"COMPARE THE EFFECTIVENESS OF CHEMICAL AND HERBAL DISINFECTANTS ON ALGINATE IMPRESSION MADE FROM DIABETIC DENTULOUS PATIENTS"

Dissertation

Submitted to

BABU BANARASI DAS UNIVERSITY

LUCKNOW, UTTAR PRADESH

In the partial fulfillment of the requirements for the degree

of

MASTER OF DENTAL SURGERY

In

PROSTHODONTICS, CROWN & BRIDGE

By

DR. ROOPALI SHARMA

Under the guidance of

DR. GARIMA AGARWAL

READER

DEPARTMENT OF PROSTHODONTICS, CROWN & BRIDGE

BABU BANARASI DAS COLLEGE OF DENTAL SCIENCES,

LUCKNOW

(Faculty of Babu Banarasi Das University)

Batch:2020-2023 **Enrollment number: 12003290350**

DECLARATION BY THE CANDIDATE

I hereby declare that this dissertation entitled "**COMPARE THE EFFECTIVENESS OF CHEMICAL AND HERBAL DISINFECTANTS ON ALGINATE IMPRESSION MADE FROM DIABETIC DENTULOUS PATIENTS"** is a bonafide and genuine research work carried out by me under the guidance of **DR. GARIMA AGARWAL**, Reader, Department of Prosthodontics, Babu Banarasi Das College of Dental Sciences, Lucknow, Uttar Pradesh.

Roopali Sharma.

DR. ROOPALI SHARMA

Place: Lucknow

Date: 14/2/23

CERTIFICATEBYTHEGUIDE

This is to certify that the dissertation entitled **"COMPARE THE EFFECTIVENESS OF CHEMICAL AND HERBAL DISINFECTANTS ON ALGINATE IMPRESSION MADE FROM DIABETIC DENTULOUS PATIENTS"** is a bonafide work done by **DR. ROOPALI SHARMA**, under my direct supervision and guidance in partial fulfillment of the requirements for the degree of Master of Dental Surgery (MDS) in Department of Prosthodontics.

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Roopali Sharma

Dr. Roopali Sharma

“UNCONDITIONAL LOVE AND SUPPORT COMES ONLY FROM PARENTS”

DEDICATED TO MY PARENTS

Mrs. VANDANA

SHARMA Late Dr. NEERAJ

SHARMA

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Best Regards

Dr. Roopali Sharma

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LISTOF ABBREVIATIONS

CHX/CH	Chlorohexidine
A-/AVG	Aloe vera
CFU	Colony Forming Unit
TSB	Tryptone Soya Broth
BAM	Blood Agar Media

AIM: To compare the effect of herbal and chemical disinfectants on alginate impression made from diabetic dentulous patients.

MATERIALS AND METHODS:

30 Age and sex matched, patients with uncontrolled diabetes (type II) were included in this study. Alginate impression was made from each patient which was further divided into 3 parts. Thus, in total 90 specimens were obtained. Microbiota colonies were studied on each specimen. For the control groups no treatment was done. Second sample was treated with chlorohexidine and the third one with aloevera disinfectant. The three samples were further processed for growth of microbiota colony, The efficacy of the disinfectants was compared between the two samples treated with disinfectants with respect to the control.

STATISTICAL ANALYSIS USED:

The statistical analysis was performed using SPSS software (version 23).
The statistical difference in CFU between three groups-
The intragroup comparison of CFU between different time interval (24hrs,48hrs,72hrs) in three groups (control, aloevera, CHX) were performed using paired t-test.
The intergroup difference in the CFU at 24hrs, 48hrs & 72hrs were analysed using the one-way ANOVA followed by post HOC analysis at 5% significance level. (p= 0.05).

RESULTS

The data obtained from the above study demonstrates that the efficacy of aloevera is comparable to chlorohexidine (a gold standard) as a disinfectant for microbial growth on alginate impression.

CONCLUSION

Efficacy of aloe vera, the herbal disinfectant is comparable to chlorohexidine, the chemical disinfectant on alginate impression made from diabetic dentulous patients.

The goal of disinfection in the dental office is to prevent the spread of infection from one patient to another and maintain the safety of the dental care providers. Prevention of cross infection has significant effect on infection control. Impression disinfectants are important consideration when it comes to preventing infection. Nowadays, various disinfectants are available in the market, but to narrow down our pick for the one with high potency and least side effects is still debatable. Use of chemical disinfectant is a commonly known practice in dentistry but use of the herbal for the same is evolving too. Aloe vera is being used as a disinfectant in medical field already. The availability of the same is just not easy but also offers higher biocompatibility and lesser side effects.

Aloe Vera belongs to Liliacea family. Aloe barbadensis miller and Aloe aborescens are available commercially. Free Anthroquinones and their derivatives like Barbaloin-IO-aloe emodin-9 anthrone, Isobarbaloin and chromones in Aloe Vera leaves exert a strong purgative effect and are potent anti-microbial agents[1].

Aloe vera has a lot to offer in the field of dentistry, a lot of studies is on the way to utilize the effective antimicrobial property of the miracle plant. Interest is gathering for the use of aloe vera in dentistry and this natural therapy is already proved its unlimited use in our field. [2]

AVG has various pharmacological actions like being antibacterial, anti-fungal.

Among the natural products, aloe vera had shown antifungal property on the heat-cured acrylic denture base. The availability, cost-effectiveness, and colossal advantages make this herb one of the best alternatives to the present denture cleansing tablet agents that are used. It is, therefore, essential to explore natural plant-based medicines in developing countries where citizens are sometimes incapable to purvey expensive devout medicine.[3]

AVG had also been used in dentistry and showed valuable results. Ikmal Hisham Ismail et al studied the use of aloe vera as an intracanal medicament and found effective results as an antimicrobial agent.[4]

Chlorhexidine (CHX) is a bisbiguanide that was developed in the 1940s in the UK and has been marketed as a general disinfectant. In the 1970s, its antiplaque activity

was discovered, and by 1976 it was available as a mouthwash. Oral biofilm and its associated bacteria have been linked to the pathogenesis of various oral diseases including halitosis, caries, gingivitis, and periodontitis. Often the bacterial contamination of implant placement may result in biofilm formation and early failure; therefore, a 1-minute pre-operative rinse of 0.2% CHX is recommended to reduce the bacterial load.[5]

It also has bactericidal effect, causing cell membrane rupture and consequent leak of intracellular fabric, including potassium (at low concentrations) or throw respiratory inhibition and nucleic acid loss (at high concentrations). CHX inhibits glycosyltransferase and a pair of phosphoenolpyruvate phosphotransferases, enzymes necessary for the function and maintenance of the bacterial glycolytic pathway.

In addition to yeast, CHX has a wide range of activity against Gram-positive and Gram-negative microorganisms. CHX is dependent on the type of microorganism. Gram-positive bacteria are more susceptible than Gram-negative bacteria.[6] In dentistry, CHX products are used therapeutically as well as prophylactically.

The subject of the study chosen are diabetic individuals as they are more prone to infections and delayed wound healing due to variable microbiota.

AIM

To compare the effect of herbal and chemical disinfectants on alginate impression made from diabetic (type II) dentulous patients.

OBJECTIVES

1- To study the efficacy of chemical disinfectant on alginate impression made from diabetic dentulous patients.

2- To study the efficacy of herbal disinfectant on alginate impression made from diabetic dentulous patients.

3- To compare the efficacy of herbal and chemical disinfectants on alginate impression made from diabetic dentulous patients.

1) L. 2. G. Touyz and M. Rosen (1991)[7] studied the disinfection of alginate impression material using disinfectants as mixing and soak solutions and concluded that the mix and soak technique described was more effective at disinfecting the alginate tested than water-mixed alginate alone, or water-mixed alginate which was subsequently soaked in disinfectant. Chlorhexidine gluconate 0.2 per cent aqueous solution was effective and was the disinfectant of choice.

2) M. R. Leonardo et al (1999)[8] did a study to evaluate the in vivo antimicrobial activity of 2% chlorhexidine gluconate (FCFRP-USP) used as a root canal irrigating solution in teeth with pulp necrosis and radiographically visible chronic periapical reactions and concluded that chlorhexidine prevents microbial activity in vivo with residual effects in the root canal system up to 48 h.

3) D E Slot et al (2007)[9] did a study to assess the effect of application of 0.12% CHX dentifrice gel on de novo plaque accumulation and concluded that application of 0.12% CHX dentifrice gel is not significantly different from application of regular dentifrice on plaque accumulation. Use of a 0.12% CHX mouthwash is significantly more effective. CHX-DGel appears a poor alternative for a dentifrice. It is not an effective inhibitor of plaque growth and does not possess fluoride.

4) J. Hintao et al (2007)[10] studied the microbiological profiles of saliva, supragingival and subgingival plaque and dental caries in adults with and without type 2 diabetes mellitus and concluded diabetic subjects had higher levels of Treponema denticola, Prevotella nigrescens, Streptococcus sanguinis, Streptococcus oralis and Streptococcus intermedius in their supragingival plaque than non-diabetic subjects.

5) Siribang-on Piboonniyom Khovidhunkitet al (2009)[11] studied Xerostomia, hyposalivation, and oral microbiota in type 2 diabetic patients and concluded that xerostomia and hyposalivation were prevalent in patients with type 2 DM and were associated with higher numbers of oral pathogens in the saliva. Patients with hyposalivation had significantly higher numbers of mutans streptococci, Lactobacillus spp., and Candida spp. in the saliva compared with those without hyposalivation.

6) Dr Reena Kulshrestha et al (2011)[12] studied comparison of oral microflora of diabetic and non-diabetic patients with periodontitis and concluded that numerous oral changes were seen in diabetic patients such as predominance of Candida sps., Hemolytic Streptococci, Staphylococci, Porphyromonas sps., Actinobacillzs sps.

7) Hanoem EH et al (2011)[13] studied the effectiveness of mimba oil (Azadirachta indica A. Juss) spray disinfectant on alginate impression and concluded that 50% mimba oil as disinfectant was already effective decreases microorganism colonies in the alginate impression.

8) Mohamad Rafiul Ahsan et al (2012)[14] studied the antimicrobial effect of disinfecting solutions on alginate impression materials and concluded that both disinfectant solutions (1% sodium hypochlorite and 2% glutaraldehyde) significantly reduced microbial count from alginate impression surface. Among them 2% glutaraldehyde showed more antimicrobial effect than 1% sodium hypochlorite. It was concluded that rate of bacterial transmission from alginate impression to cast was significantly reduced in case of 1% sodium hypochlorite solution than 2% glutaraldehyde solution.

9) V Zand et al (2012)[15] did a study to evaluate the effectiveness of different concentrations of Chlorhexidine (CHX) and sodium hypochlorite (NaOCl) in disinfecting contaminated Resilon cones within one minute and concluded that chlorhexidine is unable to disinfect Resilon cones during one-minute exposure.

10) Hamid Badrian et al (2012)[16] studied the effect of three different types of disinfectant agents (hypochlorite sodium 0.525%, epimax, deconex) on alginate impression material after 5 and 10 minutes and concluded that epimax showed effective results in 10 minutes as it completely eradicated all kinds of microorganisms.

11) Satheesh B. Haralur et al (2012)[17] studied the efficacy of sodium hypochlorite (1: 10) and iodophor disinfectants on alginate impressions along with their effect on the survived bacterium count on the gypsum cast and concluded that sodium hypochlorite (1: 10) preceded with water rinsing was the best disinfectant for disinfecting alginate impression. The least number of bacterial colonies were found on the dental cast made from these impressions.

12) Bajaj et al (2012)[18] studied oral manifestations in type-2 diabetes and related complications in total of 50 cases of DM with oral manifestations and concluded that the majority were observed to have periodontal disease- 34%, followed by oral candidiasis in 24%, tooth loss in 24%, and dental caries in 24%. Other complications included oral mucosal ulcers in 22%, taste impairment in 20%, halitosis in 16%, xerostomia and salivary gland hypofunction in 14%, and burning mouth sensation in 10%. And significant oral complications were found in patients of DM. FBG and PPBG were significantly higher among diabetics with oral manifestations compared to those without diabetes mellitus. Microvascular and macrovascular complications of DM were found to be significantly higher among diabetics with oral diseases.

13) Joana Correia-Sousa et al (2013)[19] studied the effect of water and sodium hypochlorite disinfection on alginate impressions and concluded that the sodium hypochlorite disinfection was an efficient disinfection method and tap water rinsing could reduce microbial load but does not disinfect efficiently dental impression materials, so, additional methods should be used.

14) Mohammed T. Al-Khafagy (2015)[20] studied the effects of natural disinfectant solutions on dimensional stability of silicon impression material and concluded that very high viscosity polysiloxane impression can be disinfected with apple vinegar (natural solution) for purpose of primary impression for completely or partially edentulous arches, as well as producing opposing casts in prosthodontics treatment, making interocclusal devices, and surgical guides.

15) Yoshihisa Yamashita et al (2017)[21] studied the oral microbiome and human health and concluded that most organisms in the salivary microbiota were present in almost all individuals, including Streptococcus, Neisseria, Rothia, Prevotella, Actinomyces, Granulicatella, Porphyromonas, Haemophilus, and Porphyromonas species. Data collected on bacterial composition from 2,343 participants suggested that these predominant organisms comprise two different cohabiting groups of bacteria: and one mainly composed of Prevotella histicola, Prevotella melaninogenica, Veillonella parvula, Veillonella atypica, Streptococcus salivarius, and Streptococcus parasanguinis (bacterial cohabiting group I), the other primarily assembled from Neisseria flavescens, Haemophilus parainfluenzae, Porphyromonas

pasteri, Gemella sanguinis, and Granulicatella adiacens (bacterial cohabiting group II).

16) Jirong Long et al (2017)[22] studied the comparsion of oral microbiome profiles of 98 participants with incident diabetes, 99 obese non-diabetics, and 97 normal weight non-diabetics, via deep sequencing of the 16S rRNA gene. They concluded that multiple bacteria taxa in the phylum Actinobacteriaare was associated with risk of type 2 diabetes. Some were also associated with the prevalence of obesity, suggesting that the oral microbiome may play an important role in diabetes etiology.

17) Luisa Fernanda Gómez Chabala et al (2017)[23] studied the release behavior and antibacterial activity of Chitosan/Alginate Blends with aloe vera and silver nanoparticles and concluded that the synergic effect between alginate, chitosan, aloe vera gel and the AgNps resulted in a promising alternative to be used in antibacterial applications. This alternative method could help to decrease the secondary effects of antibiotics that were commonly used in wound treatments and the advantage being the matrices developed promote wound healing through their chemical characteristics.

18) Jonathan Tam et al (2018)[24] studied obesity altering composition and diversity of the oral microbiota in patients with type 2 diabetes mellitus independently of glycaemic control and concluded that obesity was significantly associated with the oral microbial composition. The impact of glycaemic control on oral microbiota, however, could not be assured statistically.

19) Datla Durga Devi et al (2018)[25] studied the microbial load on impressions and the efficacy of various disinfectants on reducing microorganisms from the impression surface after disinfection and concluded that 2% Glutaraldehyde showed higher efficacy in reducing the microflora compared to Dimenol spray and UV radiation. There was complete removal of microorganisms with Microwave radiation.

20) Gopal Krishna Choudhury et al (2018)[26] studied the disinfection efficacy of epimax and 0.525% sodium hypochlorite on alginate impression over a period of 10 minutes and concluded that both Epimax and 0.525% sodium hypochlorite could disinfect the alginate impression material against C. albicans, P. aeruginosa, and S.

aureus. However, epimax was found to be more effective against S. aureus as compared with 0.525% sodium hypochlorite.

21) Priti Jha et al (2019)[27] studied the efficacy of organic disinfectant (Ecosan-an herbal disinfectant with primary active ingredient as natural polymer of glucosamine) which has similar characteristic and structure of honey. The presence of quaternary ammonium was used as an emulsifying and it was concluded that in comparison to using water for cleaning alginate impression Ecosan® proves to be a promising natural disinfectant for dental impressions but requires further studies & comparison with other chemical disinfectants.

22) Ameena Nausheen et al (2019)[28] studied the effect of different chemical and herbal disinfectant solutions on the mechanical and physical properties of gutta-percha: and concluded that aloe vera gel at 90% was considered as a safer GP disinfectant as it did not alter the tensile strength and topography of GP, which eventually would lead to enhanced sealing ability and reinforcement of the root canal. Sodium hypochlorite solution at 5.25% would decrease the tensile strength and left a numerous pitting on the surface of GP cones.

23) Bahare Salehi et al (2019)[29] studied plant-derived bioactives in oral mucosal lesions. They emphasized on curcumin, lycopene, chamomile, aloe vera, green tea and coffee properties and concluded that A. vera mouthwash may prevent radiation-induced mucositis by promoting wound healing and reducing inflammation. Furthermore, A. vera antifungal and immunomodulatory effects could reduce oral candidiasis severity in patients with head and neck radiotherapy.

24) Shelly Withers et al (2019)[30] studied the oral microbiome & systemic disease (diabetes) and concluded that diabetic patients with active periodontal disease could have more difficulty controlling it due to increased inflammation and insulin resistance along with reduced ability to regulate glucose. There was also a strong microbial component that made management more difficult, for instance, a study that utilized 16S rRNA gene sequencing noted significant differences between subgingival microbiota in patients with Type 2 diabetes and those without diabetes.

25) RobertoFarina et al (2019)[31] studied whole metagenomic shotgun sequencing of the subgingival microbiome of diabetics and non-diabetics with different periodontal conditions and concluded that there was significantly higher relative abundance of Anaerolineaceae bacterium oral taxon in patients with moderate to severe periodontitis vs patients without history of periodontitis, which was maintained when the comparison was restricted to type 2 diabetics.

26) Azadeh Farhang Nia et al (2020)[32] studied the comparative evaluation of antimicrobial activity of chlorhexidine (CHX) and silver nanoparticles (AgNPs) combined with irreversible hydrocolloid and concluded that the antimicrobial activity of AgNPs at 0.1 and 0.2% against the five tested bacterial strains were similar to those of pure CHX 0.2% solution and CHX 0.2% mouthwash.

27) Ayesha Al Shikh et al (2020)[33] studied the effectiveness of alcohol and aldehyde spray disinfectants on analogue dental impressions in hospital setting and concluded that alcohol-based spray disinfection of dental impressions could be less effective than aldehyde spray and full immersion of impressions was recommended. Careful wetting or soaking of all surfaces of impressions was very important when using a spray.

28) Divya Dharshini A et al (2020)[34] studied the effect of role of disinfectants on alginate impression materials and concluded that it had the most effective method of reducing the burden of microorganisms from alginate without any change in accuracy was chlorhexidine, when it was used as liquid for alginate preparation and post-setting disinfection solution.

29) Tandi Matsha et al (2020)[35] studied oral microbiome signatures in diabetes mellitus and periodontal Disease and concluded that In individuals with prediabetes or DM, Fusobacteria and Actinobacteria were significantly more abundant.

30) Daniel Belstrøm (2020)[36] studied the salivary microbiota in health and disease and concluded that as per several studies done compairing SM (salivary microbiota) in patients with diabetes to that of healthy controls by means of NGS, and in general, data showed that diabetes associates with a decrease in bacterial diversity of SM . In addition, higher salivary levels of P. gingivalis, T. forsythia and F. alocis were reported in patients with gestational diabetes, whereas only minor differences were

identified in children with T2DM, when compared to obese and healthy controls, respectively.

31) Dr.Priyanka Rathod et al (2021)[37] studied the evaluation of the anti-microbial properties of prepared herbal solution on dental impressions using irreversible hydrocolloid and concluded that herbal disinfectant solution could be considered as effective antimicrobial and could be used as a disinfectant agent and it also prevented deleterious effect on alginate impression.

32) Monica Kotwal et al (2021)[38] studied the disinfection of impression materials with glutaraldehyde, ultraviolet radiation, and autoclave and concluded that autoclave was the better method of sterilization compared to the use of glutaraldehyde, UV radiation, and herbal disinfectant.

33) Kandasamy B et al (2021)[39] studied the comparative Assessment of Sodium Hypochlorite, UV Radiation, aloe vera and microwave irradiation for disinfection of impression materials and concluded that the sodium hypochlorite was the better method of sterilization along with microwave irradiation. Whereas UV radiation and aloe vera were also effective as a disinfectant.

34) Forouzande Badooei et al (2021)[40] studied the comparison between the effects of ginger and aloe vera mouthwashes on the xerostomia in patients referred to Bandar Abbas diabetes clinic (Iran). They concluded that the ginger and aloe vera mouthwashes significantly reduced all symptoms and severity related to xerostomia.

36) Miguel Ángel González-Moles et al (2021)[41] studied the state of evidence on oral health problems in diabetic patients and concluded that DM patients had a special predisposition to the development of fungal infections, especially of the Candida sp. genus, with significantly higher rates of oral mucosa colonization by Candida sp. both in patients with DM1 (85%) and DM2 (68%) compared to non- diabetics (27%).

37) Tamanna Ali et al (2021)[42] studied Type-2 Diabetes Mellitus Individuals Carry Different Periodontal Bacteria and they aimed to identify etiologic microbiota associated periodontal diseases among diabetes patients and the factors related to the most commonly identified bacteria species and they concluded that Type-2 diabetes mellitus was associated with a higher amount of dental plaques. Periodontal plaque

samples from diabetic and non-diabetic subjects possess differential microbial communities. Diabetic plaques contained more versatile microbes predominated by gram-positive streptococci and staphylococci.

38) Yun-kunLiu et al (2021)[43] studied salivary microbiome-based auxiliary diagnostic model for type 2 diabetes mellitus and concluded that salivary microbiome for treatment-naive type 2 diabetes mellitus patients was imbalanced with certain taxa, including Slackia, Mitsuokella, Abiotrophia, and Parascardovia that being significantly dominant, while the abundance of Moraxella was high in healthy controls. Diabetic patients exhibited varying levels of Prevotella nanceiensis and Prevotella melaninogenica which were negatively correlated with glycosylated hemoglobin and fasting blood glucose levels, as well as fasting blood glucose levels, respectively.

40) Xian Peng et al (2022)[44] studied oral microbiota in human systematic diseases and concluded that when compared with non-diabetic periodontitis patients, the community structure of the subgingival microbiome of diabetic and periodontitis patients had undergone significant changes, and a variety of bacteria between the two were differentially enriched. Oral microorganisms could trigger insulin resistance by influencing the body‟s immune inflammation and oxidative stress, thereby affecting the process of diabetes.

The study was conducted in the Department of Prosthodontics and Crown & Bridge, at Babu Banarasi Das College of Dental Sciences, Lucknow, to compare the effect of herbal and chemical disinfectants on alginate impression made from diabetic dentulous patients.

Study Sample and size

Total no. of sample /specimens -30

Partially dentulous patients reporting to the Department of Prosthodontics, were selected for study as per the inclusion and exclusion criteria.

The study was approved by the ethical Committee of Babu Banarasi Das College of Dental Sciences, BBD University.

The number allotted to the study is

IEC CODE: <u>ARMAMENTARIUM</u>

The Materials and instruments that were used during the course of this study.

. Materials and equipments used in the study: -

1-Alginate (Neoalgin)

2-Disinfectants – aloe vera (Patanjali-94%) and chlorhexidine (V-consept 2%)

3-Media – blood agar plates, tryptic soy broth (TSB - Hi Media)

4-Laminar air flow (Mangat Ram and Sons)

5-Normal saline

6-Impression trays (SS White)

7-Incubator (Surgico Industries)

8- Rubber bowl

9- B.P Handle (no.4) & blade(no.23)

10-Dental probe

11- Twizzer

12- Plastic spatula

13- Cotton holder

14- Mouth mirror

FIGURE 1: Chlorhexidine (V-consept 2%)

FIGURE 2: Normal saline

FIGURE3- Alginate (Neoalgin)

FIGURE 4: Aloevera (Patanjali)

FIGURE 5: Impression trays (SS White)

FIGURE 6: TSB MEDI

FIGURE 7: Glass test tubes

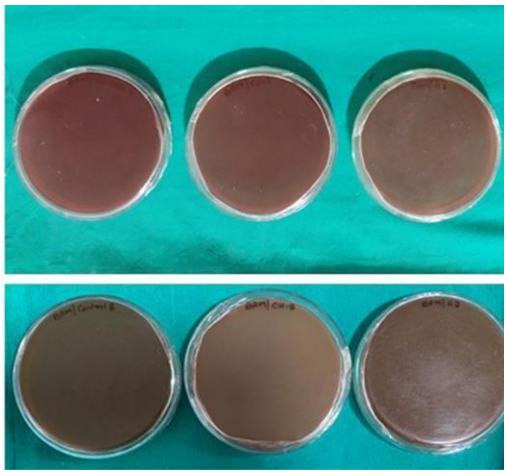
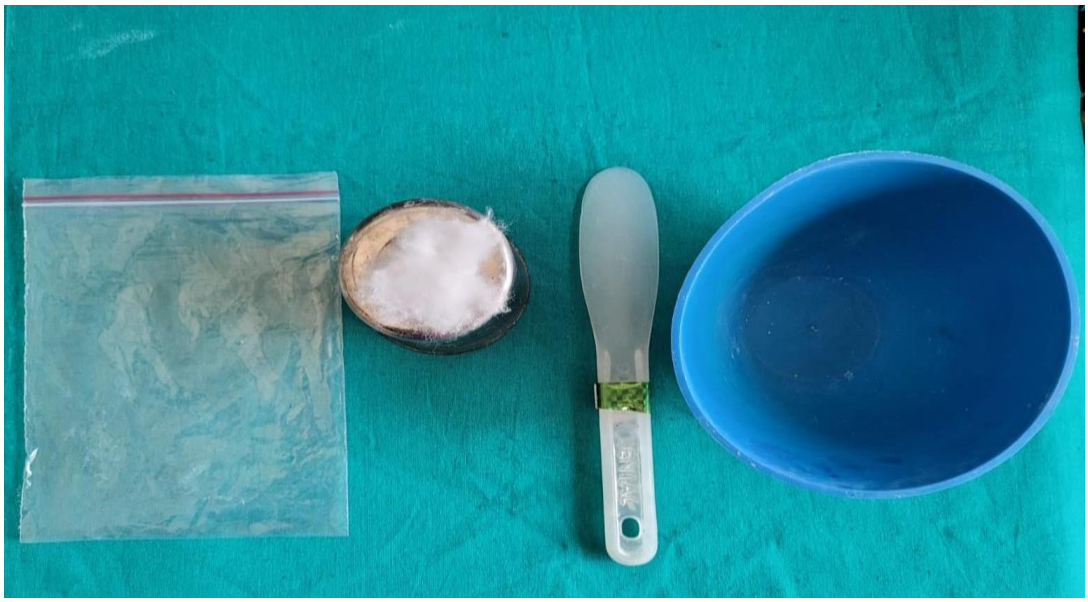


FIGURE 8: Blood agar media

FIGURE 9: Incubator

FIGURE 10: Laminar flow



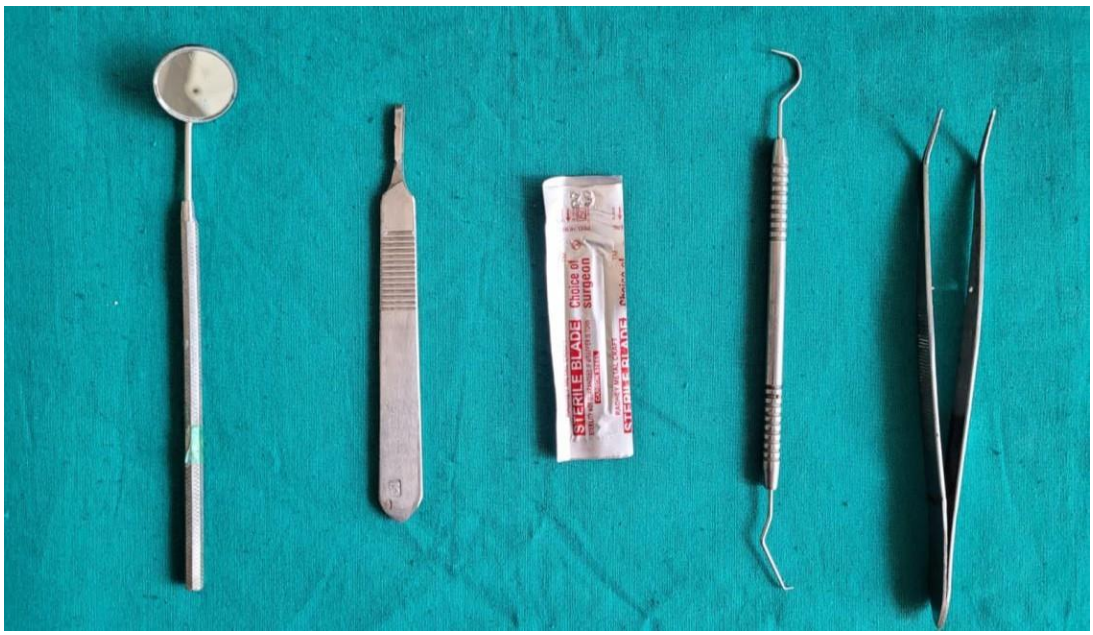


FIGURE 11: Miscellaneous instruments

Selection Criteria

Inclusion criteria

- Age (45to 60years),
- Diabetic type II,
- Dentulous patients.

Exclusion Criteria

- Non diabetic,
- Completely edentulous,
- Neuromuscular disorders.
-

METHODOLOGY

IMPRESSIONS MAKING:

Appropriate maxillary stock tray was selected and alginate impression is made from diabetic dentulous patients. Inspection of the impression was done for any errors. If no error was present, the impression was divided into three parts. Three parts were taken from palate region to maintain the standardization of the samples. Two longitudinal cuts were were treated made from different individuals following the mentioned criterias. The samples were cut and stored separately in zip lock bags.

FIGURE 12: 3 blocks of approximately similar sizes (0.5 x 0.5 inches) from the palate region.

MICROBIOLOGICAL ANALYSIS EVALUATION:

Statistical analysis would be done for the results obtained.

1. To investigate the effect of different disinfectant materials i.e., Chlorhexidine and Aloe Vera gel (Patanjali), 30 samples were used for the work.
2. All 30 samples (alginate impressions) were cut into 3 blocks of approximately similar sizes (0.5 x 0.5 inches) from the palate region.
3. The block to be treated with aloe vera were marked as “A”, chlorhexidine as “CH” and no treatments as “Control” for each sample respectively.
4. The blocks of impressions were then placed in respective disinfectants in sterile beakers marked appropriately and left for 10 minutes time period. Whereas the block marked as “control” indicating no treatment were placed in clean sterile dry beaker covered for 10 minutes.
5. 3-The antimicrobial effect of disinfectants is studied by three methods namely: rinse, spray, immersion methods. In the present study immersion method was used. It has been earlier reported that Spraying and immersing methods are almost equal while mere water rinsing showed no significant disinfection effects.[16]
6. On the other hand, TSB broth was prepared and sterilized (autoclaving at 121C, 15 psi, 25 minutes) in separate test tubes.
7. On completion of 10 minutes incubation period of blocks in disinfectant, clean and sterile forceps were used to take out the blocks and transfer to the cooled and sterilized TSB broth.
8. TSB broth were then incubated at 37°C for 24 hours.
9. Blood Agar plates were prepared by using standard composition of Hi-Media according to manufacturer‟s instruction.
10. 0.1ml of inoculum was transferred from TSB broth onto sterile Blood Agar Plates and spread using L-shaped sterile rod.
11. Plates were then sealed with parafilm and placed in inverted position in the incubator at 37°C.
12. Colonies were counted at an incubation period of 24, 48 and 72 hours on each plate.

13. CFU/ml were calculated on each plate using formula CFU/ml = No. colonies observed x Dilution factor/Volume of sample plated (In this case no dilution was made).

FIGURE 13: Testing of cut blocks by immersion method in Aloe vera & CHX

Test: Determination of Colony forming unit (CFU/ml) on disinfectant treated Alginate impressions

Test sample: Alginate impressions

Sample No./name: 30 / Control, A (Aloe Vera treated impressions), CH (Chlorhexidine treated impressions)

Sample code: Control1-30, CH1- CH30, A1-A30

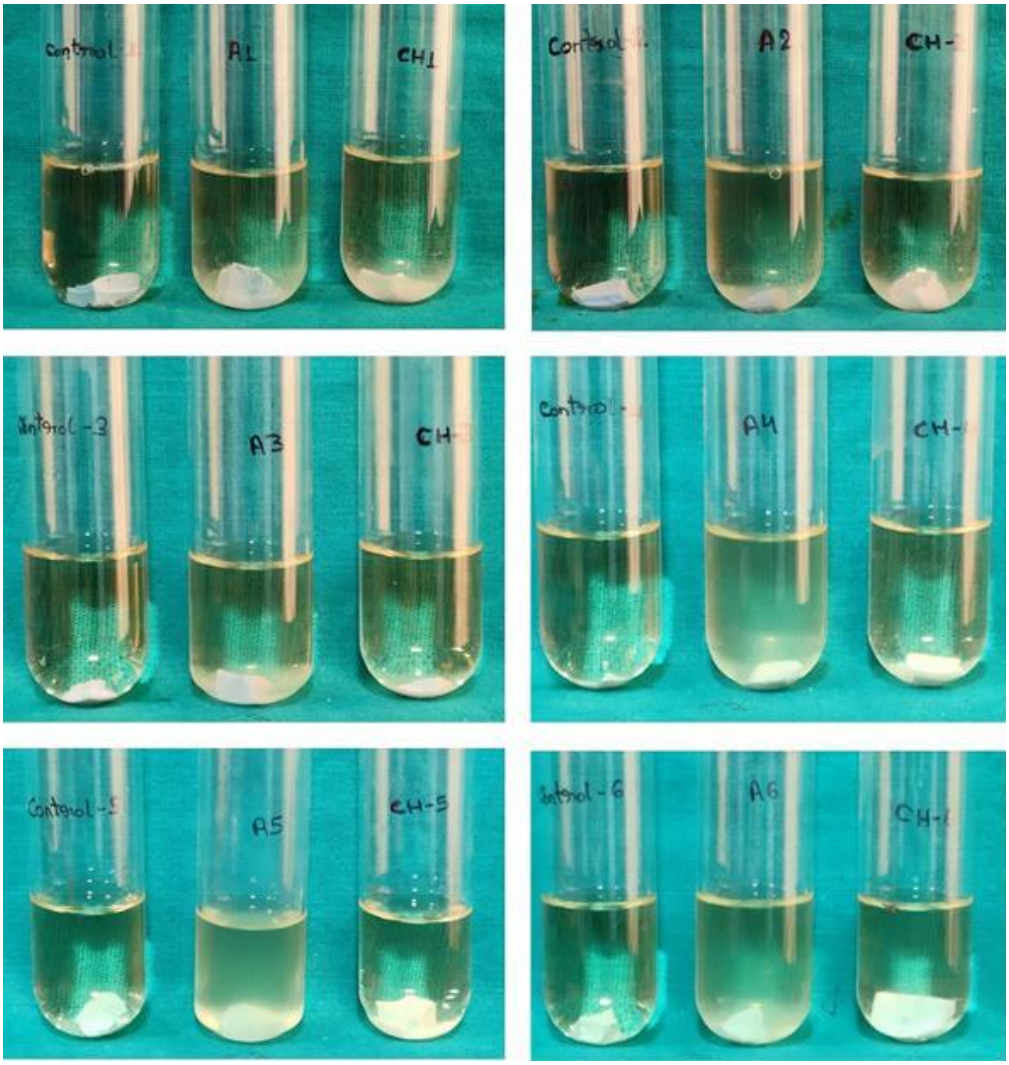


FIGURE 14: Control: No disinfectant treatment; CH: Chlorhexidine treated; A: Aloe Vera treated

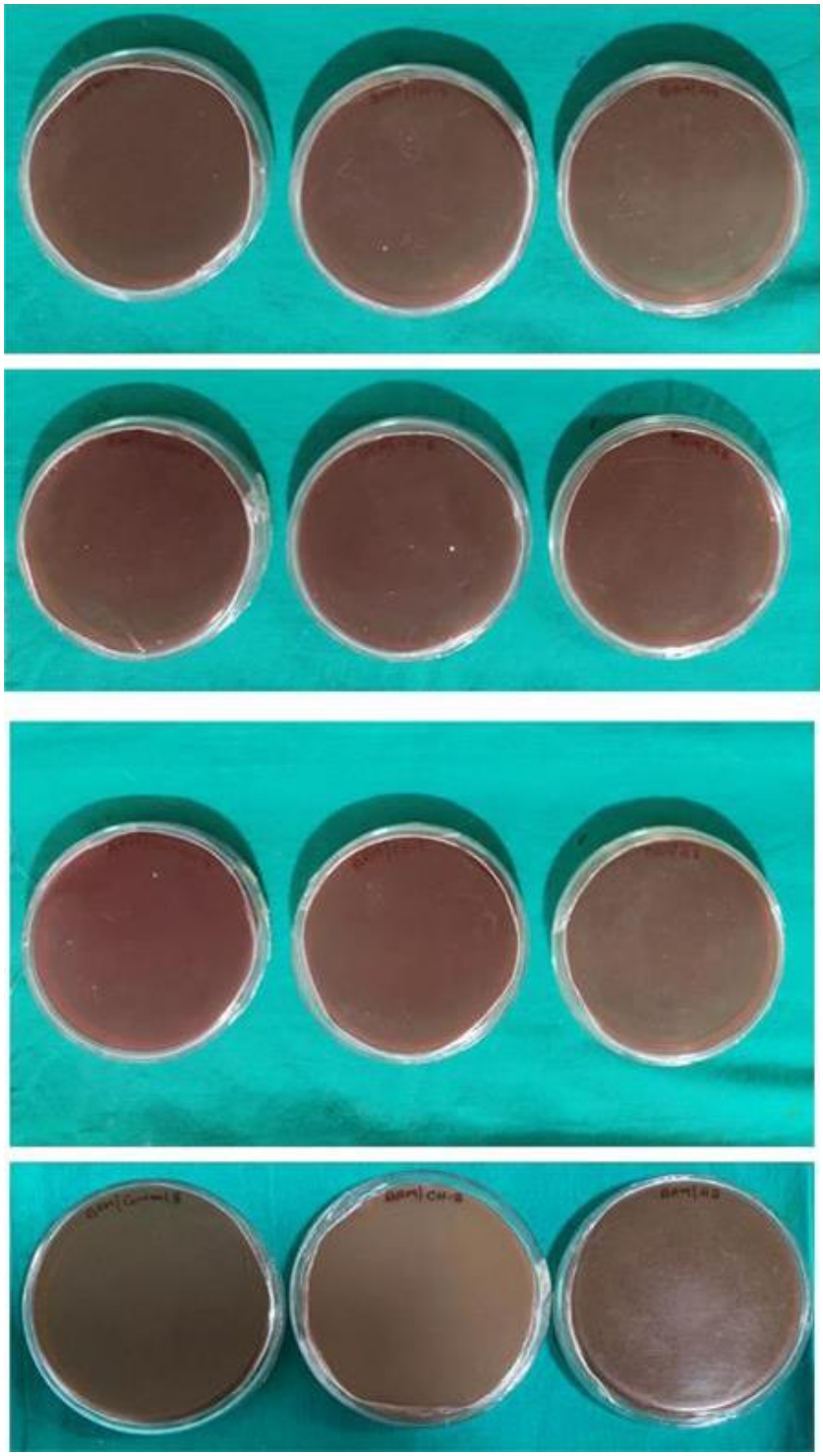


FIGURE 15: Initial Plates images before incubation: Samples Blood Agar Media

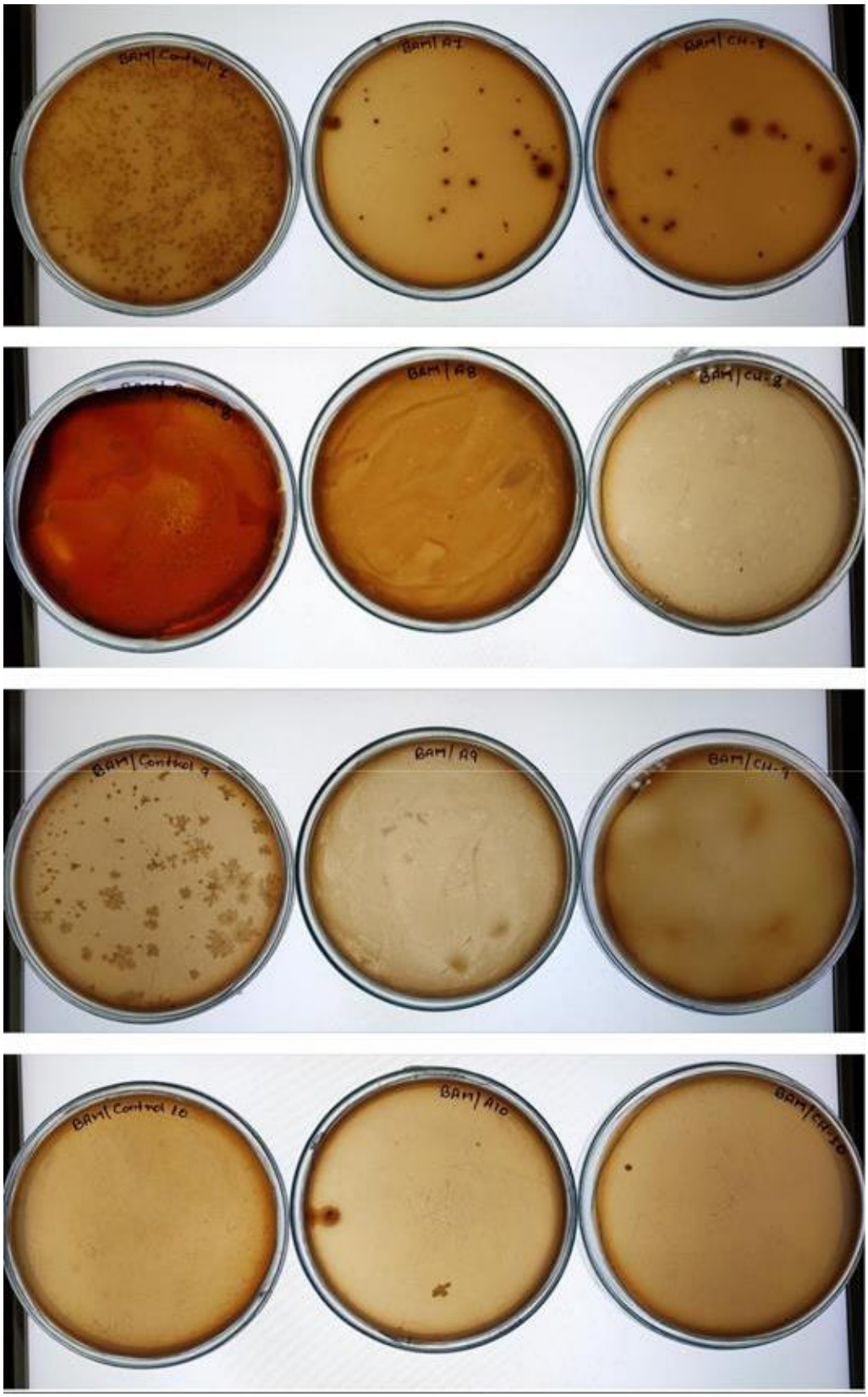


FIGURE 16: Plate images after 24 Hrs incubation

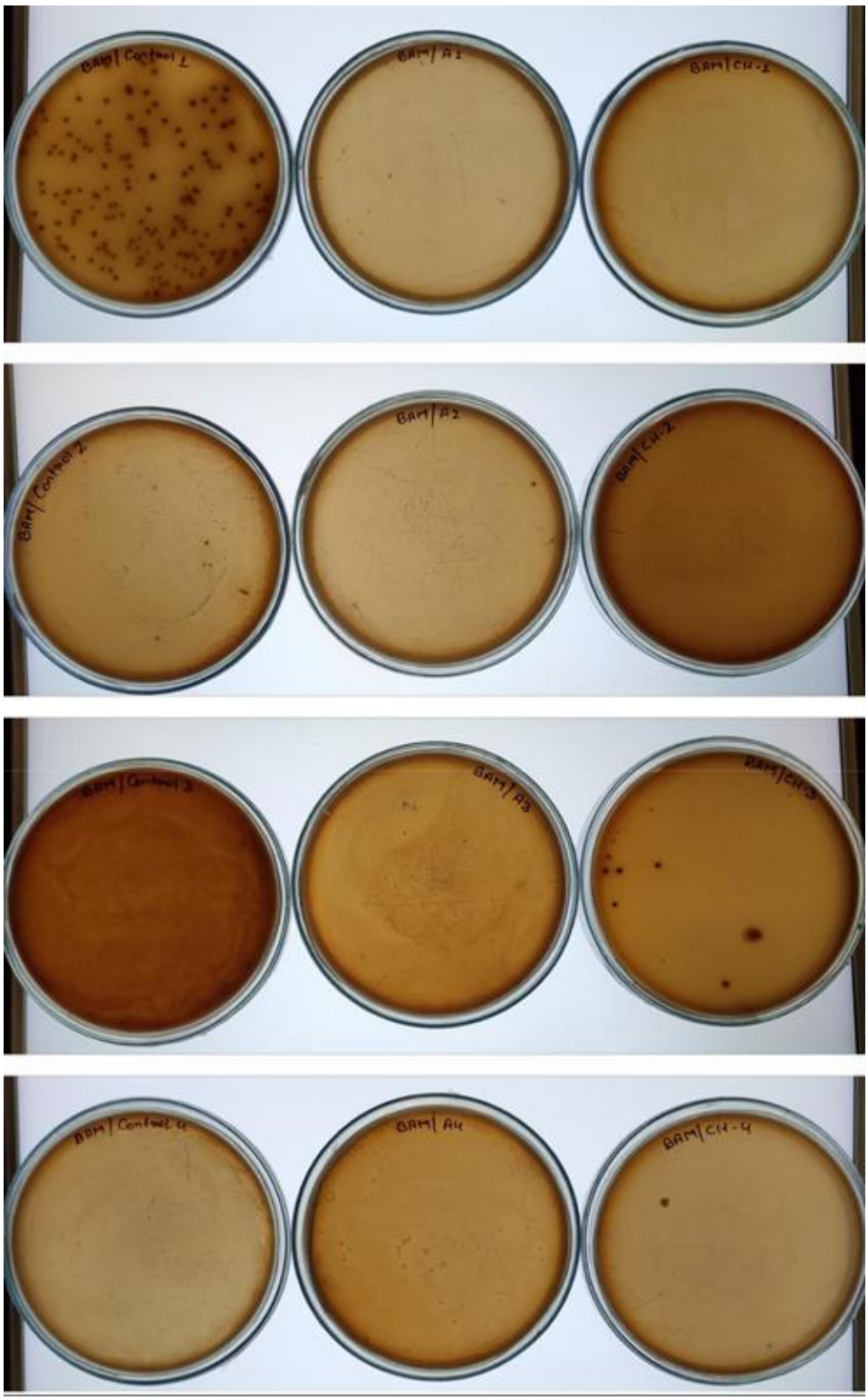


FIGURE 17: Plate images after 48 Hrs incubation

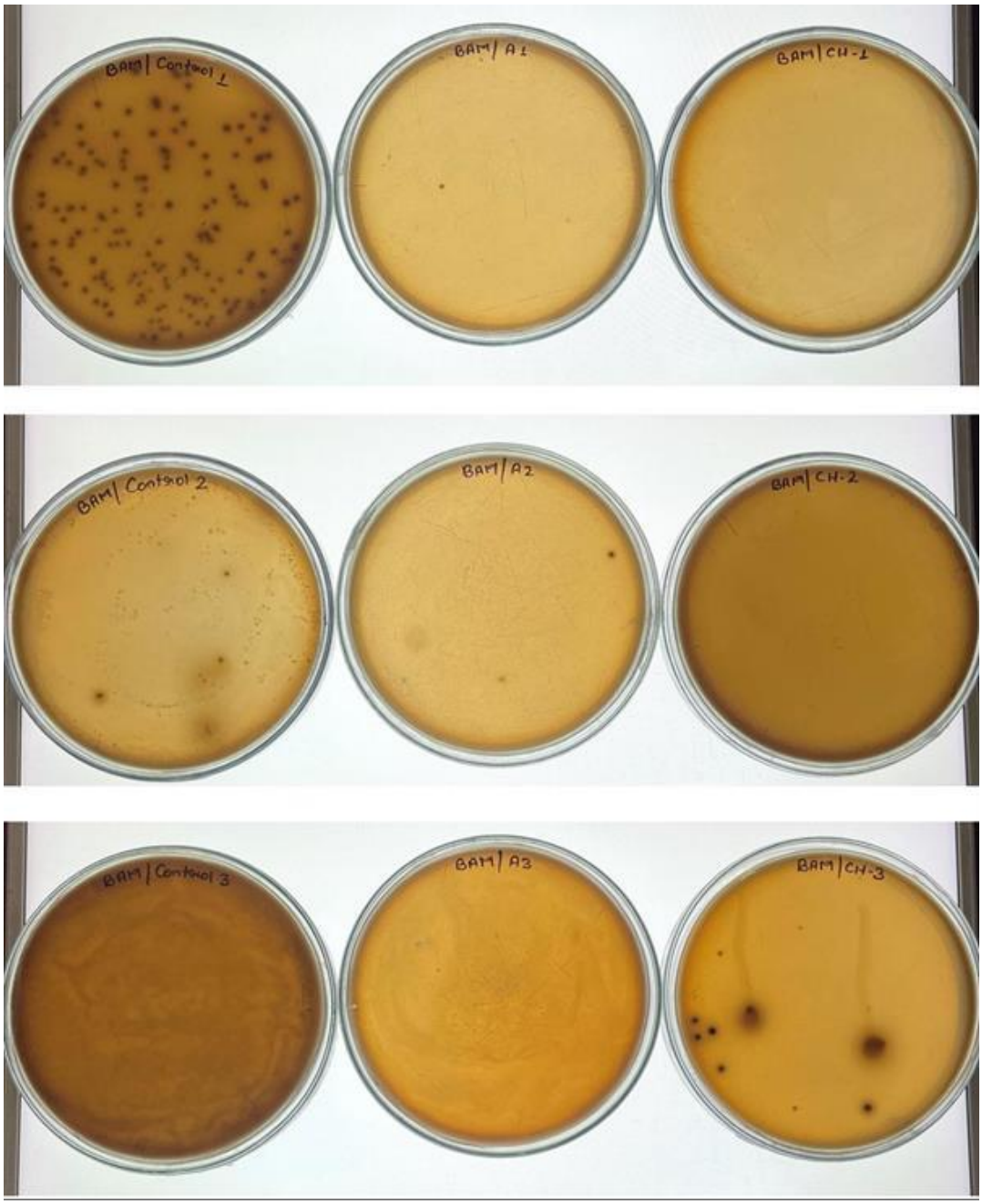


FIGURE 18: Plate images after 72 Hrs incubation

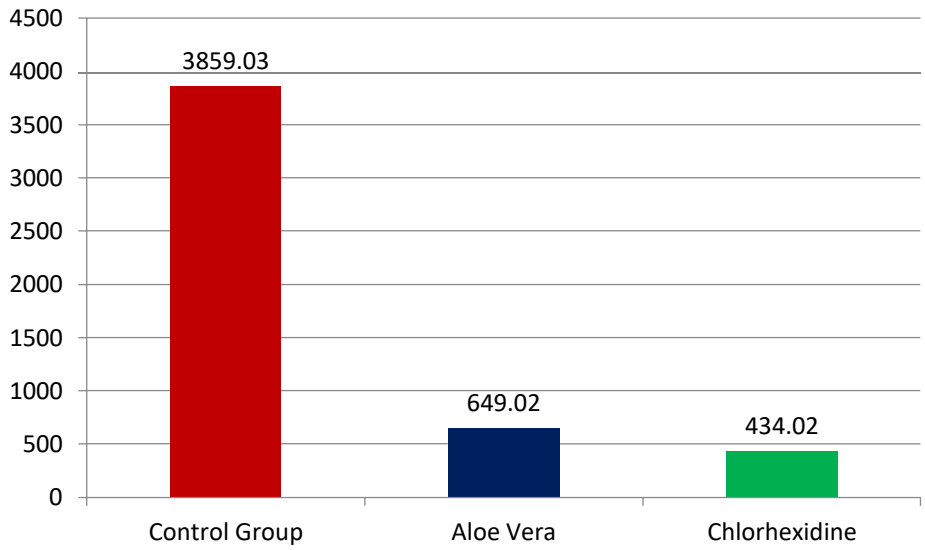
The data for the present study was entered in the Microsoft Excel 2007 and analyzed using the SPSS statistical software 23.0 Version (IBM Corp. Released 2015. IBM SPSS Statistics for Windows, Version 23.0. Armonk, NY: IBM Corp.). The descriptive statistics included mean, standard deviation. The intragroup comparison for the different time intervals was done using paired t test to find the difference between the individual time intervals The level of the significance for the present study was fixed at 5%.

The intergroup comparison for the difference of mean scores between independent groups was done using the One Way ANOVA and post Hoc Tukey analysis

The Shapiro–Wilk test was used to investigate the distribution of the data and Levene‟s test to explore the homogeneity of the variables. The data were found to be homogeneous and normally distributed. Mean and standard deviation (SD) were computed for each variable

CFU AT 24 HRS IN THE THREE GROUPS

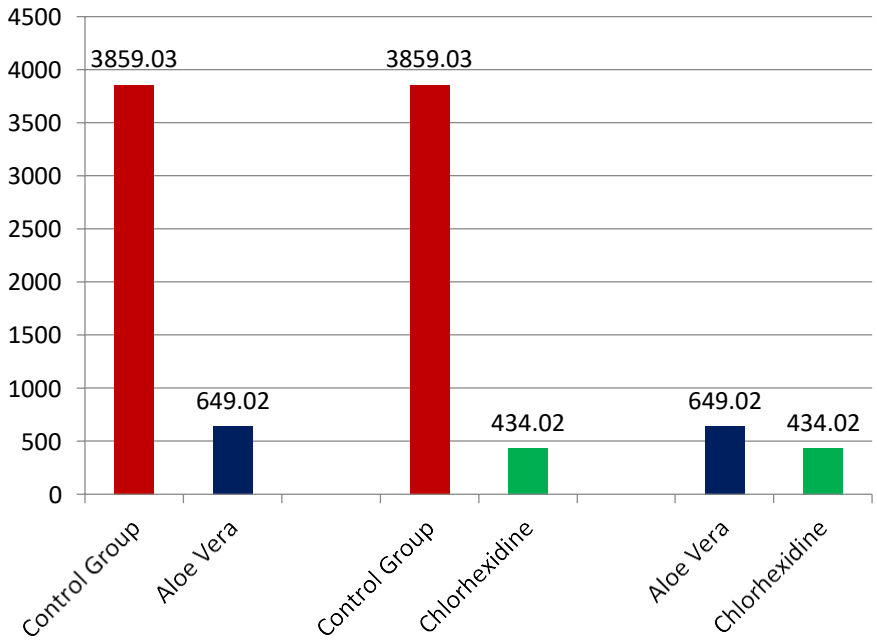
	Mean	Std. Deviation	Std. Error	Minimum	Maximum
Control Group	3859.03	1798.08	328.28	680.00	7000.00
Aloe Vera	649.02	873.46	159.47	10.00	2300.00
Chlorhexidine	434.02	846.54	154.55	.00	2560.00



The mean CFU in the control group at the 24 hrs time interval was 3859.03±1798.08. In the aloevera group the mean CFU were 649.02±873.46 and in the Chlorhexidine group the mean CFU were 434.02±846.54. The mean CFU were highest in the Control Group followed by Aloe vera and least in the Chlorhexidine group. The statistical difference in CFU between the three groups was analyzed using the One Way ANOVA followed by post hoc analysis at 5% significance level (p=0.05)

Intergroup comparison of CFU between three groups at 24 hrs

	Mean	Std. Deviation	Std. Error	P value	Significance
Control Group	3859.03	1798.08	328.28	0.001	Significant
Aloe Vera	649.02	873.46	159.47		
	Mean	Std. Deviation	Std. Error		
Control Group	3859.03	1798.08	328.28	0.001	Significant
Chlorhexidine	434.02	846.54	154.55		
	Mean	Std. Deviation	Std. Error		
Aloe Vera	649.02	873.46	159.47	0.508	Non-Significant
Chlorhexidine	434.02	846.54	154.55		

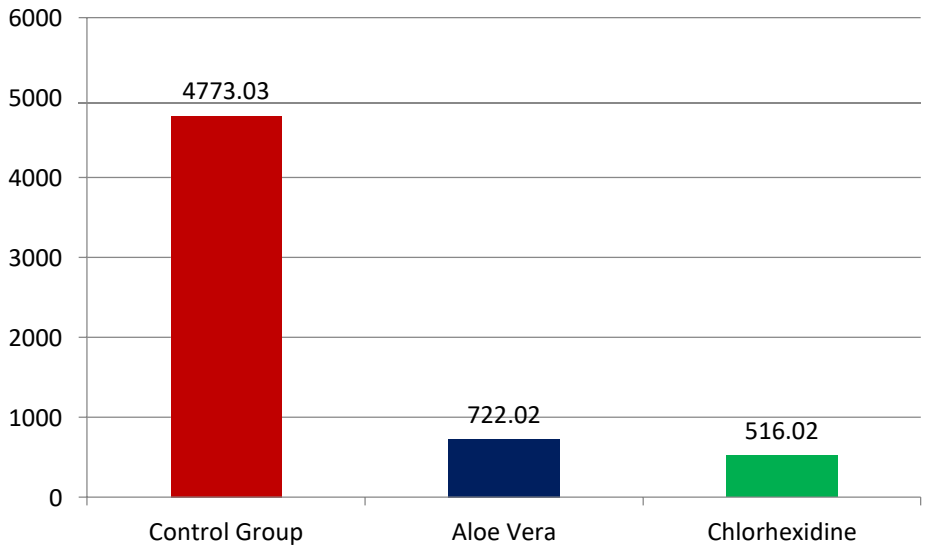


The mean CFU in the control group at the 24 hrs time interval was 3859.03±1798.08. In the aloevera group the mean CFU were 649.02±873.46 and in the Chlorhexidine group the mean CFU were 434.02±846.54. The mean CFU were highest in the Control Group followed by Aloe vera and least in the Chlorhexidine group. The intergroup difference in CFU between the Control group and Aloe vera Group was statistically significant with p value of 0.001 signifying a highly significant difference between the groups, The intergroup difference in CFU between the Control group and Chlorhexidine Group was statistically significant with p value of

0.001 signifying a highly significant difference between the groups. The intergroup difference in CFU between the Aloe Vera group and Chlorhexidine Group was statistically non-significant with p value of 0.508

CFU AT 48 HRS IN THE THREE GROUPS

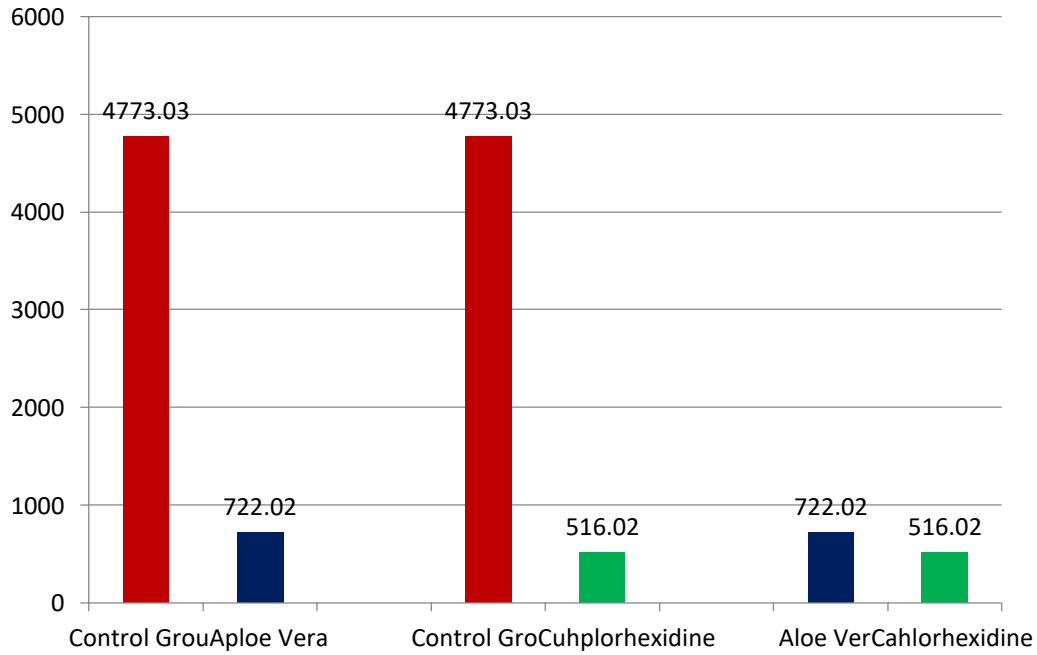
	Mean	Std. Deviation	Std. Error	Minimum	Maximum
Control Group	4773.03	2304.38	420.71	770.00	8000.00
Aloe Vera	722.02	984.69	179.77	10.00	2570.00
Chlorhexidine	516.02	1047.90	191.32	.00	3300.00



The mean CFU in the control group at the 48 hrs time interval was 4773.03±2304.38. In the aloevera group the mean CFU were 722.02±984.69 and in the Chlorhexidine group the mean CFU were 516.02±1047.90. The mean CFU were highest in the Control Group followed by Aloe vera and least in the Chlorhexidine group. The statistical difference in CFU between the three groups was analyzed using the One Way ANOVA followed by post hoc analysis at 5% significance level (p=0.05)

Intergroup comparison of CFU between three groups at 48 hrs

	Mean	Std. Deviation	Std. Error	P value	Significance
Control Group	4773.03	2304.38	420.71	0.001	Significant
Aloe Vera	722.02	984.69	179.77		
	Mean	Std. Deviation	Std. Error		
Control Group	4773.03	2304.38	420.71	0.001	Significant
Chlorhexidine	516.02	1047.90	191.32		
	Mean	Std. Deviation	Std. Error		
Aloe Vera	722.02	984.69	179.77	0.612	Not Significant
Chlorhexidine	516.02	1047.90	191.32		

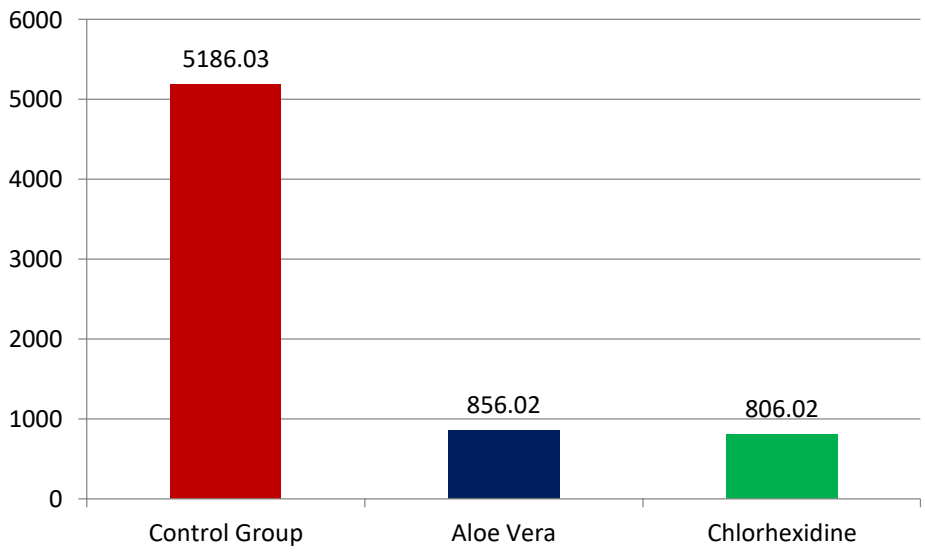


The mean CFU in the control group at the 48 hrs time interval was 4773.03±2304.38. In the aloevera group the mean CFU were 722.02±984.69 and in the Chlorhexidine group the mean CFU were 516.02±1047.90. The mean CFU were highest in the Control Group followed by Aloe vera and least in the Chlorhexidine group. The intergroup difference in CFU between the Control group and Aloe vera Group was statistically significant with p value of 0.001 signifying a highly significant difference between the groups, The intergroup difference in CFU between the Control group and Chlorhexidine Group was statistically significant with p value of

0.001 signifying a highly significant difference between the groups.The intergroup difference in CFU between the Aloe Vera group and Chlorhexidine Group was statistically non-significant with p value of 0.612

CFU AT 72 HRS IN THE THREE GROUPS

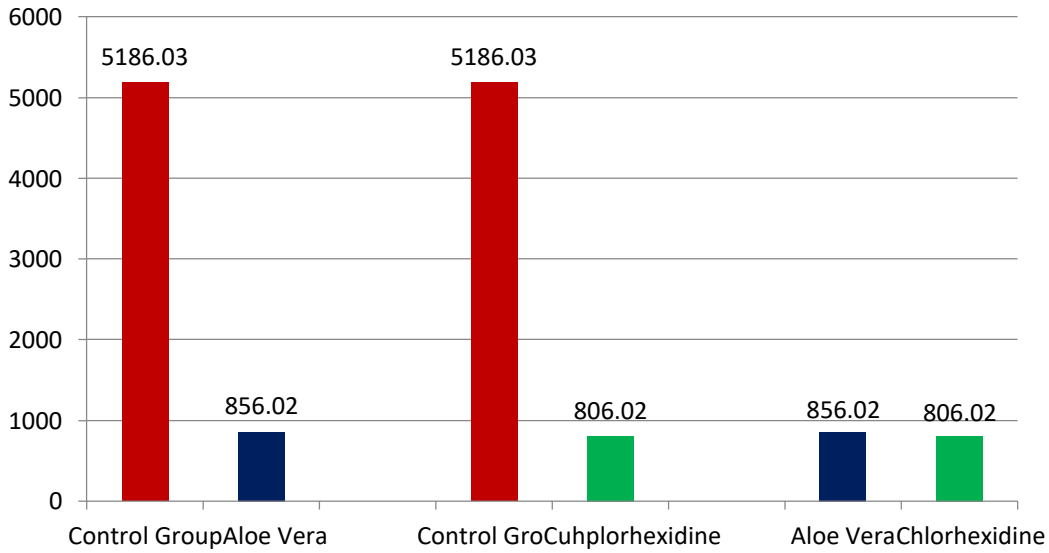
	Mean	Std. Deviation	Std. Error	Minimum	Maximum	
Control Group	5186.03	2531.99	462.27	5186.03	2531.99	
Aloe Vera	856.02	1092.11	199.39	856.02	1092.11	
Chlorhexidine	806.02	1286.02	234.79	806.02	1286.02	



The mean CFU in the control group at the 72 hrs time interval was 5186.03±2531.99. In the aloevera group the mean CFU were 856.02±1092.11 and in the Chlorhexidine group the mean CFU were 806.02±1286.02. The mean CFU were highest in the Control Group followed by Aloe vera and least in the Chlorhexidine group. The statistical difference in CFU between the three groups was analyzed using the One Way ANOVA followed by post hoc analysis at 5% significance level (p=0.05

Intergroup comparison of CFU between three groups at 72 hrs

	Mean	Std. Deviation	Std. Error	P value	Significance
Control Group	5186.03	2531.99	462.27	0.001	Significant
Aloe Vera	856.02	1092.11	199.39		
	Mean	Std. Deviation	Std. Error		
Control Group	5186.03	2531.99	462.27	0.001	Significant
Chlorhexidine	806.02	1286.02	234.79		
	Mean	Std. Deviation	Std. Error		
Aloe Vera	856.02	1092.11	199.39	0.930	Not Significant
Chlorhexidine	806.02	1286.02	234.79		

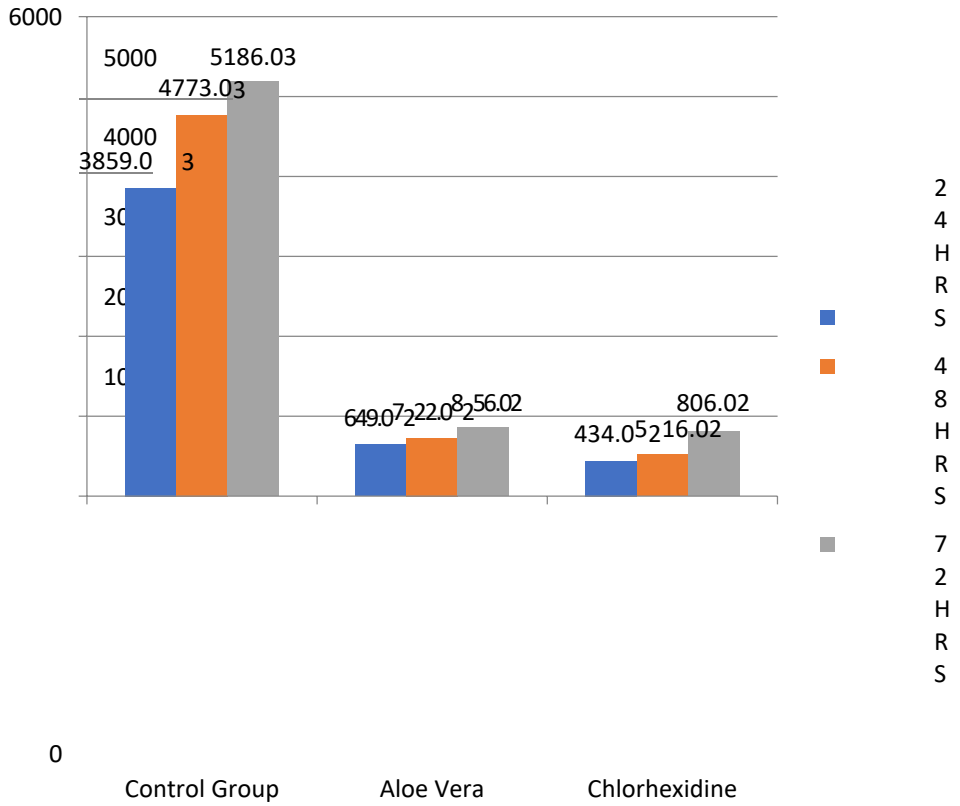


The mean CFU in the control group at the 72 hrs time interval was 5186.03±2531.99. In the aloevera group the mean CFU were 856.02±1092.11 and in the Chlorhexidine group the mean CFU were 806.02±1286.02. The mean CFU were highest in the Control Group followed by Aloe vera and least in the Chlorhexidine group. The intergroup difference in CFU between the Control group and Aloe vera Group was statistically significant with p value of 0.001 signifying a highly significant difference between the groups, The intergroup difference in CFU between the Control group and Chlorhexidine Group was statistically significant with p value of

0.001 signifying a highly significant difference between the groups. The intergroup difference in CFU between the Aloe Vera group and Chlorhexidine Group was statistically non-significant with p value of 0.930

INTRAGROUP COMPARIOSN OF CFY BETWEEN DIFFERENT TIME INTERVALS IN THREE GROUPS

	Mean	Std. Deviation	Std. Error	24-48 hrs	24-72 Hrs	48-72 hrs
Control Group	3859.03±1798.08	4773.03±2304.38	5186.03±2531.99	0.012 (Sig)	0.024 (Sig)	0.476 (non-Sig)
Aloe Vera	649.02±873.46	722.02±984.69	856.02±1092.11	0.775(non-Sig)	0.619 (non-Sig)	0.401 (non-Sig)
Chlorhexidine	434.02±846.54	516.02±1047.90	806.02±1286.02	0.768 (non-Sig)	0.184 (non-Sig)	0.289 (non-Sig)



In the control group the mean CFU at the 24 hrs time interval was 3859.03±1798.08 which increased to 4773.03±2304.38 at 48 hours and to 5186.03±2531.99 at 72 hours. The intragroup increase in CFU was statistically significant from 24 hrs to 48 hrs and from 24 hrs to 72 hrs

In the aloevera group the mean CFU were 649.02±873.46 at the 24 hrs which increased to 722.02±984.69 at 48 hrs and further increased to 856.02±1092 at 72 hours The intragroup increase in CFU was statistically non-significant from 24 hrs to 48 hrs, 24 hrs to 72 hrs and from 48 to 72 hours

In the Chlorhexidine group the mean CFU were 434.02±846.54 at the 24 hrs which increased to 516.02±1047.90 at 48 hrs and further increased to 806.02±1286.02 at 72 hours The intragroup increase in CFU was statistically non-significant from 24 hrs to 48 hrs, 24 hrs to 72 hrs and from 48 to 72 hours

Use of disinfectants is mandatory in dentistry. With time and awareness rising among people about the side effects of chemicals used in daily life as well as in dental procedures the inclination is shifting towards the use of more natural and herbal treatments available. Several studies done has shown reduced side effects as well as comparable results of herbal/natural product use in treatments to that of chemicals. Impression making is one of the primary steps in dental procedures. Among the various materials used for impression making in dentistry, irreversible hydrocolloids is among the most common. Alginate is an irreversible hydrocolloid preferred by dentists. Therefore, it is important to understand the handling the material as well maintainence of hygiene, minimizing if not eliminating the risk of cross infection from the very step.[45] Alginate is popular as the cost is low, better tolerability by patient, handling is easy, short execution time, instrumentation, very simple execution technique, and the possibility of detecting a detailed impression (even in the presence of undercuts) all in one step. Due to their low cost, they are mostly used as materials for studies related to medical and diagnostic purpose. Varying setting time alginate are available for the comfort of patient.[46] Though the material has many perks over other impression material. Maintainence of hygiene with material is a concern.[47] Jean Karl Demajo et al did a study to assess the antimicrobial activity of chemical disinfectants on alginate and silicone impression materials and concluded that alginate harbors three times more microorganisms than silicone impression material. [48]

Various disinfectants are being currently used in dentistry for disinfecting impressions. The broad category concerned here is divided into two- herbal and chemical. Some of the regular chemical disinfection solution used in dentistry includes Iodophor, Glutaraldehyde, Sodium hypochlorite, 0.25% Benzalkonium chloride (BC), Alcohols, Isopropyl alcohol, Ethyl alcohol, Chlorhexidine, Ozone water.[50] In this study the efficacy of herbal disinfectant (aloe vera) has been tested and compared with Chlorohexidine.[49]

Due to indiscriminate use of antimicrobials the resistance is also rising leading to inefficacy of treatments. Due to this rise in bacterial resistance to various synthetic antimicrobial agents, phytotherapeutics is popularizing. Phytotherapy is the medicinal use of plant extracts. Vrious natural products such as garlic extract, cinnamon oil, thyme oil, tea tree oil, Aloe vera etc., have shown anti-microbial

properties and hence been preferred.[50] Other studied done have evaluated the effect of Aloe vera and chlorhexidine as disinfectants on the success of selective caries removal technique. The use of Aloe vera extract as a cavity disinfectant has shown positive results[51]. Other studies have shown the use of herbal treatment (neem, garlic, and green tea) resulting in efficacy as chlorhexidine as potent disinfectants for toothbrushes.[52] Herbal solutions (Aloevera, Pancha Tulsi Juice and Amla juice) have been found to be effective in the disinfection of GP points as well.[53] The positive results of herbal treatments in dentistry is growing a keen interest for substituting chemical with herbal treatments.

Though CHX is one of the most popular disinfectants used in dentistry there is a raised concern regarding the drawback it holds such as high price, chemical used, burning sensation and genotoxicity. This brings in the keen interest in herbal substitute for the same. With advancement and growing popularity of herbal medicine, the aloe vera has been used in plaque control and oral health maintainence. Aloe vera or aloe barbadensis is a succulent cactus- like plant that belongs to Liliaceae family. It is already being used in cosmetic and medical industry for its various benefits. The pharmacological benefits are wound healing effects, immunomodulating activity, anti-inflammatory, antioxidant and antimicrobial properties. In dentistry, the use of aloe vera is done for treatment of several dental and oral conditions including oral lichen planus, oral submucous fibrosis, aphthous stomatitis, periodontitis and gingivitis. While the results of many trials recommend the use of aloe vera mouthwash as an effective substitute to chlorhexidine mouthwash.[54] There are various dental uses of aloe vera. The increased interest among researchers to analyze the use of aloe vera in dentistry and various studies have proved the effectiveness of this plant.[55] Studies done has showed that agar plates having sectioned alginate impression sprayed with disinfectant herbal solution showed less bacterial growth compared with the agar plates having alginate washed with water. Thus, concluding that herbal disinfectant solution is effective in controlling the bacterial growth on impressions with irreversible hydrocolloid.[38]

Besides these studies done have showed significant reduction of microbiota on alginate impressions treated with CHX. Though there was minimum effect on Candida albicans when treated with the same.[6] This raise the concern when the procedures are related to patient‟s having diabetes (prone to fungal infections).

Diabetes is one of the largest global health emergencies of this century. It ranks among the 10 leading causes of mortality together with cardiovascular disease (CVD), respiratory disease, and cancer. Type 2 diabetes susceptibility varies to a great extent around the globe, with Pacific Islanders, Asian Indians, and Native Americans having a significantly higher risk of developing the disorder. The number of people with type 2 diabetes rose globally in the 1990s, and since 2000, there is a dramatic increase in the number of people with diabetes.[56] Type II is an outcome combination of impairment of insulin resistance and defective secretion of insulin by beta cells. Factors contributing are genetics, obesity, physical inactivity and advancing age. Diabetic patients are more prone to develop caries, periodontitis, xerostomia, oral ulcers, burning mouth syndrome, candidiasis, loss of resilience of oral mucosa, residual bone resorption, periodontal abscess, gingival overgrowth and poor tolerance to prosthesis especially for complete dentures.[57] Oral manifestations are most likely due to increase glucose concentration in saliva, polyuria, impaired host resistance due to defective function of polymorphonuclear leucocyte (PMN) and microvasculsar changes.[58] Diabetic plaques have shown to contain more versatile microbes predominated by gram-positive streptococci and staphylococci.[59] Studies done have found a decrease in the biological and phylogenetic oral microbiome diversity in diabetics in comparison to non-diabetics from South Arabia, evidence that was related to an increase in the pathogenic content in the diabetic‟s oral microbiome.[60]

Considering the rising shift of disinfectants from chemical to herbal and the keen interest in the patients suffering from one of the most common disorders(diabetes) these days the following study has been done. The evidence remains suggests that aloe vera mouthwash is comparable to chlorhexidine in reducing gingival inflammation but inferior to chlorhexidine in reducing plaque.[61] Since aloe vera has shown so many benefits in dentistry, it‟s use as a disinfectant for impression has been tested considering the benefits, it‟s ease of availability and reduced or no side effects.

Chlorhexidine is also considered the gold standard for comparing against the antiplaque and anti-gingivitis agents. CHX is available in concentrations of 0.2% and 0.12% for mouthwash rinses.[62] In present study three blocks of 0.5x0.5 inch each has been cut from palatal region. Palatal region has been considered for obtaining the blocks to maintain the standardization of the samples. Specimens from ridge and

teeth bearing area in impression has been excluded to maintain the uniformity of the all three blocks.

The method used to treat the specimen with disinfectant is- immersion method. The blocks obtained were immersed in AGV and CHX and left for 5 minutes. Hamid Badrian et al in 2012 did a study to investigate the effect of three different types of disinfectant agents on alginate impression material after 5 and 10 minutes. It was concluded that the antimicrobial effect of spraying and immersing methods was almost equal while mere water rinsing showed no significant disinfection effect.[16]

As shown in Graph 1 the CFU count obtained at 24hrs in the three groups , control group had highest number of microbial colony. Aloe vera treated specimen showed microbial count far less than control group but more than CHX group. Thus, stating that aloevera and CHX both show effective results as disinfectants. The disinfection property of both showed significant results. Though the difference in the effectiveness of CHX and AGV were not found significant. R Kriplani et al did a comparative evaluation of antimicrobial efficacy of various root canal filling materials along with aloevera used in primary teeth and concluded that Aloevera + Sterile Water was found to have superior antimicrobial activity against most of the microorganisms followed by ZOE + Aloevera, calcium hydroxide + Aloevera, ZOE, calcium hydroxide, Metapex in the descending order and Vaseline showed no inhibition.[63] Tereza A. Delle Vedove Semenoff et al did a study on antimicrobial activity of 2% chlorhexidine gluconate, 1% sodium hypochlorite and paramonochlorophenol combined with furacin against S. aureus, C. albicans, E. faecalise and P. aureginosa and concluded that the antimicrobial activity of 2% chlorhexidine gluconate was greater than the other substances examined.[64]

In Graph 2 the CFU count obtained at 48 hours in the three group, control was found to hold the highest microbial count followed by AVG and CHX treated specimens. The effectiveness of AVG was significant as disinfectant. CHX showed better effectiveness but was comparable to that of AVG. Deepa G Kamath et al did a comparison of antiplaque and anti-gingivitis effects of aloe vera mouthwash with chlorhexidine in fixed orthodontic patients and concluded that although chlorhexidine is still the gold standard mouthwash, aloe vera exhibits promising results in reducing plaque and gingivitis scores, without any reported adverse

effects.[65] N T Sena et al did in vitro study on antimicrobial activity of sodium hypochlorite and chlorhexidine against selected single-species biofilms and concluded that the mechanical agitation improved the antimicrobial properties of the chemical substances tested using a biofilm model, favouring the agents in liquid presentation, especially 5.25% NaOCl and 2% chlorhexidine.[66]

In Graph 3 the CFU obtained at 72hrs in three groups showed close value results between AVG and CHX treated specimens. Both the specimens worked on rendering the microbiota ineffective suggesting that efficacy of both is worthy. The values obtained suggests that AVG and CHX showed no significant difference. Swathi Vangipuram et al did a comparative efficacy of aloe vera mouthwash and chlorhexidine on periodontal health and concluded that AloeVera shows equal effectiveness as Chlorhexidine.[67]

S C Supranoto et al studied the effect of chlorhexidine dentifrice or gel versus chlorhexidine mouthwash on plaque, gingivitis, bleeding and tooth discoloration and concluded that when daily oral hygiene cannot be performed, CHX MW is the first product of choice to inhibit plaque growth.[68]

Chlorhexidine is a broad-spectrum biocide effective against Gram-positive bacteria, Gram-negative bacteria and fungi. Chlorhexidine inactivates microorganisms with a broader spectrum than other antimicrobials (e.g., antibiotics) and has a quicker kill rate than other antimicrobials (e.g., povidone-iodine).1 It has both bacteriostatic (inhibits bacterial growth) and bactericidal (kills bacteria) mechanisms of action, depending on its concentration. Chlorhexidine kills by disrupting the cell membrane. Upon application in vitro, chlorhexidine can kill nearly 100% of Gram-positive and Gram-negative bacteria within 30 seconds.[69]

Aloe vera and its active compounds showed promising role as a cytotoxic, antitumoral, anticancer, and antidiabetic agent. In the last 6 years, most pharmacological studies have been in vitro and in vivo works.[70] Aloe vera contains 6 antiseptic agents: Lupeol, salicylic acid, urea nitrogen, cinnamonic acid, phenols and sulfur. They all have inhibitory action on fungi, bacteria and viruses.

Aloe vera has been found to be comparable to CHX in earlier studies as disinfectant in normal healthy subjects. The difference in oral microbiota of uncontrolled diabetic subjects compared to non -diabetic, has prompted us to study the efficacy of herbal

disinfectant on the same. The present study clearly demonstrates that efficacy of herbal (aloevera) disinfectant is comparable to chemical disinfectant (CHX).

The future study prompts to identify the various microbiota specific for usually found diabetic subjects and efficacy of herbal disinfectant on them.

The present study was conducted in Department of Prosthodontics, crown & bridge, Babu Banarasi Das College of Dental Sciences, Lucknow. The study conducted presented the following results.

1-The efficacy of aloevera to reduce the microbiota count on alginate impression made from diabetic dentulous patients at 24hrs was found to be statistically significant.

2- The reduction in CFU count upon aloevera treatment on alginate impression made from diabetic dentulous patients at 48hrs was found to be statistically significant.

3- The result of aloevera being effective on alginate impression made from diabetic dentulous patients at 72hrs was found to be statistically significant.

4- The efficacy of chlorohexidine to reduce the microbiota count on alginate impression made from diabetic dentulous patients at 24hrs was found to be statistically significant.

5- The reduction in CFU count upon chlorohexidine treatment on alginate impression made from diabetic dentulous patients at 48hrs was found to be statistically significant.

6- The result of chlorohexidine being effective on alginate impression made from diabetic dentulous patients at 72hrs was found to be statistically significant.

7-The microbiological efficacy of herbal disinfectant (aloevera) is comparable to chemical disinfectant (chlorohexidine as a gold standard) on alginate impressions of maxilla made from uncontrolled diabetic dentulous patients.

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ANNEXURE-

Babu Banarasi Das University
Babu Banarasi Das College of Dental Sciences,
BBD City, Faizabad Road, Lucknow – 226028 (INDIA)

Dr. Lakshmi Bala
Professor and Head Biochemistry and
Member-Secretary, Institutional Ethics Committee

Communication of the Decision of the IXth Institutional Ethics Sub-Committee

IEC Code: 24 **BBDCODS/04/2022**

Title of the Project: Compare the effectiveness of chemical and herbal disinfectants on alginate impression made from diabetic dentulous patients.

Principal Investigator: Dr Roopali Sharma **Department:** Prosthodontics and Crown & Bridge

Name and Address of the Institution: BBD College of Dental Sciences Lucknow.

Type of Submission: New, MDS Project Protocol

Dear Dr Roopali Sharma,

The Institutional Ethics Sub-Committee meeting comprising following four members was held on 07th April, 2022.

1.	Dr. Lakshmi Bala Member Secretary	Prof. and Head, Department of Biochemistry, BBDCODS, Lucknow
2.	Dr. Amrit Tandan Member	Prof. & Head, Department of Prosthodontics and Crown & Bridge, BBDCODS, Lucknow
3.	Dr. Rana Pratap Maurya Member	Reader, Department of Orthodontics, BBDCODS, Lucknow
4.	Dr. Akanksha Bhatt Member	Reader, Department of Conservative Dentistry & Endodontics, BBDCODS, Lucknow

The committee reviewed and discussed your submitted documents of the current MDS Project Protocol in the meeting.
The comments were communicated to PI thereafter it was revised.

Decisions: The committee approved the above protocol from ethics point of view.

Forwarded by:

(Dr. Lakshmi Bala)
Member-Secretary
IEC

Member-Secretary
Institutional Ethic Committee
BBD College of Dental Sciences
BBD University
Faizabad Road, Lucknow-226028

(Dr. Puneet Ahuja)
Principal
BBDCODS

PRINCIPAL
Babu Banarasi Das College of Dental Sciences
(Babu Banarasi Das University)
BBD City, Faizabad Road, Lucknow-226028

ANNEXURE-56

BABU BANARASI DAS COLLEGE OF DENTAL SCIENCES (FACULTY OF BBD UNIVERSITY), LUCKNOW

INSTITUTIONAL RESEARCH COMMITTEE APPROVAL

The project titled **"Compare the effectiveness of chemical and herbal disinfectants on alginate impression made from diabetic dentulous patients"** submitted by **Dr Roopali Sharma** Post graduate student from the **Department of Prosthodontics and Crown & Bridge** as part of MDS Curriculum for the academic year 2020-2023 with the accompanying proforma was reviewed by the Institutional Research Committee present on **11th October 2021** at BBDCODS.

The Committee has granted approval on the scientific content of the project. The proposal may now be reviewed by the Institutional Ethics Committee for granting ethical approval.

Prof. Vandana A Pant
Co-Chairperson

Prof. B. Rajkumar
Chairperson

ANNEXURE-III

Babu Banarasi Das College of Dental Sciences

(Babu Banarasi Das University)

BBD City, Faizabad Road, Lucknow – 227105 (INDIA)

Consent Form (English)

Title of the Study ……..

Study Number……..

Subject‟s Full Name…….

Date of Birth/Age ……

Address of the Subject…….

Phone no. and e-mail address……

Qualification ……

Occupation: Student / Self Employed / Service / Housewife/Other (Please tick as appropriate)

Annual income of the Subject……

Name and of the nominees(s) and his relation to the subject (For the purpose of compensation in case of trial related death).

1. I confirm that I have read and understood the Participant Information Document dated ……..for the above study and have had the opportunity to ask questions. **OR** I have beenexplained the nature of the study by the Investigator and had the opportunity to ask questions.
2. I understand that my participation in the study is voluntary and given with free will without any duress and that I am free to withdraw at any time, without giving any reason and without my medical care or legal rights being affected.
3. I understand that the sponsor of the project, others working on the Sponsor„s behalf, the Ethics Committee and the regulatory authorities will not need my permission to look at my health records both in respect of the current study and

any further research that may be conducted in relation to it, even if I withdraw from the trial. However, I understand that my Identity will not be revealed in any information released to third parties or published.

4. I agree not to restrict the use of any data or results that arise from this study provided sucha use is only for scientific purpose(s).
5. I permit the use of stored sample (tooth/tissue/blood) for future research. **Yes[ ]No[ ]**

Not Applicable [ ]

6. I agree to participate in the above study. I have been explained about the complications andside effects, if any, and have fully understood them. I have also read and understood the participant/volunteer"s Information document given to me.

Signature (or Thumb impression) of the Subject/Legally AcceptableRepresentative:……..

Signatory„s Name……. Date …….

Signature of the Investigator…… Date……..

Study Investigator„s Name..... Date……..

Signature of the witness…… Date……..

Name of the witness……

Received a signed copy of the PID and duly filled consent form Signature/thumb impression of the subject or legally Acceptable representative Date……..

Babu Banarasi Das College of Dental Sciences
(Babu Banarasi Das University)
BBD City, Faizabad Road, Lucknow – 227105 (INDIA)

सहमति पत्र

अध्ययन शीर्षक.....
अध्ययन संख्या.....
प्रतिभागी के पूर्ण नाम.....
जन्म तिथि / आयु.....
प्रतिभागी का पता .....
फोन नं. और ई-मेल पता .....
योग्यता .....
व्यवसाय: छात्र / स्व कार्यरत / सेवा / ग्रहिणी .....
अन्य (उचित रुप मे टिक करें) .....
प्रतिभागी की वार्षिक आय .....
प्रत्याशीयो के नाम और प्रतिभागी से संबंध...(परीक्षण से संबंधित मौत के मामले मे मुआवजे के प्रयोजन के लिए)

1. मेरी पुष्टि है कि मैने अध्ययन हेतु सुचना पत्र दिनांक ..... को पढ व समझ लिया तथा मुझे प्रश्न पुछने या मुझे अध्ययन अन्वेषक ने सभी तथ्यों को समझा दिया है तथा मुझे प्रश्न पुछने के समान अवसर प्रदान किए गये।

2. मैंने यहॉ समझ लिया कि अध्ययन में मेरी भागीदारी पूर्णतः स्वैच्छिक है और किसी भी दबाव के बिना स्वतंत्र इच्छा के साथ दिया है किसी भी समय किसी भी कारण के बिना , मेरे इलाज या कानूनी अधिकारो को प्रभावित किए बिना , अध्ययन में भाग न लेने के लिए स्वतंत्र हुँ ।

3. मैंने यह समझ लिया है कि अध्ययन के प्रायोजक , प्रायोजक की तरफ से काम करने वाले लोग, आचार समिति और नियामक अधिकारियों को मेरे स्वास्थ्य रिकार्ड को वर्तमान अध्ययन या आगे के अध्ययन के सन्दर्भ देखने के लिए मेरी अनुमति की जरूरत नही है, चाहे मैने इस अध्ययन से नाम वापस ले लिया है। हॉलाकि मै यह समझता हुँ कि मेरी पहचान को किसी भी तीसरे पक्ष या प्रकाशित माध्यम में नही दी जायेगी।

4. मै इससे सहमत हूँ कि कोई भी डेटा या परिणाम जो इस अध्ययन से प्राप्त होता है उसका वैज्ञानिक उद्देश्य (ओं) के उपयोग के लिए मेरी तरफ से कोई प्रतिबंध नही है।
5. भविष्य के अनुसंधान के लिए भंडारित नमूना (ऊतक/रक्त) पर अध्ययन के लिए अपनी सहमति देता हुँ।

हॉ [ ] नही [ ] अनउपयुक्त [ ]

6. मै परीक्षण की अनुमति देता हुँ। मुझे इसके द्वारा यदि कोई परेशानी होती है, इसके बारे में जानकारी दे दी गई है। मैने रोगी जानकारी सूचना पत्र को पढ तथा समझ लिया है।

प्रतिभागी / कानूनी तौर पर स्वीकार्य प्रतिनिधि का हस्ताक्षर (या अंगूठे का निशान.....

हस्ताक्षरकर्ता का नाम..... दिनांक .....अन्वेषक के हस्ताक्षर ..... दिनांक .....

अध्ययन अन्वेषक का नाम .....

गवाह के हस्ताक्षर ..... दिनांक .....गवाह के नाम .....

मैनें पीआईडी और विधिवत भरे सहमति फार्म का एक हस्ताक्षर की नकल प्राप्त की.

प्रतिभागी कानूनी तौर पर प्रतिनिधि का हस्ताक्षर/ अंगूठे का निशान ..... दिनांक.....

ANNEXURE-IV

Babu Banarasi Das College of Dental Sciences

(Babu Banarasi Das University)

BBD City, Faizabad Road, Lucknow – 227105 (INDIA)

Guidelines for Devising a Participant / Legally Acceptable Representative Information Document (PID) in English

Guideline for preparation of the participant information document	
While submitting your project report to the Institutional Ethics Committee, ensure that you have included participant information document and an informed consent form that is prepared as per the guidelines for Good Clinical Practice-Centre for Drug Candidate Optimization (GCP-CDCO2001), International Conference on Hormonization-Good Clinical Practice (ICH – GCP), ICMR ethical guidelines 2006, and the Declaration of Helsinki. The document is important because it enables the participants to make an informed choice. It also has got to be unique because no two research projects are identical. **The participant information document (PID should include only those headings listed below which are relevant to that study. Any further information you wish to add, is your choice.**	
1.	Participant information document and an consent form **in English and Hindi** (other languages if required)
2.	Font: Arial spacing of lines with 1.5
3.	Size: 12
4	All the consent forms must have Version No, Date, Page no **in the footer**
5.	In the case of participants with age≥ 18 yrs, PID and consent form should be attached while in the case of participant‟s age≤ 18 yrs and ≥ 8 yrs the above along with information document and assent form for children (minor) should be attached. In the case of ≤ 8 it will be signed by the guardian.

Potential recruits to your research/trial study must be given sufficient information to allow them to decide whether or not they want to take part. The Information Document should contain information under the headings given below, and preferably in the order specified. It should be

written in simple, non-technical terms and be easily understood by a lay person. Use short words, sentences and paragraphs.

1.Study Title

Compare the effectiveness of chemical and herbal disinfectants on alginate impression made from diabetic dentulous patients.

2. Invitation Paragraph

You are being invited to take part in a research/trial study. Before you decide it is important for you to understand why the research/study is being done and what it will involve. Please take time to read the following information carefully and discuss it with friends, relatives and your treating physician/family doctor if you wish. Ask us if there is anything that is not clear or if you would like more information. Take time to decide whether or not you wish to take part.

3. What is the purpose of the study?

The aim of this study is to bring herbal disinfectants in regular dental practice instead of chemical

disinfectants. Chemical disinfectants used to clean impressions can have side effects. Our aim is

to study efficacy of herbal disinfectants on oral microorganisms as it has less/no side effects.

4. Why have I been chosen?

You are chosen as you fulfil the criteria of stud`y. Example- age (45 to 60yrs), Type II diabetic and having teeth.

5. Do I have to take part?

It is up to you to decide whether or not to take part. If you do decide to take part you will be given this information sheet to keep and be asked to sign a consent form. If you decide to take part you are still are free to withdraw at any time and without giving a reason.

6. What will happen to me if I take part?

You will have to come for only one sitting which will take approximately 10mins. I will take impression of your upper jaw with biocompatible material. As a volunteer your responsibility will be to arrive on time (approximately will be given according to your convenience) and eat something light before visit.

7. What do I have to do?

There is nothing major you need to do. There will be no lifestyle restrictions. No dietary changes needed. You can take your regular medications, drink and eat as you wish.

8. What is the procedure that is being tested?

Procedure will include you being seated on dental chair. Alginate is the material that will be loaded on a sterilized metal/plastic tray and will be placed in your mouth for taking impression. Once the material will set, dentist will remove the tray and you will be free to go.

9. What are the interventions for the study?

Study includes no interventions.

10. What are the side effects of taking part?

Volunteer might be allergic to the material or can have gagging (feeling like vomiting). If you suffer these or any other symptoms you will report immediately. You will have the dentist‟s name and contact number to phone if you become prone to any symptom.

11. What are the possible disadvantages and risks of taking part?

Study includes minimal risk. There is no disadvantage involved in the study.

12. What are the possible benefits of taking part?

There are no intended clinical benefit to the patient.

13. What if new information becomes available?

Sometimes during the course of a research project, new information becomes available about the research being studied. If this happens, your researcher will tell you about it and discuss with you whether you want to continue in the study. If you decide to withdraw, your researcher/investigator will make arrangements for your withdrawal. If you decide to continue in the study, you may be asked to sign an updated consent form.

14. What happens when the research study stops?

If the study finishes/stops before the stipulated time, the reason for the same will be be explained to you.

15. What if something goes wrong?

Volunteer will be taken care of by the doctor expertizing in the field in BBDCODS OPD.

16. Will my taking part in this study be kept confidential?

Your name, address or any personal or other information will not be shared outside BBDCODS.

17. What will happen to the results of the research study? Identity of the participants will not be disclosed in any result/report/publication.

18. Who is organizing the research?

Study is organized by the researcher. Complete cost of the research study is bore by the researcher only.

19. Will the results of the study be made available after study is over?

If the patient wishes the result of the study will be made available to him/her.

20. Who has reviewed the study?

HOD /IRC/IEC of the institution has reviewed and approved the study.

21. Contact for further information

Name of the PI- Dr.Roopali Sharma

Address- Department of Prosthodontics & Crown & Bridge, BBDuniversity, Lucknow

e-mail address- roopalisharma1522@gmail.com

Telephone Numbers- 8354846852

Member Secretary of Ethics Committee of the institution- (Dr.Lakshmi Bala Member Secretary)

Address- Babu Banarasi University, Faizabad Road, Lucknow

e-mail address- bbdcods.iec@gmail.com

telephone numbers- (ext. no. 1291).

Thankyou for taking part in the study!

The participant will be given a copy of the information sheet and the signed consent form.

Signature of PI……

Name……..

Date……..

Babu Banarasi Das College of Dental Sciences
(Babu Banarasi Das University, Lucknow)
BBD City, Faizabad Road, Lucknow – 227105 (INDIA)

प्रतिभागी के लिए सूचना पत्र

1. अध्ययन शीर्षक

मधुमेह के दंत रोगियों से बने एल्गिनेट प्रभाव पर रासायनिक और हर्बल कीटाणुनाशकों की प्रभावशीलता की तुलना करने हेतु।

2. निमंत्रण अनुच्छेद

आपको एक शोध/परीक्षण अध्ययन में भाग लेने के लिए आमंत्रित किया जा रहा है। निर्णय लेने से पहले आपके लिए यह समझना महत्वपूर्ण है कि शोध/अध्ययन क्यों किया जा रहा है और इसमें क्या शामिल होगा। कृपया निम्नलिखित जानकारी को ध्यान से पढ़ने के लिए समय निकालें और यदि आप चाहें तो दोस्तों, रिश्तेदारों और अपने इलाज करने वाले चिकित्सक/पारिवारिक चिकित्सक के साथ इस पर चर्चा करें, हमसे पूछें कि क्या कुछ स्पष्ट नहीं है या यदि आप अधिक जानकारी चाहते हैं। यह तय करने के लिए समय निकालें कि आपको भाग लेना है या नहीं।

3. अध्ययन का उद्देश्य क्या है ?

इस अध्ययन का उद्देश्य रासायनिक के बजाय हर्बल कीटाणुनाशकों को नियमित दंत चिकित्सा अभ्यास में लाना है। कीटाणुनाशक छापों को साफ करने के लिए इस्तेमाल किए जाने वाले रासायनिक कीटाणुनाशक के दुष्प्रभाव हो सकते हैं। इस अध्ययन का उद्देश्य है मौखिक सूक्ष्मजीवों पर हर्बल कीटाणुनाशकों की प्रभावकारिता का अध्ययन करने के लिए क्यों कि इसका कम/कोई पक्ष नहीं है।

4. मुझे इस अध्ययन के लिए क्यों चुना गया है ?

आपको चुना जाता है क्योंकि आप अध्ययन के मानदंडों को पूरा करते हैं।

उदाहरण आयु (45 से 60 वर्ष), टाइप–2 मधुमेह और दांत होना।

5. क्या इसमें मुझे भाग लेना चाहिए ?

यह आपको तय करना है कि भाग लेना है या नहीं। यदि आप भाग लेने का निर्णय लेते हैं तो आपको यह सूचना पत्र रखने के लिए दिया जाएगा और सहमति प्रपत्र पर हस्ताक्षर करने के लिए कहा जाएगा, यदि आप भाग लेने का निर्णय लेते हैं तो भी आप किसी भी समय और बिना कोई कारण बताए वापस लेने के लिए स्वतंत्र हैं।

6. मुझे क्या होगा यदि मैं इस अध्ययन में भाग लेता हूँ।

आपको केवल एक बैठक के लिए आना होगा जिसमें लगभग 10 मिनट का समय लगेगा। मैं बायोकंपैटिबल सामग्री के साथ आपके ऊपरी जबड़े की छाप लूंगा। एक स्वयं सेवक के रूप में आपकी जिम्मेदारी होगी कि आप समय पर पहुंचें (लगभग आपकी सुविधा के अनुसार दिया जाएगा) और यात्रा से पहले कुछ हल्का खा लें।

7. मुझे क्या करना है ?

आपको कुछ भी बड़ा करने की जरूरत नहीं है। जीवन शैली पर कोई प्रतिबंध नहीं होगा। कोई आहार परिवर्तन की जरूरत नहीं है। आप अपनी नियमित दवाएं ले सकते हैं, अपनी इच्छानुसार पी सकते हैं और खा सकते हैं।

8. किस प्रकिया का परीक्षण किया जा रहा है ?

प्रक्रिया में आपको डेंटल चेयर पर बैठाना जाना शामिल होगा। एल्गिनेट वह सामग्री है जिसे एक निष्फल धातु/प्लास्टिक ट्रे पर लोड किया जाएगा और छाप लेने के लिए आपके मुंह में रखा जाएगा। एक बार सामग्री सेट हो जाने के बाद, दंत चिकित्सक ट्रे को हटा देगा और आप जाने के लिए स्वतंत्र होंगे।

9. इस शोध में कौन से हस्तक्षेप दिए जायेगें ?

अध्ययन में कोई हस्तक्षेप शामिल नहीं है।

10. इस अध्ययन में भाग लेने के क्या दुष्प्रभाव है ?

स्वयंसेवक को सामग्री से एलर्जी हो सकती है या उसे गैगिंग (उल्टी जैसा महसूस होना) हो सकता है। यदि आप इन या किसी अन्य लक्षण से पीड़ित हैं तो आप तुरंत रिपोर्ट करेंगे। यदि आप किसी भी लक्षण से ग्रस्त हो जाते हैं, तो आपके पास दंत चिकित्सक का नाम और फोन पर संपर्क नंबर होगा।

14. क्या होता है जब अध्ययन / शोध परीक्षण बन्द हो जाता है ?
यदि अध्ययन निर्धारित समय से पहले समाप्त हो जाता है/रूक जाता है, तो उसका कारण आपको समझाया जाएगा।

15. क्या होगा अगर कुछ गलत हो जाता है ?
बी.बी.डी.सी. ओ.डी.एस. ओ.पी.डी. में क्षेत्र विशेषज्ञ चिकित्सक द्वारा स्वयंसेवी की देखभाल की जाएगी।

16. मेरे इस अध्ययन में भाग लेने को गोपनीय रखा जाएगा ?
आपका नाम, पता या कोई भी व्यक्तिगत या अन्य जानकारी बाहर साझा नहीं की जाएगी।

17. अध्ययन/शोध परीक्षण के परिणाम का क्या होगा ?
किसी भी परिणाम/रिपोर्ट/प्रकाशन में प्रतिभागियो की पहचान का खुलासा नहीं किया जाएगा।

18. इस अध्ययन को कौन आयोजित कर रहा है और इस परीक्षण के लिए धन कहॉ से आएगा ?
अध्ययन वहां शोधकर्ता द्वारा आयोजित किया जाता है। शोध अध्ययन की पूरी लागत केवल शोधकर्ता द्वारा बोर किया जाता है।

19. क्या सेवाएं शोध खत्म हो जाने के बाद उपलब्ध रहेगी या नही ?
यदि रोगी चाहे तो अध्ययन का परिणाम उसे उपलब्ध कराया जाएगा।
20. इस अध्ययन का पुर्ननिरिक्षण किसने किया है ?
संस्थान के विभागाध्यक्ष/आईआरसी/आईईसी ने अध्ययन की समीक्षा की और उसे अनुमोदित किया।

निम्न लोगो से सम्पर्क करें

21. अधिक जानकारी के लिए
वैज्ञानिक/अन्वेषक का नाम – डॉ0 रूपाली शर्मा
पता – डिपार्टमेंट ऑफ प्रोस्थोडान्टिक्स एंड क्रॉउन एंड ब्रिज,
बी0बी0डी0 युनिवर्सिटी, लखनऊ।
ई0 मेल – **roopalisharma1522@gmail.com**
मोबाइल नं0 – **8354846852**

संस्था की नैतिकता समिति के सदस्य सचिव – डा0 लक्ष्मी बाला, सदस्य सचिव
पता – बी0बी0डी0 युनिवर्सिटी, फैजाबाद रोड, लखनऊ।
ई0 मेल – **bbdcods.iec@gmail.com**
मोबाइल नं0 – (दूरभाष नं0 1233)

भाग लेने के लिए धन्यवाद।

प्रतिभागी को इस सूचना पत्र की एक प्रतिलिपि प्रदान की जाएगी।

प्रमुख अन्वेषक के हस्ताक्षर

प्रमुख अन्वेषक का नाम ..... दिनांक.....

MASTER CHART

S.No.	GROUP	COLONY COUNT CFU/ml		
		24Hrs	48Hrs	72Hrs
1.	Control	680	770	820
2.	Control	3020	3100	3220
3.	Control	6000	6500	7000
4.	Control	5000	6000	6500
5.	Control	5000	6000	6500
6.	Control	3000	7700	8200
7.	Control	3140	3560	3800
8.	Control	7000	8000	9000
9.	Control	3190	3300	3880
10.	Control	2560	2800	2940
11.	Control	683	775	821
12.	Control	3014	3108	3267
13.	Control	6012	6487	7038
14.	Control	4976	6001	6674
15.	Control	4929	6128	6598
16.	Control	2987	7756	8237
17.	Control	3103	3530	3956
18.	Control	6976	7984	9003
19.	Control	3185	3284	3954
20.	Control	2545	2760	2943
21.	Control	683	698	865
22.	Control	3010	3108	3245
23.	Control	5982	6546	7067
24.	Control	4901	6000	6512
25.	Control	4967	5959	6569
26.	Control	2958	7654	8245

27.	Control	3108	3542	3841
28.	Control	6967	7969	9056
29.	Control	3157	3274	3880
30.	Control	2551	2768	2949

MASTER CHART

S.No.	GROUP	COLONY COUNT CFU/ml		
		24Hrs	48Hrs	72Hrs
1.	Aloe Vera	10	20	20
2.	Aloe Vera	10	10	20
3.	Aloe Vera	790	820	1100
4.	Aloe Vera	790	880	1350
5.	Aloe Vera	2230	2540	2760
6.	Aloe Vera	20	20	20
7.	Aloe Vera	270	280	370
8.	Aloe Vera	40	40	40
9.	Aloe Vera	2300	2570	2840
10.	Aloe Vera	30	40	40
11.	Aloe Vera	10	19	23
12.	Aloe Vera	9	10	20
13.	Aloe Vera	87	810	1069
14.	Aloe Vera	795	854	1325
15.	Aloe Vera	2225	2521	2865
16.	Aloe Vera	19	22	24
17.	Aloe Vera	265	286	373
18.	Aloe Vera	38	43	46
19.	Aloe Vera	2256	2586	2856
20.	Aloe Vera	29	39	43
21.	Aloe Vera	9	19	23
22.	Aloe Vera	8	12	21
23.	Aloe Vera	788	823	1127
24.	Aloe Vera	768	885	1369
25.	Aloe Vera	2240	2532	2769
26.	Aloe Vera	18	22	27

27.	Aloe Vera	257	285	387
28.	Aloe Vera	35	42	43
29.	Aloe Vera	2280	2569	2865
30.	Aloe Vera	29	43	46

MASTER CHART

S.No.	GROUP	COLONY COUNT CFU/ml		
		24Hrs	48Hrs	72Hrs
1.	Chlorhexidine	0	0	0
2.	Chlorhexidine	0	0	0
3.	Chlorhexidine	90	100	110
4.	Chlorhexidine	10	20	1920
5.	Chlorhexidine	1490	1530	1920
6.	Chlorhexidine	10	10	10
7.	Chlorhexidine	170	190	210
8.	Chlorhexidine	0	0	0
9.	Chlorhexidine	2560	3300	3880
10.	Chlorhexidine	10	10	10
11.	Chlorhexidine	0	0	0
12.	Chlorhexidine	0	0	0
13.	Chlorhexidine	87	98	110
14.	Chlorhexidine	9	19	1964
15.	Chlorhexidine	1481	1531	1924
16.	Chlorhexidine	9	10	13
17.	Chlorhexidine	167	182	217
18.	Chlorhexidine	0	0	0
19.	Chlorhexidine	2555	3281	3889
20.	Chlorhexidine	9	11	12
21.	Chlorhexidine	0	0	0
22.	Chlorhexidine	0	0	0
23.	Chlorhexidine	87	98	114
24.	Chlorhexidine	9	18	1927
25.	Chlorhexidine	1459	1532	1934

26.	Chlorhexidine	9	13	14
27.	Chlorhexidine	178	193	215
28.	Chlorhexidine	0	0	0
29.	Chlorhexidine	2538	3218	3895
30.	Chlorhexidine	9	11	13

ANNEXURE-V

Formulas used for analysis

Mean

$$\bar{X} = \frac{\Sigma X}{N}$$

Where:

$\bar{X}$ = the data set mean

$\sum$ = the sum of

X = the scores in the distribution

N = the number of scores in the distribution

Range

$$range = X_{highest} - X_{lowest}$$

Where:

$X_{highest}$ = largest score

X_{lowest} = smallest score

Variance

$$SD^2 = \frac{\Sigma (X - \bar{X})^2}{N}$$

The simplified variance formula

$$SD^2 = \frac{\Sigma X^2 - (\Sigma X)^2}{N}$$

N

Where:

SD^2 = the variance

$\sum$ = the sum of

X = the obtained score

$\overline{X}$ = the mean score of the data

N = the number of scores

Standard Deviation (N)

$$SD = \sqrt{\frac{\Sigma(X-\overline{X})^2}{N}}$$

The simplified standard deviation formula

$$SD = \sqrt{\frac{\Sigma X^2 - \frac{(\Sigma X)^2}{N}}{N}}$$

Where:

SD = the standard deviation

$\sum$ = the sum of

X = the obtained score

$\overline{X}$ = the mean score of the data

N = the number of scores

The Pearson correlation

$$r = \frac{\Sigma z_X z_Y}{N}$$

Where:

r = correlation coefficient

$\sum$ = the sum of

z_X = Z score for variable X

z_Y = Z score for variable Y

$z_X z_Y$ = the cross product of Z scores

N = the number of scores

One Way ANOVA

The formula for the one-way **ANOVA** *F*-test statistic is

$$F = \frac{\text{between-group variability}}{\text{within-group variability}}.$$

The between-group variability" is

$$\sum_{i=1}^{K} n_i (\bar{Y}_{i\cdot} - \bar{Y})^2 / (K-1)$$

where Y_i denotes the sample mean in the i^{th} group, n_i is the number of observations in the i^{th} group, $\bar{Y}$ denotes the overall mean of the data, and K denotes the number of groups.

The "within-group variability" is

$$\sum_{i=1}^{K} \sum_{j=1}^{n_i} (Y_{ij} - \bar{Y}_{i\cdot})^2 / (N-K),$$

where Y_{ij} is the j^{th} observation in the i^{th} out of K groups and N is the overall sample size.

Paired t test

$$t = \frac{\bar{x} - 0}{SE(\bar{d})} = \frac{\bar{x}}{SD(x)/\sqrt{n}}$$

A paired t-test is used to compare two population means where you have two samples in which observations in one sample can be paired with observations in the other sample. Examples of where this might occur are: - Before-and-after observations on the same subjects (e.g., students‟ diagnostic test results before and after a particular module or course) or A comparison of two diff erent methods of measurement or two diff erent treatments where the measurements/treatments are applied to the same

Post Hoc Tukey Test

Tukey's range test, also known as the Tukey's test, Tukey method, Tukey's honest significance test, or Tukey's HSD (honestly significant difference) test,[1] is a single-step multiple comparison procedure and statistical test. It can be used on raw data or in conjunction with an ANOVA (post-hoc analysis) to find means that are significantly different from each other. Named after John Tukey, it compares all possible pairs of means, and is based on a studentized range distribution (q) (this distribution is similar to the distribution of t from the t-test. Tukey's test compares the means of every treatment to the means of every other treatment; that is, it applies simultaneously to the set of all pairwise comparisons $\mu_i - \mu_j$ and identifies any difference between two means that is greater than the expected standard error. Tukey's test is based on a formula very similar to that of the t-test. In fact, Tukey's test is essentially a t-test, except that it corrects for family-wise error rate.

The formula for Tukey's test is:

$$q_s = \frac{Y_A - Y_B}{SE},$$

where Y_A is the larger of the two means being compared, Y_B is the smaller of the two means being compared, and SE is the standard error of the sum of the means. This q_s value can then be compared to a q value from the studentized range distribution. If the q_s value is *larger* than the critical value obtained from the distribution, the two means are said to be significantly different at level.

ANNEXURE-VI

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INTRODUCTION

The goal of disinfection in the dental office is to prevent the spread of infection from one patient to another and maintain the safety of the dental care providers. Prevention of cross-infection has significant effect on infection control. Impression disinfectants are important consideration when it comes to preventing infection. Nowadays various disinfectants are available in the market but to narrow down our pick for the one with high potency and least side effects is still debatable. Use of chemical disinfectant is a commonly known practice in dentistry but use of the herbal for the same is evolving too. Aloe vera is being used as a disinfectant in medical field already. The availability of the same is just not easy but also offers higher biocompatibility and lesser side effects. Aloe Vera belongs to Liliaceae family. Aloe barbadensis miller and Aloe arborescens are available commercially. Free Anthroquinones and their derivatives like Barbaloin, 10- aloe-emodin-9-anthrone, Isobarbaloin and chromones in Aloe Vera leaves exert a strong purgative effect and are potent anti-microbial agents1. Aloe vera has a lot to offer in the field of dentistry. a lot of studies is on the way to utilize the effective anti-microbial property of the miracle plant. Interest is gathering for the use of aloe vera in dentistry and this natural therapy is already proved its unlimited use in our field.2 AVG has various pharmacological actions like being antibacterial, anti-fungal. Among the natural products, aloe vera had shown antifungal property on the heat-cured acrylic denture base. The availability, cost effectiveness, and colossal advantages make this herb one of the best alternatives to the present denture cleansing tablet agents that are used. It is, therefore, essential to explore natural plant-based medicines in developing countries where citizens are sometimes incapable to survey expensive devout medicine.3