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Importance of antibiotic residues in foodstuffs of avian origin marketed in souk ahras (Algerian republic)

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Abstract
Currently in Algeria, the use of antibiotics is frequent in poultry farming. Failure to comply with the withdrawal period, the use of these molecules as growth promoters as a preventive measure and misuse without precise diagnosis results in the presence of antibiotic residues in broiler chicken.
In order to preserve food safety, a study was conducted on the possible presence of antibiotic residues in broiler chicken meat in the willaya of Souk Ahras.
A survey among private veterinarians of the Wilaya thus a total of 15 samples was analyzed using a quantitative spectrometric method to detect residues of antimicrobial substances.
From this 100% analysis, the results of this study are a wake-up call, especially when we know the harmful effects of the presence of antibiotic residues on consumer health, particularly the selection of antibiotic-resistant bacteria on the one hand, and other allergic and carcinogenic risks on the other. This is why measures must be taken at several levels by those involved in the sector (public authorities, veterinary surgeons, technicians and farmers) to guarantee the safety of foodstuffs of avian origin.

Keywords: Antibiotic residues, Algeria, broiler meat

1. Introduction

White meat production has increased significantly in Algeria in recent years, making the price of this product reasonable and very attractive to consumers and the main source of protein for the population (Allaoui, 2011) [34]. Currently, various veterinary products are used in poultry farming, under the responsibility or not of veterinarians in order to control diseases and improve yield (Alambedji *et Al.*, 2008) [41]. Among these products, antibiotics are of prime importance.Since the 1950s, antibiotics have continued to be used to prevent and treat infectious diseases that can cause significant morbidity and be associated with mortality. The use of antibiotics have continued to be used to prevent and treat infectious diseases that can cause significant morbidity and be associated with mortality. The use of antibiotics (like any veterinary drug) is intended to keep animals healthy and contribute to their well-being. These drugs are essential tools for controlling the level of health and ensuring quality and productivity in livestock farming (Dehaumont and Moulin, 2005) [41, 42]. The use of these molecules, if justified by their remarkable efficacy in the fight against infectious diseases, must be rational. However, their use without control may lead to the formation of residues in products derived from these animals, especially when withdrawal periods are not respected by users. The potential risks associated with the presence of residues in food of animal origin are of several kinds: carcinogenic risks (Nitrofurans), allergic risks (Penicillins, Streptomycin), toxic risks (Chloramphenicol), modification of the intestinal flora (Tetracyclines). In addition, poor practices based on the use of antibiotics can select strains of multi-resistant pathogenic bacteria, which can be transmitted to humans through food. On the other hand, there is an indirect risk for meat consumers: zoonoses. In April 2010, the European Food Safety Authority (EFSA) published a dossier on these diseases, which are easily transmitted from animals to humans, and their link with the use of antibiotics. The mass or inappropriate use of antibiotics is a major cause of the emergence of resistance of certain bacteria to certain classes of antibiotics. Resistant bacteria, which grow in animals, can be transmitted to humans mainly through meat.

Escherichia coli, Salmonella, Campylobacter, Enterococcus and Staphylococcus aureus can therefore cause gastrointestinal infectious diseases that can be difficult to treat. Resistance genes carried by resistant bacteria can also be transmitted to other bacterial species and make them resistant themselves.

According to the WHO, at least 61% of pathogens affecting humans are zoonoses and three quarters of the diseases that have emerged in the last ten years are of zoonotic origin. This progressive resistance of bacteria to antibiotics is a real public health problem (Claire Peltier, 2017) [39]. Residues of veterinary drugs can compromise food safety and endanger consumer health. For this reason, a threshold has been set for each drug above which the amount of residues present in a food presents a direct danger to the consumer. This is the maximum residue limit (MRL) (Kantati, 2011) [35]. In Algeria, the curative and preventive use of antibiotics in animal husbandry is not regulated and the control of the presence of maximum residue limits (MRLs) in food of animal origin is not applied, which poses a potential risk to consumers. Few scientific studies and data on this subject are available in Algeria. This is why we have undertaken this study, the general objective of which is the detection of antibiotic residues in broiler chicken in Souk Ahras (Algeria).

2. Antibiotic residue testing in the laboratory

2.1 Field investigation

The survey involved 50 poultry farmers variously located in the wilaya of Souk Ahras.

The collected data were entered and analyzed with the software Le Sphinx Plus² version v.5.1.1.5.0 and Excel. In order to reduce the cost of veterinary services, some farmers buy drugs on the market without reliable diagnostic advice that leads to drug abuse (overdose). In this study, a survey was conducted to evaluate antibiotic use in poultry farmers, detect the presence of antimicrobial residues, particularly tetracycline and erythromycin in wishbone muscles and poultry liver.

2.2 Antibiotic residue testing in the laboratory

15 samples of chickens were randomly collected from different butcher shops in the commune of M' daourouche. These samples were brought to our laboratory where the analysis of the wishbone muscles was done on 9 samples among them and the liver analysis was done on 6 samples. The standard bacteriological laboratory equipment and reagents required (Acetonitrile.2-methyl-2-propanol, Hydrochloric acid, Methylene chloride and petroleum ether) were used for the implementation of the standardized microbiological method for residue detection, known as the "turbidimetric method". The principle of this method is that the optical density of a suspension is proportional to the mass of the suspended particles, the light absorption of the suspension is measured and the biomass concentration is deduced from a reference range (LAPRENT GOURAUD S., 1992).

3. Resultant's Discussion

3.1 Field investigation

Antibiotic use without symptom on chickens

Our results show that 70% of poultry farmers use antibiotics without symptom on chickens... Our results are similar to those obtained by Albayoumi *et al* (2015) [25] n Gaza who found that 68% of farmers use antibiotics without symptom on chickens.

Table 1: Antibiotic use without symptom onset on

Use of ATBs without symptom appearance	Number	Percentage
Yes	35	70%
No	15	30%
Total	50	100%

3.2 Overdose

34% of farmers use a double dose to speed up the healing of chickens. Our result is close to those of Sinaly (2014) who found that 48.59% of the treatments performed were overdosed. The observed overdose may be related to the choice of dosage expressed in millilitres or milligrams of drug per litre of water rather than in milligrams of drug per kilogram of body weight.

On the other hand, they are lower than those obtained in 2015 by Albayoumi et al who found that 54.5% of farmers used a double dose to accelerate healing. The consequences of this practice are diverse. They can cause toxicity to the animal or generate residues in the products of treated animals. As well as the increase in the emergence of antibiotic-resistant bacteria.

Table 2: overdose and accelerated healing

Overdose	Number	Percentage
Yes	17	34%
No	33	66%
Total	50	100%

3.3 Sensitivity of poultry farmers to the adverse effect on human health of the presence of antibiotic residues in broiler chicken meat

During our study, 72% of the farmers surveyed confirmed that there is a risk to human health associated with the unreasonable use of drugs (antibiotics). On the other hand, 28% of farmers say that there is no risk to human health. The lack of awareness of the risks may be due to the ignorance of these respondents, or to a lack of awareness. Based on the results obtained, we can say that the majority of farmers are sensitized. Our results are very close to those of Albayoumi et al (2015) who found that 84.4% of farmers sensitive to the harmful effect of antibiotics in the chicken body on human health.

Table 3: Sensitivity of poultry farmers to the adverse effect of antibiotics on public health

Sensitivity of poultry farmers	Number	Percentage
Yes	36	72%
No	14	28%
Total	50	100%

3.4 Adherence to waiting times by breeders

70% of the farmers surveyed say they respect the waiting period, with 64% of farmers stopping giving drugs before marketing more than 2 days. Our result is similar to that of BADA-ALAMBEDJI et al (2004) [18] who found that 70.7% of breeders respect the waiting time. Another survey conducted by Tobi (2004) [17] in Dakar shows that 84.62% of farmers respect the waiting time. Non-compliance with waiting times by farmers was observed in 30% of respondents. This could be explained by a lack of responsibility and awareness among farmers. Antibiotics, if their withdrawal period is not respected, can leave residues in food of animal origin that are dangerous for the consumer and capable of causing hypersensitivity accidents or poisonings, but also the selection of bacteria resistant to subsequent treatments.

Table 4: Poultry Producers' Waiting Time Compliance

Waiting Time	Number	Percentage
Yes	35	70%
No	9	18%
Sometimes	6	12%
Total	50	100%

3.5 Consultation with a veterinarian to prescribe drugs

The majority of poultry farmers follow the prescribed treatments with veterinarians, especially if we consider that in 70% of farmers consult their veterinarians, while 26% of farmers consult their veterinarians only if the treatments prescribed do not give favourable results. However, our result is different from that of Messai (2006) who found that 94% of respondents said that their clients only come back to them if the treatment, already implemented, does not produce clinical results (persistence of symptoms) and 39.39% said they come back, even if the treatment is effective.

Table 5: Consultation with a veterinarian to prescribe drugs

Consultation	Number	Percentage
Always	35	70%
Sometimes	13	26%
Never	2	4%
Total	50	100%

3.6 Objective of antibiotic use

74% of poultry farmers use antibiotics as a therapeutic treatment to cure clinically ill animals and avoid mortality. The treatment also reduces bacterial excretion, in some cases leading to bacteriological healing.

However, 34% use antibiotics as a preventive measure. This type of antibiotic therapy is based on the principle of prescribing antibiotic treatment before infection occurs in subjects in a pathological situation that exposes them to a significant risk of infection. It can be implemented during certain so-called risk periods, when the probability of developing an infection is high; start-up period when general hygiene conditions are poor or, in cases where post-vaccination reactions are relatively severe. But it has a major disadvantage (due to the wide use of antibiotics it causes, it becomes an essential cause of the development of bacterial resistance).

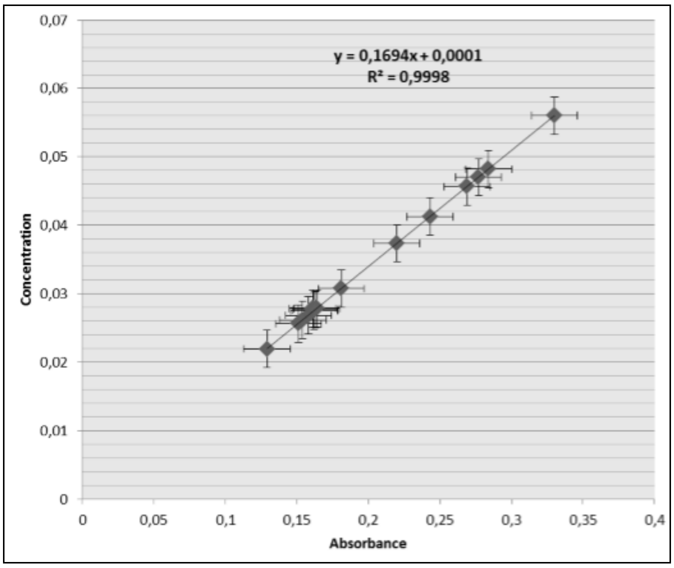

Fig 1: Tetracycline calibration curve

3.7 Detection of antibiotic residues by the turbidimetric method

Table 7: Measured absorbances and concentrations obtained for samples analysed for the presence of tetracycline

Samples	Absorbance A	Transmission (%)	Concentration
Wishbone 1	0,158	69,5	0,02682513
Wishbone 2	0,161	69,1	0,0278438
Wishbone 3	0,164	68,5	0,0278438
Wishbone 4	0,181	65,9	0,03073005
Wishbone 5	0,151	70,6	0,02563667
Wishbone 6	0,162	68,9	0,02750424
Wishbone 7	0,154	70,1	0,02614601
Wishbone 8	0,220	60,3	0,03735144
Wishbone 9	0,129	74,3	0,02190153
Liver 1	0,277	52,9	0,04702886
Liver 2	0,330	46,8	1,02167183
Liver 3	0,269	53,8	0,04567063
Liver 4	0,163	68,8	0,02767402
Liver 5	0,284	52,0	0,04821732
Liver 6	0,243	57,2	0,04125637

The table shows the presence of the antibiotic (tetracycline) in all samples analysed at a concentration ranging from 216 to 482ng/g with a level exceeding the maximum residue limits (100ng/g) for wishbone muscle and (300ng/g) for the liver.

Table 8: Measured absorbances and concentrations obtained for samples analyzed for erythromycin

Echantillons	Absorbance	Transmission (%)	Concentration
Wishbone 1	0,201	71,8	0,03412564
Wishbone 2	0,159	62,9	0,02190153
Wishbone 3	0,129	69,4	0,02190153
Wishbone 4	0,157	74,4	0,02665535
Wishbone5	0,146	69,6	0,02478778
Wishbone 6	0,137	71,4	0,02325976
Wishbone 7	0,246	73,0	0,0417657
Wishbone 8	0,143	56,8	0,02427844
Wishbone 9	0,120	72,0	0,02037351
Liver 1	0,231	58,8	0,03921902
Liver 2	0,176	66,8	0,54489164
Liver 3	0,245	56,8	0,04159593
Liver 4	0,166	68,2	0,02818336
Liver 5	0,169	67,8	0,0286927
Liver 6	0,210	61,7	0,03565365

Under our experimental conditions, erythromycin was detected at a concentration above the maximum residue limit (MRL) of 200ng/g for wishbone muscle and poultry liver in all samples analysed.

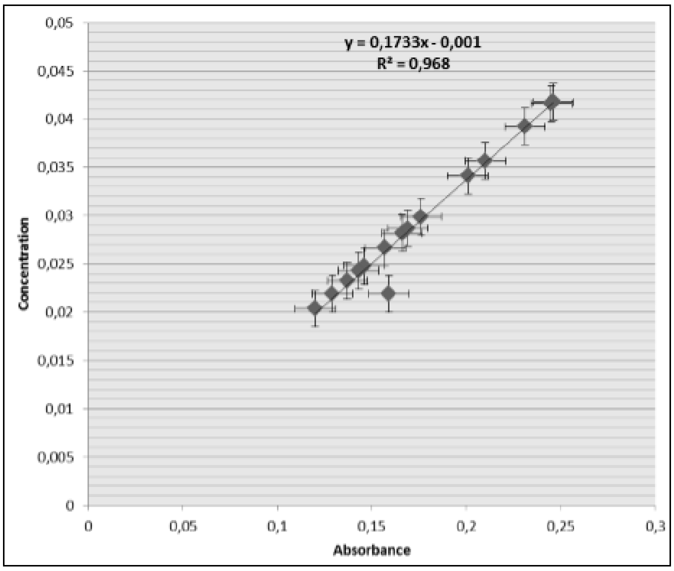

Fig 2: Erythromycin calibration curve

4. Discussion

The results obtained from the present study on the detection of antibiotic residues in broiler meat in the municipality of M'daourouch show that of the 15 chicken samples tested, all are positive, i.e. a percentage of 100%. Our results revealed the presence of tetracycline in all samples analyzed at a concentration ranging from 216 to 482 ng/g with a level exceeding the maximum residue limits. This is in accordance with the result found in Tizi Ouzou by Ramdane (2015) [16], who reported that all samples analysed are positive for the presence of oxy tetracycline residues with a concentration above the MRL. This result is comparable to that observed in Congo by OKOMBE (2016) [27] where traces of tetracycline with a rate of 100% were found.In a study in Pakistan, tetracyclines were detected in 29% of broiler samples after HPLC confirmation, six out of 89 were above the MRLs. Using the HPLC technique, 60% of Wishbone muscles were found positive for the presence of tetracycline residues in Iran. The concentration of tetracycline in ten percent of the samples was significantly higher than the MRLs (100 μg / kg). AL-GHAMDI *et al* (2000), in searching for tetracycline residues in poultry products in the eastern province of Saudi Arabia, found 69.7% antibiotic residues in broilers.A lower result than that detected in this study was recorded in Kuwait. Using the Charm II test to investigate the presence of tetracycline residues in 263 parts of chicken, 12 (5%) of the samples contained tetracyclines. All detected samples were confirmed by LC / MS / MS / MS and with a rate exceeding the maximum residue limits. Another result lower than that obtained in our study is that obtained by ALBAYOUMI (2015) in Gaza, which showed the presence of 43.15% tetracycline. Another study carried out in Senegal by ABIOLA (2005) [20] found a rate of 14% of tetracycline residues, the levels obtained ranged from 77 to 376 μg/kg, therefore with concentrations sometimes above the MRL which is 100 μg/kg. As for macrolides, A study carried out in Brazil (Nonaka, 2009) revealed that 212 samples were positive for the presence of antibiotic residues (macrolide and aminoglycoside), i.e. a rate of 21.9%, after confirmation by liquid chromatography, 45% mass spectrometry were macrolides, but none of these residues exceeded the established MRLs. Our results are higher than those obtained by BEN MOHAND (2008) [15] in Algiers, which found a percentage of positivity of 6.66% of beta lactam (penicillin G) and macrolides (erythromycin). In Europe, prevalence rates of contamination of food of animal origin with drug residues are less than 1% (Serratosa et al. 2006) [11]. In Africa, updated studies on the presence of antibiotic residues in food of animal origin are very limited. For example, a study on antibiotic residues in chicken meat and offal in Dakar, Senegal, showed the presence of residues of prohibited substances such as nitrofuran and chloramphenicol in the different matrices (Abiola et al. 2005) [20]. In Ghana, the prevalence rates of antibiotic residues are 30.8% for beef, 29.3% for kid meat, 28.6% for pork, 24% for sheep meat and 6.8% for eggs (Donkor *et al*. 2011) [1]. In Nigeria, antibiotic residue prevalence rates are 0.1% to 1% for eggs (Idowu *et al*. 2010 [4], {Kabir, 2004 #500), 23.6% for laying hens, 4.8% for local breed chickens and 21.8% for chicken droppings {Kabir, 2004 #500}. Higher rates of 33.1% have been reported in Nigeria for broilers (Kabir *et al*. 2004) [5], 52% in gizzards and 81% in chicken livers in Senegal (Abiola *et al*. 2005) [20], Kenya (Kang'ethe *et al*. 2005) and Tanzania (Kurwijila *et al*. 2006) [7].

Risks related to the consumption of food contaminated with antibiotic residues: The prevalence rate of veterinary drug residues in food of animal origin is less than 1% in Europe, while it reaches 94% in some African countries. The presence of antibiotic residues in food of animal origin can cause allergies, cancers, changes in the intestinal flora, bacterial resistance (Mensah *et al*. 2014) [9] Tetracycline in meat can potentially stain young children's teeth. Penicillin in chicken is reported to have caused a severe anaphylactic reaction in a consumer (Teh and Rigg 1992) [12]. Skin allergies in eggs containing sulphonamide residues have also been reported. In addition to these health risks, the presence of veterinary drug residues in food of animal origin is a significant economic barrier to international trade with the new WHO health rules.

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