

WORLD RABIES DAY –SEPTEMBER 28

Face to face with RABIES



Dr.Jeny K John & Dr. Jobin Jose Kattoor

INTRODUCTION

- ▶ Rabies is a fatal disease posing serious public health concern, particularly in **Asia and Africa**

(Singh *et al.*, 2017)

- ▶ Due to under-reporting, rabies falls among the eighteen Neglected Tropical Diseases

(Hampson *et al.*, 2015)

- ▶ Word rabies has its origin from the Latin word “**rabere**” which means “to be mad”.
- ▶ First reference to animal rabies was recorded in **the laws of Eshunna in Mesopotamia in twenty third century BC**
- ▶ **First century BC – Canine rabies was described in the Susrutasamhita (India)**
- ▶ Demonstration of neurotropism and development of a rabies vaccine followed by its successful administration by **Louis Pasteur** during 1880’s was a great triumph in the history of rabies.

- ▶ Rabies is **completely preventable** through **exhaustive mass immunization of dogs and post-exposure prophylaxis (PEP)** of humans as well as animals.
- ▶ It is imperative to **create awareness among the vulnerable population to avoid exposure**
(Balaram et al., 2016)
- ▶ Manifestation : mild to severe neurological signs described as furious and dumb forms, followed by paralysis, coma and death.
- ▶ Rabies virus circulates in two interrelated cycles,
 - involving domestic mammals (predominantly dogs, cats)
 - involving wild mammals (mongoose, fox, raccoon, wolf, jackal, badger, bat, etc.)(Kuzmin et al., 2012)

- ▶ Stray dogs and probably jackals in India are the main vectors or reservoirs that sustain the disease in humans and other animals through bite of the infected ones
(Singh *et al.*, 2017)
- ▶ Bat mediated rabies, which is most prevalent in Latin American countries, has not been reported in India.
- ▶ With the aim to eradicate the dog-borne rabies in humans by 2030, Global Alliance for Rabies Control (GARC), a non-profit organization in collaboration with international rabies stakeholders (WHO/OIE/FAO) and various other communities is engaged in rabies control activities.

Etiology

- ▶ Rabies virus (RABV) is a **negative sense, single-stranded, enveloped, bullet shaped RNA virus** of the genus *Lyssavirus* under the family Rhabdoviridae



7 distinct genotypes of RABV

- ▶ **Classical rabies virus genotype 1** (street and laboratory strains) is the globally prevalent and causing disease in >99% cases in humans as well as animals

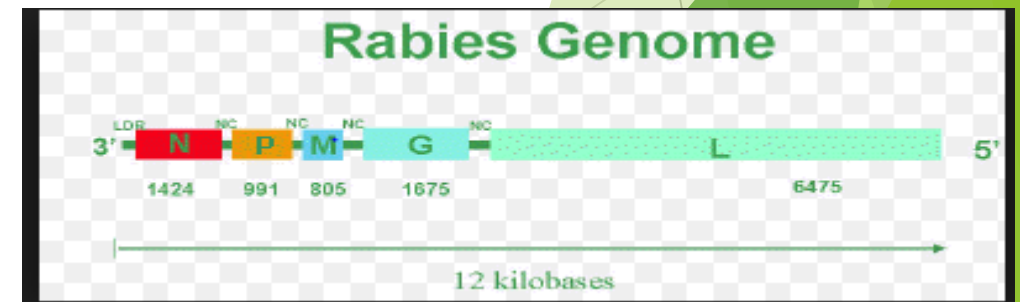
(King *et al.*, 2011)

- ▶ Other 6 genotypes named as rabies- related viruses (RRVs) such like Lagos bat virus (genotype-2), Mokola bat virus (genotype-3), Duvenhage virus (genotype-4), European bat Lyssaviruses (genotypes-5 and 6), and Australian bat Lyssavirus (genotype-7), are prevalent in certain areas of Africa, Western and Eastern Europe and Australia
(Gould *et al.*, 1998; Heaton *et al.*, 1999).
- ▶ The fatal classical rabies like disease in humans as well as animals is known to be caused by all the RRVs, except Lagos bat virus.

- ▶ Genome of RABV contains 5 highly conserved genes in the following order from 3' leader sequence **N**, **NS (P)**, **M**, **G** and **L**

(Yousaf *et al.*, 2012)

- ▶ **N gene** encodes a highly conserved **nucleoprotein** with “group specific” antigenic determinants shared by all rabies viruses and is **exploited for diagnosis and virus identification**.
- ▶ **G gene** encodes trans-membrane glycoprotein, a “serotype specific” antigen responsible for pathogenesis and induces neutralizing antibodies in the host (Singh *et al.*, 2017)
- ▶ **M protein** –assembly and budding of virus



2 strains of virus – fixed & street virus.

- ▶ Fixed virus is obtained after several passaging of virus in rabbit, which fixes its properties like incubation period, disease producing ability etc
- ▶ Street virus occurs normally in nature and has variable long incubation period

Epidemiology of rabies

- ▶ Affects all warm blooded animals
- ▶ Incidences have been reported from all continents except Antartica
(Singh *et al.*, 2017)
- ▶ Asia remains the hot spot in terms of both incidences and reservoirs.
- ▶ Mortality rate in children below 15 years of age is more.
- ▶ South Asian region -India and Bangladesh, Nepal, Bhutan, Myanmar, Thailand and Indonesia
- ▶ Prevalence across domesticated species of animals in the region is in the range of 20-50%.

- ▶ In India, rabies is widely prevalent except Andaman and Nicobar
- ▶ Increasing trend in stray dog population, urbanization and the lack of environmental hygiene among the densely populated rural communities are considered as critical risk factors in the predominance of rabies in Asian as well as African countries
- ▶ On the contrary, European nations zero down rabies in humans by compulsory vaccination in animals, especially in dogs
- ▶ Prevalence of rabies as 61.4% in cattle and buffaloes, 48.7% in goats, 48% in dogs, 45% in horses and 21.9% in cats

(WHO, 2005; Sudarshan et al., 2006; Singh et al., 2017)

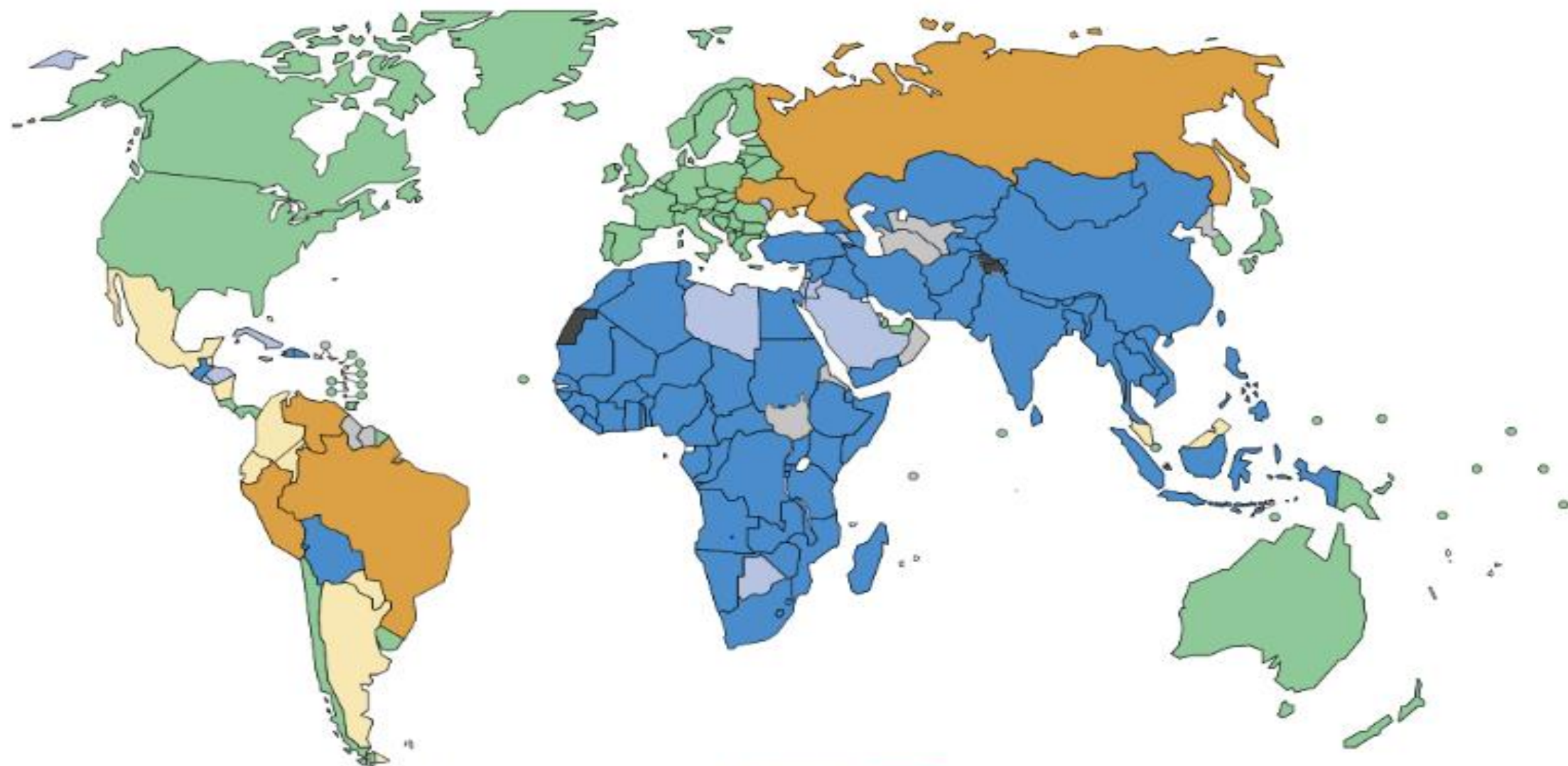
- ▶ Rabies outbreaks in domestic and wild animals in India are reported round the year with all such cases traced back to the bite of rabid dogs

(Singh et al., 1990; Singh et al., 1995)

- ▶ A study conducted from 11 states/union territories of India during January 2012 to December 2014 showed about 52.3% of rabies cases were from Karnataka, 18% from Maharashtra, 7.8% from Tamil Nadu, 6.3% from Kerala and 3.1% from Andhra Pradesh states.
- ▶ West Bengal was the worst affected state, with 47 rabies deaths in 2016, followed by Karnataka with 19 deaths ([Mani et al., 2016](#))

Endemicity of dog rabies and dog-transmitted human rabies, 2016

Endémicité de la rage canine et de la rage humaine à transmission canine, 2016



- Endemic dog-transmitted human rabies: dog rabies and dog-transmitted human rabies present in the country – Endémie de la rage humaine transmise par les chiens: la rage canine et la rage humaine à transmission canine sont présentes dans le pays
- Endemic dog rabies: dog rabies in the majority of the country, but no dog-transmitted human rabies cases – Endémie de la rage canine: rage canine présente dans la majeure partie du pays, mais aucun cas de rage humaine transmise par les chiens
- Sporadic dog-transmitted rabies: dog rabies in few areas of the country with sporadic human cases – Cas sporadiques de rage transmise par les chiens: rage canine présente dans quelques zones du pays, accompagnée de cas humains sporadiques
- Controlled dog rabies: few cases of dog rabies in limited areas of the country but no dog-transmitted human rabies cases –

- No dog rabies: zero dog rabies and zero dog-transmitted human rabies cases (except from imported) – Absence de rage canine: aucun cas de rage canine et aucun cas de rage humaine transmise par les chiens (sauf cas importés)
- No information – Aucune information
- Not applicable – Sans objet

TRANSMISSION

- ▶ Most common way of entry through **saliva or infected neural tissues via bite wounds or open cuts in the skin and mucus membranes and not through intact skins**
- ▶ Dog catching, handling, loading transportation – increases risk
- ▶ Non bite exposure methods-inhalation of aerosolized rabies virus, cornea/organ transplant, contamination of abrasions, open wounds, mucus membranes with virus laden saliva

- ▶ Rabies in human beings develops from the bites of infected animals, predominantly dogs (91.5%)
- ▶ Fatality depends on the site of bite and the quantum of virus in the saliva of bitten rabid animal
(Hemachudha *et al.*, 2013)
- ▶ Transmission through gastro-intestinal tract has also been documented
(CDC, 1999).
- ▶ Exposure to live attenuated vaccines during the production stage may have potential risk

Pathogenesis

- ▶ Incubation period: 2 weeks to 6 years
- ▶ Greater risk if bite on head, face, neck and hand
- ▶ Rabies virus remain in muscles for prolonged period - give the chance for post exposure treatment and clearance of virus by immune system

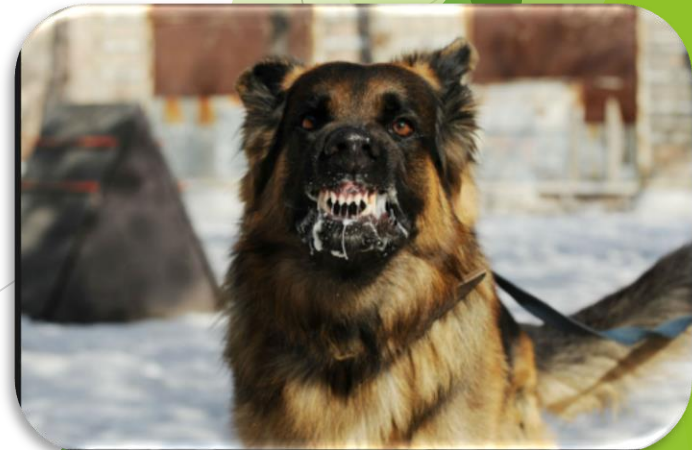
Sequence of events in the disease process involves:

- ▶ 1) replication of the virus in the peripheral tissues (virus attaches by G protein and
- ▶ 2) spread along the peripheral nerves and the spinal cord to the brain- infect nerve cells
- ▶ 3) dissemination within the central nervous system (CNS)
- ▶ 4) centrifugal spread along nerves to various tissues and organs.
- ▶ Receptors are nicotinic acetylcholine receptor, neural cell adhesion molecule (CD56) and low-affinity nerve growth factor receptor (NTR75)
- ▶ **SPEED OF TRANSPORT-12-100MM/DAY**

- ▶ In brain it damages nerve cells & vascular endothelium, grows in ganglion cells.
- ▶ Irritation to the nerve cells cause excitability producing furious form.
- ▶ Due to affection of medulla, fever, polyuria & glycosuria are observed.
- ▶ Subsequently, nerve cells under go degeneration & so the muscle paralysis sets in.
- ▶ Due to paralysis of deglutination muscles, animal is unable to swallow & saliva dribbles, jaw hangs down.
- ▶ Paralysis of respiratory muscles leads to respiratory failure (asphyxia) & death.
- ▶ From brain, virus spread centrifugally along the nerves supplying salivary glands (i.e. facial & glossopharyngeal nerve) and then excreted through saliva.
- ▶ May spread to milk, foetus in utero.

Symptoms

- ▶ Clinical signs of rabies are rarely definitive.
- ▶ Rabid animals of all species exhibit typical signs of CNS disturbance, with minor variations among species.
- ▶ The most reliable signs, regardless of species, are **behavioral changes and unexplained paralysis**.



In dogs, it is manifested in 2 forms:-

Furious form - Animal is very excitable, bites everybody, snaps & barks at imaginary objects, chews all objects – sticks, stones etc.

- ▶ Eyes are red, have a vacant look, champing of jaws & dribbling of saliva

Dumb form - Animal is in dumb, sad condition.

- ▶ It does not obey orders, does not recognizes its master & lower jaw hangs, with dribbling of saliva.
- ▶ Death occurs in 3-4 days, or within 10 days

- ▶ Horses - There is mania along with colic symptoms.
- ▶ Cattle & Sheep – Cattle with furious rabies are dangerous, attacking and pursuing man and other animals.
- ▶ There is bellowing & bleating. Bulls & ram show excessive sexual urge.
- ▶ Cat - Only Furious form noticed.
- ▶ Rabbits – Only dumb form is noticed.



Humans

- ▶ Swelling of the brain and the spinal cord
- ▶ Frequent headache
- ▶ Feeling of Nausea
- ▶ Severe stomach pain
- ▶ Develop muscle cramps
- ▶ It causes drowsiness
- ▶ Causes Insomnia
- ▶ Develop severe fever
- ▶ Feeling of Anxiety
- ▶ Develop a state of confusion
- ▶ Causes hallucination
- ▶ excess secretion of saliva
- ▶ Develop problem during swallowing
- ▶ Develop fear from water

Lesions

These are limited to CNS only.

- ▶ Grossly, hyperemia, edema of meninges with few petechiae.

H/P: - Changes are seen in Hippocampus, the brain stem and the gasserion ganglion.

- ▶ Non – suppurative encephalitis is noticed.
- ▶ Perivascular cuffing by lymphocytes is noticed.
- ▶ Endothelial cell injury and vacuolation in endothelium
- ▶ Around the ganglion cells, microglial cells proliferate & form nodules called as “Babes nodule” & then neuronophagia occurs.
- ▶ Degeneration of the salivary and lachrymal glands, pancreas, and adrenal medullae

- ▶ Ganglioneuritis in paraveterbral ganglion is constantly present, which are swollen, red and reveal infiltration of lymphocytes, degeneration, neuronophagia and satellitosis.
- ▶ Inclusions bodies (I/C acidophilic) appear in neuron cytoplasm called as Negri bodies.
- ▶ These are found abundantly in hippocampus region in dogs and in purkinje cells of cerebellum in ruminants.

Diagnosis

- ▶ 1) Symptoms
- ▶ 2) Isolation of suspected dog for 10 dogs (Death occurs)
- ▶ 3) Impression smears from hippocampus region stain by “Seller’s Stain” and Manns stain for Negri bodies.
- ▶ 4) Habel’s Mouse Inoculation test – 2-3 wk old mice inoculated with suspected material in skull.
- ▶ Examine brain after sacrificing mice at 5, 6, 7th day and look for Negri bodies.



- ▶ 5) Biological inoculation test in Rabbit – Intra cranially can produce the disease.
- ▶ 6) Immunofluorescence microscopy on fresh brain tissue, which allows direct visual observation of a specific antigen-antibody reaction, is the test of choice.
- ▶ When properly used, it can establish a highly specific diagnosis within a few hours.
- ▶ Brain tissues examined must include **hippocampus, medulla oblongata, and cerebellum** (and should be preserved by refrigeration with wet ice or cold packs).
- ▶ 7) CFT

- ▶ PCR assay and sequencing

Samples : saliva, serum, spinal fluid, and skin biopsies of hair follicles at the neck region

- ▶ Isolation of RABV in albino mouse, cell lines like Neuro2a/CCL 131, BHK-21/C13, Vero and McCoy are the most dependable and consistent methods in diagnosis of rabies

CCL 131 cell line without any adaptation is most susceptible to street RABV.

- ▶ Lateral flow technique employing immunochromatographic principle having 100% specificity and more than 88% sensitivity enabled the early diagnosis of diverse rabies virus strains (RABV species 1, 5, 6 and 7)

- ▶ Approaches currently recommended by WHO to estimate RABV neutralizing antibodies are rapid fluorescent focus inhibition test (RFFIT) and fluorescent antibody virus neutralization test (FAVN)
- ▶ A minimum measurable protective antibody titre of 0.5IU/ml represents the level of immunity in humans and animals to rabies infection (OIE, 2016).

Treatment

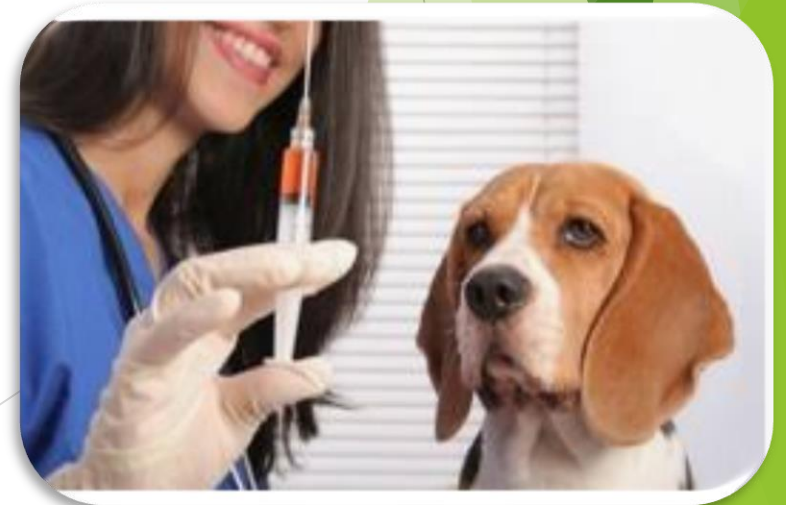
Categories of contact with suspected rabid animal and Post-exposure prophylaxis measures

- ▶ **Category I** – touching or feeding animals, licks on intact skin-
None
- ▶ **Category II** – nibbling of uncovered skin, minor scratches or abrasions without bleeding- Immediate vaccination and local treatment of the wound
- ▶ **Category III** – single or multiple transdermal bites or scratches, licks on broken skin; contamination of mucous membrane with saliva from licks, contacts with bats- Immediate vaccination and administration of rabies immunoglobulin; local treatment of the wound

- ▶ Being an enveloped virus, thorough cleansing of the wound should be carried out as soon as possible with soap/ detergent and water for 10 min
- ▶ Application of ethanol or viricidal agents such as aqueous solution of iodine or povidone dissolves the envelope of virus and hence destroys majority of virus around the wound site
- ▶ In category III exposures, local infiltration of rabies immunoglobulin should be done in the depth and around the wound to neutralize the virus.
- ▶ Suturing of wound should be avoided as far as possible.
- ▶ Tetanus and antibiotic prophylaxis should be given to prevent sepsis in the wound

Prevention and control

- ▶ Strict licensing and vaccination of pet dogs
- ▶ Control of stray dog population - mass vaccination and animal birth control program
- ▶ Elimination of suspected infected animals
- ▶ Education of public about post exposure vaccination
- ▶ Public health educational program
- ▶ **Pre-exposure vaccination should be included in routine immunization schedule????**
- ▶ Intradermal vaccination



Vaccination approaches in control of rabies:

- ▶ To stimulate the host immunity against rabies infection, normally different forms (live intact, inactivated, attenuated) or purified components of pathogens (outer coat proteins of rabies virus) with high immunogenicity are used.
- ▶ This generates immune response within 2 weeks.
- ▶ These vaccines are administered by IM or ID routes.
- ▶ If dog population is sufficiently (>70%) covered by rabies vaccination, it will reduce the disease burden in humans.
- ▶ The vaccines administered within first few days of postexposure are reported to decrease the disease by attenuating the virus considerably. Later on the 4 dose series (day 0, 3, 7 and 14)

- ▶ Vaccination in wild animals is an essential part in control of rabies so as to prevent its spread to domestic animals and human.
- ▶ **Oral Rabies Vaccine (ORV) laden baits** have been successfully established for red, Arctic, and gray foxes; coyotes, raccoon dogs, raccoons, skunks, and domestic dogs (Yang *et al.*, 2013).
- ▶ ORV was explicated to vaccinate free-ranging wild animals in geographically large enzootic areas, to subjugate the development and extent of a rabies epizootic, or to establish a rabies-free buffer zone (Muller *et al.*, 2001).

Vaccination for rabies : Pre- or post-exposure



- Pre-exposure vaccination is performed to safeguard those who are at high risk of rabies exposure.

Pre-exposure immunization should be offered to **rabies researchers, certain laboratory workers and other persons in high-risk groups, such as veterinarians and their staff, and animal handlers**

Vaccination is recommended for children living in or visiting countries where exposure to rabid animals is a constant threat

- Post-exposure vaccination is performed after the bite of rabies suspected animal to prevent the development of disease.
- The vaccines used for pre-exposure and post-exposure prophylaxis remains the same, but the schedule varies

- ▶ Pre-exposure prophylaxis for humans consists of three doses on Days 0, 7, and 21 or 28.
- ▶ Post-exposure vaccination five doses on 0,3,7,14 and 28
- ▶ Rabies immune globulin (RIG) 20 IU/kg on Day 0 in conjunction with the first vaccine dose.
- ▶ Dog and cat: 3 mth
- ▶ Booster after 1 month
- ▶ Annually revaccination

Rabies research centers in India

- ▶ CADRAD, ICAR-IVRI, Bareilly
- ▶ ICAR-NIVEDI, Bengaluru
- ▶ Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), Ludhiana
- ▶ NIMHANS (human samples), Bangalore
- ▶ National Centre Disease control (NCDC), formerly National Institute of Communicable Disease (NICD), New Delhi

- ▶ The NIMHANS (human rabies) and NCDC (human and animal rabies) are the two World Health Organization (WHO) collaborating centers for rabies epidemiology and reference and research in rabies, respectively.

A large number of pink piggy banks with white wings are flying through a blue sky with white clouds. The piggy banks are of various sizes and are scattered across the frame, creating a sense of movement and abundance. The text "Thank you..." is written in a red, cursive font across the center of the image.

Thank you...