At the age of one week Swedish infants with atopic eczema have less diverse intestinal microbiota compared to non-atopic infants

C. Karlsson\textsuperscript{1}, M. Wang\textsuperscript{1}, C. Olsson\textsuperscript{2}, G. Molin\textsuperscript{3}, A. Wold\textsuperscript{4}, B. Hesselmar\textsuperscript{5}, R. Saalman\textsuperscript{6}, I.-L. Strannegård\textsuperscript{7}, N. Åberg\textsuperscript{8} and S. Ahrné\textsuperscript{9}

\textsuperscript{1}Laboratory of Food Hygiene, Department of Food Technology, Engineering and Nutrition, P.O. Box 124, Lund SE-22100, Sweden
\textsuperscript{2}Department of Surgery, Universitetssjukhuset MAS, SE-20502, Malmö, Sweden
\textsuperscript{3}Department of Clinical Bacteriology, Gothenburg University, SE-41346 Göteborg, Sweden
\textsuperscript{4}Department of Paediatrics, Gothenburg University, SE-41685 Göteborg, Sweden

INTRODUCTION

The prevalence of allergy is rising, especially in developed countries. Before birth the human gastrointestinal tract is sterile and the immune system is not fully developed. The main part of our immune system is located in the gastrointestinal lymphoid tissue, and the intestinal microbiota is considered important for a correct maturation of the immune system in children. Thus, it is of great interest to investigate how the composition differs between healthy individuals and patients with immunological diseases.

AIMS

To analyse differences in the composition of the dominating intestinal microbiota in Swedish non-atopic and atopic infants (diagnosed at 18 months), at the age of one week.

To test if Terminal Restriction Fragment Length Polymorphism (T-RFLP) of the 16S rRNA gene is a suitable method for analysing microbial diversity.

RESULTS

The microbiota of atopic infants generated a lower number of peaks compared to that of non-atopic infants after digestion with \textit{Alu} I (\textit{P}=0.05), Figure B and C.

The prevalence of allergy is rising, especially in developed countries. Before birth the human gastrointestinal tract is sterile and the immune system is not fully developed. The main part of our immune system is located in the gastrointestinal lymphoid tissue, and the intestinal microbiota is considered important for a correct maturation of the immune system in children. Thus, it is of great interest to investigate how the composition differs between healthy individuals and patients with immunological diseases.

Material and Methods

Since many species are unculturable, molecular genetic methods are useful when analysing the complex intestinal microbiota. In T-RFLP the 5'-end of the universal forward primer was fluorescently labelled with Cy5, Figure A. In this study, eight non-atopic and eight atopic infants were enrolled. The parents collected stool samples at home, according to a standardized procedure. The faecal samples were stored under anaerobic conditions and refrigerated until delivered to the laboratory.

Bacteria from faecal sample

DNA extraction

PCR with universal primers in which one is fluorescently labelled

Detection of labelled fragments, information transferred to a computer and are presented by peaks

Separation of fragments in a polyacrylamide gel

Digestion, by restriction endonucleases, of PCR products gives only fluorescent label on terminal fragments

Figure A. Schematic view of Terminal Restriction Fragment Length Polymorphism.

MATERIAL AND METHODS

Since many species are unculturable, molecular genetic methods are useful when analysing the complex intestinal microbiota. In T-RFLP the 5'-end of the universal forward primer was fluorescently labelled with Cy5, Figure A. In this study, eight non-atopic and eight atopic infants were enrolled. The parents collected stool samples at home, according to a standardized procedure. The faecal samples were stored under anaerobic conditions and refrigerated until delivered to the laboratory.

MATERIAL AND METHODS

Since many species are unculturable, molecular genetic methods are useful when analysing the complex intestinal microbiota. In T-RFLP the 5'-end of the universal forward primer was fluorescently labelled with Cy5, Figure A. In this study, eight non-atopic and eight atopic infants were enrolled. The parents collected stool samples at home, according to a standardized procedure. The faecal samples were stored under anaerobic conditions and refrigerated until delivered to the laboratory.

CONCLUSIONS

High bacterial diversity might be one important parameter for not developing atopic eczema.

T-RFLP was a suitable method for analysing the bacterial diversity of intestinal microbiota in infants.