



Antibiotic Residues in Meat and Eggs in Taiwan: A Local Surveillance

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Authors' contributions

This work was carried out in collaboration between all authors. Authors JLW and SCY designed the study, wrote the protocol and wrote the first draft of the manuscript. Authors SCY, MCY and YHL managed the experimental process. All authors read and approved the final manuscript.

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ABSTRACT

Aims: To estimate the prevalence and identify risk factors of antibiotic residues in meats and eggs in Taiwan through a cross-sectional study.

Methodology: From 1 Jan 2013 to 31 Dec 2014, we collected 175 samples from supermarkets (eggs, $n = 50$; chicken meat, $n = 50$; pork meat, $n = 50$; and fish meat, $n = 25$) and 175 samples from wet markets (eggs, $n = 50$; chicken meat, $n = 50$; pork meat, $n = 50$; and fish meat, $n = 25$) in Taiwan. Most samples were purchased in southern Taiwan.

Results: In total, 350 samples were collected, and 8% were determined by the Premi@Test to contain antibiotic residues. Univariate analysis showed that the meat and eggs purchased from wet markets were more likely to have antibiotic residues than those purchased from supermarkets (12% vs. 4%, $P = .006$). Multivariate analysis showed that the suburban locale (odds ratio: 4.59; 95% confidence interval: 1.21–17.42) and the wet market type (odds ratio: 3.51; 95% confidence interval: 1.43–8.62) were two independent risk factors for meats and eggs to be positive for antibiotic residues.

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Conclusion: The 2013–14 meat and egg surveillance for antibiotic residues found 8% rate of positive samples. The suburban locale and wet market type were two independent risk factors for antibiotic residues to be detected in meats and eggs in Southern Taiwan. Further surveillance of antibiotic residues in meats and eggs in these high risk areas may be necessary.

Keywords: Antibiotic residues; poultry products; meat; market; Taiwan.

1. INTRODUCTION

The emergence of antibiotic-resistant bacteria in humans may be linked to antimicrobial use in food animals [1]. Antimicrobials are used in animals for disease treatment, prevention and control and for growth promotion/feed efficiency [1]. The main factors related to the presence of antibiotic residues in food animals include animal's age, use and failure to comply with withdrawal times for regular and off-label uses of antibiotics [2]. The withdrawal time is the number of days required for the drug to be fully eliminated from the animal's body. Several methods are available, such as liquid chromatography–mass spectrometry and liquid chromatography–tandem mass spectrometry, for the detection of antibiotic residues and could be used for a national surveillance program [3–6]. For national campaigns, screening can be performed by quicker, easier and cheaper tests. Four-plate test, Nouws antibiotic test, Fast, Cast, Stop and Delvotest are widely used by national surveillance programmes [7]. The Premi®Test is a microbial screening test for the detection of antibiotic residues in food. It is based on the growth inhibition of *Bacillus stearothermophilus*, a thermophilic bacterium that is highly sensitive to many antibiotics and sulpha compounds [8,9]. Assay results are available within 4 h, and the use of spores instead of vegetative cells allows prolonged shelf life of the kit, making its commercial distribution feasible [10].

Antibiotic residues present in animal food products at levels above the legal limit clearly have an impact on human health, and thus reducing antibiotic residues in foods is important [4]. For example, antibiotic residues in foods of animal origin not only cause concerns about their direct toxicity to humans but also concerns that low levels of antibiotic exposure may result in alterations of microflora in humans, leading to the development of antibiotic-resistant strains [11]. Antibiotics may also cause increased incidence of resistant *Helicobacter pylori* in the mucosa of stomach. By this several diseases such as peptic ulcer, gastric cancer and Dieulafoy lesion occurs

[12,13]. Different prevalence rates of multidrug-resistant bacteria have been found in raw meat from different animal species [14], and bacterial contamination rates also differ between supermarkets and wet markets [15]. Netherlands surveillance found that less than 5% of food samples had antimicrobial residue contamination using fast screening method [16].

It is unknown if the prevalence of antimicrobial residues differs among foods from different species of animals or between supermarkets and wet markets in Taiwan. In this study, we used the Premi test to screen for antibiotic residues in meats and eggs and to determine if there are risk factors for the presence of antibiotic residues in foods.

2. METHODOLOGY

This study complied with the World Medical Association Declaration of Helsinki on medical research protocols and ethics. We collected meat (pork, chicken and fish) samples and eggs in 2013 and 2014 from different markets, with a 1:1 ratio of samples from supermarkets and wet markets. We also recorded the date and place of the purchase for all meat and egg samples. Most of the supermarkets were in urban areas. After collecting the meats and eggs, we performed the Premi test to test for antibiotic residues following the manufacturer's protocol. We collect data including source of meats and eggs (wet markets or supermarkets, urban or suburban) and season of collection in to further univariate and multivariate analysis

The Premi test is a commercially available agar diffusion test based on the principle of growth inhibition of microorganisms, similar to other microbiological tests. The Premi test is convenient, easy to use and suitable as an initial screening test for antibiotic residues. Specificity (95.3%) and sensitivity of the Premi®Test (72.5%) were very satisfactory [8]. Briefly, we thawed the frozen meat, took approximately 2 cm³ of the meat and extracted the meat juice using the meat press delivered with the starter kit

of the Premi test. Then, we slowly pipetted 100 µL of the meat juice onto the agar in Premi test ampoules. For the negative control, 100 µL of distilled water was pipetted onto the agar in Premi test ampoules.

The ampoules were incubated at 64°C in an incubator, which was pre-heated for 10 min. After 3 h, we withdrew the ampoules from the incubator and determined the colour of the lower two-thirds of the solid agar. For eggs, the procedure was similar, i.e. we collected 100 µL of the yolk, inoculated it onto Premi test ampoules and incubated the ampoules at 80°C in a water bath for 10 min and later at 64°C for 3 h as described by the manufacturer. The ampoules that remained purple after the incubation were recorded as positive for antimicrobial residues, and those that turned yellow were recorded as negative for antimicrobial residues. The Premi test kit was obtained from Nizawa International Hi-Tech Corp.

Chi-square test was performed for the univariate analysis and the logistic regression test for the multivariate analysis using the SPSS 16.0 software. *P* values less than .05 are reported as statistically significant. The results of the logistic regression models are presented as adjusted odds ratios (ORs) with 95% confidence intervals (CIs).

3. RESULTS

From 1 Jan 2013 to 31 Dec 2014, we collected 175 samples from supermarkets (eggs, *n* = 50; chicken meat, *n* = 50; pork meat, *n* = 50; and fish, *n* = 25) and 175 samples from wet markets (eggs, *n* = 50; chicken meat, *n* = 50; pork meat, *n* = 50; and fish, *n* = 25). Ninety % were from southern Taiwan. In total, 350 samples were collected, and 8% were positive for antibiotic residues. We compared the different risk factors for the presence of antibiotic residues using the univariate analysis, and the results are presented in Table 1. The rates of positive antibiotic residues were not different between the meat types or seasons of collection (Table 1). For the samples from supermarkets, the rate of positive antibiotic residues was lower than that for the samples from wet markets (4% vs. 12%, *P* = .006). The rate of positive antibiotic residues for the samples from suburban areas was higher than for those from urban areas (19% vs. 7.3%);

however, the *P* value (0.054) obtained by the Chi-square test in the univariate analysis was greater than .05, which indicated that this difference was not statistically significant.

Table 1. Risk factors of foods being positive for antibiotic residues by univariate analysis

Risk factors	Antibiotic residues positive	Antibiotic residues negative	P-value
Food source			0.715
Chickens	8 (8%)	92 (92%)	
Eggs	7 (7%)	93 (93%)	
Fish	6 (12%)	44 (88%)	
Pork	7 (7%)	93 (93%)	
Markets			0.006
Wet markets	21 (12%)	154 (88%)	
Supermarkets	7 (4%)	168 (96%)	
Area			0.054
Suburban	4 (19%)	17 (81%)	
Urban	24 (7.3%)	305 (92.7%)	
Season			0.390
Summer	5 (5.8%)	81 (94.2%)	
Non Summer	23 (8.7%)	241(91.3%)	

The rates of positive antibiotic residue test results for the raw meat and egg samples purchased from different sources are shown in Fig. 1. There were no antibiotic residues detected in any eggs purchased from the supermarkets. Among the samples from the wet markets, chicken meat (12%), eggs (14%) and fish meat (16%) had a higher prevalence of antibiotic residues than pork meat (8%); however, these differences were not statistically significant (*P* = .72). In the multivariate analysis using the logistic regression test, the suburban locale (OR: 4.59; 95% CI: 1.21–17.42; *P* = .03) and the wet market type (OR: 3.51; 95% CI: 1.43–8.62; *P* = .006) were two independent risk factors for meats and eggs to be positive for antibiotic residues after an adjustment (Table 2).

Table 2. Risk factors of foods being positive for antibiotic residues by multivariate analysis

Variables	Odds ratio	95% confidence interval	P-value
Suburban area	4.59	1.21-17.42	0.025
Wet market	3.51	1.43-8.62	0.006

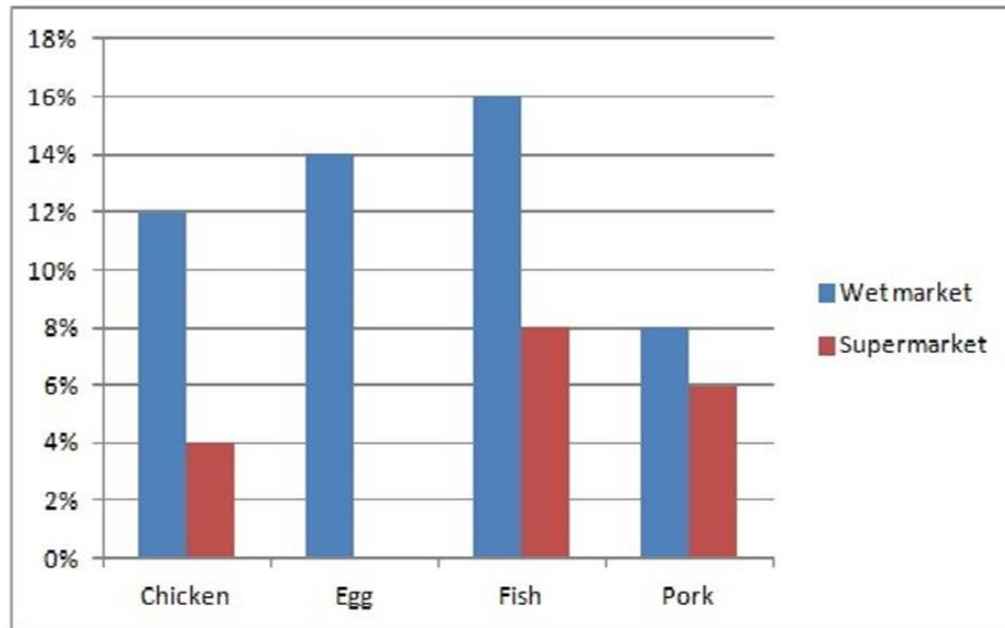


Fig. 1. The rates of positive antibiotic residue test results for the raw meat and egg samples purchased from different sources (Y axis, positive rate of antimicrobial residues)

4. DISCUSSION

A total of 8% of the meats and eggs sampled in this study were positive for antibiotic residues, and this rate was higher for the samples from suburban areas than for those from urban areas and for the samples from conventional wet markets than for those from supermarkets in south Taiwan. The rate of antibiotic-residue positive samples in Taiwan was not high compared to those in African tropical countries (e.g. 21.4% in Tanzania, 89.3% in Nigeria and 37.2% in Madagascar) but was similar to that obtained in a study from Vietnam [17–21].

Similar to the data from the surveillance carried out by the Taiwanese government, the prevalence rates of antibiotic residues detected were higher for eggs, chicken meat and fish meat. Our rate of antibiotic residue violations was similar to that found by longitudinal surveillance (2004–2013) of drug residues in food animals in Taiwan [22]. The above study found that the average rate of samples that were negative for drug residues, including antimicrobials, in Taiwan was 93.4% (95% CI: 92.6–94.2%). Similar to our data, the Taiwan government surveillance in 2013 found that poultry products and fish meat had higher rates of positive antibiotic residue test results than pork meat [22]. In the surveillance, 11% of poultry products, including chicken meat

and eggs, were positive for antibiotic residues, but no pork meats were positive for antibiotic residues. The most commonly observed antibiotic residues in 2013 were trimethoprim and chloramphenicol [23], and further monitoring and the determination of the predominant classes of antimicrobials used are needed.

Traditional wet markets have existed in Asian countries, including Taiwan, long before the invention of the refrigerator. Wet markets have the advantage of selling the freshest produce, meats and other food products because the majority of the goods sold there are grown locally. However, there are concerns regarding the safety of fresh meat products sold at traditional markets in Taiwan [24]. The agricultural administration has established programs to raise consumer awareness of potential risks of fresh meats that were stored at room temperature for several hours. Although supermarkets sell chilled and frozen meat products, the majority of grocery shoppers in Taiwan still prefer purchasing fresh meat products at traditional markets where meat is displayed on counters or hangs on hooks [24]. Some studies have shown higher *Salmonella* contamination rates at non-integrated poultry companies and wet markets [15]. Additionally, a study in Africa showed a high prevalence of antibiotic residues in meat samples from

suburban and rural areas [21,25]. A study in Vietnam showed that the proportion of meat samples with antibiotic residues was significantly higher for suburban areas than for urban areas [20]. The results of this study indicated that antibiotic residues in products sold at conventional wet markets might be a food safety concern. The number of samples from suburban and rural areas and the number of fish meat samples were relatively small; thus, further studies are required to confirm the validity of the results of this study. It is unclear whether the differences between supermarkets and wet markets in the rates of samples positive for antibiotic residues were related to other factors, such as a herd size, a higher frequency of use of medicated feeds and producers' attitudes [26].

Our study had limitations, since neither random nor systematic sampling was taken and since it focused on southern Taiwan. Furthermore, the antibiotic residue detection method used in this study could not identify specific antibiotics. Further studies may be required to identify specific antibiotics present in fish meat and in food products sold in suburban areas.

5. CONCLUSIONS

This local meat and egg surveillance performed in 2013–14 found an 8% rate of antibiotic-residue positive samples. The rate is higher than that in the US but lower than the rates reported for African tropical countries. Previous surveillance data did not focus on the source of animal foods, such as the market type. In our study, the multivariate analysis showed that the suburban locale and wet market type were two independent risk factors for the presence of antibiotic residues in meats and eggs in Taiwan. Further surveillance of antibiotic residues in meats and eggs in these high risk areas may be necessary.

CONSENT

This article does not contain a study performed with human participants or animals. Informed consent was not given because all samples including meat and eggs were originally designated for human consumption in the market.

ETHICAL APPROVAL

This study does not involve human participants or animals; therefore no ethical approval was required.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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