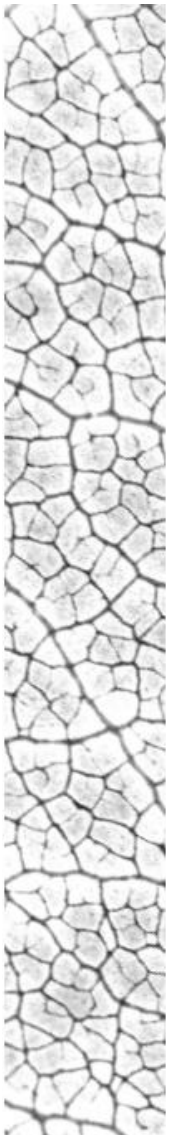

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The effectiveness of Taro (*Colocasia esculenta L. Schott*) leaf extract as a denture cleanser against *Candida albicans* in acrylic plate

Olivia Ananto Putri [1], Utari Kresnoadi [1*], Agus Dahlan [1]

[1] Department of Prosthodontics, Faculty of Dental Medicine, Universitas Airlangga, INDONESIA
*Corresponding author: utari-k@fkg.unair.ac.id

Abstract
Denture wearers use cleanser with chemical cleaning solution to eliminate the formation of plaque, tartar and food scraps, as well as bad breath. Thus, many natural materials used as alternative synthetic materials have recently been developed. One of natural materials developed is Taro Leaf. To determine the effectiveness of Taro leaf extract as a denture cleanser against *Candida albicans* on acrylic plate. The acrylic plate was soaked in sterile saliva and then soaked in *Candida albicans* for 24 hours. Those samples then were divided into four groups, namely one control group and using Taro leaf extract at concentrations of 40%, 60%, and 80%. After the treatment, they were planted on Sabaroud Dextrose Agar and then incubated for 24 hours. After 24 hours, the visible growth of *Candida albicans* colonies was measured. The results then were analyzed by using the Kolmogrov Smirnov, Levene test, one way ANOVA, and Tukey HSD test. There was a significant difference in the growth of *Candida albicans* colonies between the treatment group using Taro leaf extract at the concentration of 40%, 60% and 80%. This study can be concluded that the immersion of acrylic resin plate in taro leaf extract (*Colocasia esculenta L. Schott*) with a concentration of 80% can effectively decrease the number of colonies of *Candida albicans* on acrylic resins.

Keywords: antifungal, taro leaf extract, denture cleanser, resin acrylic, *Candida albicans*

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INTRODUCTION

Denture wearers who continually equip the denture without cleaning it regularly can increase the accumulation of plaque and can cause inflammation in the oral mucosa, such as denture stomatitis. *Candida albicans* is known as pathogenic microorganisms that are able to produce hydrolytic enzymes that are toxic and can cause denture stomatitis (Bhat, et al. 2011). Index usage of protesa in Indonesia, based on the data of the national health research, was 4.5% with 5.0% on men and 5.6% women (Widati, K. C. 2017).

Removable partial dentures can be cleaned mechanically, chemically, or a combination of both. The materials that are often used as cleaning agent is alkaline peroxide and alkaline hipochloride. Test the effect of alkaline cleaning agents hypochlorite and alkaline peroxide showed discoloration of the denture base (Gajwani-Jain, et al. 2015; Kazanji, & Ahmad, 2004). Evaluation of the effect of some denture cleansers on the colour of acrylic resin denture base materials (Kazanji, & Ahmad, 2004).

Denture cleansing should have characteristics such as non-toxic, the ability to resist the deposit of organic or inorganic materials on the denture, harmless for all types of denture including denture base polymer, aluminum, acrylic, dental porcelain and lining material, stable when stored, and bactericidal and fungicidal (Naeem, et al. 2015).

Thus, many natural materials are developed as a healthier alternative to synthetic materials. One of the natural cleanser is Taro leaf. The leaf contains some useful compounds, such as flavonoids, saponins, and tannins which have antifungal effects. The taro plant is one of the tubers are cultivated in the area started in Papua and are found most frequently in Bogor, Malang and Bali (Talas, 2003; Holli, et al, 2017).

The purpose of this research is to determine the effectiveness of Taro leaves extract as a denture cleanser against *Candida albicans* on acrylic plate by using a reference concentration of 40%, 60% and 80%. Previous experiments reveal that a concentration of 40% and 60% decline the colonies of microorganism and in

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the concentration of 80% there is no growth of microorganism (Yusticia, 2018).

MATERIAL AND METHODS

This research is an experimental laboratory research, and have received an ethical clearance from the Ethic Commite of Faculty of Dental Medicine, Universitas Airlangga No. 057/HRECC.FODM/V/2018. The research is divided into five steps. Step one, Taro leaf extracting (Bagus Satriya, 2019)., taro leaves were taken and washed out, then cut into parts and dried for 6 hours. The leaves were inserted into extractor machine, then added solvent ethanol 96% twice the weight of the taro leaves and shaken for 24 hours. Following after, the leaves were filtered so that clear filtrate is obtained. The extract was evaporated with the evaporator machine at 50-60° for 3-5 hours and it will be obtained a thick greenish taro leaves extract which has been free of solvents of ethanol which is considered 100%.

Step two is the dilution of Taro leaves (Bagus Satriya, 2019). to obtain extract with a concentration 40%, 60% and 80% dilution by using solvent aquades. To obtain concentration 40% dilute 40 ml of taro leaves extract with 60 ml aquades, for 60% concentration dilute 60 ml of taro leaves extract with 40 ml aquades, and for 80% concentration, dilute 80 ml of taro leaves extract with 20 ml aquades.

Step three is the sample treatment (Fakhriyana, & Salim, 2010). Acrylic plate was sterilized in 121^{o} of autoclave machine during 18 minutes, and then soaked the acrylic plate into sterile saliva for 1 hour and rinsed with PBS as much as 2 times. After that, the acrylic plate was soaked into *Candida albicans* for 24 hours. In Tube 1, the acrylic is soaked in 5ml of aquades for 15 minutes as control; in tube 2, the acrylic plate is soaked in 5ml of 40% taro leaves extract in 15 minutes; in tube 2, the acrylic plate is soaked in 5ml of 60% taro leaves extract in 15 minutes; ; in tube 2, the acrylic plate is soaked in 5ml of 80% taro leaves extract in 15 minutes. After that, PBS is used to rinse the plate twice to removed the taro leaves extract from the surface of the plate.

Step four is the cultivation of *Candida albicans*. This is done by putting the acrylic plates that have been rinsed in 5ml of SDB (Sabouraud Dextrose Broth) and soaked into vibrator machine to remove the *Candida albicans* from the surface of the plate. After that, put the liquid into media of SDA (Sabaroud Dextrose Agar) then spride the liquid and incubate for 48 hours with 37°C.

And the last step is the calculation of *Candida albicans* (Wijaya, Utari, & Yudianingsih, 2015). After incubation for 48 hours, the calculation of colony is done using a Quebec Colony Counter colony count. The results then were analyzed by using the Kolmogrov Smirnov, Levene test, one way ANOVA, and Tukey HSD test.

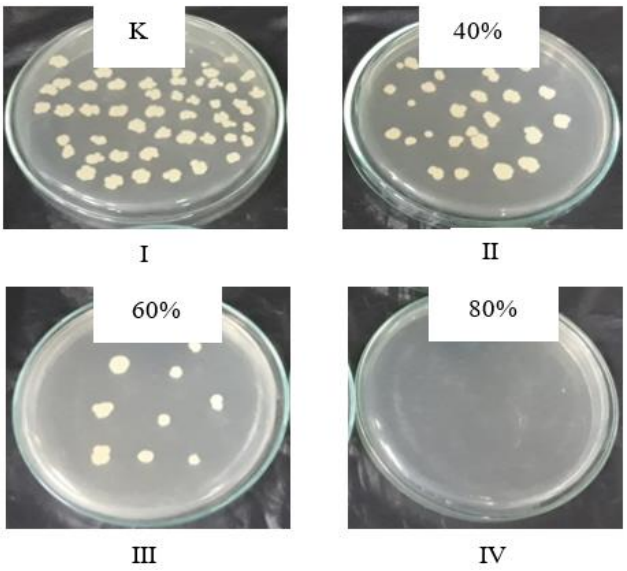

Fig. 1. The colonies of *Candida albicans* after being soaked with an extract of taro leaves in 15 minutes, Plate I: control (immersion in aquades), Plate II: Results of immersion with a concentration of 40%, Plate III: Results of immersion with a concentration of 60%, Plate IV: Results of immersion with a concentration of 80%.

Table 1. The mean value and Standard Deviation of colonies *Candida albicans*

Treatment group	N	R	SB	Sig
1	10	68.3000	3.30151	0.000
2	10	31.6000	2.22111	
3	10	11.0000	1.76383	
4	10	0	0	

N = Number of treatments
R = Mean of research results
SB = Standard deviation
1 = Number of colonies of *Candida albicans* that were soaked in aquades
2 = Number of colonies of *Candida albicans* that were soaked in 40% taro leaf extract
3 = Number of colonies of *Candida albicans* were soaked in 60% taro leaf extract
4 = Number of colonies of *Candida albicans* were soaked in 80% taro leaf extract

RESULTS

The number of *Candida albicans* colonies on heat-cured acrylic resin after immersion in taro leaves extract with a concentration of 40%, 60%, 80% and distilled aquades as a control. Results can be seen in **Figure 1**.

In **Figure 1**, it is known that in the results of plate II (concentration 40%) and plate III (concentration 60%), there are still colonies of Candida albicans, while the results of plate IV that used the concentration of 80% taro leaves extract showed that there is no growth of Candida albicans colonies.

From the research data in **Table 1**, the data is processed to see the mean and standard deviations growth of *Candida albicans* colonies of each group od taro leaves concentration.

The results showed that there is inhibition of taro leaves extract against *Candida albicans* colonies.

Table 2. Tukey HSD test

Treatment group	Control	40%	60%	80%	Sig.
Control	-	*	*	*	0000
40%	*	-	*	*	0000
60%	*	*	-	*	0000
80%	*	*	*	-	0000

The results showed that there were significant differences of the effectiveness between groups differences in concentration.

DISCUSSION

Taro (*Colocasia esculenta*) is a tuberous medicinal plant is a rich source of tarin, a GNA-related lectin. The effectiveness of taro leaf extract as a denture cleaning agent against *Candida albicans* in the oral cavity in acrylic resin, the acrylic resin plate was soaked first on sterilized saliva for 1 hour to bind *Candida albicans.* After that, the acrylic plate was soaked into *Candida albicans* for 24 hours. Then the acrylic plate was soaked in a tube containing taro leaves extract with a concentration of 40%, 60%, 80% and a tube containing sterile aquades used as a control for 15 minutes. After that, rinsed the acrylic plate with PBS twice to removed the taro leaves extract from the surface of the plate and inserted into the Sabouraud Dextrose Broth (SDB) and soaked into vibrator machine to removed the *Candida albicans* from the surface of the plate. After that, the liquid was put into media of SDA (Sabaroud Dextrose Agar) then spride the liquid and incubation for 48 hours to determine the number of colonies from *Candida albicans.*

The results of acrylic plate immersion at the concentration of taro leaves extract at 40% showed 31.6 cfu/ml growth of *Candida albicans* colonies, the 60% extract concentration showed 11 cfu/ml growth of *Candida albicans* colonies was, and the immersion in 80% taro leaves extract did not show any growth of *Candida albicans* colonies, while the results of the control (using sterile aquades) showed the highest growth of *Candida albicans* colonies was 68.3 cfu/ml. These results indicate that the higher the concentration of taro leaves extract, the less the number of colonies from Candida albicans that grow in the research media.

According to other researchers, the British Pharmacopoeia states that the use of chemical disinfectants must be able to damage pathogenic organisms, and in practice can work as both bactericides and fungicides; which means they have the ability to kill microorganisms perfectly (Regezi, Sciubba, & Jordan, 2003). Other researchers also concluded that taro leaves extract is useful for the treatment of diseases caused by bacteria and fungi. The use of taro leaves extract as an antibacterial and antifungal can be emphasized as herbal medicines in various treatments (Dutta, & Aich, 2017).

The results of this research are consistent with the results of research conducted at the Industrial Research and Consultation Center that taro leaves extract contains a chemical compound like 2.15% tannins, 4.57% saponins, and 3.06% flavonoids, which has been known in previous studies that these chemical compounds can kill and reduce the growth of fungi (Wijaya, 2014).

As an antifungal, flavonoids have genestein compounds which function is to inhibit cell division or proliferation. (Machmud, 2018). The antifungal mechanism possessed by tannins is due to their ability to inhibit chitin synthesis which is used to form cell walls in fungi and damage cell membranes so that fungal growth is inhibited. And the antifungal of saponins originate from the formation of polar saponin compound bonds with lipoproteins and non-polar saponin group bonds with fungal plasma membrane fat cells.

Natural or manufactured food fortified by tarin could contribute to health maintenance and human well-being. Tarin activity can be explored as a putative immunomodulatory by bringing new evidence regarding potential boosting of the immunological system in order to partially reverse immunosuppressive effects. Immunosupressive states may take advantage of tarin effects on hematopoietic and progenitor cells. Tarin may also be part of future cancer therapeutics, since it may interact directly with cancer cells to inhibit tumor proliferation and the migration of malignant cells. Tarin, in its purified form described herein, could be a promising adjuvant in chemotherapeutic treatment if animal and clinical trials support its effectiveness with no serious side effects to human cells associated with the protein preparation (Pereira, et al. 2018).

CONCLUSION

It can be concluded from this research that immersion of acrylic resin in Taro leaves extract (*Colocasia esculenta L. Schott*) with a concentration of 80% is the most effective concentration as an acrylic plate cleaner against *Candida albicans* in the oral cavity.

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