

## Bacteriological prevalence of salmonellosis due to *Salmonella gallinarum* (typhosis) in exotic and local hens in peri-urban areas of Bamako, Ségou and Sikasso in Mali

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### Abstract

In Mali, poultry farming is the main source of table eggs and broiler chickens. However, among the constraints to its development are those of a health nature in general and salmonellosis in particular. The objective of this study conducted in 2014-2015 was to assess the bacteriological prevalence of salmonellosis caused by *Salmonella gallinarum* in exotic and local hens in the peri-urban areas of Bamako, Ségou and Sikasso. 526 samples (515 cloacal swabs and 11 organ samples) from 182 chicken farms (exotic and local) collected and tested in culture. An overall bacteriological prevalence rate of 10.2% obtained with 9.7% in Bamako against 15.1% in Ségou and 8.0% in Sikasso. These rates varied according to the nature of the samples (9.5%, 14.2% and 6.2% for cloacal swabs against 25%, 100% and 50% for organ samples respectively in Bamako, Ségou and Sikasso) and the type of poultry (9.2% in exotic hens and 27.5% in local chickens). Statistical analysis revealed no significant variation ( $p=0.128>0.05$  in Ségou,  $p=0.335>0.05$  in Sikasso) in prevalence at the Ségou and Sikasso sites. On the other hand, it revealed a significant variation according to the types of samples and poultry ( $p=0.003<0.05$ ). This study confirmed the persistence of avian salmonellosis in poultry farms located in peri-urban areas of Bamako, Ségou and Sikasso. 5% in local hens). Statistical analysis revealed no significant variation ( $p=0.128>0.05$  in Ségou,  $p=0.335>0.05$  in Sikasso) in prevalence at the Ségou and Sikasso sites. On the other hand, it revealed a significant variation according to the types of samples and poultry ( $p=0.003<0.05$ ). This study confirmed the persistence of avian salmonellosis in poultry farms located in peri-urban areas of Bamako, Ségou and Sikasso. 5% in local hens). Statistical analysis revealed no significant variation ( $p=0.128>0.05$  in Ségou,  $p=0.335>0.05$  in Sikasso) in prevalence at the Ségou and Sikasso sites. On the other hand, it revealed a significant variation according to the types of samples and poultry ( $p=0.003<0.05$ ). This study confirmed the persistence of avian salmonellosis in poultry farms located in peri-urban areas of Bamako, Ségou and Sikasso.

**Keywords:** Poultry farming; Bamako; Prevalence; Salmonellosis; Ségou; Sikasso

### 1. Introduction

Poultry farming plays an important role in socio-economic and cultural development in Mali. In 2020, the number of poultry at the national level was estimated at around 55952026 subjects, including 49617572 subjects (88.68%) for the traditional sector and 6 334454 subjects (11.32%) for the modern sector (1). This number has increased significantly compared to that of 2017, which was approximately 45004600 subjects (2). Several sources have reported that poultry farming is one of the main activities generating income and sources of protein for the population in Mali [2, 3, 4, 5]. Indeed, the various commercial transactions related to poultry farming estimated at approximately 5,019,232,000 CFA francs per year. In addition, in 2019, the production of poultry meat estimated at 12,250,191 tons, or 14, 55% of total meat

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production<sup>[1]</sup>. Hence its obvious contribution to reducing poverty and achieving food and nutritional security in Mali. Chicks mainly imported from other countries, even if the embryos of industrial units producing local day-old chicks are beginning to emerge. Prophylaxis plans also made available to poultry farmers by chick suppliers. Despite the application of these prophylaxis plans, high mortality rates are often recorded (20% or more in layers compared to an acceptable 5% during the rearing period and 2 to 3% per month during the production period)<sup>[6]</sup>. Among the constraints to the development of modern poultry farming are those of a health nature in general and the losses following the numerous cases of salmonellosis in particular<sup>[6]</sup>. According to the results of a diagnostic study carried out in Mali during the period 2003 – 2007 and relating to the identification of the main constraints in semi-industrial poultry farming, salmonellosis is one of the dominant pathologies (Typhosis: 20.09%, case infection of eggs by *Salmonella pullorum/gallinarum*: 19.53%)<sup>[6]</sup>. The results of the study in question are fragmentary, as it did not make it possible to establish the prevalence of the disease according to the sites and the types of chickens (exotic and local). According to the diagnostic report on breeding in Mali by the technical team, traditional poultry farming was the results of a diagnostic study carried. Out in Mali during the period 2003 – 2007 and relating to the identification of the main constraints in semi-industrial poultry farming, salmonellosis is one of the dominant pathologies (Typhosis: 20.09%, case infection of eggs by *Salmonella pullorum/gallinarum*: 19.53%)<sup>[6]</sup>. The results of the study in question are fragmentary, as it did not make it possible to establish the prevalence of the disease according to the sites and the types of chickens (exotic and local).<sup>[6]</sup>. The results of the study in question are fragmentary, as it did not make it possible to establish the prevalence of the disease according to the sites and the types of chickens (exotic and local). According to the diagnostic report on breeding in Mali by the Alive Technical Team, traditional poultry farming being p salmonellosis is one of the dominant pathologies (Typhosis: 20.09%, cases of infection of eggs by *Salmonella pullorum/gallinarum*: 19.53%)<sup>[6]</sup>. The results of the study in question are fragmentary, as it did not make it possible to establish the prevalence of the disease according to the sites and the types of chickens (exotic and local). According to the diagnostic report on breeding in Mali by the Alive Technical Team, traditional poultry farming being p salmonellosis is one of the dominant pathologies (Typhosis: 20.09%, cases of infection of eggs by *Salmonella pullorum/gallinarum*: 19.53%)<sup>[6]</sup>. The results of the study in question are fragmentary, as it did not make it possible to establish the prevalence of the disease according to the sites and the types of chickens (exotic and local). According to the diagnostic report on breeding in Mali by the Alive Technical Team, traditional poultry farming being pratified without any form of improvement, experiences very high losses (20% and more) due to diseases and predators (Team Alive Mali)<sup>[7]</sup>. The control strategy for avian salmonellosis caused by *Salmonella gallinarum* must be based on the implementation of control measures aimed at reducing the risk of contamination by salmonella. Many strategies can be used in on-farm control programs, including antibiotic therapy<sup>[8]</sup> recommending the application of vaccination, biosecurity measures and all efforts necessary to achieve *Salmonella*-free food production. Vaccine protection against any *Salmonella* infection will always be insufficient if biosecurity is also insufficient<sup>[9]</sup>. This study carried out during the period 2014-2015 and covered 182 mixed poultry farms infected with *Salmonella* in peri-urban areas of the District of Bamako (96) and the cities of Ségou (43) and Sikasso (43). It made it possible to determine the bacteriological prevalence rates of salmonellosis in exotic laying hens and subjects of local breeds in the different study sites.

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## 2. Material and methods

The study was carried out during the period from August 2014 to December 31, 2015 in two essential phases: a phase of clinical-epidemiological monitoring and collection of appropriate samples and a phase of implementation of laboratory tests (isolation of strains in culture). Sampling by purposive choice was used to select the study areas, the farms and the subjects to be sampled (sick or recently dead laying hens having presented suspicious clinical signs of salmonellosis).

### 2.1. Type and area of study

This prospective study was carried out in the peri-urban areas of the District of Bamako and the cities of Ségou and Sikasso. The development and importance of modern poultry farming in these sites justified the choice of these sites. In the peri-urban area of Bamako, poultry farms have been chosen on 06 main axes including Bamako-Koulikoro, Bamako-Siby, Bamako-Kangaba, Bamako-Ségou, Bamako-Kati and Bamako-Bougouni. In Ségou, the farms chose on 03 axes including Ségou-Markala, Ségou-Cinzana and Ségou-Bamako. In Sikasso, they chose on 04 axes including Sikasso-Koutiala, Sikasso-Nièna, Sikasso-Bobo Dioulasso and Sikasso-Missiri Koro.

### 2.2. Choice of farms

The study covered 182 modern poultry farms located in peri-urban areas of Bamako, Sikasso and Ségou. Thus, the choice of farms made on the basis of objective criteria, such as the accessibility of the farm, the promoter's adherence to the spirit of the study, the frequency of suspected clinical cases of salmonellosis in poultry. 96 farms visited in Bamako, 43 in Ségou and 43 in Sikasso (Tables 3, 4 and 5).

### 2.3. Choice of poultry and collection of samples

As part of this study, fortnightly visits made during which cloacal swabs taken from sick laying hens showing signs of depression, anorexia, whitish diarrhea, dehydration and anemia and Cyanosis apparent on comb and wattles. Autopsies performed on sick or recently dead subjects. In the event of the presence of lesions specific to salmonellosis (discoloration of the liver, ovaritis or salpingitis, splenomegaly, enteritis, myopericarditis, pneumonia), samples of livers, ovarian clusters, spleen, intestines, heart and lungs. The various samples identified, kept under ice and sent to the Central Veterinary Laboratory for laboratory examinations.

### 2.4. Laboratory diagnostic methods

The samples collected were prepared before cultured for the selective search for Salmonella. To do this, the organs first crushed, while the swabs directly inoculated. Thus, all the samples subjected to prior pre-enrichment on peptone water followed by enrichment on tetrathionate broth or on Rappaport-Vassiliadis medium. *Salmonella* isolation performed on *Salmonella-Shigella* (SS) and Mac Conkey (MC) agars from the enrichment broth cultures. Salmonella suspect colonies subcultured onto soybean tryptose agars and incubated for 24 hours at 37°C to obtain pure Salmonella cultures.

### 2.5. Statistical analysis method

Data analyzed with Stata version 6.0 software. Logistic regression used to verify the existence of a significant difference in prevalence rates depending on the sites and types of samples.

## 3. Results

During this study, 526 samples including 515 cloacal swabs and 11 organ samples collected from 182 farms and subjected to laboratory examinations. Culture of the various samples made it possible to establish an overall bacteriological prevalence rate of 10.3% for *Salmonella gallinarum*. The prevalence rate did not vary significantly ( $p>0.05$ ) depending on the study sites (9.7% in Bamako against 15.1% in Ségou and 8.0% Sikasso). On the other hand, it varied significantly ( $p<0.05$ ) depending on the nature of the samples and the type of poultry. The results obtained are illustrated in Tables 3, 4, 5 and 6.

**Table 1** Results by site of bacteriological examinations (Bamako)

Site	Farms visited	Specimens			
		Cloacal swabs		Organs	
		Tested	Positives	Tested	Positives
Dialakorobougou	01	03	01	01	0
Tabacoro	04	0	0	0	0
Baguineda	05	19	03	0	0
Kasela	02	12	04	0	0
Sadiouroubougou	03	05	01	0	0
Senou	01	03	0	0	0
Banankoroni	05	18	02	0	0
Sanankoroba	14	30	0	01	0
Sikulu	03	13	02	0	0
Missabougou	14	26	0	0	0
Morocco	03	0	0	01	0
Tiètiguibougou	02	0	0	0	0
Dogobala	02	11	01	0	0
Sebenikoro	01	03	01	0	0

Konobougou	01	03	01	0	0
Missala-Gouana	13	20	04	0	0
Soundougouba	01	10	01	01	01
Mountougoula	11	40	01	0	0
soro	01	09	02	0	0
Yayabougou	01	09	0	0	0
Dialakoroba	01	10	0	0	0
Samaya	02	12	0	0	0
Balandougou	05	17	02	0	0
Total	96	273	26	04	01

In the peri-urban area of the District of Bamako in the 96 farms visited, 277 samples were collected and cultured, of which 27 proved positive (i.e. 9.7%), including 26 cloacal swabs (26/273 i.e. 9.5%.) and 01 single organ (01/04 or 25.0%).

**Table 2** Results by site of bacteriological examinations (Ségou)

Site	Farms visited	Specimens			
		Cloacal swabs		Organs	
		Tested	Positives	Tested	Positives
Kirangowere	03	04	0	0	0
Sekoro	01	03	0	0	0
Pelengana	05	10	01	0	0
Guerefebougou Road	01	04	0	0	0
Sebougou	07	14	04	0	0
Banankoro	01	05	01	0	0
Hamdallaye	02	01	0	01	01
Medina	07	13	03	0	0
Bagadadji	03	09	0	0	0
Sebougou	01	02	0	0	0
Hamdallaye	02	02	0	0	0
Emgoulem	02	04	01	0	0
Sakoiba (Missira)	04	17	04	0	0
City COMATEX	01	04	00	00	00
bamananking	01	01	00	00	00
millionking	01	03	00	00	00
Sokolakono	01	02	00	00	00
Total	43	98	14	01	01

In Ségou, in the 43 farms visited, 99 samples took, including one organ sample. Of which, 15 found to be positive (i.e. 15.1%), including 14 swabs (14/98 i.e. 14.2%) and 01 organ.

**Table 3** Results by site of bacteriological examinations (Sikasso)

Site	Farms visited	Specimens			
		Cloacal swabs		Organs	
		Tested	Positives	Tested	Positives
Wayèrèma II	01	07	0	0	0
Sanoubougou	04	0	0	03	02
Kafela	04	11	0	0	0
Mamabougou	05	16	01	01	0
Ouahibera	01	02	0	0	0
Doniana	02	02	0	0	0
Zangaradougou	02	03	03	0	0
Manabougou	01	05	01	0	0
nankoundiassa	08	34	0	01	01
Zanso sirakoko	01	07	01	01	0
Zanblala	01	01	0	0	0
Sirakoro	04	23	02	00	00
Wayèrèma I	04	11	00	00	00
Farako Fenkolo	01	06	00	00	00
Medina	03	09	01	00	00
Koro dam	01	07	00	00	00
Total	43	144	09	06	03

Of the 526 samples, 44 found to be positive. This represents a bacteriological prevalence rate of 8.3%. No significant variation ( $p>0.05$ ) in prevalence was demonstrated between the sites. On the other hand, there is a significant variation in prevalence according to the nature of the samples and the breeds of poultry ( $p=0.003<0.05$ ).

**Table 4** Results by type of poultry of bacteriological examinations

Site	Samples collected	Types of poultry					
		Exotics			local		
		Culture	Positives	%	Culture	Positives	%
Bamako	277	272	26	9.5	05	01	20.0
Segou	99	88	13	14.7	11	02	18.1
Sikasso	150	137	07	5.1	13	05	38.4
Total	526	497	46	9.2	29	08	27.5

The statistical analysis revealed a significant difference between the prevalence rates according to the types of samples ( $\text{Chi}^2(3) = 13.62$ ;  $p=0.003<0.05$ ).

**Table 5** Results of the statistical analyzes according to the site

Comments	526					
Chi2(3)	13.62					
prob	0.0035					
Nickname R2	0.0391					
					95% Confidence Interval	
Headings	Coefficients	standard error	Z	p	Terminal <	Terminal >
Segou	0.53	0.35	1.52	0.128	-0.15	1.22
Sikasso	-0.35	0.38	-0.93	0.335	-1.09	0.39
Organs	2.25	0.64	3.51	0.000	0.99	3.51
Constant	-2.29	0.21	-11.08	0.000	-2.7	-1.89

Site (s)	Cloacal swabs			95% Confidence Interval		Standard Deviation
	Tested	Positives	Prevalence	Terminal<	Terminal>	
Bamako	273	26	9.52	6.04	13	3.48
Sikasso	144	9	6.25	2.3	10.2	3.95
Segou	98	14	14.29	7.36	21.22	6.93
Total	515	49	9.51	6.98	12.04	2.53

Swabs: 9.52±3.48 % (Bamako), 6.25±3.95% (Sikasso), 14.29±6.93% (Ségou) withPrev. Overall: 9.51±2.53%

**Table 6** Results of statistical analyzes on the link between the bacteriological prevalence of salmonellosis and the type of samples (organs)

Site (s)	Organs			95% Confidence Interval		Standard Deviation
	Tested	Positives	Prevalence	Terminal<	Terminal>	
Bamako	4	1	25	-17.44	67.44	42.44
Sikasso	6	3	50	10	90	40
Segou	1	1	100	-	-	-
Total	11	5	45.45	16.03	74.87	29.42

Organs: 25±42.44% (Bamako), 50±40% (Sikasso), 100% (Ségou) withPrev Global: 45.45±29.42%

#### 4. Discussion

Avian salmonellosis caused by *Salmonella pullorum/Gallinarum* distributed worldwide, especially in developing countries (10). Mali is no exception to this reality as several studies have shown.

This study has established an overall bacteriological prevalence rate of 10.2% for *Salmonella gallinarum*. There is no significant difference ( $p>0.05$ ) between the prevalences at the site level (9.7% in Bamako, 15.1% in Ségou and 8.0% in Sikasso). On the other hand, there is a significant difference depending on the nature of the samples (9.5%, 14.2% and 6.2% for cloacal swabs against 25%, 100% and 50% for organ samples respectively) in Bamako, Ségou and Sikasso), as well as depending on the type of chicken.

Similarly, the study showed that the prevalence of salmonellosis is much higher in subjects of local breeds (27.5%) compared to exotic poultry (9.2%). Thus, it obtained a prevalence of 9.5% for subjects of exotic breed against 20% for those of local breed in Bamako, 14.7% for subjects of exotic breed against 18.1% for those of local breed in Ségou, 5.1% for exotic poultry against 38.4% for local poultry. This militates in favor of the great extent of the phenomenon in local hens, which do not generally benefit from appropriate care compared to exotic hens. These results show that *Salmonella gallinarum* infections are a reality in modern poultry farms in Mali. Indeed, it shown, that avian salmonellosis is one of the major constraints both in Mali [6, 11] and in other countries of the sub-region such as Senegal [12, 13, 14, and 15] and Côte d'Ivoire [16]. Various authors report that the prevalence of *Salmonella* spp. vary considerably depending on the country and the type of direct debit. Thus, in Mali, Satigui and al. [17] obtained bacteriological prevalence of 9.9% following analysis of cloacal swabs and organs. Long before the previous study, Sidibe and al. [6] obtained from the organs, a bacteriological prevalence 20.48% of which 18.91% in Bamako, 50% in Sikasso and 20% in Ségou. They also obtained following the analysis of eggs from Bamako, an infection rate of 7.87%. Sidibe and al. [6] were able to isolate *Salmonella pullorum/gallinarum* from cloacal swabs. A study on the evaluation of the risks linked to the presence of salmonella in eggs from improved breeding in the peri-urban area of the District of Bamako 2008-2009 gave an 11.79% of *Salmonella pullorum/gallinarum* infection rates. Arbelot and al. [13] obtained 38% in layers in the Niayes area in Senegal. In Somalia, Prosperi et al. [18] obtained 54.8% serological prevalence for *Salmonella gallinarum* infection and 74.2% bacteriological prevalence in intensive poultry farming. The same authors report that in the rainy season, 41 to 45% of batches of laying hens infected with *Salmonella gallinarum/pullorum*. The small size of organ samples compared to swabs may also explain the difference in prevalence encountered depending on the type of sample. The high prevalence encountered with organs explained by the very small size of the samples taken. The prevalence is not precise compared to that where the prevalence obtained on a large sample size. In addition, due to the small size of the samples collected at the Ségou and Sikasso sites, it is difficult to draw a clear conclusion from the results relating to bacteriological prevalence rates in these sites. The persistence of salmonellosis in exotic chickens from the study sites could explained on the one hand by the often-frequent cohabitation of laying hens with local poultry. It recognized as potential reservoirs of salmonellosis, and on the other hand, by the very poor hygiene and biosecurity conditions in poultry houses due to the small size of the samples collected at the Ségou and Sikasso sites, it is difficult to draw a clear conclusion from the results relating to the bacteriological prevalence rates in these sites. The persistence of salmonellosis in exotic chickens from the study sites could be explained on the one hand by the often frequent cohabitation of laying hens with local poultry recognized as potential reservoirs of salmonellosis, and on the other hand by the very poor hygiene and biosecurity conditions in poultry houses.

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## 5. Conclusion

This study made it possible to highlight the persistence of salmonellosis in modern poultry farms in the peri-urban areas of the District of Bamako and the cities of Ségou and Sikasso. The overall bacteriological prevalence rate varied significantly depending on the types of samples (swabs and organs) and the types of poultry (exotic and local). On the other hand, the prevalences did not vary significantly according to the study sites.

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## Compliance with ethical standards

### *Disclosure of conflict of interest*

No conflict of interest disclosed.

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