Correlation between caries related bacteria in plaque and saliva in different age group children

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ABSTRACT

Background: Dental plaque contains bacteria that are both acidogenic and acidoduric. Different types of streptococci were identified in saliva. Although many bacterial subspecies have been shown to be associated with caries, streptococcus mutans is still believed to be the most important bacterium in the initiation of this disease while lactobacillus is correlated with the active caries episode. This study was conducted in order to estimate the correlation between caries related bacteria in plaque and saliva in different age group children.
Materials and methods: Fifty three children aged 3-10 years old were chosen for this study. Recording of dental caries was carried out by the dmfs index for primary teeth and DMFS index for the permanent teeth according to the criteria suggested by the WHO. One ml of unstimulated (resting) whole saliva was collected from the children using spitting method then diluted and applied on the surface of agar media specific for streptococcus mutans and lactobacilli growth. Dental plaque sample was taken from the buccal surface of the maxillary second primary molar by a clean toothpick and store in Epindorf tube which contain 1 ml. normal saline then inoculated in the same growth media that were used with the salivary samples. Colonies of the bacteria were counted with the aid of dissection microscope (15 X) on the basis of their characteristic morphology.
Results: Strong positive significant correlation was found between dmfs and ds components of the primary teeth, Positive results was found when correlating dmfs or ds with streptococcus mutans in dental plague while a negative correlation was found with lactobacilli. Negative correlation was found when correlating dmfs with streptococcus mutans in saliva while the relation is positive with lactobacilli. In dental plaque and in saliva there was a strong positive highly significant correlation between the DMF and the DS. Correlation coefficient between DMFS with the bacterial counts of the caries related microorganisms (streptococci and lactobacilli) in the dental plaque and in saliva revealed weak, negative not significant correlations.
Conclusion: The relation is not significant between the caries related microorganisms with each other in different media; either the dental plaque or saliva.
Key words: Correlation, streptococcus mutans, lactobacilli, saliva, plaque, dmfs, DMFS. (J Bagh Coll Dentistry 2012;24(3):140-144).

INTRODUCTION

Dental caries is a microbial disease of the calcified tissues of the teeth characterized by demineralization of the inorganic portion and destruction of the organic portion of the tooth [(1)]. Several interrelated factors such as: the tooth and saliva (host), microorganisms and cariogenic substrate [(2)].These factors interact in a certain period of time, causing an imbalance in the demineralization and remineralization between tooth surface and the adjacent plaque (biofilm)[(3)].
Plaque is a complex mixture of dense microbial elements enmeshed within a gel like matrix of bacterial polysaccharide, salivary proteins and cellular components of the oral mucosa [(4)].
Dental plaque contains bacteria that are both acidogenic and acidoduric. Although many bacterial subspecies have been shown to be associated with caries, streptococcus mutans is still believed to be the most important bacterium in the initiation of this disease while lactobacillus is correlated with the active caries episode [(2, 5)].

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Saliva has several critical roles in the caries process; it is excreted at different rates and with different constituents depending on the presence or absence of stimulatory factors and it helps to balance the caries process and has critical role in remineralization as it provides a stabilized supersaturated solution of calcium and phosphate ions as well as fluoride ions from extrinsic sources [(2)].Different types of streptococci were identified in saliva [(6)].
Different studies have shown a correlation between counts of streptococcus mutans in the oral cavity and both the prevalence and incidence of caries [(7, 8, 9, 10)].However, in other studies, no correlation has been found between the quantity of streptococcus mutans and the incidence of caries [(11, 12)]. On the other hand, some researchers have suggested the importance of streptococci other than streptococcus mutans in the generation of dental caries [(13)].Usually there are only few lactobacilli in saliva, their number increases if mutans streptococci start to colonize in the oral cavity, since they produce a favorable acid environment for lactobacilli, so the pH-value decreases [(14)]. Lactobacilli preferably settle in niches with a low pH-value and in the vicinity of plaque accumulation [(15)]. Koroluk et al and

Ziotopolus et al were found significant correlations between the plaque and salivary levels of mutans streptococci and caries experience in the primary teeth[16, 17].Concerning permanent teeth E. Bramilla et al found a statistically positive relationship between the levels of salivary streptococcus mutans and lactobacilli and both were significantly correlated to caries[18].

MATERIALS AND METHODS

Fifty three children aged 3-10 years old were chosen for this study, all of them were with carious lesion, attended the department of Pediatric and Preventive Dentistry, Baghdad teaching hospital for Dentistry, and neighboring primary schools and kindergartens. Recording of dental caries was carried out by the dmfs index for primary teeth and DMFS index for the permanent teeth according to the criteria suggested by the WHO in 1987[19], with no history of any systemic disease.

Salivary samples were collected in the morning between (10-11) a.m. at least one hour after breakfast, and then the children were asked to rinse out their mouths with water. The first mouthful of saliva was discarded, and then one ml unstimulated (resting) whole saliva was collected into small labeled plastic polyethylene tubes using spitting method for collection.

The following points were followed for collection whole saliva according to Fejerskov and Thylstrup [20].

1. The patient should not eat or drink (except water) one hour before saliva collection.
2. A pre – sampling period of one minute is recommended.
3. A fixed collection time (10-15 min. for unstimulated saliva) should be used.
4. The patient should sit in a relaxed position in an ordinary chair.
5. Samples containing blood should be discarded if chemical analyses of saliva are planned.

After collection of the saliva, it was diluted with normal saline in the bacteriology laboratory of Baghdad university college and then by using micropipette the saliva was applied on the surface of Mitis salivaris agar and Rogosa agar medium which were used for mutans streptococci and lactobacilli growth. The plates were incubated in an anaerobic atmosphere for 48 hours at 37°C. CFU with morphology characteristic of s.mutans and lactobacilli were counted and expressed as numbers of CFU per milliliter of saliva [21].

Dental plaque sample was taken from the buccal surface of the maxillary second primary molar which should be sound, otherwise the sample was taken from the maxillary first primary molar**,** by a clean toothpick and store in Epindorf tube which contain 1 ml. normal saline to prevent dryness while reaching the laboratory to be disperse for 30 seconds by vortex mix[22]**,** then tenfold dilutions was made by normal saline to have a full colony counting for the caries related microorganisms (streptococcus mutans and lactobacilli), then inoculated in Mitis-Salivarius-Bacitracin agar which is the special selective media for the streptococcus mutans, and Rogosa agar media which is the special selective media for the lactobacilli. Colonies of the bacteria were counted with the aid of dissection microscope (15 X) on the basis of their characteristic morphology [21].

Statistical analysis was done by using correlation test by the aid of the SPSS application version 13. Correlation is significant at 0.01 levels (2-tailed).

RESULTS

Descriptive statistics revealed that the caries experience for primary teeth was 31.5±17.56 with range 10(minimum) -88(maximum), while for the permanent dentition was 1.125±1.39 with a range 0.0-4.0 (Table 1), age range was 3-10 years demonstrated in table 2. Correlation coefficient revealed that there was a weak negative not significant correlation between the caries related microorganisms (streptococcus mutans and lactobacillus) in the dental plaque. Also, the same result was found between the streptococcus mutans bacterial count in different media (plaque and saliva). On the other hand, when we correlated the lactobacillus bacterial counts in dental plaque and saliva, we found a weak positive not significant correlation. The same result was found when we correlate the caries related microorganisms (streptococcus mutans and lactobacillus) with each other in saliva (Table 3).

Data analysis of the present study showed that there was a strong positive significant correlation between dmfs and ds components of the primary teeth. The results revealed that there was a weak positive not significant correlation between the caries experience of the primary teeth (dmfs or ds) and the streptococcus mutans bacterial counts in the dental plaque, while there was a weak negative not significant correlation between the caries experience in the primary teeth (dmfs or ds) with the lactobacillus bacterial counts in the dental plaque (Table 4).

Different pictures were found concerning the relation of caries experience for the primary teeth with the bacterial count (streptococcus mutans and lactobacilli),the relation was a weak negative not significant with the streptococcus bacterial counts, and a weak positive non significant with the lactobacillus bacterial counts(Table 5).

Concerning permanent teeth, data analysis of the present study showed that there was a strong positive significant correlation between DMFS and DS components. Correlation coefficient between caries experience in the permanent teeth with the bacterial counts of the caries related microorganisms (streptococci and lactobacilli) in the dental plaque revealed weak, negative not significant correlations (table 6).

The same results were found when correlating the caries experience in the permanent teeth with bacterial counts of the caries related microorganisms (streptococci and lactobacilli) in saliva (Table 7).

DISCUSSION

There is a concept that the streptococcus mutans are responsible for the initiation of dental caries and lactobacilli are responsible for its progression [2, 5], the same result was found in the present study as in dental plaque, negative correlation was found between streptococcus mutans and lactobacilli.

Negative, weak, not significant correlation was found when correlating the streptococcus mutans bacterial count in dental plaque and saliva which agrees with Orstavik et al who stated that when bacteria were suspended in whole saliva, the adherence of streptococcus mutans was inhibited [23]. Opposite results was found when correlating lactobacilli bacterial count in dental plaque and saliva, the outcome was positive but weak not significant, this is in agreement with Nancy and Dorignac[24], while it was found that the correlation is positive between streptococcus mutans and lactobacilli in saliva, and this is in accordance with Brambilla et al[18].This may be due to the caries activity in dental carious patients[2,5].

Strong positive significant correlation between dmfs and ds and the same result is obtained concerning the permanent teeth(DMFS,DS), may give a clue that the most apparent component of dmfs of the patients is ds and not the filling or missing surfaces, which indicates that these patients are with urgent need for motivation and visit their dentists for treatment. Positive results when correlating dmfs or ds with streptococcus mutans in dental plague, this may be explained by that the accumulation of plaque on teeth surface is the main causative factor of dental caries. While a negative correlation was found with lactobacilli, which could be attributed to the fact that the lactobacilli are responsible for the progression and not the initiation of dental caries.

Negative correlation was found when correlating caries experience in the primary teeth with streptococcus mutans in saliva and this disagree with Ge et al [25]; it may be due to different age group and different sample size.

Positive correlation was found between caries experience in primary teeth with lactobacilli in saliva which can be explained by the fact that lactobacilli appear during the first years of a child's life, and are present in high numbers in saliva [26].

Correlation coefficient between caries experience in the permanent teeth with the bacterial counts of the caries related microorganisms (streptococci and lactobacilli) in the dental plaque and in saliva revealed weak, negative not significant correlations. These results could be due to that this study involves different age group children which contain mixed dentition.

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Table 1: Caries experience for primary and permanent dentition.

Caries experience	N	Minimum	Maximum	Mean	SD
dmfs*	53	10.00	88.00	31.5094	17.55718
DMFS**	24	.00	4.00	1.1250	1.39292

*** decayed, missed, filled, surfaces in the primary teeth.**
**** decayed, missed, filled, surfaces in the permanent teeth.**

Table 2: Distribution of the age groups

Age group	N	percent
3-6	35	66
7-10	18	34
	53	100

Table 3: Correlation coefficient of the bacterial counts (streptococcus mutans and lactobacillus) in the dental plaque with the bacterial counts in the dental plaque and saliva.

		r	P	N
Streptococcus mutans-plaque	**Lactobacillus-plaque**	-.010	.959	32
	Streptococcus mutans -saliva	-.068	.711	32
Lactobacillus- plaque	**Lactobacillus- plaque**	.008	.964	32
Streptococcus mutans -saliva	**Lactobacillus- plaque**	.247	.174	32

Table 4: Correlation coefficient between the caries experience and the bacterial colony counts (streptococcus mutans and lactobacillus) in the dental plaque.

Caries experience		r	P	N
dmfs	Streptococcus mutans-plaque	.289	.109	32
	Lactobacillus- plaque	-.107	.559	32
ds*	Streptococcus mutans-plaque	.320	.074	32
	Lactobacillus- plaque	-.195	.284	32

*** decayed surfaces in the primary teeth.**

Table 5: Correlations between the caries experience in the primary teeth and the bacterial counts (streptococcus mutans and lactobacillus) in saliva.

Caries experience	Saliva	r	P	N
dmfs	Streptococcus mutans	-.097	.597	32
	Lactobacillus	.110	.551	32
ds	Streptococcus mutans	-.083	.651	32
	Lactobacillus	.200	.273	32

Table 6: Correlations between the caries experience in the permanent teeth and the bacterial counts (streptococci and lactobacilli) in the dental plaque:

Caries experience	plaque	r	P	N
DMFS	Streptococcus mutans	-.346	.174	17
	Lactobacillus	-.198	.446	17
DS*	Streptococcus mutans	-.346	.174	17
	Lactobacillus	-.198	.446	17

***decayed surfaces in the permanent teeth.**

Table 7: Correlations between the caries experience in the permanent teeth and the bacterial counts (streptococci and lactobacilli) in saliva:

Caries experience	Saliva	r	P	N
DMFS	Streptococcus mutans	-.081	.757	17
	Lactobacillus	-.117	.654	17
DS	Streptococcus mutans	-.081	.757	17
	Lactobacillus	-.117	.654	17