

MICROBIAL ECOLOGY

Influence of intestinal anaerobes and organic acids on the growth of enterohaemorrhagic *Escherichia coli* O157:H7

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A suspension of human faeces (FS) and its anaerobic culture (FC), bacterial metabolic products and organic acids were examined for inhibitory effects on growth and verotoxin 2 (VT2) production of *Escherichia coli* O157:H7 *in vitro*. FS and FC showed a marked inhibitory activity to growth and production of VT2 by *E. coli* O157:H7 under anaerobic conditions. They may have both bacteriostatic and bactericidal effects on *E. coli* O157. The growth of *E. coli* O157 was markedly suppressed by acetic, propionic and butyric acids compared with hydrochloric acid and lactic acid at concentrations between 25 mM and 40 mM, being proportional to the pH values. At pH 5.5, 40 mM of short-chain fatty acids (SCFAs) almost completely inhibited the growth of *E. coli* O157. SCFAs markedly inhibited the growth of *E. coli* O157 at pH 6.0 rather than pH 7.0. Propionic acid is likely to be more suppressive to *E. coli* than acetic and butyric acids. The production of VT2 was approximately proportional to the growth of *E. coli* O157. However, incubation for 24 h *in vitro* showed that the growth and VT2 production of *E. coli* O157 decreased but were not completely inhibited at pH 6.5 and 7.0 in a mixture of acetic, propionic and butyric acids at a physiological concentration (110 mM, 60:25:25, respectively, in molar ratio). It is probable that the colonic microflora could contribute to a reduction of *E. coli* O157:H7 infections *via* the activation of intestinal fermentation by dietary manipulation or something similar to give pH 6.0 or <6.0 and that factors such as age, chemical therapy and body condition, which have effects on the balance of the intestinal microflora, would be associated with the incidence rates of *E. coli* O157 infections.

Introduction

In 1996, large outbreaks of enterohaemorrhagic *Escherichia coli* O157:H7 disease occurred in Japan. According to the Annual Report of Food Poisoning Outbreaks in 1996 (Welfare Ministry of Japan), the prevalence of *E. coli* O157:H7 infections in five school outbreaks ranged from 15.7 to 50.3% in children who ate school lunches. The prevalence rates in two homes for the elderly were 11.5% and 18.5%. However, in the same year, the prevalence rate for *E. coli* O157 infection in a company outbreak was only 1.5% in the staff who ate canteen food. Adults, excluding elderly individuals, are considered to be more resistant to *E. coli* O157 infection than children. Other reported

risk factors, apart from being young or elderly, include recent antimicrobial therapy and previous gastrectomy [1]. These factors are considered to be associated with changes in the composition of the intestinal microflora, or with reduced activity of the microflora, which provides protection against bacterial infections of the intestine. The factors associated with this protective activity are pH, redox potential and fermentation products such as acetic, propionic, butyric and lactic acids [2–5]. The present study was undertaken to examine the effect of human faecal mixed cultures and organic acids on the production of verocytotoxin 2 (VT2) and the growth of *E. coli* O157:H7 *in vitro*.

Materials and methods

Bacterial strains and culture media

E. coli O157:H7 strain HYM, a human isolate from Hiroshima, Japan, was kindly provided by Dr K.

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