Epidemiology of Newcastle Disease in Village Poultry Farming in Ambatolampy, Madagascar

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Abstract: *A descriptive cross-sectional study was conducted in 2019 among 41 households in Ambatolampy district to describe the epidemiological profile of Newcastle disease in the site. For this purpose, an epidemiological survey with, ELISA serological tests of chicken blood samples were performed. The data from the survey and the test results were analyzed with multiple logistic regression to determine the factors associated with Newcastle disease. The serological test showed that Newcastle disease is devastating village poultry farming with a seroprevalence of 64% in chickens and 58.5% in households. Furthermore, logistic regression analysis confirmed that species mix (p=0.01; OR=132.9), flock size (p=0.02; OR=11.2), and restocking method (p=0.03; OR=51.6) were the factors significantly associated with Newcastle disease (p≤0.05). Newcastle disease remains a serious threat in village poultry farming in Ambatolampy district until now and vaccination is the best way to control it.*

I. INTRODUCTION

Newcastle disease (ND) is a viral infection of domestic poultry and other bird species characterized mainly by respiratory, digestive and nervous system disorders [1, 2]. It occurs worldwide and is under control in Canada, the United States of America and some Western European countries [3], but is the main cause of mortality in village poultry (70%) in developing countries, mainly in Asia, Africa and Eastern Europe [4]. Its socio-economic impact and rapid spread in birds indicate that it should be immediately notified to the World Organization for Animal Health (WOAH) upon identification [5].

ND was first detected in Madagascar in August 1946 at the port of Toamasina (in the east) and spread throughout the island [6]. Since then, different ND seroprevalences have been reported in different regions of Madagascar and ND is currently the main constraint in Malagasy village poultry farming [7, 8]. It is responsible for 84% of morbidity [9] and 44.3% of mortality in village chickens [10]. The losses caused by this disease were estimated at more than 4 million euros in 2013 based on a mortality rate of village chickens and palmipeds of 7.14% [9].

ND reached Ambatolampy district (in the center) via the railway axis in December 1946 [11]. Two studies conducted in 2019 showed seroprevalences of 54.2% and 63.07% in the area. They did not report the involvement of livestock management or practices on the disease [12, 13]. However, village poultry farming plays an important role in the area, particularly duck farming for foie gras production, as more than 70% of households in Behenjy (one of the district's communes) have been living from this activity for at least 40 years [14]. The total amount of foie gras supplied by these households was 3010 kg per month to restaurants, food processing companies, households, exports, etc. [13]. Secondly, palmipeds (domestic ducks and geese) are known to be reservoirs and excretors of ND virus and thus could be a source of exposure of other poultry to the virus[9, 15]. Hence the question: is there a statistically significant association of husbandry practices with ND in Ambatolampy District? The hypothesis of this study was that poultry husbandry practices with a high presence of palmipeds were involved in the persistence of ND in the district. Thus, the objective of the study was to determine the husbandry factors that may be associated with ND.

II. Material and Methods

Study site

The study was conducted in the communes of Ambatolampy and Behenjy,which are located in the district of Ambatolampy (Figure 1). The district has a total of eighteen communes. Its surface is 30 km^2.The commune of Behenjy represents the capital of foie gras in Madagascar where more than 70% of households raise ducks for the production of this commodity. It produces up to more than 3,010kg of foie gras per month for various ranges of local or international consumers after hand force-feeding the ducks for 21 days.

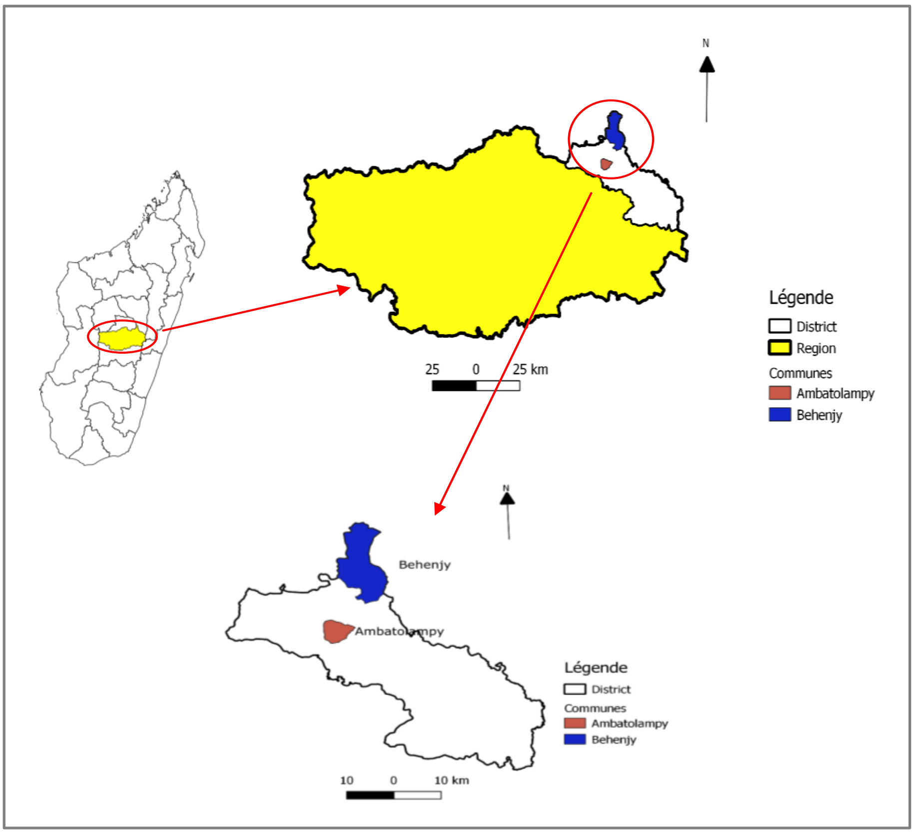

Figure 1: Study sites in the district of Ambatolampy, Madagascar

Samples and household sampling method

The study population consists of poultry and village poultry farming households in the communes of Ambatolampy and Behenjy. A household is the equivalent of a village poultry farm according to the studies of Rasamoelina, Koko and Maminiaina teams [8, 16, 17]. The sampling is divided into two parts:

- The first consists of selecting the herders surveyed by the snowball sampling, as there is no list of poultry farmers available[18];
- The second sampling concerns the animals to be tested that are either unvaccinated or vaccinated less than seven days or more than 12 months.

A total of 41 households were surveyed, 26 in the commune of Ambatolampy and 15 in the commune of Behenjy. Thus, 100 blood samples were collected and analyzed for the two communes.

Study parameters and data analysis

Data entry, sorting, and verification were performed on Microsoft Office Excel 2013. The data were presented with descriptive statistics and a part was analyzed to test the association of the husbandry parameters (factors) with ND (dependent variable). This association test consists of multiple logistic regression analysis under R software version 4.1.1.

Study parameters

The variable to be explained (dependent variable) was represented by the seroprevalence of ND in the households and the explanatory variables (independent variables) by the different husbandry practices. The independent variables were the flock size, the species mix, the method of restocking, the hygiene of the chicken houses, the fence, the disinfection, the sanitary emptying, the husbandry conduct in the case of disease and the husbandry conduct in the case of mortality.

The size of the flock is a qualitative variable divided into 2 modalities: low or high. Based on the number of flocks reported in previous studies by Koko *et al.* and Andriamaroarison *et al.* [6, 7], a household with 20 or more chickens was classified as having a high flock size and otherwise the size was considered low.

Multiple logistic regression analysis

Multiple logistic regression was used to analyze the factors associated with a response variable (e.g., disease). Its aim is to evaluate the association between the response variable (disease) and the explanatory variables studied (factors) by means of the calculation of p or the Odds-Ratios. This model makes it possible to express the relationship between a response variable Y (dichotomous) and one or more factors Xi (nominal or quantitative) as a probability (between 0 and 1) or risk.The statistical analysis and interpretation of the results were carried out according to the method presented by El Sanharawi [19].

Thus, eight independent variables were chosen for the determination of the association factors. They were taken from the literature [20] and were flock size, species mix, restocking method, poultry house hygiene, fencing, husbandry practices in case of disease, husbandry conduct in case of mortality and sanitary emptying.

III. Results

Forty-on (41) households were interviewed, 26 in the commune of Ambatolampy and 15 in the commune of Behenjy. Thus, 100 blood samples were collected and analyzed for the two communes. The majority (47.58%) of blood samples from poultry were from chickens (4-8 months old).The poultry species present in households include mostly chickens (82%), ducks (5%), turkeys (2%), geese (2%) and other species (9%).

General poultry house structure and hygiene

The households generally build poultry houses with minimum financial outlay. Most farmers (51.2%) tend to build brick walls, sheet metal roofs and design dirt floors that are difficult to clean. Seventy-three percent (73%) of the poultry houses are located near the farmers' homes for the safety of the poultry from thieves. More than half of the farmers have a flock size of less than 21.Sixty-three point four percent (63.4%) of the households do not regularly clean their chicken houses. The seroprevalence of ND differs between households that clean their poultry houses (17.1%) and those that do not (41.5%).

Seroprevalences of ND

According to the result of the ELISA serological test for ND virus antibodies, the ND seroprevalence are 58.5%at the household level (Table I) and 64% at the household level (Table II).

Table I: ND serological results at household level

Municipalities	Number of households sampled	Number of positive households	Percentage of positive households (%)
Ambatolampy	26	15	57.6
Behenjy	15	9	60
Total	41	24	58.5

Table II: Serological results of ND at animal level

Municipalities	Number of animals collected(n=100)	Number of positive animals (n=64)	Percentage of positive animals (%)
Ambatolampy	58	36	56
Behenjy	42	28	44
Total	100	64	64

Mixed poultry species in households

More than half of the poultry farming households (51.2%) are mixed. They keep both chickens and other bird species such as ducks, geese, turkeys, etc. in the same household. The frequency of ND was higher in mixed farms (39.0%) than in single farms (19.5%) showing a significant difference with a $p = 0.02$.

Flock size

The average size of the poultry flock is 20 head of chickens per household. Fifty-one percent (51.2%) of households have a flock equal or less than 20 chickens and 48.8% have more than 20. From this result, the modalities on flock size were divided into 2: high for those with 21 or more chickens and low for those with less than 21. The frequency of ND is higher for households with more chickens (36.6%) than for households with fewer chickens (22%).The Chi^2 test showed that the difference in proportions was significant with a $p = 0.04$.

Presence of fence

Fifty-eight point five percent (58.5%) of the farms were not fenced, but allowed the chickens to roam. The proportion of ND seropositive and seronegative households remained the same (29.3%). No significant association was observed between ND and the presence of fencing ($p = 0.19$).

Method of restocking

Purchase and reproduction are the two methods of restocking poultry, with a higher proportion of 58.5% for restocking by reproduction. ND seroprevalence increased from 24.4% to 34.1% among farmer-households when farmers chose to restock by purchase.A significant association of ND with the method ofis observed with a $p = 0.01$.

Conduct in case of disease

Eighty-four point two percent (84.2%) of herder households did not slaughter sick animals, but consumed them for animal protein. The rest (15.8%) sold sick animals in markets or restaurants to earn some money to face small emergencies such as buying rice, medical expenses, or school fees. The results show that ND seroprevalence increased from 12.2% for good management to 46.3% for poor management in the face of a disease outbreak in herder households.The Chi^2 test showed that there was a significant associationbetween ND and disease management ($p = 0.03$).

Sanitary emptying and disinfection

Sanitary emptying is practiced by only 48.8% of farmers and its duration varies from one week to one month. The prevalence of ND increases from 24.4% among households who empty their barns after an illness to 34.1% among households who do not empty their barns during a given time. However, there was no association between ND and the practice of fallowing ($p = 0.69$). The survey results also show that 78% of households do not practice disinfection either.

Conduct in case of mortality due to a disease

In case of poultry mortality due to disease, more than 51% of households consumed dead poultry and 39% did not burn soiled poultry materials.

Table III: Husbandry practices in case of mortality

Variables	Modalities	Number of households surveyed (n=41)	Percentage of households surveyed (%)
Burying of dead birds	Yes	19	46.3
	No	22	53.7
Cusomption of dead poultry	Yes	21	51.2
	No	20	48.8
Treatment of the materials	Burn	16	39
	Do not burn	25	61

Factors associated with ND

Simple logistic regression analysis (univariate analysis) selected 5 variables associated with ND at the 20% error level ($p \leq 0.20$) for multivariate analysis(Table IV).Indeed, the multivariate logistic regression analysis showed that 3 farm variables were significantly associated with ND. These were species mix, flock size and stocking method:

- First, for species mix, the prevalence of ND was significantly higher in mixed farms (39.0%) than in single farms (19.5%) with a $p = 0.02$;
- Second, for flock size, ND was significantly more prevalent in households with more than 20 chickens (36.6%) than in those with less than 20 chickens (22.0%) with a $p = 0.04$;
- And third, with respect to the method of restocking, ND affected more households that restocked their flocks by purchasing new birds (34.1%) than those that restocked by breeding (24.4%) with a $p = 0.01$.

Apart from these three factors, the difference in ND seroprevalence was not significant for poultry house maintenance, presence of fencing, sanitation, disinfection and disease management. The p values were greater than 0.05.

Table IV: Multivariate analysis of variables

Variables	Categories	*n*	Seropositive *ns*	Seropositive %	CR	OR	95%CI	*p*
Flock size	≤ 20	21	9	22.0	-	11.29	1.73-121	0.02
	> 20	20	15	36.6	2.42			
Species mix	Unique	20	8	19.5	-	132.99	6.31-14670	0.01
	Mixed	21	16	39.0	0.01			
Presence of fence	Yes	17	12	29.3	-	0.07	0-0.96	0.08
	No	24	12	29.3	-2.54			

Restocking method	Reproduction	24	10	24.4	-	51.66	2.63-4474.6	0.03
	Buy	17	14	34.1	3.94			
Conduct in case of disease	Good	14	5	12.2	-	0.37	0.01-6.13	0.50
	Bad	27	19	46.3	-0.97			

n:number per category; *ns:* number of seropositive; RC: logistic regression coefficient; 95% CI: 95% Confidence Interval

IV. Discussion

Seroprevalence and factor associated with the ND

The results showed that ND prevalences in Ambatolampy district are very high, 64% and 59% at animal and household level, respectively (Tables I and II). Seroprevalences close to those of the present study were reported in Lac Alaotra in 2008 (70.94%) and in 2012 (73%) [21]; in Ambohimangakely and Moramanga with 72-100% seropositive in 2000 [17]. A higher seroprevalence of 92.34% was also reported in the village poultry farm of Vatomandry in 2017 [7]. In 2015, Rasamoelina's team found a seroprevalence of 88% in ND outbreaks in Lac Alaotra with an average mortality rate of 44% [8]. In Africa and other continents, results showed lower seroprevalences of 22% in Côte d'Ivoire and 27.4% in Ethiopia [22, 23] while seroprevalences of 12.6% and 33.8% were reported in Australia in 2006 and Brazil in 2008 respectively [24, 25]. In the United States of America, a seroprevalence close to that of the present study (79%) has been recorded [24, 26].

According to multivariate logistic regression analysis, this high seroprevalence of Newcastle disease in village poultry farming at the site was associated with non-compliance with household husbandry practices. The factors significantly involved were species mix (p=0.01; OR=132.9), stocking method (p=0.02; OR=51.6) and flock size (p=0.03; OR=11.2).

Mixed species in the household was the first variable implicated because it increased exposure to ND by 132.9 times for farmer-households raising both chickens with palmipeds, turkeys and other poultry species. This practice is very common in the Ambatolampy area because the district represents the capital of foie gras production. In addition to mixing poultry, the majority of farmers in the area neglect the rules of animal husbandry most of the time and take care of the chickens without cleaning their hands with soap after force-feeding the ducks. And even if there is a separation of species, it consists only of a simple wooden fence that does not prevent contact between the birds and thus the transmission of the disease in the chickens. This mixing of different poultry species has also been reported in poultry-farming households in other regions of Madagascar, including Vatomandry District, where results showed that a village poultry farmer had an average of 20 chickens, three ducks, two muscovy ducks, one goose and one turkey in his household [7]. However, in 2003, Alexander reported that waterfowl or palmipeds are receptive to ND virus and shed the virus without usually developing any apparent clinical signs even with velogenic strains. In 2002, Capua et al. reported that households raising mixed species of poultry were at higher risk of ND than other households[27]. Faced with such exposure, Rasamoelina et al. suggested that control of ND in palmipeds could cut the cycle of viral transmission to chickens [9].

After species mix, flock size is the second factor associated with ND. A farmer household with more than 20 chickens contracted ND 11.29 times more than one with fewer chickens. This association of high flock size with ND is consistent with studies by Ban-Bo et al. in Chad, Alders and Spradbrow and Maminiaina et al. in Madagascar [10, 28, 29]. Indeed, in these countries, a large number of flocks or a high density of the avian population leads to very close contact between healthy and sick individuals and/or poor ventilation in the farm, which causes the concentration of infectious viral particles to rise. In addition to this, the risk of infection in Malagasy village poultry is also caused by the proliferation of roaming and intermingling animals in the backyard. Since most households do not vaccinate their poultry, the frequency of ND increases significantly from 22.0% in households with low livestock populations to 36.6% in households with high livestock populations.

Finally, the third factor is presented by the method of restocking. A purchase of new chickens significantly increases ND exposure by 51.66 times and the prevalence increases from 24.4% to 34.1%. This is because either the newly purchased chickens are immediately mixed with the flock without prior quarantine, or they are tethered but not isolated from the rest of the flock. As a result, the flock will always be in contact with animals of uncertain health status. Similar cases have been reported by Riise et al. and have shown that poultry farmers often mix new birds with the rest of the flock without prior quarantine [30].. This practice of restocking is implicated in the transmission of ND virus according to Getabalew et al. [1].

This lack of respect for husbandry practices is characteristic of village poultry farming and has become a common practice in many regions of Madagascar over the years, which is difficult to remedy due to the lack of will and knowledge on the part of poultry farming households. In most African countries, village poultry farming is a secondary activity to agriculture and provides a safety net in emergencies in vulnerable households.

Limitations of the study

This study allowed for a serological study of ND exposure in village poultry in Ambatolampy district. For this purpose, 100 blood samples of chickens from the commune of Ambatolampy and Behenjy were collected and showed an overall seroprevalence of 64%. Rigorous methods were used in this study with respect to the sampling method (snowball sampling) and the serological test (ELISA test). However, the significance of this result is limited both by the number of communes involved in the district and the number of farmer households considered (41 farmer households in total).

Vaccination against ND

Vaccination is the best way to control ND. Vaccination is regular in industrial poultry farming, but the rate is low in Malagasy village poultry farming, especially in remote areas. At the national level, it has been estimated at 5.62% in 2021[31]. This low vaccination rate (46%) is due to the reluctance of poultry farmers in the Ambatolampy district. A study in Lake Alaotra showed that refusal to vaccinate was a function of the age, sex and education level of the farmers and their perception of the effectiveness of the vaccine[32]. Furthermore, for village poultry farmers, this lack of interest is reflected in the fact that poultry can still be valued whether it is sick or dead by eating or selling it [9].

Given also that mixing of poultry species is very common in Malagasy village poultry farming and that wild birds can sometimes carry the virus wherever there are poultry farms[33], it is very important to cut the transmission cycle of ND virus from palmipeds to chickens. Thus, it would be necessary to conduct vaccine research to reduce viral shedding as much as possible by designing an inactivated vaccine from the circulating strain and vaccinating the palmipeds. To this end, Rasamoelina et al. have already initiated an animal experiment on vaccination of domestic palmipeds (ducks) in Madagascar to reduce viral shedding, but it was not successful due to the insufficient biosecurity level of the farming infrastructure [9]. In contrast, in China, the same experiments showed that ducks were protected from clinical signs of ND and no virus was isolated from the vaccinated duck groups after one month of inoculation with pathogenic strains of ND. These results demonstrated that vaccination is very important in reducing viral shedding and even the spread of the disease [34, 35].

V. Conclusion

ND remains a serious threat to Malagasy village poultry farming, and vaccination of poultry is the best way to control it, since poultry farmers are not accustomed to following good husbandry practices. Thus, in perspective, it would be necessary to continue vaccine research on the design of new vaccines preventing viral excretion and research on the immunization of palmipeds in order to cut the transmission cycle of the virus.

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