



**Effect of Tooth Paste Containing Lactoferrin on Reversal of White Spot Lesion & Reduction in S. Mutans Count & Its Comparision with Clin Pro Tooth Crème in Children with Stage 1 ECC: A Clinical Study**

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**Type of Publication:** Original Research Article

**Conflicts of Interest:** Nil

**Abstract**

**Introduction:** ECC is the most prevalent disease worldwide with prevalence of 48% globally. It poses a clear risk to the general health, also is associated with a marked functional, aesthetic & psychological impact on the quality of life in children and their families.

**Aim:** To Evaluate effect of toothpaste containing lactoferrin in reversal of WSL & reduction in S. mutans count in children (3 to 5 years of age) with stage 1 ECC & its comparison with clin pro tooth crème

**Method:** A total of 32 children aged 3-5 with stage 1 ECC were randomly divided into 2 groups, group 1- BioXtra Tooth paste & group 2- Clin pro tooth crème. Salivary samples were collected and analyzed for streptococcus mutans at base line and after 4 weeks. Visual assessment of white spot lesions was done according to Ekstrand et al. The collected samples were

immediately transported to lab and inoculated on MSB agar.

**Result:** There was reversal of white spot lesion and reduction in S. mutans count in both groups but was not statistically significant.

**Conclusion:** Both the toothpaste can be used for reversal of white spot lesion and reduction of S. mutans count as there was statistically significant difference between the two toothpastes.

**Keywords:** lactoferrin, saliva, remineralization

**Introduction**

Dental caries is a dynamic process that occurs continuously affecting all population worldwide. It is the most prevalent disease worldwide with prevalence of 48% globally and has several unique characteristics in clinical appearance such as rapid development of caries, which affects a number of teeth soon after they emerge

in oral cavity. These lesions involve tooth surfaces that are less prone to caries development. Also, poses a clear risk to the general health and has a marked functional, aesthetic, and psychological impact on the quality of life in children and their families. Moreover, primary teeth caries is the strongest predictor of caries in the permanent dentition.

ECC usually progresses in phases as a result of multiple risk factors, including enamel defects, microbial risk factors, dietary risk factors, environmental factors, high sugar intake, poor oral hygiene, and lack of fluoride exposure. It has a pathognomic clinical progression with classic etiologic factors. The incorrect feeding habit of putting a child to sleep with milk is one of the main contributing reasons. The majority of primary teeth are rapidly becoming carious with early pulp involvement due to milk pooling, decreased salivary flow, and swallowing; occasionally, even the most resilient mandibular incisors are affected.<sup>4</sup>

Understanding the normal dynamics of oral cavity is the goal in preserving the primary tooth till the time of exfoliation. Saliva has a significant role in remineralization of dental enamel.

It not only has a buffering capacity to neutralize the oral cavity's low pH generated after acidic encounters, but also acts as a carrier of essential ions, such as fluoride, calcium and phosphate, which have a positive role in enamel's remineralization. Additionally, it contains a variety of other substances, including proteins including immunoglobulins, glycoproteins, amylase, lactoferrin, and albumin. Amino acids, urea, uric acid, creatine, lipids, water-soluble vitamins, lysozyme, and esterases.<sup>5</sup>

One among them is the Lactoferrin, a non-enzymatic antibacterial protein which is present in concentration of 0.008 mg /ml in saliva is an important modulator of immune response and inflammation and represents an

important defensive element by inducing broad spectrum of antibacterial effects against microbial infections. It can compete with bacterial iron-chelating molecules & deprive the bacteria of this essential element.

It is proven that decreased levels of lactoferrin can lead to increased susceptibility to ECC that endorses the idea of adding lactoferrin to oral health care products to partially restore saliva's own antimicrobial capacity and to add physiological salivary antimicrobial agents into the mouth.

As prevention is the mainstay in pediatric practice so every effort should be done to either reduce the microorganism, reduction of substrate and to increase the resistance of teeth so that the ECC can be reversed in the initial stage itself and rampant spread of caries can be prevented. Prevention of the progress of the ECC can be achieved with the aid of restorations, diet counselling, educating parents regarding decay promoting feeding behaviors, maintain good oral hygiene, and the use of preventive agents like topical fluorides.<sup>9</sup>

Clinpro tooth crème (3M ESPE) is composed of 0.21% sodium fluoride (NaF) and it is considered as anti caries dentifrice that contains 950 ppm fluoride and a functionalized tricalcium phosphate (f-TCP) ingredient. One of the most important advantages is being stable in an aqueous environment and also does not affect the fluoride activity added in the dentifrices according to Karlinsey et al.<sup>11</sup>

Therefore, the present study aimed to evaluate effect of toothpaste containing lactoferrin in reversal of white spot lesion & reduction in *S.mutans* count & its comparison with clin pro tooth crème in 3-5 years old children with stage I ECC.

**Materials and Method:** A total of 32 children were selected according to inclusion criteria from the

outpatient department of pediatric & preventive dentistry from KVG dental college & hospital, Sullia.

### Inclusion Criteria

- Children with stage I early childhood caries
- Children of 3 to 5 years of age of both sexes
- Children who have not received any antibiotic therapy for last 3 months

### Exclusion Criteria

- Children with special health care needs
- Children who have systemic disease
- Children with emergency treatment needs
- Parents who are not willing for informed consent
- Parents were informed regarding the study and consent was obtained from them.
- Subjects were randomly selected & divided into two groups with 16 children in each group. Salivary samples were analyzed for streptococcus mutans. Visual assessment of white spot lesions was done according to Ekstrand et al. (figure 10)
- Group I: used BioXtra tooth paste
- Group II: used clin pro tooth crème tooth paste
- All the children were examined using a mouth mirror with good reflecting surface in adequate day light. The patients were instructed to use pea sized toothpaste with the scrub- technique of brushing for a time period of 2-3 minutes twice daily. The saliva samples were collected from the children before and after 4 weeks of brushing & again Visual assessment of white spot lesions was done according to Ekstrand et al to check for the efficacy of different treatment regimes.
- The subjects were instructed to chew on paraffin wax for 2-3 minutes and expectorate 1-2 ml of saliva directly into the sterile collection bottles.

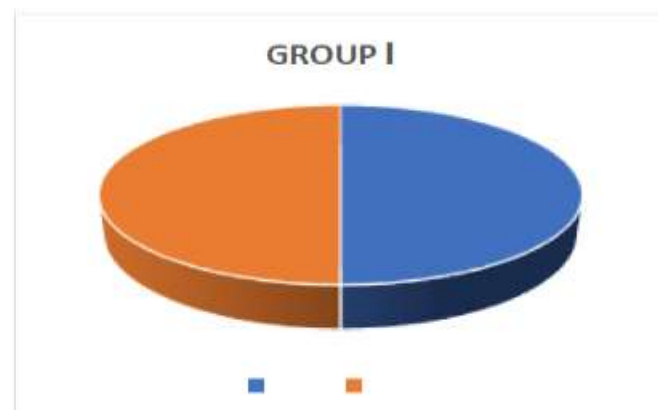
- The collected samples were immediately transported to Microbiology lab and inoculated on Mitis Salivarius Bacitracin agar (MSB). The inoculated plates of Mitis Salivarius Bacitracin agar were incubated at 37 0 C in a candle jar environment for 48 hours. Colony Forming Units (CFU) were counted and the results were tabulated and subjected to statistical analysis.

### Results

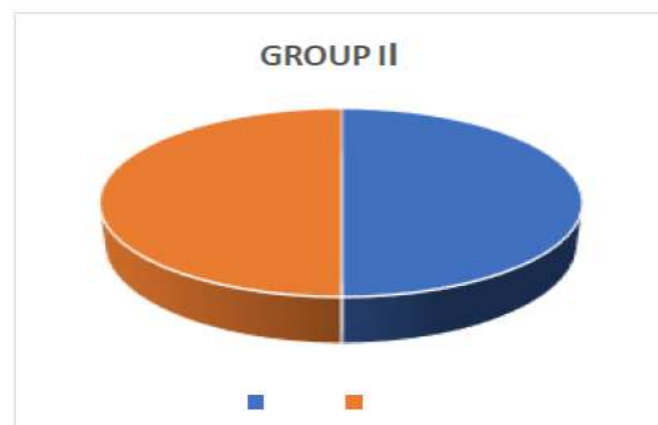
Table 1:

Table No 1			GENDER DISTRIBUTION		
			Group		Total
			Group I	Group II	
	Male	Count	8	8	16
		%	50.0%	50.0%	50.0%
	Female	Count	8	8	16
		%	50.0%	50.0%	50.0%
Total		Count	16	16	32
		%	100.0%	100.0%	100.0%

Graph 1: Distribution of gender- group I

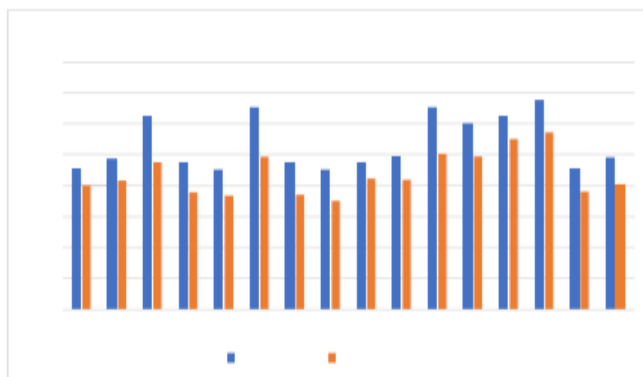


Graph 2: Distribution of gender- group II



**Interpretation:** A total of 32 samples taken for the study were randomly divided into two groups using simple random sampling. 8(50%) males and 8(50%) females in group 1 & 8(50%) males and 8(50%) females in group 2 participated in the study. (Graph 1,2) GROUP I GROUP II

#### Group I- S Mutans Count At Baseline & After 4 Weeks



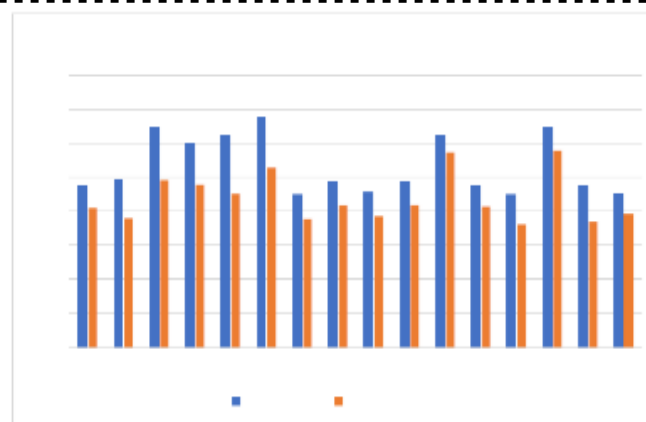
Graph 3: s mutans count at baseline & after 4 weeks for group

Table 2:

Table No 2	Group	N	Mean	Std. Deviation	Z
SM count day 1	Group I	16	106548.750	17230.487	.038
SM count after 4 weeks	Group I	16	87395.000	13720.112	.038

**Interpretation:** BioXtra toothpaste group showed Mean salivary S Mutans count of 106548 at baseline and 87395 post intervention suggestive of significant reduction of salivary S Mutans count post intervention after 4 weeks

#### Group II- S Mutans Count At Baseline & After 4 Weeks



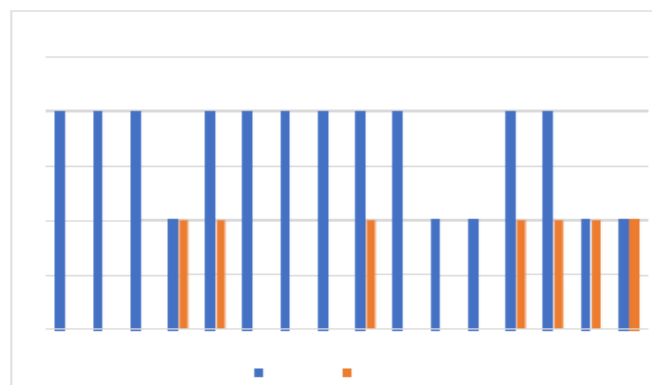
Graph 4: s mutans count at baseline & after 4 weeks for group 2

Table 3:

Table No 3	Group	N	Mean	Std. Deviation	Z
SM count day 1	Group II	16	106560.625	17216.947	p=0.97 ns
SM count after 4 weeks	Group II	16	87779.437	14186.651	p=0.97 ns

**Interpretation:** Clin pro tooth crème toothpaste group showed Mean salivary S Mutans count of 106560 at baseline and 87779 post intervention suggestive of significant reduction of salivary S Mutans count post intervention after 4 weeks

#### Group I- White Spot Lesion at Baseline & After 4 Weeks



Graph 5: WSL at baseline & after 4 weeks for group 1

Table 4:

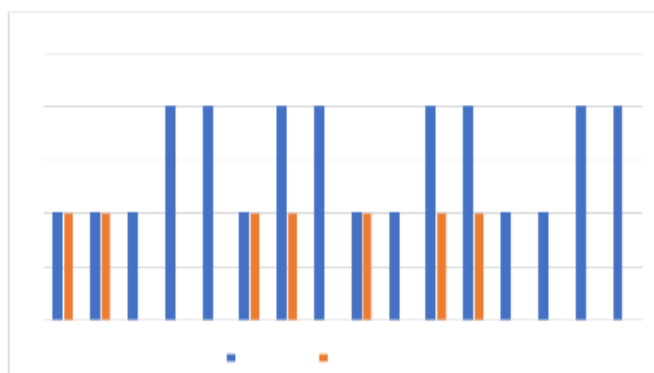
Table No 4	WHITE SPOT LESION AT BASELINE- GROUP I			
SCORE	1	Count	5	
		%	31.3%	
	2	Count	11	
		%	68.8%	
Total	Count		16	
	%		100.0%	

Table 5:

Table No 5		WHITE SPOT LESION AFTER 4 WEEKS- GROUP I	
SCORE	0	Count	9
		%	56.3%
	1	Count	7
		%	43.8%
Total		Count	16
		%	100.0%

**Interpretation:** Group 1 showed white spot lesion score of 2 for 68.8% and score of 1 for 31.3% of samples at baseline according to criteria by Ekstrand et al. Post intervention after 4 weeks 56.3% samples showed score 0 and 43.8% showed score 1 suggestive of significant reversal of white spot lesions post intervention

#### Group II- White Spot Lesion At Baseline & After 4 Weeks



Graph 6: WSL at baseline & after 4 weeks for group 2

Table 6:

Table No 6	WHITE SPOT LESION AT BASELINE- GROUP II		
SCORE	1	Count	8
		%	50.0%
	2	Count	8
		%	50.0%
Total		Count	16
		%	100.0%

Table 7:

Table No 7		WHITE SPOT LESION AFTER 4 WEEKS - GROUP II		
SCORE	0	Count	9	
		%	56.3%	
	1	Count	7	
		%	43.8%	
Total		Count	16	
		%	100.0%	

**Interpretation:** Group II showed white spot lesion score of 2 for 50.0 % and score of 1 for 50.0 % samples at baseline according to criteria by Ekstrand et al. Post intervention after 4 weeks 56.3% of samples showed score 0 and 43.8% showed score 1 suggestive of significant reversal of white spot lesions post intervention.

#### Group I: pre and post intervention





## Group II: pre and post intervention



## Discussion

Prevention is better than cure is a very appropriate strategy in management of ECC. Caries in primary dentition if untreated can be a risk factor for permanent dentition and the aftermath could be very disastrous to the child. So, every effort should be taken to curtail the risk of progress of ECC, as it is a devastating disease of childhood that not only destroys the oral health but also affects general health causing malnutrition, development of oral habits, malocclusion, mocking from peer leading to psychological trauma.

ECC is initially observed as dull, white spot lesion on the cervical margins of maxillary primary incisors which advance to cavitated enamel surfaces in short span of time due to demineralization of enamel subsurface that can be reversed if detected at this stage.

Hence, early detection followed by intervention of ECC at stage 1 plays a key role in preventing further demineralization and destruction of tooth structure as these lesions are in reversible stage so any step taken to favor remineralization in the dynamic process of demineralization and remineralization happening in the

child's oral cavity can be a very effective step in fighting ECC.

Oral cavity with saliva supersaturated with calcium and phosphate ions constantly favours remineralization and substances like IgA and lactoferrin are the main anticariogenic factors promoting remineralization. Our study intended to evaluate role of lactoferrin by incorporating it in a toothpaste to evaluate the reversal of white spot lesion also, as *S Mutans* is the main causative cariogenic bacteria, we evaluated the levels of *S mutans* and compared it with Clin pro tooth crème toothpaste in children aged 3 to 5 years with stage 1 ECC. Stage I ECC was included in our study as at this stage the carious process can be reversed back with the aid of diet counselling, educating parents regarding decay promoting feeding methods, restorations, maintain good oral hygiene, and the use of preventive agents like fluorides.

In our study salivary *S mutans* count was checked pre and post intervention as it is the most common microorganisms associated with ECC. Therefore, reduction of salivary *S mutans* is directly proportional to decrease in caries susceptibility. this is in accordance with Avery et al.

Post intervention a significant reduction in salivary *S mutans* count was noted with use of BioXtra toothpaste because of presence of Antimicrobial salivary proteins such as lactoferrin that have cumulative/synergistic effect resulting in an efficient molecular defense network of the oral cavity. It can compete with bacterial iron-chelating molecules & deprive the bacteria of this essential element. Also, similar results were seen in Clin pro tooth crème toothpaste group in which the *s mutans* count had significantly reduced post intervention as it is an advanced formula containing an innovative tri-calcium phosphate, fluoride as well as calcium and

phosphate, which are components naturally found in saliva.<sup>11</sup> Tri-calcium phosphate maximises the performance of both calcium and fluoride to provide enhanced fluoride delivery, resulting in strengthening of enamel and reversal of white spot lesions, which is in accordance with the study done by chung et al. In our study reversal of white spot lesion was examined using visual assessment criteria by Ekstrand et al and only lesions scoring 2- opacity / discolouration visible even without air drying was included.

In the present study, there was reduction of score from 2 to 1 or 0 post intervention in BioXtra toothpaste group suggesting the reversal of white spot lesion. This is because of presence of Antimicrobial salivary protein such as lactoferrin. Also, the fluoride which is present forms fluorapatite by inhibiting the formation of highly soluble calcium phosphate. The higher the amount of fluorapatite crystals, the greater the resistance of the enamel surface to acid attack. Fluorapatite promotes remineralization of initial demineralized areas and prevents the formation of new lesions.

Similarly, in Clin pro tooth crème toothpaste group after 4 weeks of intervention there was reduction of score from 2 to 1 or 0 which shows that the reversal of white spot lesion is due to presence of tri-calcium phosphate, fluoride as well as calcium and phosphate in clin pro tooth crème toothpaste which are also the components naturally present in the saliva. This Tri-calcium phosphate maximises the action of fluoride and calcium to supply fluoride more effectively thereby strengthening enamel and reversing white spot lesions.

There was noticeable reduction in the salivary S. mutans count and white spot lesion in both BioXtra toothpaste group as well as in Clin pro tooth crème toothpaste group but was not statistically significant as p value was greater than 0.05 which is parallel with the similar

studies done by Ravi Kumar et al, Vijay Kumar et al chung et al and Suresh et al.

### Conclusion

As most of the areas in India are in fluoride belt, already the risk of systemic fluorosis is severe. Since it is debatable whether to prescribe the fluoride tooth paste in fluorosis areas, BioXtra toothpaste or clin pro tooth crème toothpaste can be proposed as a substitute to normal tooth paste to reduce the salivary bacterial activities and caries progression in children with early childhood caries.<sup>6</sup>

Moreover, additional clinical assessments, particularly longitudinal studies, are necessary to fully understand the significant impact of these alternatives. Extensive clinical trials and long-term in vivo studies are essential to substantiate this perspective.

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TRICALCIUM PHOSPHATE VERSUS CASEIN  
PHOSPHOPEPTIDE- AMORPHOUS CALCIUM  
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