

**Estimation of salivary glutathione levels and lipid peroxidation in caries free and caries active children.**

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**Conflicts of Interest:** Nil

**Abstract**

**Aims and objectives:** The aim of present study was to evaluate the salivary glutathione levels and lipid peroxidation levels in caries free and caries active children.

**Materials and methods:** The study included one hundred children who were divided in two groups i.e., caries free group and caries active group. Unstimulated saliva was collected and sent for laboratory analysis and children DMFT score was recorded.

**Results:** Both salivary glutathione levels and lipid peroxidation levels were increased in caries free group as compared to caries active group.

**Conclusion:** Within the limitations of the study, we concluded that the antioxidant enzymes of saliva can play an important role in development of caries.

**Keywords:** Salivary Glutathione, Lipid Peroxidation, Caries, Saliva, Antioxidant Enzymes

**Introduction:**

Dental caries is the most common chronic disease of mankind<sup>1</sup> and is profoundly affected by various factors

like oral hygiene, saliva, diet, bacteria etc.<sup>2</sup> Oral fluid or saliva is often called as mirror of body's health and is a perfect medium to be explored for health and disease surveillance.<sup>3</sup> It mainly comprises proteins, glycoprotein, electrolytes, small organic molecules and compounds transported from blood, constantly bathes the teeth and oral mucosa. It acts as a cleansing solution, an ion reservoir, a lubricant and a buffer and in addition, it also act as first line defense against free radical-mediated oxidative stress, since the process of mastication and digestion of ingested foods promotes a variety of reactions releasing free radicals.<sup>4</sup> Healthy individuals secrete about a liter and a quarter of saliva per day<sup>5</sup> and is easily available, includes non-invasive techniques and can be a useful diagnostic fluid in various clinical situations,<sup>3</sup> especially very good media to assess various analytes in children. Recently it has been claimed that imbalances in levels of free radicals, reactive oxygen species and antioxidants in saliva may play an important role in development of caries.<sup>4</sup> Hence, evaluation few low-molecular weight antioxidant enzymes i.e., salivary

glutathione levels and lipid peroxidation levels was done in caries free and caries active children to assess its effect on dental caries.

### Materials and methods

Hundred children aged between 11-14yrs were included in the study and were divided into two groups; group I consisting fifty children who were caries free, group II consisting of fifty children who were caries active. Informed consent was taken from parents and unstimulated saliva was collected<sup>4</sup> using cotton swab method. Caries was checked using DMFS index. The inclusion and exclusion criteria were caries active and caries free group are as follows:

#### Inclusion criteria:<sup>5</sup>

- Free from systemic or local disease, which affect salivary secretions.
- Caries active children should have at least five decayed tooth surfaces.
- Caries free children having no caries, DMFS =0.

#### Exclusion criteria:<sup>5</sup>

- Patients who were physically and medically compromised.
- Patients who were on medications.
- Patients who have arrested carious lesions.

All the subjects were instructed to refrain from drinking and eating 12hr prior to saliva collection and to brush their teeth in the morning.<sup>2</sup> Before saliva collection, the mouth was rinsed with distilled water and then unstimulated saliva was collected using cotton swab method<sup>14</sup> and sent to laboratory for analysis. Salivary glutathione levels was analyzed Beutlers method<sup>6</sup> and lipid peroxidation using ledwozyw method.<sup>6</sup>

### Results

Salivary glutathione (GSH) levels which were determined by Beutler method were lower in caries active children as

compared to subjects who were caries free (table 1, table2) indicating their absence can damage the tissues.

Lipid peroxidation (LPO) level which was determined using Ledwozyw method is an indicator of oxidant damage and was higher in caries active children as compared to caries free children (table 3, table 4).

**Table 1: Caries active group: GSH**

Descriptives			Statistic	Std. Error
CaGGSH	Mean		3161.067	268.2742
	95% Confidence Interval for Mean	Lower Bound	2621.949	
		Upper Bound	3700.184	
	5% Trimmed Mean		3103.852	
	Median		3165.333	
	Variance		3598552	
	Std. Deviation		1896.985	
	Minimum		832.0000	
	Maximum		6525.333	
	Range		5693.333	
	Interquartile Range		3410.000	
	Skewness		.329	
	Kurtosis		-1.184	
				.337
				.662

**Table 2: Caries free group: GSH**

Descriptives			Statistic	Std. Error
CfGGSH	Mean		6877.333	178.8473
	95% Confidence Interval for Mean	Lower Bound	6517.926	
		Upper Bound	7236.740	
	5% Trimmed Mean		7001.630	
	Median		6898.667	
	Variance		1599318	
	Std. Deviation		1264.641	
	Minimum		2592.000	
	Maximum		8512.000	
	Range		5920.000	
	Interquartile Range		1196.667	
	Skewness		-1.517	
	Kurtosis		3.806	
				.337
				.662

**Table 3: Caries active group: LPO**

Descriptives			Statistic	Std. Error
CAGLPO	Mean		30546.15	723.7613
	95% Confidence Interval for Mean	Lower Bound	29091.7	
		Upper Bound	32000.6	
	5% Trimmed Mean		30732.9	
	Median		30769.2	
	Variance		2.6E+0	
	Std. Deviation		5117.76	
	Minimum		19038.4	
	Maximum		38269.2	
	Range		19230.7	
	Interquartile Range		7644.23	
	Skewness		-.449	
	Kurtosis		-.539	
				.337
				.662

**Table 4: Caries free group: CFG-LPO**

Descriptives			Statistic	Std. Error
CFG LPO	Mean		26809.98	566.7965
	95% Confidence Interval for Mean	Lower Bound	25670.96	
		Upper Bound	27949.00	
	5% Trimmed Mean		26897.85	
	Median		26923.08	
	Variance		1.6E+07	
	Std. Deviation		4007.857	
	Minimum		14423.08	
	Maximum		34038.46	
	Range		19615.39	
	Interquartile Range		6826.923	
	Skewness		-.407	.337
	Kurtosis		.355	.662

## Discussion

Free radicals can be defined as molecules or molecular fragments with an unpaired electron which imparts certain characteristics to the free radicals such as reactivity.<sup>7</sup> Reactive free radicals are able to damage proteins, lipids, carbohydrates and nucleotides in the tissues<sup>8</sup> and can damage cells by lipid peroxidation that causes profound alteration in structural integrity and functions of cell membranes.<sup>8</sup>

Antioxidants are found in all biological species and protect against the potentially harmful effects of processes or reactions that cause excessive oxidations.<sup>9</sup> Certain inflammatory processes in the mouth, such as periodontal disease, have been related to an increase in oxidative damage in mouth and decrease in antioxidants.<sup>10</sup>

In past few decades saliva is used as a new diagnostic fluid, which is rich in antioxidant enzymes.<sup>3</sup> Many authors have related the saliva with periodontal disease but limited is known about its relation with caries.<sup>9</sup> Hence this study is attempted to correlate caries with salivary Glutathione levels and lipid peroxidation levels.

GSH is a tripeptide containing SH group whose salivary levels are decreased in periodontal diseases<sup>11,12</sup> and in our study its levels are also reduced in caries active children.

LPO, which is one of the indicators of oxidant damage, causes degeneration of cell membrane of oral tissue during

inflammatory processes.<sup>6</sup> Any imbalances in levels of free radicals and reactive oxygen species may play important roles in caries.<sup>4</sup> Saliva may constitute a first line of defense against free radical mediated oxidative stress.<sup>4</sup> Hence in present study LPO levels are more in caries active children. Rai et al determined the LPO product malonaldehyde in some oral diseases such as leukoplakia, oral submucous fibrosis, candidiasis, dental caries.<sup>13</sup>

As caries is a multifactorial disease many factors contribute to its occurrence so within the limitations of present study, we determined GSH levels which was less in caries active children whereas the LPO levels were higher in caries active children. It has been also found that various fruits and vegetables are good source of antioxidants and we can expect that intake of such foods may be important in reducing caries and also they can small volumes in various cements, mouthwashes etc.<sup>5</sup> However, before prescribing it becomes mandatory to definitely establish role of these enzymes in etiopathogenesis of caries.

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