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EFFECT OF BETA-LACTAM ANTIBIOTICS ADMINISTERED PERORALLY ON HUMAN INTESTINAL MICROFLORA

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Oral kullanılan beta-laktam antibiyotiklerin insan barsak mikroflorası üzerine etkisi.

INTRODUCTION

The composition of the normal human intestinal microflora is stable. However, this ecosystem can be disrupted by different factors such as antimicrobial treatment, diet, pathological processes and surgical procedures. The most common cause of disturbances in the normal microflora is the administration of antimicrobial agents. When the normal flora is reduced during treatment, resistance to microbial colonization is lowered which may induce several ecological changes. One is overgrowth of already resistant microorganisms such as *Candida* species which may lead to systemic infections in immunocompromised patients and *Clostridium difficile* which may cause diarrhoea/colitis. A second effect is the development of resistance among the bacteria in the normal flora and a third consequence is transfer of resistance factors between the bacteria in the intestinal tract. Different factors influence the extent to which an antimicrobial agent can change the normal intestinal microflora. The incomplete absorption of antimicrobial agents perorally administered is an important factor. Poorly absorbed agents may reach the large intestine in active form where they suppress susceptible bacteria and disturb the ecological balance. Antimicrobial agents that are secreted from the intestinal mucosa or in the bile may also have an impact on the intestinal microflora.

This paper reviews the effect on the ecological balance of the intestinal human microflora after oral administration of different beta-lactam antibiotics. All reviewed studies have been published in international journals.

IMPACT OF PENICILLINS ON INTESTINAL MICROFLORA

Phenoxymethylpenicillin. Heimdahl and Nord (4) studied the impact of phenoxymethylpenicillin on the intestinal microflora in volunteers. Ten subjects received 800 mg phenoxymethyl penicillin capsules bid for 7 days. No changes in the intestinal aerobic microflora were observed. No antibiotic activity was noticed in the faecal samples during the observation period (Table).

Ampicillin. The influence of peroral daily administration of 1 to 3 g ampicillin on the intestinal microflora in 10 volunteers during 5 days was investigated by Knothe and Wiedemann (8). A significant reduction in the numbers of *Escherichia coli,* enterococci, bifidobacteria, and anaerobic gram-negative rods was observed and subsequently increased numbers of ampicillin-resistant *Citrobacter, Klebsiella,* and *Proteus* species were isolated (Table).

Leigh (10) investigated the impact of peroral administration of ampicillin in a dosage of 500 mg bid for 5 days given to 10 volunteers. Changes in the total numbers of aerobic bacteria and *Candida* species were observed and 5 volunteers developed diarrhoea which lasted for 1-2 days (Table).

Amoxicillin. The impact of amoxicillin in a dosage of 2 g/day for at least 15 days on the intestinal microflora in 8 patients with respiratory tract infections was studied by Gipponi et al. (3). A reduced number of microorganisms was observed in four of the patients. Th-

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Table. Impact of perorally administered beta-lactam antibiotics on intestinal microflora.

Agent	Dose mg/day	Days of administration	Number of patients	Impact on Enterobacteria	Impact on Enterococci	Impact on Anaerobic bacteria	Overgrowth of resistant strains	Reference
Phenoxymethyl-penicillin	800x2	7	10	–	–	–	–	Heimdahl & Nord 1979
Ampicillin	1000-3000	5	10	↓↓	↓↓	↓↓	+	Knothe & Wiedemann 1965
	500x3	5	10	↑	–	–	+	Leigh 1979
Amoxicillin	2000	≥15	8	↓	↓	↓	+	Gipponi et al. 1985
	250x3	5	10	↑	–	–	–	Leigh 1979
	500x3	7	6	↑	–	–	+	Mittermayer 1983
Bacampicillin	400x3	7	12	–	–	↓	–	Heimdahl et al. 1979
	1600	≥15	8	–	–	–	–	Gipponi et al. 1985
Pivampicillin	700x4	3	10	–	–	–	+	Knothe & Lembke 1973
Talampicillin	250x4	2	10	↑	–	–	+	Leigh et al. 1976
	250x3	5	10	↑	–	–	–	Leigh 1979
Pivmecillinam	600x4	7	10	↓↓	↑	↓	+	Knothe 1976
	400x3	7	5	↓	↑	↓	+	Knothe 1976
Cefaclor	250x3	14	6	–	–	–	+	Finegold et al. 1987
	250x3	7	10	–	–	–	–	Nord et al. 1987
Cephrocile	500x2	8	12	↓	↑	–	+	Lode et al. 1990
Cefixime	400	14	6	↓	↑	↓↓	+	Finegold et al. 1987
	200x2	7	10	↓↓	↑	↓↓	+	Nord et al. 1988
Cephradine	1000x2	7	6	–	–	–	–	Brumfitt et al. 1986
Amoxicillin/	500/125x3	7	6	↑	–	–	+	Mittermayer 1983
Clavulanic acid	27.5mg/kgx4	10-11	11	–	–	–	+	Lambert-Zechovsky et al. 1984
	500 / 250x3	3	6	–	–	–	–	Wise et al. 1984
	187.5x3 (2/1)[b]	5	4	–	–	–	+	Motohiro et al. 1985
	375x3 (2/1)[b]	5	4	↑	↓	–	+	Motohiro et al. 1985
	250/125x3	7	6	–	↓	–	+	Brumfitt et al. 1986

↓↓ strong suppression >4 $\log_{10}$ CFU/g faeces; ↓ mild to moderate suppression 2-4 $\log_{10}$ CFU/g faeces; ↑ increase in number of microorganisms during therapy; - no significant chang; [a]decrease in number of anaerobes were seen in 6 subjects receiving bacampicillin as syrup; [b] ratio amoxicillin/clavulanic acid.

ree patients had increased levels of *Candida* and one patient increased levels of enterococci (Table).

Ten healthy volunteers received 250 mg amoxicillin tid perorally for 5 days and the impact on the intestinal microflora was investigated (10). Four volunteers showed a significant increase in the aerobic microflora, but none of the volunteers developed diarrhoea (Table).

Bacampicillin. The effect of bacampicillin on the colon microflora was investigated by Heimdahl et al. (5). Bacampicillin was given as tablets or syrup in doses of 400 mg tid for 7 days to 12 subjects. No changes in the colon microflora were observed in the volunteers receiving tablets, while there was a decrease in the numbers of anaerobic bacteria in those volunteers taking syrup. No ampicillin was detected in the faecal samples during the investigation period. No increased resistance to ampicillin was observed in the colon microflora (Table).

Gipponi et al (3) investigated the impact of bacampicillin on the intestinal microflora in 8 patients receiving 1.6 g daily for at least 15 days. Moderate microbial changes were observed in 2 of the patients. No *Clostridium difficile* strains or toxins were isolated (Table).

Pivampicillin. Knothe and Lembke (7) investigated the impact of pivampicillin in a dose of 700 mg qid for 3 days on the intestinal microflora of 10 volunteers. Pivampicillin caused only minor changes in the flora during and after administration. Thus an increase in the numbers of *E.coli* was seen in 7 of the 10 subjects and numbers of *Candida* increased in 3 volunteers (Table).

Talampicillin. Leigh et al. (11) investigated the impact of talampicillin administration on the intestinal microflora. Ten healthy volunteers took 250 mg talampicillin qid for 2 days. Four subjects had minor changes in the aerobic and anaerobic microflora, while 5 subjects had a considerable increase in the numbers of aerobic bacteria and 2 subjects had a decrease in the numbers of anaerobic bacteria. Two volunteers developed mild diarrhoea (Table).

Leigh (10) studied the impact of talampicillin given perorally on the intestinal microflora in 10 volunteers at a dosage of 250 mg tid for 5 days. Six volunteers showed changes in the aerobic intestinal microflora and 1 volunteer developed diarrhoea (Table).

Pivmecillinam. Knothe (6) compared the influence on the intestinal microflora of the administration of pivmecillinam 600 mg qid or 400 mg tid for 7 days to two parallel groups of 10 and 5 volunteers, respectively. There was a marked reduction in the number of *E.coli,* lactobacilli, and *Bacteroides* and an increase in the number of enterococci. The changes were more pronounced after the higher dose of pivmecillinam (Table).

IMPACT OF CEPHALOSPORINS ON INTESTINAL MICROFLORA

Cefaclor. The impact of cefaclor on the intestinal microflora of 6 volunteers receiving cefaclor orally in a dosage of 250 mg tid for 14 days was studied by Finegold et al. (2). No decrease in *E.coli* was seen and there was little change in counts of enterococci. On the other hand, there were 13 new strains of enterobacteria and 2 new strains of *Staphylococcus aureus* in the intestinal microflora during cefaclor treatment. In the anaerobic flora only bifidobacteria were eliminated in 2 subjects. Three subjects were colonized by *C.difficile,* but none developed pseudomembranous colitis. There was no evidence of development of resistance or β-lactamase induction (Table).

In another study, cefaclor was given orally in doses of 250 mg every 8 h for 7 days to 10 volunteers (15). Faecal specimens were taken up to 16 days for cultivation of aerobic and anaerobic microorganisms and for assay of cefaclor. Cefaclor was not detected in faeces. The aerobic intestinal microflora was unchanged during and after cefaclor administration while a minor impact on the anaerobic intestinal microflora was observed. The anaerobic intestinal flora returned to its normal state within 1 week. No new colonization with cefaclor

resistant microorganisms was observed and no side effects were registered during the investigation period (Table).

Cephrocile. The pharmacokinetics of cephrocile and the influence on faecal flora were determined in 12 volunteers after oral administration of 500 mg every 12 h over an 8-day period (12). Bioavailability parameters (AUC_{tot}, C_{max}, urinary recovery) indicated an excellent absorption. No cumulation over the 8 day period was registered. The analysis of the faecal flora showed an ecological impact of cephrocile on the intestinal microflora such as a moderate decrease of enterobacteria and a slight increase of enterococci and staphylococci as well as *Bacteroides* during the study. *C.difficile* was detected in 3 volunteers during the cephrocile administration but no toxins could be identified. The number of all bacterial species was normalized within 4 days after the administration period (Table).

Cefixime. Finegold et al. (2) investigated the impact of 400 mg perorally administered cefixime daily for 14 days on the bowel flora in 6 healthy male subjects. A significant decrease in the numbers of *E.coli* occurred. In 4 of the subjects enterococci increased 3 logs or more. The impact on the anaerobic microflora was pronounced. Bifidobacteria disappeared from 2 of 5 subjects, clostridia from 3 of 4 subjects, and *B.fragilis* from 1 subject. Ingrowth of *C.difficile* was noted in 4 subjects but no subject developed severe gastrointestinal symptoms. No development of resistance among the aerobic or anaerobic bacteria was seen and there was no evidence of β-lactamase induction in the microflora (Table).

The ecological effects on the normal intestinal microflora after cefixime tablets in doses of 200 mg twice daily for 7 days were studied in 10 healthy volunteers (16). Stool specimens were collected before and 2, 4, 7, 14 and 21 days after start of treatment. The concentrations of cefixime in faeces increased during treatment. One subject had detectable concentrations in faeces on day 2, 3 subjects on day 4 and 8 subjects on day 7 in the order of 237-912 mg/kg faeces. There was a marked decrease in the numbers of streptococci and *E.coli* and an increase in the numbers of enterococci during the administration of cefixime. In the anaerobic microflora, the numbers of cocci, clostridia and *Bacteroides* were suppressed while there were minor changes in the numbers of bifidobacteria. *C.difficile* was isolated in 5 subjects on day 7 but cytotoxin was only detected in 1 subject. The intestinal microflora was normalized within 2 weeks after treatment had stopped (Table).

IMPACT OF BETA-LACTAMASE INHIBITORS ON INTESTINAL MICROFLORA

Amoxicillin plus clavulanic acid. Mittermayer (13) compared the effect of amoxicillin alone with amoxicillin plus clavulanic acid on the intestinal microflora. Six volunteers received 500 mg amoxicillin and 6 volunteers 500 mg amoxicillin plus 125 mg clavulanic acid tid for 7 days. In both groups of volunteers the number of amoxicillin-resistant enterobacteria increased significantly. Selection of amoxicillin-clavulanic acid-resistant enterobacteria occurred only in those volunteers receiving amoxicillin-clavulanic acid. The total number of aerobic and anaerobic bacteria was not affected by either treatment. No significant changes in the number of *Pseudomonas, Bacteroides,* or yeasts were observed during or after treatment (Table).

Lambert-Zechovsky et al. (9) investigated the effect of amoxicillin-clavulanic acid on the intestinal flora in 11 children. The antibiotic combination was given orally to 7 patients and parenterally to 4 patients in a dosage of 27.5 mg/kg body weight qid over 10 to 11 days. The impact on the intestinal microflora was more pronounced in those patients receiving amoxicillin-clavulanic acid perorally than parenterally. After treatment had stopped, an increase in amoxicillin-resistant *E.coli* strains was observed and an overgrowth of *Klebsiella* strains occurred. There were no other significant changes in the numbers of aerobic and anaero-

bic bacteria (Table).

Wise et al. (17) investigated the impact of cefuroxime axetil and amoxicillin-clavulanic acid, respectively, on the faecal microflora in 6 volunteers. The subjects first received 600 mg cefuroxime axetil 8 hourly for 10 doses and then 6 weeks later 500 mg amoxicillin and 250 mg clavulanic acid 8 hourly for 10 doses. Cefuroxime caused a decrease in the number of enterobacteria in 3 volunteers who developed diarrhoea. In 2 of these 3 volunteers, the enterococci were also significantly suppressed. The counts of *Candida* species increased significantly in the 3 subjects with diarrhoea. *Bacteroides,* peptococci, and peptostreptococci decreased significantly in 3 volunteers while no significant changes in the number of clostridia were observed. When the subjects receieved amoxicillin-clavulanic acid, no major changes in the intestinal flora were observed. Thus no significant changes in the number of enterobacteria, staphylococci, and clostridia were registered, while minor changes in the number of enterococci, peptostreptococci, peptococci, and candida took place. One volunteer taking amoxicillin-clavulanic acid developed diarrhoea (Table).

The impact of amoxicillin-clavulanic acid on the faecal microflora in 8 subjects was studied by Motohiro et al. (14). Four volunteers received 187.5 mg tablets (ratio amoxicillin - clavulanic acid 2:1) tid for 5 days. The number of *E.coli* strains did not change while there was an increase in the numbers of *Klebsiella* strains. The staphylococci disappeared in 3 of 4 subjects. There was an increase in the number of enterococci after the administration had stopped. No effect on the anaerobic part of the microflora was observed. In the volunteers receiving 375 mg tablets (ratio amoxicillin-clavulanic acid 2:1) tid for 5 days, enterobacteria increased significantly, while the numbers of staphylococci dropped to undetectable levels. In 2 volunteers the enterococci disappeared during treatment. The anaerobic intestinal microflora was not affected by the administration of amoxicillin-clavulanic acid. The faecal concentrations of amoxicillin were between 12.9 and 44.4 mg/kg faeces in volunteers receiving the dose of 375 mg tid. One subject in each dosage group developed diarrhoea (Table).

In another study, Brumfitt et al. (1) determined the side effects and changes in the faecal microflora composition in volunteers given cephradine or amoxicillin-clavulanic acid. Six subjects took 1 g cephradine bid and another 6 subjects 250 mg amoxicillin plus 125 mg clavulanic acid tid for 7 days. The staphylococci were virtually eliminated in both treatment groups while streptococci decreased significantly in the group receiving amoxicillin-clavulanic acid treatment. No other significant changes were observed. The total number of enterobacteria, enterococci, and anaerobic bacteria was unaffected. No overgrowth of yeasts was noticed. Some amoxicillin-resistant enterobacteria were isolated after treatment in both groups, while cephradine or amoxicillin-clavulanic acid-resistant enterobacteria were rarely recovered. No cephradine or amoxicillin activity was found in faecal samples taken immediately after the end of treatment (Table).

CONCLUSIONS

It has become evident with the introduction of new beta-lactam antibiotics that their suppressive activities are directed not only against pathogenic bacteria but also against the host's normal microflora. The changes in the intestinal microflora may result in overgrowth of bacteria and yeasts, proliferation of antibiotic resistant microorganisms and increased susceptibility to colonization by new microorganisms.

It has been stated that narrow beta-lactam agents should always be used in preference to broad beta-lactam agents in order to avoid these ecological problems. This statement is an oversimplification and other factors such as mode of excretion, activity, inactivation and development of resistance should also be considered. These ecological impacts are often difficult to predict when beta-lactam agents are developed, and the clinical studies of new agents should always include an investigation of their effects on the intestinal microflora.

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