Said, F., Rahmawati, I., & Ningsih, N. S. (2023). Revolution of Bone and Teeth Health: Study of Aloe Barbadensis Instant Powder Formulation. JURNAL INFO KESEHATAN, 21(4), 782-794. <u>https://doi.org/10.31965/infokes.Vol21Iss4.1336</u>

 Jurnal Info Kesehatan
 782

 Vol. 21, No. 4, December 2023, pp. 782-794
 Image: Control of the second secon

Revolution of Bone and Teeth Health: Study of Aloe Barbadensis Instant Powder Formulation

Fahmi Said^{1a*}, Ida Rahmawati^{1b}, Neny Setiawaty Ningsih^{2c}

- ¹Department of Dental Health, Poltekkes Kemenkes Banjarmasin, Banjarmasin, South Kalimantan, Indonesia
- ²Department of Dental Health, Poltekkes Kemenkes Pontianak, Pontianak, West Kalimantan, Indonesia
- ^a Email address: fahmialai1959@gmail.com
- ^b Email address: idarahmawati102@gmail.com
- ^c Email address: nenysetiawaty26@gmail.com

Received: 11 September 2023 Revised: 21 December 2023 Accepted: 31 December 2023

Abstract

Aloe barbadensis is a plant with many applications such as anti-inflammatory, anti-fungal, antibacterial, and aiding in cell regeneration. Understanding aloe barbadensis' phytochemical profile and pharmacological action is essential since it is believed to have an impact on the formation of teeth and bones. The stability and bioavailability of Aloe barbadensis can be improved by formulating it as an instant powder. The research's objectives are to ascertain the Aloe barbadensis instant powder's qualitative and quantitative phytochemical profile, dosage formulations, and activity testing on hemoglobin, cholesterol, and red blood cell parameters. Samples of aloe barbadensis were washed, and they were then dried for 72 hours at 50°C. Following a maceration process using a 70% ethanol solvent, the extract was dried. Phytochemical screening, TLC profile, and extract description were employed to test the extract qualitatively. The quantities of total flavonoids, total anthraquinones, and total phenolics were determined to quantitatively test the extract. The formulation of the instant powder was then completed and evaluated on female mice using metrics associated with red blood cells, hemoglobin, and cholesterol levels. Furthermore, observations were made on the mice's liver organs. The study's findings revealed a qualitative profile of Aloe barbadensis extract, which included a tasteless, unique odor, milky white hue, and liquid shape. Aloe barbadensis has been demonstrated to contain flavonoids, phenolics, tannins, saponins, and anthraquinones, according to the results of phytochemical screening. Three spots, identified as Rf 3.2 and Rf 8.5 in the Rf 2.3 area, are visible in the chromatographic pattern. Total anthraquinones were discovered to be 4.59%, total flavonoids to be 0.24%, and total phenolic content to be 1.42%. The third formula of instant Aloe barbadensis pollen has been demonstrated through preclinical examinations to have the capacity to reduce cholesterol, boost hemoglobin, and enhance red blood cell count—all of which are associated with the growth of teeth and bones. An SPSS statistical study demonstrating statistically significant differences with other groups supports this. Mice liver histopathological examinations revealed no liver damage in any of the test groups.

Keywords: Phytochemical Analysis, Oral Health, Clinical Trials.

*Corresponding Author:

Fahmi Said

Department of Dental Health, Poltekkes Kemenkes Banjarmasin, Banjarmasin, South Kalimantan, Indonesia Email: fahmialai1959@gmail.com



©The Author(s) 2023. This article is distributed under the terms of the Creative Commons Attribution 4.0 International License (<u>http://creativecommons.org/licenses/by/4.0/</u>), which permits unrestricted use, distribution, and reproduction in any medium, provided you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons license, and indicate if changes were made. The Creative Commons Public Domain Dedication waiver (http://creativecommons.org/publicdomain/zero/1.0/) applies to the data made available in this article, unless otherwise stated.

1. INTRODUCTION

In contrast to other varieties of aloe, Aloe barbadensis (also known as Aloe barbadensis L.) is a plant from a tribe that is widely used and farmed (Sánchez et al., 2020). People have been using aloe as medicine for hundreds of years, starting in ancient Egypt and continuing through Greece, Rome, China, and India (Sadoyu et al., 2021). The plant Aloe barbadensis possesses several qualities, such as anti-inflammatory, anti-fungal, anti-bacterial, and aids in cell regeneration. Aloe barbadensis has been identified in other studies to have anti-inflection properties for skin and burn wounds, as well as laxative properties for its leaves (Guo & Mei, 2016). There is also substantial evidence that aloe barbadensis may affect the growth of human teeth and bones (Kumar et al., 2019). This provides many opportunities for the use of Aloe barbadensis in the field of dentistry.

In Indonesia, aloe barbadensis is prevalent throughout the country, however, studies have indicated that the most optimal locations for discovering it are in West Kalimantan Province (Said et al., 2023). When compared to other plants from South Kalimantan, aloe barbadensis has the highest concentration of active chemicals. The plant's active components are influenced by the environment in which it grows (Adawiyah & Rizki, 2018). The temperature, humidity, rainfall, and environmental factors surrounding the growing region all have an impact on this. To ensure optimal activity, carefully selected plants should originate from growth locations that are optimum for ideal growing conditions.

Standardization is required for natural ingredients intended for use as pharmaceuticals (Sari et al., 2023). Analyzing the plant's unique characteristics can help achieve standardization (Rizki, 2020). Qualitative profiles, chromatographic profiles, phytochemical screening, and quantitative analysis of plant active component levels are examples of specific parameters (Zainab et al., 2022). This is crucial to guarantee the quality of the extract that will be examined for toxicity and pharmacological activity. The effectiveness of a natural element will depend on several factors (Sari et al., 2023). Acquiring the right amount of aloin is crucial because it is the primary ingredient that provides many of the health advantages of aloe barbadensis.

The body's ability to endure and absorb natural compounds is limited (Fitriana et al., 2022). According to Chabib et al. (2015), it is crucial to formulate natural components in dosage form to guarantee their stability and bioavailability. Furthermore, the dosage form will help patients accept the substance. An instant powder formulation is a powdered solid preparation that comprises sugar as one of its ingredients. Instant powder has many benefits, such as ease of formulation, production, and packaging (Deglas & Apriliani, 2022). In addition, users simply need to dissolve instant powder in water because it contains sugar, which provides it a delicious taste (Lubis et al., 2023). Since it can be ingested warm with hot water or cold with ice cubes, its application is more adaptable. It will be simpler for both adults and children to use Aloe barbadensis according to its quick powder format.

It is possible to assess a preparation's pharmacological activity both in vitro and in vivo (Rizki et al., 2021). Because it is conducted directly on live things, in-vivo testing is recommended (Utami et al., 2023). Test animals are available for the test for aloe barbadensis utilizing a variety of techniques and specifications (Utami et al., 2022). As a result of slowing down the body's metabolism, high cholesterol can contribute to a postponement of children's tooth and bone growth. Bone and tooth growth can be impacted by high or low cholesterol levels, as cholesterol performs an essential role in controlling bone and tooth metabolism (Yin et al., 2019). The development of teeth and bones is influenced by hemoglobin and red blood cells. There is a correlation between the prevalence of stomatitis in the teeth and the incidence of iron deficiency, which results in anemia (Mersil, 2021). Bone development is slowed by decreased red blood cell counts (Bernado et al., 2016). Determining the extract's qualitative-quantitative profile, preparation formulation, and activity test on hemoglobin, red blood cell,

and cholesterol parameters from Aloe barbadensis instant powder are the primary objectives of this study.

2. **RESEARCH METHOD**

This research is experimental research involving instant powder testing on test animals. The following tools were used in this research: a hot plate stirrer (Stuart), a measuring flask (Herma), a UV lamp with a wavelength of 254 and 366 nm, a macerator, a microscope, an object glass, an oven (Vinco), a volume pipette (Iwaki), a propipet (Vitalab), an analytical balance (Pioner), a vortex (Jeio Tech), and a water bath (SMIC).

Aloin fruit peel (Sigma-Aldrich), quercetin (Sigma-Aldrich), distilled water (CV.Viana), Cd(NO2)3 (Merck), ethanol (pa) (Sigma-Aldrich), ethanol (CV.Viana), ethyl acetate (Merck), FeCl3 (Merck), fluoroglucin (Merck), 1% gelatin, potassium hydroxide (Merck), paper Whatman strain, Dragendorff reagent (Merck), Folin-Ciocalteau reagent 5% (Sigma-Aldrich), Mayer reagent, toluene (Merck), alpha mangostin (Sigma-Aldrich), and pregnant female rats were the ingredients in this study. The research will be conducted at the Banjarmasin Pucuk Sirih Herbal Medicine Factory Laboratory which already has a certificate on Good Traditional Medicine Manufacturing Methods (CPOTB) and the Banjarbaru Industrial Research and Standardization Center (Baristan).

The first thing that has to happen when conducting research with live things is research ethics testing. The Muhammadiyah University of Banjarmasin Ethics Committee conducted the ethical evaluation using Statement of Research Ethics Eligibility No. 492/UMB/KE/VI/2023.

Preparation and Manufacture of Extracts. Aloe barbadensis is cleaned and divided into two sections with a knife, and the interior flesh is removed. It is then weighed and allowed to dry for 72 hours at 50°C in a drying cabinet (Rizki et al., 2023). Aloe barbadensis which had been dried was weighed and ground into a powder. After weighing the powder, it was combined with pro-analysis ethanol solvent and heated to 70°C for six hours. The filter paper was used to filter the mixture. Repeated extraction of the filtered residue is required to obtain a clear solution. The filtrate is gathered, dried above a water bath, and then evaporated using a solvent-rotary evaporator (Ikalinus et al., 2015).

Qualitative Phytochemical Profile. The extract's color, taste, smell, and establishment are all highlighted. After the substance under examination had been exposed to air for fifteen minutes, the extract description test was conducted using all five senses. Descriptive explanations are provided for the analysis and display of color, taste, smell, and form data (Marliana et al., 2005). Phytochemical screening incorporates testing using reagents. Tests were conducted for the presence of alkaloids, flavonoids, terpenoids, steroids, saponins, tannins, phenolics and anthraquinones (Panchal & Parvez, 2019; Rizki et al., 2021).

Chromatogram Pattern. A total of 0.1 g of the extract was weighed, and 1 mL of 96% ethanol was applied to dissolve it (Sonam et al., 2017). Spotted on GF TLC plate 254, the solution was eluted with a mobile phase-hexane: ethyl acetate at 4:6 (v/v) and 3:7 (v/v) ratios, respectively. The spots were observed using UV lamps with wavelengths of 254 and 366 nm. For each examined sample, the Rf value was computed and compared (Marliana et al., 2005).

Qualitative Content Up to Phenolic Total. Total phenolic content was ascertained by applying gallic acid standards. Gallic acid standard solution and sample were combined in 0.5 mL with 2.5 mL of 5% Folin Ciocalteau reagent, shaken until smooth, and allowed to sit for 8 minutes. Afterwards, 2 mL of NaOH solution was added, vortexed until smooth, and the mixture was allowed to sit for 30 minutes. A UV-Vis spectrophotometer operating at a wavelength of 742 nm was utilized to measure the absorbance (Rizki et al., 2022).

Determination of total flavonoid levels utilizing quercetin standard. A total of 0.5 mL of solution was added with 1.5 mL of ethanol p.a., 0.1 mL of AlCl₃ 10%, 0.1 mL of 5% acetic acid, and 2.8 mL of distilled water in a test tube. Leave it for 20 minutes, absorbance is read employing UV-Vis spectrophotometry at a maximum wavelength of 418 nm (Anwar et al., 2017).

Aloin standards are applied to determine the levels of total anthraquinone. A 5 ml measuring flask was filled with 4 ml of solution, 100 μ L KOH 5% was added, and then methanol was added gradually until the mark was reached. The mixture was shaken until it was homogenous, and allowed to stand within the operating time range, and the absorbance was determined using UV-Vis spectrophotometry at the maximum wavelength (Mutiara et al., 2007).

The process of formulation is performed by trial and error. The recipe calls for popular instant powder components, such as lychee flavoring, distilled water, and granulated sugar. For pharmacological testing, a formula that can produce immediate powder will be utilized (Lubis et al., 2023).

Pharmacological Test. White pregnant mice (the rat was born) and twenty Wistar females were utilized as test subjects. The female test subject weighed between 250 and 300 grams. Mice were acclimated (adjusted to their surroundings) for seven days. The test animals were placed in the following treatment groups:

- a. Normal Control Group (KN), that is a group of test animals that were given standard food and drink for 14 days.
- b. Normal Treatment Control Group (KNP0), that is a group of test animals that were given standard food and drink for 14 days.
- c. Treatment Group 1 (KP1), that is a group of test animals that were given standard food and drink, as well as a dose of instant powder 500 Mg/KgBW/Day on days 13-19.
- d. Treatment Group 2 (KP2), that is the group of test animals that were given standard food and drink, as well as a dose of instant powder 1000 Mg/KgBW/Day on days 13-19.
- e. Treatment Group 3 (KP2), that is the group of test animals that were given standard food and drink, as well as a dose of instant powder 2000 Mg/KgBW/Day on days 13-19 (Wijayatri, 2017).

Before initiating the medication, the mice's blood was extracted and their total blood hematological was examined. To assess the effect of the treatment, a complete blood hematological examination was carried out after it was finished. Next, following the ethical treatment of animals, the mice were put to death. Mice were operated upon, and their liver organs were examined under a microscope to determine the toxicity of the instant powder (Yolanda et al., 2022).

The investigation produced quantitative and qualitative data on extract samples that were descriptively examined as the research's output. Hematology tests were utilized in preclinical testing to compare pre-and post-test findings, and rat organs and fetuses were observed visually.

3. RESULTS AND DISCUSSION

The extract's qualitative characteristics include a tasteless, unique odor, milky white hue, and liquid shape. The Aloe barbadensis meat that has been suspended in distilled water is what provides it its signature milky white hue. The reason the extract has no taste is because Aloe barbadensis does not normally have any taste. The smell that was detected is the characteristic scent of Aloe barbadensis; it is similar to the scent of grass and is unique to Aloe barbadensis alone. The reason for the liquid extract form is that distilled water is added to Aloe barbadensis to increase its dominance, resulting in a liquid extract (Quaye et al., 2023).

Phytochemical Screening Profile. The extract obtained was then identified for the presence of secondary metabolite compounds. This presence was measured utilizing phytochemical screening as presented in the table below.

No	Phytochemicals	Reactor	Results
1.	Flavonoid	Magnesium and Hydrochloric Acid	+
2.	Phenolic	Iron Chloride	+
3.	Alkaloid	Reagan Meyer	-
4.	Tannin	Gelatin Solution	+
5.	Saponin	Aquades	+
6.	Steroid	Lieberman Burchard	-
7.	Terpenoids	Lieberman Burchard	-
8.	Anthraquinone	KOH 10%	+

 Table 1. Phytochemical Screening.

Chemicals are employed in tubes for phytochemical screening. As a result, the tube method or reagent method are other names for the phytochemical screening procedure. Aloe barbadensis is believed to contain flavonoids, phenolics, tannins, saponins, and anthraquinones, according to the results of phytochemical screening. Steroids, terpenoids, and alkaloids are absent from aloe barbadensis. These findings are consistent with phytochemical screening results from prior studies (Guo & Mei, 2016), which discovered that Aloe barbadensis includes flavonoids, anthraquinones, saponins, and tannins. Since Aloe barbadensis is known to be high in water, it is unlikely to contain terpenoid or steroid chemicals, which tend to be non-polar or insoluble in water. In Aloe barbadensis, alkaloids are not recognized, despite the reality that they can be identified in some fruit flesh (Ngibad, 2019). Alkaloids are not found in several fruit flesh types. Since every plant is unique, it is possible that a group will not be present in a certain species of plant (Said et al., 2023).

The growth of teeth and bones will be supported with aloe barbadensis. Pathogens can cause problems in the development of teeth and bones. Because bacteria in teeth can harm tooth components, it's critical to employ natural compounds with antibacterial properties, such as aloe barbadensis, which contains flavonoids, phenolics, and tannins. Natural components have also been demonstrated to be safe on a large scale, therefore using them in the mouth is not extremely dangerous. It has also been demonstrated that these secondary metabolites lessen the development of oral illnesses and tooth plaque (Kumar et al., 2021).

Chromatographic Pattern. The objective of chromatography pattern testing is to ascertain the extract's separation pattern on the silica plate. One of the extract's identities—along with a component of extract standardization—will be this pattern of separation. A feature of an extract can be its chromatographic pattern, which can serve as an identifying marker. The table below displays the chromatographic pattern's results.

Condition	Chromatography Results	Nilai Rf	
Mobile phase	00	$Rf_1 = 2,3$	
n-hexane: ethyl acetate (7:3)		$Rf_2 = 3,2$	
-		$Rf_1 = 8,5$	
Silent phase silica gel 60 F254			

Table 2. Chromatographic Pattern.

| 786

The results of the extract's separation or elution on a chromatography plate or silica plate employing a certain mobile phase (solution) generate the chromatographic pattern. Aloe barbadensis extract is chromatographed using a stationary phase comprising a silica plate that fluoresces at a wavelength of 254 and a mobile phase of n-hexane and ethyl acetate at a composition of 7 to 3. Three spots appear on the plate once the extract has been spotted there and the mobile phase has been eluted. The three sets of spots represent the division of active chemicals. These patches show up in the regions of Rf 2.3, Rf 3.2, and Rf 8.5. This is the identification of the Aloe barbadensis extract, which has three spots with spot positions Rf 2.3, Rf 3.2, and Rf 8.5 in the silica gel stationary phase and the mobile phase above. If you ever want to verify the accuracy of the Aloe barbadensis extract, you can utilize this data as identity data (Rizki, 2020).

It is evident from the spot results that certain compound groups are highly patterned (2 bottom spots) and non-polar (1 top spot). According to Marliana et al. (2005), the results demonstrate that Aloe barbadensis still contains some somewhat nonpolar compounds. These compounds are most likely derived from flavonoids, some of which are less polar. On the other hand, the majority of the spots at the bottom are indicative of the presence of groups of compounds that tend to be polar, such as phenolics, saponins, tannins, and anthraquinones.

As part of the quantitative analysis, the amounts of Aloe barbadensis extract are ascertained. One of an extract's quality parameters includes this analysis. Quercetin is employed to determine total flavonoid content, aloin is used to estimate anthraquinone content, and gallic acid standards are utilized to determine total phenolic content. Table III displays the assay's findings.

No	Phytochemicals	Standard	Replication	Much	Percent Content
1.	Phenolic	Gallic Acid	1	14,09 µg/mg	1,40 %
			2	14,58 µg/mg	1,45 %
			3	14,19 µg/mg	1,41 %
	Rate			14,29 µg/mg	1,42 %
2.	Flavonoid	Quercetin	1	2,45 µg/mg	0,24 %
			2	2,59 µg/mg	0,25 %
			3	2,33 µg/mg	0,23 %
	Rate			2,46 µg/mg	0,24 %
	Anthraquinone	I started	1	46,10 µg/mg	4,61 %
			2	45,78 µg/mg	4,57 %
			3	46,10 µg/mg	4,61 %
	Rate			45,99 µg/mg	4,59 %

Table 3. Extract Active Ingredients Rate.

As per the Indonesian Herbal Pharmacopoeia, level determination is an integral component in standardizing extracts of natural ingredients. The total phenolic content analysis revealed that the extract had an average total phenolic content of 1.42% of its total weight. Typically, the extract's total flavonoid content is less than 0.24% of its total weight. It is known that the anthraquinone content averages 4.59%. These findings demonstrate that anthraquinones predominate in Aloe barbadensis flesh. This is consistent with other studies showing that the anthraquinone group is the dominating group in Aloe barbadensis. Phytochemical screening also revealed the presence and amounts of anthraquinones, which is consistent with the results. The anthraquinone levels found were somewhat higher than the 4.48% anthraquinone levels observed in prior investigations including Aloe barbadensis (Mutiara et al., 2007). This indicates that the findings in this study and those from related studies are not all that different.

| 788

Phenolics are identified to be greater than the flavonoid group. This is because the phenolic group of chemicals is present in an extensive range of plants. Flavonoid and tannin chemicals are likewise included in total phenolics, and they similarly build up. Therefore, the phenolic group will often have a higher content than the flavonoid group. Although not all phenolics will be discovered in the flavonoid group, the flavonoid group is a component of the phenolic group. The amount of Aloe barbadensis utilized will be revealed by the data gathered for this study. When materials from different locations are used, one of the quality parameters will be the amount of active chemicals. To preserve the extract's effectiveness, the concentration of each element used—especially when using it in multiple locations—should ideally be ascertained. Low levels will affect the low efficacy that follows (Rizki, 2020).

The pharmacological activity of the sample will be correlated with high levels of total flavonoids and total phenolics. Aloe barbadensis is used in conjunction with phenolics and flavonoids to suppress the formation of bacteria in the mouth and prevent plaque on teeth. Aloe barbadensis can be used to avoid dental issues and promote healthy tooth growth. Elevated phenolic and flavonoid content will boost metabolic functions, speeding up the process of bone formation. The primary chemical in aloe barbadensis that produces the action is anthraquinone. According to Guo and Mei (2016), anthraquinones have anti-inflammatory, antibacterial, and anti-caries effects. Its anti-inflammatory properties will shield teeth wounds from discomfort and hasten their healing. This ability will support aloe barbadensis in its use as a supplement to support tooth and bone growth (Quaye et al., 2023).

Preparation of Formulation. The extracted aloe barbadensis is then combined with other ingredients to create a dosage form. The active components' stability will be preserved and their use will be made easier by the dosage formulation. To facilitate the acceptance or consumption of the chemicals employed, formulation is crucial. The table below displays the dose formula that was derived from the optimization results.

No	Material	Function	Material Weight	
1.	Aloe barbadensis flesh	Active Ingredients	100 grams	
2.	Granulated sugar	Filling Material	500 grams	
3.	Aquades	Solvent	1000 mL	
4.	Lychee Flavoring	Seasoning	1 mL	

Table 4. Preparation of Formulation

Direct consumption of aloe barbadensis tends to be less acceptable because of its strong odor. It is hard to eat because the fragrance is similar to that of grass. Patients have to discover the dose form to be convenient to use and agreeable. The preparation that was selected is a powder that includes lychee flavoring to mask the off-putting taste and smell of aloe barbadensis, granulated sugar as a filler and sweetener, distilled water as a solvent, and aloe barbadensis as the active ingredient (Deglas & Apriliani, 2022). Through trial and error and compositional tinkering, the formulation that was arrived at was achieved. Following several tests, the mixture indicated in the formula table proved to be the ideal formula.

The powder formulation of aloe barbadensis will make it easier to use. Aloe barbadensis has a disagreeable smell, and when placed in the open it becomes less stable; this formulation will help to keep it fresher and simpler to use. During storage, the powder dose form exhibits greater stability. To use it, simply add water and consume. Aloe barbadensis is easily absorbed by the body because it enters the digestive tract in solution form. Rapid and simple absorption will expedite the intended outcome. Aloe barbadensis powder is a supplement that promotes the growth of teeth and bones. Children who are in the stage of tooth and bone growth will be more receptive to this preparation if it is easy to use. The calcium and magnesium contained in Aloe barbadensis tend to dissolve in water, making it easier to formulate preparations (Fiorentini et al., 2021).

Test animal preparation is the first step in preclinical testing. The test subjects were Wistar strain pregnant female white rats. For seven days, the test animals were acclimated to their cages. Blood hematology, including hemoglobin, cholesterol, and red blood cell counts, are measured prior to testing. It is well-recognized that high cholesterol exacerbates bone disorders. An excessive amount of cholesterol raises the risk of malnutrition (Yin et al., 2019). Anemia is associated with quantities of red blood cells and hemoglobin. The spine produces red blood cells, which are correlated with the spine's health (Quaye et al., 2023). Figure 1 below illustrates the duration of the test animals' treatment and the procedure used for collecting blood from them.



Figure 1. Treatment and Blood Collection

An oral probe was employed to provide therapy to test animals in both the three-dose treatment group and the normal control group. Giving takes place up until the fourteenth day. After the course of treatment, another blood sample was obtained. The mouse orbital sinus in its eye is utilized for blood collection, which has the benefit of allowing for the extraction of 1-2 mL of blood, which facilitates hematological analysis (Sharma et al., 2014). The table below displays the research findings as data from hematological test results.

The study of mouse blood samples using parameters related to cholesterol levels was performed both before and after the treatment. Before the study, the values of cholesterol in all groups were quite similar. The cholesterol levels of each test animal were normal. Furthermore, treatment 1, treatment 2, and treatment 3 groups got Aloe barbadensis at varying doses, whereas the positive control group mice received simvastatin as a cholesterol reducer. Hematological test results collected after the fourteenth day revealed that the normal group's cholesterol levels increased as a result of the mice's constant feeding while they received no medical attention. The delivery of simvastatin, which can lower cholesterol levels, caused the levels of cholesterol in the positive control group to decrease.

No	Group	Replication	Up to Cholesterol		Up to Hemoglobin		Red Blood Cell Rate (Million/mm3)	
			Before	After	Before	After	Before	After
1	Normal	1	45	53	12,3	12,4	8,1	8,2
	Control	2	46	54	13,4	12,8	7,9	7,5
	Group	3	46	56	12,8	13,1	8,3	7,8
	(KN)	4	45	58	12,9	12,6	7,4	8,1
		Rate	45,5	55,25	12,8	12.7	7,9	7,9

Table 5. Rat Cholesterol Test Results

Said, F., Rahmawati, I., & Ningsih, N. S. (2023). Revolution of Bone and Teeth Health: Study of Aloe Barbadensis Instant Powder Formulation. JURNAL INFO KESEHATAN, 21(4), 782-794. <u>https://doi.org/10.31965/infokes.Vol2</u>1lss4.1336

790

								1790
2	Positive	1	44	38	12,4	12,3	8,2	7,4
	Control	2	45	36	12,5	12,5	8,1	8,4
	Group	3	45	38	13,4	13,1	7,6	8,3
	(KKP)	4	48	37	13,1	12,8	8,4	8,1
		Rate	45,5	37,25	12,8	12,6	8,0	8,0
3	Treatment	1	44	47	13,4	12,4	7,6	8,1
	Group 1	2	48	47	12,3	13,1	7,9	8,1
	(KP1)	3	46	48	12,5	12,5	8,4	8,3
	_	4	47	49	13,1	13,2	8,6	7,9
		Rate	46,25	47,75	12,8	12,8	8,4	8,3
4	Treatment	1	44	43	13,4	13,1	7,9	8,1
	Group 2	2	46	43	13,1	13,4	7,8	8,2
	(KP2) [–]	3	46	42	12,7	13,4	8,4	8,7
	-	4	47	41	12,9	13,5	8,1	8,8
		Rate	45,75	42,25	13,0	13,3	8,1	8,4
5	Treatment	1	47	38	12,4	15,6	8,1	10,2
	Group 3	2	45	36	13,2	15,1	7,9	9,5
	(KP2)	3	47	38	13,1	15,2	8,1	9,4
	-	4	47	37	12,6	14,9	7,6	9,8
		Rate	46,5	37,25	12,8	15,2	7,9	9,7

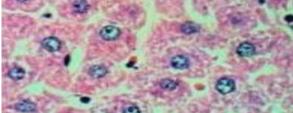
The results of statistical analysis using SPSS demonstrated that there were significant differences between treatment groups 1, 2, and 3 and the normal control group. This demonstrates how administering instant powder affects variations in cholesterol levels. When comparing the positive control group to treatment 3, statistical analysis using SPSS revealed no significant changes. This indicates that treatment 3 can perform on par with the positive control. Children's teeth and bone development are believed to be slowed down in those with high cholesterol. Elevated cholesterol levels cause the body's metabolism to slow down, which hinders the growth of teeth and bones. Low or high cholesterol can have an impact on tooth and bone formation because cholesterol regulates the metabolism of bones and teeth (Yin et al., 2019). Children will have stunted bone and dental growth if they receive inadequate nutrition, particularly in the areas of calcium and magnesium. The eruption status of the mandibular permanent central incisors correlates significantly with poor nutrition (Rahmawati et al., 2014). Previous studies have indicated that aloe barbadensis contains calcium and magnesium, both of which are essential for the development of teeth and bones. Teeth are critical to the body since they are primarily composed of calcium and magnesium (Said et al., 2023).

Hemoglobin level parameters from mice blood analysis were performed both before and after treatment. Before the trial, there were not many differences in the hemoglobin levels among all groups. Before testing, the hemoglobin levels of each test animal were normal. Furthermore, because the same mice were utilized for all treatments, the mice in the positive control group received simvastatin, but the mice in the treatment 1, treatment 2, and treatment 3 groups received varying amounts of aloe barbadensis for each treatment. Hematology test results received after the fourteenth day indicated that the normal group's hemoglobin levels had not significantly changed from the previous day. Simvastatin had no influence on hemoglobin levels, which explains why hemoglobin levels in the positive control group likewise remained unchanged.

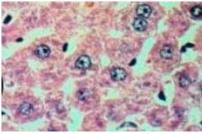
In treatment groups 1 and 2 there were no significant differences between before and after treatment. It was discovered that the mice in treatment group 3 had higher hemoglobin levels. These findings suggest that giving mice three doses of Aloe barbadensis for a period of 14 days

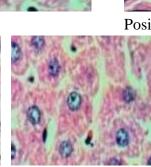
can raise their hemoglobin levels. There were noteworthy distinctions between treatment group 3 and the other treatment groups, according to the findings of the statistical analysis conducted with SPSS. This demonstrates how fast powder may raise mice's hemoglobin levels. The incidence of anemia, which is correlated with the growth of teeth and bones, is related to hemoglobin. Patients with low hemoglobin levels have poor bone and tooth growth potential. This is also correlated with the rate of wound healing in patients with low hemoglobin levels following tooth extraction. Hemoglobin also affects bone mineral density. Low hemoglobin levels can cause low bone density. Low hemoglobin levels can also escalate the risk of bone fractures (Yoon et al., 2016).

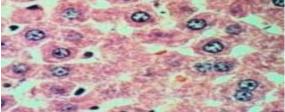
Red blood cell parameters from mouse blood analysis were measured both before and after therapy. The objective is to ascertain how the medication affects the levels of red blood cells in rats. Before the trial, the red blood cell counts in each group were essentially the same. Prior to testing, the red blood cell counts of all test animals were normal. Furthermore, because the same mice were utilized for all treatments, the mice in the positive control group received simvastatin, but the mice in treatment 1, treatment 2, and treatment 3 groups received varying amounts of aloe barbadensis for each treatment. Hematological test results received after the fourteenth day indicated that the normal group's red blood cell levels had not significantly changed from the initial values. In the positive control group, red blood cell levels also did not change, this is because simvastatin did not have any effect on red blood cell levels. There were no discernible differences between the pre- and post-treatment periods in treatment groups 1 or 2. There was a rise in red blood cell counts in treatment group 3. The statistical analysis conducted using SPSS revealed significant differences between all groups and treatment 3. This demonstrates that treatment 3 can raise the number of red blood cells in mice. These findings suggest that mice's red blood cells were able to proliferate by receiving a third dosage of Aloe barbadensis. Bone development and red blood cells are closely connected processes. Because hematopoiesis and bone homeostasis are interconnected, low red blood cell counts may be a sign of bone health issues.



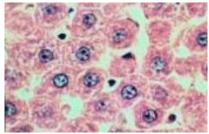
Control Normal







Positive Control



Treatment 1Treatment 2Treatment 3Figure 2. Rat Liver Histopathology Results in the Research Group

The results of histopathology tests conducted on mice are displayed in the figure above. One mouse per research group was employed for histopathological assessment. The study had five groups in total. The phases of euthanizing and necropsying the mice, followed by surgery, liver removal, sectioning the organ, paraffin block creation, hematoxylin-eosin staining, and 40x magnification examination under a microscope are all included in the results of the histological testing. There were no histological changes among the five rat livers observed by the five study groups, according to the findings of their observations. Mice's livers exhibited neither necrosis nor inflammatory development. Additionally unharmed and without granule formation are the livers of rats (Yolanda et al., 2022). These results illustrate that in the five test groups, no toxicity occurred in the rat liver. This confirmed that the Aloe barbadensis administered had no toxic effects at the first, second and third doses. Aloe barbadensis is safe to consume within reasonable limits as needed for 14 days.

4. CONCLUSION

Aloe barbadensis includes flavonoids, phenolics, tannins, saponins, and anthraquinones, according to the results of the qualitative profile analysis. Total anthraquinones were present in amounts of 4.59%, total flavonoids in 0.24%, and total phenolics in 1.42% (quantitative). A combination of 100 g of Aloe barbadensis flesh, 500 g of granulated sugar, 1 mL of lychee flavoring, and 1000 mL of distilled water provides the best results for immediate pollen production. Mice liver histopathological examinations revealed no harm in any of the test groups. The third formula of instant Aloe barbadensis pollen has been demonstrated through preclinical tests to have the capacity to reduce cholesterol, enhance hemoglobin, and improve red blood cell count—all of which are associated with the growth of teeth and bones. Further research can focus on isolating and characterizing specific bioactive compounds from Aloe barbadensis, particularly those identified in the qualitative phytochemical profile. This can provide a more in-depth understanding of the individual components responsible for the observed pharmacological effects.

REFERENCES

- Adawiyah, R., & Rizki, M. I. (2018). Aktivitas Antioksidan Ekstrak Etanol Akar Kalakai (Stenochlaena palustris Bedd) Asal Kalimantan Tengah. *Jurnal Pharmascience*, *5*(1), 71-77. https://doi.org/10.20527/jps.v5i1.5788
- Anwar, K., Beny, R., Triyasmono, L., Rizki, M. I., Halwany, W., & Lestari, F. (2017). The Influence of Leaf Age on Total Phenolic, Flavonoids, and Free Radical Scavenging Capacity of Aquilaria beccariana. *Research Journal of Pharmaceutical Biological and Chemical Sciences*, 8(1S), 129–133.
- Bernado, P., Rahardjo, R., & Rahajoe, P. S. (2016). Pengaruh Recombinant Human Erythropoietin Terhadap Jumlah Osteoblas, Osteoklas Dan Kadar Hemoglobin Pada Penyembuhan Fraktur Tulang Mandibula. *Jurnal Kedokteran Gigi*, 7(2), 131–137.
- Chabib, L., Indrati, O., & Rizki, M. I. (2015). Formulasi Tablet Effervescent Ekstrak Lidah Buaya (Aloe vera). *Jurnal Pharmascience*, 2(1), 72–80.
- Deglas, W., & Apriliani, F. (2022). Pembuatan Minuman Serbuk Instan Lidah Buaya dengan Penambahan Kacang Hijau. *BIOFOODTECH: Journal of Bioenergy and Food Technology*, 1(1), 1–8. https://doi.org/10.55180/biofoodtech.v1i1.233
- Fiorentini, D., Cappadone, C., Farruggia, G., & Prata, C. (2021). Magnesium: Biochemistry, Nutrition, Detection, and Social Impact of Diseases Linked to Its Deficiency. *Nutrients*, 13(4), 1136.
- Fitriana, M., Halwany, W., Kartika, Y., Anwar, K., Siswadi, S., Rizki, M. I., Rahmanto, B., & Andriani, S. (2022). Formulation and physical stability of syrup containing gaharu (Aquilaria microcarpa Baill.) leaves extract. *Jurnal Riset Industri Hasil Hutan*, 14(1), 33. https://doi.org/10.24111/jrihh.v14i1.7647

- Guo, X., & Mei, N. (2016). Aloe Vera: A Review Of Toxicity And Adverse Clinical Effects. Journal of Environmental Science and Health, Part C, 34(2), 77–96. https://doi.org/10.1080/10590501.2016.1166826
- Ikalinus, R., Widyastuti, S. K., & Setiasih, N. L. E. (2015). Skrining Fitokimia Ekstrak Etanol Kulit Batang Kelor (Moringa oleifera). *Indonesia Medicus Veterinus*, 4(1), 71–79.
- Kumar, M., Prakash, S., Radha, Kumari, N., Pundir, A., Punia, S., Saurabh, V., Choudhary, P., Changan, S., Dhumal, S., Pradhan, P. C., Alajil, O., Singh, S., Sharma, N., Ilakiya, T., Singh, S., & Mekhemar, M. (2021). Beneficial Role of Antioxidant Secondary Metabolites from Medicinal Plants in Maintaining Oral Health. *Antioxidants*, 10(7), 1061. https://doi.org/10.3390/antiox10071061
- Kumar, R., Singh, A. K., Gupta, A., Bishayee, A., & Pandey, A. K. (2019). Therapeutic Potential of Aloe vera—A Miracle Gift of Nature. *Phytomedicine*, 60, 152996. https://doi.org/10.1016/j.phymed.2019.152996
- Lubis, R. T., Lubis, M. S., Dalimunthe, G. I., & Yuniarti, R. (2023). Formulasi Sediaan Minuman Serbuk Jeli Lidah Buaya (Aloe vera (L.) Burm.f.). FARMASAINKES: Jurnal Farmasi, Sains, dan Kesehatan, 2(2), 178–188.
- Marliana, S. D., Suryanti, V., & Suyono, S. (2005). The Phytochemical Screenings and Thin Layer Chromatography Analysis of Chemical Compounds in Ethanol Extract of Labu Siam Fruit (Sechium edule Jacq. Swartz.). *Biofarmasi Journal of Natural Product Biochemistry*, 3(1), 26–31.
- Mersil, S. (2021). Stomatitis sebagai Manifestasi Oral dari Anemia Defisiensi Zat Besi disertai Trombositosis. *e-GiGi*, 9(2), 181. https://doi.org/10.35790/eg.v9i2.34481
- Mutiara, M., Verdia, E., & Pramono, S. (2007). Pengaruh Komposisi Pelarut Pada Pembuatan Ekstrak Daun Lidah Buaya (Aloe vera L.) Dengan Parameter Kadar Antrakinon Total Dihitung Sebagai Aloin. *Tesis*. Universitas Gadjah Mada.
- Ngibad, K. (2019). Phytochemical Screening of Sunflower Leaf (Helianthus annuus) and Anting-Anting (Acalypha indica Linn) Plant Ethanol Extract. *Borneo Journal of Pharmacy*, 2(1), 24–30.
- Panchal, P., & Parvez, N. (2019). Phytochemical Analysis Of Medicinal Herb (Ocimum sanctum). *International Journal of Nanomaterials, Nanotechnology and Nanomedicine*, 5(2), 008–011.
- Quaye, B., Opoku, O., Benante, V., Adjei-Mensah, B., Amankrah, M. A., Ampadu, B., Awenkanab, E., & Atuahene, C. C. (2023). Nfluence Of Aloe Vera (Aloe barbadensis M.) As An Alternative To Antibiotics On The Growth Performance, Carcass Characteristics And Haemato-Biochemical Indices Of Broiler Chickens. *Veterinary Medicine and Science*, 9(3), 1234–1240. https://doi.org/10.1002/vms3.1099
- Rahmawati, A. D., Retriasih, H., & Medawati, A. (2014). Hubungan antara Status Gizi dengan Status Erupsi Gigi Insisivus Sentralis Permanen Mandibula. *IDJ*, *3*(1), 16–21.
- Rizki, M. I. (2020). Farmakognosi dan Metabolit Sekunder. CV. IRDH.
- Rizki, M. I., Nurlely, N., Fadlilaturrahmah, F., & Ma'shumah, M. (2021). Skrining Fitokimia Dan Penetapan Kadar Fenol Total Pada Ekstrak Daun Nangka (Artocarpus heterophyllus), Cempedak (Artocarpus integer), dan TARAP (Artocarpus odoratissimus) Asal Desa Pengaron Kabupaten Banjar. Jurnal Insan Farmasi Indonesia, 4(1), 95-102. https://doi.org/10.36387/jifi.v4i1.667
- Rizki, M. I., Sari, A. K., Kartika, D., Khairunnisa, A., & Normaidah. (2022). Penetapan Kadar Fenolik Total dan Uji Aktivitas Antioksidan Fraksi dari Ekstrak Etanol Daun Cempedak (Artocarpus integer) dengan Metode DPPH. *MPI (Media Pharmaceutica Indonesiana)*, 4(2), 168–178. https://doi.org/10.24123/mpi.v4i2.4937

- 794
- Rizki, M. I., Triyasmono, L., & Rizky, R. A. (2023). Antioxidant Activity And Total Flavonoid Content Kalangkala Leaf Extract (Litsea angulata). *Journal of Current Pharmaceutical Science*, 7(1), 696–701.
- Sadoyu, S., Rungruang, C., Wattanavijitkul, T., Sawangjit, R., Thakkinstian, A., & Chaiyakunapruk, N. (2021). Aloe Vera and Health Outcomes: An Umbrella Review of Systematic Reviews and Meta-Analyses. *Phytotherapy Research*, 35(2), 555–576. https://doi.org/10.1002/ptr.6833
- Said, F., Rahmawati, I., & Ningsih, N. S. (2023). Clinical Study and Toxicity Tests of Disclosing Agent Aloe Vera Gel. Asian Journal of Pharmaceutical Research and Development, 11(2), 15–19.
- Sánchez, M., González-Burgos, E., Iglesias, I., & Gómez-Serranillos, M. P. (2020). Pharmacological Update Properties of Aloe Vera and its Major Active Constituents. *Molecules*, 25(6), 1324. https://doi.org/10.3390/molecules25061324
- Sari, A. K., Rizki, M. I., Akbar, N. H., Amaliya, M., Fadlilaturrahmah, & Normaidah. (2023). Determination of Specific and Non-Specific Parameters of Cempedak Leaf Simplisia (Artocarpus Integer) . *International Journal of Pharmaceutical and Bio Medical Science*, 3(09), 454–459. https://doi.org/10.47191/ijpbms/v3-i9-03
- Sharma, A., Fish, B. L., Moulder, J. E., Medhora, M., Baker, J. E., Mader, M., & Cohen, E. P. (2014). Safety and Blood Sample Volume And Quality of A Refined Retro-Orbital Bleeding Technique In Rats Using A Lateral Approach. *Lab Animal*, 43(2), 63–66. https://doi.org/10.1038/laban.432
- Sonam, M., Singh, R. P., & Saklani, P. (2017). Phytochemical Screening and TLC Profiling of Various Extracts of Reinwardtia indica. *International Journal of Pharmacognosy and Phytochemical Research*, 9(4), 523–527.
- Utami, N. K., Amperawati, M., & Rizki, M. I. (2022). Uji In Vivo Terhadap Ekstrak Kayu Secang (Caesalpinia sappan L/Biancaea sappan) Sebagai Disclosing Agent. *An-Nadaa Jurnal Kesehatan Masyarakat*, 9(2), 203. http://dx.doi.org/10.31602/ann.v9i2.9031
- Utami, N. K., Amperawati, M., & Rizki, M. I. (2023). Stability Test And Clinical Trial Of Secang Wood Extract Gel (Biancaea sappan) As Disclosing Agent. *AMJ*, *63*(01), 8609–8618.
- Wijayatri, R. (2017). Pengaruh Fraksi Etil Asetat Ekstrak Etanol 96% Akar Pasak Bumi (Eurycoma longifolia Jack) Terhadap Gambaran Histopatologis Hepar Tikus Betina Sprague dawley yang Diberi Karsinogen 7,12-Dimetilbenz(A) Antrasen (DMBA). Indonesian Pharmacy and Natural Medicine Journal, 1(1), 34–47.
- Yin, W., Li, Z., & Zhang, W. (2019). Modulation of Bone and Marrow Niche by Cholesterol. *Nutrients*, 11(6), 1394. https://doi.org/10.3390/nu11061394
- Yolanda, S., Masyitha, D., & Sayuti, A. (2022). Gambaran Histopatologi Hati Tikus Putih (Rattus norvegicus) Setelah Pemasangan Implan Wire Material Logam. *Jurnal Ilmiah Mahasiswa Veteriner*, 6(4), 234–242.
- Yoon, J., Kim, D.-J., Sung, H.-H., & Jo, Y.-K. (2016). Analysis of Bone Mineral Density according to Hemoglobin in University Students. *The Korean Journal of Clinical Laboratory Science*, 48(4), 296–303. https://doi.org/10.15324/kjcls.2016.48.4.296
- Zainab, Z., Nurlailah, N., & Rizki, M. I. (2022). Identification of Active Compound and Antibacterial Activity Against Gram-Positive and Gram-Negative Bacteria of Chromolaena odorata Leaf Extract. *Research Journal of Pharmacy and Technology*, 15(10), 4720–4726. https://doi.org/10.52711/0974-360X.2022.00793