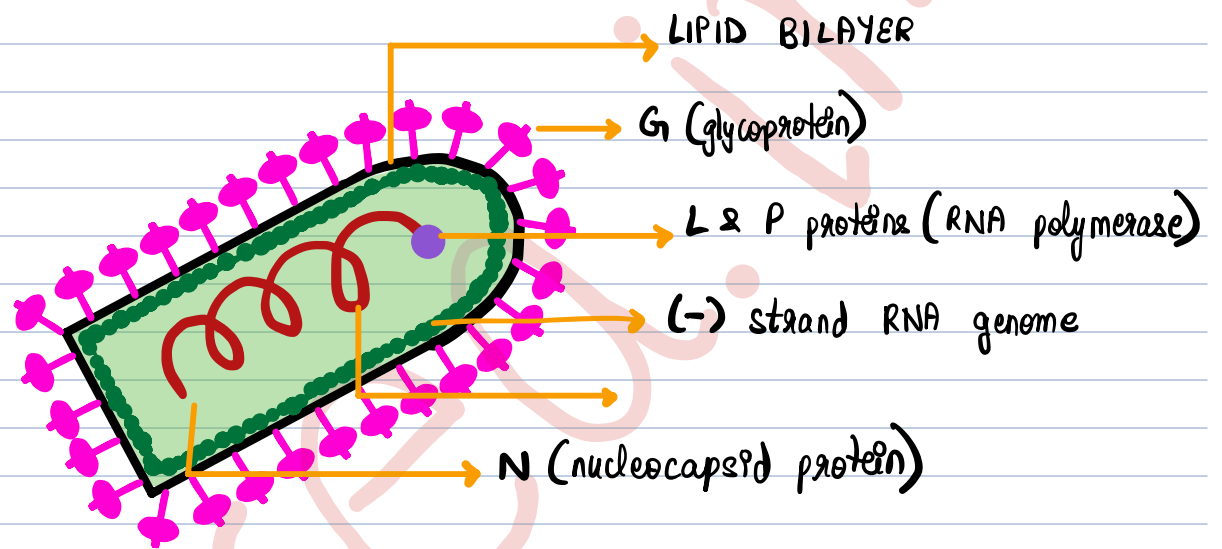
i] Furious Rabies / Mad Dog Syndrome

ii] Dumb / Paralytic Rabies:

Control of Rabies in Dogs: elimination of stray dogs & mass immunization of at least 80% dogs in an area

- vaccines commonly used in dogs are:

- BPL inactivated neural vaccine
- Oral recombinant glycoprotein vaccine.



Major Antigens of Rabies Virus:

Glycoprotein G	Nucleoprotein
→ polymers or spikes embedded in lipid envelope	→ capsid proteins associated with viral RNA
→ species-specific	→ group-specific
→ it binds to Ach receptors in neural tissues which is the 1st step of pathogenesis	→ does not have any role in pathogenesis.
→ used in haemagglutination inhibition test to detect Ab in patient's serum	→ Ag detected in patient's serum by direct - IF test
→ it induces neutralizing antibodies which are protective	→ antibodies are not protective.
→ purified form of the antigen can be used in subunit vaccine	→ Not used for vaccination.

Slow Virus & Prion Disease

- a group of neurodegenerative conditions
- long incubation period (months to years) because of long doubling time
- Predisposition for CNS, invariably fatal
- strong genetic predisposition
- they lack antigenicity ∴ there is lack of immune response & lack of associated inflammation.

Slow Virus Diseases: (In humans)

- Progressive multifocal leukoencephalopathy (JC virus)
- Subacute sclerosing panencephalitis (measles)
- Progressive rubella panencephalitis.

Prion Disease: (In human)

- Kuru
- Gurtzfeldt - Jakob Disease (CJD)
- Gerstmann - Straussler - Scheinker disease
- Fatal Familial insomnia.

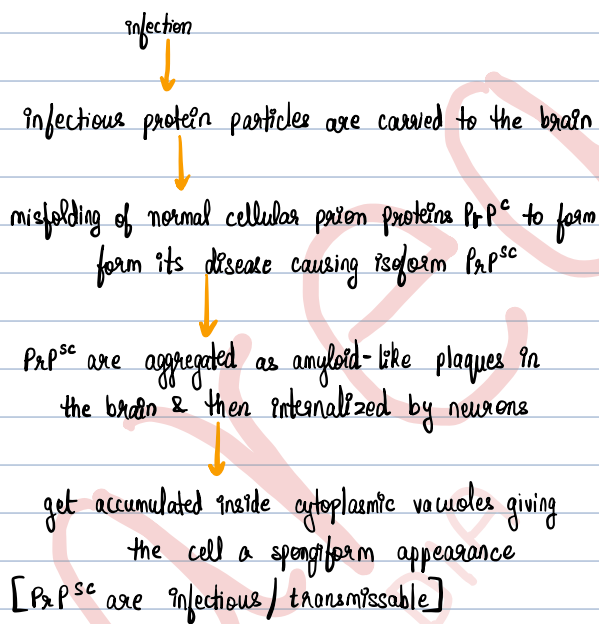
Slow Virus Disease:

- chronic form of encephalitis/encephalopathy (chronic progressive demyelinating disease of CNS)
- Progressive Multifocal Leukoencephalopathy (PML):
 - caused by a polyoma virus called JC virus (John Cunningham)
 - characterized by multifocal areas of demyelination distributed throughout the brain with deformation of both astrocytes & oligodendrocytes.
 - Risk factor: immunodeficient individuals with comorbidities such as AIDS, hematologic malignancy, transplantation, etc.
 - Manifestations: visual deficits, mental impairment, weakness, ataxia, seizures
 - Diagnosis: MRI scan, PCR targeting JC virus DNA (CSF is typically normal)
 - no definitive treatment; cidofovir may be effective
- Measles Encephalitis: Measles produces various CNS complications like Subacute Sclerosing Panencephalitis (SSPE).
 - slowly progressive disease
 - characterized by seizures & progressive deterioration of cognitive & motor functions caused by a defective measles virus
 - Age: children < 2 years of age acquiring the primary measles infection are usually prone to develop SSPE
 - SSPE usually develops after 7-13 years after primary measles infection
 - Manifestations: progressive intellectual deterioration, focal or generalized seizures, myoclonus, ataxia, visual disturbances (fatal within 1-3 years of onset)
 - Diagnosis: CSF analysis shows markedly elevated gamma globulin level (> 20% of total CSF protein), high titres of antibody to measles virus in CSF
 - no effective treatment available.
- Rubella Encephalitis: [Progressive Rubella Panencephalitis]
 - primarily affects males with congenital rubella syndrome (manifests after a latent period of 8-19 years)

Prion Disease:

- infectious protein particles that lack any nucleic acid
- resistant to wide range of sterilants

Mechanism of Prion Disease:



Clinical Manifestations:

- prodromal phase lasts for 3-5 months
- manifestations: loss of muscle control, shivering, myoclonic jerks, tremors, loss of coordination & rapidly progressive dementia
- death occurs within one year of onset of disease.

Prion Diseases of Animals:

- Scrapie: prototype of prion disease
 - ↳ affects sheep
- Mad Cow Disease/ Bovine Spongiform Encephalopathy (BSE)

Human Prion Diseases:

- Kuru: seen only in Eastern Highlands of New Guinea
 - spread by customs associated with ritual cannibalism of dead relatives infected with the disease.
- Gurtzfeldt - Jakob Disease [CJD]: most common form of prion disease in humans presenting with dementia & myoclonus
 - Types of CJD:
 - i] Classical or sporadic [sCJD]
 - ii] Familial [fCJD]
 - iii] Iatrogenic [iCJD]
 - iv] Variant [vCJD]
 - Laboratory Diagnosis:
 - i] Measurement of PrP^{sc} by conformation dependent immunoassay.
 - ii] Brain biopsies — spongiform degeneration with lack of inflammatory response
 - iii] Sequencing the PRNP gene to identify mutations.
 - iv] Abnormal EEG [high-voltage, triphasic sharp discharges are observed]
 - Treatment: no known effective therapy

Borna Disease:

- Borna disease virus (BDV) is a highly neurotropic virus
- causes neuropsychiatric disorders in horses & sheep
- seen in certain areas of Germany
- disorder is immune-mediated; characterized by deposition of inflammatory cells in the brain

Arboviral Encephalitis:

{arthropod transmitted viruses}

→ Families under arbovirus:

- Togoviridae
- Flaviviridae
- Bunyaviridae
- Reoviridae
- Rhabdoviridae

Japanese Encephalitis:

→ leading cause of vaccine-preventable viral encephalitis in Asia (including India).

→ belongs to family flaviviridae.

→ enveloped virus containing ssRNA.

Epidemiology: JE virus is transmitted by bite of Culex mosquito [C. tritaeniorhynchus & C. vishnu]

→ JE virus infects several non-human hosts; two transmission cycles are predominant:

- Pigs → Culex → Pigs
- Aedes birds → Culex → Aedes birds [Humans are considered as dead end].

→ About 85% of cases occur in children below 15 years

→ infection is common in rainy season

→ JE is endemic in Southeast Asia region.

Clinical Manifestations: incubation period: 5-15 days.

↳ Subclinical infection is common; it typically shows iceberg phenomenon [cases are much less compared to subclinical/inapparent infections]

Clinical Course: 3 stages -

- Prodromal stage: febrile illness
- Acute encephalitis stage: acute onset of fever, mental confusion, disorientation, delirium, seizures, or coma
- Late stage & Sequelae: convalescent stage in which the patient may be recovered fully or retain some neurological defect permanently.

→ Case fatality rate: 20-40%.

Laboratory Diagnosis: • IgM capture antibody (MAC) ELISA

• RT-PCR / qPCR.

Treatment: only supportive measures (no specific antiviral drugs available)

Vaccine Prophylaxis: for children residing in rural area & for individuals living in endemic areas.

• Live Attenuated SA 14-14-2 Vaccine: primary hamster kidney cell line - derived

→ given to children (1-15 years)

→ 2 doses • 1st at 9 months - 12 months
• 2nd at 16-24 months

→ 0.5 ml / dose, s.c., at left upper arm

• Inactivated JE Vaccine: vero cell culture - derived vaccine

→ 2 doses, 4 weeks interval

→ for children > 3 years & adults ≥ 18 years

→ 0.5 ml / dose, i.m.

• Catch up Vaccine: given to all susceptible children up to 15 years during an outbreak or prior to an anticipated outbreak

• Combined Vaccine: genetically engineered vaccine

↳ combines JE (attenuated SA 14-14-2 strain) & yellow fever vaccine.

Nipah & Hendra Viral Encephalitis:

→ Nipah & Hendra viruses are zoonotic paramyxoviruses.

→ fruit bats (flying foxes) are the natural host for both.

→ Hendra virus is confined to horses in Australia

→ Nipah viruses cause infection of pigs in Malaysia

→ Human infection of Nipah virus is emerging especially in Southeast Asia.

Transmission: Hendra virus is transmitted by exposure to infected body fluids & excretions of horses.

→ Nipah virus: after direct contact with fruit bats, pigs, or persons.

(Human-to-human transmission has been reported)

Clinical Manifestations: incubation period = 4-14 days.

↳ encephalitis in humans

→ presents with fever, headache, myalgia, dizziness, drowsiness, altered consciousness, seizures

→ severe cases progress to death within 48 hours & death.

Laboratory Diagnosis: Real-time PCR from throat & nasal swabs, CSF, urine & blood

→ Antibody (IgM & IgG) detection by ELISA

→ these viruses are prone to cause laboratory acquired infections & are classified as biosafety level 4 pathogens.

Treatment: no antiviral drug available

Vaccine: Subunit Vaccine - using Hendra G protein produces cross-protective antibodies against Hendra & Nipah viruses.

Viral Myelitis

Definition of Myelitis: Inflammation of the spinal cord which can result in disruption of the connection from the brain to the rest of the body & vice versa

→ Viral myelitis is the most common infectious cause of myelitis which is of two types:

• **Grey matter Myelitis:** caused by infection of anterior horn of spinal cord

→ acute flaccid paralysis

→ mostly caused by poliovirus

• **Transverse myelitis:** (Leukomyelitis / White matter myelitis)

→ often caused by herpes virus & influenza virus (rarely by HIV & HTLV)

Poliomyelitis

Morphology: Polioviruses are a type of enteroviruses

→ family: Picornaviridae

→ Icosahedral symmetry

→ capsid is composed of 60 subunits, each consisting of 4 viral proteins (VP1 - VP4)

→ single-stranded positive sense linear RNA

Antigenic Types:

Polioviruses
 { Wild polioviruses (cause natural disease)
 Vaccine derived poliovirus (VDPV) which are vaccine strains that have regained neurovirulence

WPV: 3 strains (WPV1, WPV2, WPV3)

→ these 3 strains cause identical illness but are genetically & immunologically distinct & differ from each other in VP1 region

→ antibody response is type-specific & not cross-protective ∴ each strain needs to be eradicated individually

→ WPV2 & WPV3 are globally eradicated in 1999 & 2019 respectively

[currently all the natural cases are caused by WPV1]

Pathogenesis: transmission by feco-oral route is most common, respiratory droplets via inhalation or rarely by conjunctival contact

→ virus binds to CD155 receptors present on host cell surface & is internalized.

Spread to CNS/Spinal Cord:

• Hematogenous spread (most common): spread to regional lymph nodes ⇒ spill over to bloodstream (primary viremia) ⇒ multiplication in RES ⇒ re-entry into bloodstream (secondary viremia)

• Neural spread: spread directly through nerves

→ occurs following tonsillectomy where virus spreads via glossopharyngeal nerve present in tonsillar fossa

Site of Action: motor nerve ending, i.e.,

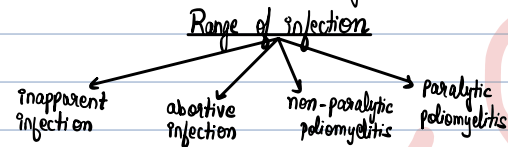
anterior horn cells of the spinal cord which leads to muscle weakness & flaccid paralysis

Neuron-degeneration: virus-infected neurons undergo degeneration

→ earliest change: degeneration of Nissl body

Clinical Manifestations:

→ incubation period = 7-14 days



→ Proximal muscles are affected earlier than distal muscles

→ Paralysis starts at hip & proceeds towards the extremities which leads to Tripod Sign (child sits with flexed hip, both arms extended towards the back for support)

→ Sites involved: spinal, bulbospinal & bulbar.

• respiratory insufficiency or dysphagia are common in bulbar involvement

→ Biphasic course: aseptic meningitis occurs first ⇒ recovery ⇒ return of fever with paralytic features 1-2 days later

→ NO SENSORY LOSS INVOLVED.

Laboratory Diagnosis:

→ antibody detection - by neutralization test

→ virus isolation in primary monkey kidney cell line from throat swabs, rectal swabs & stool samples

[detection of viral growth by CPE, viral antigen or viral gene in cell line]

• virus isolation from CSF/blood is very rare

→ real-time multiplex reverse transcriptase PCR

Polio Vaccine	Injectable Polio Vaccine [IPV] - Salk	Oral Polio Vaccine [OPV] - Sabin
Formulations	- IPV contains 3 serotypes (1,2,3) - can be given in 2 doses: full dose IPV & fractional-dose IPV	- OPV is available as: - Trivalent OPV (serotypes 1,2,3) - Bivalent OPV (serotypes 1,3) - Monovalent OPV (any one serotype)
National Immunization Schedule	2 fractional doses I.D. at upper arm [0.1 mL/dose] - given at 6th & 14th week along with bivalent OPV	- Total 5 doses [2 drops/dose P.O] of bivalent OPV - Zero dose at birth 1st, 2nd & 3rd dose ⇒ 6th, 10th, 14th week - booster at 16-24 weeks - Safe except in immunocompromised patients, pregnancy, old age
Safety	Relatively safer than OPV	Economical
Economy	Relatively expensive	Long-lasting
Duration of Protection in epidemics	Short, need booster doses periodically	Can be used safely
Herd immunity	Can precipitate paralysis	Prevalent due to feco-oral spread of vaccine virus
Can prevent VAPP, VDPV	Not provided	- Paralysis & intestinal re-infection - Relatively more chance
	- Only paralysis - zero chance	

{ VAPP: Vaccine-associated paralytic poliomyelitis }
 { VDPV: vaccine-derived polioviruses }

VAPP: occurs following OPV administration in OPV patients & their close contacts due to feco-oral spread

→ VAPP strains are not capable of circulating in the community

VDPV: live-attenuated vaccine strains present in OPV may undergo genetic changes to become VDPV

→ VDPV is capable of producing poliomyelitis (indistinguishable from WPV)

→ Three types of VDPVs:

• circulating VDPVs (cVDPVs)

• Immunodeficiency-associated VDPVs (iVDPVs)

• ambiguous VDPVs (aVDPVs)

Clostridium tetani

- obligate anaerobe, gram-positive
- terminal round spore (drum stick appearance)
- produces two toxins - tetanolysin & tetanospasmin
- Tetanolysin: hemolysin, no role in pathogenesis
- Tetanospasmin (or Tetanus toxin): neurotoxin
 - responsible for disease manifestations.
 - it gets toxoided spontaneously or by formaldehyde (toxoid form is antigenic but avirulent ⇒ used for vaccine preparation)
 - plasmid coded.

MoA: tetanus toxin binds to receptors (polysialogangliosides) on motor nerves

↓
toxin internalisation

↓
retrograde transport to GABA & glycine producing inhibitory neuron terminals

↓
prevents presynaptic release of inhibitory neurotransmitters - glycine & GABA

↓
spastic muscle contraction

Clinical Manifestations:

- Incubation period = 6-10 days. (shorter the incubation period ⇒ graver is the prognosis)
- Muscles of the face & jaw are affected first (due to shorter distances for toxin to reach presynaptic terminals)

First Symptom: increase in masseter tone ⇒ Lock jaw / Trismus

→ muscle pain & stiffness

→ back pain, difficulty in swallowing

In neonates: difficulty in feeding (initial presentation)

Disease Progression: painful muscle spasm

Localised - affects the limb involved

Generalised - descending spastic paralysis.

→ deep tendon reflexes are exaggerated

Autonomic Disturbance: maximal during 2nd week

- low/high BP • tachycardia • intestinal stasis
- sweating • increased tracheal secretions
- acute renal failure

Complications:

- Risus Sardonius - abnormal, sustained spasm of the facial muscles (appears to produce grinning)
- Opisthotonus position - bow shaped position (backward arching of body) due to generalised spastic contraction of the extensor muscles
- Respiratory Muscles spasm - may cause airway obstruction.

Epidemiological Risk Factors:

- warm climate
- rural area with fertile soil
- unhygienic surgeries/deliveries

Mode of transmission:

- Injury (superficial abrasions, punctured wounds, RTAs)
- Surgery done without proper asepsis
- Neonates - following abortion/delivery due to unhygienic practices
- Otitis media (otogenic tetanus)

Tetanus

Laboratory Diagnosis:

Specimen - excised tissue bits from the necrotic depths of wounds (more reliable than wound swabs)

Gram-staining: gram-positive bacilli with terminal round spores (drumstick appearance).

→ microscopy alone cannot distinguish C. tetani from morphologically similar non-pathogenic species like C. tetanomorphum, C. sphondiloides.

Culture: (more reliable than microscopy)

Robertson's cooked meat broth: C. tetani, being proteolytic, turns meat particles black & produces a foul odour.

Blood agar with polymyxin B: (incubated at 37°C for 24-48 hrs under anaerobic conditions)

→ C. tetani produces characteristic SWARMING GROWTH

Toxigenicity Test: toxin assay by in vivo mouse inoculation test on specimens such as serum & urine

Treatment:

I Passive Immunization (tetanus Ig)

→ TREATMENT OF CHOICE

→ Available preparations:

- HTIG (human tetanus Ig)
- ATS (antitetanus serum)

Dosage: 250 IU HTIG / 1500 IU ATS

→ single IM dose

→ intrathecal route is more effective

→ duration of protection:

- 30 days (HTIG)
- 7-10 days (ATS)

→ ATS is associated with side effects - serum sickness, anaphylactoid reaction.

II Combined Immunisation (Active + Passive)

→ In non-vaccinated person:

- first dose of tetanus toxoid (TT) in one arm
- ATS / HTIG in the other arm
- complete course of TT vaccine

III Antibiotics: play a minor role as they cannot neutralize the already-released toxins.

→ Useful in:

- early infection, before expression of toxin (< 6 hrs)
- to prevent further release of toxin

Metronidazole ⇒ DRUG OF CHOICE ⇒ 7-10 days (alternative ⇒ penicillin / doxycycline)

IV other measures: endotracheal intubation, early tracheostomy, anti-spasmodic (diazepam), β-blockers (to control sympathetic hyperactivity)

→ Surgical debridement - entry wound should be identified, cleaned & debrided of necrotic material, so as to remove the anaerobic foci of infection.

Prevention:

① Tetanus Toxoid (TT):

- monovalent vaccine
- combined vaccine - DPT
 - Td
 - Pentavalent vaccine

② Primary Immunization of Children:

→ Total 7 doses

- 3 doses of pentavalent vaccine at 6, 10, 14 weeks.
- 2 booster doses of DPT at 16-24 weeks & 5 years.
- 2 additional doses of Td at 10 years & 16 years.

③ Adult Immunization: if primary immunization not administered in childhood.

→ immunization with Td

→ deep IM route at anterolateral aspect of thigh (children) & in deltoid (in adults)

Protective tetanus titre ≥ 0.01 IU/mL.

Prevention of Tetanus After Injury

Vaccine Course	Non-tetanus prone wound	Tetanus-prone wound
< 5 years	-	-
> 5, < 10 years	Td 1 dose	Td 1 dose
> 10 years back	Td 1 dose	Td 1 dose + HTIG
unknown / incomplete immunisation	Td complete dose	Td complete dose + HTIG.

Neonatal Tetanus:

an illness occurring in a child who loses ability to suck & cry between Day 3 & 28 of life & becomes rigid & has spasms. a.k.a 8th day disease (∵ symptoms usually start after 1 week)

→ Most common cause: unhygienic practices during delivery

→ Seasonal: more common in July - Aug - Sep.

Prevention of Neonatal Tetanus:

- discouraging home deliveries, promoting institutional deliveries
- following aseptic clean practices during deliveries (clean hands, clean surfaces, clean blades, clean cord tie, clean cord stump, clean towel, clean water)
- Td (2 doses) ⇒ given to all pregnant women during 2nd trimester at 4 weeks gap.

India has achieved the elimination status for neonatal tetanus (as declared by WHO in 2016).