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Antibiotic resistance of *Escherichia coli* pathogenic for poultry

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ABSTRACT

The Regional Veterinary Laboratory Center of Tlemcen, in collaboration with veterinary centers of Oran monitor together the network of epidemiological surveillance of antibiotic resistance among key bacterial pathogens in poultry. This monitoring is based on the collection and processing of results of susceptibility tests routinely performed by veterinary laboratories network members. Regarding the poultry industry, 2615 results of susceptibility testing were recorded in 2002 nearly 80% were performed for strains of *Escherichia coli*. The percentages of strains susceptible to tetracycline, amoxicillin and the association sulfonamides/trimethoprim were respectively 16.1%, 45.6% and 55.9%. The percentages of susceptible strains for the remaining 13 molecules studied vary between 71.3% and 99.7%. Serovar O78K80 represents 18.7% of *E. coli* that were the subject of sensitivity. The percentages of susceptible strains belonging to this serovar are statistically lower than those of serovars O1 and O2K1K1 to 9 antibiotics among 16 studied. The center veterinarian can monitor the proportion of bacteria that have acquired one or more resistance mechanisms the different anti-infective used in animal husbandry and detect the possible emergence of new resistant phenotypes.

Key words : *Escherichia coli*, poultry, antibiotic resistance, serotype, serovar, Tlemcen.

INTRODUCTION

In many countries, the evolution of bacterial resistance to antibiotics has been monitored since many years, both in human and veterinary medicine (Stelling and O'Brien, 1997, Wray and Gnanou, 2000, Sanders, 2001). The regional veterinary center of Tlemcen has established a surveillance network for antimicrobial resistance of major bacterial pathogens isolated in poultry.

MATERIALS AND METHODS

The bacteria are isolated from viscera of diseased animals by veterinary laboratories. It is essentially *Escherichia coli*, *Salmonella*, *Staphylococcus*, *Streptococcus* and *Haemophilus*. When susceptibility testing is performed, laboratories send the results to members of the network coordinators (Neuman, 1990), Data associated with bacteria and antibiograms (date and nature of sampling, pathology observed) are also recorded. Antimicrobial drug susceptibility testing was done by the Kirby-Bauer disk diffusion method (Barry, Thornsberry, 1985) on Iso-

Sensitest agar (Oxoid Canada, Nepean, Ontario), which compares well with Mueller-Hinton agar for testing *Enterobacteriaceae* (Flandrois, Peyret M, Zindel J, 1991). *Escherichia coli* strain ATCC 25922 (American Type Culture Collection, Manassas, Virginia, USA), which gave reproducible growth inhibition zones, was used as a susceptible control throughout (Feillou, Martel, 1996, Bateman, 2000). The results of susceptibility testing are diameters of inhibition zones whose size is inversely proportional to the concentration (MIC) of antibiotic against bacteria (Figure 1).

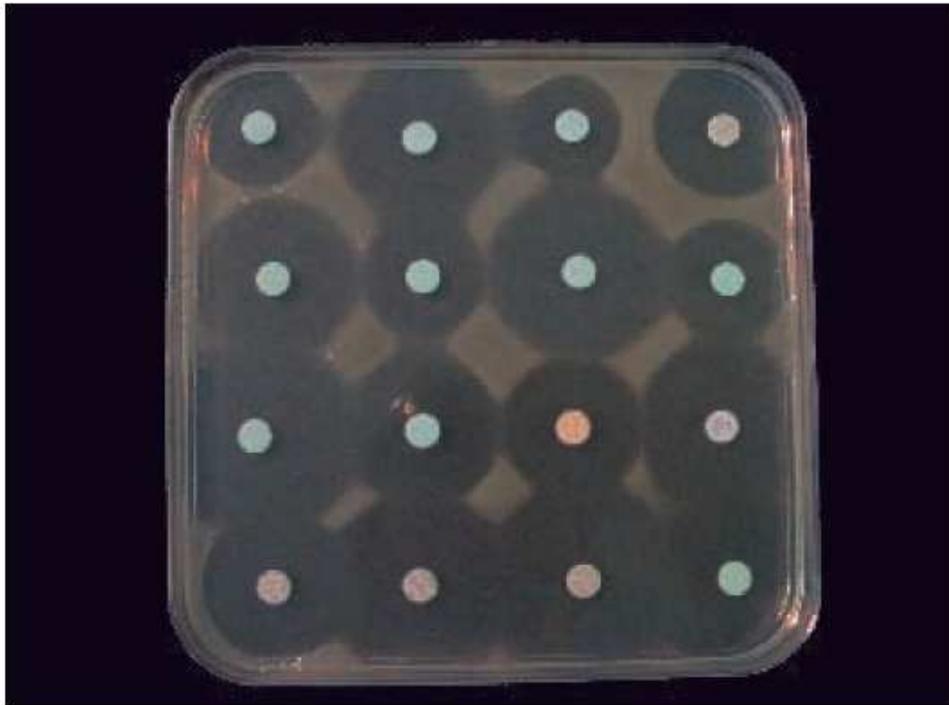


Figure 1: Susceptibility test results of the reference strain *E. coli* ATCC 25922 regarding 16 antibiotics (Diffusion technique in agar medium)

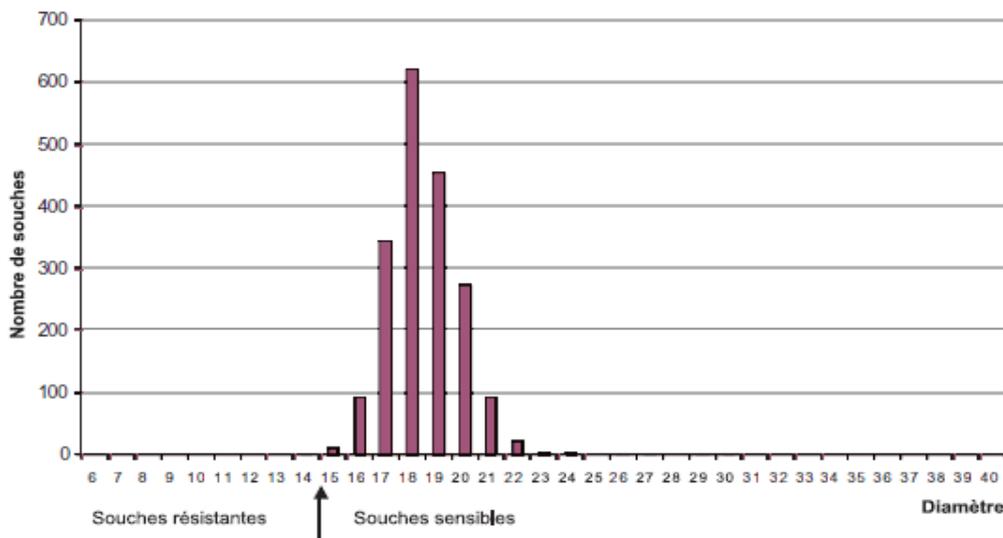


Figure 2 : Distribution example: *E. coli* resistivity/ sensibility against colistine (1934 strains)

The sensitivity is subject to internal quality controls according to the standards and compared to reference strains ATCC. The antibiotics used in susceptibility correspond to families of anti-infectives prescribed in veterinary medicine (Gyles, Thoen, 1986, Prescott, 2000, Sanders, 2001, Stelling, 1997).

The sensitivity of a bacterial population against an antibiotic is therefore characterized by the frequency of different diameters whose distribution inhibition (Figure 2) allows to highlight the possible existence of a subpopulation (s) resistant (s). The critical diameters therapeutic, determined in human medicine for each antibiotic (Wray, Gnanou, 2000), can be used so epidemiological veterinary medicine if they also separate sub-populations of bacteria of animal origin. For molecules with specific veterinary critical diameter are indicated by the manufacturing laboratories.

RESULTS AND DISCUSSION

The majority of susceptibility (67%) was achieved for bacteria isolated from deep organs (septicemia). (Figure 3) shows the different bacteria isolated from poultry that have been submitted to an antibiogram in 2002. Nearly 80% of bacteria whose susceptibility results are *E. coli*. Serovars sought in poultry are O78K80, and O2K1 O1K1. They represent respectively 18.7%, 11.5% and 1.5% of all bacteria that have been subject of sensitivity.

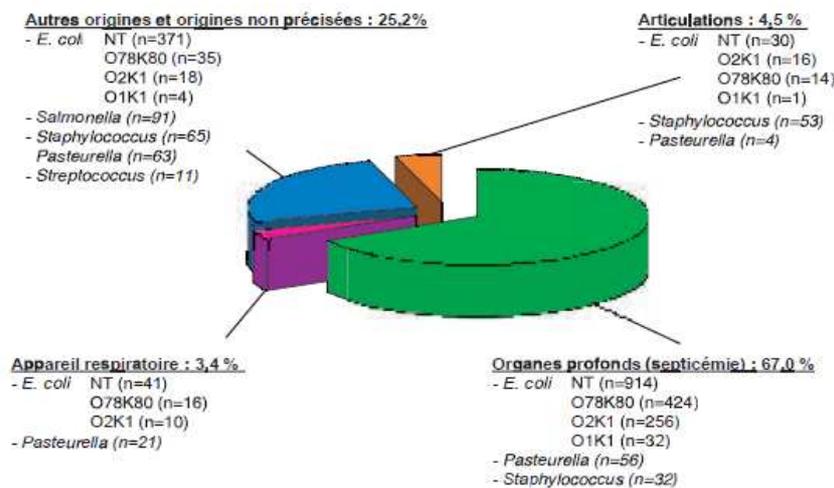


Figure 3 : Origin and bacterial identification

Table 1 shows the results of antibiotic susceptibility of *E. coli* isolated from poultry in 2002 all samples combined regarding 16 molecules commonly used in susceptibility testing. The percentages of strains that have acquired no resistance mechanism against tetracycline, amoxicillin and the association sulfonamide / trimethoprim were respectively 16.1%, 45.6% and 55.9%. The percentages of the highest sensitivity (> 85%) are obtained with fluoroquinolones (enrofloxacin and difloxacin), aminoglycosides, cephalosporins, florfenicol and colistin. The percentages of susceptible strains belonging to serotype O78 K80 are statistically lower than those of serovars O1 and O2 K1 K1 for nine of the 16 molecules studied.

With the exception of four molecules (ceftiofur, florfenicol, enrofloxacin and difloxacin), the number of susceptible strains was determined with critical diameters established to human medicine. The therapeutic impact of these percentages in veterinary medicine is limited. However, these figures allow to show the proportion of bacteria that have acquired one or more resistance mechanisms. The lab can follow the evolution of these percentages over time and their variation for different bacteria and different serovars of the same bacterium.

The new resistance phenotypes to be identified early possible in order to effectively limit their distribution (food chain, environment) and to preserve the therapeutic action of antibiotics used in animal husbandry (CA-SFM, 2007). On *E. coli*, the existence of rare animal strains resistant to colistin and ceftiofur could be confirmed. It is now more specifically to study the mechanisms of resistance of these strains and closely monitor their numbers in all *E. coli*.

Coordinators in collaboration with the heads of laboratories are also working to improve the standardization of susceptibility testing techniques. Indeed, the reliability of the surveillance of antibiotic resistance and therapeutic success in farming depends on the quality of the results of susceptibility testing and interpretation (Michel-B., 1986).

Table 1: Autopsy findings of animals, number of chickens on six autopsies with lesional index (Nb* index)

Antibiotiques	E. coli Total		E. coli 078 K80		E. coli 02 K1 et 01 K1		Test Chi-2 Différence significative (5 %) *
	n	% S	n	% S	n	% S	
Amoxicilline	2212	45,6	501	36,5	348	60,6	oui
Amox. + clav.**	307	71,3	76	56,6	65	70,8	non
Céfalexine	180	97,2	30	100,0	35	97,1	non
Ceftiofur	1923	99,4	434	100,0	330	99,4	non
Néomycine	1594	89,7	425	78,8	325	98,2	oui
Gentamicine	1926	93,5	434	94,5	329	97,0	non
Spectinomycine	1524	87,1	411	86,9	308	90,3	non
Florfenicol	574	99,7	66	100,0	65	100,0	non
Tétracycline	2149	16,1	469	6,6	354	28,8	oui
Doxycycline	1445	14,3	304	5,9	230	25,2	oui
Colistine	1934	99,5	438	99,8	332	99,7	non
Sulfa. + trim.***	2225	55,9	498	53,8	354	67,2	oui
Fluméquine	2216	70,9	500	58,8	349	80,2	oui
Acide oxolinique	1927	76,0	438	67,8	325	81,8	oui
Enrofloxacin	1273	87,1	301	85,4	210	92,3	oui
Difloxacin	1273	88,0	301	85,0	210	92,3	oui

n = nombre de souches testées % S = pourcentage de souches sensibles

* Test du Chi-2 réalisé entre le sérovar 078 K80 et les sérovars 02 K1 et 01 K1

** Amoxicilline + acide clavulanique *** Sulfamides + triméthoprime

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