BRIEF NOTE

Isolation of \textit{Yersinia enterocolitica} from Cow’s Intestinal Contents and Beef Meat

Masanao INOUE and Masuko KUROSE

Hygienic Laboratory of Okayama Prefecture, Furugyo-cho, Okayama-shi, Okayama 700

(Received for publication August 1, 1974)

Many cases of the disease with \textit{Yersinia enterocolitica} (\textit{Y.e.}) are reported by European investigators [1, 3–5]. Zen-yoji et al. [12], Tsubokura et al. [7] and Asakawa et al. [2] also have reported about the diseases in Japan. Particularly, the community outbreak by \textit{Y.e.} in 1972 at a primary school in Shizuoka Prefecture was perhaps the first case in the world. Human infection with \textit{Y.e.} has been overlooked by the most investigators and only a few have studied \textit{Yersinia pseudotuberculosis} [8] in Japan. Since then, Asakawa et al. [2] have been interested in \textit{Y.e.} to be an agent of food poisoning, which is one of the important problems in public health, but ecology of \textit{Y.e.} remains unclarified. The present studies have mainly been initiated in order to obtain more information concerning the ecology of \textit{Y.e.} in Japan.

Attempts have been made to isolate \textit{Y.e.} from foods, water and animals, and Ahvonnen [1], Mollaret [5] and Tsubokura et al. [7] have suggested an increase of the distribution of \textit{Y.e.} in animals, especially in pigs. But the papers dealt with the isolation of \textit{Y.e.} from bovine intestinal content and beef meat are very few [10, 11].

In the present studies, \textit{Y.e.} have been isolated from cow’s intestinal contents and beef meat purchased from butchers.

One hundred and fifteen specimens of cow’s intestinal content were collected from the slaughterhouse in Okayama City, and the other sixty-one specimens of beef meat were collected at butchers in Okayama City during the period from July, 1973 to April, 1974.

One hundred-gram specimen was divided into two parts, one of which was enriched by placing it in 500 ml selenite medium, and the other half was also enriched by placing it in 500 ml of 1/15 M phosphate buffer solutino (P.B.S., pH 7.6) in each trial.

The selenite medium was prepared by adding 40 mg novobiocin per liter, which was incubated at 37°C for 24 hours, and P.B.S. medium was incubated at 5°C for 3 weeks. The materials were subcultured aerobically at 25°C for 48 hours on the SS agar plates, and SSK or SSB agar plates by triple plate methods. Biochemical characterization of isolates was investigated by the conventional methods of \textit{Enterobacteriaceae}, biotyping was done by the methods of Nilehn [6], and serotyping by the methods of Winblad et al. [9].

As shown in Table 1, out of the 115 speci-
mens of cow's intestinal contents, 9 specimens (7.9%) proved positive to \( Y.e. \), and out of 61 specimens of beef meat, 15 specimens (24.6%) were positive to \( Y.e. \). The materials of enrichment culture with selenite medium tended to lower the positive rate of \( Y.e. \), i.e. none of these materials was found to positive, except a single specimen of beef meat. In contrast, the materials of enrichment culture with P.B.S. enhanced positive rate, especially in the case of beef meat. Namely, 9 specimens of the cow's intestinal contents and 15 specimens of the beef meat proved positive (including one specimen from the selenite medium). As shown in Table 1, serologically the isolates from the cow's intestinal contents consisted of O-4, O-5, O-7, O-10, O-12, O-14, O-22, O-27, O-31 and untypable strains, while the isolates from the beef meat gave O-3, O-5, O-6, O-7, O-8, O-13, O-15, O-22, O-30, O-31 and untypable strains. It has been reported generally that the human type of \( Y.e. \) could not be isolated from the animals except pig.

In our studies, however, the human type of \( Y.e. \) (O-5) could be isolated also from the cow's intestinal contents so that it was possible to assume that cows might be the carrier of human type \( Y.e. \), which might also be found in other animals, but the isolation rate of \( Y.e. \), whether high or low, might depend on the quantity of \( Y.e. \) in the reservoir of animal species. Isolates of \( Y.e. \) from beef meat varied in O-group, moreover these isolates from both materials were not consistent with each other in O-group except for O-5, O-7, O-22 and O-31 types. This fact suggests that the beef meat is likely to be contaminated by the contaminants identical with \( Y.e. \), that is, by the intestinal contents of animals at the slaughterhouse or by the other contaminants of environments during the transportation of beef from slaughterhouse to butcher.

On the other hand, cow's intestinal contents were sampled directly from cow's intestine as soon as possible after slaughtering. As mentioned, groups O-3, O-5 and O-9 organisms are the strains involving in human infection, and it is said that O-8 group is isolated from the person infected with \( Y.e. \) in U.S.A. [11]. The beef meat contaminated with O-3, O-5 and O-8 groups seems to be the very cause of sporadic or mass poisoning outbreak, especially, because the wide use of refrigerators in many houses would enhance the proliferation of \( Y.e. \) in contrast to other bacteria.

Therefore, it is to be noted that out of the 11 specimens collected on the sixth of March, 1974, ten specimens proved to be positive of \( Y.e. \). Anvonen [1] has stated that human types of \( Y.e. \) have rarely been encountered in animals with the exception of pigs.

However, in our findings only one out of 115 cow's intestinal specimens yielded human type of \( Y.e. \), and this fact seemed to prove that the cows could also serve as reservoir of \( Y.e. \) of human type.

Acknowledgments: The authors are much indebted to Assistant Professor Misao Tsukokura, Department of Veterinary Microbiology, Faculty of Agriculture, Tottori University, for valuable advices throughout this
work and painstaking proof reading of the paper.

References