PULSE FIELD GEL ELECTROPHORESIS ANALYSIS OF SALMOELLA ISOLATED FROM THREE CHINESE BEEF ABATTOIRS

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I. INTRODUCTION

Salmonella infection is a very common foodborne illness in China. Most of these infections are associated with the consumption of contaminated meat products [1]. Animal feces and hide are the major reservoirs of *Salmonella*, therefore, the contamination of carcasses can be formed through the feces-animal contact, animal-animal contact, and the puncture of gastrointestinal tract during processing [2]. Our previous study has provided some epidemiological data such as the prevalence incidence, serotype and phage type distribution in three beef abattoirs. However, the prevalence in beef abattoirs was quite low. It is necessary to adopt some molecular typing approaches to explore more information on the contamination source of *Salmonella*. Pulsed field gel electrophoresis (PFGE) is currently considered as a gold standard method for epidemiological subtyping of *Salmonella* strains [3]. Thus, in this study, PFGE was applied based on the epidemiological data of our previous findings, to gain a better insight of the distribution of *Salmonella* in beef processing plants.

II. MATERIALS AND METHODS

Forty-eight *Salmonella* isolates collected from seven processing points at three beef plants in China were used for the molecular typing in this study. Of which 45 isolates belong to six serotypes and 3 isolates couldn't be typed. PFGE was performed according to the internationally standardized PulseNet protocol using 15 U XbaI restriction enzyme [4]. *S. Braenderup* (H9812) served as a DNA size marker. Banding patterns were analyzed using BioNumerics v.6.6 (AppliedMaths, Kortrijk, Belgium). An unweighted pair group method with arithmetic mean (UPGMA) dendrogram was constructed using a dice coefficient.

III. RESULTS AND DISCUSSION

Eleven to eighteen stripes ranging from 20 to 1100kb were found in each PFGE lane. Five large clusters (I to V) which were high correlated to the original plant were divided at the dice coefficient of 73% (Fig.1). This result showed PFGE has a high capacity of discriminating on the isolates from different origins. Thirteen types of different patterns (named A-M) were found by visual inspection. The results of banding patterns showed 22 different profiles (PT1-PT22) were yielded from 48 isolates of *Salmonella* at the similarity level of 96%. This result provided more detailed information on the cross contamination in each plant. In profile PT8, six highly similar strains were isolated from three different processing points, including post-washing carcasses, hide and feces (Fig.1). It can be deduced that the *Salmonella* strain isolated from the "post-washing carcasses" of animal 10 derived from the hide of the same animal during the hide removal (strain 134-A-**D10**-AGO and 137-A-**A10**-AGO, naming rules: internal lab code-plant code-**processing point and animal code**-abbreviations for serotypes). Furthmore, it showed *Salmonella* isolated from the feces of animals 6 and 9 (136-A-**G06**-AGO, 071-A-**G09**-AGO) had the same profile as that isolated from the hide of animal 10 (137-A-**A10**-AGO), indicating that feces are the final source of the carcasses contamination, and the intervention measures in this plant failed to cut off the transmission.

A more widespread of cross contamination between hide and feces were also found after the analysis of PT1, PT4, PT5, PT11, PT15, PT18 and PT22 using the same method. These results indicate a serious cross contamination among animals before entering the slaughter house. Good pre-slaughter management such as the adequate sterilization of lairage and vehicle, enough space and appropriate fasting time should be emphasized. Additionally, as there is a high level of cross contamination between the hide and feces, it

is of great importance to apply an effective intervention after the hide removal process to cut off the fecal - hide-carcass cross contamination route.

Compared to serotype, more details can be provided by PFGE and a high accuracy traceability analysis can be implied in beef plants. For example, a carcass isolated strain 620-D-B08-AGO showed the PT13 PFGE pattern (Fig.1), which only has a 50.9% similarity with PT4. This indicates that the strain originates only from the hide of animal A07, not from the animal A01 and A04, although those isolates had the same serotype.

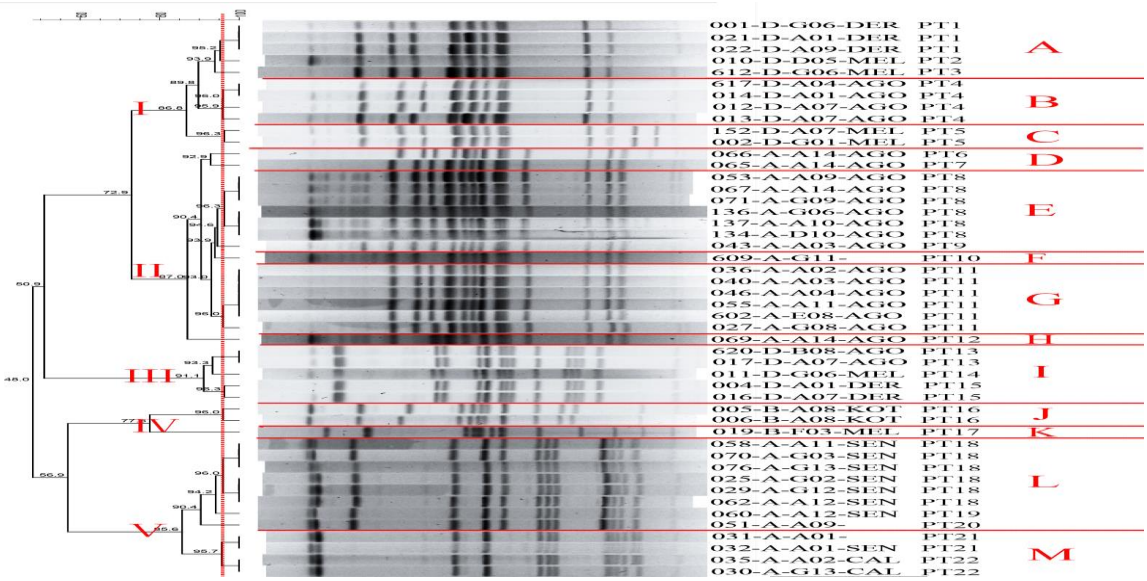

Fig.1 Dendrogram of 48 *Salmonella* strains isolated from 7 processing points in 3 beef abattoirs

Note: strain number contains four data items: the first item is the internal lab code; three letters (A, B or D) in the second item stand for sampling plant; letter A-G in item 3 stands for seven sampling points (hide, pre-evisceration carcasses, post-evisceration, post-washing carcasses, chilled carcasses, meat, feces) and Arabic numerals stand for animals; a combination of 3 letters in item 4 stands for different serotypes.

IV. CONCLUSION

The PFGE typing data showed a better insight into the distribution of *Salmonella* in three Chinese beef abattoirs. A carcass-hide-fecal cross contamination was verified and a widespread of cross contamination between different animal's hide and feces were found.

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