



WOAH Reference Laboratory for
Peste des Petits Ruminants



中国动物卫生与流行病学中心
China Animal Health And Epidemiology Center



FAO Reference Centre for
Peste des Petits Ruminants

Control of PPR through vaccination and sero-monitoring in China

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8th PPR-GREN Meeting, 25-27 Nov, 2025, Qingdao, PR.China

Outlines

- Brief Introduction to the PPR history in China
- Mandatory Vaccination Program
- Post Vaccination Evaluation



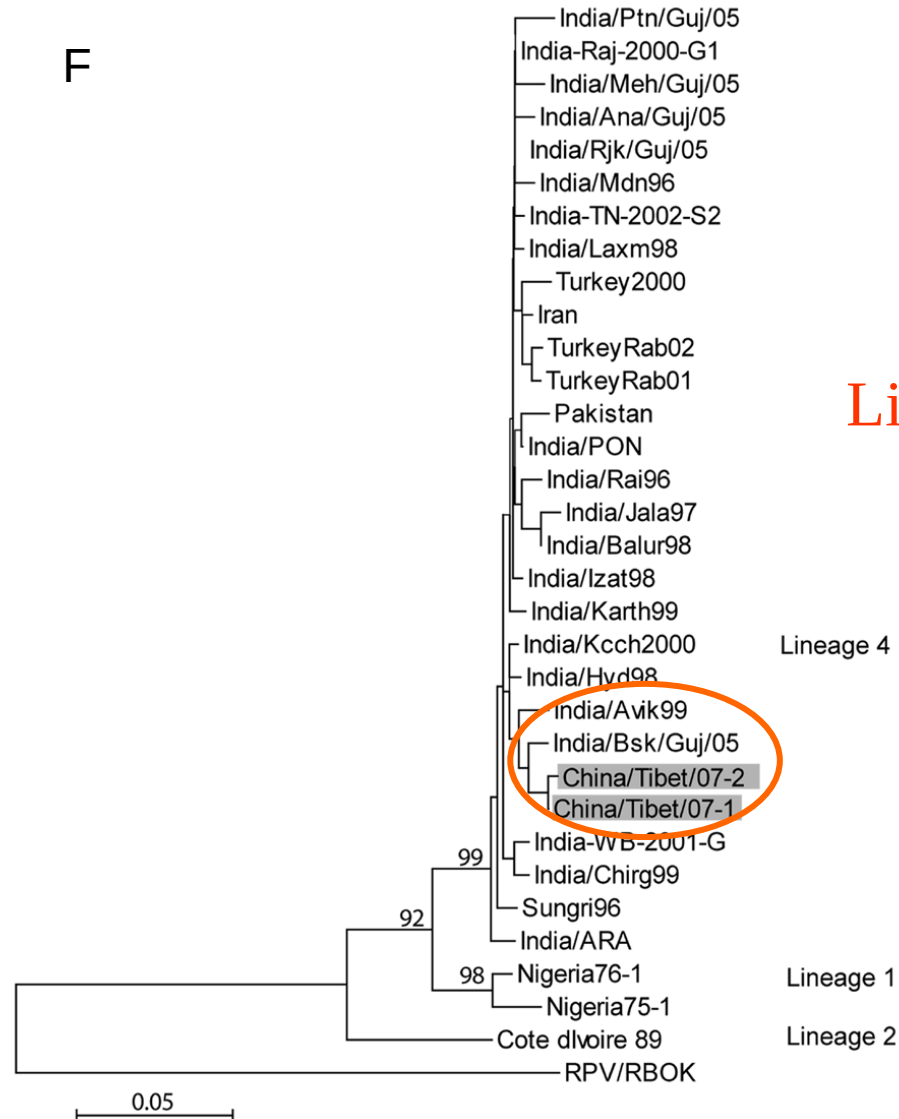
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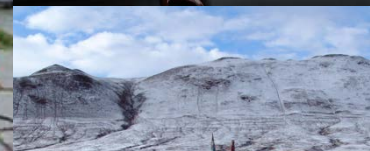




Phylogenetic analysis



Lineage IV close to Indian strains



Field Investigation



Epidemiology: Spatial-temporal evolution

Largely based on
serological and
molecular evidence from
field investigation

Serological and Virological tests

ELISA	1721
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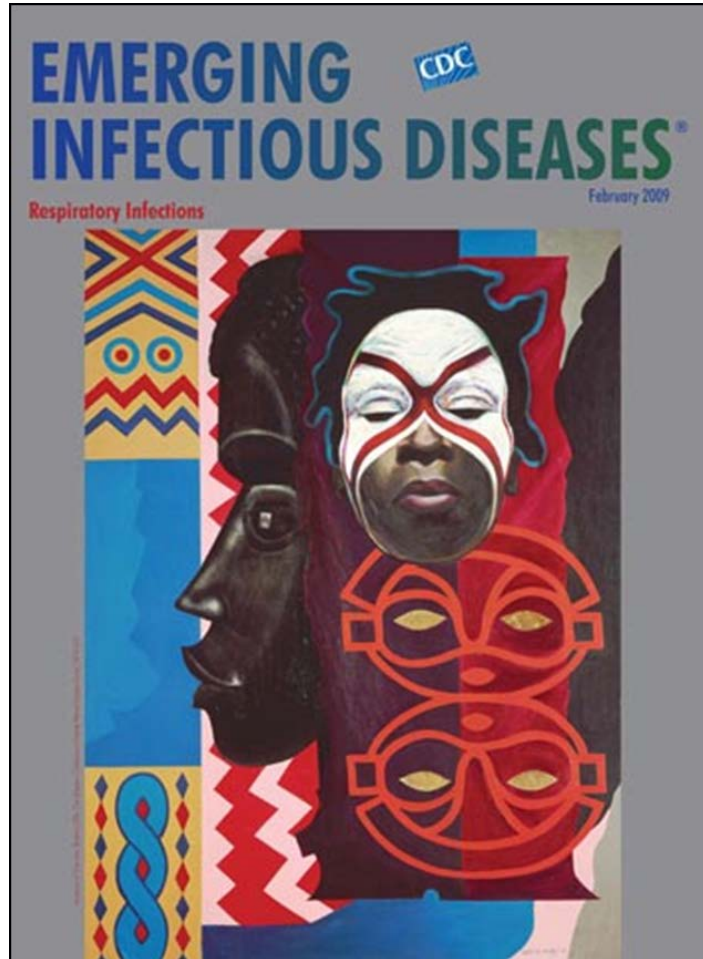
Infected zone	279 / 837
Out of the zone	0 / 884

PCR	51
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Positive	35 / 51
Sequencing	19 / 35



Publications on the First Outbreak



Peste des Petits Ruminants Virus in Tibet, China

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Serologic and molecular evidence indicates that peste des petits ruminants virus (PPRV) infection has emerged in goats and sheep in the Ngari region of southwestern Tibet, People's Republic of China. Phylogenetic analysis confirms that the PPRV strain from Tibet is classified as lineage 4 and is closely related to viruses currently circulating in neighboring countries of southern Asia.

Peste des petits ruminants (PPR) is an acute and highly contagious viral disease contracted by small ruminants such as goats and sheep causing high rates of illness and death. The disease is endemic in parts of sub-Saharan Africa, the Middle East, and Asia. The PPR virus (PPRV) genome consists of 4 lineages (1,2). PPRV infection was officially reported in the Ngari region of western Tibet, People's Republic of China, in July 2007. Our study assesses the prevalence of PPRV infection in goats and sheep by region in Tibet. We also characterize strains of the virus by comparing part of the genome sequences with other PPRV sequences available in the GenBank database.

The Study

Small ruminants in regions throughout Tibet were examined for PPRV antibody from July 2007 through November 2007. The sampling procedure focused on 3 groups of animals. The first comprised 718 animals in 4 counties (Rutog, Ge'gyai, Gexue, and Zada) in the Ngari region, where animals having clinical signs of PPRV infection had been reported by local authorities. The second group included 208 animals in Gar and Bulang counties in the same region and in 2 counties bordering the Ngari region (Nyima in Nagu region and Zhonglu in Shigatse region). The third group comprised 520 animals in 5 counties within 3 separate regions (Nyima and Yading in Shigatse region, Gexue and Lushan in Shannan region, and Zayu in Nyingchi region).

Author affiliations: China Animal Health and Epidemiology Center, Qinghai People's Republic of China (Z. Wang, J. Bao, X. Wu, Y. Liu, L. Li, C. Liu, Z. Xia, W. Zhao, W. Zhang, N. Yang, J. Li, S. Wang, J. Wang), and Tibet Center for Animal Disease Control, Lhasa, Tibet, People's Republic of China (J. Suo).

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Table 1. PPRV antibody in animals sampled in Tibet, China, 2007*

Region	County	No. samples	No. (%) positive
Ngari	Gexue	131	59 (45.0)
	Ge'gyai	214	90 (42.1)
	Rutog	209	122 (58.4)
	Zada	64	0
	Gar	50	0
Nyingchi	Bulung	65	0
	Shyri	60	0
Nyima	Nyima	60	0
	Nyima	120	0
Shigatse	Yading	60	0
	Zhonglu	120	0
Shannan	Gexue	135	0
	Lushan	135	0
Total		1,538	271 (17.6)

We examined 1,538 animals (771 goats and 767 sheep) and collected serum samples from each. A competitive ELISA that used a monoclonal antibody to the N protein (3) identified 271 animals (17.6%) having antibody to PPRV. The PPRV-positive sera were collected from Rutog (122/209), Gexue (59/131), and Ge'gyai (90/214) counties in the Ngari region (Table 1). Rates of PPRV infection were higher in goats than in sheep. Of 763 goats examined in the Ngari region, 262 (34.5%) were seropositive for PPRV. The highest seroprevalence (61.6%, 121/198) was found in goats in Rutog County. Only 8 (1.1%) of 73 sheep examined in the Ngari region were seropositive for PPRV (Table 2).

Field samples, including organs (lymph node, spleen, lung, and intestine) and swab specimens, were obtained from 49 goats and sheep suspected of being infected with PPRV. These animals inhabited 4 counties in the Ngari region (Ge'gyai n = 33, Zada n = 7, Gexue n = 5, and Rutog n = 4). Two reverse transcription-PCR (RT-PCR) and 1 newly developed and validated real-time quantitative RT-PCR (qRT-PCR) were conducted to determine whether the animals had viral RNA (4–6). The first RT-PCR (N RT-PCR), which amplified a 351-bp fragment in the N protein gene, detected virus in 28 samples. The second RT-PCR (F RT-PCR), which amplified a 448-bp fragment in the F protein gene, detected virus in 27 samples. The qRT-PCR detected virus in 37 samples. Use of qRT-PCR and 1 of the 2 RT-PCRs showed that 31 animals were found to contain viral RNA. In goats, 23 (73%) of the 30 samples contained viral RNA, and 2 (29%) of 7 sheep samples contained viral RNA. Most (51%) infected animals showed a high viral load with individual cycle threshold (Ct) values <30. At least one third (29%) had a moderate viral load (Ct 30–35), and 10% had a Ct value >35. The distribution of Ct values differed slightly according to the infected animals' origins. All animals from Gexue County had low Ct values (Ct 19–

*These authors contributed equally to this article.

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试验研究

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我国首例小反刍兽疫诊断报告

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摘要: 2007年7月我国西藏发生不明山羊疫情, 国家外来动物疫病诊断中心对当地动物疫病控制中心送检的14只病死山羊病料和一批血清样品分别进行病原学和血清学检测。利用较敏感的小反刍兽疫病毒(PPRV)特异性荧光定量 RT-PCR 方法, 在 11 只山羊组织中检测到小反刍兽疫病毒核酸。利用常规 RT-PCR 方法, 从 8 只山羊组织中检测到 PPRV 核酸。针对 1 号样本病原核酸 N 基因和 F 基因片段进行遗传分析, 该病原属于 4 系。利用竞争 ELISA 试剂盒对送检的 13 份血清样本进行检测, 结果全部阴性。将 1 号组织样本接种 Vero 细胞分离病毒, 透射电镜观察下, 发现了 500 纳米左右的病毒粒子。对分离病毒进行 RT-PCR 检测, 证实其为 PPRV。

关键词: 小反刍兽疫, 荧光定量 RT-PCR, 普通 RT-PCR, 病毒分离, RT-PCR 特异性抗体

Diagnosis of the First Outbreak of Peste des Petits Ruminants in Tibet

Abstract: The first outbreak of peste des petits ruminants in China in 2007 was confirmed by multiple laboratory tests including one step real-time RT-PCR, conventional RT-PCR, competitive ELISA and virus isolation. The presence of PPRV was detected in 11 out of 13 field samples by one step real-time RT-PCR, among which 8 were further detected by conventional RT-PCR. Sequence analysis showed that the virus fell into the lineage 4 which was epidemic in Middle East and certain western Asian countries. One strain of PPRV was isolated in Vero cell lines from field samples and confirmed by RT-PCR test.

Keywords: Peste des petits ruminants, Real-time RT-PCR, Conventional RT-PCR, Virus isolation, PPRV-specific antibody

小反刍兽疫(PPR)是感染小反刍兽(特别是山羊和绵羊)的严重的烈性、接触性传染病, 临床上以高热、坏死性肺炎、胃肠炎和肺炎为特征, 发病率和病死率分别高达 100% 和 90%。该病流行于非洲赤道到撒哈拉地区, 大部分中东国家和印度、尼泊尔、塔吉克斯坦等与我国接壤的国家。近年来呈蔓延趋势, 包括牲畜、野兔、野马等上从来没有发生的记载。但至今为止, 我国尚未发现该病。

2007 年 7 月 8 日国家外来动物疫病诊断中心接到西藏地区发生不明山羊疫情的报告并收到采集的样品。病羊主要症状: 高热, 甚至失明; 浆液性和脓性鼻液; 体温高达 40–41°C; 严重腹泻。剖检变化: 肺炎变化, 支气管炎和肺脏出现干酪样灶灶, 肺脏表面, 支气管黏膜, 肾脏、肠系膜

等器官充血、水肿、出血。国家外来动物疫病诊断中心接到样品后立即进行病原学和血清学检测。结果表明, 本病是由小反刍兽疫病毒引起, 是历史上首次在我国发现。现将诊断报告如下。

材料和方法

1.1 病料 分别将 14 只山羊的剖检病料, 包括肺脏、脾脏、淋巴结等器官组织送检。

1.2 血清 西藏地区送检发病后期羊 13 份血清, 1 份来自外表健康羊, 2 份来自临床发病后期的羊。

1.3 病料组织总 RNA 提取 在加有酶的裂解液匀浆 (Roche 公司产品) 中, 加入 600 μL RLT 缓冲液 (RNeasy mini Kit 试剂盒, QIAGEN 公司产品), 加入剪碎的肺脏或淋巴结组织 30 mg, 使用 RNeasy RiboLysor 组织匀浆机进行匀浆处理, 再使用 RNeasy mini Kit 试剂盒提取总 RNA。

1.4 荧光定量 RT-PCR 用本实验室建立的 PPRV 特异的荧光定量 RT-PCR 方法进行小反刍兽疫病毒 N 基因片段的扩增。PPRV 特异性引物为 PPRNP/PPRNP, TaqMan 探针为 PPRNP, 产物长度为 115 bp。同时以本室合成的 cRNA 标准品为阳性对照。

1.5 普通 RT-PCR 根据参考文献合成 PPRV 特异性引物 NP3/NP4, 利用提取的 RNA 进行 PPRV N 基因的特异性 RT-PCR 扩增。同时以本室合成的 cRNA 标准品作为阳性对照。RT-PCR 产物大小为 351 bp。另外, 根据参考文献合成 PPRV 特异性引物 PPRV F1/PPR F2, 利用提取的 1 号山羊肺脏组织 RNA 样本进行 F 基因的特异性 RT-PCR 扩增, RT-PCR 产物大小为 372 bp。

1.6 RT-PCR 产物的序列测定 利用乙醇沉淀法对 1 号山羊肺脏组织 RNA 样本 N 基因和 F 基因

*通讯作者

前五位作者对本文具有相同贡献

Critical Control Measures

- Stamping out of all infected herds
- Vaccination (N 75/1) in all affected districts in Tibet region
- Establishing a buffer zone by ring vaccination along the neighboring counties
- Movement control of SR and products

Only one outbreak at a boundary village in May 2010,
no other outbreaks during 2009-2013



The second PPR Outbreaks

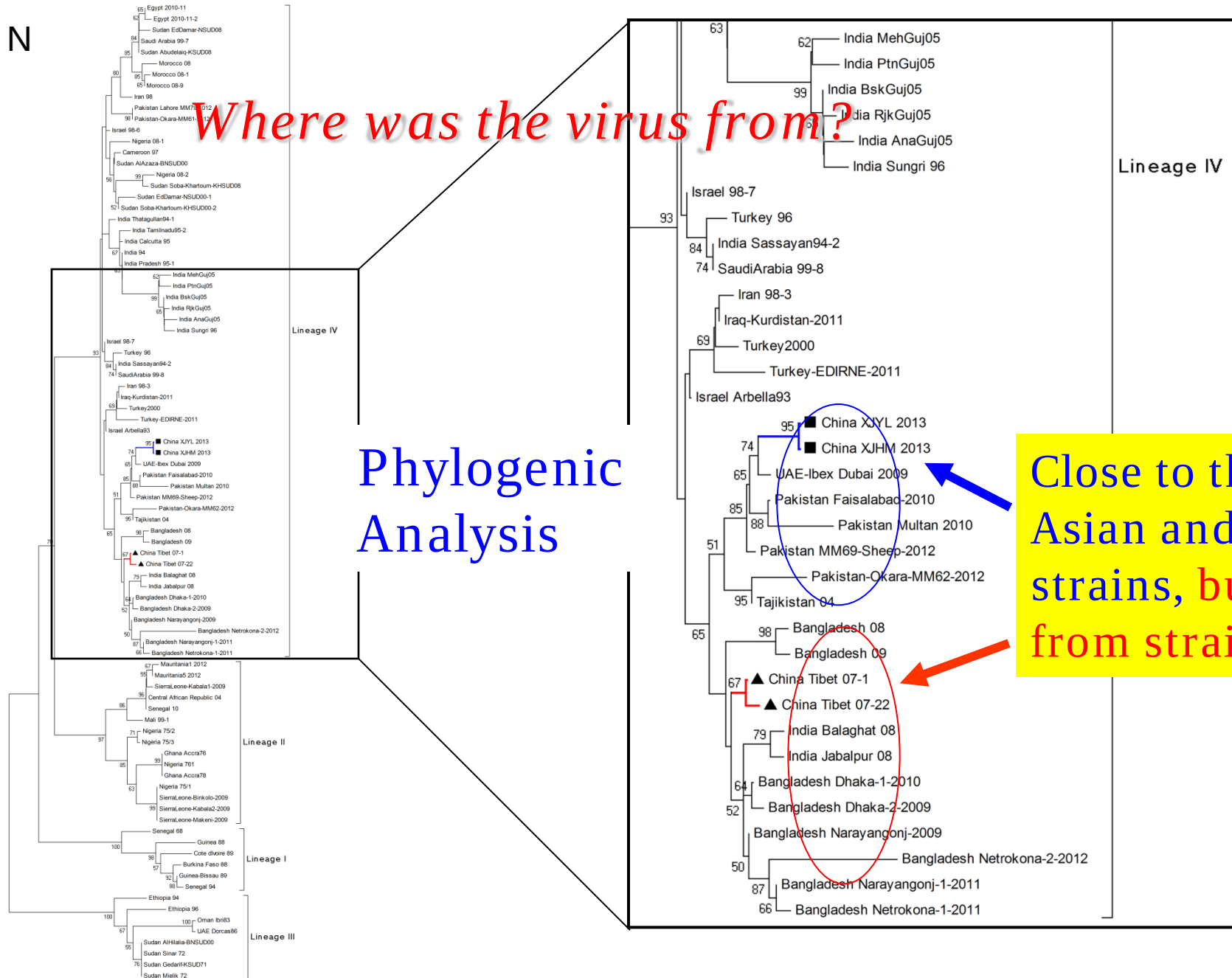
—Dec 2013 to May 2014



N

Where was the virus from?

Phylogenetic Analysis





Critical Control Measures

- Stamping out of all infected herds
- Vaccination campaign in most provinces
- Temporarily ban the movement of live SR, except for slaughter
- Temporarily close all SR markets within the Country

Outlines

- Brief Introduction to the PPR history in China
- **Mandatory Vaccination Program**
- Post Vaccination Evaluation



National Mandatory Vaccination Program

Regulated by

- **Law on Animal Epidemic Prevention, P.R. China**
- **National Guidelines for Mandatory Vaccination against Animal Diseases (revised every five year)**
- **National Technical Guidelines for Animal Disease Vaccination (reviewed annually)**
- **Quality Standards for Veterinary Bio-products**
- **National Animal Disease Surveillance Plan**
- **Technical Specification for the Prevention and Control of PPR**

Guidelines for Mandatory Vaccination



- All eligible sheep and goats should be vaccinated, except those in a establishing or established PPR free zone or compartment.
- Mass vaccination conducted yearly in Spring and/or Autumn, with the catch-up vaccination in following months. (since Dec. 2013)
- Herd immunity level: no less than 70% individuals are sero-positive
- Each province may develop their own vaccination program based on their conditions and in accordance with the national guidelines.

Guidelines for Mandatory Vaccination

➤ Financial Supports

- All farms can receive PPR vaccine and vaccination service from the local governments for free.
- SAV (Subsidy After Vaccination) : An extra choice for large intensive farms.
- Local government may procure vaccination services from a third-party in case of HR shortage.

Vaccine Production and Quality Control

Supervised and authorized at batch level
by China Institute of Vet Drug Control (IVDC)

➤ PPR Live Vaccine

PPRV N75/1 $\geq 10^3 \text{TCID}_{50}$ per dose

➤ PPR and GP Combined Live Vaccine

PPRV N75/1 $\geq 10^3 \text{TCID}_{50}$ per dose

GPV AV41 $\geq 10^{3.5} \text{TCID}_{50}$ per dose



Other Attributes and Requirements

Supervised and Delivered by Local Vet Authorities

➤ **Storage, Transport and Usage**

$\leq -20^{\circ}\text{C}$

Cold Chain

Diluted and kept on ice, used within 1 hour avoid UV light

➤ **Target Species**

Healthy sheep and goats

➤ **Administration Route**

Subcutaneous injection at the neck (1 ml)

➤ **Shelf Life**

24 months

➤ **Duration of Immunity**

36 months

Vaccination Schedules (From National Guidelines)

Farming Type	Vaccination Schedule
Intensive Farm	-Initial vaccination at 2-3 months of age, to avoid maternal antibody -Annual booster in case of high risk -Vaccination schedule can be adjusted as per herd immunity and maternal antibody levels
Backyard/Smallholder	-Mass vaccination of the adults annually, in the spring or autumn. -Catch-up vaccination: Individuals not fit for mass vaccination should be vaccinated in the following months -Local government may intensify the schedule based on risk.

Outlines

- Brief Introduction to the PPR history in China
- Mandatory Vaccination Program
- **Post Vaccination Evaluation**



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Post Vaccination Evaluation

Rationale

- PPRV sero-positive individuals are deemed to be protected.
- Threshold for successful vaccination is 70% of the individuals in an epi-unit are sero-positive after 21 days post vaccination.

Policy – *National Surveillance Plan*

- **Provincial Level**

Conducted by local authorities annually, in Spring and/or Autumn

- **National level**

Conducted by national labs annually, in Autumn

农业农村部文件

农牧发〔2021〕11号

农业农村部关于印发《国家动物疫病监测与流行病学调查计划(2021—2025年)》的通知

各省、自治区、直辖市及计划单列市农业农村(农牧、畜牧兽医)厅(局、委),新疆生产建设兵团农业农村局,部属有关事业单位,各有关单位:

为做好非洲猪瘟等动物疫病防控,持续加强监测和流行病学调查工作,我部组织制定了《国家动物疫病监测与流行病学调查计划(2021—2025年)》,现印发你们,请遵照执行。

农业农村部
2021年4月13日

Minimum Sample Size Requirements at National Level

➤ Epi-units

100 units/province

Calculations are based on a 95% level of confidence and 10% standard error, assumed that at least 50% of vaccinated epi-units will have at least 70% of its population sero-positive.

➤ Animals

9 animals/epi-unit

Assumed that sero-negative prevalence within each epi-unit is at least 30% , with 95% confidence intervals.

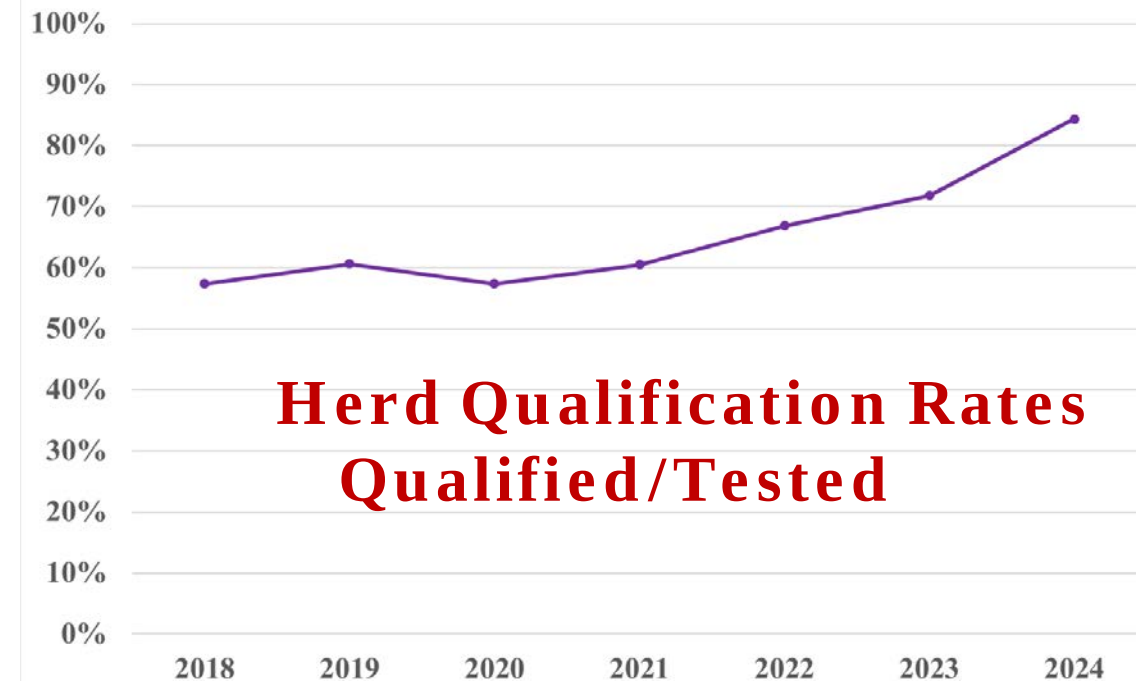
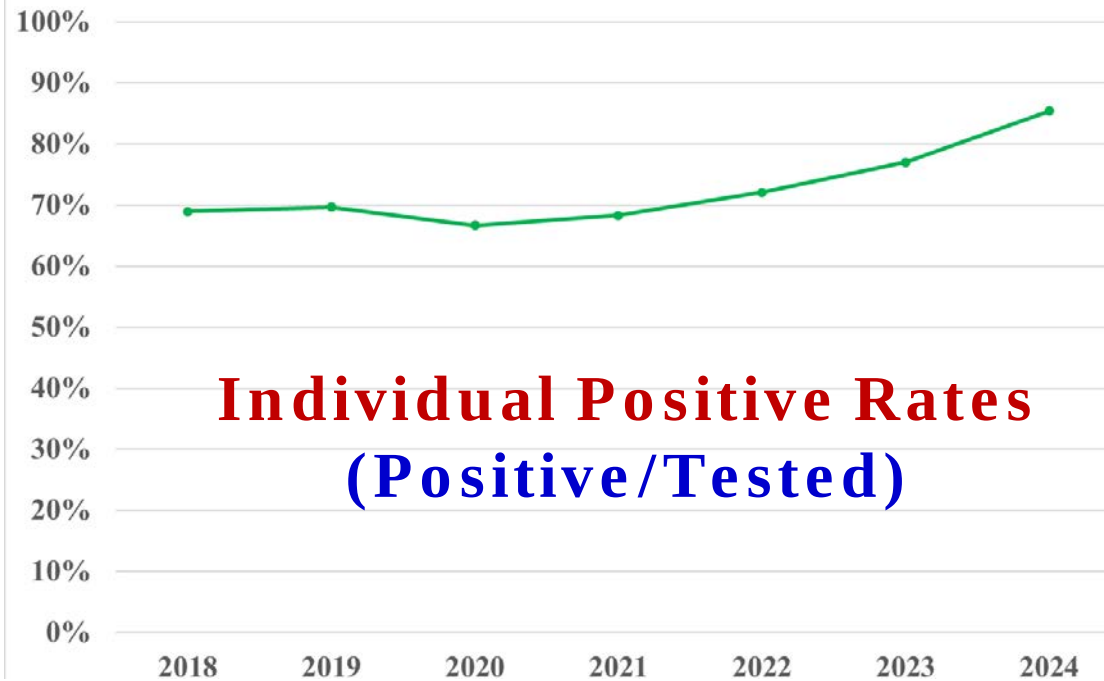
➤ Sampling in Practice

Number animals in an epi-unit	≤6	7-10	11-25	26-55	≥56
Sample size	all	≥6	≥7	≥8	≥9

➤ PPRV Antibody Tests

- Blocking ELISA or Competitive ELISA

Results of sero-surveillance at national level (2018-2024)



Looking into the Future

- **Big Country with many priorities**
- **Big number of sheep and goats**
- **Big challenge to movement control after vaccination**
- **Big number of neighboring countries**
- **When and how to cease vaccination still needs to be addressed.**

PPR vaccination

- **Big number of sheep and goats**
- **One of the key tools available for PPR control**
- **Not sufficient but normally necessary for eradication**
- **A higher herd immunity pursued and lower incidence expected**



Thank you for your attention

