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THE INFLUENCE OF GENERAL ANESTHESIA ON ORAL FLORA

Genel Anestezinin Oral Floraya Etkisi

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ARAŞTIRMA

ÖZET

Amaç: Alt solunum yolu enfeksiyonları sıklıkla oral flora bakterilerinden kaynaklanmaktadır. Entübasyon oral flora bakterilerinin alt solunum yoluna taşınması açısından risk faktörü kabül edilir. Çalışmamızda genel anestezinin bu olaydaki yerini belirlemek amacıyla oral florada gözlenen değişiklikler araştırılmıştır.

Hastalar ve Yöntemler: İntratekal genel anestezi ile ameliyat olan ve 53'ü proflaktik antibiyotik kullanan 90 hastanın (ASA I-II), (43 kadın, ortalama yaş 30. 53 + 1. 98 ve 47 erkek, ortama yaş 35.53 ± 2.61) ameliyat öncesi ve ameliyat sonrasında dil arkasından alınan örneklerde, oral flora bakterileri kültür yöntemiyle araştırılmıştır. Bakteriyel kolonizasyon log 10 tabanına göre belirlenmiştir.

Bulgular: Genel anestezi alan hastalarda ameliyat sonrası oral flora bakterilerinde anlamlı derecede azalma olmuştur. Oral streptokok kolonizasyonu cinsiyete göre farklılık göstermesine rağmen penisiline duyarlı MIC konsantrasyonuna sahip oral streptokok oranı kadınlarda anlamlı düşük bulunmuştur. Ameliyat öncesinde proflaktik olarak tek doz cefuroxime axetil alımı total aerob ve total anaerob bakteriyel kolonizasyonda anlamlı azalmaya neden olmuş fakat oral streptokok kolonizasyonu etkilenmemiştir. Preoperatif anaerob bakteri kolonizasyonu ve postoperatif oral streptokok kolonizasyonu sigara içenlerde içmeyenlere göre anlamlı azalma göstermiştir.

Sonuç: Genel anestezinin oral floradaki bakteriyel kolonizasyonu azalttığı, proflaktik antibiyotik kullanımının bu azalmaya yardımcı olduğu gözlenmiştir. Sigaranın oral anaerob bakteriyel kolonizasyonda yaptığı azalmanın nikotinin olası antibakteriyel etkinliğine bağlı olabileceği düşünülmüştür. Tek doz 1 gr cefuroxime axetil kullanımının, oral streptokoklara etkisiz olması nedeniyle, oral cerrahi girişimler öncesinde yeterli proflaktik değere sahip olmadığı görülmüştür.

Anahtar kelimeler: Oral flora, genel anestezi, bakteriyel kolonizasyon.

RESEARCH

ABSTRACT

Aim: Lower respiratory tract infections are frequently originated from oral flora bacteriae. Entubation is accepted as a risk factor for transmisson of oral flora bacteria to lower respiratory tract. In this study, changes observed in the oral flora were investigated to determine the part of general anesthesia in this situation.

Patients and Methods: Oral flora bacteria were investigated in samples taken from the posterior tongue pre and post operation from 90 patients (ASA I-II), (43 women, median age 30.53± 1.98 and 47 men, median age 35.53±2.61) operated with intratracheal general anesthesia and 53 of them had profilactic antbiotics. Bacterial colonization were evaluated according to log 10 base.

Results: Significant reduction was seen in oral flora bacteria after operation in patients who had general anesthesia. Despite oral streptococcus colonization did not show difference between sexes. Oral streptococcus ratio which had penicilin sensitive MIC concentrations was found significantly low in women. Preoperative profilactic single dose cefuroxime axetil intake caused signigicant reduction of total aerob and anaerob bacterial colonization but oral streptococcus colonization was not affected. Preoperative anaerob colonization and postoperative oral streptococcus colonization were showed significant reduction in cigarette smokers than non smokers.

Conclusion: It was observed that general anesthesia was reduced oral flora bacterial colonization and profilactic antibiotic management helped in this reduction. It was thought that reduction of oral anaerob bacterial colonization achieved by cigarette was due to possible antibacterial effect of nicotine. Because single dose 1 gr cefuroxime axetil management did not have effect on oral streptococci. It was considered that it did not have adequent profilactic value before oral surgery.

Key words: Oral flora, general anesthesia, bacterial colonization.

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INTRODUCTION

Community acquired or hospital acquired (nosocomial) bacterial pneumonia has been originated from resident and transient bacterial flora of oropharyngeal mucosa. Respiratory infections are of particular concern in hospitals and other health care facilities such as nursing homes, especially in intubated patients. These infections often prolong hospital stays, increase patient care costs, and cause significant morbidity and mortality (1).

It was determined that there was a relationship between developments of respiratory infection and bad oral hygiene. Streptococci have been important part of oral flora and may play role at the beginning and progression of pneumonia. For example viridians streptococci were found to be the cause of pneumonia in 4% of chronic obstructive pulmonary disease patients (2).

Anaerob bacteria in oral flora led to pneumonia in lower respiratory tract by means of salivary secretion (1) and were responsible for 30-40% of aspiration pneumonia, necrotizing pneumonia, or lung abscess (3).

Intubation greatly increases the risk of bacterial pneumonia because the endotracheal tube allows direct entry of colonized oropharyngeal bacteria into the lower respiratory tract (4). Determination of the changes in oral flora from which several infections were originated in patients undergoing general anesthesia with endotracheal intubation was aimed in this study.

PATIENTS AND METHODS

Subject

The changes in oral flora of 90 patients (ASAI-II) aged 6 to 78 yr old (43 female with mean age, 30.53+1.98 years and 47 male with mean age, 35.53+2.61 years) undergoing otorhinolaryngologic surgery were investigated in this study which was performed during 6 months (January-June) in 2006. The weight (kg) of patients in female was 64.23+11.44 and in male 79.04+10.20.

Anesthesia was induced with thiopental (5-7mg/kg) and midazolam (0.15 mg/kg), intravenously (IV). Vecuronium 0.1mg/kg IV was administered to facilitate tracheal intubation and 50% nitrous oxide in oxygen with 2% sevoflurane was used for maintenance. Thirty-seven percent of patients received prophylactic single dose of cefuroxime axetyl 1gr IV at premedication. No patients used dental prostheses and had carious tooth. Smoker patients had one pack-year for at least one year history.

Microbiology

Samples were taken from the dorsum of the tongue by rotating the swab approximately 360º and two swab specimens (preoperative and postoperative) for every patient were used. Samples were transported to laboratory in Shadler Anaerob Broth-2mL (Oxoid Ltd, Basingstoke, Hampshire, England) and studied in one hour. After being vortexed for 30 seconds tenfold serial dilutions (10^{-1} -10^{-5}) were prepared in sterile serum psychology solution 0.9 mL and 0.1 mL aliquots of the appropriate dilutions were inoculated onto both selective and non-selective media. Mitis Salivarius Agar (MSA) (Difco, Becton, Dickinson and Company, Spark, USA) supplemented with 0.1% potassium telluride (Sigma-Aldrich Co. Ltd, Poole, Dorset) was prepared for the growth of the oral streptococci (5). Mutans streptococci (*S. mutans* and *S. sobrinus*) were isolated from MSA with bacitracin 0.2 U/mL (Sigma-Aldrich Co. Ltd, Poole, Dorset) and sucrose 15%w/v (Sigma-Aldrich Co. Ltd, Poole, Dorset) (BMSA). The plates were incubated anaerobically at 37 ºC for 3 days (1). Shadler Anaerob Agar supplemented with 5% (v/v) defibrinated horse blood was prepared to determine the aerobic and anaerobic counts. The plates for the aerobic counts were incubated in air supplemented with 5% CO_2 at 37 ºC for 3 days. The other plates were incubated anaerobically in an anaerobic gas pack (90% nitrogen, 5% hydrogen, 5% carbon dioxide) at 37 ºC for 3 days.

Eight dilutions in mitis-salivarius agar (from 0.0625 U/mL to 8 U/mL) were prepared to detect the penicillin resistance of oral streptococci (□0.125= S and □4= R). For isolation resistant oral streptococci standard agar dilution antimicrobial susceptibility test approved by the National Committee for Clinical Laboratory Standards was applied. The strains which were sensible to penicillin with middle degree were accepted as resistant strains. A reference strain was *Enterococcus faecalis* (ATCC 29212) (6).

Statistics

All statistical analyses were performed with SPSS version 11.5. Viable counts were transformed $\log_{10}$ cfu; results are as the mean of the $\log_{10}$ cfu. The difference in number of preoperative and postoperative bacterial colony ($\log_{10}$ cfu) was assessed with paired *t* test. The interrelationship among the parameters including age, smoking and gender, and the difference in logarithmic values of bacterial colony at preoperative and postoperative period, were evaluated using independent simplest *t* test. The difference in number of preoperative and postoperative bacterial colony was compared with smoking, gender, surgery time, age and the usage of prophylactic antibiotic, using linear regression analysis.

RESULTS

The bacteria of oral flora were decreased significantly in patients undergoing general anesthesia (Table 1). There was no relationship between the differences in number of colony at preoperative and postoperative period, and smoking, gender, surgery time, age and the usage of

Table 1— Preoperative and Postoperative Bacterial Colonization log_{10} (Mean±SD)

Bacteria	Preoperative	Postoperative	P
Aerob bacteria (n: 90)	4.64±0.84	4.18±1.08	<0.001
Anaerob bacteria (n: 90)	4.53±0.82	4.31±0.87	0.001
Oral streptococci (n: 89)	3.90±0.68	3.58±0.92	<0.001
Mutans streptococci (n: 84)	3.03±0.78	2.70±0.90	<0.001

prophylactic antibiotic when compared using linear regression analysis.

Although the number of aerob and anaerob colonization decreased significantly preoperatively in patients who received prophylactic antibiotic when comparing with the patients who did not receive prophylactic antibiotic (p=0.01 and p=0.044, respectively) the significant decrease was only observed in aerob bacterial colonization postoperatively (p=0.025) (Table 2). Bacterial colonization in smokers was found to be significantly lower than in non-smokers preoperatively (p=0.047). Gender and age had no effect on oral aerob and anaerob bacterial colonization.

The number of total aerob bacteria in patients received prophylactic antibiotic decreased significantly more than the patients with no antibiotic usage after surgery with the duration of 120 minutes (p=0.003).

Even though there was no effect of smoking on oral streptococci preoperatively colonization of oral streptococci was found to be lower in smokers when compared with non-smokers postoperatively (p=0.01) (Table 3), but other parameters investigated did not affect colonization of oral streptococci.

Oral streptococci colonization consisting of sensible penicillin MIC (Minimal inhibitory concentrations) concentration was significantly lower in female when compared with male pre and postoperatively (p=0.028 and p=0.044, respectively) (Table 4).

The rate of resistant streptococci in patients received prophylactic antibiotic was significantly higher than the patients without prophylaxis. The usage of prophylactic antibiotic was determinative and the significant decrease in oral streptococci colonization consisting of resistance MIC concentration was observed in these patients (p=0.017).

When investigated the effects of gender, age, smoking and antibiotic prophylaxis on preoperative aerobic colonization number using linear regression analysis, the usage of antibiotic was found to be a determinant parameter, besides other parameters had effects on colonization number whereas gender had no effect (p<0.001). Similar results were obtained for preoperative anaerobic colonization number (p=0.002). Age, smoking and antibiotic prophylaxis affected postoperative anaerobic colonization number while no activity for these parameters on postoperative aerobic colonization.

Table 2— The Factors Affecting Aerob and Anaerob Bacterial Colonization log_{10} (Mean±SD)

	Aerob bacteria			Anaerob bacteria		
	Preop.	**Postop.**	**Preop-Postop**	**Preop.**	**Postop.**	**Preop-Postop**
Sex						
Female (n: 43)	4.63±0.85	4.22±1.11	0.41±0.59	4.54±0.88	4.20±0.15	0.34±0.57
Male (n: 47)	4.64±0.84	4.14±1.05	0.50±0.77	4.52±0.77	4.40±0.11	0.12±0.63
Smoking						
Yes (n: 23)	4.42±0.71	3.93±0.92	0.49±0.70	*4.25±0.73	4.06±0.78	0.19±0.77
No (n: 67)	4.71±0.88	4.27±1.11	0.44±0.69	4.63±0.83	4.39±0.89	0.24±0.55
Age (year)						
<35 (n: 48)	4.66±0.82	4.22±1.03	0.44±0.70	4.50±0.82	4.21±0.90	0.29±0.70
≥35 (n: 42)	4.61±0.87	4.14±1.13	0.48±0.68	4.57±0.83	4.41±0.82	0.15±0.49
Surgery time (min)						
<120 (n: 43)	4.70±0.81	4.21±1.01	0.49±0.72	4.47±0.82	4.32±0.77	0.16±0.60
≥120 (n: 47)	4.58±0.87	4.16±1.14	0.43±0.66	4.58±0.83	4.29±0.96	0.29±0.62
Prophylactic antibiotic						
Yes (n: 37)	4.28±0.81†	3.89±1.12†	0.40±0.59	4.32±0.81†	4.19±0.99	0.13±0.59
No (n: 53)	4.89±0.78	4.39±1.00	0.49±0.75	4.68±0.81	4.39±0.76	0.29±0.62

*Significantly lower when compared smoker and non-smoker (p<0.05)
†Significantly lower when compared the usage of prophylactic antibiotic and without prophylaxis (p<0.05)
Preop; preoperative, Postop; postoperative, Preop-Postop; the difference between preoperative and postoperative colony number.

Table 3— The Factors Affecting Colonization of Streptococci log_{10} (Mean±SD)

	Oral streptococci			Mutans streptococci		
	Preop.	**Postop.**	**Preop-Postop**	**Preop.**	**Postop.**	**Preop-Postop**
Sex						
Female	3.84±0.72	3.53±0.83	0.31±0.63	3.08±0.69	2.84±0.90	0.24±0.70
Male	3.95±0.64	3.62±1.00	0.33±0.75	3.98±0.86	2.56±0.89	0.42±0.85
Smoking						
Yes	3.71±0.84	*3.16±1.44	0.56±1.04	2.84±0.95	2.48±1.04	0.36±0.97
No	3.96±0.61	3.72±0.60	0.24±0.51	3.09±0.71	2.77±0.84	0.32±0.72
Age (year)						
<35	3.91±0.69	3.47±1.12	0.45±0.86	3.02±0.85	2.58±1.05	0.45±0.92
≥35	3.88±0.67	3.70±0.61	0.18±0.40	3.04±0.69	2.84±0.65	0.20±0.56
Surgery time (min)						
<120	3.85±0.73	3.51±1.01	0.34±0.63	3.01±0.86	2.74±0.79	0.27±0.72
≥120	3.94±0.64	3.63±0.84	0.31±0.75	3.05±0.71	2.66±0.99	0.39±0.84
Prophylactic antibiotic						
Yes	3.74±0.61	3.44±0.88	0.31±0.74	2.99±0.62	2.56±0.75	0.43±0.89
No	4.01±0.71	3.68±0.95	0.33±0.66	3.06±0.88	2.79±0.99	0.27±0.70

*Significantly lower when compared smokers and non smokers ($p<0.05$)
Preop; preoperative, Postop; postoperative, Preop-Postop; the difference between preoperative and postoperative colony number.

Whereas gender, age, smoking and antibiotic prophylaxis had no effect on preoperative oral streptococci and mutans streptococci colonization, smoking and antibiotic usage were found to be effective on postoperative oral streptococci colonization and smoking was found to be more serious determinant parameter (p=0.002).

DISCUSSION

Anesthesia and surgery have activity on immune response decreasing lymphocyte number impairing their function and decreasing serum immunoglobulin concentrations (7). Besides anesthetics may have local and systemic activity against infections. For example some anesthetics like midazolam and ketamine have dose-dependent inhibitory effect on mast cell exocytose. The impairment of mast cell function by anesthetics may influence the defense against infection (8). The decrease in salivary secretion but the increase in salivary Ig (secretary IgA, IgG, IgM) concentrations and antimicrobial factors (like amylase and lysozym) were observed postoperatively in one study related to the changes in oral cavity of patients undergoing general anesthesia (9). The number of salivary streptococci did not change whereas anaerobic bacteria decreased postoperatively in the same study. We found that the

Table 4— The Factors Affecting Oral Streptococci Colonization Consisting Sensitive Penicillin MIC Concentration of (≤0.125 U/mL) log_{10} (Mean±SD)

	Sensitive oral streptococci			Sensitive mutans streptococci		
	Preop.	**Postop.**	**Preop-Postop**	**Preop.**	**Postop.**	**Preop-Postop**
Sex						
Female	*1.21±0.61	*0.92±0.74	0.29±0.51	0.59±0.47	0.38±0.34	0.29±0.49
Male	1.82±0.96	1.51±0.92	0.31±0.39	0.92±0.55	0.78±0.71	0.09±0.85
Smoking						
Yes	1.43±0.46	0.99±0.64	0.43±0.68	0.86±0.32	0.40±0.33	0.47±0.47
No	1.57±0.99	1.32±0.95	0.25±0.32	0.70±0.61	0.71±0.69	0.03±0.78
Age (year)						
<35	1.53±1.13	1.19±1.08	0.34±0.42	0.66±0.55	0.67±0.65	0.01±0.92
≥35	1.53±0.51	1.27±0.65	0.26±0.47	0.83±0.51	0.54±0.56	0.35±0.43
Surgery time (min)						
<120	1.70±0.83	1.25±0.96	0.45±0.47	0.83±0.56	0.62±0.66	0.26±0.70
≥120	1.38±0.88	1.22±0.83	0.16±0.38	0.65±0.49	0.58±0.54	0.08±0.74
Prophylactic antibiotic						
Yes	1.22±0.84	0.97±0.91	0.25±0.47	0.69±0.60	0.63±0.76	0.12±0.78
No	1.78±0.81	1.44±0.82	0.34±0.43	0.80±0.48	0.58±0.48	0.23±0.68

*Significantly lower in female when compared with male ($p<0.05$).

colonization of aerob and anaerob bacteria, and oral streptococci decreased postoperatively (Table 1). This decrease in colonization of bacteria may be evaluated as a result of the increase in salivary Ig and antimicrobial factors.

Some anesthetics are well known inhibitors of the oxidative metabolism of various drugs like antibiotics and antifungal agents in the liver (10). Such interactions may induce adverse effects by elevating the plasma concentration of the interacted drugs (10). Especially midazolam increased the activity of some cephalosporin (11). We explained that more decrease in aerob bacterial colonization of patients receiving single dose of cefuroxime as prophylactic antibiotic when compared with the patients without prophylaxis, postoperatively was caused by the increase of cefuroxime activity with anesthetics (Table 2). The number of total aerob bacteria decreased depending on surgery time in patients received prophylactic antibiotic, and but this decrease was significant only for colonization of aerob bacteria in operations lasting more than 120 minutes. No activity of prophylactic antibiotic on colonization of anaerob bacteria postoperatively was commended that anesthetic agents did not increase further the effect of antibiotic on anaerob bacteria because of the reasons originated from affectivity spectrum of antibiotic.

Pneumonia or infective endocarditic caused by oral streptococci may occur after oral surgical interventions, that's why prophylactic antibiotic is advised before oral surgery. Although single dose of cefuroxime prophylaxis decreased the number of aerob and anaerob bacteria significantly, this decrease was not current for oral streptococci. But it is possible that repeated doses may be effective on endocarditic originated from streptococci instead of single dose, because the rate of oral streptococci that have resistant penicillin MIC concentration was significantly lower in patients received prophylactic antibiotic than in patients with no antibiotic. In other words, cefuroxime sensitize oral streptococci for antibacterial effect of penicillin. Contrarily, in a previous study, immediately after application of single dose amoxicillin 3 gm, streptococci became sensitive against antibacterial effect of penicillin and total number of streptococci decreased significantly (12). There may be differences related to antibiotic used, doses and application access.

Nicotine affected all oral streptococci with the exception of mutans streptococci (Table 3). We thought that nicotine increased the antibacterial activity of general anesthetics on oral streptococci because this difference occurred postoperatively not preoperatively. In some previous studies, no statistically significant difference in the prevalence of any of the bacteria was found between smokers and the non-smoker sub sample (13). In another study, smoking exerts little, if any, influence on the sub gingival occurrence of several of the bacteria most commonly associated with periodontal disease (14). Even though nicotine has antibacterial activity and has the potential to be used as a disinfectant the data are insufficient (15). The reason of decrease in preoperative colonization of total anaerob bacteria was thought as a result of antibacterial activity of nicotine especially on anaerob bacteria.

Oral or systemic biotransformation of some antibiotics shows difference depending on general anesthetics and sex (16). Oral streptococci colonization did not change depending on sex in our study (Table 3), but we thought that the reason of significant decrease in the rate of oral streptococci which had penicillin sensitive MIC concentration in female (Table 4) was to apply cefuroxime preoperatively with the doses of 1 gr not arranging for weight. Having lower mean weight of female when compared with male supported this comment.

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