IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)
e-ISSN: 2279-0853, p-ISSN: 2279-0861.Volume 16, Issue 6 Ver. VI (June. 2017), PP 06-12
www.iosrjournals.org

The Effect of Immersing Alginate Impression in Chloroxylenolon Level of Staphylococcus AureusandDimensional Change of Cast

SuryantoPurba[1],Syafrinani[2],LiaKusumawati[3]
[1,2]*Departmentof Prosthodontics, Faculty of Dentistry, Universitas Sumatera Utara, Medan*
[3] *Departement of Microbiology, Faculty of Medicine, Universitas Sumatera Utara, Medan*

Background
Aim: *The study aims to determine effect of immersing alginate impression in chloroxylenol for 1 minute, 5 minutes, 10 menits on level of Staphylococcus aureus and dimensional change of cast.*
Materials and methods :*The study was conducted on 2 types of sample, i.e.alginate impression and cast, 28 samples respectively. Microbial test was performed to count colonies of Staphylococcus aureus cultured in blood agar by using colony counter, and dimensional change was measured by using digital capiler. Kruskal-wallis test was performed to determine effect of chloroxylenol on level of Staphylococcus aureus, and Mann-Whitney test was performed to determine difference of the effectin control group and chloroxylenol immersed groups. One-way ANOVAtest was performed to determine effect of chloroxylenol on dimensional change and LSD test was performed to determine difference of the effect in control group and chloroxylenol-immersed groups.*
Results :*The study showed significant effect of immersing alginate impression in chloroxylenol on level of Staphylococcus aureus anddimensional change of cast.*
Conclusion :*The study suggested chloroxylenol disinfection by 5-minute 5% dettol liquid immersion, as it reduced Staphylococusaureusby 100% and maintained dimensional changes to < 0.20% that is consistent to ADA specification.*
Keywords :*alginate, chloroxylenol,Staphylococus aureus, dimensional change*

I. Introduction

One of the objectives of denture fabrication for edentulous patient is increase of quality of life. The first step of denture fabrication process includes impression procedure, i.e. anatomic impression to obtain study and working cast. The study cast is used as diagnostic cast, while working cast is used in fabrication of physiologic impression tray and denture for cases of Kennedy Class III and IV. Anatomic impression can be obtained by using irreversible hydrocolloid (alginate). Several advantages of alginate include easy manipulation, comfortable impression process, relatively inexpensive price of material, flexible properties and accurate impression.[1]However, one of the disadvantages of the material includes its imbibition property, which is absorbtion of liquid when placed in moist environment leading to easy swelling and syneresis of water when placed in open-air environment leading to shrinkage. The pressure of impression tray during impression-taking process could injure oral mucosa and gingiva causing bleeding, which leads to contact of blood and impression material. Blood, saliva and other exudates in the oral cavity contain many microorganisms. Miller and Cottone stated that a drop of saliva contained 50,000 of potentially pathogenic bacteria and could easily spread. *Staphylococusaureus*is one of the most common microorganims that is found in high level in the oral cavity. It causes several diseases such as pneumonia, skin lesions and osteomilitis. Thus, disinfection is needed to prevent cross infection of impression material containing this bacteria. To obtaincompatible and safe disinfectant, study is required to determine effect of immersing alginate impression in chloroxylenol for 1 minute, 5 minutes and 10 minutes on level of *Staphylococcus aureus*and dimensional change of alginate during its immersion in chloroxylenol. Impression material, particularly alginate, contains substantial number of bacteria that could cause cross infection in operator and other dental carers.[2,3,4] Alginate has 2-5 times more potential in becoming microorganism carrier compared to elastomer as it possesses more poreus texture that increases adherence ability of oral microorganisms leading to increase of cross infection.[5]

Disinfection of alginate impression in particular to prevent cross infection is an essential procedure. Disinfection could be performed by immersion in disinfectant substance, such as chloroxylenol, which is widely marketed as *Dettol*. However, immersion of alginate in liquid will lead to dimensional change of impression due to its imbibition property. Studies on the use of chloroxylenol in certain concentration to disinfect alginate impression remained scarce, which motivated the author to conduct the study to determine level of *Staphylococusaureus* after chloroxylenol disinfection and its effect on dimensional changes of impression after immersion.

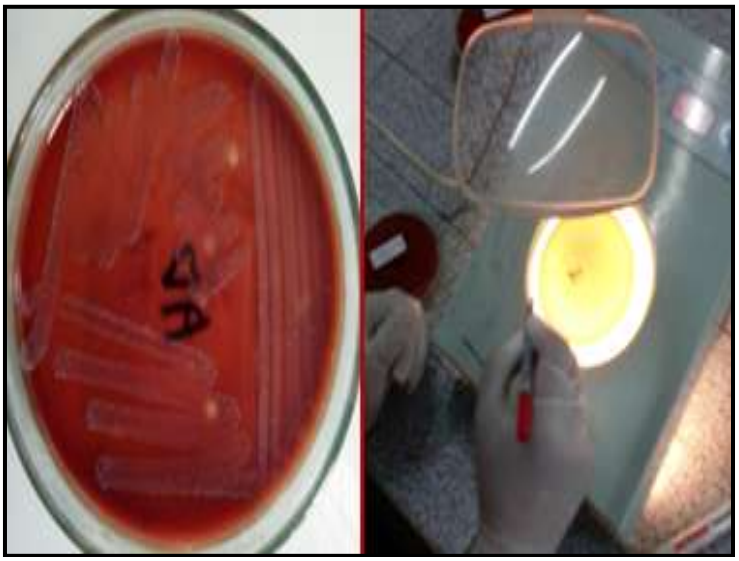
Several studies using different disinfectant material showed no dimensional changes in 5 and 10-minute immersion, which could possible occur with use of choloroxylenol in suggested concentration of medical use.[6] To obtain compatible and safe disinfectant, study is needed to determine effect of immersing alginate impression in chloroxylenol for 1 minute, 5 minutes and 10 minutes on level of *Staphylococcuesaureus*and dimensional change of alginate in its immersion.

II. Materials And Methods

Samples of the study were 28 master-cast alginate impressions, contaminated with *Staphylococcus aureus*and tested microbially to calculate number of *Staphylococcus aureus*according to ISO 6888-1 2003 and 28 casts that was fabricated by filling alginate impression with type III dental gyps to measure dimensional change. Microbial test was performed by retrieving master-castsfrom beaker glass filled with *Staphylococcus aureus*suspension, the casts were drained to create an equally contaminated surface. The alginate impression of the casts were made, rinsed with aquadest for 15 seconds and immersed in Dettol 5% for 1 minute, 5 minutes, and 10 minutes respectively. The surface of the casts was then swabbed with sterile cotton buds and blood agar culture wasmade(Figure 1). The blood agar media was placed in 37^{o}C incubator for 24 hours. The blood agar media wasretrivedand count of bacterial colony was performed with colony counter (Figure 2). Samples of control group in microbial test were alginate impressions that were only rinsed with sterile aquadest for 15 seconds and were not immersed in chloroxylenol solution. Kruskal-Wallis test was performed to determine effect of chloroxylenol on level of *Staphylococcus aureus*. Mann-Whitney test was performed to determine difference of effect in control group and other chloroxylenol-immersed groups.

(a) (b)

Figure 1.(a) Swab of alginate impression surface, (b)Bacteria culture in blood agar

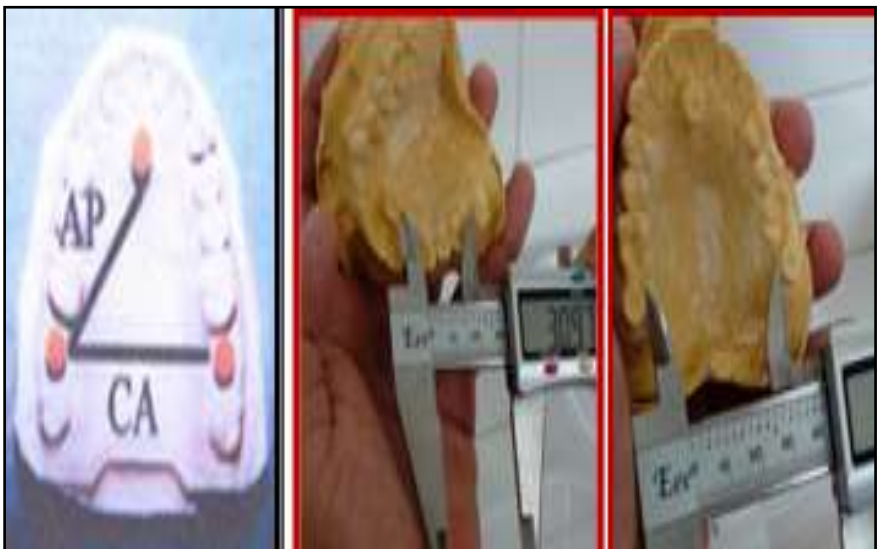


(a) (b)

Figure2.(a) Colony of *Staphylococcus* formed after incubation(b)Bacterial count usingcolony counter.

To measure dimensional changes, water and alginate impression material were put in alginator following factory instructions, centrifuged for 10 minutes with 2-bar pressure and loaded on the impression tray. Impression of the master cast was performed and once set, removed and rinsed with flowing water for 15 seconds, then dried with air spray for few seconds. Gyps were mixed by pouring 30 ml of water and 100 gr of type III gyps powder in vacuum mixer, mixed for 30 seconds and loaded to the alginate impression on vibrator to aid flow of material and release of air bubbles, then placed on secure place and left for 2 hours to set (factory instructions). Once set, the cast was removed from the alginate impression and dimensional changes was measured.Samples of control group in microbial test were alginate impressions that were only rinsed with flowing water for 15 seconds and were not immersed in chloroxylenol solution. Measurement of dimensional changes on casts was performed as following (Figure 3):[8,9]

1) Determination of measurement point on the outermost part of cylinder die circle
2) Length of space between the first and second point is antero-posterior dimension line (AP)
3) Length of space between the second and third point is cross-arch dimension line (CA)
4) Length of space between CA and AP of master-cast was measured before performing impression



(a) (b) (c)

Figure 3. Dimensional change measurement technique on working cast

(a)Determination of measurement points on outermost part of cylinder die circle
(b)Length of space between the first and second point is antero-posterior (AP)dimension line
(c) Length of space between the second and thirf point iscross arch (CA) dimension line

One-way ANOVA test was performed to determine effect of chloroxylenol on dimensional change, whildLSD test was performed to determine difference of the effect in control group and chloroxylenol-immersed groups.

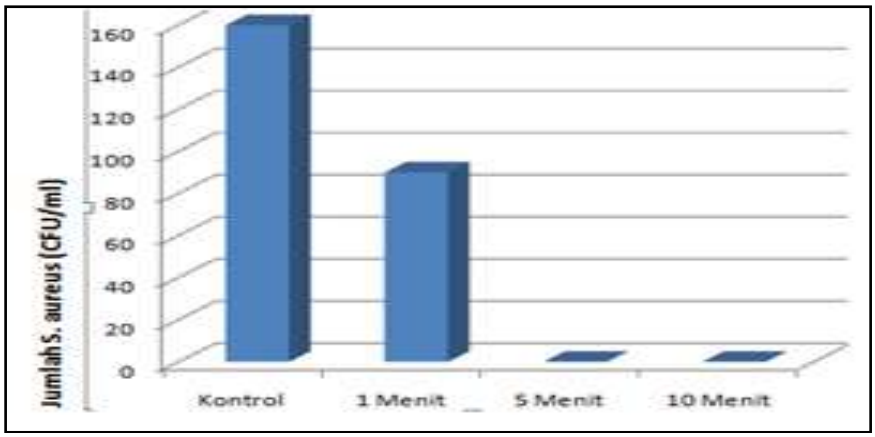
III. Results

Effect of Immersing Alginate Impression in Chloroxylenol on Level of Staphylococcus aureus

Results showed that median value of control group was 167 with interquartile range of 23. Median value of 1-minute chloroxylenol immersion group was 93 with interquartile range of 16. Median value of 5-minute chloroxylenol immersion group was 0 with interquartile range of 0. Median value of 10-minute chloroxylenol immersion group was 0. Effect of immersing alginate impression in chloroxylenol for 1 minute, 5 minutes, and 10 minutes on level of *Staphylococcus aureus* obtained by using Kruskal-Wallis test showed significant difference between control group and 1-minute, 5-minute, and 10-minute chroloxylenol immersion groups, p=0,0001 (p<0,05) and interquartile range of 0. Mann-Whitney test was performed to analyze difference of the effect of immersing alginate impression in chloroxylenol for 1, 5, and 10 minutes on level of *Staphylococcus aureus.* The test showed significant difference of control group and 1-minute chloroxylenol immersion group with p=0,002 (p<0.05), control group and 5-minute chloroxylenol immersion group with p=0,001 (p<0.05), and control group and 10-minute chloroxylenol immersion group with p=0,001 (p<0.05). Significant difference between 1-minute and 10-minute chloroxylenol immersion groups was also observed with p=0,001 (p<0.05). There was no significant difference between 5-minute and 10-minute chloroxylenol immersion group with p=0,1 (p<0,05) (Table 1) (Graphic 1)

Table 1. Effects of immersing alginate impression in chloroxyenol for 1 minute, 5 minutes, and 10 minutes on level of *Staphylococcus aureus*

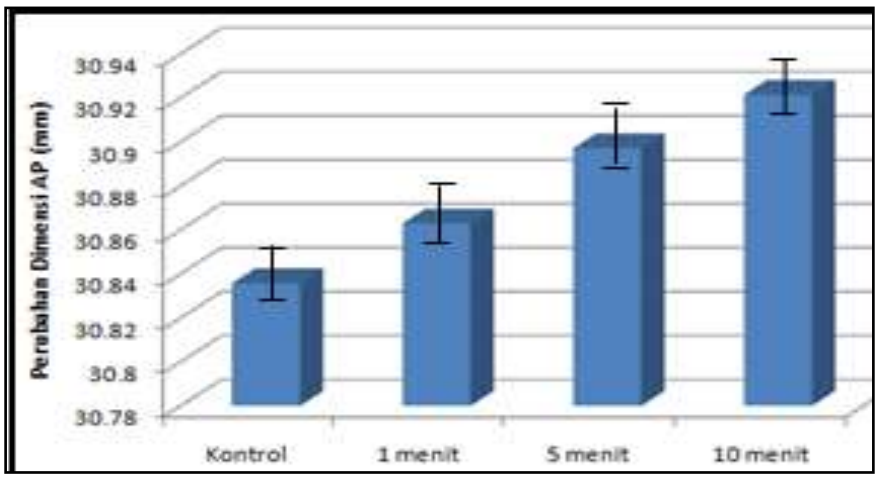
Group	Staphylococusaureus		p
	n	Median ± *interquartile range*	
control	7	167 ± 23	0,0001*
1 minute	7	93 ± 16	
5 minute	7	0	
10 minute	7	0	
The mean the dimensional change is different between:			
Control with 1 minute			0,002*
Control with 5 minute			0,001*
Control with 10 minute			0,001*
1 with 5 minute			0,001*
1 with 10 minute			0,001*
5 with 10 minute			0,1



Graphic1. Level of *Staphylococcus aureus*in control group and groups with 1-, 5-, and 10-minute chloroxylenol immersion

Effect of Immersing Alginate Impression in Chloroxylenol on Dimensional Change

Result showed mean value of AP line dimensional change measurements analyzed by univariant test. Mean value of control group was 30.84 mm with 0.007 deviationstandard and 0.0069% with 0.071 deviationstandard. Mean value of 1-minute chloroxylenol immersion group was 30.86 mm and 0.011 deviation standardand 0.073% with 0.036 deviationstandard. Mean value of 5-minute chloroxylenol immersion group was 30.89 mm and 0.011 deviation standard and 0.184% with 0.035 deviationstandard.Mean value of 10-minute chloroxylenol immersion group was 30.92 mm and 0.049deviationstandard and 0.236% with 0.159 deviationstandard. (Graphic 2)



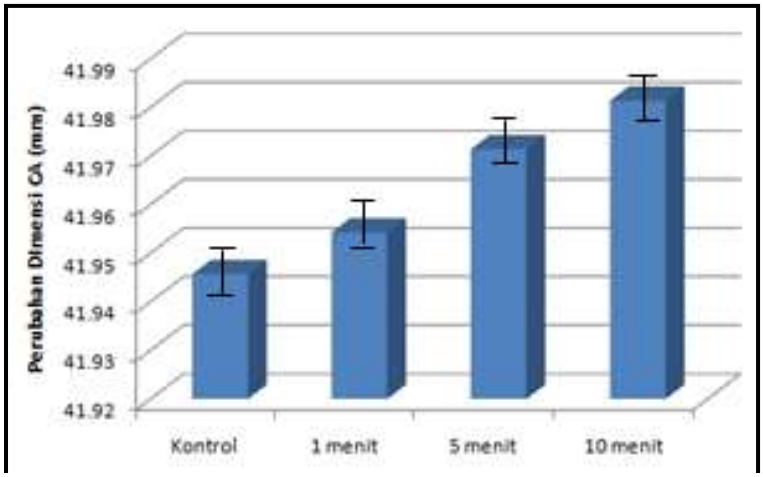
Graphic 2.Mean value of AP line dimensional change in control group and groups with 1-, 5-, and 10-minute chloroxylenol immersion

One way ANOVA test showed effect of immersing alginate impression in chloroxylenol for 1 minute, 5 minutes and 10 minutes on dimensional change by measuring dimensional change in AP line of working cast. The test showed significant difference in control group chloroxylenol immersion for 1, 5, and 10 minutes with p=0,001 (p<0.05) and followed by LSD test. (Table 2)

Table 2. Effect of Immersing Alginate Impression in Chloroxylenol on dimensional change in AP line dimension of working cast

Group	Dimensional change in AP line of Working Cast			p
	n	$\overline{X}$ ± SD (mm)	X ± SD (%)	
control	7	30,84 ± 0,007	0,069 ± 0,071	
1 minute	7	30,86 ± 0,011	0,073 ± 0,036	
5minute	7	30,89 ± 0,011	0,184 ± 0,035	
10 minute	7	30,92 ± 0,049	0,263 ± 0,159	0,001*
The mean the dimensional change is different between:				
Control with 1 minute				0,926
Control with 5 minute				0,026*
Control with 10 minute				0,001*
1 with 5 minute				0,031*
1 with 10 minute				0,001*
5 with 10 minute				0,118

Mean value of dimensional change in CA line of working cast was analyzed by univariant test. Mean value of control group is 41.94 mm with 0.025 mm deviation standard and 0.197% with 0.059 deviation standard. Mean value of 1-minute chloroxylenol immersion group is 41.95 mm with 0.007 deviation standard and 0.170% with 0.076 deviation standard. Mean value of 5-minute chloroxylenol immersion group is 41.97 mm with 0.018 deviation standard and 0.074% with 0.044 deviation standard.Mean value of 10-minute chloroxylenol immersion group is 41.98 mm with 0.019 deviation standard and 0.104% with 0.045 deviation standard. (Graphic 3)



Graphic 3. Mean value of CA line dimensional change in control group and groups with 1-, 5-, and 10-minute chloroxylenol immersion

One way ANOVA test on effect of immersing alginate impression in chloroxylenol for 1 minute, 5 minute, and 10 minutes on CA line dimensional change of cast and followed by LSD test, showed significant difference of control group and other groups with 1-, 5-, and 10-minute chloroxylenol immersion with p=0.002 (p<0.05). (Table 3)

Table 3. Effect of Immersing Alginate in Chloroxylenol on dimensional change in CA line of working cast

Group	Dimensional Change in CA line of Working Cast			p
	n	$\overline{X}$ ± SD (mm)	X ± SD (%)	
control	7	41.94 ± 0,025	0,197 ± 0,059	
1 minute	7	41.95 ± 0,007	0,170 ± 0,076	0,002*
5minute	7	41.97 ± 0,018	0,074 ± 0,044	
10 minute	7	41.98 ± 0,019	0,104 ± 0,045	
The mean the dimensional change is different between:				
Control with 1 minute				0,389
Control with 5 minute				0,001*

Control with 10 minute	0,006*
1 with 5 minute	0,005*
1 with 10 minute	0,043*
5 with 10 minute	0,340

IV. Discussion

Effect of immersing alginate impression in chloroxylenol on level of *Staphylococcusaureus* in 1-minute immersion group was found highest at 100 CFU/ml and lowest at 77 CFU/ml, median value of 93 and interquartile range of 16. This showed significant decrease. There were various theories of *Staphylococcus aureus*tolerance to chemical substances, among them were death within 15 minutes in 2% phenol, death within 3 minutes in 3% hydrogen peroxide and death within 1 minute in iodine tincture (Dzen SM *et al,* 2003). Decrease in level of *Staphylococcuesaureus*after 1-minute chloroxylenol immersion showed activity of the substance though not optimum. This finding is consistent to theory of Cottone*et al* which stated that length of immersion time was one of the factors determining effectivity of disinfectant material.[10] Despite its significant result, it is hoped that chloroxylenol could reduce level of *Staphylococcus aureus* by 100% on the surface of alginate impression.

Level of *Staphylococusaureus*in 5-and 10-minute chloroxylenol immersion groups showed equal decrease of zero, which meant no finding of *Stapylococcusaureus*colony. This result is consistent with study of Ghasemi*et al* in 2012 on microorganism contamination with *Staphylococcus aureus*suspension culture which showed that 5- and 10- minute of immersion in deconex disinfectant cleared up all microorganisms.[10] Study of El-Kholy and Sedky (2012) showed similar effectivity of Dettol, Lysoformin 3000 and sodium hypochlorite in 100% disinfecting impression by spraying it on the surface.[12] The current possible disinfection method for alginate impression is chemical disinfection by immersinf or spraying.[13] Immersing method provides several benefits such as the possibility of disinfectant substance to cover entire surface of impression, including undercuts. One of the materials used to disinfect alginate impression is chloroxylenol. Chloroxylenol (4-chlor-3,5-dimethylphenol) is an active halogen phenol material in Dettol, which is an intermediate level of disinfectant. This compound contains halogen, in particular chlor and works effectively against positive-gram bacteria.[14, 15, 16]Chloroxylenol promotes death of bacteria by disrupting its cell membrane which will lower its ability to produce ATP as source of energy. Kruskal-Walls test showed significant effect of Dettol 5% immersion incontrol group and group with 1-, 5-, 10-minute immersion on level of Staphylococcus aureus with $p=0.0001$ ($p<0.05$).

Effects of chloroxylenol immersion in dimensional changes of working casts between control group and group with 1-, 5-, 10-minute immersion shower various results in AP and CA line. There are several factors contributing to dimensional changes of alginate, including properties of alginate and gyps itself.One of them includes setting process of material, both chemical and temperature changes. The setting reaction will create pressure in the material which causes flow in the material to release the pressure of the material. Flow of impression material is ability of material to resist its force that is called viscosity. Skinner stated that dimensional changes could occur due to certain pressure or stress during gelation process of hydrocolloid impression material in impression taking process. Masri*et al* stated that pressure or force by impression material can be found on palatum area during impression takin process. The process of removing impression from the oral cavity is said to have cause dimensional change. Alginate is a viscoelastic material and snap removal technique is required to obtain elastic response. Several compression strain of material will occur during removal of impression in undercut area (10%) and could lead to permanent deformation (1.5%). [3] Dimensional changes in alginate could also be caused by its own imbibition property. Similar to other hydrocolloid material, alginate contains large amount of water and distorts easily.[17]

Dimensional change of gyps was contributed by several factors: room temperature, water temperature, water-powder ratio, speed of mixing, retarder, and accelerator. Property of gyps that caused dimensional changes is a result expansion process during setting due to growth of its crystal. Gyps expands during process of setting, which means that size of casts will be bigger than its impression and cause dimensional change of gyps casts.Variation in sizes of working cast, both AP and CA line, may be due to alginate properties during impression taking, when force given during impression on master cast holding board will combine viscoelastic strain (reversible) and viscous flow (irreversible)[3]. Graphic illustration showed that longer immersion leads to bigger dimensional changes both in AP and CA lines. This shows that dimensional changes of alginate impression material is affected by contact of the material with air, and its immersion in water and disinfectant solution. [21, 22] The result is different with theory of Oderinu*et al* on alginate that showed no dimensional changes of alginate immersed in 1% sodium hychrolite solution for 10 minutes, but significant changes when immersed for 20 and 30 minutes.[3]

In the study, AP line represented vertical plane and CA line represented horizontal planes of working casts[23] (Sari RF *et al*, 2013). Results showed dimensional change in AP line for $p=0.001$ and CA line for

p=0.002, which meant that dimensional change of alginate in both vertical and horizontal plane increased along with the length of immersion time.

V. Conclusion.

The use of *chloroxylenol* (Dettol 5%) in 5 minutes immersion could reduce level of *Staphylococcus aureus* by 100 and cause 0,184% dimensional changes which is acceptable based on ADA Specification No. 25, where tolerated dimensional change of type III gyps is 0-0.20%.[24]

References

[1]. Annusavice KJ. Bukuajarilmubahankedokterangigi. AlihBahasa Johan ArifBudimandan Susi Purwoko.Jakarta .Penerbitbukukedokteran. 2004: 103-114
[2]. Hatrick DC, EakleWS,Bird WF. Dental materials: Clinical appliance for dental assistant and dental hygienists. 2nd ed.new York: Elsevier. 2003: 179 – 80
[3]. Sastrodihardjo S. Perubahandimensihasilcetakangigidanmulut. USU Press. 2010: 71–82.
[4]. Pang SK,Millar BJ. Cross infection control of impressions a questionnaire survey of practice among private dentists in hongkong'. Hongkong dental journal. 2006:3: 89-93.
[5]. Haralur SB, Aldowah OS, GanaNS.AlHytam A. Effect of alginate chemical disinfection on bacterial count over gypsum cast. J.Adv Prosthodont.2012: 484-8
[6]. Amin WM. The Effects of Disinfectants on Dimensional Accuracy and Surface Quality of Impression Materials and Gypsum casts. J Clin Med Res.2009;1 (2); 81 - 89
[7]. Notoatmojo S, ,Metodologipenelitiankesehatan. RinekaCipta. Jakarta 2002:157-67
[8]. Farzin M, Panahandeh H. Effect of pouring time and storage temperature on dimensional stability of cast made from irreversible hydrocolloid. Journal of Dentistry. Tehran University of Medical science. 2010: 179-184
[9]. Izadi A.,Badamchizadeh S, Kahnamouyi HM, Marefat H, Dimensional stability of polyether, alginate, and silicone impression materials after disinfection with 2 % sanosil through the immersion technique' ,Avicenna J Dent Res. 2014;(2): 1-6
[10]. Cottone, J.A., Geza T. Terezhalmy, John A. Molinari. Mengendalikanpenyebaraninfeksipadapraktikdoktergigi. ahlibahasaLilianYuwono, 2000; (3):154-5
[11]. Ghasemi E, Badrian H, Khalighinejad N, 'Investigating the effects of spraying deconex on disinfection of three different impression materials'.J Dent Shiraz univ Med Scien, 2012: 13(3) 97-102
[12]. El-Kholy S.M. Sedky.N.A. 'Aplication of Infection Control Procedures in Dental Laboratories in Alexandria Governorate and The Efficacy of various Disinfectants on The Mostly Used Impression Materials. Egyptian journal 2012; (58):5-8
[13]. Kumar RN, Karthik KS, MallerSV.Infection Control in Prosthodontics.JIADS2010; (1):22-3
[14]. EL Mahmood, A.M. Doughari, J.H. Effect of dettol on viability of Some Microorganisms Associated with Nosocomial Infections. African Journal of Biotechnology 2008;Vol.7 (10): 1554-1562
[15]. Walter S, Klaus M, Manfred H. Senyawaobat. Penerjemah Joke R wattimena,SriwoelanSoebito, Gadjahmada university Press. 1990 :768 -771
[16]. American Cleaning Institute (ACI). Chloroxylenol antimicrobial efficacy & resistance.2014:1-15
[17]. ImberyTA,NehringJ,janus C, Moon PC. Accuracy and dimensional stability of extended pour and conventional alginate impression materials. J Am Dent Assoc 2010: 141 32-39
[18]. Hatrick DC, Eakle WS, Bird WF. Dental materials clinical applications for dental assistants and dental hygienists.2nd ed. Missouri: Saunders Elsevier.2011:203-6.
[19]. Power J.M, SakaguchiR.L.Restorative dental materials. 11th ed. Philadelphia: Mosby Elsevier 2006,: 270–283
[20]. Anusavice KJ. Philips' science of dental materials. 11th ed. St. Louis: Elsevier Inc 2003: 256- 77
[21]. Muzzafar D, ahsan SH, Afaq A. Dimensional changes in alginate impression during immersion in a disinfectant solution;.JPMA 2011: 61 (756)
[22]. Kohn WG, Harte JA, Malvitz DM, Collins AS, Cleveland JL, Eklund KJ. Guidelines for infection control in dental health care setting. JADA2004: 135 : 33 – 6
[23]. Sari RF, Parnaadji R, SumonoA.PengaruhTeknikDesinfeksidenganberbagaimacamlarutandesinfektanpadahasilcetakanalginatterhadapstabilitas dimensional. JurnalPustakaKesehatan. 2013 1(1): 29-34
[24]. American National Standards Institute. Revised american national standards/American dental association specification no.25 for dental gypsum products.