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Research Article

COMPARISON ANTI-BACTERIAL EFFICACY OF OCTENIDINE AND CHLORHEXIDINE AS A MOUTH WASH

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ABSTRACT

To evaluate the antimicrobial activity of Octenidine (OCT) 0.1%, Chlorhexidine (CHX) 0.2% using BANATM kit.

Microbiological techniques demonstrated that the combination of SRP and repeated professional plaque removal could have a beneficial effect on the subgingival microbiota.1,2 This has led to use of antimicrobial agents as an adjunct to periodontal therapy.One of the most frequently used antimicrobial agents is chlorhexidinegluconate (CHX), it is a broad spectrum antiseptic with a pronounced antimicrobial effect on both gram negative and gram positive bacteria as well as on some yeast and lipophillic viruses and its prolonged substantivity is still recognized as the gold standard for chemical plaque control.4 0.2% chlorhexidine solution was the first clinically effective mouth rinse that inhibited supragingival plaque formation .5 . There are not many studies done on OCT The results showed that OCT 0.1% was found to be the most effective in substantially reducing total bacterial counts after 42 days time interval. OCT 0.1% was found to be the more effective in substantially reducing total bacterial counts.

INTRODUCTION

Periodontitis is result of cumulative exposure of dental plaque, thus the main aim of periodontal therapy is the prevention of plaque accumulation and maintain periodontal health. The clinical effect of scaling and root planing (SRP) are well documented.6-8These studies indicated that SRP decreased pocket probing depth and attachment level measurements particularly at the deeper sites. Coccoid cells are more predominant at normal sites and motile rods and spirochetes are more frequently associated with the periodontically diseased sites.[9] Microbiological studies on effect of SRP indicated that proportion of spirochetes and motile rods decline after SRP while cocci and non motile rods increased .[10]

Although mechanical treatment significantly decreases the prevalence and levels of subgingival microorganisms, it does not necessarily eliminate all pathogens.[11] As probing depth increases, the effectiveness of scaling and root planing decreases leaving subgingival plaque and calculus on the root surfaces,[12]and repopulation of scaled teeth from bacterial reservoirs in dentinal tubules.[13]Haffajee *et al*. reported that SRP alone has limited affect on some pathogenic species .Microbiological techniques demonstrated that the combination of SRP and repeated professional plaque removal could have a beneficial effect on the subgingival microbiota[14,15] This has led to use of antimicrobial agents as an adjunct to periodontal therapy.

Clinical improvements after SRP are associated with microbiological changes that include a decrease in microbial load and a mean percentage change of certain periodontal pathogens, such as *Treponemadenticola*, *Porphyromonasgingivalis and Tannerella forsythia.*[16] Studies have shown that a significant antibody response is directed to *Porphyromonasgingivalis* and *Tannerella forsythia* in periodontal patients[17]*Treponemadenticola*have also been demonstrated in deep pockets from adult periodontitis patients compared to healthy subjects.[18]

These species are gram negative anaerobes which possess, invivo an enzyme capable of hydrolyzing synthetic trypsin substrate, BANA (N-Benzoyl D-L Arginine -2 Naphalamide) .BANA a colourless substrate, it releases β- naphthylamide , which turns orange red when a drop of fast garnet is added to the solution. Several Bacteroides and Capnocytophaga species were occasionally BANA positive, only when in large CFU's .[19]

Loesche proposed the use of this BANA reaction in subgingival plaque samples to detect the presence of any of these periodontal pathogens and thus serve as a marker of disease activity.19 Beck *et al* found positive BANA test was

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highly associated with the severity of attachment loss and was a significant predictor of attachment loss over a long period. [20]
Thus in this study we will aim to evaluate a) the effects of Scaling and Root planing with or without 0.2 % Chlorhexidine rinse on clinical parameters b) use of BANA as a test for evaluating microbiological parameters in Generalized Chronic Periodontitis.

To improve the outcome of mechanical oral hygiene procedures several antimicrobial agents , delivered by rinsing , irrigation , systemic administration and local devices, have been used to overcome the limited efficacy of conventional treatment of periodontitis.[3] One of the most frequently used antimicrobial agents is chlorhexidinegluconate (CHX), it is a broad spectrum antiseptic with a pronounced antimicrobial effect on both gram negative and gram positive bacteria as well as on some yeast and lipophillic viruses and its prolonged substantivity is still recognized as the gold standard for chemical plaque control.[4] 0.2% chlorhexidine solution was the first clinically effective mouth rinse that inhibited supragingival plaque formation .[5]

During the past few years, there has been a dramatic increase in the use of mouthwashes. These are perceived by users to maintain oral health and have a fresh "dental" taste.[1]Some health care professionals recommend their use as an adjunct to conventional mechanical removal of plaque and this advice has been supported by studies which have shown that tooth brushing is only poorly carried out. [2, 3]

Octenidine hydrochloride (OCT) Octenidine, is a bispyridine derivative, i.e., N,N-[1,10-decanediyldi-1(4H)-pyridinyl – 4 pylidene] bis (1-octanamine) dihydrochloride a new bipyridine antimicrobial compound, has been developed as a potential antimicrobial/antiplaque agent for use in mouthwash formulations.[24] The existing data suggest that a mouthrinse containing 0.1% OCT may be capable of exerting beneficial clinical effects upon plaque accumulation and gingivitis. OCT used in the form of mouthrinse was reported to inhibit dental plaque and caries both in rats and humans. It has been demonstrated that OCT appears to be more effective than chlorhexidine as a means for prolonged bacterial anti-adhesive activity.[25]

Aim

The aim of the study was to compare and evaluate the as antibacterial action of octenidine hydrochloride and chlorhexidinegluconate as a mouthwash.

MATERIALS AND METHODS

Mouthwash

The mouthwash tested in the study were Octenidine hydrochloride (OCT) 0.1% (ORAHEX PRO, Abbott) and Chlorhexidinegluconate (CHX) 0.2% (Chlorhex , Dr Reddy's). 60 Subjects randomly selected comprising of both the sexes, visiting outpatient department of Periodontology, Govt. Dental College and Hospital Jammu, were considered for the present clinical study after meeting inclusion and exclusion criteria.

Inclusion Criteria

The criteria for inclusion in the study were:- Patients of age between 25-50 years. Patients diagnosed as suffering from generalized chronic periodontitis determined on clinical and radiographic examination .Minimum of 4 teeth with one site with pocket depth ≥5mm& ≤7mm .Cooperative patients who are able to attend the hospital at frequent intervals.

Exclusion Criteria

Patient who had received any type of invasive periodontal therapy for past 4 months. Presence of any systemic disease that would influence the course of periodontal disease. Pregnancy and lactation. Smoking habit. Allergic to chlorhexidine.Subjects having periapicallesions , gingival abscess , periodontal abscess .Patients with history of antimicrobial drug intake for 7days or longer in previous 3 months

BEFORE THE SELECTED SUBJECTS WERE TAKEN UP FOR THE STUDY, THEY WERE MADE TO SIGN A WRITTEN CONSENT FROM REGARDING THE BENEFITS AND PROTOCOL OF THE STUDY.

METHODOLOGY EMPLOYED

After the selection of subjects for the study based on the inclusion and exclusion criteria, the periodontal examination was done. Subjects were randomly assigned into two groups:- 30 Subjects in Control Group(Group A) and 30 Subjects in Treatment Group (Group B) i.e.:-
Group A (SRP + 0.2%CHX)
Group B (SRP + 0.1% OCT)

Parameters Selected

Plaque Index (Sillness and Loe ,1964).[26]

1. Baseline
2. DAY 42

Sulcus Bleeding Index (Muhlemann H.R and son ,1971).[27]

1.Baseline
2.DAY 42

BANA (N-benzoyl-d L-arginine-2-napthylamide)[28]

DAY 42

BANA (N-benzoyl-d L-arginine-2-napthylamide)[28]

Subgingival plaque sample was collected at 42ndDAY from the 4 selected site with Gracey curettes then was placed on BANA impregnated strips at the lower half of the strip . The upper reagent matrix contains a chromogenic diazo reagent (fast black K) was activated by moistening it with distilled water . The upper reagent matrix, which reacts from BANA by bacterial enzyme reacts with fast black K forming a permanent blue color when incubated at 55°C for 15 mins. The blue color of a positive or weak positive reaction appears in the upper matrix and is permanent.

Blood, and saliva do not hydrolyze BANA and do not interfere with the test, but blood in the sample may obscure the visualization of the blue color. The BANA strips were subsequently examined for the presence (positive) or absence (negative) of blue colour . No colour change was marked for negative , light faint blue was marked for weak positive and evident blue marked for positive test result.

Statistical Methodology Employed

Statistical analysis was done by using Statistical Software SPSS (Version 20.0) and Microsoft Excel. Data was analysed by applying descriptive statistics viz., means, standard deviations and percentages and presented by means of Bar Diagrams.

Inter group analysis was done by applying Student's Independent t-test and Chi-square test.. P-value less than 0.05 was considered statistically significant. P-value less than 0.001 was considered statistically highly significant.

RESULTS

Table 1 Comparison of mean Plaque Index reduction scores between two groups

Plaque Index	Group A		Group B		Difference between groups	P-value
	Mean	SD	Mean	SD		
Baseline	1.8805	0.0745	1.9164	0.1687	0.0359	0.490#
42 Days	1.2195	0.0883	0.9871	0.1242	0.2324	<0.001*

On comparison between the two groups at the baseline the difference was statistically not significant. The test group (group B) showed greater improvements in plaque control index scores than control group (group A) .The difference in results showed a statistically significant decrease at day 42 ($p < 0.001$).

Table 2 Comparison of mean Sulcus Bleeding Index reduction scores between two groups

Sulcus Bleeding Index	Group A		Group B		Difference between groups	P-value
	Mean	SD	Mean	SD		
Baseline	1.8708	0.0650	1.9168	0.0840	0.0460	0.131#
42 Days	0.9479	0.1216	0.7900	0.0917	0.1580	0.001*

On comparison between the two groups at the baseline the difference was statistically not significant. The test group (group B) showed greater improvements in sulcus bleeding index scores than control group (group A). The difference in results showed a statistically significant decrease at day 42 ($p < 0.001$).

Table 3 Comparison between two groups based on BANA test score

BANA	Group A (n=54 sites)		Group B (n=54 sites)		P-value
	No.	%age	No.	%age	
Baseline	0	0	0.0	0	0.0	0.749#
	1	9	15.0	10	16.7	
	2	51	85.5	50	83.3	
42 Days	0	39	65.0	44	73.3	0.017*
	1	13	21.7	12	20.0	
	2	8	13.3	4	6.7	

***Statistically Significant Difference (P-value by Independent t-test)**

#Statistically Non-significant Difference (P-value by Independent t-test)

On comparison, the results (**Table 3)** dictated that the improvement in the results were statistically significant (p 0.017) on the 42nd day. Group B showed a significant increase in the BANA negative sites in comparison to the control group on the 42nd day. The no of BANA positive sites were also statistically decreased more in test group than in the control group.

DISCUSSION

In this study, antibacterial effect of OCT in comparison with CHX was evaluated . In the present study, the plaque index scores were more loweredin group B when compared to group A. The difference was statistically significant. The scores of sulcus bleeding index were also more decreased in group B than group A. The difference was statistically significant .Therefore the study indicates that OCT mouthwash is more potent antiplaque and anti inflammatory agent in comparison to CHX.Microorganisms levels were tested by BANA . The BANA negative sites were more in OCT group than the CHX group and the difference was statistically significant. Thus the antimicrobial effect of OCT is more than CHX.

Gjermo *et al.* [29] reported that rinsing twice a day with 10 ml of a 0.2% CHX inhibited the dental plaque formation. Furthermore, its antigingivitis efficacy was also well documented.[-30-32] Unfortunately, these positive effects are accompanied by side effects, the most disturbing being extrinsic tooth staining.[33, 34] In few cases, the occurrence of gingival desquamation and painful mucosa were reported.[32, 34]Flotra *et al.*[34] have implicated chlorhexidine in altered taste sensation, superficial desquamation of the oral mucosa, brownish discoloration of the tongue and teeth, and increased calculus formation. [35]Chlorhexidine has also been associated with potential anaphylactic reactions. [36-37] Studies with radiolabeled chlorhexidinemouthrinse have shown its ability to penetrate the intact mucosal barrier of the oral cavity or intestinal tract.[38, 39]Ohtoshi *et al.* [41] reported more than 30 cases of anaphylactic shock after the topical application of chlorhexidine.

OCT is a mouth rinse capable of exerting beneficial clinical effects upon plaque accumulation and gingivitis development. Octenidine is an excellent antimicrobial mouth-rinse having properties to support this inference. Although OCT has significant antibacterial activity, additional studies will be needed to investigate OCT's relative safety, biocompability and absence of unfavourable cosmetic and organoleptic properties. Octenidinedihydrochloride is a cationic antimicrobial substance, which as a result of the two positive charges in relation to the molecular weight of 437 daltons is strongly adsorbed onto negative cell surfaces. It reacts with polysaccharides in the cell wall of microorganisms, attacks the enzymatic systems there, destroys cell function and leads to leakage of the cytoplasmic membrane.

As a result, the mitochondrial function is also disturbed. Furthermore, interaction with salts of the fatty acid glycerol phosphate in bacterial cell membranes serving as main binding partners is discussed. Some findings indicate strong adherence to lipid components in cell membranes (e.g. cardiolipin), which might explain the high antimicrobial activity together with good tolerability for human epithelium and wound tissue. Octenidinedihydrochloride shows a broad antimicrobial activity against Gram-positive and Gram-negative bacteria, chlamydiae and fungi. Microbiostatic and microbicidal efficacy ranges about 10 times higher than that of chlorhexidine.[42]Beiswanger *et al.* [43] (1990) conducted a three-month clinical trial of 0.1 % Octenidinemouthrinse in which

450 adults participated, using their normal oral hygiene practices. Octenidine reduced plaque by one-third and gingivitis by one-half compared with the placebo. One of the recent studies showed that a 0.1% octenidine mouth rinse provided statistically significant reductions of 39% less plaque, 50% less gingivitis, and 60% fewer gingival bleeding sites.
Dogan *et al* [44] compared the short-term relative antibacterial effects of OCT and CHX. Their results were similar with our study, OCT was found favorably more effective than CHX in its antibacterial activity, both *in vitro* and *in vivo*.

CONCLUSIONS

Two different mouth rinse solutions i.e. OCT 0.1% and CHX 0.2% were compared with each other for their antibacterial effects. From the tested rinsing solutions, OCT 0.1% was found to be the more effective in substantially reducing total bacterial counts.

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