

Effectiveness of different Decontamination Methods on Implant Surfaces – A Microbiological Study.

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Abstract-

Introduction: From ancient time, different alternatives have been developed for replacement of missing teeth, out of which dental implants are considered as the most stunning evolution. Being crucial in dentistry, although dental implants have foreseen long-term success, the complications and failures do occur. One of the most supreme reasons for its failure is the bacteria which can lead to peri-implant diseases like peri-implant mucositis and peri-implantitis. Thus, removal of microbial plaque biofilm becomes a prerequisite. Different surgical and nonsurgical treatment modalities have been suggested for the same. Our study compares between various decontamination methods like air-polishing with glycine, erythritol and irrigation with doxycycline and tetracycline.

Objective: To evaluate the efficacy of different decontamination methods on implant surface by microbiological analysis.

Methodology: Seven sterile, threaded, tapered, titanium coated implants with machined surface were used, out of which 6 were contaminated with plaque sample obtained from chronic periodontitis patient. After 24 hours of incubation, 4 implants were decontaminated using various agents and 1 implant was irrigated using normal saline. All the 7 implants were again incubated for 24 hours and microbial analysis were performed.

Results: All the decontamination methods showed significant results with respect to the reduction in microbial load. 0.5% Doxycycline irrigation showed best results whereas 0.5% tetracycline irrigation showed less satisfactory results in terms of CFU comparatively with other decontamination agents in study.

Conclusions: Decontamination of implant surface using doxycycline is more effective in biofilm removal. An efficient removal of biofilm is critical as it may play a role in re-osseointegration.

Keywords- dental implant, decontamination, peri-implantitis, doxycycline, tetracycline, air- polishing device, chemotherapeutic agents.

Introduction :

In spite of the increasing survival rates of the oral implants, failures do occur which further require a substantial periodontal and prosthodontics maintenance over time. Traditionally, Implant failure has been described as early/ late when biological factors are considered: Early failure (Prior to implant loading) is because of surgical, implant or host-related factors. Late failure (after prosthodontics rehabilitation) is because of peri-implant disease or biomechanical overload which lead to bone loss around the implant and loss of osseointegration⁽¹⁾. Peri-implant mucositis and peri-implantitis account for the two most prevalent peri-implant diseases. Peri-implant mucositis is when inflammatory response is restricted to the soft tissues around a functioning oral implant and peri-implantitis is when inflammatory response includes the marginal bone loss around a functioning oral implant.

At first European workshop on periodontology (1993), peri-implantitis is defined as destructive inflammatory process

affecting the soft and hard tissues around osseointegrated implants, leading to the formation of a peri-implant pocket and loss of supporting bone. Bleeding on probing (BOP), suppuration, mobility, peri-implant probing and radiographic bone level evaluation are some of the diagnostic parameters for peri-implantitis. Presence of bleeding on probing indicates soft-tissue inflammation which is considered a significant parameter for the diagnosis of peri-implant disease⁽²⁾. Progressive bone loss is another important factor for implant failure. Peri-implantitis start progressing from the coronal part of the implant while simultaneously the apical portion of the implant maintains an osseointegrated status, resulting with a non-mobile implant until bone loss involves the complete implant surface⁽³⁾.

Etiological factors for peri-implantitis are microbial plaque accumulation, biomechanical overload and other factors like patient related factors (systemic disease, social factors, parafunctional habits, inadequate host bone support), iatrogenic factors (traumatic surgical techniques, lack of

primary stability, premature loading during the healing period), retrograde peri-implantitis and / or genetic polymorphism. Most significant factor in pathogenesis of implant failure is microbial plaque accumulation. Peri-implantitis is an infection which involves a large number of anaerobic microbes⁽⁴⁾. Dental implant can harbour bacteria which are not part of normal periodontopathic microbiota.

Commonly found microflora are *A. actinomycetocomitans*, *P. gingivalis*, *T. forsythia*, *P. intermedia*, *C. rectus* and less commonly found are *P. aeruginosa*, *Enterobacteriaceae*, *C. albicans*, *Staphylococci sp.* There are evidences for a bacterial cause of peri-implantitis. In development of a peri-implantitis *Staphylococcus aureus* plays the most important role. This bacterium has a large affinity to titanium⁽⁵⁾. It has a large positive (80%) and negative (90%) predictive value. Peri-implant microflora is established shortly after implant placement and for successful implants experience no shifts in microbial composition over time is required.

Prevalence of peri-implant mucositis ranged between 19-65%, whereas the prevalence of peri-implantitis varied from 10-40%. Removal of microbial plaque biofilm is a main pre-requisite in the management of peri-implantitis. There are three different methods to achieve this purpose, namely non-surgical (mechanical and chemical) and surgical methods. Mechanical cleansing using different prophylaxis methods such as ultra-sonic scalers, plastic tip ultrasonic scalers, stainless steel curettes, expanded polytetrafluoroethylene (e-PTEE) curettes, air-polishing systems, polishing rubber cups and brushes, abrasive pumice, airborne particles abrasive units (sodium bicarbonate, sodium carbonate, erythritol, glycine), plastic scalers. Chemical decontamination is done using local application of antimicrobial chlorhexidine solution or gel, stannous fluoride, tetracycline, minocycline, citric acid, hydrogen peroxide, 35% phosphoric acid etching gel. Photodynamic therapy and laser can be used for decontamination of implant surface to treat peri-implantitis by doing microbial reduction and biofilm control.

Removal of bacterial biofilm along with disinfection of implant surfaces is main aim of peri-implantitis treatment. However, because of special surface conditions and structures it is more difficult to remove bacterial biofilm from implant surfaces than teeth. Screw thread implant and intraosseous part of modern dental implants aggravates biofilm removal⁽⁶⁾. In this study erythritol and glycine are used as air-polishing agents and tetracycline, and doxycycline are used as chemical irrigants.

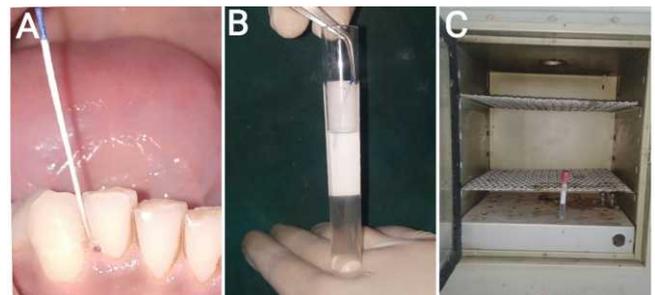
Erythritol is a natural sugar alcohol produced by the reduction of erythrose. That erythritol altered the microstructure and metabolomic profile of the biofilm produced by *S.gordonii* and *P.gingivalis* under in-vitro

conditions⁽⁷⁾. The amino acid glycine has shown to induce minimum tooth and implant surface alterations while still removing biofilm with efficacy in vitro and in vivo. It has been used in the treatment of peri-implantitis, demonstrating to be safe and to provide good clinical results^(8,9). Tetracycline is a bacteriostatic antibiotic that inhibits protein synthesis. It has been shown in many studies to allow re-osseointegration with arrest of disease and radiographic bone fill of the defect⁽¹⁰⁾. Doxycycline has been an effective antimicrobial for the treatment of periodontal diseases and peri-implantitis. It can be used systemically or locally. Application of Atridox gave significant benefits over mechanical debridement in terms of pocket probing depths and probing attachment levels⁽¹¹⁾.

Materials & methods –

Implant sample: A total of 7 threaded, tapered, titanium coated implants with machined surface were used (IMPLUS Cylindrical Implant, LEADER ITALIA S.r.l). All implants used were sterile and packed from company.

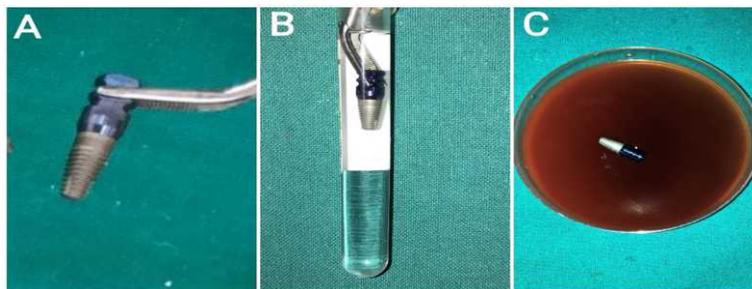
Plaque sample collection: A patient of generalized chronic periodontitis visiting outpatient department of periodontology and oral implantology was taken for collecting plaque sample. Sterile absorbent paper point was inserted in pocket (> 5mm deep), using sterile tweezer for 30 seconds. The same absorbent paper point was dipped into sterile normal saline (5 ml) stored in a sterile test tube. The test tube was kept on agitator so that the microbes attached to paper point gets detached and released in saline solution. The test tube was subsequently placed into incubator stored (at 37°C) in department of microbiology for 24 hours to allow incubation of micro-organism. (Figure 1)



(Figure 1 **Plaque sample collection** A: Sterile paper point insertion into periodontal pocket (> 5mm). B: Paper point dipped into test tube containing normal saline. C: Test tube placed into incubator.)

Contamination of implants: After 24-hour incubation, inoculated test tube was removed from incubator and 6 out of 7 implants were sequentially dipped in incubated test tubes for intentional contamination. (Except one implant – which is left sterile as a **control group I**). Contaminated implants

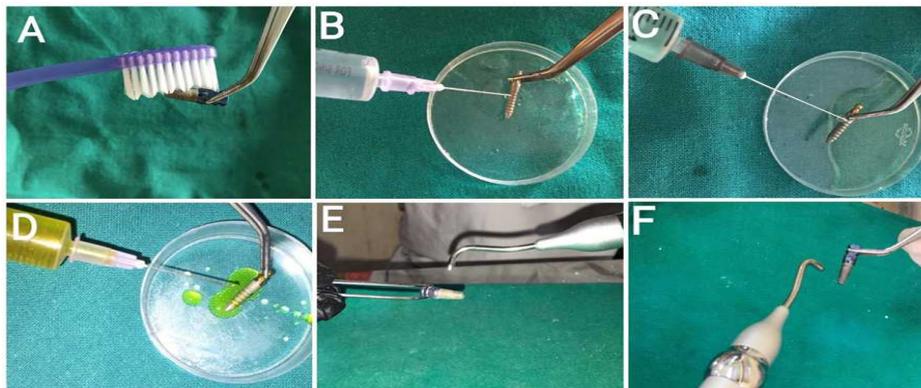
were then placed on sterile blood agar and then incubated for 24 hours at 37°C in an incubator. (Figure 2)



(Figure 2 **Contamination of implant** A: Implant sample used. B: Contamination of implant by dipping into incubated saline. C: Contaminated implant placed on sterile blood agar for incubation)

Decontamination of implants: Out of 6 contaminated implants 5 were cleaned with a soft nylon brush as mechanical debridement (Figure 3.A). And one was left as it is without mechanical debridement and decontamination to verify the amount of microbial deposits on implant surface. 4 implants were decontaminated using different decontaminating agents and 1 implant was decontaminated using normal saline irrigation which serves as control group II (Figure 3.B).

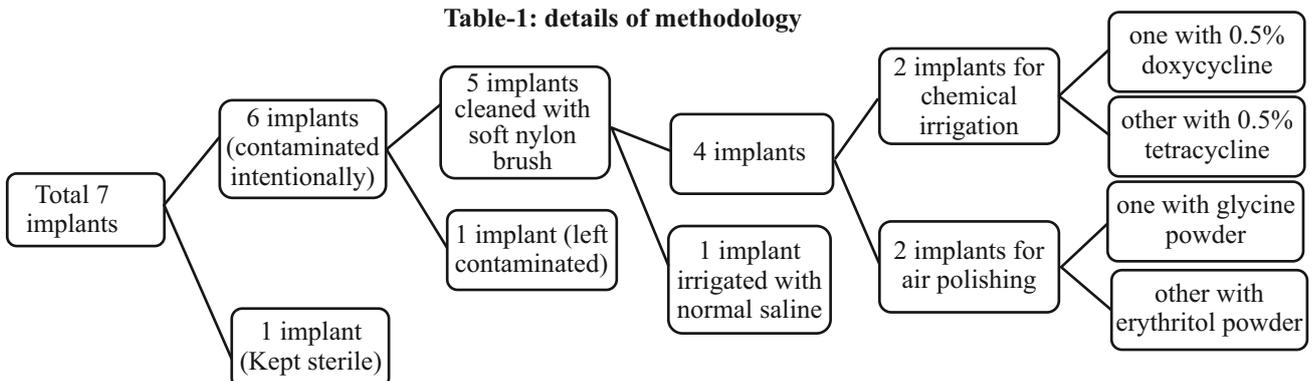
Decontamination agents used in this study were tetracycline, doxycycline, erythritol, and glycine. 1st implant was decontaminated using 5 ml of tetracycline (0.5%) solution irrigated for 30sec (Figure 3.C). 2nd implant was decontaminated using 5ml of doxycycline (0.5%) solution irrigated for 30sec (Figure 3.D). 3rd implant was decontaminated with erythritol using air- polishing device for 20sec (Figure 3.E). 4th implant was decontaminated with glycine using air- polishing device for 20sec (Figure 3.F).



(Figure 3 **Decontamination of implant** A: Mechanical debridement done using soft nylon brush. B: Irrigation with normal saline (Control group II). C Irrigation with tetracycline (0.5%) solution irrigation. D Irrigation with doxycycline (0.5%) solution irrigation. E: Decontamination using erythritol air polishing. F: Decontamination using glycine air polishing.)

After all the procedures 7 implants were kept in separate sterile blood agar medium and incubated for 24 hours. Details of methodology is explained in Table 1.

Table-1: details of methodology



Results:

All the decontamination methods showed significant results with respect to the reduction in microbial load. Doxycycline (0.5%) irrigation showed best results with least CFU (colony forming units), whereas 0.5% tetracycline irrigation showed

least satisfactory results with more CFU formation comparatively with other decontamination agents in study. Total microbial colony forming units (CFU) after decontaminating implant with different agents and incubating them for 24 hour is detailed in Table-2.

Table-2: Total microbial colony forming units (CFU) after 24hr

SR.NO	DECONTAMINATION METHODS	CFU at 24 hrs. incubation	
1	STERILE (Control group I)	0	
2	WITHOUT DECONTAMINATION	116	
3	IRRIGATION	TETRACYCLINE SOLUTION IRRIGATION	28
4		DOXYCYCLINE SOLUTION IRRIGATION	23
5	AIR POLISHING	ERYTHRITOL	31
6		GLYCINE	39
7	NORMAL SALINE (Control group II)	74	

Discussion:

Implant related periodontal diseases includes peri-implant mucositis which is reversible, presence of probing depth > 4mm with bleeding or suppuration with no sign of bone loss . On the other hand peri-implantitis involves probing depth > 4mm with bleeding, suppuration and radiographic bone loss beyond remodelling. Pathogenesis of peri-implantitis includes chronic inflammation starting as peri-implant mucositis, initially plaque formation at titanium surface and formation of biofilm, in which mostly gram negative anaerobes are involved. Increased number of neutrophils, lymphocytes and macrophages was found due to the inflammatory infiltration in connective tissue. Rate of disease progression is faster along with tissue destruction⁽¹²⁾. Treatment of peri-implantitis is determined by the amount of bone loss. If there is minimal bone loss (3 threads or less) one must proceed with treatment similar to peri-implant mucositis, along with that decontamination of prosthetic components is also needed. However surgical access with resective or regenerative components will need to be used if bone loss is advanced or progressive⁽¹³⁾.

Experimental study on dogs, which was on progression of peri-implantitis on different implant surfaces found that peri-implantitis was more progressive on rough surface when compared with machined one⁽¹⁴⁾. Because of this reason, use implants with smooth collar or completely machined surface mostly advised. That is why sterile, threaded, tapered, titanium coated implants with machined surface were used in our study. Out of 7 implants one was left sterile as control group I, to ensure proper sterilisation and the implants used in this study were devoid of any microbes. From all

contaminated implants (6), one was left contaminated (control group II) to know the CFU (colony forming units) before decontamination and compare with CFU values after use of different decontamination methods. Soft nylon brush is used for mechanical debridement and to avoid further surface roughness.

Results of air polishing systems are depending on the used medium and are significantly better in the following order: hydroxyl apatite/tricalcium phosphate > hydroxyl apatite > glycine > titanium dioxide > water and air (control group) > phosphoric acid. After air polishing therapy there was between 39% and 46% of re-osseointegration of titanium implants noted with increased clinical implant attachment and pocket depth reduction⁽¹⁵⁾. Greater amount of bacterial endotoxin removal with air powder abrasive treatment than citric acid, stannous fluoride, tetracycline HCL, chlorhexidine gluconate, hydrogen peroxide, chloramine T, sterile water and a plastic sonic scaler tip⁽¹⁶⁾. In their review (9 studies) on systemic and local antibiotic applications (e.g. tetracycline, doxycycline, amoxicillin, metronidazole, hydrochloride, ciprofloxacin, sulfonamide + trimethoprim) led to significant reductions of pocket depth in a period of between one to six years⁽¹⁷⁾. According to our study, doxycycline (0.5%) shows least CFU formation which was in accordance with in-vitro study in which they concluded that adjunctive use of 14% doxycycline gel might be viable option in management of peri-implantitis and peri-mucositis when used for decontamination of machined and sandblasted acid-etched implants. It also concluded that 14% doxycycline is effective in reducing bacterial colonization⁽¹⁸⁾.

Locally applied resorbable doxycycline releasing Nano spheres over 15 months period noticed the same⁽¹⁹⁾. After treating peri-implantitis with surgical debridement and the use of various antibiotics and combinations of them (including clindamycin, amoxicillin + metronidazole, tetracycline, ciprofloxacin) found success rate of 58%. Local or systemic antibiotics are considered as an options for additional therapy. Just administration of antibiotics is not treatment option⁽²⁰⁾. It is a supportive treatment which can be used in combination with other conservative or surgical treatments. Clinical peri-implantitis symptoms were more efficiently reduced⁽²¹⁾. There is increasing evidence of peri-implant inflammations representing one of the most frequent complications affecting both the surrounding soft and hard tissues which in future can lead to the loss of the implant. Therefore, it is necessary in this decade to add different strategies for prevention and treatment of peri-implant disease in modern rehabilitation concepts in dentistry for long term success and survival of dental implant. Also, there is need to understand the pathogenesis, etiology, risk factors, prevention of peri-implantitis to avoid further progression of peri-implantitis and implant failure.

Conclusion:

Decontamination of implants is effective in biofilm removal which in turns reduce the bacterial load. Tetracycline (0.5%), doxycycline (0.5%) solution irrigation and air-polishing with glycine, erythritol can be used as effective methods for decontamination. Within the limitations of our study, we found that **5 ml of 0.5% doxycycline irrigation for 30sec is more effective** than other decontamination methods when used as an irrigant on machined titanium surface.

Conflict of Interest: None

Funding source: None

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