

Westhampton isolates incubated in LB for 48 h. There was no significant correlation between antimicrobial resistance and biofilm formation of the isolates. Biofilm cell survival and recovery growth after sanitization exhibited most isolates were highly susceptible to all sanitizers tested.

Conclusions: Our results provide baseline information on the distribution of *Salmonella* serotypes in chicken slaughterhouses, and indicate the need for improved farming practice and for more cautious use of antimicrobial agents. Also, this study suggests that the sanitization during the slaughtering process may be necessary to reduce *Salmonella* contamination in chicken carcasses.

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Distribution of serovars of *Salmonella* in pet turtles being sold in South Korea

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Introduction: Salmonellosis in children caused by pet turtles has gradually increased public health issues. Reptile associated salmonellosis (RAS) is a serious disease among the infants and kids who contact reptiles like a pet turtles. In the United States in 1975, it was banned to sell the pet turtle whose size was under 4 inch in order to protect kids from *Salmonella* infection from turtles. After then, the number of infection was effectively decreased by that process. Although *Salmonella* is a kind of bacteria in rod shape and gram negative, it has many types of serovars which were over 2,600 serotypes. The objective of this study was to conduct the investigation of *Salmonella* spp. in pet turtles which were sold in commercial market in South Korea and to find out the route of infection.

Materials and Methods: Pet turtles were collected by purchasing from small pet vender or super market. Isolation of *Salmonella* was isolated from fecal swabs and total organs of turtles and environmental samples including water and feed. *Salmonella* was identified using multiplex PCR (multiplex polymerase chain reaction) method and VITEK II bacterial identification system. Serotypes and minimum inhibitory concentrations (MICs) of isolates were determined following conventional method.

Results: *Salmonella* isolate identified were *Salmonella enterica* subsp. *enterica* serovars, Minnesota, Pomona, Kottbus, Newport, Berta, and Uzaramo, and *S. enterica* subsp. *diarizonae* from swab and total organ samples. However there were no isolates from environmental samples including water and feed.

Conclusions: This study showed that turtles were potential reservoirs of *Salmonella* which can cause salmonellosis in humans, especially children and kids. It also suggests that proper quarantine programs are required to protect

young children from salmonellosis caused by pet turtles. Further studies are underway to find out pathways of *Salmonella* transmission in South Korea after import of turtles.

References

- [1] Pedersen K., et al. *Zoonoses and Public Health*, 2009, 56:238-242.
- [2] Piasecki T., Chrzastek K. and Wieliczko A., *ACTA VET. BRNO*, 2014, 83: 287-294.
- [3] Lee K. et al. *Journal of Applied Microbiology*, 2009, 107: 805-811.

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Characterization of *Escherichia coli* isolated from Korean native cattle

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Introduction: *Escherichia coli* is one of the most important cause of diarrhea in cattle. Pathogenic *E. coli* is designated as enterotoxigenic *E. coli* (ETEC), Shiga toxin-producing *E. coli* (STEC), enteropathogenic *E. coli* (EPEC), and enterohemorrhagic *E. coli* (EHEC). Each pathotype can cause severe diarrhea in cattle via the production of variable toxins (ETEC - fimbriae and STa; STEC - shiga toxins; EPEC - intimin; EHEC - shiga toxins and intimin). This study was conducted to determine the prevalence and characteristics of pathogenic *E. coli* strains from Korean native cattle.

Materials and Methods: From 2013 November to 2015 May, fecal samples were collected from 852 cattle with diarrhea ($n = 771$) and without diarrhea ($n = 81$). Among them, calves aged below six months were 711. The samples were plated onto sheep blood and MacConkey agar and incubated overnight at 37°C. From each sample, four suspected colonies were selected and subcultured, and the isolates were screened by PCR of virulence genes (f5, f17, f41, stx1, stx2, eae, hly, saa and sta). O serogroup determination of 145 *E. coli* isolates was performed by slide agglutination technique using 181 different rabbit antisera (Joongkyeom, Korea).

Results: In total, *E. coli* encoding one or more of virulence genes were isolated from 420 cattle with diarrhea ($n = 369$) and without diarrhea ($n = 51$). Overall, the most frequently identified virulence factors were f17 (249/852, 29.2%), followed by hly (9.5%), eae (9.0%), saa (8.3%), stx1 (7.8%), stx2 (5.8%) and sta (2.7%), whereas the f5 and f41 was not detected in any of the isolates. Among

isolated *E. coli*, the prevalence of pathogenic strain was 17.3%; 63 STEC, 36 EPEC, 41 EHEC, 7 ETEC and 705 non-toxigenic strains. The most prevalent pathotype was STEC that carried the stx1 or stx2, three isolates were stx1 only, 15 were stx2 only and most complex combinations were stx1-saa (15/63, 23.8%) and stx2-sta (14/63, 22.2%). The combinations of eae-hly (21/36, 58.3%) and stx1-eae-hly (33/41, 80.5%) were frequently observed among EPEC and EHEC, respectively. As a result of O serogrouping with 145 isolates, 87 strains (60.0%) were typed in one of serogroups. The majority of strains ($n = 34$) belonged to three kinds of O serotype, which included O26 (17/87, 19.5%), O168 (9/87, 10.3%), and O15 (8/87, 9.2%). In particular, EHEC and STEC make up the majority of the O26 (70.6%) and O168 (77.8%) serotype, respectively. There is no difference of the distribution of pathotype according to cattle ages, however, the highest prevalence was observed in cattle aged 7-12 months. Regional prevalence also showed no difference except the EHEC which showed a higher prevalence in the central region. Seasonal prevalence of pathotype was confirmed to be infected more in spring-summer (21.7%) than in the fall-winter (10.1%).

Conclusions: This study was performed to investigate the prevalence and characteristics of pathogenic *E. coli* strains from Korean native cattle. The results showed the prevalence of pathogenic *E. coli* was 17.3% however, there was no distinction between diarrheic and non-diarrheic cattle. The high frequency of STEC from cattle implies this animal species can be potential reservoir of pathogenic *E. coli* for humans. Further studies are needed to form the epidemiological basis using PFGE or MLVA.

References

- [1] T. D. Nguyen, T. T. Vo, H. Vu-Khac. 2011. Virulence factors in *Escherichia coli* isolated from calves with diarrhea in Vietnam. *Journal of Veterinary Science* 12(2): 159-164.
- [2] G. I. Andrade, F. M. Coura, E. L. S. Santos, M. G. Ferreira, G. C. F. Galinari, E. J. Facury Filho, A. U. de Carvalho, A. P. Lage, M. B. Heinemann. 2012. Identification of virulence factors by multiplex PCR in *Escherichia coli* isolated from calves in Minas Gerais, Brazil. *Trop Anim Health Prod* 44: 1783-1790

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Molecular identification and detection of major pathogens from bovine mastitis

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Introduction: Bovine mastitis has been one of the most important diseases in the dairy industry and caused mainly

by *Staphylococcus aureus*, *E. coli* and *Streptococcus agalactiae*. In case of subclinical mastitis, coagulase negative *Staphylococci* (CNS), Gram negative rods and *S. aureus* were identified at high frequency in order. For molecular epidemiological study, multilocus sequence typing (MLST) has been used; however, recently rpoB sequence typing (RST) was reported to be more useful in case of *S. aureus* study. In the present study, we identified bacteria from bovine mastitis cases from nationwide dairy farms and performed RST for molecular epidemiological study of *S. aureus*. In addition, we tested a SYBR Green dye-based real time PCR to identify major pathogens in raw milk specimens.

Materials and Methods: In total, 383 raw milk specimens from 28 dairy farms were used in the present study. Sheep blood agar was used for bacteria culture and VITEK2 (Biomerieux) was used for bacterial identification. RST was performed as previous (Seong et al., 2013) and SYBR green dye-based real time PCR was conducted by using rpoB-based pathogen-specific primer sets and StepOne™ Real-time PCR system. (Applied Biosystem).

Results: Raw milk specimens 24 (85.7) out of 28 from dairy farms were infected with more than 2 bacteria. *Staphylococcus* spp. were most frequent followed by *Streptococcus* spp. *S. aureus* was the most frequent species and its 7 isolates were classified into RST 8-1 (2), 10-2 (2), 11-4 (1) and 22-1 (2). RST 10-2 were clustered with RST 11-1 which were composed of *S. aureus* isolates from humans. According to our result, major pathogens of bovine mastitis showed specific melting temperatures (T_m) which could be easily differentiated each other.

Conclusions: The bovine isolates of RST 10-2 may be genetically related to human isolates of RST 11-1, and the rpoB-based pathogen-specific real time PCR may be promising to detect major pathogens of bovine mastitis at the same time.

References

- [1] Seong W. et al. J. Comparison of complete rpoB gene sequence typing and multi-locus sequence typing for phylogenetic analysis of *Staphylococcus aureus*. *Gen. Appl. Microbiol.*, 59, 335-343 (2013)

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Genotypic analysis of *Salmonella* isolated from poultry in South Korea

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Introduction: *Salmonella* is widely concerned harmful organism which can affect humans and animals. Besides it has been known to be a zoonotic pathogen mainly found in animal sources such as meat, poultry and eggs, etc.