

# Paramyxoviridae

Newcastle Disease (Ranikhet Disease)

Peste des petits ruminants (PPR)

Canine Distemper (CD)

Greek **para-** “by the side of” and **myxa** “mucus”

# Paramyxovirus

## Group V: Negative sense ssRNA viruses

**Order:** *Mononegavirales*

**Family:** *Paramyxoviridae*

**Genus:** *Aquaparamyxovirus*

*Avulavirus*

*Ferlavirus*

*Henipavirus*

*Morbilivirus*

*Respirovirus*

*Rubulavirus*

### **Important Species**

*Newcastle disease virus*

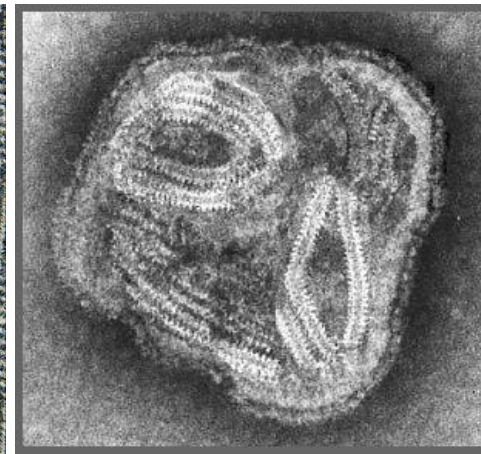
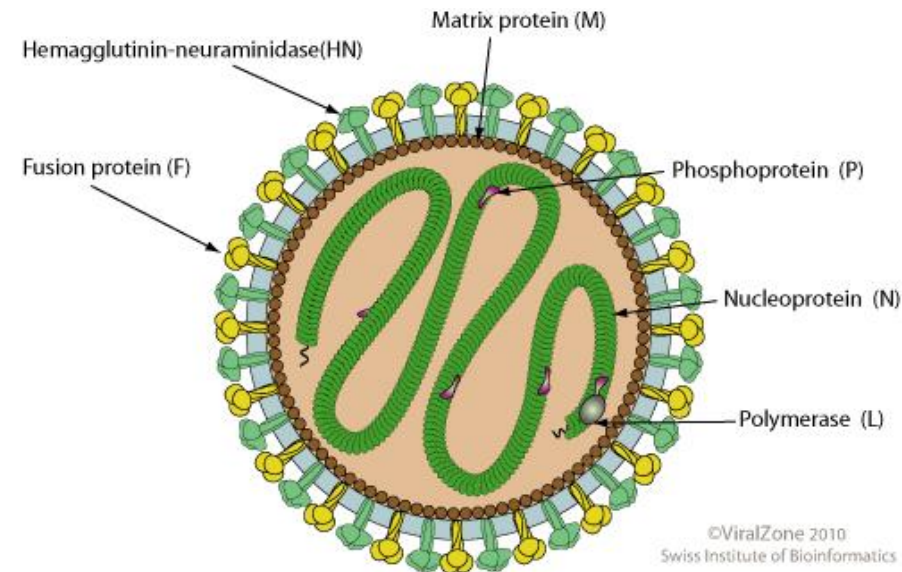
*Caniner Distemper virus*

*Peste des petits ruminants virus*, **Measles virus**

*Human Rubula virus* – **Mumps virus**

# Properties of Paramyxovirus

- Virions are **enveloped**
- **Size: 150-300 nm in diameter.**
- Shape: **Pleomorphic (spherical as well as filamentous forms occur).**
- Covered with large **peplomers**
- Virions contain a **"herringbone-shaped" helically symmetrical nucleocapsid**
- Virion envelope contains **two viral glycoproteins Haemagglutinin & Neuraminidase.**
- Genome consists of a **single linear molecule of negative sense, single-stranded RNA,**
- **Cytoplasmic replication, Syncytium formation, intracytoplasmic and intranuclear inclusion bodies** (genus Morbillivirus)



## Ranikhet Disease



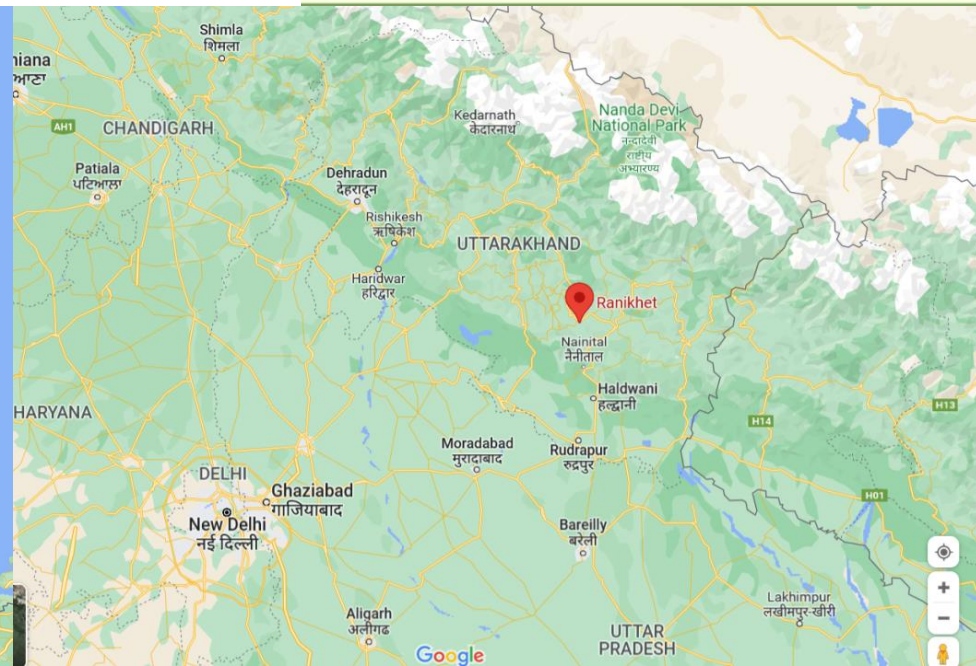


# Newcastle Disease

## History

First seen-Java, Indonesia in 1926 and in the same year it was reported in **Newcastle, England** in 1926

In India - First recorded by Edwards in 1927 in **Ranikhet, Uttaranchal, India**



# Newcastle Disease Virus Strains

The virulence of ND virus varies .

Based on the virulence the NDV isolates can be divided into three groups viz.

**Lentogenic (Lasota, Hitchner, England F)**  
**Mesogenic (R2B Mukteshwar) &**  
**Velogenic (Virulent ND) strain.**

The intracerebral pathogenicity index (ICPI), is the World Animal Health Organization protocol for defining NDV pathogenicity, consists of inoculating virus into the cerebrum of one-day-old SPF (or NDV-antibody free) chickens and deriving a clinical weighted score that ranges from 0.0 to 2.0.

Scores  $\geq 0.7$  classify a strain as virulent (vNDV).

Typically, ICPI values for mesogenic strains are from 0.7 to 1.5, and from 1.5 to 2.0 for velogenic strains

# Physical Properties of **Newcastle** Disease virus

**Temperature:** Inactivated by 56°C/3 hours, 60°C/30

**pH:** Inactivated by acid pH

**Chemicals:** Ether sensitive

**Disinfectants:** Inactivated by formalin and phenol

**Survival:** Survives for long periods at ambient temperature, especially in faeces

# Newcastle Disease : Transmission

- Direct contact with faeces and respiratory discharges
- Contamination of the environment
  - Feed, water
  - Equipment
  - Human clothing
  - Contaminated or incompletely inactivated vaccines





## Pathogenesis

Virus replicates in respiratory and intestinal respiratory tract



Within 24 hours virus spread via blood



Viraemia



Spleen, bone marrow-other target organs proventriculus,  
gizzard



Cross brain barrier-destroy respiratory center in brain-paralysis  
& death.

# Newcastle Disease



## Clinical Signs

Incubation period : 4-6 days

### Viscerotropic velogenic/Asiatic Form ND (Doyle's Form)

Hyperpnea, greenish blood stained diarrhea,  
dehydration, tremor, torticollis and paralysis of wings or leg.  
Haemorrhagic lesions are prominent in the digestive tract.  
Mortality is close to 90%.

### Neurotropic velogenic ND (Beach's Form)

Severe respiratory and nervous signs predominate.  
Coughing and gasping,  
Head tremors, wing and leg paralysis and twisted necks.  
Depression, loss of appetite and a drop in egg production also occur.  
Mortality: 10% - 50% of adults .



## Clinical Signs

### Mesogenic ND (**Beaudette's form**)

Mainly respiratory signs, with coughing but no gasping.

Depression, loss of weight and decrease in egg quality and production for up to 3 weeks.

Nervous signs may develop late in the course of the disease and death rates are about 10%.

### Lentogenic ND (**Hitchner's Form**)

Symptoms are mild or absent and include mild respiratory signs, impaired appetite and a drop in egg production.

No nervous signs occur and deaths are usually negligible.

# Newcastle Disease

## Clinical Signs

- Edema of head, especially around eyes
- Greenish-dark watery diarrhea
- Respiratory and neurological signs
- Signs vary with species and virulence





## Post mortem Lesions

- Indistinguishable from highly pathogenic avian influenza
- Hemorrhagic internal lesions
  - Tracheal mucosa
  - Proventriculus
  - Intestinal mucosa



# DIAGNOSIS

## 1. Isolation & Identification of the agent

**Samples:** Tracheal and cloacal swabs (or faeces) from live birds or from pools of organs and faeces from dead birds

Serological tests :Clotted blood samples or serum

Inoculation of 9-11-day-old embryonated chicken eggs .

## 2.Pathogenicity assessment

Plaque test in chicken embryo fibroblast cultures

Mean death time of embryonated chicken eggs

Intracerebral pathogenicity index in 1-day-old chickens

Intravenous pathogenicity index (IVPI) in 6-week-old chickens

## 3.Serological tests

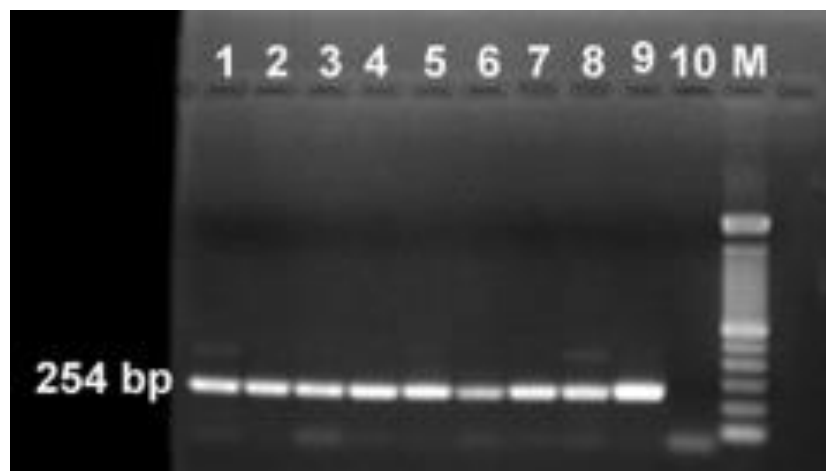
Haemagglutination inhibition test ,

ELISA .

## 4.Molecular detection

Reverse Transcriptase-PCR

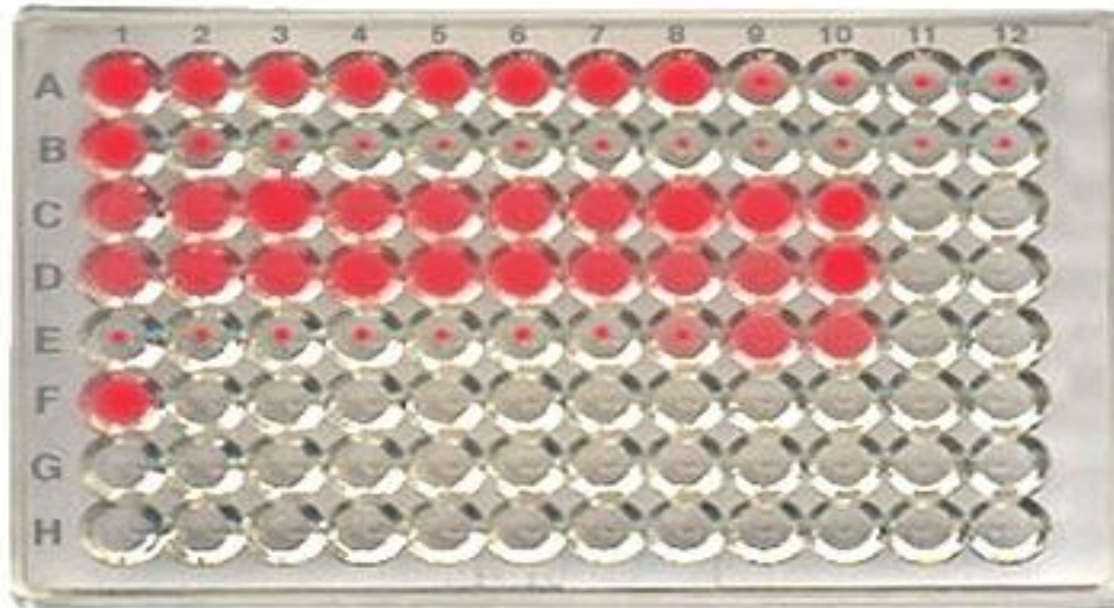
## Molecular Detection by RT-PCR



Analysis of RT-PCR products of different NDV isolates by gel electrophoresis  
1. La Sota, 2. Australis, 3. Hitchner B1, 4. Ulster 2C, 5. Herts 33, 6. PMV1-301/90, 7. PMV1-442/00, 8. PMV1-G94, 9. PMV1-758/00, 10. allantoic fluid, M- 100 bp DNA step ladder (Promega)

NDV-F328: 5'-CCTTGGTGAITCTATCCGIAGG-3',  
NDV-R581: 5'CTGCCACTGCTAGTTGIGATATACC-3'.

# Haemagglutination Inhibition Test



## PREVENTION & CONTROL

1. Strict biosecurity measures
2. All in all out strategy
3. Balanced feed, Clean water
4. Regular vaccination

### Vaccination

**Broiler** – 7<sup>th</sup> day, 28<sup>th</sup> day (**Lasota strain**)

**Layer** – 7<sup>th</sup> day, 28<sup>th</sup> day, 9 weeks (**Mukteshwar strain**), 16<sup>th</sup> week (**ND Killed Vaccine**), 40<sup>th</sup> week

**Breeder** – 7<sup>th</sup> day, 28<sup>th</sup> day, 9 weeks (**Mukteshwar strain**), 16<sup>th</sup> week (**ND Killed Vaccine**), 40<sup>th</sup> week

Booster after every 10 weeks (**ND Killed Vaccine**)



**THANKS**



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**Canine Distemper (CD)**

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# Paramyxovirus

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*Respirovirus*

*Rubulavirus*

### **Important Species**

*Newcastle disease virus*

### ***Caniner Distemper virus***

*Peste des petits ruminants virus, Measles virus*

*Human Rubula virus – Mumps virus*

THIS STORY IS FROM DECEMBER 11, 2019

# 34 lions had died in canine distemper virus outbreak: Gujarat govt

Himanshu Kaushik / TNN / Updated: Dec 11, 2019, 15:27 IST



## ARTICLES



34 lions had died in canine distemper virus outbreak: Gujarat govt



Cause of Asiatic lioness Radha's death still a mystery



Seven arrested for thefts in two districts



Protesting LRD candidates detained in Gandhinagar



File photo of Asiatic lion at Kankaria zoo in Ahmedabad

## SPOTLIGHT

- 1 Manav Rachna Announces Admission Expo
- 2 Samsung Galaxy F23 5G arrives with a bang!
- 3 Shahid & Mira get hilarious in latest OnePlus ad

TOI  
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<https://indianexpress.com/article/india/1000-doses-of-vaccine-imported-says-official-lions-that-died-early>

ENGLISH தமிழ் বাংলা മലയാളം हिंदी मराठी



Monday, March 21, 2022

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Home / India / 1,000 doses of vaccine imported, says official; 'Lions that died early this year had canine distemper virus too'

# 1,000 doses of vaccine imported, says official; 'Lions that died early this year had canine distemper virus too'

The confirmation comes after the detection of babesiosis among lions in parts of Gir forest and establishes that the combination of the viral and protozoal infections observed in 2018 has occurred again and claimed around 32 big cats.



# LIFE & DEATH



## POPULATION CONCERNS

- Lions have spilled outside the Gir forest, as was reflected in the lion census figures of 2015
- One in three lions lives outside the sanctuary area, the survey showed
- The increasing population is forcing lions to leave the protected area
- There are an estimated 800 lions in Gujarat right now

## WHAT HAPPENED IN GUJARAT?

- The lion deaths last September sparked fears that an epidemic could devastate the last surviving population of Asiatic lions
- Lions were quarantined in the Gir sanctuary area
- As a precaution, 33 lions from an adjoining forest range in Gir were also captured and quarantined. Their return to their natural habitat remains uncertain
- A drive to vaccinate dogs in the forest area was also carried out, as they could be carriers of canine distemper virus (CDV)
- The government carved out a new Shetrunji wildlife division, outside the Gir wildlife sanctuary, to give the lions better cover





# Canine Distemper

Canine distemper is a **highly contagious acute febrile disease**.

Canine distemper virus (CDV) is a paramyxovirus closely related to the measles and rinderpest viruses;

Canine distemper has been known since at least 17<sup>th</sup> century and has a worldwide distribution.

**1746** Well-described in by **Antonio de Ulloa**

**1905 Carre** demonstrated the filterable agent in disease.

**1926 Laidlaw and Dunkin** confirmed the causative agent.

Canine distemper is a contagious and serious disease caused by a virus that attacks the **respiratory, gastrointestinal and nervous systems of puppies** and dogs.

## Canine Distemper virus

- Virus survives heating at 55 degree C for 30 minutes.
- Inactivated at 60 degree C for 30 minutes.
- pH stability- 4.5 to 9.0
- Inactivated readily by Formalin 0.1% for 1-2 hrs., 0.5 Phenol for 48-72 hours and 0.3% Chloroform for 10 minutes
- **Eleven distinct genetic lineages of CDV are recognized** worldwide, based on phylogenetic sequence analysis of the H gene.
- These CDV lineages are known as America-1, America-2, Arctic, Asia-1, Asia-2, Asia-3, Asia-4, Europe-1/South America-1, Europe wildlife, Rockborn-like, and Africa-1.
- **Despite genetic differences among field strains of CDV, cross-neutralization studies show only minor antigenic differences.**

# Canine Distemper : Transmission

Puppies and dogs most often become infected through **airborne exposure** (through sneezing or coughing) to the virus from an infected dog or wild animal.

The virus can also be transmitted by **shared food and water bowls and equipment.**

Infected dogs can shed the virus for months, and **mother dogs can pass the virus through the placenta to their puppies.** The virus may be transmitted *in utero* and may persist in the brain.

Contact between **wild animals and domestic dogs** can facilitate the spread of the virus. The domestic dog has largely been responsible for spreading the disease to new geographic areas and for introducing canine distemper to previously unexposed wildlife.

As human populations expanded, **increased domestic dog contact with wild carnivores** exacerbated the risk for disease transmission

# Host

The disease occurs in a wide variety of terrestrial carnivores including

**Canidae** (dog, fox, wolf, raccoon dog),

**Mustelidae** (ferret, mink, skunk, wolverine, marten, badger, otter),

**Procyonidae** (raccoon, coatimundi),

**Viverridae** (palm civet),

**Ailuridae** (red panda),

**Ursidae** (bear), and

large **Felidae** (lions, tigers, leopards, cheetahs), as well as a few other mammals such as Asian elephants and some primates.

**Domestic and feral dogs** are considered to be the **main reservoir host species**. All dogs are at risk **but puppies younger than four months old and dogs that have not been vaccinated against canine distemper are at increased risk** of acquiring the disease.

CDV is a **serious threat to endangered wildlife** and this threat is expected to increase with increased encroachment of humans (along with their dogs) into undeveloped areas of the world.

# Canine Distemper - Pathogenesis

- Virus enters via respiratory or alimentary passage
- Amplification of virus in reticuloendothelial cells
- **Viraemia** (Lymphopenia)
- **Increase in temperature** 3-6 days post infection
- If immune response sets in, virus fail to infect epithelial tissues and viraemia ceases. If not
  - 10-18 days post infection virus infects epithelial cells of intestine, respiratory, urogenital skin and exocrine endocrine glands
  - **Second viraemia**
  - **Rise of Temperature (Diphasic Fever)**
- Neurotropic strains invade CNS (meningeal macrophages, neurons. Recovered dogs suffer from subacute diffuse sclerosing encephalomyelitis (Old Dog Encephalitis)
- CDV is shed in respiratory exudates, feces, saliva, urine, and conjunctival exudates for up to 90 days after natural infection.



# Canine Distemper - Clinical Signs

- Incubation Period: 3-7 days
- Initially, infected dogs will develop **watery to pus-like discharge from their eyes**. They then develop **fever, nasal discharge, coughing**, lethargy, reduced appetite, and vomiting.
- As the virus attacks the nervous system, infected dogs **develop circling behavior, head tilt, muscle twitches, convulsions** with **jaw chewing movements** and salivation (“**chewing gum fits**”), **seizures**, and partial or complete paralysis.
- In cutaneous form- Appearance of rashes, vesicular lesion, pustules on the ventral aspect of abdomen and inner side of thighs and causes **footpads to thicken and harden**, leading to its nickname “**hard pad disease**”.
- In wildlife, infection with canine distemper **closely resembles rabies**.
- Distemper is often fatal, and dogs that survive usually have **permanent, irreparable nervous system damage**.

# Clinical Signs

- **Respiratory**

- Nasal & Ocular Discharge
- Coughing
- Dyspnea
- Pneumonia

- **Gastrointestinal (GI)**

- Anorexia
- Vomiting
- “Distemper Teeth”
- Diarrhea (May be bloody)

- **Dermatological**

- Abdominal Pustules
- Nasal & Digital Hyperkeratosis

- **Ocular**

- **Anterior Uveitis**

(Inflammation of the front chamber of the eye; may cause the cornea to appear cloudy and/or cause changes in the appearance of the virus.)

- **Keratoconjunctivitis Sicca**

- **Optic Neuritis**

- **Retinal Degeneration**

- **Neurological**

- **“Chewing Gum” Seizures**

- **Weakness or Paralysis**

- **Loss of Balance**

- **Muscle Twitching**

- **Hypersensitivity**

- **Neck Pain**

- **Behavioral Changes**

# Clinical Signs

- Nasal & Digital Hyperkeratosis
- -often found in dogs with neurological manifestations.



Hyperkeratosis of nasal planum



Hyperkeratosis of foot pad



NORMAL PAWS



AFFECTED PAWS

## **HYPERKERATOSIS OF THE DIGITAL PADS**





Mucous secretion from conjunctiva  
Ulceration of cornea  
Panopthalmitis-inflammation of the interior of the eye that also extends into the uvea and sclera  
Retinal atrophy of all layers  
**OLD DOG ENCEPHALITIS**



# Distemper Teeth



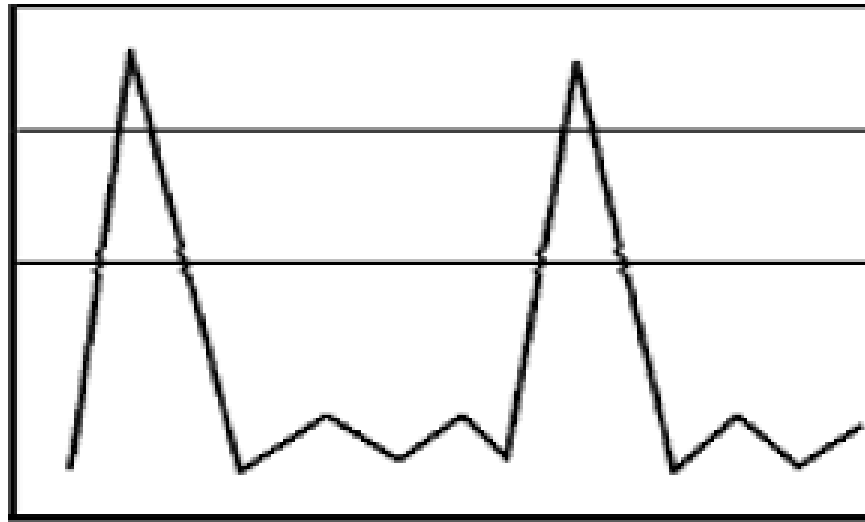
The pitted, discolored teeth that may result when young dogs are infected with distemper virus prior to the eruption of their permanent teeth.



**Enamel hypoplasia**



# DIPHASIC FEVER CURVE



3-4<sup>th</sup> day      11<sup>th</sup> –      13<sup>th</sup> day high fever  
High fever      12th  
due to  
viraemia

## Epileptic Seizures and Excessive Chewing Movements Accompanied By Salivation



# Diagnosis of Canine Distemper

## Material Collection:

- Buffy coat cells are the most rewarding specimen for ante-mortem diagnosis of canine distemper.
- 6 ml of EDTA-blood from suspect distemper dogs.
- If cerebral distemper is suspected, a CSF sample submitted along with serum (red top tube/clotted blood) can lead to a definitive diagnosis.

# Diagnosis of Canine Distemper

- 1) **Virus Isolation:** vero cells expressing canine signalling lymphocyte activation molecule (Vero.DogSLAM). Cytopathic effects (CPE) in the form of **syncytia formation** and cell necrosis were observed in 33 (20.4%) specimens within 24 h of inoculation and the presence of CDV was confirmed with the aid of the direct fluorescent antibody test and electron microscopy (EM).
- 2) **Immunofluorescence assay** (IFA) which looks for inclusion bodies on conjunctival scrapes, in urine sediment, in transtracheal washes and cerebrospinal fluid (with neurological signs).
- 3) **Blood tests (serology) look for antibodies** (titers) to distemper. It is usually necessary to take serial titers on 2 serum samples taken two weeks apart to detect rising titers as single titers do not have much diagnostic value. However, they can help to do risk assessment for exposed dogs in a shelter
- 4) **RT-PCR** (polymerase chain reaction) detects virus in respiratory secretions, CSF, feces, urine (depending on localization of virus). False positives are possible within 1-3 weeks of vaccination.

## PREVENTION & CONTROL

Vaccine	Primary Dose Puppy	Primary Dose Adult	Booster	Recommendation
<i>Distemper</i>	3 doses, 2-3-4 months	2 dose, 3-4 wk apart	Annual	Highly recommended for all ages

Usually a puppy can get it's first Vaccination at 45 Days of Age..

### Core Vaccination Schedule :

**1st Vaccination** : DHPP( Against Canine Distemper, Parvo, Hepatitis, Para Influenza) and Along with it Vaccination against leptospirosis is also given.

After 21 Days..

**2nd Vaccination** : Repeat Booster Dose of 1st Vaccination of DHPP and Lepto is given.

After 21 Days.

**3rd Vaccination** : Vaccination against Rabies is administered and 3rd Booster dose of DHPP is given. (90th Day)

After which every year the dog should be brought to the vet for an annual booster dose of Anti Rabies and DHPP Vaccination.

# MEGAVAC-7<sup>®</sup>

single shot protection against multiple canine infectious agents

- Consistent with current Indian pharmacopoeia
- Cross-protection against CPV-2, CPV-2a and CPV-2b
- High quality, trusted efficacy and safety

## Composition

- Canine Distemper Virus (Live) I.P
- Canine Contagious Hepatitis Virus CAV-2 (Live) I.P
- Canine Parainfluenza Virus (Live) I.P
- Canine Parvo Virus (Live) I.P
- Canine Contagious Hepatitis Virus (CAV-1), Inactivated I.P
- *Leptospira canicola* inactivated I.P
- *Leptospira icterohemorrhagiae* inactivated I.P

## Dosage

1 mL S/C or I/M

## Vaccination Regimen

- Primary Vaccination : 2 Months of age
- Booster Dose : 3 Months of age
- 2<sup>nd</sup> Booster Dose : 4 Months of age
- Re-vaccination : Annually

2

\* Additional information available on request





**THANKS**



# Paramyxoviridae

Newcastle Disease Virus, *Peste des petits ruminants*, Canine Distemper

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## SYNONYMS - *Peste des petits ruminants*

- Pseudo-rinderpest
- Kata
- Goat Plaque
- Pests of Sheep & Goat
- Pneumo-enteritis Complex
- Goat Catarrhal Fever

## **ETIOLOGY** - *Peste des petits ruminants*

- PPR disease is caused by Genus Morbillivirus of Paramyxoviridae family.
- The virus is antigenically similar to RP virus, CD virus & Measle virus.

## *Peste des petits ruminants*

**Peste des petits ruminants** (PPR), is an acute contagious disease caused by a Morbillivirus of Paramyxoviridae family. It affects small ruminants, especially goats, which are highly susceptible, and occasionally wild animals.

Resembles rinderpest in cattle.

**PPR is characterised** by severe pyrexia, which can last for 3-5 days, erosive lesions, which occur in the mouth, diarrhoea and pneumonia, serous ocular and nasal discharges. At necropsy, characteristic '**zebra markings**' may occur in the large intestine, but is not a consistent finding

## Susceptible Host - *Peste des petits ruminants*

1. Goat (markedly evident)
2. Sheep (less susceptible)
3. White tail deer ( less susceptible)
4. Cattle ( subclinical)



## Transmission - *Peste des petits ruminants*

Transmission primarily through

- Direct contact
- Contaminated food
- Water
- Beddings & other appliances.
- Secretion & excretions from affected animal

# CLINICAL SIGNS - *Peste des petits ruminants*

Incubation period is 3-10 days.

## Acute form :

- Sudden rise in body temperature (40-41°C)
- Restlessness, dull coat, dry muzzle, loss of appetite
- Serous nasal discharge becoming mucopurulent and resulting, at times in a profuse catarrhal exudate leads to respiratory distress
- Small areas of necrosis on the visible nasal mucous membrane
- Necrotic stomatitis with halitosis is common
- Severe non-haemorrhagic diarrhoea
- Bronchopneumonia evidenced by coughing is a common feature
- Abortion
- Dehydration, emaciation, dyspnoea, hypothermia and death within 5-10 day

## *Peste des petits ruminants*



## *Peste des petits ruminants*





## ***Peste des petits ruminants***



# LESIONS - *Peste des petits ruminants*

- Emaciation, conjunctivitis, erosive stomatitis inside the lower lips and the free portion of the tongue
- **'Bran like deposits'** on oral mucosa
- Lesions on the hard palate, pharynx and upper third of the oesophagus in severe cases
- Small streaks of haemorrhages and erosions in the first portion of the duodenum and the terminal ileum
- Extensive necrosis and sometimes severe ulceration of Peyer's patches
- **'Zebra stripes'** of congestion in the posterior part of the colon
- Small erosions and petechiae on the nasal mucosa, turbinates, larynx and trachea
- Bronchopneumonia
- Congestion, enlargement and oedema of most of the lymph nodes



## *Peste des petits ruminants*

### **DIFFERENTIAL DIAGNOSIS**

- Contagious caprine pleuropneumonia
- Rinderpest
- Bluetongue
- Pasteurellosis **LESIONS -**
- Contagious ecthyma
- Foot and mouth disease
- Heartwater
- Coccidiosis
- Mineral poisoning

# DIAGNOSIS OF PPR

## 1. Identification of the agent :

- ***Antigen detection*** -
  - Agar gel immunodiffusion
  - Counter immunoelectrophoresis
  - Indirect fluorescent antibody test
  - ELISA
  - Immunohistopathology

## **MATERIAL COLLECTION** - *Peste des petits ruminants*

- Swabs of the conjunctival discharges and from the nasal, buccal and rectal mucosa
- Whole blood collected on heparin (blood and anticoagulant should be mixed gently)
- Lymph nodes, especially the mesenteric and bronchial nodes
- Spleen , large intestine and lungs

**NOTE-** Samples should be transported under refrigeration

# Diagnosis - *Peste des petits ruminants*

## Virus isolation and identification \_

- **CELL LINE-**

- Primary lamb kidney cells or VERO cell line

- Virus neutralization

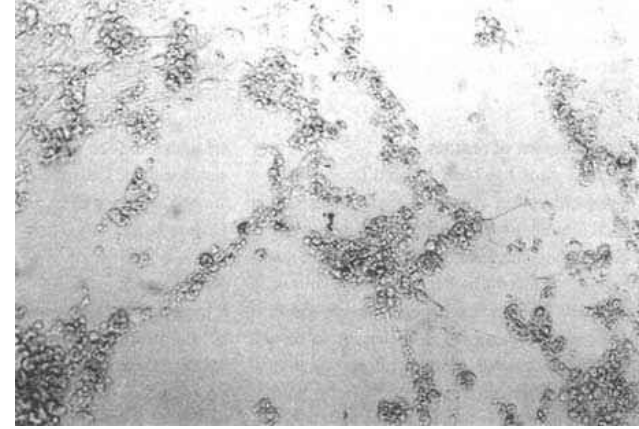
- Electron microscopy

## Virus RNA detection \_

- PPR-specific cDNA probes

- Amplification by RT-polymerase chain reaction (RT-PCR)

## *Peste des petits ruminants virus* - **CPE**



- The CPE produced by PPRV can develop within
- Cell rounding and aggregation culminating in **syncytia formation** in lamb kidney cells.
- In **Vero cells**, it is sometimes difficult to see the syncytia. If they exist, they are very small. However, in stained, infected Vero cells, small syncytia are always seen.
- Syncytia are recognised by a circular arrangement of nuclei giving a '**clock face**' appearance. Cover-slip cultures may give a CPE earlier than day 5. There are also intracytoplasmic and intranuclear inclusions.

# *Peste des petits ruminants*

## RT-PCR

- The primers were designed based on the published
- **N gene sequence of PPR virus**
- (Genbank accession number L39878) as follows:

Primer 1 **N gene** sequence (Forward primer):

5'-AAGGCGCCATGGCGACTCTCCTCAAAAG-3'

Primer 2 (Reverse primer):

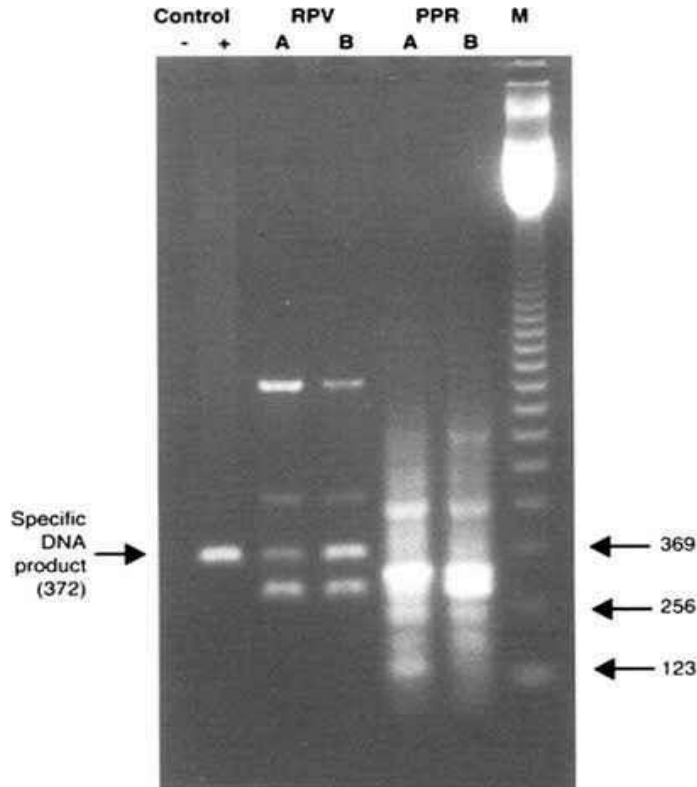
5'-AAGAGCTCTCAGCTGAGGAGATCCTTGT-3'

- A PPR specific set based on the equivalent



# *Peste des petits ruminants*

## RT-PCR



# Diagnosis - *Peste des petits ruminants*

## 2.Serological tests :

- Virus neutralisation
- Competitive ELISA
- Counter immunoelectrophoresis
- Agar gel immunodiffusion
- Immunodiffusion inhibition test



## *Peste des petits ruminants*

### **PREVENTION AND CONTROL**

- Strict sanitation & hygienic measures to be adapted in flock.
- Quarantine measures should be followed.

#### **Medical prophylaxis**

- Rinderpest vaccine is commonly used
- A homologous PPR vaccine is also available and is preferable.
- Both vaccines give strong immunity

**THANKS**



[www.veterinarymicrobiology.in](http://www.veterinarymicrobiology.in)

