

# Preliminary Research Study for Antibiotics in Eggs Produced and Sold in Bamako, Mali

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## Abstract

The quality of agri-food products, in particular those of poultry origin, has now become an imperative. Because of the antibiotics used in poultry production, we are seeing the presence of antibiotic residues in products from poultry farming, especially eggs. Residues in eggs do not always comply with regulatory requirements, which can have harmful consequences for consumers such as: risk of allergies or risk of antibiotic resistance. This study was carried out with the aim of evaluating the presence of residues of veterinary antibiotics in eggs produced and marketed in the district of Bamako and surroundings. All samples were analyzed using the Premi "Test which is a rapid detection kit for antibiotic residues in different materials. We collected 900 eggs from 10 farms and 10 markets in Bamako. Out of 900 eggs, 228 were tested. One hundred and eighty-nine (189) eggs tested negative for the presence of antibiotic residues, *i.e.* 82.9%; on the other hand, 39 were positive, *i.e.* 17.1%. This study clearly shows the presence of antibiotic residues in eggs from farms and various markets in Bamako and the surrounding area. This will allow us to continue our study at a later date to identify the types of antibiotic and to dose them.

# **Keywords**

Antibiotic Residues, Eggs, Mali

# **1. Introduction**

Meeting the food needs of a growing population is becoming an increasingly

acute problem. Animal husbandry has therefore been modernized to provide the appropriate quantities of food to this population. The use of veterinary drugs, especially anti-infective drugs in modern farming has contributed to the increase in production [1].

Antibiotics therefore have an important place in modern animal husbandry today. They are administered to farm animals, either by injection or via feed, and pass into the muscles, kidneys, liver and in the case of laying hens, into the eggs [2]. These antibiotics are used as drugs or as growth promoters. These generate residues for a variable duration. The presence of these non-standard residues can have an impact on consumer health and can also contribute to antibiotic resistance in humans.

The prevalence of drug residues in foods of animal origin is less than 1% in Europe, and reaches 94% in some African countries [1].

In Africa, updated studies on the presence of antibiotic residues in foods of animal origin are very limited [3] [4].

With regard to residues of veterinary drugs in foods, WAEMU does not yet have a functional system for researching drug residues in foods of animal origin [5].

However, whatever the nature of the antibiotic administered, there is a risk of finding residues in tissues (meat) and excretion products (milk, eggs). It is for this reason that a threshold has been set for each drug beyond which the quantity of residues present in a food presents a direct danger for the consumer: this is the maximum residue limit (MRL) [6]. In Mali, the curative and preventive use of antibiotics in livestock farming is not well regulated and the control of the presence of maximum residue limits (MRLs) in foodstuffs of animal origin is not applied, which poses a potential risk to consumers. The importance of this problem and the lack of scientific documentation on experimental work dealing with this subject in our country led us to take an interest in this theme. Our objective is to look for the presence of antibiotic residues in eggs produced and marketed in Bamako. This study will therefore be used to determine the prevalence of antibiotic residues in eggs.

#### 2. Materials and Methods

#### 2.1. Place and Period of Study

The study was carried out over a period of 4 months in the district of Bamako due to the concentration of poultry farms from February 01, 2020 to May 31, 2020. It was carried out in two phases: the first consisted of making egg samples; and the second to the analysis of samples in the laboratory.

#### 2.2. Survey Materials

A survey sheet has been developed to collect the information necessary for a better understanding of our theme: Identification of the farm, Characterization of the different antibiotics.

### 2.3. Sample Collection and Storage Equipment

To take and store the samples, we used as equipment:

- A cooler and cold accumulators;
- Sampling sheets;
- A permanent marker;
- A refrigerator.

## 2.4. Sampling

The sampling steps are:

- We took the list of farms with FIFAM;
- We divided the farms into zones and made a draw of the farms to visit;
- Calculate sample size;
- Perform weighted sampling per farm.

#### 2.5. Laboratory Equipment, Reagents

The material used in the laboratory consisted of glassware and apparatus:

- Glassware: 5 ml hemolysis tube; Beakers;
- Equipment: Oven brand memmmert model 30 1060.
  - Reagents:

Screening methods can be qualitative or quantitative. A qualitative method allows the detection of the presence of an analyte and gives a result in the form of presence/absence. A quantitative method quantifies the analyte present and gives a result in the form of analyte concentration. We used Le Premi \*Test, which is a qualitative method.

The Premi <sup>®</sup>Test: For the detection of residues:

It is a test based on the inhibition of the growth of Bacillus stearothermophilus, a bacterium very sensitive to many antibiotics. Standardized spores are embedded in agar supplemented with selected nutrients. Covering a wide range of antibiotics, the Premi \*Test is a rapid, sensitive, reliable, ready-to-use test. Popelka *et al.*, (2005) revealed, during method validation studies on poultry, that the detection limits of the Premi Test are equal to or higher than the MRLs for most antibiotics (Macrolides, Tetracyclines, Sulfonamides), with the lowest detection limits for  $\beta$ -lactams [7]. Since 2006, it has been recognized as an official method in many countries such as France and has been validated by the AgenceFrançaise de Normalization [8].

The sample is added to the premi-test vial and incubated at 64° the spores will germinate. These germinated spores will multiply and acidify the medium in the absence of inhibiting substances. This will result in a color change of the indicator from purple to yellow. If the antimicrobial residues are present in sufficient quantity (above the detection threshold) the germ will not develop and the color will remain purple.

#### 3. Results

We have divided the farms into 7 zones according to the geographical location.

On each axis we visited at least 2 farms. During the study period, we took samples from 10 farms and the rest from a few markets in Bamako. We collected 900 eggs, including 600 from farms and 300 from markets (**Table 1**). We worked on a total of 228 samples, *i.e.* 25.3% of the eggs collected. The samples were tested in 2 batches. We carried out a screening test to see the presence or absence of residues in the eggs (**Figure 1**). Of 228 eggs analyzed, 39 were detected positive for antibiotic residues, *i.e.* 17.1%, and 189 were negative, *i.e.* 82.9% (**Figure 2**).

We detected the presence of antibiotics in 30 cases, *i.e.* a percentage of 18.9% of the 158 eggs analyzed from the farms. Out of 70 eggs analyzed from the markets, the test was positive in 09 cases, *i.e.* 12.9%. The overall results are shown in **Table 2**.

We submitted a questionnaire to the farms and we found that all the poultry farmers had notions about waiting times. Non-compliance with these waiting times before placing on the market after treatment was observed in 84% of poultry farms. Antibiotics are used in prevention, treatment and also growth factor in cases. We observed the presence of no regulatory authority.

Table 1. Number of eggs according to the collection site.

Sampling point	Number of eggs collected	Percentage
Farms	600	66.7
Markets	300	33.3
Total	900	100

Table 2. Overall results of the tests according to the sampling site.

Sampling point	Analyzed	Positive test	Percentage	Negative test	Percentage
Farms	158	30	18.9	128	81.1
Markets	70	9	12.9	61	87.1
Total	228	39	17.1	189	82.9



Figure 1. "Premi "Test" staining ranges. Source: National health laboratory of Bamako.

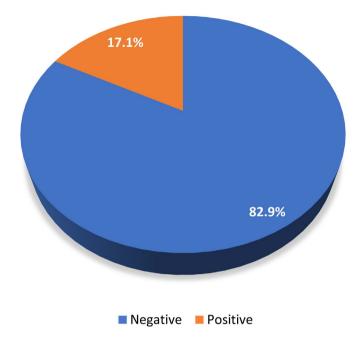


Figure 2. Results of the search for residues.

## 4. Discussion

The egg can be defined as a low-energy source of perfectly balanced proteins and very easily digestible lipids, also providing 20% to 30% of man's daily need for many minerals and vitamins (per 100 g of egg) [9].

For humans, the egg is the reference protein source because it contains many molecules of a protein nature with various biological activities. The egg is a widely consumed food, the study of the residues it may contain is not completely meaningless. It must even question the waiting times practiced, in order to offer healthy and safe eggs to consumers. It turns out that poultry is among the top three species most exposed to antibiotics (along with rabbits and pigs) [10]. Screening is a critical and essential step, which must detect antibiotic residues as well as possible at regulatory limits (e.g. MRLs) to avoid the presence of residues in food.

Within WAEMU, in the collection of references for harmonized methods of analysis of food products, methods of analysis of residues of veterinary drugs are not taken into account. This results in differences between the analysis methods from one country to another and even from one laboratory to another [1]. For our study, we used the Premi \*Test, which is based on the inhibition of spore growth by antibiotics. We chose it because it is a simple and economical test for the qualitative screening of a broad spectrum of antibiotics.

Of 228 eggs analyzed, 39, or 17.1%, were detected positive for antibiotic residues. In Africa, some investigations on the presence of residues have been carried out using different methods from one study to another. For example, FAGBAMILA *et al.* [11] in Nigeria reported that out of 900 egg samples analyzed using the disk diffusion test, 32 (3.6%) contained antibiotic residues and 18 eggs (2%) gave positive results with the Premi \*Test method. In Senegal, the presence of antibiotic residues was qualitatively confirmed in eggs at 12% [12].

A study conducted in Ivory Coast showed the presence of residues in almost 26% of table egg samples tested [12].

Although the results of these studies are somewhat different, they clearly show that the problem of antibiotic residues in food remains a concern for consumer health on the continent.

The potential risks linked to the presence of residues in foodstuffs of animal origin are of several types: carcinogenic risks (Nitrofurans), allergic risks (Penicillins, Streptomycin), toxic risks (Chloramphenicol), modification of the intestinal flora (Tetracyclines), selection of bacteria resistant to antibiotics (several antibiotics are concerned) [13].

In our study, 84% of poultry farmers did not respect the waiting period. It is therefore important to pay particular attention to the use of antibiotics, in particular respecting the withdrawal period.

To better limit the dangers that antibiotic residues can pose to egg consumers, it is necessary to draw up standards and regulations in terms of residues (rational use of antibiotics, knowledge of authorized and prohibited drugs, and control of the withdrawal period).

This study needs to be continued to identify the types of antibiotics and their content likely to be harmful for human consumption.

## **5.** Conclusions

The development of resistance to antibiotics is a global problem that must require research, monitoring and education actions aimed at developing prudent use of this essential therapeutic class for doctors and veterinarians. Veterinary medicinal products should be used in the context of infection control on prescription from a veterinarian and should in no way be a palliative.

The results of this study cannot represent the general situation. They are, however, alarming and reflect the poor use of antibiotics in animal husbandry. This misuse results in the presence of residues in the eggs.

We can say that breeders as well as veterinarians have some knowledge about dose limits and withdrawal times. However, for economic reasons, eggs produced during antibiotic treatment end up on the market and ultimately on our plates.

The method used to obtain these results is qualitative. Subsequent studies with more advanced quantitative methods should however be carried out to clearly identify the nature of the antibiotic(s) present in these foodstuffs, as well as the exact levels.

We all have an interest in monitoring animal health because human health is closely linked to it.

# **Conflicts of Interest**

The authors declare no conflicts of interest regarding the publication of this paper.

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