

Effect Of Heat-Cured Acrylic Resin Soaking In Star Fruit Leaf Extract Solution (*Averrhoa bilimbi*) On *Candida albicans* Colony

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Abstract

Denture stomatitis is a common inflammatory condition among elderly denture wearers and is closely associated with biofilm accumulation on heat-cured acrylic resin dentures, which promotes the proliferation of *Candida albicans*. Star fruit leaves (*Averrhoa bilimbi* L.) contain tannins, flavonoids, and saponins with known antifungal activity. This study aimed to evaluate the effect of a 50% star fruit leaf extract solution on the growth of *C. albicans* colonies on heat-cured acrylic resin. A laboratory experimental design with a pre-test–post-test control group was used. Colony counts were obtained before and after treatment, and statistical analysis included the Shapiro–Wilk test, Levene’s test, and paired t-test. The mean number of *C. albicans* colonies on acrylic resin plates soaked in 50% extract was 1.356×10^5 , compared with 1.587×10^6 in the aquadest control group. The paired t-test showed a significant reduction in colony count in the treatment group ($p < 0.05$). These findings indicate that soaking heat-cured acrylic resin in a 50% star fruit leaf extract solution effectively inhibits the growth of *C. albicans* colonies.

Keywords: Heat-cured acrylic resin, Star fruit leaf extract, *Candida albicans*

Introduction

All individuals will eventually enter the elderly phase, a stage where people expect to continue living in healthy, meaningful, productive, and dignified conditions. According to the Law of the Republic of Indonesia No. 13 of 1998 Article 1 Paragraph 2 concerning elderly welfare, an elderly person is defined as an individual aged sixty years or older. During this stage, the aging process gradually reduces the ability of body tissues to repair, replace, and maintain normal function, leading to various biological and functional changes [1].

Tooth decay, loss of periodontal attachment, and alveolar bone resorption are common periodontal tissue changes in the elderly. If left untreated, these

conditions can lead to increased tooth mobility and even tooth loss. As a result, oral function and daily activities may be affected, ultimately lowering the individual's quality of life. Individuals who experience tooth loss often require dentures to restore oral function and improve comfort [1].

Dentures are a common treatment option used to replace missing teeth and restore oral function in edentulous patients. The denture base, which directly contacts the oral mucosa, plays an essential role in supporting the denture, distributing occlusal forces to the underlying tissues, and maintaining retention and stability. Heat-cured acrylic resin is the most widely used denture base material because it is economical,

non-toxic, non-irritating, insoluble in oral fluids, aesthetically acceptable, and easy to manipulate [1].

However, heat-cured acrylic resin has several disadvantages, including susceptibility to fracture and a porous surface, which facilitates food debris retention and plaque accumulation. These conditions can increase the colonization and growth of *C. albicans* when denture hygiene is inadequate. As a result, denture users are at risk of developing denture stomatitis, an inflammatory condition of the oral mucosa caused by contact with the fitting surface of the denture [1, 2]. To prevent microbial accumulation, dentures must be disinfected routinely. Chemical disinfectants are commonly used for this purpose; however, limited accessibility and cost may restrict their use among some individuals. As an alternative, herbal-based disinfectants may be considered, including extracts from star fruit (*Averrhoa bilimbi* L.) [1, 2, 3, 4]. Star fruit (*Averrhoa bilimbi* L.) has traditionally been used as a medicinal plant due to its bioactive compounds, including flavonoids, which possess antimicrobial properties effective against viruses, bacteria, and fungi [5, 6].

Methods

This study employed a laboratory experimental design using a pre-test–post-test control group method. The samples consisted of heat-cured acrylic resin blocks measuring 10 mm × 10 mm × 2 mm. Two groups were assigned: an experimental group and a control group, each comprising 16 samples, for a total of 32 samples. The experimental group was immersed in 50% star fruit (*Averrhoa bilimbi* L.) leaf extract, while the control group was immersed in sterile distilled water, each for 30 minutes.

The star fruit leaf extract was prepared using the infusion method, in which 300 g of crushed leaves were boiled in 300 mL of distilled water. The mixture was then filtered to obtain a 50% extract concentration. Prior to treatment, the initial colony count of *C. albicans* was determined. The acrylic resin samples were subsequently incubated in *C. albicans* cultures prepared in Sabouraud Dextrose Broth (SDB) for 24 hours. After the 30-minute immersion, the soaking solutions were subjected to serial dilution and cultured on Sabouraud Dextrose Agar (SDA) to assess *C. albicans* colony growth. Colony-forming units (CFUs) were recorded before

and after treatment to evaluate the antifungal effectiveness of the test solution.

Result

The study showed that the 50% star fruit leaf extract group had fewer *C. albicans* colonies than the aquadest control group. Normality testing for both the pre-test and post-test data indicated normality, with p-values of 0.131 and 0.134, respectively. Homogeneity testing also showed that both groups were homogeneous, with significance values greater than 0.05. The paired t-test showed a p-value of 0.003 ($p < 0.05$), indicating a statistically significant difference between the pre-test and post-test colony counts in the experimental group soaked in 50% star fruit leaf extract compared with the aquadest control group. These statistical results support the research hypothesis that 50% star fruit leaf extract effectively reduces *C. albicans* colony growth.

Discussion

The lowest number of *C. albicans* colonies was observed in the heat-cured acrylic resin samples immersed in 50% star fruit leaf extract. These findings are consistent with existing theories and previous studies, indicating that immersion of heat-cured acrylic resin denture plates in 50% star fruit leaf extract is effective in inhibiting *C. albicans* growth. The inhibitory effect is attributed to the presence of flavonoid compounds, which possess antimicrobial and antifungal properties and play a significant role in suppressing the proliferation of *C. albicans* colonies [4, 6].

This result aligns with the study by Sari M. (2014), which demonstrated that star fruit leaf extract inhibits *C. albicans* growth and that higher extract concentrations (20%, 40%, 60%, and 80%) produce larger inhibition zones and greater antifungal effectiveness. The antifungal activity is associated with the presence of bioactive compounds such as flavonoids, tannins, and saponins, which suppress fungal development by inhibiting pseudohyphae formation during colony growth [7].

Puspitasari (2017) reported that immersion of removable orthodontic appliances in star fruit leaf extract (*Averrhoa bilimbi* L.) at concentrations of 12.5%, 25%, 50%, and 100% demonstrated antifungal activity against *C. albicans*, with larger inhibition zones observed at higher extract concentrations. These

findings support the antifungal potential of star fruit leaf extract and are consistent with the results of the present study [8].

The findings of this study also align with Rakhmatullah et al. (2018), who compared the antifungal activity of methanolic star fruit leaf extract with 0.2% chlorhexidine. Their study showed that star fruit leaf extract at concentrations of 20%, 40%, 60%, and 80% effectively inhibited the growth of *C. albicans*, although the highest effectiveness was observed at 100%, which still did not surpass the antifungal activity of 0.2% chlorhexidine [9]. The antifungal mechanism of star fruit leaf extract is largely attributed to its flavonoid content, which functions as an antibacterial and antifungal agent. According to Rakhmatullah et al. (2018), flavonoids exert antifungal effects by disrupting fungal cell membrane permeability and damaging extracellular hydroxyl proteins, leading to toxic effects that suppress the growth of *C. albicans* [9].

The acrylic resin used in this study was heat-cured polymethyl methacrylate (PMMA). According to Annusavice and Craig (2015), heat-cured acrylic resin has several disadvantages, including its susceptibility to fracture and its porous surface, which facilitates the retention of food debris and promotes plaque accumulation. These conditions can increase the density and growth of *C. albicans* colonies when denture hygiene is inadequate, ultimately leading to denture stomatitis [2, 3, 10, 11, 12].

Hernawati (2017) states that poor denture hygiene is a major contributing factor to denture stomatitis. Daily denture cleaning traditionally involves immersion in alkaline peroxide solutions. However, several studies have shown that heat-cured acrylic resin dentures can also be disinfected using herbal-based solutions, as many herbal ingredients contain bioactive compounds with antimicrobial and antifungal activity, particularly flavonoids [13].

According to Ningsih et al. (2023), flavonoids are the largest group of natural phenolic compounds and are widely distributed in various plant parts, including roots, wood, bark, leaves, stems, fruits, and flowers. Flavonoids consist of several subclasses, one of which is flavanones. Flavanones such as naringenin and pinocembrin are commonly found in plants and exhibit notable antibacterial and antifungal bioactivity. Flavonoid compounds also contain genistein-like

structures that act as inhibitors of fungal cell division and proliferation by penetrating the fungal cell wall and reaching the cell membrane. The phenolic components within flavonoids can disrupt cytoplasmic integrity, causing leakage of intracellular contents, including the fungal cell nucleus, ultimately leading to cell damage and inhibition of *C. albicans* growth [14].

Conclusion

Based on the findings of this study, immersion of heat-cured acrylic resin in 50% star fruit leaf (*Averrhoa bilimbi*) extract demonstrated a significant effect on the reduction of *C. albicans* colonies. The number of colonies in the treatment group immersed in star fruit leaf extract was markedly lower compared with the aquadest control group. This antifungal activity is attributed to the presence of flavonoid compounds in starfruit leaf extract, which exert antibacterial and antifungal effects by disrupting fungal cell membrane permeability and damaging extracellular hydroxyl proteins. These mechanisms create toxic conditions for fungal cells, thereby suppressing the growth and proliferation of *C. albicans*.

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Table 1. Average number of *Candida albicans* colonies after 30 minutes of immersion

Group	N	Pre-Test	Post-Test
Aquades	16	1.0×10^6	$1,587 \times 10^5$
Star Fruit Leaf Extract 50%	16	1.0×10^6	$1,356 \times 10^5$

Table 2. Shapiro Wilk Test Normal Distribution Test Results

Soaking	Mr	Information
Pre Test	0,131	Normal
Post Test	0.134	Normal

Table 3. Levene Test Homogeneity Test Results

Information	Levene's test	Mr
Pre-Test	2,251	0,144
Post -Test	3,798	0,061

Table 4. Hasil Uji Paired T-Test

Variable	Average	Standard Deviation	95% Interval	Sig
Pre-Test-Post Test	225,000	389,375	84,164 to 365,385	0,003