

3 Title: Quantitative Changes in Fecal Bacterial Flora

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Objective To quantitate the bacterial flora of individuals with and without diarrhea and to attempt to relate changes of bowel flora to disease. Additional studies were made on the influence of antimicrobial agents administered in therapeutic doses to individuals without diarrhea.

Description Fecal specimens were obtained from inpatients with acute diarrhea at Children's Hospital and from SMRL personnel. Processing the specimens for aerobic and anaerobic incubation usually took one hour. Serial 10-fold dilutions and plate counts were made on 10 media each designed to favor certain bacterial genera. In those instances when the same organism grew on more than one medium, the highest count was recorded. Routine bacteriologic and serologic procedures were utilized for final identification of organisms.

Progress During the period covered by this report daily specimens were taken from individuals with acute diarrhea with the intent of relating flora changes to clinical improvement. For this study specimens taken during the first 2 days of hospitalization were regarded as "acute" and subsequent specimens were considered "convalescent". Certain species of bacteria were recovered from almost every specimen. These included obligately anaerobic gram negative bacilli, coliforms, lactobacilli, alpha hemolytic streptococci, Streptococcus fecalis and yeasts. The ranks of numerical prevalence of organisms recovered are presented in Table 10. In both categories of specimens coliforms and obligately anaerobic gram negative bacilli respectively composed the largest components of the aerobic and anaerobic flora (10^8 – 10^9 per gram of wet feces). The mean count of alpha hemolytic streptococci in "acute" specimens was 3 logs greater than in "convalescent" specimens while the converse was true for St. fecalis. Clostridia and Proteus spp were rarely recovered from these specimens.

The effect of oral neomycin on the bowel flora of adults was studied. Three Thai and 3 U.S. adult males with no apparent illness volunteered for this study. Bacterial counts of stools relative to taking 1 gm/day oral neomycin sulfate were noted. Few changes were noted in the stools of the Thais. There were no appreciable decreases in either group except that one had 10^2 or greater increases of coliforms, lactobacilli, gram negative anaerobic rods, Staphylococcus epidermidis and alpha hemolytic streptococci. No consistent changes occurred in the specimens from U.S. personnel. One developed a moderate diarrhea which started while taking neomycin and continued throughout the remainder of the study. This was coincident with an abrupt increase in the numbers of Proteus mirabilis in the stool. The same organism appeared in the stool of a second American in cultures taken 8–10 days after starting oral neomycin. This increase was accompanied by a mild diarrhea which persisted for 3 days.

Changes in the numbers of neomycin-resistant organisms in the same specimens were determined by inoculating the same dilutions onto plates containing 5 mcg/ml neomycin sulfate. While there was no consistent pattern there were many instances of increases of neomycin-resistant organisms which persisted for at least 10 days after cessation of oral neomycin. Neomycin-resistant coliforms increased in all individuals. Neomycin-resistant St. fecalis increased only in the Americans and the Proteus mirabilis which emerged in 2 of the Americans was neomycin-resistant. There were no examples of overgrowth of yeasts

and only one person had an increase of neomycin-resistant Staphylococcus aureus. It is concluded that this dosage of neomycin did not cause appreciable imbalance of bowel flora. However the possibility of P. mirabilis being partially responsible for diarrhea in two instances cannot be discounted.

Summary Qualitative and quantitative studies were carried out on the microflora of the feces of Thai children during the acute and convalescent phases of diarrhea. Although great variations were noted in the numbers of bacteria in most patients, coliforms were usually the predominant aerobes and gram negative anaerobic bacilli the predominant anaerobes. Feces of patients, with diarrhea tended to contain larger numbers of alpha hemolytic streptococci than did feces from convalescent patients. Low doses of oral neomycin in normal adult Thai and U.S. males resulted in few changes of their fecal flora except that some neomycin-resistant organisms, notably P. mirabilis emerged and persisted for at least 10 days after cessation of therapy.