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Consumption habits of broiler offal and its contamination by antibiotic residues in the Northeast of Algeria

Hábitos de consumo de vísceras de pollos de engorde y sus niveles de residuos de antibióticos, en el nordeste de Argelia

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ABSTRACT

Antibiotic use has promoted poultry production intensification. However, irrational use of these drugs results in an accumulation of toxic and harmful residues in the edible products of the treated birds. These residues eventually affect the consumer's health by transferring antibiotic–resistant bacteria and causing allergies and other pathologies. This study aims to describe the consumption habits of broiler offal in two Algerian Provinces: Batna and Sétif (North–East of Algeria). In addition, a qualitative study was conducted to screen antibiotic residues in broiler offal commercialized in these regions. A total of 250 consumers were randomly surveyed using a questionnaire including questions regarding their socio–demographics, their consumption habits of offal, and their knowledge of the risks associated with the presence of antibiotic residues in poultry products. The study was conducted from 5th February to 25th March 2019. Antibiotic residues were investigated in 66 broiler samples (36 livers and 30 gizzards) using an agar diffusion test inspired by the conventional microbiological method of the four plates. The survey revealed high consumption rates of offal: 83.3% in Batna and 92.0% in Sétif. The liver was the most preferred offal in both regions due to its palatability (86.5%) and nutritional value (67.3%). Most respondents (68% in Batna and 52% in Sétif) are aware of the risks of antibiotic residues in offal. The results of the antibiotic residue investigation showed an overall contamination rate of 33.3%. Livers were more contaminated than gizzards. In both regions, the widespread consumption of broiler offal and its contamination by antibiotic residues constitute a potential health hazard for consumers.

Key words: Antibiotic residues; broiler offal; health; consumer survey

RESUMEN

El uso de antibióticos ha promovido la intensificación de la producción avícola. Sin embargo, el uso irracional de estos fármacos provoca una acumulación de residuos tóxicos y nocivos en los productos comestibles de las aves tratadas. Estos residuos acaban afectando a la salud del consumidor al transferir bacterias resistentes a los antibióticos y provocar alergias y otras patologías. Este estudio pretende describir los hábitos de consumo de vísceras de pollos de engorde en dos provincias argelinas: Batna y Sétif (noreste de Argelia). Además, se realizó un estudio cualitativo para analizar los residuos de antibióticos en las vísceras de pollos de engorde comercializados en estas regiones. Se encuestó aleatoriamente a 250 consumidores mediante un cuestionario que incluía preguntas sobre sus características sociodemográficas, sus hábitos de consumo de vísceras y su conocimiento de los riesgos asociados a la presencia de residuos de antibióticos en productos avícolas. El estudio se llevó a cabo del 5 de febrero al 25 de marzo de 2019. Se investigaron los residuos de antibióticos en 66 muestras de pollos de engorde (36 hígados y 30 mollejas) mediante una prueba de difusión en agar inspirada en el método microbiológico convencional de las cuatro placas. La encuesta reveló altos índices de consumo de vísceras: 83,3% en Batna y 92,0% en Sétif. El hígado fue la víscera preferida en ambas regiones por su palatabilidad (86,5%) y su valor nutritivo (67,3%). La mayoría de los encuestados (68% en Batna y 52% en Sétif) son conscientes de los riesgos de los residuos de antibióticos en las vísceras. Los resultados de la investigación sobre residuos de antibióticos mostraron una tasa global de contaminación del 33,3%. Los hígados estaban más contaminados que las mollejas. En ambas regiones, el consumo generalizado de vísceras de pollos de engorde y su contaminación por residuos de antibióticos constituyen un peligro potencial para la salud de los consumidores.

Palabras clave: Residuos de antibióticos; vísceras de pollos de engorde; salud; encuesta de consumidores

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INTRODUCTION

Offal is an important by-product of the meat industry. The main factors affecting their consumption Worldwide are consumer preferences, income, cultural and religious considerations [1]. In many Countries, offal helps reduce protein malnutrition and food insecurity and can be used regularly in a low-cost approach to obtain high-quality protein and nutrition [2]. All slaughterhouse meat by-products (including the spleen, pancreas, and uterus) are commonly used in human food. Depending on the species from which they originate, these products are consumed differently in several nations [3].

In Algeria, poultry livers, hearts, and gizzards are commercialized individually and in larger quantities. In 2021, chicken (*Gallus gallus domesticus*) meat was the predominant animal production, with more than 257,644 tons [4]. The consumption of offal is rooted in the Country's culinary traditions. Today, offal (liver, gizzard, and heart) is available for sale individually to reduce the risk of salmonella cross-contamination [5].

The industrialization of poultry farming in the World has been accompanied by the widespread use of antibiotics to prevent and treat infections. Antibiotics have also been added to the feed at a sub-therapeutic level to improve growth rate and feed use efficiency [6]. This practice provides some economic benefits to producers in general. However, it is also associated with a few concerns [7].

The residues and their breakdown products have several side effects on the human body and, in a broader sense, on the environment. The repeated exposure of farm animals to small doses of antibiotics contributes significantly to the selection of multi-resistant pathogenic bacteria strains. Since many of these antibiotics are the same or substitutes for antibiotics used in human therapeutic practices, selecting such strains is a real Public Health problem [8, 9, 10].

In addition to antibiotic resistance, the presence of these residues in foodstuffs of animal origin is associated with several risks: damage of bone marrow (Chloramphenicol), carcinogenic risks (Sulfamethazine, Oxytetracycline, and Furazolidone) [11], allergic risks (Penicillin, Streptomycin) and modification of the intestinal flora (Tetracyclines) [12].

Algeria is an example of such Countries where the risks of developing antibiotic resistance are very high due to the unregulated access to veterinary antibiotics, the non-respect of waiting periods, the non-respect of dosing, and the lack of awareness concerning antibiotic resistance [13]. Furthermore, no legal regulations set the maximum residue limits (MRLs) for antibiotics in foodstuffs of animal origin.

In Algeria, several studies have focused on the detection and quantification of antibiotic residues in chicken meat [14], milk [15], and honey [16], but very few have studied their presence in poultry offal. Indeed, there is currently a lack of knowledge regarding the consumption patterns of chicken offal and the extent of its antibiotic residue contamination.

In this context, this study was conducted in two Algerian Provinces (Batna and Setif) to survey the dietary practices associated with broiler offal consumption and the level of awareness among consumers regarding the hazards of antibiotic residues to their health. This study also assessed the level of antibiotic residue contamination of broiler offal (liver, gizzard) commercialized in these Provinces.

MATERIALS AND METHODS

Study area

The study covered communes in the Provinces of Batna and Sétif (North-Eastern Algeria). Batna is located in North-Eastern Algeria, in the Aurès Region. Sétif is considered a crossroads between East and West, North and South Algeria [17]. Together with the Provinces of Média and Bouira, they account for a quarter of national production, i.e., 1.6 million tons of poultry products [4].

Survey on consumption of broiler offal:

Type of study

This descriptive cross-sectional survey used direct interviews with the respondents (face-to-face). A pilot study was conducted in January 2019 on 20 respondents residing in Batna and Setif Provinces. The pilot study allowed us to test the questionnaire and improve its content by adding, removing, or reformulating some questions.

Study population

Respondents were randomly selected and interviewed from February 5th to March 25th, 2022. The only inclusion criterion was the age of the respondents (at least 18 years old). The participants were interviewed in urban areas and in different locations: markets, universities, polyclinics, and administrative establishments.

Questionnaire

The final version of the questionnaire contained 16 questions divided into three sections: The first one was devoted to the respondent identification; the second section focused on habits of consumption of chicken offal, and the third section aimed to evaluate the respondents' level of knowledge about the risks of contamination of foodstuffs by antibiotic residues.

Microbiological screening of antibiotic residues in broiler offal:

Sample collection

The samples were collected from legal businesses chosen for the availability of broiler offal, and compliance with regulatory slaughtering conditions.

A total of 66 samples of broiler offal (liver and gizzard) were collected (TABLE I). For each sample, a minimum weight of 30 g was collected aseptically. Each sample was placed in a separate, sterile, hermetically sealed, clearly marked, and labeled bag. The collected samples were transported in a cooler with ice packs to the microbiology laboratory and were frozen at -18°C for 24 h (Fiocchetti brand freezer 489-2013, Model ULV0481M1BS1856, class C, S/N: 47020 made in Italy).

TABLE I
Collection of samples from different locations in the two willayas

	Batna	Setif
Slaughterhouses	06 livers / 06 gizzards	06 livers / 06 gizzards
Butchers, Sales outlets, Markets	12 livers / 09 gizzards	12 livers / 09 gizzards
Total	66 Samples	

Residue screening

The microbiological screening for antibiotic residues was carried out using an agar diffusion test inspired by the Agence Française de Sécurité Alimentaire 2010 [18] reference method (four-box method). This method allows for the detection of the following families of antibiotics: beta-lactams, Tetracyclines, Sulphonamides, Aminoglycosides, and Macrolides [19]. This study used *Bacillus subtilis* strain ATCC 6633 grown at three pH levels (pH 6.0; pH 7.2; pH 8.0). It is a ubiquitous telluric germ from the Bacillaceae family. Its optimal growth temperature is between 30 and 37°C. To rule out antibiotic resistance of the used strain, a pre-test was conducted by cultivating the germ in the antibiotic discs used in this study, namely Penicillin, Tetracycline, Tylosin, and Colistin. The germ was inhibited by all 4 antibiotics.

Preparation of the culture medium

The culture medium used was Mueller Hinton. To prepare 1 L of this medium, 1 L of distilled water and 51 g of Muller Hinton powder were used. The solution obtained was divided into two (0.5 L) and was then adjusted with HCL to obtain a pH of 6, and with NaOH to obtain a pH of 8. The solution was autoclaved at 120°C for 15 min.

Sample processing

A few minutes before use, liver and gizzard samples were removed from the freezer for thawing. A cylindrical core (6 mm diameter × 2 cm length) was taken from each sample and then sliced into small discs (6 × 2 mm) using a scalpel. Sample discs were placed on a circle approximately 1cm from the periphery of each Petri dish inoculated with *Bacillus subtilis* [20]. After 20 h of incubation at 30°C, the diameters of the inhibition zones around the sample discs were measured. The samples with an annular zone size (distance between the edge of the disc and the outer limit of the inhibition zone) of at least 2 mm were considered positive (FIG.1 and FIG.2).

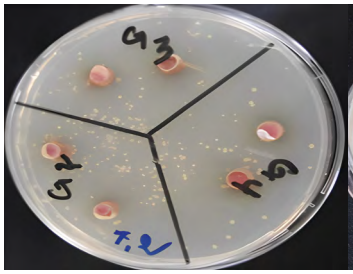

FIGURE 1. Negative result

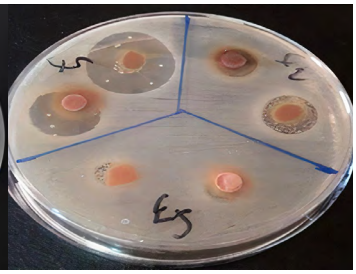

FIGURE 2. Positive result

Data processing and analysis

The data from both studies were analyzed using EPI INFO software version 3.3.2 [21]. Descriptive statistics were used to determine the means and percentages of the survey data and antibiotic contamination rates of the analyzed samples.

Chi-square statistics were used to determine the relationship between age, gender, level of education, antibiotic residue awareness, and consumption of broiler offal. $P < 0.05$ was considered statistically significant. The graphs were produced using Excel 2013 (Microsoft©).

Ethical aspect

This study was approved by the scientific council of the Institute of Nutrition, Food and Agri-Food Technologies (INATAA), University des Frères Mentouri Constantine 1 (Algeria). The respondents of the survey were interviewed after explaining the purpose of the interview and obtaining their verbal consent.

RESULTS AND DISCUSSIONS

Survey on consumption of broiler offal:

Consumers' demographics attributes

This study looks at the consumption habits of broiler offal in two Provinces of North-East Algeria. No study on this subject has yet been published in Algeria.

A total of 250 respondents were interviewed (TABLE II). The study included male and female respondents with close percentages (49.6% female vs. 50.4% male). The participants' average age was 37.1 ±11.6, with 30 years as the highest frequency (8.4%). The eldest participant was 68, and the youngest was 20 years old. More than half of the respondents had a university education (58.8%), while 6.8% had a low education level not exceeding primary education. The largest group of respondents had their source of income working as civil servants or in the private sector or were retired. The majority of participants were from an urban area (72.8%). The analysis of the demographic characteristics of the studied population shows that the distribution by sex and age corresponds to the national average. The percentage of respondents with a high level of education exceeds the national average. In fact, according to the latest available data (2015), 68.2% of the Algerian adult population has a low level of education, 19.3% a medium level, and 12.5% a high level [22]. This bias is probably due to the higher levels of self-confidence and willingness to participate found in people with higher levels of education, as reported by Claret *et al.* [20].

Levels and motivations of chicken offal consumption in the studied regions

In both studied Regions, the consumption of chicken offal was widespread (86.8%). These consumption levels were high for the whole household regardless of age category.

Worldwide, the amount of edible offal available to slaughterhouses, meat processors, and wholesalers has expanded significantly over the past decades [23]. The consumption of chicken by-products has increased due to their low cost, low-fat content, and quick preparation time [24]. Indeed, chicken offal is considered an attractive source of iron and protein because it is cheaper than other animal sources like red meat and fish.

According to this survey (TABLE II), the nutritional value of broiler offal has a significant influence ($P<0.003$) on the consumption of this foodstuff. However, gender, education level, residential area, and profession have no significant influence on the consumption of chicken offal in either Province ($P> 0.05$). In Somalia, offal is traditionally consumed mainly by women, apart from liver and kidney, which are also consumed by men [25]. In South Africa, in all Districts, the frequency of liver intake (times per month) was higher in households where mothers had less than 12 years of schooling compared with households where mothers had higher years of schooling [26]. Even though more than half (61.6%) of the respondents were well aware of the potential dangers

TABLE II
Factors influencing the consumption of broiler offal

	Consumers		Non-consumers			*P*
	Freq	%	Freq	%	Total	
Gender						
Female	110	44.0%	14	5.6%	124	0.37
Male	107	42.8%	19	7.6%	126	
Total	217	86.8%	33	13.2%	250	
Residential area						
Rural	59	23.6%	9	3.6%	68	0.99
Urban	158	63.2%	24	9.6%	182	
Total	217	86.8%	33	13.2%	250	
Profession						
Civil executive	10	4.0%	3	1.2%	13	0.58
Civil servant	85	34.0%	13	5.2%	98	
Liberal function	47	18.8%	7	2.8%	54	
Retired	62	24.8%	9	3.6%	71	
Unemployed	11	4.4%	0	0.00%	11	
Undeclared profession	2	0.8%	1	0.4%	3	
Total	217	86.8%	33	13.2%	250	
Level of education						
None	3	1.2%	1	0.4%	4	0.16
Primary	15	6.0%	2	0.8%	17	
Middle	32	12.8%	1	0.4%	33	
Secondary	45	18.0%	4	1.6%	49	
University	122	48.8%	25	10.0%	147	
Total	217	86.8%	33	13.2%	250	
Assessment of nutritional value						
Yes it has	146	58.4%	71	28.4%	217	0.003
No it has not	6	2.4%	27	10.8%	33	
Total	152	60.8%	98	39.2%	250	
Awareness of the presence of ATB residues						
Aware	130	52.0%	24	9.6%	154	0.15
Not aware	87	34.8%	9	3.6%	96	
Total	217	86.8%	33	13.2%	250	

%: Percentage. Freq: Frequency

associated with antibiotic residues in offal, this knowledge did not deter them from consuming offal. These high consumption rates are probably encouraged by the low price and availability of offal compared with other animal proteins like meat.

Consumption of each type of broiler offal according to the consumer's age category

Regardless of their age category, the majority of the respondents consider the liver as their preferred offal, followed in second place by the heart. The gizzard was the least popular offal, particularly among children and older people (TABLE III).

TABLE III
Consumption of broiler offal according to the consumer's age category

	Liver		Gizzard		Heart	
	Freq	%	Freq	%	Freq	%
Children	176	70,4%	31	12,4%	49	19,6%
Adults	216	86,4%	161	64,4%	167	66,8%
Elderly	157	62,8%	61	24,4%	85	34,0%

%: Percentage. Freq: Frequency

Motivations to consume each type of offal (liver, gizzard, and heart)

Nutritional value plays an important role in offal consumption. It is the prime motivation to consume liver and the second motivation to consume both hearts and gizzards (FIG. 3). Palatability is also a determining consumption factor. It comes as the first motivation for heart and gizzard consumption and the second for liver consumption. Availability and affordability were also mentioned as motivators to consume the three types of offal.

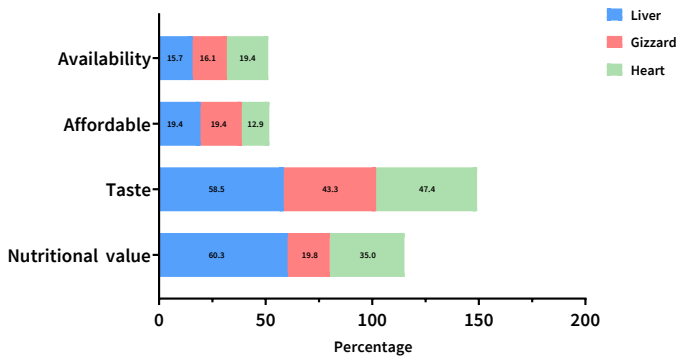

FIGURE 3. Motivations to consume each type of offal (liver, gizzard, and heart)

The liver is considered a palatable offal with known benefits for the body. In Iran, the liver is traditionally the most consumed offal and is considered a quality source of protein [19, 27]. The liver is rich in vitamins A, B, and D [28] and contains minerals and trace elements such as iron and zinc [14]. According to some studies, it may have a higher nutritional value than meat [29]. This survey showed that the liver is the most consumed offal by people of all ages, including children (70.4%). However, the use and value of offal, edible, and inedible meat by-products depend entirely on the culture and the country [23]. In some parts of the World, like Somalia, the liver is rarely provided to young children because local people believe that consuming broiler liver by children could affect their learning to talk correctly [25]. In other parts of the World, liver consumption by children is encouraged, like in the Northern Cape Province of South Africa, where 73% of preschool children eat liver at least once a month and as many as 20% once a week or more. While only 60–75 g of liver per month is needed to meet the vitamin A requirement of preschool children [26].

In this survey, the gizzard was less appreciated by children and old people (TABLE III), probably because of its organoleptic qualities. Indeed, Wani and Majeed [30] reported that gizzard is less preferred

by consumers due to their peculiar flavor and texture. The texture of chicken gizzards is tough and rubbery due to their characteristic muscular construction and a high level of connective tissue [31]. Tenderness is important for the sensory quality [32, 33]. Chicken gizzards need proper tenderization for efficient utilization [30]. According to the results obtained by Güven *et al.* in 2021 [31], the protein content of chicken gizzards is comparable to the protein content in other consumed meats (muscle tissues). Chicken and turkey gizzards are good sources of macro and trace elements, such as iron and zinc. Furthermore, the consumption of gizzards could be beneficial for human health since gizzards contain lower total saturated fatty acids (SFA) levels and higher total monounsaturated fatty acids (MUFAs) as well as polyunsaturated fatty acids (PUFAs) levels compared to muscle tissues.

Frequency of offal consumption and their culinary preparation modes

Twenty-one-point two percentage (21.2%) of the surveyed households consumed broiler offal once a week, while 12.4% consumed it several times a week. According to a study conducted in Iran in 2019, 38.1% of the respondents consume chicken liver, and most consume it once every three months [34].

Among this households, chicken offal is consumed mainly fried (73.7%) or as the main ingredient of a local sauce called Kamounia (57.1%).

Around the World, the liver is prepared and cooked in different ways with other ingredients in traditional cooking, such as fried liver and liver pate [35]. In Iran, 80.8% of consumers prefer cooking liver on a barbecue [34].

Microbiological screening of antibiotic residues in broiler offal

A microbiological screening analysis using the agar diffusion method was carried out on 66 samples of broiler offal (36 livers and 30 gizzards) to detect the antibiotic residues, in particular residues of Tetracyclines, Sulphonamides, and Aminoglycosides.

TABLE IV shows the absence of antibiotic residues in all the samples taken from public slaughterhouses. However, out of 42 samples of offal taken from private sales outlets, 52.3% were contaminated with antibiotic residues, including 12 samples of liver and 8 of gizzard. Representing respectively 33.3 and 26.7% of the total analyzed samples.

The contamination level revealed in the present study (30.3%) was lower than those recorded in other Regions of Algeria. A study on chicken liver conducted in 2017 in the Souk Ahras region revealed a contamination incidence of 70.6% [13]. In the Tizi-Ouzou Region, the study conducted by Hakem *et al.* (2013) indicated a rate of 86.2% [36], and Ramdane in 2015 reported a rate of 60% of contaminated samples in the Central Region [37].

The high percentage of positive cases is due to the abusive use of antibiotics, a characteristic of poultry farming in Algeria. Most veterinary practitioners and producers use antibiotics to prevent infectious diseases without respecting the prophylactic or therapeutic dosage, length of treatment, and withdrawal time indicated on the product label [13, 28, 38]. Internationally, contamination with veterinary drugs in foodstuffs of animal origin, particularly meat, is a real problem. Several studies in Africa revealed such contaminations: in Lubumbashi (DR Congo), Okombe *et al.* showed that out of 144 samples analyzed, 43 (36 from cattle and 7 from poultry) were contaminated with antibiotic residues, and the overall contamination rate was 29.9% [39].

Investigations conducted by Olatoye *et al.* at Akure municipal slaughterhouse in Nigeria revealed 54.4% contamination with antibiotic residues [40]. In Morocco, Chaiba *et al.* study revealed the presence of 11 livers contaminated with ATB residues out of 18 liver samples, representing a percentage of 61.1% [41]. In Asia, contamination levels are also high. In Iraq, Shareef *et al.* reported 39 positive samples out of 75 [42]. In Iran, the results obtained by Javadi *et al.* showed 100% positive cases out of 40 livers collected [43]. In Europe, studies reported lower contamination levels. In Bulgaria, for example, they recorded (15.8%) [44].

In France, the results for all species combined show that the maximum level was 1.1% in fresh meat and products of bovine (*Bos taurus*) and ovine (*Ovis aries*) species, and the minimum level was 0.08% in poultry meat [45].

The higher rates of liver contamination compared with the gizzards could be explained by the fact that many drugs are mainly metabolized by the liver and eliminated via the bile duct, leading to the persistence of these molecules in this organ. Additionally, the enterohepatic cycle followed by these medications slows down their elimination. Furthermore, many molecules have an affinity for richly vascularized organs like the liver [46].

Nature of the antibiotic residue detected in the contaminated samples

TABLE V shows the nature of the antibiotic residue detected in the contaminated samples (livers and gizzards). Nine out of 24 contaminated liver samples (37.5 %) and 8 out of 18 contaminated gizzard samples (44.4%) were suspected to contain Tetracycline residues. Liver samples were also contaminated by Sulfonamide and Aminoglycoside residues that were not detected in any gizzard sample.

In order of importance, the most incriminated antibiotic families were Tetracyclines (40.5%), followed by Sulphonamides (9.52%), while Aminoglycosides were involved in only 2.38%.

These results are consistent with those reported in Algeria by Mokhtar Rahmani *et al.* [27] and with those reported in Morroco by Chaiba *et al.*

TABLE IV
The number and the rate of broiler offal samples suspected to contain antibiotic residues

	Liver		Gizzard		Total
	Private sales outlets	State slaughterhouses	Private sales outlets	State slaughterhouses	
Number of samples contaminated	12/24	0/12	8/18	0/12	20/66
Rate	50%	0%	44.4%	0%	30.3%
Total	12/ 36 (33.3%)		8/30 (26.7%)		

TABLE V
Nature of the antibiotic residue detected in the contaminated samples (livers and gizzards)

Antibiotics	Liver	Gizzard	Rate %
Tetracyclines	9/ 24 37,5%	8/18 44,4%	17/42 40,5%
Sulfonamides	4/24 16,7%	0	4/42 9,52%
Aminoglycosides	1/24 4,16%	0	1/42 2,38%

[41] and Tassist *et al.* [47], who indicated that β-Lactams, Tetracyclines, and Macrolides were the most used antibiotics in broiler production.

CONCLUSION

This study indicated a widespread consumption of broiler offal by all households in the investigated Regions, mainly because consumers attach great nutritional value to this relatively affordable foodstuff. The liver is the preferred offal because of its palatability and high nutritional value. The study also revealed that broiler offal is contaminated with antibiotic residues, reflecting antibiotic misuse in poultry farming. Considering the quantity and frequency of offal consumption by the population, these results are alarming because of the potential risks to the consumers' health.

Conflicts of interest

The authors declare no competing interests.

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