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Health and nutritional quality assessment of *salmonella*-contaminated poultry products in sub-Saharan Africa; A case of cote d'ivoire

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The poultry sector is developing rapidly in sub-Saharan Africa but it remains informal in many countries, including Côte d'Ivoire. This situation, which is favourable to the health and nutritional insecurity of food of avian origin, deserves special attention, because of *Salmonella*, which has poultry as its outbreak. The aim of this study was to help reduce the risk of food poisoning linked to the consumption of poultry products contaminated by *Salmonella*. A characterisation of avian production from the farm to the fork has been carried out. It covered 1860 samples of avian origin from 20 farms and 630 samples of diarrhoeal stool from human patients. The strains isolated were characterized by antibiotyping, serotyping and molecular typing. The frequency of isolation of *Salmonella* was 6.8% in poultry products, 5.8% in gizzards, 1.9% in carcasses, 8.2% in eggs and 4.2% in drinking water. In diarrhoeal patients, this frequency was 11.9%, with 15% in children and 10% in adults. Frequently isolated *Salmonella* strains were distributed among *S. Typhimurium* (27.6%), *S. Enteritidis* (20%) and *S. Hadar* (10.6%). A resistance of these microorganisms to β-lactamines was between 70% and 83%. Clonal links have been identified between the serotypes of *S. Typhimurium* and *S. Heidelberg*, isolated in both avian and human matrices. The study shows that unsanitary poultry products could be responsible for diarrhoeal *Salmonella* infections in humans; therefore, preventive provisions are needed for consumer health protection.

Key words: Food insecurity, *Salmonella*, serotype, antibiotic resistance.

INTRODUCTION

Diseases caused by microbial contamination of food are a major and growing public health problem. The situation is all the more worrying because despite awareness campaigns on food hygiene and good food preparation

practices advocated by sub-Saharan health authorities, the number of *Salmonella* isolated in laboratories remains significant (Dibi et al., 2017). Poor hygine practice in the industrial preparation of food and its storage outside the required standards facilitate the proliferation of microorganisms (WHO, 2005a). Most countries with a foodborne disease reporting system have demonstrated that for several decades there has been an increase in the incidence of diseases caused by microorganisms present in food, including the genus *Salmonella* (Jay et al., 2000). Worldwide, 90% of salmonellosis cases are foodborne (CNRSS, 2007); Africa leads with over 80% of cases (Newell et al., 2010). In Tunisia, this disease is still endemic, especially in rural areas, with an incidence of 5 per 100,000 inhabitants (WHO, 2005b).

Salmonellosis and particularly non-tyhoidal salmonellosis are responsible for sporadic or epidemic infections, most often due to food contamination or asymptomatic carrying (Rostagno and Callaway, 2012). Meat from poultry and pigs, eggs, dairy products, and green vegetables contaminated by manure or water are the most frequently cited vehicles or risk factors in the transmission of this bacterial agent (Filbert et al., 2012). The occurrence of salmonellosis is more frequent in persons at risk, in particular immuno compromised persons, including those infected by HIV/AIDS (Gordon, 2008).

In Côte d'Ivoire, studies show that *Salmonella* plays an important role in bacterial diarrhoea (Coulibaly et al., 2015). *Salmonella* promotes the occurrence of certain diseases such as meningitis (Sangaré et al., 2007) and various forms of rheumatism (Dakoury-Dogbo et al., 2001).

This study aimed to serotype *Salmonella* strains and determine their level of resistance to antibiotics generally used in Côte d'Ivoire.

MATERIALS AND METHODS

The study included 2490 sample strains, including 1860 from poultry products and 630 from human diarrheal stools. The stools were received in a reference laboratory for the processing of human samples, with the consent of the participants. An average of 157.5 human samples were expected per year.

The non-biological material mainly included analytical equipment commonly used in bacteriology and molecular biology laboratories (culture media and reagents), as well as survey sheets used to assess the health characteristics and practices of poultry farms, poultry markets, slaughterhouses and their environment. The survey sheets also collected data on the type of meals consumed by some patients before the onset of infectious diarrhea (Table 1).

The isolation and identification of *Salmonella* strains from these samples was carried out in accordance with NF ISO 6579 (2002). Serotyping was carried out according to the scheme defined by Kaufmann and White, and then reviewed by Le Minor (Le Minor and Popoff, 1987 and 1997). The antibiotic susceptibility test was performed with standard antibiotics on all *Salmonella* spp strains (Kirby-Baeur method). The resistance study of the strains was determined by measuring the diameters of the inhibition zones, in accordance with the recommendations of the Antibiogram Committee of the French Society of Microbiology (CA-SFM, 2018).

RESULTS

Farm environment characteristics

The characteristics of the farm environment are divided into two groups of farms: those smaller than 3000 head (size 1) and those larger than 3000 head (size 2). Farms are generally characterized by the presence of rodents (rats, mice, etc.), insects and reptiles, also called pests (60%). These farms are generally located not far from the houses (55%). Among the farms studied, some were near traffic lanes (20%), landfill (15%) or slaughterhouses (10%). The farms in Adjamé commune are closer to the houses (20) than those in Cocody and Yopougon communes (Table 2).

Characteristics of poultry markets

The study shows that all markets are open every day of the week. Where poultry sold is usually temporary (80%). These markets are characterized by facilities that do not meet health safety standards. The markets are also marked by the presence of rats, mice, insects and reptiles called pests (60%), and raving poultry (60%). These markets are often used as poultry breeding sites (60%). Two of the poultry markets visited are located near garbage dumps (Table 3).

Characteristics of the last meals

The last meal taken by the people having made the diarrhoea gave the results contained in Table 4 which shows the diversity of the local menu composed mainly of attiéké, rice of alloko, foutou (banana, taro, yam, manioc), placali and kabato. This table shows that rice and fish are highly valued by the study population. Eggs come in second place, in terms of animal proteins.

Identified serotypes

Five different serotypes were identified in this study. They

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Table 1. Distribution of avian origin samples.

Years	2006	2007	2008	2009
Eggs	125	125	125	125
Carcasses	120	120	120	120
Gizzards	120	120	120	120
Water	100	100	100	100

Table 2. Farm environmental characteristics.

Indicator	Farms concerned per municipality N (%)					Total N (%)
	Abobo	Cocody	Port-Bouët	Yopougon	Adjamé	
Near dwelling	3 (15)	1 (5)	2 (10)	1 (5)	4 (20)	11 (55)
Watercourse	0 (0)	2 (10)	2 (10)	1 (5)	0 (0)	5 (25)
Pest access	3 (15)	2 (10)	2 (10)	2 (10)	3 (15)	12 (60)
Animals in divagation	2 (10)	2 (10)	1 (5)	2 (10)	1 (5)	8 (40)
Fully covered farmhouse	3 (15)	3 (15)	2 (10)	3 (15)	3 (15)	14 (70)
Proximity to high-traffic lanes	1 (5)	0 (0)	0 (0)	1 (5)	2 (10)	4 (20)
Near a landfill	0 (0)	2 (10)	0 (0)	0 (0)	1 (5)	3 (15)
Near slaughterhouse	1 (5)	0 (0)	1 (5)	0 (0)	0 (0)	2 (10)

are: *S. Typhimurium*, *S. Enteritidis, S. Heidelberg, S. Dublin* and *S. Anatum*.

Table 4 shows the distribution of *Salmonella* strains from 2006 to 2009. Overall, it indicates a stationary evolution during these four years. However, in 2007, a significant number (25) were isolated. On average, 19 strains were isolated each year.

The distribution of these serotypes according to the 75 *Salmonella* strains isolated is shown in Table 5.

Level of resistance of *Salmonella* strains to common antibiotics

Table 6 expresses the frequency of resistance of the *Salmonella* strains studied; thus, despite the existence of a high number of strains resistant to families of antibiotics, there are still antibiotics whose strains showed no resistance.

Table 7 shows the frequency of resistance of identified strains to common antibiotics. Thus, one notes a strong resistance of the strains (72%) to ampicillins and amoxicillin + association. However, all *Salmonella* strains studied were susceptible to the following antibiotics: cefotaxime, imipenem, aztreonam, gentamicin, colistine. The resistance rates of *Salmonella* strains isolated according to the antibiotics tested are shown in Table 7.

DISCUSSION

The farm investigation showed that undesirable animals such as mice, margouillats and rodents were accessible at these facilities. However, it is known that these animals may carry pathogens such as *Salmonella* (Corry et al., 2002). Cleaning farms is an effective way to reduce and even eliminate certain pathogenic microorganisms. However, in 30% of the farms, the soil consists of beaten earth, the cleaning of which does not guarantee the reduction of pathogens. Clay soils can provide favourable temperature and humidity conditions for the development of enterobacteriaceae, including *salmonella*. According to Cogan et al. (1999), concrete floors are easy to clean and disinfect and do not promote the survival and

Table 3. Characteristics of poultry markets.

Indicators	Number of markets (N)	(%)
Market opening 7/7 days	5	100
Pests	3	60
Temporary site	4	80
Divagating animals	3	60
Animal husbandry	3	60
Near dwelling	2	40
Bare floor	2	40
No changing rooms	1	20
Traffic nearby	2	40
Near a garbage dump	2	40
**Market cleaning	0	0

The study focused on N markets, N= 5. The cleaning of cages and sales areas is done individually and once a week

Table 4. Survey of last meal taken per person before diarrhoea.

Food		Men		Women	
		Children (Nb)	Adults (Nb)	Children (Nb)	Adults (Nb)
Starchy	Attiéké	35	115	57	199
	Alloko	20	10	43	72
	Placali	16	12	19	24
	Foutou	05	13	11	16
	Kabato	15	32	08	11
	Rice	45	196	30	241
	Other	06	03	05	08
Animal protein	Fish	85	203	44	192
	Meat	15	112	73	215
	Egg	38	57	49	153
	Other	04	09	07	11

NB / Child: between 4 and 17 years, adult: 18 years and over, other: no precise information on the food consumed before diarrhoea.

Table 5. Distribution of *Salmonella* isolates from 2006 to 2009.

Serotype	2006 (%)	2007 (%)	2008 (%)	2009 (%)	Total (%)
S. typhimurium	06 (08.0)	11(14.7)	07 (09.3)	09 (12.0)	33 (44.0)
S. enteritidis	04 (05.3)	08 (10.7)	08 10.7)	03 (04.0)	23 (30.7)
S. heidelberg	00 (00.0)	04 (05.3)	06 (08.0)	02 (02.7)	12 (16.0)
S. dublin	03 (04.0)	00 (00.0)	00 (00.0)	01 01.3)	04 (05.3)
S. anatun	00 (00.0)	02 (02.7)	01 (01.3)	00 (00.0)	03 (04.0)
Total (%)	13 (17.3)	25 (33.3)	22 (29.3)	15(20.0)	75 (100.0)

Table 6. Frequency of antibiotic resistance in *Salmonella* spp.

Families	Antibiotic (concentration)	Resistance (%)
ß-lactamine	Ampicilin (10 ug)	56 (74.7)
	Amoxicillin + Acide clavulanic (20 +10 ug)	54 (72.1)
	Imipenem (10 ug)	00 (0.0)
	Cefalotin (30 ug)	52 (69.4)
	Cefotaxim (30 ug)	00 (0.0)
	Aztreonam (30 ug)	00 (0.0)
Aminosides	Kanamycin (30 ug)	01 (01.6)
	Gentamicin (15 ug)	00 (0.0)
Phenicols	Chloramphenicol (30 ug)	38 (50.8)
Tétracyclines	Tetracycline (30 ug)	30 (40.1)
Polypeptides	Colistine (50 ug)	00 (0.0)
Nitrofuranes	Furans (300 ug)	33 (44.1)
Quinolones	nalidixic Acid (30 ug)	04 (05.6)
	Pefloxacin (5 ug)	03 (04.2)
	Ciprofloxacin (5 ug)	01 (01.6)
Sulfamides + combinations	Triméthoprim + Sulfamethoxazole (SXT) (1.25ug + 23.75 ug)	45 (60.1)

multiplication of bacteria. According to Huys et al. (2013), *Salmonella* strains are highly pathogenic bacteria and the presence of a single strain of *Salmonella* in a food product results in the food being declared unfit for

Table 7. Antibiotic resistance of *Salmonella* serotypes.

Antibiotics	Resistant strains (%)	*S. Typhimurium (%)*	*S. Enteritidis (%)*	*S. Heidelberg (%)*	*S. Dublin (%)*	*S. Anatun (%)*
Ampiciline	54 (72.0)	27 (82.0)	18 (78.3)	05 (42.0)	03 (75.0)	01(33.3)
Amoxicillin+ Clavulanique Ac.	54 (72.0)	27 (82.0)	18 (78.3)	05 (42.0)	03 (75.0)	01(33.3)
Cefalotine	52 (69.3)	25(76.0)	20 (87.0)	04 (33.3)	02 (50.0)	01 (33.3)
Kanamycin	01(01.3)	01(03.0)	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)
Chloramphenicol	38 (50.7)	26 (79.0)	08 (34.8)	02 (16.7)	01 (25.0)	01 (33.3)
Tetracycline	30 (39.9)	20 (61.0)	07(30.4)	02 (16.7)	00 (00.0)	01(33.3)
Nitrofurans	33 (43.9)	18 (54.5)	11(47.8)	03 (25.0)	00 (00.0)	00 (00.0)
nalidixic Acid	04 (05.3)	03 (09.1)	01(04.3)	00 (00.0)	01 (25.0)	00 (00.0)
Pefloxacin	03 (04.0)	02 (06.1)	01(04.3)	00 (00.0)	00 (00.0)	00 (00.0)
Ciprofloxacin	01 (01.3)	01 (03.0)	01(04.3)	00 (00.0)	00 (00.0)	00 (00.0)
Triméthroprime + Sulfamides (SXT)	45 (59.8)	30 (90.9)	13(56.5)	00 (00.0)	00 (00.0)	02 (66.7)

consumption. Sanitary measures must therefore be taken to protect all products intended for human consumption). The relatively high rate of positive results of this work can be explained by several factors: non-compliance with standards, the absence of a general policy for monitoring farms, even private farms, the absence of *salmonella* surveillance and control programmes in hatcheries and private farms. The multiplication of carcass handling, the humidity and heat conditions offered in slaughterhouses, but also the slaughter and hygiene conditions throughout the slaughter chain allow the multiplication of *Salmonella* strains (Garnier, 2008).

Several other factors may be involved in *Salmonella* transmission; the persistence of infection in farm buildings and hatcheries certainly plays an important role (Gradel and Rattenborg, 2003). Rats and mice can carry infections and contaminate buildings and food (Van Immerseel et al*.,* 2005).

Diarrhea has been observed in human patients after consumption of food of questionable hygienic quality. The dietary profile of their last meals, before the onset of diarrhoea, indicates overall that eggs are highly prized. However, it is recognized that eggs are an excellent reservoir of *Salmonella*, which is responsible for many diarrheal diseases in humans (De Knegt et al., 2015). Although in this study, it is difficult to say exactly which foods or other components of the meal or containers are responsible for infectious diarrhea. The positive *Salmonella* infection rate reported here is consistent with the ANSES Opinion (2012). This report showed that human cases of salmonellosis in *S. Enteritidis* were most often associated with the consumption of contaminated eggs and poultry meat, while cases of salmonellosis in *S. Typhimurium* are mainly associated with the consumption of contaminated pork, poultry and beef (Hugas et al*.,* 2014). The analysis of *Salmonella* diarrhoeal stools isolated 75 strains of *Salmonella* grouped into five serotypes. The most frequent were *S. Typhimurium* (40.0%) and S. Enteritidis (30.7%). This result, which shows the prevalence of these two human *Salmonella* serotypes, is in line with the data from Grimont et al. (2007). In addition to *S. Typhimurium, S. Enteritidis*, *S. Heidelberg*, *S. Anatum* and *S. Dublin*, there are other serotypes such as S. Essen, S. Derby which are isolated from human biological products in Côte d'Ivoire, as shown by Coulibaly et al*.* (2010). According to a study conducted in Abidjan by the Microbiology Laboratory of the University of Cocody's Pharmaceutical and Biological Sciences UFR in 2001, the age group of people with typhoid fever out of 103 cases is 18 to 40 years old with a sex ratio of 0.9 (Mwamakamba et al*.,* 2012). Tables 6 and 7 show respectively the families of antibiotics whose *Salmonella* strains are resistant and the resistance phenotypes observed in the isolated and identified strains. These results are all the more alarming as their resistance to β-lactamines, in particular ampicillin, amoxicillin + clavulanic acid and ticarcillin are important. Similarly, resistance of *Salmonella* isolates to tetracycline, Trimethroprime + sulfonamides and fluroquinolones was observed. This same observation is made by Ouédraogo et al*.* (2017), who questioned the main cause of this emergence of resistance. According to these authors, it is due to unsustainable consumption of antibiotics.

Conclusion

The characterization of production systems in the study shows that the majority of selected poultry farms are deficient in terms of sanitation, protection of farm areas and farmer hygiene. The study showed sanitary deficiencies at poultry slaughter sites and poultry markets. Strains of *Salmonella* isolated both from poultry products and humans belong to different serotypes. The consumption survey showed several cases of *Salmonella*

diarrhoea following the consumption of certain food products. Although this prospective study cannot place direct responsibility for the food origin of *Salmonella* infectious diarrhoea in humans, it should be noted that diets combining poultry products have been identified as possible risk factors for *Salmonella* diarrhoea. The resistance to common antibiotics of *Salmonella* serotypes isolated is remarkable for some families of β-lactamines, particularly for Penicillins such as Ampicillin, Ticarcillin and Amoxicillin.

The uncontrolled use of antibiotic therapy in the treatment of certain diseases in humans and especially in agricultural practices could represent a real threat to public health.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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