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Evaluation of PPR Seroconversion in the Provinces of N'Djamena, Chari Baguirmi, Hadjar Lamis, Bahr Elgazal, and Lac

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Abstract

Objective: The objective of this study was to evaluate the post-vaccination seroconversion of small ruminants after the 2022-2023 vaccination campaign against peste des petits ruminants, as part of the national PPR control and eradication strategy. **Methodology and Results:** The study was conducted in the study area comprising the provinces of Chari-Baguirmi, N'Djamena, Bahr Elgazale, Kanem and Lac. It involved the collection of 1687 samples of sera from small ruminants, which were analysed in the laboratory using the Eliza competition diagnostic method. Statistical analyses of the data obtained were carried out using the R Studio 4.1.2 software. The data analyses made it possible to assess post-vaccination seroconversion at the level of the study area, regions and municipalities, as well as to see the effect of sex, marking and the "vaccinated and unvaccinated animal" aspect on PPR seroconversion. At the level of the study area, an overall seroconversion of 73.56% (higher than the rate set by the national strategy) was obtained, but it was distributed differently between provinces and municipalities. **Conclusion and Application of Results:** Provinces such as Bahr Elgazal (69.84%) and Lac (68.78%) experienced a seroconversion to decaf of the rate set by the national strategy for the control and eradication of PPR. The municipalities with low seroconversion rates were the 1st district of the city of N'Djamena (54.29%) and Amsileb (49.02%). Thus, it would be suggested that the General Directorate of Veterinary Services, through its Animal Health Directorate, intensify its efforts in municipalities with a low seroconversion rate and maintain the gains made in those with high rates. In this context, it is important to ensure that vaccines are administered appropriately and that the cold chain is functioning properly to achieve the objective of the national strategy for the control and eradication

of peste des petits ruminants in Chad.

Keywords

Plague, Small Ruminants, Seroconversion, Post-Vaccination, Chad

1. Introduction

Chad has an estimated herd of 91,522,170 small ruminants. These livestock provide livelihoods and food security as well as employment opportunities for 300 pastoralist families [1].

However, the Chadian herd is subject to multiple constraints that hinder its development. Among these, the pathological constraints are not the least. In this case, the constraints related to peste des petits ruminants, which is enzootically rampant on the national territory. Peste des Petits Ruminants (PPR) is a highly contagious transboundary viral disease of small ruminants, considered to be one of the most damaging to livestock. PPR now affects nearly 76 countries in Africa, the Middle East and Asia [2], China, most of the Indian subcontinent and some European countries, such as Türkiye [3]. The risk period for PPR is between January and April, with a peak observed in February. The key risk factors for PPR are the environment in which the farm is located, the introduction of new animals into the farm, the gathering of animals to pasture or water together, the wandering of animals and transhumance with respective opinion percentages of 54%, 33%, 27%, 22% and 7%. PPR is generally characterized by the onset of depression, mouth sores, respiratory problems [4], fever, oculonasal discharge, stomatitis, diarrhoea and pneumonia with foul-smelling breath leading to death [5]. It can lead to high morbidity and mortality rates, reaching up to 100% and more than 90% respectively in naïve herds. Mortality occurs between 5 and 10 days after infection, and convalescent animals develop strong lifelong immunity. In Chad, the disease has been endemic since 2013, and the isolation of its virus was carried out in 2014 [6] [7]. PPR control mainly relies on vaccination, isolation of infected animals and restriction of animal movements. As a prelude to the global and pan-African strategies for the control and eradication of PPR, Chad has set up its national strategy for the control and eradication of this disease on the model of these two strategies. Given that vaccination is the key element in the control and eradication of PPR [8] [9], Chad, through the Ministry of Livestock and Animal Production (MEPA), organized two initial mass vaccination campaigns of small ruminants throughout the national territory, in 2017 and 2018 respectively. This was followed by two more mass campaigns in 2019 and 2023. Chad has made vaccination against PPR compulsory (Order No. 009 of March 24, 2023) [10]. During the last mass vaccination campaign against PPR, 9,485,373 small ruminants were vaccinated throughout the national territory [1].

This study is undertaken with a view to estimating the seroconversion of PPR

Virus (PPRV), through the analysis of small ruminant sera collected in the field from sheep and goats in the study area consisting of the provinces of N'Djamena, Chari-Baguirmi, Bahr El Gazal, Kanem and Lac. The small ruminant herd in this area is estimated at 4,881,959 head in 2022. The 2022-2023 mass vaccination campaign reached 2,155,649 heads of small ruminants [1].

2. Material and Methods

2.1. Study Area

The study area as presented in (Figure 1) is made up of the provinces of N'Djamenna, Chari Baguirmi, Hadjar Lamis, Bahr Elgazal and that of Lac.

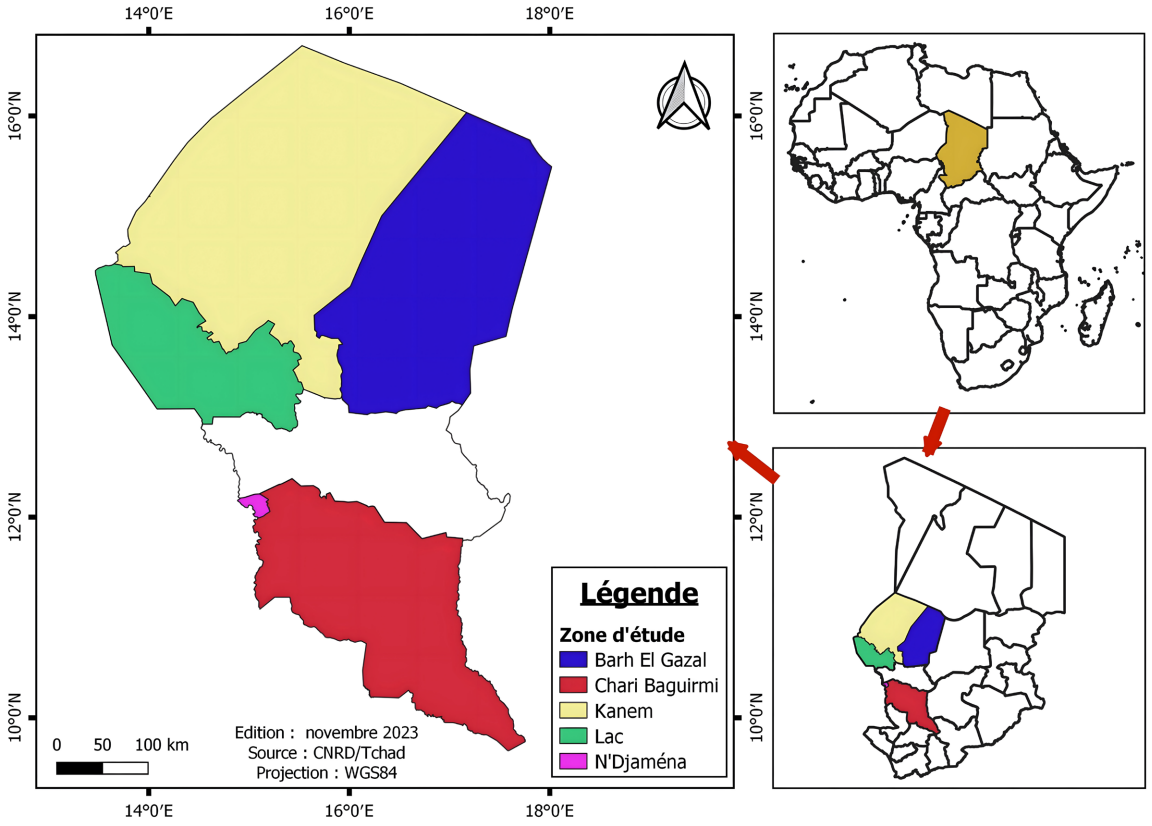

Figure 1. Map of the study area (colors green, light gray, blue, and red).

2.2. Duration of the Study

The duration of the study is four months (August-November 2023).

2.3. Methodology

Vaccination is considered effective, and the small ruminant population is considered to be protected against transmission of the PPR virus if post-vaccination immune coverage is greater than 70%. To measure this immune coverage, it is proposed to calculate the proportion of Protected Epidemiological Units (EPUs) (*i.e.* those for which at least 70% of animals are positive for PPR antibodies). If the

proportion of protected EUs is greater than 70%, then the immune coverage (whether natural or post-vaccination) can be considered sufficient to stop the circulation of the virus, provided that the risk of introduction of the disease is controlled. In the context of the post-vaccination assessment, the ideal EU is the largest spatial unit within which the probability of vaccination can be considered identical. However, the lack of available data forces us to fall back on an arbitrary spatial unit, such as an administrative unit. The EU population concerned by the post-vaccination evaluation (target population) is all the UEs in the Province in order to be able to compare vaccination coverage with immune coverage for protected UEs [8].

2.4. Study Type

This is a descriptive and cross-sectional study that was carried out in two stages: the first stage consists of collecting sera samples from small ruminants and the second stage is that of the analysis of these samples at the IRED serology laboratory using the ELIZA competition method.

2.5. Sampling Type and Sample Size

The sampling used is that developed by CIRAD at PRAPS. It is a simple random sampling stratified by epidemiological units (epidemiological unit 1: the commune and epidemiological unit 2: the village or the herd)

The inclusion criteria are sheep and goats over 3 months of age, sedentary or transhumant without distinction of breed and vaccinated or unvaccinated.

Herds will be randomly selected from each village and/or camp to collect sera from the target animals through the following cohort:

- Pets 0 - 1-year-old;
- Pets aged 2 - 3 years;
- Animals aged 4 - 5 years;
- Pets over 5 years of age.

2.6. Sample Size per Stratum

The sample size for stratum 1 is 17 communes and that for stratum 2 is 52 villages or camps. According to the methodology used by the PRAPS, 32 animals will be taken per village or camp. The calculated sample size is therefore 1612 sera to be collected. But since the breeders were cooperative, the size of the serums collected was 1687.

2.7. Conduct of the Study

The animals are drawn at random from at least three different herds in the village or camp. The collection of blood samples is carried out after having the approval of the owner.

Blood samples are taken in dry tubes using the "venoject" device consisting of a needle and a needle holder. The blood test takes place by puncturing the jugular

vein. The tubes containing blood are left at room temperature for 2 hours. Once the clots are detached from the wall of the tube, they are removed. After removal of the clots, the contents of the tube are transported to the cold laboratory where the serum is obtained by centrifugation at 3000 rpm for 15 minutes. The serums are then frozen at minus 20˚ until they are analysed at the IRED serology laboratory in N'Djamena. The analysis of the sera was carried out using the competition ELISA method against PPR virus antibodies according to the protocol specified by the manufacturer iD.Vet.

2.8. Statistical Analyses

The ELIZA competition test data (Optical Densities, ODs) of the serum samples are exported in Excel format by the ELIZA reader software. However, the Excel spreadsheet and the R Studio 4.1.2 software allowed us to carry out descriptive analyses of these data, the results of which are presented in the form of frequency tables and histograms.

3. Results and Discussion

The results of PPR seroconversion across the study area are presented in Table 1. Out of a total of 1687 sera analyzed, 1246 are positive. The post-vaccination seroconversion for the study area was 73.56%.

Table 1. PPR seroconversion in the study area.

Study area	Positive	Negative	Total	Seroconversion (%)	IC
Samples	1241	446	1687	**73.56**	[0.71; 0.75]

This result is in line with the rate set by the national PPR control and eradication strategy as well as with the sub-regional and international PPR control and eradication strategies. This rate is higher than that obtained in Senegal (67.3%) [11], that obtained in Niger (73%) [12], Tanzania (71%) [13] and Rajasthan (54.95%) [14]. However, this rate is lower than that obtained in Burkina Faso (76.3%) [15].

Table 2. PPR post-vaccine seroconversion by province.

Provinces	Samples	Positive	Negative	Seroconversion (%)	IC
Char Baguirmi	422	307	115	72.74	[0.68; 0.76]
N'Djamena	214	151	63	70.56	[0.63; 0.76]
Bahr Elgazal	378	264	114	69.84	[0.64; 0.74]
Kanem	359	303	56	84.40	[0.79; 0.87]
Lac	314	216	98	68.78	[0.63; 0.73]

The results of PPR seroconversion at the provincial level are presented in Table 2. The post-vaccination seroconversion of PPR by province is not homogeneous

and has some differences between them. The results indicate a distribution that varies between 68.67% and 84.21%. Thus, 4 out of 5 provinces have a rate of more than 70% and one province whose seroconversion is less than 70%. To this end, the results obtained for the Provinces of Chari-Baguirmi, N'Djamena, Kanem and Lac meet the first indicator of the 3 PPR control and eradication strategies (national, sub-regional and international strategy), all of which aim to achieve a seroconversion greater than or equal to 70%. The rates obtained respectively of 68.78% and 69.84% in Lac and Bahr Elgazal are close to that set by the national strategy for the control and eradication of PPR. This difference could be linked to constraints during the execution of vaccination campaigns.

The results of PPR seroconversion according to sex presented in Table 3 show that the seroconversion of males exceeds the rate set by the MEPA and is 72.44%, which is lower than that of females (73.36%). These results show that sex does not influence PPR seroconversion (p-value greater than 0.05).

Table 3. Sex-specific seroconversion.

Sex	Positive	Negative	Total	Seroconversion (%)	IC
Male	205	78	283	72.44	[0.31 - 0.45]
Females	1030	374	1404	73.36	[0.33 - 0.38]
Total	**1235**	**452**		**1687**	

The results of seroconversion according to the performance of the vaccination act are presented in Table 4. These results show a very significant difference between the seroconversion rate of vaccinated animals, which is 74.92% and that of the unvaccinated, which is 7.89%. The presence of immunized subjects among the unvaccinated could be a confusion on the part of the owners of the animals during the investigation, *i.e.* they were already in contact with the virus.

Table 4. Seroconversion according to the completion of the vaccination procedure.

Vaccination	Positive	Negative	Total	Seroconversion (%)	IC
Vaccinated	1233	416	1659	**74.92**	[0.72 - 0.76]
Unvaccinated	3	35	38	**7.89**	[0.02 - 0.22]
Total	**1236**	**451**	**1687**		

The results of PPR seroconversion according to the municipalities presented in Table 5 show that out of 17 communes, 7 have a rate above 80%, 4 have a rate above 71%, 4 have a rate above 61% and which is close to 70% (indicator of the national and global strategy) and 2 communes have the lowest rate: Amsileb with a rate of 49.02% and the first district of the city of N'Djamena with a rate of 54.29%. Seroconversion rates that are less than 70% could be due to the absence of some of the animals during vaccination campaigns, either due to the vaccination act which may not be well accomplished and could be due to technical constraints to ensure

the cold chain.

Table 5. Seroconversion by municipality.

Communes	Positive	Negative	Total	Seroconversion (%)	IC
1st Arrondissement	38	32	70	**54.29**	[0.41 - 0.65]
7th Arrondissement	64	7	71	**90.14**	[0.78 - 0.94]
9th Arrondissement	50	21	71	**70.42**	[0.57 - 0.79]
Amsileb	75	78	153	**49.02**	[0.40 - 0.56]
Dourbali	90	30	120	**75.00**	[0.65 - 0.81]
Isseirom	81	31	112	**72.32**	[0.70 - 0.86]
Kaiga Kindjiria	70	24	94	**74.46**	[0.63 - 0.81]
Kekedena	71	13	84	**64.89**	[0.73 - 0.90]
Kouno	61	33	94	**64.21**	[0.53 - 0.73]
Mai Aiche	80	15	95	**84.21**	[0.74 - 0.89]
Massenia	77	36	113	**68.14**	[0.58 - 0.76]
Moussoro	106	25	131	**80.91**	[0.72 - 0.87]
Ngarangou	66	42	108	**61.11**	[0.51 - 0.70]
Nokou	63	21	84	**75.00**	[0.64 - 0.83]
Rig Rig	83	12	95	**87.36**	[0.78 - 0.93]
Salal	84	11	95	**88.42**	[0.79 - 0.93]
Wadjigui	87	10	97	**89.69**	[0.81 - 0.94]
Total	**1246**	**441**	**1687**		

The results of seroconversion of marked and unmarked animals are shown in Table 6. Marked animals have a seroconversion rate of 71.67% while unmarked animals have a rate of 84.13%. These results are in line with the national, sub-regional and international strategy for the control and eradication of PPR. This high rate of seroconversion of unlabelled animals could be the result of previous vaccinations, during which the animals were not tagged, or it is a post-infection immunity.

Table 6. Seroconversion by marking.

Animals	Positive	Negative	Total	Seroconversion (%)	IC
Marqués	1060	419	1479	**71.67**	[0.69 - 0.74]
Non-marqués	175	33	208	**84.13**	[0.78 - 0.88]
Total	**1235**	**452**	**1697**		

The results of PPR seroconversion according to the age of the animals are presented in Table 7. These results show a PPR seroconversion of between 72% and 75%. These results are significantly higher than the rate set (70%) by the national

strategy for the eradication and control of PPR in Chad.

Table 7. Age-specific seroconversion.

Age (year)	Positive	Negative	Total	Seroconversion (%)	IC
0 - 1	286	93	379	**75.46**	[0.70 - 0.79]
2 - 3	625	235	860	**72.67**	[0.69 - 0.75]
4 - 5	266	97	363	**73.28**	[0.68 - 0.77]
>5	63	22	85	**74.12**	[0.63 - 0.82]
Total	**1240**	**447**	**1687**		

4. Conclusion

A total of 1687 sera were collected from 17 communes and 52 villages across the study area. These samples were analysed at the Institute of Research in Livestock for Development (IRLD) serology laboratory. The results obtained show a good performance of the effectiveness of PPR vaccination campaigns. The objectives set by the Ministry of Livestock and Animal Production, in its strategy for the control and eradication of PPR by 2030, have been partially achieved. It would be suggested that the General Directorate of Veterinary Services (GDVS), through its Directorate of Animal Health, intensify its efforts in municipalities with low seroconversion and maintain the achievements in those with seroconversion above the target of 70%. In this context, it is recommended that the GDVS to ensure that vaccines are administered appropriately and that the cold chain functions properly.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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