STUDY OF THE TRANSFER OF SHIGA TOXIN-PRODUCING *ESCHERICHIA COLI* DURING THE SLAUGHTER OF CATTLE USING MOLECULAR TYPING COMBINED WITH EPIDEMIC DATA

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I. INTRODUCTION

Shiga toxin-producing Escherichia coli (STEC) is a pathogenic bacterium that produces Shiga toxin and causes diarrhea, hemorrhagic colitis (HC), and even life-threatening hemolytic uremic syndrome (HUS) in humans. Beef cattle are the important reservoir of STEC, and cross-contamination can easily occur during beef production, which has potential food safety risks. Despite the risk associated with beef, limited information is available regarding the prevalence and characteristics of common foodborne pathogens in beef in China in recent years except our previous survey conducted 6 years ago [1]. In the present study, samples from eight processing points in two beef cattle slaughtering plants were collected and further characterized to clarify the prevalence of STEC at different operating points in the slaughter process and conduct traceability analysis, to make a further expand and longitudinal extension of our previous study, the results of this study is anticipated to useful for the development of food safety objectives and the improvement of control measures for beef safety control.

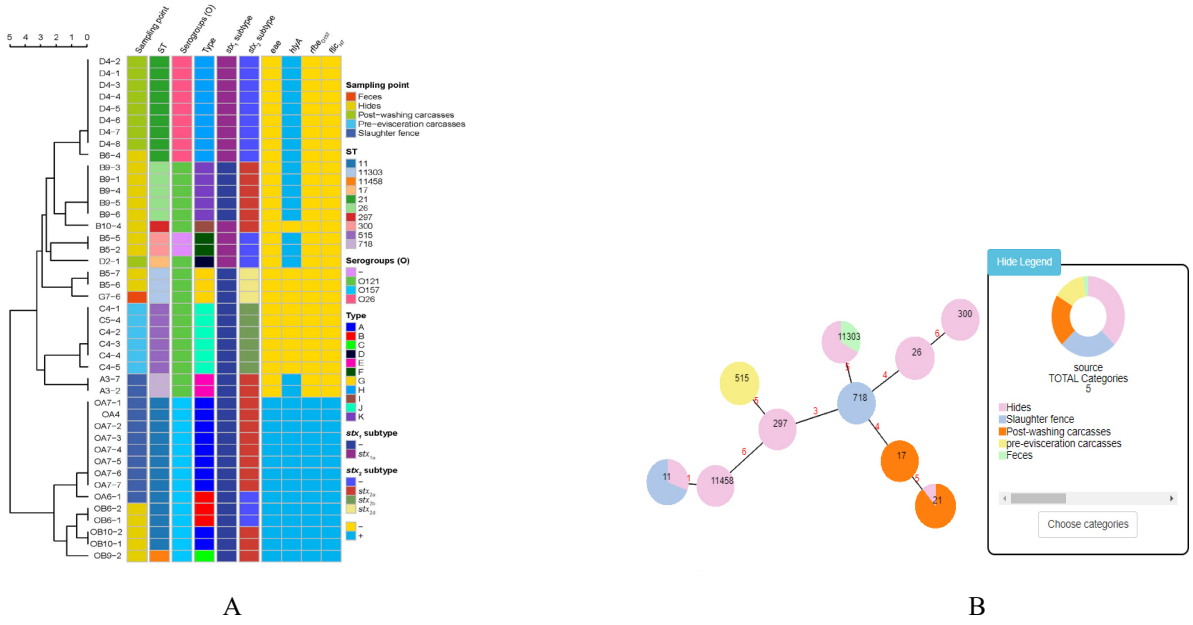
II. MATERIALS AND METHODS

From April 2021 to November 2021, 435 samples were collected from two beef processing plants with a processing capacity of 60–70 animals per day. Eight critical points including feces, slaughter fence, hide, pre-evisceration carcasses, post-washing carcasses, chilled carcasses, environmental samples (knives and spray water), and meat. Swab samples were collected on 13 points of carcass [2]. Immunomagnetic separation (IMS) was carried out using immunomagnetic beads coated with an anti-*E. coli* O157:H7 anti-body. After separation, detection of $flic_{h7}$ and *rfbE* by PCR, secondary screening and isolation of the PCR positive samples using CHROMagar™ O157. For non-O157 STEC samples, two genes stx_1, stx_2 were detected by multiplex PCR, the *stx*-positive samples were selected for a secondary screening by CHROMagar™ STEC. Each STEC isolate was characterized for O serogroups and *stx* subtypes by PCR. MLST fractal analysis of *E. coli* strains was based on the MLST database of *E. coli*.

III. RESULTS AND DISCUSSION

In this study, the PCR-positive (isolation rate) of feces, slaughter fence, hide, pre-evisceration carcasses, post-washing carcasses, chilled carcasses, meat, environment samples were 10.9% (1.80%), 36.4% (7.30%), 27.3% (12.70%), 14.6% (3.60%), 12.0% (8.0%), 1.8% (0%), 13.0% (0%), and 2.5% (0%). The prevalence in hides is much higher than the other critical points indicating severe cross-contamination in hides, which was also confirmed by the subsequent molecular typing and traceability analysis. O121 and O26 of the “TOP SIX” serotypes and O157 were found in the strains isolated, as well as the presence of virulence factor stx_{2a}, which has a high pathogenic capacity, thus showing a potential high risk to public health. 11 strains isolated in this study that harbored both *eae*,

stx$_2$ and *hlyA* were all further serotyped as *E. coli* O157:H7. All the above results indicate the strong pathogenic ability of these *E. coli* O157 strains and there is a correlation between serotype and virulence genes. The 43 STEC isolates were classified into 11 types of A-K by combining virulence factors, O serotypes and ST types (Figure 1A). The presence of the same type in different processes reveal cross-contamination during the slaughter process, the results indicate that hides play an important role in cross-contamination, at the same time, strains isolated from hide samples showed the most diverse MLST types (Figure 1B), this confirm that hides play an important role in the transmission of microorganisms among cattle, slaughter fence, and carcasses. The cleaning, sterilization, and shower processes of beef cattle before slaughter should be strictly controlled in the processing plants to reduce the amount of STEC carried in the hide.



A B

Figure 1. Distribution of ST types, O serotype and virulence factors in STEC isolates (A) and ST types of strains isolated from the samples of different processes (B).

IV. CONCLUSION

In the present study, the prevalence and biological properties of STEC strains isolated from beef cattle slaughtering plants were determined. Two of the “TOP SIX” serotypes were found in the strains isolated, thus showing a potential high risk to public health. The animal’s hide appears to play a significant role in cross-contamination. The results of the present study are expected to provide a baseline guide for improving beef safety control.

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REFERENCES

1. Dong, P., Xiao, T., Nychas, G. E., Zhang, Y., Zhu, L. & Luo, X. (2020). Occurrence and characterization of Shiga toxin-producing *Escherichia coli* (STEC) isolated from Chinese beef processing plants. Meat Science 168, 108188.
2. Barkocy-Gallagher, G. A., Berry, E. D. & Rivera-Betancourt, M. (2002). Development of Methods for the Recovery of *Escherichia coli* O157:H7 and *Salmonella* from beef carcass sponge samples and bovine fecal and hide samples. Journal of Food Protection 65, 1527-1534.