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Health practices and antibiotic use on dairy farms in Côte d'Ivoire

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Abstract

This research focuses on describing antibiotic treatment practices on dairy farms in Côte d'Ivoire and assessing the presence of antibiotic residues in raw milk. For this purpose, a survey was conducted among 90 dairy farmers in the southern (Abidjan), central (Bouaké) and northern (Korhogo) regions of Côte d'Ivoire. Then, antibiotic residues were analyzed in 21 samples, using the Delvotest T kit and antibiotics were identified and quantified using high-performance liquid chromatography-mass spectrometry (HPLC-MS). The results showed that between 43% and 60% of farmers systematically resorted to self-medication in the event of infection of the cow herd for various reasons: experience, availability of products, saving time and economic reasons. On most farms, pathologies were treated with antibiotics obtained from pharmacies, vets or the parallel market. The antibiotics used on the dairy farms visited were macrolides, penicillins, sulphonamides, tetracyclines, aminoglycosides and first- and second-generation cephalosporins. The antibiotics identified were sulfadoxine, sulfadimethoxine, gentamicin, streptomycin, erythromycin and spiramycin. Concentrations of amoxicillin and erythromycin above the MRL (Maximum residual limit) were observed in at least 50% of samples. These findings highlight the need to educate stakeholders on the responsible use of antibiotics to minimise the risk of antimicrobial resistance and ensure food safety.

Keywords: Raw milk; Antibiotics; Antibiotic therapy; Dairy cows; Survey; Health practices

1. Introduction

Bovine milk is one of the most widely consumed foods in the world due to its high nutritional value [1]. It is one of the main dietary sources of calcium, protein, lipids and vitamins, helping to balance consumers' diets. Demand for cow's milk is increasing in cities in developing countries due to population growth and urbanisation [2]. In Côte d'Ivoire, national production covers only 17% of dairy consumption [3]. The dairy sector plays a vital role in rural areas, where it is an important source of income for local people [4]. The Ivorian cattle herd consists of 1,645,175 head of cattle, of which the main breeds are N'dama, Baoulé and lagoon taurins, zebus and their crossbreeds. In many developed countries, milk production is systematically and rigorously controlled because of the potential risks to human health [5]. However, in Côte d'Ivoire, the informal organisation of the industry and the weakness of the regulatory system and quality control structures mean that the hygienic quality of dairy products is not sufficiently high [6]. Local milk is generally not controlled and is most often consumed raw or fermented. Raw milk is mainly produced from cows reared using traditional methods, and certain producer practices relating to animal health, milking hygiene and handling influence its nutritional and microbiological quality [7, 8]. Consumption of poor-quality dairy products can be a health risk to consumers. Raw milk is highly susceptible to contamination by a variety of pathogenic micro-organisms. It has been implicated in numerous outbreaks of infection and food poisoning throughout the world [9]. Milk is a particularly

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favourable environment for the development of micro-organisms, especially salmonella, which can cause serious food poisoning. The pathogens present in milk can come from the cows themselves, from their skin products or from their environment, as well as from the gut flora of the animals.

In addition to known biological contaminants (bacteria, viruses, yeasts, moulds), there are non-biological contaminants, including antibiotics and anti-inflammatories used to treat common pathologies in dairy cows such as mastitis, endometritis and bronchopathies [10]. The excessive and inappropriate use of antimicrobials in agriculture, particularly antibiotics, leads to the contamination of dairy products and the appearance of treatment-resistant bacteria [11]. Failure to comply with the recommended withdrawal periods for these drugs increases the risk of residues in raw milk, exacerbating the public health problems associated with antimicrobial resistance. The presence of antibiotic residues in dairy products can have adverse effects on human health, such as allergies, intoxications and disruption of intestinal flora [12]. Knowledge of the production practices implemented by farmers is essential for developing relevant recommendations for improving product quality. Thus, the aim of this study is to highlight the poor practices of farmers that can influence the quality of milk from dairy farms in Côte d'Ivoire.

2. Material and methods

2.1. Characterization of farms

A survey was carried out between July 2021 and November 2022 among 90 farmers in three districts of Côte d'Ivoire which are Abidjan, Bouaké and Korhogo. The systematic sweep method [4] was used for random sampling of cow's milk production farms in the different regions. In each district, 30 farms were selected on the basis of criteria that mainly included the sedentary nature of livestock farming, accessibility and availability to provide information and the presence of dairy cows on the farm. The questions dealt with the characteristics of the farm (13 questions), milking practices and conditions (14 questions) and preventive and curative treatments to ensure the health of the animals (17 questions).

2.2. Determination of antibiotic residues in cow's milk

2.2.1. Samples Collection

A total of 90 samples of raw cow's milk were collected from farmers on farms visited during the survey in Abidjan, Bouaké and Korhogo. Samples of raw milk were also taken from 15 randomly selected retailers in each of the three towns. The samples were collected by taking 500 mL of raw milk in a sterile bottle, then transported to the Biotechnology and Food Microbiology laboratory at Nangui Abrogoua University for antibiotic residue analysis.

2.2.2. Detection of antibiotic residues

The Delvotest T kit was used in accordance with the manufacturer's instructions (DSM, Netherlands) for the qualitative detection of antibiotics in the milk samples taken. Delvotest consists of an ampoule containing nutrient agar encrusted with Bacillus stearothermophilus spores and a violet bromocresol indicator. Using a pipette, 100 µL of milk sample was transferred to the ampoule and incubated at 64±2°C for 2 to 3 hours. The colour change of the agar from purple to yellow indicated that the antimicrobial compounds were either below detection limits or absent from the sample. Furthermore, a purple colour indicated the presence of antibiotics at or above the antimicrobial detection limit, while a yellow-violet colour indicated that the antibacterial substances in the milk sample were close to the detection limit.

2.2.3. Identification and quantification of antibiotic residues

Delvotest T positive samples were selected for identification and quantification of antibiotic residues by high performance liquid chromatography-mass spectrometry (HPLC-MS) following the protocol described by [13]. These molecules were quantified using physico-chemical analytical methods based on liquid chromatography coupled to an ultra-violet detector or mass spectrometry (UPLC-MS/MS, HPLC-UV) [13].

The antibiotics to be researched were those belonging to the β-lactam family (amoxicillin, ampicillin and cefadroxil), sulphonamides (sulphadoxine and sulphadimethoxine), aminoglycosides (gentamycin, neomycin and streptomycin) and macrolides (spiramycin, erythromycin and azithromycin). These antibiotics were chosen after analysis of the data collected during the survey of dairy farms.

2.3. Sample preparation

The sample preparation methods differed depending on the antibiotics being tested for. For the beta-lactam assay, sample preparation consisted firstly in ultrasonically treating 10 mL of each milk sample taken in 25 mL flasks for 15 min. The samples were then filtered through 0.45 µm syringe filters and used directly for the assay. For the determination of sulphonamides, preparation consisted firstly of adding 5 mL of correctly homogenised milk to 25 mL of a solution of dichloromethane:acetone (1:1, v/v). The mixture was then mixed with 5 g of sodium chloride and a pinch of anhydrous sodium sulphate. After homogenisation using a mixer-disperser (Heildolf Silent Crusher, Germany) for 30 s at 11 rpm, the solution was centrifuged at 4°C at a speed of 3,000 rpm for 15 min. The supernatant was transferred to a glass test tube and evaporated to dryness in a water bath at 50°C. The dry residue was taken up in 1 mL methanol and 8 mL hexane and centrifuged again. The upper hexane phase was discarded while the lower phase was filtered through syringe filters into vials. For aminoglycosides and macrolides, preparation consisted of liquid/liquid extraction after acidification followed by ultracentrifugation.

2.4. Operating conditions

The Shimadzu HPLC was equipped with two pumps, a degasser, a manual injector, a system controller and a variable wave length UV detector with integrator and data acquisition system. The operating conditions for each antibiotic are given in Table 1. The compounds of interest were identified on the basis of their retention times. Concentrations were determined from the calibration lines established for each compound.

Table 1 Operating conditions for the determination of antibiotics by HPLC

antibiotics investigated	Mobile phase	Mobile phase flow (mL/min)	Detector	Column	Injection volume (µL)
Ampicillin	Acetonitrile + monobasic potassium phosphate (10 :15, v/v)	1.5	UV at 230 nm	C18, 4 mm x 25 cm	10
Amoxicillin					
Cefadroxil	(A): acetonitrile (B): water + formic acid 99% (1000 :1 ; v/v)	0.3	UV at 230 nm	C18, 4 mm x 25 cm	10
Sulfadoxin	Methanol + potassium dihydrogenophosphate 0.1 M (25 :75 ; v/v)	1.0	UV at 268 nm	SUPERCOSILTM LC_18 (C18), 3.0 mm x15 cm	20
Sulfadimethoxin					
Gentamycin	Heptafluorobutyric acid 1 mmol/L (0.013 %) + pentafluoro propionic acid 9.5 mmol/L (0.1 %)	1.0	UV at 220 nm	Kinetex C18, 2.1 mm x 5 cm	10
Neomycin					
Streptomycin					
Spiramycin	Acetonitrile +2-methyl-2-propanol–+ potassium phosphate buffer 0.025 M (pH 6.5) (33 :7: 60 ; v/v/v)	1.0	UV–vis SPD –10AVP at 210 nm	RP18, 4.6 mm x 25 mm	10
Erythromycin					
Azithromycin					

2.5. Data Analyses

Multiple correspondence analyses (MCA) and hierarchical clustering analyses on principal components (HCPC) were conducted. The R Statistical Software were used, specifically the FactoMineR package [14]. MCA examined the relationships existing between antibiotic use practices and concentrations of antibiotic in raw cow's milk. Six variables were used for the analysis: antibiotic residue, respect of deadline waiting, compliance with the protocol for the use of antibiotics, antibiotic sources, Procedure in case of illness Animal and frequency of visits to the vet.

3. Results

3.1. Characteristics of dairy farms

Most of the dairy farm managers surveyed were illiterate (66.7%) and their main activity was livestock rearing (62.2%). 58.9% of managers had owned their farms for more than 10 years. In Korhogo, 70.0% of the farms were set up personally and 30.0% were leased (30.0%). In contrast, the majority of farms in Abidjan and Bouaké were acquired through family takeovers (33.3% for Abidjan and 36.7% for Bouaké). Other methods of acquiring farms in these regions varied between 10 and 30%. Farming was semi-extensive for 86.7% of farms in Bouaké and 76.7% of farms in Korhogo. In Abidjan, the type of farming was divided between semi-intensive and semi-extensive, each of which accounted for 40% of the farms surveyed. Different types of livestock were reared on the sites alongside dairy cows. Poultry was reared on 50% of farms in Abidjan and 43.3% in Bouaké, while 60% of farms in Korhogo had sheep and goats. The number of people working on all the farms surveyed was between 1 and 10, with herds of between 1 and 100 head on 86.71% of farms in Bouaké and 96.7% in Korhogo. In Abidjan, 50% of the farms surveyed had more than 100 head. In addition, the dairy herd consisted of 1-50 head on more than 90% of the farms surveyed. Local breeds accounted for 83.3% of cattle on the farms, although exotic and mixed breeds were found on some farms.

Table 2 Socio-economic profiles of dairy farms

Attributes		**Numbers (%)**			
		Abidjan	**Bouaké**	**Korhogo**	**Total**
Education level	Illiterate	15 (50.0%)	22 (73.3%)	23 (76.7%)	**60 (66.7%)**
	Primary	6 (20.0%)	5 (16.7%)	4 (13.3%)	**15 (16.7%)**
	Secondary	2 (6.7%)	2 (6.7%)	3 (10.0%)	**7 (7.8%)**
	University	7 (23.3%)	1 (3.3%)	0 (0.0%)	**8 (8.9%)**
Main activity of the manager	Agriculture	0 (0.0%)	0 (0.0%)	3 (10.0%)	**3 (3.3%)**
	livestock	15 (50.0%)	24 (80.0%)	17 (56.7%)	**56 (62.2%)**
	Other	15 (50.0%)	6 (20.0%)	10 (33.3%)	**31 (34.4%)**
Period of acquisition of the farm	Under 5 years	10 (33.3%)	1 (3.3%)	3 (10.0%)	**14 (15.6%)**
	5-10 years	3 (10.0%)	11 (36.7%)	9 (30.0%)	**23 (25.6%)**
	Over 10 years	17 (56.7%)	18 (60.0%)	18 (60.0%)	**53 (58.9%)**
Acquisition mode	Family transfer	10 (33.3%)	11 (36.7%)	0 (0.0%)	**21 (23.3%)**
	Personal installation	3 (10.0%)	8 (26.7%)	21 (70.0%)	**32 (35.6%)**
	Purchase	9 (30.0%)	6 (20.0%)	0 (0.0%)	**15 (16.7%)**
	Rental	8 (26.7%)	5 (16.7%)	9 (30.0%)	**22 (24.4%)**
Type of breeding	Semi-intensive	12 (40.0%)	3 (10.0%)	3 (10.0%)	**18 (20.0%)**
	Extensive	6 (20.0%)	1 (3.3%)	4 (13.3%)	**11 (12.2%)**
	Semi-extensive	12 (40.0%)	26 (86.7%)	23 (76.7%)	**61 (67.8%)**
Other breeding on the site	Sheep/goat	5 (16.7%)	6 (20.0%)	18 (60.0%)	**29 (32.2%)**
	Poultry	15 (50.0%)	13 (43.3%)	5 (16.7%)	**33 (36.7%)**
	Other	10 (33.3%)	14 (46.7%)	4 (13.3%)	**28 (31.1%)**
Staff working on the farm	1-10 person	30 (100.0%)	30 (100.0%)	30 (100.0%)	**90 (100.0%)**
Livestock size	1-100 head	15 (50.0%)	26 (86.7%)	29 (96.7%)	**70 (77.8%)**
	+ de 100 head	15 (50.0%)	4 (13.3%)	1 (3.3%)	**20 (22.2%)**

Size of dairy herd	1-50 head	27 (90.0%)	27 (90.0%)	30 (100.0%)	**84 (93.3%)**
	50 - 100 head	3 (10.0%)	3 (10.0%)	0 (0.0%)	**6 (6.7%)**
Race	Local	17 (56.7%)	28 (93.3%)	30 (100.0%)	**75 (83.3%)**
	Exotic	10 (33.3%)	9 (30.0%)	0 (0.0%)	**19 (21.1%)**
	Mixed	3 (10.0%)	2 (6.7%)	3 (10.0%)	**8 (8.9%)**

3.2. Milking and marketing of raw milk on surveyed farms

Milking was carried out manually on all the farms visited, and once a day on more than 88% of the farms. First milk spouts were eliminated on all farms in Bouaké, while in Abidjan and Korhogo they were eliminated on 60% and 50% of farms respectively. Milk was collected exclusively in plastic containers in Bouaké and Korhogo. In Abidjan, only 10% of farmers used non-plastic equipment. Average daily production was over 10 litres on 90% of farms in Abidjan, compared with only 16.7% in Bouaké and 20.0% in Korhogo. The milk collected was packaged in plastic containers by all producers in Bouaké and Korhogo and by over 97% of producers in Abidjan. All Korhogo producers consumed part of their production, compared with 90% in Bouaké and 73.3% in Abidjan. Produce from the farms surveyed was sold to agro-industrial firms, resellers or consumers. Only 3.3% of farmers in Abidjan sold to agro-industrial firms, whereas 40% of farmers in Bouaké and 60% in Korhogo did so. At least 70% of farmers sold their milk to resellers. Only 3.3% of producers in Bouaké sold milk directly to consumers, compared with 50% in Korhogo and 80% in Abidjan.

Table 3 Milking and marketing of milk on dairy farms

Attributes		**Numbers (%)**			
		Abidjan	**Bouaké**	**Korhogo**	**Total**
Number of milking per day	1 time	25 (83.3%)	27 (90.0%)	28 (93.3%)	**80 (88.9%)**
	2 time	5 (16.7%)	3 (10.0%)	2 (6.7%)	**10 (11.1%)**
Milking method	Manual	30 (100.0%)	30 (100.0%)	30 (100.0%)	**90 (100.0%)**
Elimination of the first sprays of milk		18 (60,0%)	30 (100.0%)	15 (50.0%)	**62 (68.9%)**
Milking utensils	Plastic material	27 (90.0%)	30 (100.0%)	30 (100.0%)	**87 (96.7%)**
	Calabash	3 (10.0%)	0 (0.0%)	0 (0.0%)	**3 (3.3%)**
Average daily quantity of milk	1-10 L	15 (50.0%)	15 (50.0%)	24 (80.0%)	**54 (60.0%)**
	10-20 L	8 (26.7%)	10 (33.3%)	6 (20.0%)	**24 (26.7%)**
	over 20 L	07 (23.3%)	5 (16.7%)	0 (0.0%)	**12 (13.3%)**
Packaging of collected milk	Plastic material	29 (96.7%)	30 (100.0%)	30 (100.0%)	**89 (98.9%)**
	Non-plastic material	1 (3.3%)	0 (0.0%)	0 (0.0%)	**1 (1.1%)**
Use of collected milk	Auto-consumption	22 (73.3%)	27 (90.0%)	30 (100.0%)	**79 (87.8%)**
	Sales to agro-industrial companies	01 (3.3%)	12 (40.0%)	18 (60.0%)	**31 (34.4%)**
	Sales to resellers	27 (90.0%)	30 (100.0%)	21 (70.0%)	**78 (86.7%)**
	Direct sales to consumers	24 (80.0%)	1 (3.3%)	15 (50.0%)	**40 (44.4%)**

3.3. Hygienic conditions during milking

To prepare the teats, the majority of farmers (63.3% in Abidjan, 73.3% in Bouaké and 83.3% in Korhogo) carried out dry cleaning by hand. No method was used by 14.4% of the farmers surveyed. The other methods, pre-soaking with soap and washing with water using a sponge, were used by 3.3% and 8.9% of producers respectively. In addition, 63.3% and 96.7% of farmers in Abidjan and Korhogo took no precautions at all at the end of milking. However, in Bouaké, at least 60% of farmers disinfected their cows. The utensils used for milking cows were disinfected by more than 78% of the producers surveyed. When producers knew that cows in their herd were infected, no particular hygiene measures

were taken during milking by 66.7% in Abidjan, 83.3% in Korhogo and 46.7% in Bouaké. The treatments applied to the milk after collection were dominated by filtration, which was carried out on 46.7% of farms. Filtration was combined with pasteurisation on 32.2% of farms, while pasteurisation was used alone on 26.2%. In Abidjan and Bouaké, some producers (3.3%) added additives. To check the quality of the milk collected, microbiological and biochemical tests were used by 33.3% of farmers, while 49.2% and 24.6% applied either the biochemical or the microbiological method respectively. In Bouaké and Korhogo, 10% and 30% of farmers respectively did not use any method of milk quality control.

Table 4 Hygiene practices during milking and milk quality control

Attributes		**Numbers (%)**			
		Abidjan	**Bouaké**	**Korhogo**	**Total**
Method used to prepare the teats	Pre-soaking with soap	03 (10.0%)	0 (0.0%)	0 (0.0%)	**3 (3.3%)**
	Sponge wash with water	06 (20.0%)	2 (6.7%)	0 (0.0%)	**08 (8.9%)**
	Hand dry clean	19 (63.3%)	22 (73.3%)	25 (83.3%)	**66 (73.3%)**
	No method	2 (6.7%)	6 (20.0%)	05 (16.7%)	**13 (14.4%)**
Precautions at the end of milking	Systematic disinfection	01 (3.3%)	01 (3.3%)	01 (3.3%)	**03 (3.3%)**
	Disinfection of a section	11 (36.7%)	18 (60.0%)	0 (0.0%)	**29 (32.2%)**
	No precaution	19 (63.3%)	10 (33.3%)	29 (96.7%)	**58 (64.4%)**
Disinfection of utensils used		22 (73.3%)	22 (73.3%)	27 (90.0%)	**71 (78.9%)**
Precautions for cows infected during milking	Washing and/or disinfecting hands after handling udders	03 (10.0%)	14 (46.7%)	0 (0.0%)	**17 (18.9%)**
	Milking these cows last	01 (3.3%)	0 (0.0%)	0 (0.0%)	**01 (1.1%)**
	Others precautions	06 (20.0%)	2 (6.7%)	5 (16.7%)	**13 (14.4%)**
	No precaution	20 (66.7%)	14 (46.7%)	25 (83.3%)	**59 (65.6%)**
Treatment of milk after collection	Filtration only	18 (60.0%)	12 (40.0%)	12 (40.0%)	**42 (46.7%)**
	Pasteurisation only	02 (6.7%)	6 (20.0%)	9 (30.0%)	**17 (26.2%)**
	Filtration and pasteurisation	9 (30.0%)	11 (36.7%)	9 (30.0%)	**29 (32.2%)**
	Adding additives	01 (3.3%)	01 (3.3%)	0 (0.0%)	**2 (3.1%)**
Quality control of collected milk	Microbiological	5 (16.7%)	3 (10.0%)	8 (26.7%)	**16 (24.6%)**
	Biochemical	20 (66.7%)	7 (23.3%)	5 (16.7%)	**32 (49.2%)**
	Microbiological and Biochemical	5 (16.7%)	17 (56.7%)	8 (26.7%)	**30** (33.3%)
	No method	0 (0.0%)	3 (10.0%)	09 (30.0%)	**12 (13.3%)**

3.4. Monitoring and preventive treatment of dairy cows

Dairy cows were vaccinated on all the farms surveyed. Deworming was also common practice on all farms in Bouaké and on 90% of farms in Abidjan and Korhogo. The percentage of farms applying tuberculin treatment varied from town to town. It was highest in Bouaké with 60%, followed by Korhogo with 33.3% and Abidjan with 20%. Vaccination against brucellosis was less frequent, being carried out on only 22.2% of the farms surveyed. Farmers in Abidjan and Korhogo had not received any training in animal health and only 3.3% had been trained in Bouaké. All the farmers in Bouaké and Korhogo and almost all in Abidjan (90.0%) had recourse to vets or technicians to monitor their cows. The frequency of interventions was mostly between 1 and 5 times a year for 96.7% of farmers.

In terms of pathologies, the most common on the farms visited were foot ailments, with a percentage of 90% in Bouaké, 80% in Abidjan and 60% in Korhogo. These ailments were followed by respiratory ailments and digestive ailments,

which were observed on 55.6% and 45.6% of the farms respectively. Mastitis was reported on 20% of farms. Urinary disorders were reported on 6.7% of farms in Bouaké and Korhogo. Apart from these ailments, other ailments were reported on 60% of the farms visited in Korhogo. Between 43% and 60% of farmers systematically resorted to self-medication in the event of infection of cows in the herd. Automatic recourse to the technician concerned 40% of farmers in Abidjan, 56.7% in Bouaké and 50% in Korhogo.

The main reasons for self-medication varied from town to town. In Abidjan, 66.7% of farmers self-medicated out of experience and 26.7% to save time. In Bouaké, the availability of products in stock was the main reason for self-medication (46.7%), followed by experience (40%). In Korhogo, 43.3% of farmers practised self-medication for economic reasons and 36.7% because of the availability of products in stock. To combat disease, antibiotics were used by all farmers in Abidjan and Korhogo and by 90% of farmers in Bouaké.

Table 5 Preventive health monitoring of animals on dairy farms in Côte d'Ivoire

Attributes		**Numbers (%)**			
		Abidjan	**Bouaké**	**Korhogo**	**Total**
Cow health monitoring	Vaccination	30 (100.0%)	30 (100.0%)	30 (100.0%)	**90 (100.0%)**
	Parasite control	27 (90.0%)	30 (100.0%)	27 (90.0%)	**84 (93.3%)**
	Tuberculination	6 (20.0%)	18 (60.0%)	10 (33.3%)	**34 (37.8%)**
	Brucellosis vaccine	4 (13.3%)	9 (30.0%)	7 (23.3%)	**20 (22.2%)**
Animal health training		0 (0.0%)	1 (3.3%)	0 (0.0%)	**1 (1.1%)**
Use of vets/technicians to monitor cows		27 (90.0%)	30 (100.0%)	30 (100.0%)	**87 (96.7%)**
Frequency of vet / technician visits per year	1-5 time	27 (90.0%)	30 (100.0%)	30 (100.0%)	**87 (96.7%)**
	5-10 time	3 (10.0%)	0 (0.0%)	0 (0.0%)	**3 (3.3%)**
Frequent pathologies	Mammites	7 (23.3%)	7 (23.3%)	4 (13.3%)	**18 (20.0%)**
	Respiratory disorders	19 (63.3%)	23 (76.7%)	8 (26.7%)	**50 (55.6%)**
	Foot disorders	24 (80.0%)	27 (90.0%)	18 (60.0%)	**69 (76.7%)**
	Digestive disorders	15 (50.0%)	14 (46.7%)	12 (40.0%)	**41 (45.6%)**
	Urinary disorders	0 (0.0%)	2 (6.7%)	2 (6.7%)	**4 (4.4%)**
	Other disorders	0 (0.0%)	0 (0.0%)	18 (60.0%)	**18 (20.0%)**
procedure for infected cows in the herd	Automatic technician call-out	12 (40.0%)	17 (56.7%)	15 (50.0%)	**44 (48.9%)**
	Self-medication	3 (10.0%)	8 (26.7%)	12 (40.0%)	**23 (25.6%)**
	Self-medication then veterinary	15 (50.0%)	5 (16.7%)	3 (10.0%)	**23 (25.6%)**
Main reason for self-medication	Product available in stock	1 (3.3%)	14 (46.7%)	11 (36.7%)	**26 (28.9%)**
	Economic reasons	1 (3.3%)	4 (13.3%)	13 (43.3%)	**18 (20.0%)**
	Save time	8 (26.7%)	0 (0.0%)	2 (6.7%)	**10 (11.1%)**
	Experience	20 (66.7%)	12 (40.0%)	4 (13.3%)	**36 (40.0%)**
Disease control methods	Antibiotics	30 (100.0%)	27 (90.0%)	30 (100.0%)	**87 (96.7%)**
	Other	25 (83.3%)	7 (23.3%)	10 (33.3%)	**42 (46.7%)**

3.5. Antibiotic use practices on dairy farms

The primary source of antibiotics was the pharmacy. It was used by 73.3% of producers in Abidjan, 70% in Bouaké and 40% in Korhogo. In addition, supplies could be obtained from vets (25.6%) and the parallel market (13.3%). Protocols for the use of antibiotics were followed by 50% of farmers in Abidjan, 50% in Bouaké and 60% in Korhogo. Of those who did not follow the protocol, 51.1% felt that the protocols were not effective, 30% preferred to rely on their experience or habit of use, and 18.9% felt that the protocol was too restrictive. The primary criterion for choosing antibiotics was experience or habit (66.7%), followed by efficacy (52.2%). Some farmers in Abidjan (30%) and Bouaké (23.3%) supplemented animal feed with antibiotics. The waiting period following treatment of the cows was respected by 70% of producers in Abidjan, 80.0% in Bouaké and 90% in Korhogo. Among producers who respected the waiting period, 63.3% in Abidjan, 60.0% in Bouaké and 83.3% in Korhogo gave milk to their calves during this period. Non-compliance with the waiting period was mainly due to ignorance of its existence in Abidjan (63.3%) and Bouaké (96.7%).

Table 6 Practices relating to the use of antibiotics on dairy farms

Attributes		**Numbers (%)**			
		Abidjan	**Bouaké**	**Korhogo**	**Total**
Sources of supply for antibiotics	Parallel market	1 (3.3%)	3 (10.0%)	8 (26.7%)	**12 (13.3%)**
	Pharmacy	22 (73.3%)	21 (70.0%)	12 (40.0%)	**55 (61.1%)**
	Veterinary	7 (23.3%)	6 (20.0%)	10 (33.3%)	**23 (25.6%)**
Monitoring protocol for the use of antibiotics		12 (40,0%)	15 (50.0%)	18 (60.0%)	**45 (50.0%)**
If not, why?	Protocol too restrictive	3 (10.0%)	7 (23.3%)	7 (23.3%)	**17 (18.9%)**
	Protocol not too effective	15 (50.0%)	23 (76.7%)	8 (26.7%)	**46 (51.1%)**
	Experience of use	12 (40.0%)	0 (0.0%)	15 (50.0%)	**27 (30.0%)**
Criteria for choosing antibiotics to administer	Spectrum of activity	3 (10.0%)	21 (70.0%)	15 (50.0%)	**39 (43.3%)**
	Cost	0 (0.0%)	1 (3.3%)	1 (3.3%)	**2 (2.2%)**
	Efficiency	22 (73.3%)	17 (56.7%)	8 (26.7%)	**47 (52.2%)**
	Availability	9 (30.0%)	15 (50.0%)	6 (20.0%)	**30 (33.3%)**
	Experience	28 (93.3%)	16 (53.3%)	16 (53.3%)	**60 (66.7%)**
Supplementing animal feed with antibiotics		9 (30.0%)	7 (23.3%)	0 (0.0%)	**16 (17.8%)**
Respecting the waiting period for treatment of cows		21 (70.0%)	24 (80.0%)	27 (90.0%)	**72 (80.0%)**
If yes	Disposed of with waste water	2 (6.7%)	1 (3.3.0%)	0 (0.0%)	**3 (3.3%)**
	Fed to calves	19 (63.3%)	18 (60.0%)	25 (83.3%)	**62 (68.9%)**
	Other	9 (30.0%)	11 (36.7%)	5 (16.7%)	**25 (27.8%)**
If no	Economic reasons	9 (30.0%)	1 (3.3%)	0 (0.0%)	**10 (11.1%)**
	Ignorance of the waiting period	19 (63.3%)	29 (96.7%)	4 (13.3%)	**52 (57.8%)**
	Other	2 (6.7%)	0 (0.0%)	26 (86.7%)	**28 (31.1%)**

3.6. Antibiotic therapy on surveyed dairy farms

Different families of antibiotics were used in response to different ailments in dairy cows. Sulphonamides were used on 21.1% of farms surveyed to treat mastitis. In Abidjan, macrolides were also used by 3.3% of farmers to treat these conditions. In the case of respiratory diseases, macrolides were used by farmers in all the areas surveyed. Penicillins

were used in Abidjan and tetracyclines in Abidjan and Korhogo. In Bouaké, the antibiotics most commonly used for foot infections were cephalosporins 1 and 2 G (40%), followed by sulphonamides (26.7%). In Bouaké and Korhogo, the most commonly used antibiotics were sulphonamides. In the treatment of digestive disorders, aminoglycosides and sulphonamides were the most widely used on the farms visited, with 22.2% and 23.3% respectively. Macrolides, penicillins and tetracyclines were also used to treat these conditions.

With regard to the method of antibiotic administration, intravenous (IV) antibiotics were most commonly used. It accounted for 83.3% of cases in Abidjan, 66.6% in Bouaké and 33.3% in Korhogo. The oral and intramuscular routes were used in 23.3% and 4.4% of farms respectively.

Table 7 Antibiotics used on dairy farms depending on the disease

Antibiotics used		**Numbers (%)**			
		Abidjan	**Bouaké**	**Korhogo**	**Total**
Mammites	Macrolides	1 (3.3%)	0 (0.0%)	0 (0.0%)	1 (1.1%)
	Sulfamides	8 (26.7%)	6 (28.6%)	5 (16.7%)	19 (21.1%)
Respiratory disorders	Macrolides	3 (10.0%)	10 (33.3%)	4 (13.3%)	17 (18.9%)
	Penicillins	5 (16.7%)	0 (0.0%)	0 (0.0%)	5 (5.6%)
	Tetracyclines	1 (3.3%)	0 (0.0%)	3 (10.0%)	4 (4.4%)
Foot disorders	Aminoglycosides	01 (3.3%)	0 (0.0%)	0 (0.0%)	1 (1.1%)
	Cephalosporins 1st and 2nd G	2 (6.7%)	12 (40.0%)	0 (0.0%)	14 (15.6%)
	Penicillins	2 (6.7%)	2 (6.7%)	0 (0.0%)	4 (4.4%)
	Sulfamides	8 (26.7%)	8 (26.7%)	5 (16.7%)	21 (23.3%)
	Tetracyclines	0 (0.0%)	5 (16.7%)	0 (0.0%)	5 (5.6%)
Digestive disorders	Aminoglycosides	10 (33.3%)	6 (20.0%)	4 (13.3%)	20 (22.2%)
	Macrolides	2 (6.7%)	5 (16.7%)	3 (10.0%)	10 (11.1%)
	Penicillins	1 (3.3%)	2 (6.7%)	0 (0.0%)	3 (3.3%)
	Sulfonamides	10 (33.3%)	7 (23.3%)	4 (13.3%)	21 (23.3%)
	Tetracyclines	1 (3.3%)	6 (20.0%)	0 (0.0%)	9 (10.0%)
	Aminoglycosides + Penicillins	2 (6.7%)	0 (0.0%)	0 (0.0%)	2 (2.2%)
Antibiotic administration	Intramuscular	2 (6.7%)	2 (6.7%)	0 (0.0%)	4 (4.4%)
	Intravenous	25 (83.3%)	20 (66.7%)	10 (33.3%)	55 (61.1%)
	Oral use	8 (26.7%)	10 (33.3%)	3 (10.0%)	21 (23.3%)

3.7. Prevalence of antibiotic residues in raw cow's milk

Of the 135 samples analysed, 34 (23.6%) contained antibiotic residues. The percentages of contaminated samples were higher in samples taken from retailers than in samples taken from farmers. For samples taken from farmers, 23.3% were contaminated in Bouaké and Korhogo, compared with 16.7% in Abidjan. In the case of samples taken from resellers, the percentages contaminated were 26.7%, 33.3% and 40.0% in Abidjan, Bouaké and Korhogo respectively.

Table 8 Prevalence of antibiotic residues in cow's milk

Attributes	Numbers (%)			
	Abidjan	Bouaké	Korhogo	Total
Farmer	5/30 (16.7%)	7/30 (23.3%)	7/30 (23.3%)	19/90 (21.1%)
Reseller	4/15 (26.7%)	5/15 (33.3%)	6/15 (40.0%)	15/45 (33.3%)
Total	9/45 (20.0%)	12/45 (26.7%)	13/45 (28.9%)	34/135 (23.6%)

3.8. Antibiotic residue concentrations in raw milk

Of the 11 antibiotics tested in samples of raw cow's milk containing antibiotic residues, ampicillin, neomycin and azithromycin were not detected. The eight antibiotics identified were sulfadoxine, sulfadimethoxine, gentamicin, streptomycin, erythromycin and spiramycin. The concentrations of amoxicillin obtained were above the maximum residual limit (MRL) for all samples from Abidjan and Bouaké, and for 50% of samples from Korhogo.

Values ranged from 74.8 to 138.8 µg/L, from 53.5 to 127.6 µg/L, and from 2.87 to 32.9 µg/L for the Abidjan, Bouaké and Korhogo samples respectively. Erythromycin was also detected at concentrations above the MRL in all the Abidjan samples and in 50% of the Bouaké and Korhogo samples. Apart from cefadroxil, for which the MRL is not known, the concentrations of the other antibiotics were below the MRL.

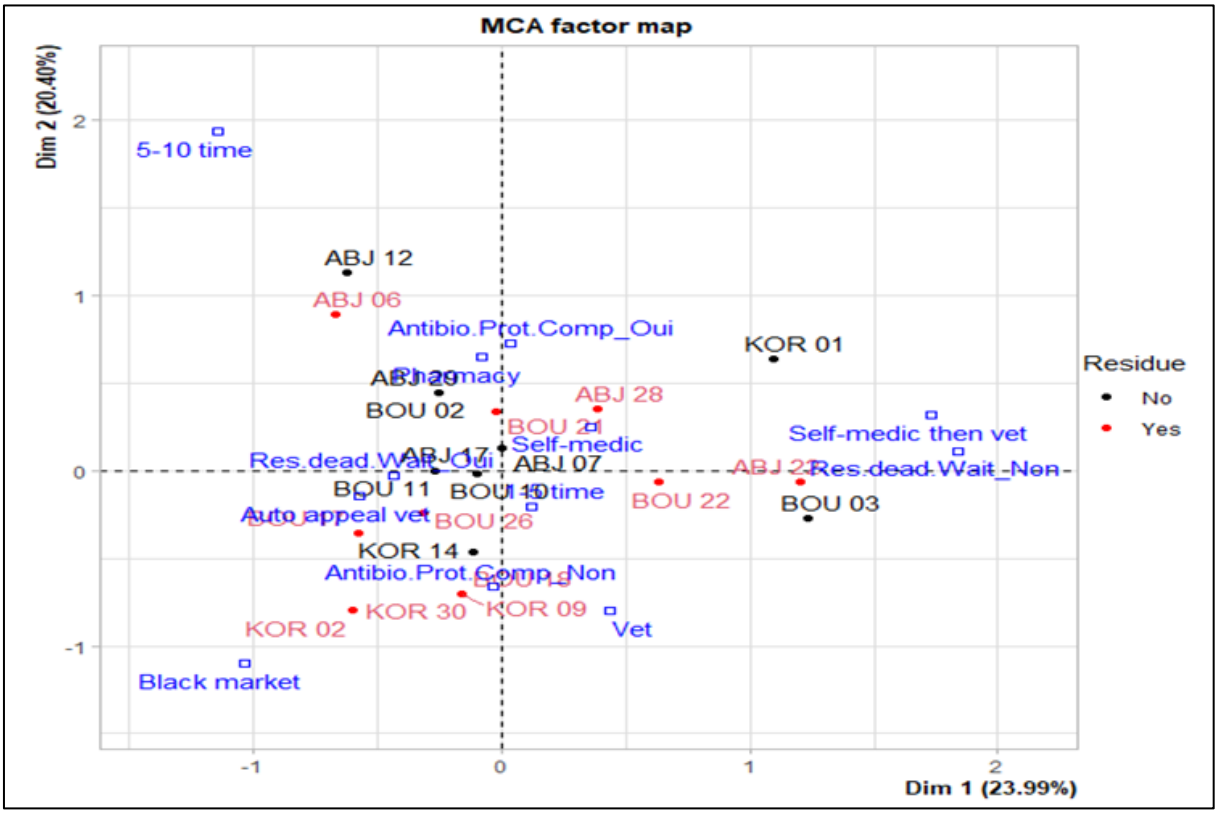
Table 9 Antibiotic concentrations in raw cow's milk

Localities	Antibiotics identified	Minimum concentration (µg/L)	Maximum concentration (µg/L)	Average concentration (µg/L)	MRL Authorised (µg/L)	% exceeding MRL
Abidjan	Amoxicillin	74.8	138.8	96.5	4	100 %
	Cefadroxil	6.8	13.1	11.1	Ind	-
	Sulfadoxine	2.1	6.9	4.9	100	0 %
	Sulfadimethoxine	2.5	5.3	4.5	100	0 %
	Gentamicin	7.2	71.8	29.7	100	0 %
	Streptomycin	10.1	41	30.8	200	0 %
	Erythromycin	25.9	133.9	65.3	40	100 %
	Spiramycin	20.6	145.2	57.8	200	0 %
Bouaké	Amoxicillin	53.5	127.6	75.9	4	100 %
	Cefadroxil	8.3	14.6	12.0	Ind	-
	Sulfadoxine	4.8	8.9	5.9	100	0 %
	Sulfadimethoxine	3.2	4.4	4.0	100	0 %
	Gentamicin	34.3	54.1	39.5	100	0 %
	Streptomycin	15.9	36.2	29.6	200	0 %
	Erythromycin	10.5	48.8	32.3	40	50 %
	Spiramycin	17.4	65.6	24.7	200	0 %
Korhogo	Amoxicillin	2.87	32.9	11.4	4	50 %
	Cefadroxil	11.2	15.1	13.1	Ind	-
	Sulfadoxine	2.2	2.9	2.53	100	0 %
	Sulfadimethoxine	3.1	3.4	3.23	100	0 %

	Gentamicin	42.3	62.8	52.27	100	0 %
	Streptomycin	12	12.9	12.43	200	0 %
	Erythromycin	32.8	41.5	37.13	40	50 %
	Spiramycin	26.6	57.7	38.7	200	0 %

3.9. Relation between antibiotic use practices and concentrations of antibiotic in raw cow's milk

Dimensions 1 and 2, generated by multiple component analysis, explained 44% of the total variability. Dimension 1 explained 23.99%, compared with 20.48% for dimension 2. The arrangement of samples containing or not containing antibiotic residues showed that the presence of residues in milk was not related to its origin. Furthermore, the presence of antibiotic residues was not specific to any particular practice. However, the plot confirmed that most farmers in Korhogo did not follow the protocol for antibiotic use and purchased antibiotics on the black market.



1-5 time, 5-10 time= frequency of visits to the vet; Proc Sick Anim= Procedure in case of illness Animal; Antibio Sour= antibiotic sources; Antibio Prot= compliance with the protocol for the use of antibiotics; Res dead Wait= respect of deadline waiting; residue= antibiotic residue

Abj= Abidjan ; Bou= Bouaké ; Kor= Korhogo

Figure 1 Distribution of farms and their respondents in the first two factors of the MCA performed according antibiotic practice

4. Discussion

The aim of this study was to identify the impact of antibiotic treatment practices on dairy farms on the sanitary quality of raw cow's milk in Côte d'Ivoire.

The survey carried out in dairy farms in the Ivorian cities of Abidjan, Bouaké and Korhogo showed that most of the dairy farm managers surveyed were illiterate and had livestock farming as their main activity. The lack of education among farm managers may be a major obstacle to understanding and adopting good dairy production practices. These managers should therefore be supported through awareness-raising campaigns. Authors have reported that awareness campaigns could play a crucial role by providing essential information visually, verbally and interactively [15]. Livestock farming was predominantly semi-extensive, in line with studies conducted in several West African countries, including

Senegal, Niger and Burkina Faso [16]. In addition, the farms were generally characterised by a herd of between 1 and 100 head, with a dairy herd of between 1 and 50 head on more than 90% of farms [6]. These authors already reported on a traditional dairy farming system with small, low-production dairy farms in the urban and peri-urban area of Abidjan. According to these authors, this low productivity is specific to local breeds in West Africa. These observations were confirmed in this study, in which 83.3% of the cattle encountered on the farms were local breeds, with a daily milk production of between 1 and 20 litres. These data are similar to those of [17] who, following a survey in the Poro region of northern Côte d'Ivoire, estimated the quantity of milk produced per day at around 24 litres. Dairy farms are also characterised by their diversification, in particular by the integration of poultry, sheep and other animal husbandry. This strategy aims to mitigate the economic risks associated with a single activity.

On all farms visited, milking was done manually and took place once a day on more than 88% of farms. Manual milking once a day is common on African family farms, mainly due to resource and labour constraints, as well as animal performance. In the case of a local cow reared under traditional conditions, increasing the milking frequency to two or three times a day quantitatively improves the cow's productivity without noticeable variations in milk composition [18]. Sales to agro-industrial firms were made by only 3.3% of producers in Abidjan, whereas they were made by 40% of producers in Bouaké and 60% of producers in Korhogo. The relatively low percentages in Abidjan can be explained by the high demand for raw milk among consumers. Consumption of dairy products is increasing rapidly in West Africa, driven by population growth, urbanisation and changing eating habits, particularly among the middle classes [19]. However, rearing conditions, in particular feeding, milking hygiene and the handling of raw milk, influence its nutritional and microbiological quality [20, 7]. In this study, the hygienic conditions observed during milking showed that the majority of farmers (63.3% in Abidjan, 73.3% in Bouaké and 83.3% in Korhogo) carried out dry cleaning by hand, and that no method was applied by 14.4% of the farmers surveyed. In addition, when producers found that cows were part of their herd, no particular hygiene measures were taken during milking by more than 50% of producers. In addition, plastic equipment was predominantly used for collecting and packaging the milk. These practices could lead to contamination of the milk with pathogens. Measures such as mechanised milking and the systematic washing of udders with stainless steel equipment could help to improve milk quality. Before sale, 58% of producers pasteurise the milk and 46% carry out microbiological testing. Pasteurisation and microbiological control should be encouraged and applied by all producers, because half of consumers in Abidjan consume raw milk directly after purchase, without heat treatment [21].

Medical prophylaxis measures (vaccination and deworming) are applied by at least 90% of farmers. Similar studies in other parts of Africa, such as Kenya and Nigeria, have also shown strong adherence to these preventive practices [22, 23]. Despite the application of these measures, pathologies such as foot ailments, respiratory ailments, digestive ailments, mastitis and urinary ailments have been observed. Between 43% and 60% of farmers systematically self-medicate in the event of infection in the cow herd for various reasons: experience, availability of products in stock, saving time and economic reasons. Cattle farmers who self-medicate often rely on previous veterinary prescriptions, without considering the potential risks to human health [24]. Nearly half of producers did not follow antibiotic use protocols.

On most farms, pathologies were treated using antibiotics from pharmacies, vets or the parallel market. Other authors [25, 26, 4] reported that no veterinarian was used to purchase and administer antibiotics. The antibiotics used on the dairy farms visited were macrolides, penicillins, sulphonamides, tetracyclines, aminoglycosides and first- and second-generation cephalosporins. These results are in agreement with those of [4] who reported that tetracyclines, penicillins, sulphonamides and macrolides were the most commonly used families of antibiotics. In this study, antibiotic residues were observed in 21.1% of samples collected from farmers and in 33.3% of samples obtained from retailers. The antibiotics identified were sulfadoxine, sulfadimethoxine, gentamicin, streptomycin, erythromycin and spiramycin. Bagré et al. [27] detected aminoglycosides and/or quinolones and/or macrolides and beta-lactams and/or sulphonamides and/or tetracyclines in 17.24% and 51.72% of raw milks respectively. In addition, in our study we observed concentrations of amoxicillin and erythromycin above the MRL in at least 50% of the samples. According to Kurjogi et al. [28], antibiotic residues in raw cow's milk are a major concern for human health and food safety. Gaouar et al. [29] reported that the presence of these residues could be due to improper use of antibiotics, particularly in the case of self-medication and failure to respect the waiting period between treatment of cows. The presence of antibiotic residues could lead to a risk of direct toxicity for consumers, an allergic risk, a carcinogenic risk, a risk of pathology linked to changes in digestive flora and a risk of the appearance, selection and spread of bacterial resistance to antibiotics within human and animal populations [30]. Several authors have reported that strains of E. coli and Salmonella spp. isolated from dairy products in Greece, Burkina Faso and other countries were resistant to amoxicillin, amoxicillin-clavulanic acid, trimethroprime sulphamethoxazole, erythromycin, tobramycin, chloramphenicol and sulphonamides [31, 32, 33].

5. Conclusion

The aim of this study was to describe antibiotic treatment practices on dairy farms in Côte d'Ivoire and to assess the presence of antibiotic residues in the raw milk produced on these farms. Risky practices were identified, including self-medication, failure to comply with regulatory withdrawal times and the use of unauthorised antibiotics. Antibiotics from the beta-lactam, aminoglycoside, tetracycline, macrolide and sulphonamide families are used to treat diseases in dairy cows. However, inappropriate use of these antibiotics can lead to high levels of residues in raw milk, sometimes in excess of legal limits. These findings highlight the need to educate those involved in the responsible use of antibiotics in order to minimise the risks of antibiotic resistance and guarantee the safety of raw milk.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare no conflict of interest.

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