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Effects of Lidocaine Concentration on *Streptococcus Mutans* Bacteria

Muhammad Wildan Hari Subagya Haryono[1]
Poerwati Soetji Rahajoe[2]

[1]Department of Dentistry, Faculty of Medicine, Muhammadiyah University, Yogyakarta, Indonesia.
[2]Department of Oral & Maxillofacial Surgery, Faculty of Dentistry, Univrsitas Gadjah Mada, Yogyakarta, Indonesia.

E-mail: ibmfkgugm@yahoo.co.id
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Abstract

Lidocaine is one of the local anesthetics agents that are frequently used in dentistry. *Streptococcus mutan* is a gram-positive, facultatively an aerobic bacterium commonly found in human oral cavity. The study aims to find out the effects of Lidocaine concentration against *Streptococcus mutans* bacteria. The study used experimental design with liquid and solid dilution method to find out bacteriostatic and bacteriocidal effects of Lidocaine concentration. The test bacteria used in the study are the ones, which have been compared with Mcfarland standard solution with 10µCFU/ML concentration. The Lidocaine concentrations were 2%, 1%, 0.5%, 0.25%, 0.125%, 0.0625%, 0.03125%, 0.015625%, and 0.007812%. Kruskal-Wallis and Mann-Whitney methods were used for the statistical test. The result of the study suggests that Lidocaine concentration has effects against *Streptococcus mutans* bacteria in the concentrations of 2%, 1% and 0.5%, to inhibit bacterial growth ($p<0.05$), but there was no bacteriocidal potential of Lidocaine concentration against *Streptococcus mutans* bacteria ($p>0.05$).

Keywords: Lidocaine, *Streptococcus mutans*, anti-bacterial.

1. Introduction

Local anesthetics have some contents in its preparation, one of which is anesthetics agent. An example of the agent is Lidocaine. Due to its beneficial characteristics, Lidocaine is commonly used in dentistry at present.

Meanwhile, awareness towards the danger of contamination like infection, which can be transmitted through direct contact with blood, saliva and contaminated instruments has been growing up. Human oral cavity is basically not free from germs. *Streptococcus mutans* is one of the normal germs in the human body. A germ can be said as normal if it is not harmful to human body. *Streptococcus mutans* can be harmful germs for human body if the number of harmful and beneficial germs in the human body is imbalanced.

Antibacterial power is the power of agents which inhibits and kills the growth of germs. Proper anesthetics agent is the one which has antibacterial power, no matter how small it is. Administration of anesthetics agent with anti-bacterial power in dental and oral treatment serves as a preventive treatment for the possible infection and bacteremia.

2. Methods

This study used laboratory-based experiment method. The subjects of the study were standard *Streptococcus mutans* bacteria available in the Microbiology Laboratory of The Faculty of Medicine, Muhammadiyah University, Yogyakarta. Lidocaine in 2% concentration was provided by Oral and Maxillofacial Surgery Laboratory of the Faculty of Dentistry, Gadjah Mada University taken from a certain trademark which is commercially available.

The independent variable during the study was Lidocaine concentration, whereas the independent variable was bacteriostatic potential and/or bacteriocidal potential against *Streptococcus mutans* in the Trypticase Soy Agar (TSA) as the media.

The study used two methods; finding Lidocaine concentration with bacteriostatic potential against the growth of *Streptococcus mutans* bacteria using liquid dilution method, and finding Lidocaine concentration with bactericidal potential against *Streptococcus mutans* bacteria using solid dilution method.

The collected data was analyzed by Kruskal-Wallis and Mann-Whitney Test.

3. Results

Table 1 indicates that 2% and 1% Lidocaine concentrations have bacteristatic potential against *Streptococcus mutans* bacteria. The 0.5% concentration was the border line between the concentration with inhibiting power and the one without, as suggested by 3 times repetition with 2 results indicating bacteriostatic potential. The lower concentration than 0.25% indicates that there was no effect on the bacterial growth inhibition against *Streptococcus mutans*. The baceriocidal effect was only found once within 3 times repetition which refers to 2% concentration.

The Kruskal-Wallis analysis on Table 2 suggests that there was a difference among various Lidocaine concentrations towards inhibiting potential against *Streptococcus Mutans* ($p<0.050$). Thus, it can be concluded that various Lidocaine concentrations have effects to bacteristatic potential against *Streptococcus mutans*.

Table 1. Bacteriostatic and bacteriocidal effects of Lidocaine against *Streptococcus mutans.*

Concentration (%)	Bacteriostatic (Σ)		Concentration (%)	Bacteriocidal (Σ)	
	+	-		+	-
2	3	0	2	1	2
1	3	0	1	0	3
0.5	2	1	0.5	0	3
0.25	0	3	0.25	0	3
0.125	0	3	0.125	0	3
0.0625	0	3	0.0625	0	3
0.03125	0	3	0.03125	0	3
0.015625	0	3	0.015625	0	3
0.007812	0	3	0.007812	0	3

(+) indicates that test concentration has both bacteriostatic/ bacteriocidal effects
(-) indicates that test concentration has no bacteriostatic/ bacteriocidal effects
Σ Bacteristatic/ bacteriocidal effects were results of 3 times repetition

Table 2. Summary of the statistical test on various Lidocaine concentrations as bacteriostatic agent against *Streptococcus mutans.*

Concentration %				P*
2	1	0.5	0.25	0.003

* Significance value based on Kruskal-Wallis Test.

Table 3 shows the results on the differences between two Lidocaine concentrations as bacteriostatic agent againts *Streptococcus mutans* by Mann-Whitney Test.

A significant difference was obtained by 2% - 0.25% concentration and 1% - 0.25% concentration, meanwhile no significant difference was found in 2% - 1% concentration , 2% - 0.5% concentration , 1% - 0.5% concentration, and 0.5% - 0.25% concentration. This finding suggests that significant difference between two test concentrations will generate different effects to inhibiting the growth of *Streptococcus mutans*, whereas no significant

difference indicates that two test concentrations generate the same effects to inhibiting potential against the growth of *Streptococcus mutans*.

Table 3. Analysis-based summary of comparison between two Lidocaine concentrations towards inhibiting power against *Streptococcus Mutans.*

Concentration %	P*
2% - 1%	1.000
2% - 0.5%	0.317
2% - 0.25 %	0.025*
1% - 0.5%	0.317
1% - 0.25%	0.025*
0.5% - 0.25%	0.114

* Significance value based on Mann-Whitney Test

Table 4. Summary of the statistical test on various Lidocaine concentrations as bacteriocidal agent against *Streptococcus mutans.*

Concentration %				P*
2	1	0.5	0.25	0.433

* Significance value based on Mann-Whitney Test

The Kruskal-Wallis analysis on Table 4 indicates that no difference among various Lidocaine concentration on its bacteriocidal effect against *Streptococcus mutans* ($p>0.05$). Thus, conclusion can be drawn that various Lidocaine concentrations bring no effects on bacteriocidal potential against *Streptococcus mutans*.

4. Discussion

Lidocaine-based research against Streptococcus mutans indicates that 2%, 1% and 0.5% Lidocaine concentrations bring bacteriostatic effect against *Streptococcus mutans*. This finding was proven by observing the baseline between test concentration and positive control, as well as negative control. The positive control serves as bacterial suspension on BHI media, meanwhile the negative control contains residue of dilution. All positive controls resulted (+) by 3 times repetition, which suggested that *Streptococcus mutans* - containing positive control remains potential to inhibit bacterial growth. The negative control also resulted (-) during by 3 times repetition indicating that Lidocaine may able to act as anti-bacterial agent.

Comparative test between two Lidocaine concentrations tested against *Streptococcus mutans* suggests that the administration of 2%, 1% and 0.5% concentration will bring the same bacteriostatic effects against the growth of *Streptococcus mutans*, whereas 0.25% concentration will generate different effects from the 3 higher concentrations as it has no inhibiting potential against the growth of *Streptococcus mutans*.

The result of the study indicates that Lidocaine has anti-bacterial potential against *Streptococcus mutans* with its capability of inhibiting the growth of bacteria (bacteriostatic). The result corresponds to another study[1] which states that Lidocaine has anti-bacterial effect with bacteriostatic characteristic against test bacteria[1], and is supported by a study conducted by Gajraj *et al.*[2] which suggest that Lidocaine has anti-bacterial power against 2 test bacteria when it is mixed with Diprivan. Lidocaine, however, is less potential compared to Procaine and Prilocaine.

The mechanism of anti-bacterial potential of local anesthetics agent has not been clearly known so far, but there are two possible mechanisms to explain the phenomena; first, mechanism of agent's effect towards structure or function of membrane, and second, mechanism of pH alteration of bacterial cells[2], which is supported by the study done by Kaya *et al.*[1] suggesting that effects of Lidocaine's inhibiting power represented by synthesis inhibition against bacterial cell wall[1]. This phenomenon corresponds to the action from mechanism of anti-bacterial potential of certain drugs against bacteria. Anti-bacterial that works by inhibition of bacterial cell wall synthesis is assumed to be influenced by peptidoglycan synthesis. Here, peptidoglycan is considered as compound needed to synthesize

bacterial wall, which is composed from glucose and amino acid. The inhibition mechanism of peptidoglycan synthesis is done by preventing transport of peptidoglycan monomer synthesis produced by cytosol passing through cytoplasm membrane[3].

Lidocaine is a local anesthetics agent formed by combination of weak base and strong base. The agent can easily be hydrolyzed on cellular tissue which is later bound by fatty layer on cytoplasm membrane to produce selective membrane permeability[4]. Selectivity of membrane permeability on bacterial cell will prevent the transport of peptidoglycan monomer synthesis, which is a composing compound for bacterial wall. The inhibition of bacterial wall composer will inhibit the growth of bacterial cell caused by weakened cellular tissue in cell.

Streptococcus mutans is a Gram-positive bacteria. Gram-positive germs have multi-layered veiled cells, which consist of cell wall and cytoplasm membrane. The wall consists of relations between peptidoglycan, teichoic acid, polysaccharide and protein. The wall of Gram-positive bacteria is thicker than that of Gram-negative with 40: 1 in comparison. The thick wall is hydrophobic. Teichoic acid serves as a connector between membrane and peptidoglycan which binds with glycerol in the tip of membrane[5,6]. Cytoplasm membrane of Gram-positive bacteria consist of phospolipid bilayer which is hydrophilic on surface and hydrophobic in core.

The result of the study indicates that Lidocaine at test concentration has no bactericidal effect against *Streptococcus mutans*. This finding corresponds to other study[1] which suggests that Lidocaine at tested concentration gives no bacteriocidal effects to some pathogenic bacteria generating nosocomial infection.

5. Conclusion

Based on the result of the study, it can be concluded that: Lidocaine has effects of anti-bacterial potential against *Streptococcus mutans*. The antibacterial potential is shown by bacteriostatic potential at 2%, 1% and 0.5% concentrations and minimum bacteriostatic potential at 0.5% concentration. Lidocaine has no minimum bacteriocidal potential against *Streptococcus mutans* at tested concentration.

6. References

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