



Antimicrobial Resistance in Zoonotic Pathogens

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Antibiotic use in animal agriculture has resulted in healthier animals, and these health-promoting effects, from which at least some of the growth-promoting effects arise, deserve more attention. However, since the development of antimicrobial resistance in animals, it has been raised as a 'hazard' in that the organism itself may be zoonotic, or may transfer its genetic material to a human pathogen. Thus, the crucial question is the degree of risk posed by the hazard. This has been the subject of debate, with a division between one group who concerns about the risk and the other group who believes that the evidence for a substantial risk remains sparse. This debate should be solved only by science approaches not by political or emotional bias. Here, we would like to introduce the current situation on antimicrobial resistance in major zoonotic pathogens in Korea.

Vancomycin-resistant enterococci (VRE) in Korea

According to the Korean Nationwide Surveillance of Antimicrobial Resistance (KONSAR) study, the percentage of VRE increased from 4% in 1997 to 16% in 2002 in human hospitals. Many *vanA* VRE were isolated from poultry meat (40%) as well. This *vanA* type of VRE, the most common one found both in humans and animals, is mediated by *Tn1546* or closely related with transposons, which confers gene cluster composed of the following seven genes; *vanR*, *vanS*, *vanH*, *vanA*, *vanX*, and *vanZ*. The heterogeneity of *Tn1546*-like elements has been previously reported and comprises point mutations, deletions, and the integration of insertion sequence (IS) elements. Among these variations, the presence of IS elements accounts for most of the heterogeneity of *Tn1546*-like elements. Until now, six kinds of IS elements have been found in *Tn1546*-like elements; *IS1542*, *IS19*, *IS1216V*, *IS1251*, *IS1216V-IS3*-like, and *IS1476*.

VRE isolates from humans (23 isolates) and poultry (20 isolates) were divided into 9 main types based on the patterns of ISs inserted into *Tn1546*-like elements. Most human isolates had insertion sequences whereas most poultry isolates showed indistinguishable pattern with the prototype *vanA* transposon, *Tn1546*. Interestingly, we found a novel IS element located in *Tn1546*-like elements. This IS element designated as *IS1678* was detected in the *vanX-vanY* intergenic region of a poultry isolate of *Enterococcus faecium*, which is resistant to vancomycin. Moreover, the transferability of vancomycin resistance between enterococci was confirmed in most human isolates (18/23) via mating experiments, whereas only three poultry isolates showed the transferability with comparatively low transfer rate. PFGE revealed extensive heterogeneity with no PFGE-deduced genetic overlap between human and

poultry isolates. It might be concluded that no evidence of clonal or horizontal dissemination of VRE between humans and poultry was found in Korea.

Methicillin-resistant *Staphylococcus aureus* (MRSA) in Korea

Fourteen *mecA* carrying *S. aureus* were isolated from various sources; 3 pre-methicillin resistant *S. aureus* (pre-MRSA) and 1 silent *mecA* carrying methicillin susceptible *S. aureus* (smMSSA) were isolated from retail chicken meat, and 3 and 7 MRSA were isolated from hospitalized dogs and humans, respectively. On the basis of staphylococcal cassette chromosome *mec* (*SCCmec*) analysis, human hospital strains had *SCCmec* types II, III as well as IV, and animal hospital strains carried *SCCmec* types II. Three chicken meat originated pre-MRSA had *SCCmec* types III, and one smMSSA isolate was not classified. All isolates except the smMSSA were multi drug resistant and had hospital acquired MRSA (H-MRSA) characteristics, although 1 human isolate, SN1, which harbored *SCCmec* types IV was shown as community acquired MRSA (C-MRSA). PFGE revealed that livestock product isolates and animal isolates had genetic relatedness between them, but not with human isolates although smMSSA strain from poultry meat was genetically similar with human isolates than livestock isolates. On the basis of multi-locus sequence typing (MLST) and BURST analyses, MRSA from humans with *SCCmec* type III and IV showed ST239 and ST254, respectively and the smMSSA showed ST8, which were all included in group 3. All the *SCCmec* type II and three pre-MRSA strains showed ST5 and they were included in group 1. No close relatedness between human isolates and veterinary fields' isolates has been found yet, however, there are possibilities of transmission of between them, and from hospital strains to the community or vice versa.

Antimicrobial resistant *Escherichia coli* in Korea

E. coli have been found in the intestine of many animals including humans. Each serotypes has particular pathogenicity to human; O1 may cause meningitis in infants, O157:H7 and some other types cause hemolytic-uremic syndrome (HUS), urinary tract infection as well as gastroenteritis in children or travelers. Extended spectrum beta-lactamases (ESBLs) are heterogenous groups of beta-lactamase enzyme that is found in a variety of members of the family *Enterobacteriaceae* including *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, other *Klebsiella* species, and *Escherichia coli*. Today, more than 150 different ESBLs have been described. Nearly all belong to one of four ESBL families; TEM, SHV, CTX-M, and OXA (for *P. aeruginosa* strains) with the majority belonging to TEM and SHV. ESBL-producing strains are resistant to beta-lactam antibiotics including third-generation cephalosporins such as cefotaxime, ceftriaxone, and ceftazidime. The clinical relevance of ESBLs is paramount due to the limited therapeutic options and increased risk of treatment failure in patients infected with such strains. Since the ESBL problem began in Western Europe in the early 1980s, the phenomenon subsequently spread to other countries and has been reported nearly worldwide. Hemolytic *E. coli* is known to cause porcine dysenteries. Among 56 *E. coli* which were isolated from swine fecal sample, 38 isolates had hemolytic characteristic. None of them produced ESBL, however they are highly resistant to tetracycline, ampicillin and chloramphenicol which have been usually used in animal husbandry. Antimicrobial susceptibility test was conducted for a total of 103 isolates composed of 38 from swine, 43 from human hospitals, and 22 from the Han River. Among 233 human isolates, 17 isolates (7.3%) produced ESBL. All of them except one isolate had TEM, and several isolates possessed AmpC, OXA

or CTX-M, additionally. Human clinical isolates were highly resistant to quinolone. Twenty-two ESBL producing *E. coli* were isolated from environment. Sixteen out of 22 isolates were found to have both of TEM and AmpC whereas human strain usually had only TEM. Moreover, TEM of ESBL producing human clinical *E. coli* isolates was found to be able to transfer to animal originated *E. coli* under experimental condition by filter binding method. There is no evidence of the transfer of *E. coli* from animal to human in this study.

Antimicrobial resistant *Salmonella* spp. in Korea

Salmonellosis is the most common food-borne disease in Korea. The major pathogens are *Sal. enterica* serovars *enteritidis* and *typhimurium*. Seventeen isolates (47.2%) of *Sal. Enteritidis* and 8 isolates (22.2%) of *Sal. typhimurium* were included in 36 non-typhoidal *Sal. spp.* whereas 64 livestock isolates were composed of 15 isolates (23.44%) of *Sal. typhimurium*, 14 isolates (21.88%) of *Sal. enteritidis* and other *Sal. spp.* Despite the efforts to control salmonella including resistant strains, *Sal. spp.* have still been common in animal-derived foods. No significant difference in antimicrobial resistance profiles between human and animal isolates was found, however human isolates were usually resistant to more antimicrobials. According to the PFGE, all isolates were divided into 6 types (A-F). Type A was classified to 13 subtype. Nine human isolates and 11 poultry isolates belong to the same subtype. This suggests there are possibilities of transmission of between two sources, human and poultry.

Conclusion

Resistance in bacteria isolated from food animal has been suspected as a potential source of resistance in human pathogens. Where resistance is present among zoonotic organism, then it is by definition possible for resistant bacteria from animals to be transmitted to a human.

All the situations persuade that antibiotic resistance of bacteria in humans and animals can be undoubtedly selected by the use of antibiotics such as in organisms that are part of the normal flora as well as in pathogens including zoonotic pathogens. The possibility of cross contamination of antibiotic resistant bacteria itself or mixing genetic material of resistance bacteria from different origins (among animal, animal products, and human) seemed to be very low based on the results in our study by comparing their phenotypic and genotypic characteristics.

However, it is still unclear whether superbacteria of animal reach to human or not. Interestingly, many ESBL producing *E. coli* were isolated from environment, the Han River. Therefore, antimicrobial resistance in food producing animal and hospitalized human cannot be explained by simple assumption, because of the complexities associated with the epidemiology of antimicrobial resistance. At this point, we should concentrate instead on minimizing the transmission of all zoonotic pathogens regardless of their antibiotic susceptibility, by insistence on good hygiene practices on farms, in slaughter houses, during distribution and marketing of food, in food preparation, and, finally, by the consumer.

Equally, the judicious use of antibiotics and continuous screening for resistant bacteria are foremost to prevent the emergence and spread of resistant bacteria.

References

1. Bywater RJ. Veterinary use of antimicrobials and emergence of resistance in zoonotic and sentinel bacteria in the EU. *J Vet Med.* 2004;B51:361-363.
2. Bywater R, Deluyker H, Deroover E, Jong A, Marion H, McConville M, Rowan T, Shryock T, Shuster D, Thomas V, Valle M, Walters J. A European survey of antimicrobial susceptibility among zoonotic and commensal bacteria isolated from food-producing animals. *J Antimicrob chemother.* 2004;54:744-754.
3. Jung WK, Lim JY, Kwon NH, Ike Y, Tanimoto K, Kim JM, Hong SK, Koo HC, Kim SH, Park YH. Occurrence of vancomycin-resistant enterococci from various animal sources in Korea. *Appl Environ Microbiol Submitted.* 2004.
4. Jung WK, Hong SK, Kwon NH, Koo HC, Park YH. Nucleotide sequence of IS1678: an insertion sequence identified within *vanA* cluster of enterococci. *Antimicrob Agents Chemother.* Accepted. 2005.
5. Kwon NH, Kim SH, Park KT, Bae WK, Kim JY, Lim JY, Ahn JS, Lyoo KS, Kim JM, Jung WK, Noh KM, Bohach GA, Park YH. Application of extended single-reaction multiplex polymerase chain reaction for toxin typing of *Staphylococcus aureus* isolates in Korea. *Intern J Food Microbial.* 2004;97:137-145.
6. Kwon NH, Park KT, Moon JS, Jung WK, Kim SH, Kim JM, Hong SK, Koo HC, Joo YS, Park YH. Staphylococcal Cassette Chromosome *mec* (SCC*mec*) Characterization and Molecular Analysis for Methicillin Resistant *Staphylococcus aureus* Isolated from Bovine Milk in Korea and Noble SCC*mec* type VI. *Antimicrob Agents Chemother.* Submitted 2004.
7. Mendelson M, Hait V, Ben-Israel J, Gronich D, Granot E, Raz R. Prevalence and risk factors of extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae* in an israeli long-term care facility. *Eur J Clin Microbiol Infecti. Dis.* 2005;24:17-22.
8. Phillips I, Casewell M, Cox T, Groot BD, Friis C, Jones R, Nightingale C, Preston R, Waddell J. Does the use of antibiotics in food animals pose a risk to human health? A critical review of published data. 2004;53:28-52.
9. Seo KS, Lim JY, Yoo HS, Bae WK, Park YH, Comparison of Vancomycin-Resistant Enterococci Isolates from Human, Poultry and Pigs in Korea. *Vet microbiol Accepted.* 2004.
10. Seo KS, Song DS, Gwyther M, Park YH Development of multiplex PCR for detection of vancomycin resistant enterococci (VRE) and epidemiological application in Korea. *J Vet Sci.* 1999;39(2):247-256.
11. Yang SJ, Park KY, Kim SH, Noh KM, Besser TE, Yoo HS, Kim SH, Lee BK, Park YH Antimicrobial resistance in *Salmonella enterica* serovars *enteritidis* and *typhimurium* isolated from animals in Korea : comparison of phenotypic and genotypic resistance characterization. *Vet Microbial.* 2002;86:295-301.