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Epidemiology and Origins of Antimicrobial Resistance in Animals

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The use of antibiotics in food animals selects for bacteria resistant to antibiotics used in humans, and these might spread via the food to humans and cause human infection, hence the banning of growth-promoters. The actual danger seems small, and there might be disadvantages to human and to animal health. The low dosages used for growth promotion are an unquantified hazard. Although some antibiotics are used both in animals and humans, most of the resistance problem in humans has arisen from human use. Resistance can be selected in food animals, and resistant bacteria can contaminate animal-derived food, but adequate cooking destroys them. How often they colonize the human gut, and transfer resistance genes is not known. 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 scientific approaches not by political or emotional bias. Here, we would like to introduce the current situation on antimicrobial resistant bacteria in animal and animal products in Korea.

Vancomycin-resistant enterococci (VRE) in Korea

Vancomycin resistant enterococci (VRE) that are resistant to high level of vancomycin and teicoplanin cause serious illness resulting in limited therapeutic options in hospitalized patients. VRE are frequently isolated the human hospital as well as outside the hospital from materials such as sewage, food, animals, and healthy human fecal samples throughout the world.

In this study, 52 *vanA* carrying *Enterococcus faecium* which showed high level resistant to vancomycin were isolated from meat, feces, and raw milk samples collected in Korea from March to November 2003. Further genotyping of the *E. faecium* isolates carrying *vanA* using pulsed-field gel electrophoresis (PFGE) revealed extensive heterogeneity. The vancomycin resistance transferability test revealed that only two of the 52 VRE were able to transfer vancomycin resistance to other enterococci.

Phenotypic and genetic characteristics of the 43 VRE isolates from humans and animals in Korea were investigated to verify the origin and transmission of these VRE between animals and humans. VRE from 23 human isolates and 20 poultry isolates showed multi-resistance to antibiotics, but they had different resistance patterns; human isolates were significantly more resistant to ampicillin and high-level resistant to gentamicin than animal isolates. The transferability of vancomycin resistance between enterococci was determined in most human isolates (18/23) via mating experiments, whereas only three poultry isolates showed the transferability with significantly low transfer rate ($P < 0.05$). PFGE of *E. faecium*, overlapping PCR of Tn1546-like elements, and sequencing analyses were performed to compare genetically the human and poultry isolates. PFGE revealed extensive heterogeneity with no PFGE-deduced genetic overlap between human and poultry isolates. Furthermore, the Tn1546-like elements in humans and poultry were different based on the results of the overlapping PCR and sequencing analysis. Twenty out of 23 human VRE isolates had insertion sequences, IS1216V-IS3-like, IS1542, IS19, and IS1216V in the *orf1*, *orf2-vanR*, *vanS-vanH*, and *vanX-vanY* regions, respectively. However, only 4 out of 20 animal VRE isolates had insertion sequences, IS1216V-IS3-like, and IS1216V or IS1678 in the *orf1* and *vanX-vanY* regions, respectively. Interestingly, IS1678 in *vanX-vanY* intergenic region and IS1216V-IS3-like in the *orf1* region of Tn1546 with a novel intervening sequence, *orf1*, were newly found from poultry isolates through this study. 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

MRSA is one of the most important pathogens in human and veterinary hospitals. The isolation of MRSA from

animals and foodstuffs has been reported with an increased incidence. However, methicillin is not used in animal husbandry or in animal hospitals in Korea. Fourteen MRSA and a silent *mecA* carrying methicillin susceptible *S. aureus* (smMSSA) were isolated from bovine milk suspected to be infected with *S. aureus* in Korea (isolation rate: 0.18%). In addition, three pre-MRSA and one silent *mecA*-carrying methicillin susceptible *S. aureus* (smMSSA) were isolated from retail chicken meat, and three MRSA were isolated from hospitalized dogs in Korea. In bovine isolates, SCC*mec* of 14 strains was classified as subtype IVg which was designated as new SCC*mec* type in this study, and one smMSSA strain was not classified. The three pre-MRSA isolates from chicken meat were determined to have a staphylococcal cassette chromosome *mec* (SCC*mec*) type III, and the smMSSA isolate was not classified. The animal hospital isolates were found to contain SCC*mec* type II. Those isolates and seven *S. aureus* isolated from hospitalized humans were examined in order to determine the epidemiological origins of MRSA. Multilocus sequencing typing (MLST) revealed that the chicken meat and bovine milk isolates were closely related to the animal hospital isolates. The SCC*mec* characteristics and MLST analyses indicated the possibility of the human to animal transmission of MRSA. These results highlight the importance of identifying MRSA carriers as well as intercepting MRSA transmission because MRSA is becoming increasingly widespread without any plausible relationship with the use of methicillin.

Campylobacter spp. from meat in Korea

Campylobacter species are some of the most common causes of bacterial diarrhea in humans worldwide. It is considered to be mainly a food-borne pathogen that is found in raw or undercooked poultry and serves as an important source of sporadic campylobacteriosis. A total of 770 samples of retail raw meat were examined for the presence of *Campylobacter* spp. The samples were obtained randomly from 232 retail stores in Korea from September 2001 to April 2006. The highest contamination rates were observed in chicken meat (81.4%, 220 of 270 samples) whereas the rates of contamination in pork and beef were extremely low (1.6% and 1.2%, respectively). The antibiotic resistant patterns of the 317 *Campylobacter* isolates were examined using the agar dilution method. Resistance to doxycycline was the most common (97.5%), followed by ciprofloxacin (95.9%), nalidixic acid (94.6%), tetracycline (94.6%), enrofloxacin (84.2%), and erythromycin (13.6%). All *Campylobacter* isolates from the retail raw meat were resistant to at least one of the six antibiotics tested, and 296 isolates (93.4%) showed multi-drug (4 or more antibiotics) resistance. This demonstrates that the multi-drug resistant *Campylobacter* spp. are widespread in meats in Korea. Therefore, more strict HACCP applications and governmental strategies to diminish *Campylobacter* contamination in raw chicken meats should be employed.

Antimicrobial resistant bacteria from small animal hospitals in Korea

Number of people live with companion animals has dramatically increased and antimicrobial agents used in human medicine are frequently used in small animal veterinary practice. Therefore, companion animals may be an important reservoir for transmission of antimicrobial resistance. The occurrence of important antimicrobial resistant bacteria, methicillin-resistant *S. aureus* (MRSA), methicillin-resistant coagulase negative staphylococci (MR-CNS), vancomycin-resistant enterococci (VRE), and extended-spectrum- β -lactamase (ESBL)-producing *E. coli* and *K. pneumoniae*, and the possible relatedness among human, animal, and environmental isolates were investigated in small animal hospitals. A total of 64 samples were taken from 15 veterinary staff, 33 hospitalized animals, and 6 environmental surfaces of animal hospitals and antimicrobial disk susceptibility tests were performed according to CLSI guideline. A total of 36 MR-CNS from 5 staff, 7 dogs, and 3 environmental sites, and 1 VRE and 2 ESBL-producing *E. coli* and 1 *K. pneumoniae* from dogs were isolated. One MR-CNS isolated from healthy dog living in a hospital was resistant to vancomycin. MRSA was not detected. Genotyping of the isolates and further study to identify the specific mode of transmission between human and animals are currently ongoing. Currently, national surveillance programs only focus on food animals and data on companion animals are needed for assessing the risk of transmission of antimicrobial resistance.

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 possibly transmitted from animals to human, or vice versa.

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. 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 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 monitoring resistant bacteria are foremost to prevent the emergence and spread of resistant bacteria.

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