



EARLY CHILDHOOD CARIES (ECC) AND NITRIC OXIDE (NO)

Biochemistry

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ABSTRACT

BACKGROUND- Dental caries is unique not only in terms of the pathological mechanism; other aspects, social and economical, are also worthy of note.¹ The concentration of nitrate in saliva varies according to dietary nitrate intake, the activity of bacterial nitrate reductase, salivary flow rate, and endogenous production of nitrate.²

AIM- To comparatively evaluate the salivary nitric oxide levels in ECC and non-ECC children. **MATERIAL AND METHODS-**

100 children were included in the study, group I was of 50 non-ECC children and group II was of 50 children with ECC. Nitric oxide was estimation done by using Griess Reagent.

RESULT- The nitric oxide levels in Group I was 54.07 ± 15.68 and Group II was 34.84 ± 10.79 .

CONCLUSION- Based on the present study and available literature it can be concluded that nitric oxide is decreased in the saliva of ECC patient

KEYWORDS

Early Childhood Caries, Nitric Oxide

INTRODUCTION:

The uniqueness of dental caries makes it a fascinating study from a scientific standpoint. Dental caries is unique not only in terms of the pathological mechanism; other aspects, social and economical, are also worthy of note. Dental Caries is a biosocial disease rooted in the technology and economy of our society¹.

Many of the details of the dental caries process have been known for a long time. Even though new research findings are continually being published on various aspects of dental caries such as the microbiology of caries, the biofilm, demineralization and remineralization, fluoride applications, dietary components, saliva, and fluoride-containing dental materials, apparently it is still not possible to embrace the science and fully implement it to reduce the level of dental caries in the population or even try to implicate a particular cause for the same³.

The concentration of nitrate in saliva varies according to dietary nitrate intake, the activity of bacterial nitrate reductase, salivary flow rate, and endogenous production of nitrate⁴. Accordingly, when salivary nitrate comes into contact with the acid environment around the teeth provided by acid-producing bacteria such as *S. mutans*, a bolus of antimicrobial compounds, including nitric oxide, is formed and which results in bacteriostatic and possibly bactericidal effects⁵.

Consequently, it is considered that caries incidence might be low in subjects with high nitric oxide levels⁷. Also, the etiology behind the sudden appearance of Rampant Caries (RC) is still unclear and suggests that an overwhelming imbalance of the oral environment seems to accelerate it so that it becomes uncontrollable when compared to the normal caries pattern⁶. This study was planned to evaluate salivary nitric oxide (NO) levels in ECC and non-ECC children.

MATERIAL AND METHODS:

This study included two groups, one had children with Early childhood caries, another will had non-Early childhood caries children.

One hundred (100) children were included in this study. They were divided into two groups, with 50 in each group:

- Group I was of 50 non-early childhood caries children less than 71 months of age.
- Group II was of 50 children with early childhood caries of the age group of up to 71 months were included in the study (as per American Academy of Pediatric Dentistry guidelines).⁷

INCLUSION CRITERIA

- Patients in the age group of up to 71 months with ECC were included in the study (as per American Academy of Pediatric Dentistry guidelines) consent from the parents and the children were taken.
- Children without ECC of same age group were included for the control group.

EXCLUSION CRITERIA

- Patients who were currently on medications and systemic diseases.
- Patients who were mentally and physically disabled.

NITRIC OXIDE ESTIMATION

The collected saliva was stored in the chiller at 4°C temperature and the NO evaluation will be done within 24 hours. Salivary nitric oxide was measured by ELISA reader (LISA Plus Microplate Reader, Aspen Diagnostic Pvt. Ltd) at 545 nm by using Griess Reagent (Sisco Research Laboratories Pvt.Ltd). 2NO⁻ concentration was determined by using the classical Griess reaction with Griess reagent. Briefly, 0.1 ml of saliva was pipetted into the wells of flat bottomed 96 well microtitration plates, followed by the addition of 0.1 ml of Griess reagent. The plate was kept at room temperature for 10 min, after which purple color developed in positive plates. A standard curve was established using NaNO₂ in a range between 10 and 30 mM. After 10 min. the microplate was read on ELISA reader at 545 nm and the data derived from the slope of the NaNO₂ curve was presented as the median.

ETHICS:

The study was conducted after taking the written informed consent form from patient and all the procedure follows standard protocol.

RESULT:

In a sample size of 100 children of age less than 72 months of age, the mean nitric oxide levels of the saliva of group I are much higher when compared with group II which is statistically very highly significant. The nitric oxide concentration of saliva was higher in Group I when compared to that of Group II. It was found that nitric oxide levels in Group I was 54.07 ± 15.68 and that of study Group II was 34.84 ± 10.79 and it was statistically significant ($p < 0.0001$).

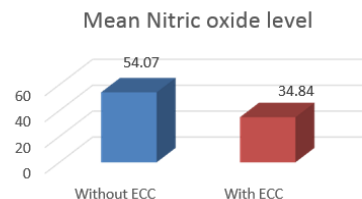


Fig: Mean nitric oxide level

Table: Nitric oxide level in both the groups

GROUP	NITRIC OXIDE LEVEL (µm/l)	
I (Without ECC)	Mean	54.07
	N	50.00
	Std. Deviation	15.68

II (with ECC)	Mean	34.84
	N	50.00
	Std. Deviation	10.79
p value	<0.0001	

DISCUSSION:

Dental caries is an infectious process involving the breakdown of the tooth enamel.⁸ American Academy of Pediatric Dentistry defined early childhood caries (ECC) as the presence of 1 or more decayed (non cavitated or cavitated lesions), missing (due to caries), or filled tooth surfaces in any primary tooth in a child 71 months of age or younger.

Saliva can affect the prevalence of dental caries in four general ways-first, through the cleansing effect which results in less accumulation of plaque; second, by reducing enamel solubility by means of calcium, phosphate, and fluoride; third, by the buffering effect saliva buffers and neutralizes the acids produced by the cariogenic microflora and finally, by antibacterial activity.⁹

The origin of nitric oxide (NO) in oral cavity appears in two ways. It can occur chemically, by physiological reduction of nitrates from food and enzymatically from L-arginine by inducible nitric oxide synthase (iNOS), an enzyme expressed in salivary- glands. In humans, dietary nitrates are absorbed in the duodenum and upper parts of ileum in the blood circulation and then concentrated in salivary glands by the mechanisms of active transport, reaching the concentration up to ten times higher than the concentration in plasma. After secretion into the oral cavity, its fast reduction into nitrites occurs. Nitric oxide is a highly reactive radical, taking part in nonspecific natural defense mechanisms of the oral cavity, aiming to prevent bacteria growth and development. Numerous authors prove the ability of salivary nitrites to have an inhibitory effect on the growth and survival of cariogenic bacteria in an acid environment. Nitric oxide easily passes through cell membranes and can provoke damage of microorganisms by different mechanisms, such as impairment of biological oxidation in mitochondria, DNA damage, and formation of highly toxic peroxynitrite¹⁰.

SUMMARY:

This study was conducted to comparatively evaluate the nitric oxide levels in children with ECC and non- ECC children. The saliva was collected from children aging less than 72 months of age stored at a 4-degree centigrade temperature which was used to estimate the nitric oxide levels of the samples of saliva by Griess reaction.

The results showed a significant difference between the groups. The results stated that salivary nitric oxide levels were higher in children without ECC (group I) than compared to children with ECC (group II).

CONFLICT OF INTEREST:

There is no conflict of interest.

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