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Evaluation of Aloe Vera (*Aloe barbadensis*) Instant Powder on Bone and Tooth Development in Pregnant Women

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Abstract: This study aimed to evaluate the physical, chemical, biological, stability, and clinical effects of aloe vera (*Aloe barbadensis*) instant powder on pregnant women. The research involved formulating the powder, assessing its physical properties (flow time, dwell angle, compressibility index), and conducting chemical analysis (phytochemical screening). Microbial contamination was tested using the Most Probable Number and Total Plate Count methods, and stability was assessed over two months. Clinical trials were conducted with two groups: one receiving the aloe vera instant powder and the other receiving calcium supplements (positive control), each consisting of 5 pregnant women for 10 days. The results showed that the powder had a flow time of 8.63 seconds, a dwell angle of 34.33°, and a compressibility index of 7.70%. Chemical screening revealed the presence of flavonoids, phenolics, tannins, saponins, steroids, and interquinones. No microbial contamination was detected, and the powder remained stable. Clinical trials showed a significant increase in red and white blood cells, hemoglobin, potassium, calcium, phosphorus, and magnesium levels in the group receiving the aloe vera instant powder. In conclusion, aloe vera instant powder is stable and enhances hematological parameters and mineral levels in pregnant women.

Keywords: Aloe vera; Bone growth; Calcium; Hematological parameters; Instant powder; Microbial contamination; Phosphorus; Pregnant women; Stability; Tooth development

Introduction

Stunting is a growth and development disorder due to chronic malnutrition and recurrent infections, characterized by below-standard length or height. According to the Indonesian Ministry of Health, toddlers can be stunted if their length or height has been measured and then compared to the standard, and the results of this measurement are below average. So, it can only be estimated or guessed using measurement. In addition to a body short for children of the same age, there are other characteristics. Growth slows; the face looks younger than the child's age, and tooth growth is delayed (Laksono et al., 2022; Rahayu et al., 2018; Titaley et al., 2019).

Stunting data for South Kalimantan Province in 2021 is a percentage of 10.64 percent above the national average of 8.8 percent. Data for August 2022 percent (the number of stunted toddlers was 21,279 out of the target number of 215,230 toddlers). In September 2022, it decreased by 10% based on the Indonesian Toddler Nutrition Status Survey (SSGBI). Still, it exceeded the national figure of 24.4% while South Kalimantan Province was 30%; the Deputy Governor of South

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Kalimantan conveyed this at the Coordination meeting of the Provincial, District, and City Stunting Reduction Acceleration Team (TPPS) (Triawanti et al., 2022).

Aloe vera is a commonly found plant because it is easy to plant and cultivate. Aloe vera contains minerals such as calcium, iron, zinc, and vitamins C and E. It also includes secondary metabolites such as phenolics, flavonoids, and anthraquinones, responsible for the plant's health benefits. Aloe vera also contains secondary metabolites such as phenols, flavonoids, and anthraquinones, which are accountable for efficacy. Aloe vera can also be used as an anti-inflammatory, wound healer, antibacterial, and anticancer. Aloe vera has a unique texture because it is gel-shaped and has been proven safe to use in the mouth because there are aloe vera preparations for mouth ulcers or wounds on the market. The plant has antimicrobial activity and can support bone and tooth growth (Anarkhis et al., 2023; Melliawati, 2018; Salehi et al., 2018).

Direct consumption of aloe vera needs to be improved. Aloe vera tends to be tasteless, making it less desirable when consumed directly. Fresh aloe vera generally cannot last long, so it is easily rotten, and the active ingredients become quickly damaged. It is necessary to make innovations in aloe vera dosage forms to overcome some of the above. One innovation that can be done is to formulate aloe vera juice into an instant powder dosage form. Instant powder preparations allow aloe vera juice to be more durable, easy to use, and acceptable to patients. Aloe vera juice instant powder only needs to add water, so it can be consumed using warm or cold water. The study aimed to evaluate the physical, chemical, biological, stability, and clinical trials in pregnant women of aloe vera instant powder.

Method

This study utilized an experimental research design with a pre-post approach, carried out prospectively from May to August 2024 at the Pucuk Sirih Jamu Company in Banjarmasin. The purpose of this design was to evaluate the effects of aloe vera instant powder on bone and tooth growth, particularly in pregnant women. Ethical clearance for the research was granted by the Research Ethics Commission of Universitas Muhammadiyah Banjarmasin, with approval number 350/UMB/KE/V/2024, dated May 24, 2024.

The first step in the research process was sample preparation. Aloe vera meat, weighing 1 kg, was thoroughly cleaned under running water, blended with 500 mL of sterile water, and combined with an additional 2000 mL of sterile water. The mixture was boiled on low heat for 30 minutes and then filtered using a stainless-steel sieve. After filtering, 2 kg of sugar was added to the juice, and the mixture was continuously heated and stirred until it formed an instant powder. The powder was then stored in a tightly sealed container for further testing. An organoleptic evaluation was performed to assess the smell, color, taste, and texture of the instant powder.

Following this, the physical properties of the aloe vera instant powder were evaluated. Key assessments included flow time, angle of repose, and compressibility index. The flow time was measured in seconds, with an ideal flow time of under 10 seconds. The angle of repose was used to assess the flowability of the powder, with acceptable values ranging between 25 and 40^{o}. The compressibility index provided insights into the powder's compactness. Chemical evaluation involved phytochemical screening to ensure that essential chemical compounds, such as flavonoids, phenolics, tannins, and saponins, were retained in the instant powder after the processing stages. This evaluation ensured the product's consistency in delivering health benefits.

Biological testing was conducted to assess microbial contamination. The Most Probable Number (MPN) method was employed to detect coliform contamination, while the Total Plate Count (ALT) method was used to assess aerobic bacteria contamination. The MPN method consisted of three stages: presumptive testing, confirmatory testing, and complementary testing. The stability of the aloe vera instant powder was tested over a period of three months to determine its durability and reliability. Physical properties, such as organoleptic characteristics, flow properties, compressibility, and angle of repose, were tested at the beginning, after one month, and after two months to ensure stability.

A clinical trial was conducted on 10 pregnant women to assess the effects of aloe vera instant powder. The participants were divided into two groups: the treatment group, which consumed aloe vera instant powder, and a positive control group, which received calcium supplements. Both groups consumed their respective treatments twice a day for 10 consecutive days. Blood samples were collected from the participants on day 0 (before treatment) and day 11 (after treatment) for hematological analysis, which included measurements of red and white blood cell counts, hemoglobin levels, and calcium concentrations.

This comprehensive research process was designed to evaluate the potential of aloe vera instant powder as a nutritional supplement for improving bone and tooth health in pregnant women, ensuring its safety, stability, and effectiveness through various stages of testing.

Result and Discussion

This is part of a multi-year research entitled Qualitative and Quantitative Phytochemical Studies, Formulation of Preparations, and Preclinical Tests on Instant Powder Preparations of Aloe Vera Barbadensis Juice as a Support for Bone and Dental Growth. This research consists of research conducted in 2023 and 2024. In 2023, research was carried out related to the qualitative profile of phytochemical content, the percentage of quantitative content of chemical compounds, preparation formulations, preclinical toxic tests on animals, and preclinical tests on test animals from aloe vera instant powder. The results of the 2023 research have been published in the Health Info Journal, Volume 21, Edition 4, with the article titled Revolution of Bone and Teeth Health: Study of *Aloe barbadensis* Instant Powder Formulation. This research is a continuation of previous research. The 2024 study aimed to evaluate aloe vera instant powder's physical, chemical, and biological properties. Stability tests and clinical trials were conducted on pregnant women with aloe vera instant powder.

This research has received approval for Research Ethics Feasibility from the Research Ethics Commission of Muhammadiyah Banjarmasin University with number 350/UMB/KE/V/2024 dated 24 May 2024. The research stages include the formulation of instant powder preparations, evaluation of the physical properties of instant powder, and chemical evaluation of powder preparations with phytochemical screening methods. The next stage was biological evaluation with microbial contamination parameters using the Most Probable Number (MPN) Total Plate Count (ALT) method. Next, an accelerated stability test was conducted using the physical properties of the instant powder on the 0^{th}, 1^{st}, and 2^{nd} months. The last stage, namely clinical trials, was conducted on women for 10 days in the treatment group (instant powder) and positive control group (calcium supplement), totaling 5 people. Parameters in the clinical trial used the patients' hematological (blood) data.

This study formulated aloe vera instant powder, which facilitates the utilization of aloe vera in the community. Instant powder is a formula that dissolves easily in water and has a flavor acceptable to patients. The formulation results of the aloe vera instant powder preparation are presented in Table 1.

Table 1. Aloe vera instant powder formulation results

Parameters	Results
Flavor	Sweet
Smell	Odourless
Color	White
Shape	Powder

Aloe vera instant powder resulting from the formulation process has a sweet taste, odorless, white color, and powder form. The resulting form is a powder form. The resulting powder dissolves quickly in hot or cold water to optimize its utilization (Adhayanti & Ahmad, 2019; Ramadhia et al., 2012). Sweetness is produced from white sugar, one of the ingredients used. The use of granulated sugar will facilitate the process of powder crystal formation, so there is no need to add sugar at the time of use. This will make this preparation more practical. However, it should be noted that patients with diabetes mellitus cannot use this preparation (Niazi, 2009).

The resulting preparation is not fresh because flavoring ingredients are not added. In the future, in the development of formulas, flavoring preparations can be added to the formulas according to market needs (Cohen et al., 2019; Edam et al., 2016; Mwale, 2023; Tang et al., 2023). The color of the resulting powder is white. The color comes from white aloe vera meat and white granulated sugar. In this formula, there is no addition of dye, so the color produced is original. In the future, a natural coloring can be added as needed. The instant powder is then analyzed for a physical evaluation. The physical assessment describes the preparation's physical properties, affecting its physical quality, durability, and ease of handling during production. The results of the physical evaluation of instant powder preparations are presented in Table 2.

Table 2. Results of physical evaluation of instant powder preparations

Parameters	Replication	Testing Results	Average	Requirements
Flow Time (seconds)	1	8.5	8.63	$\leq$ 10
	2	8.6		
	3	8.8		
Angle of Silence (°)	1	36	34.33	25 – 40
	2	35		
	3	32		
Compressibility Index (%)	1	7.4	7.70	$\leq$ 10
	2	7.8		
	3	7.9		

The evaluation results of aloe vera instant powder showed an average flow time of 8.63 seconds, an angle of repose of 34.33°, and a compressibility index of 7.70%. Aloe vera instant powder flow time is known to average

8.63 seconds. Compared to the requirements of < 10 seconds (Edy, 2020), these results are by the requirements. Flow time is the time required for the powder to flow. If the flow time is too slow, the powder preparation will be difficult to flow. This affects the large-scale production process, especially the packaging process, which requires an optimum flow speed to facilitate the automatic setting of equipment in the production process.

The angle of silence of the instant powder preparation is 34.33°, compared to the requirements for a good angle of silence, which is 25 – 40° (USP, 2012); the results obtained in this study are appropriate. The angle of repose is influenced by the size of the powder and the friction between powders that produce electrical power between particles. Particle sizes that are too large will cause the angle of silence to be too small, while particle sizes that are too small will cause the angle of silence to be too large. A dwell angle meeting the requirements will facilitate powder flow during production and packaging. The results of the evaluation of the angle of repose obtained in this study have shown that the powder produced has met the requirements based on the angle of repose parameter.

The resulting instant powder was evaluated for its chemical content. The formulation process of aloe vera is expected not to remove the active compounds. The heating process during manufacture, including mixing with sugar and distilled water, is expected not to affect the properties of aloe vera (Herawati et al., 2012; López et al., 2022). The results of the chemical evaluation of aloe vera instant powder preparations are presented in Table 3.

Chemical evaluation results show that aloe vera instant powder still contains active compounds. The powder still contains efficacious chemical compounds, so efficacy will be obtained when consuming instant powder preparations (Mawardi, 2016; Wariyah et al., 2024). The compounds are flavonoids, phenolics, tannins, saponins, steroids, and interquinones. These results are not very different from previous studies that show that aloe vera contains this group of compounds.

The biological evaluation of aloe vera instant powder preparation included coliform and aerobic microbial contamination. Coliform contamination was evaluated using the Most Probable Number (MPN) method, while aerobic microbial contamination used Total Plate Numbers (ALT). The results of the biological evaluation of the preparation are presented in Table 4.

Table 3. Results of chemical evaluation of instant powder preparations

Phytochemistry	Reagents	Results
Flavonoids	Magnesium and Hydrochloric Acid	+
Phenolic	Iron Chloride	+
Alkaloids	Reage Meyer	-
Tannins	Gelatin Solution	+
Saponins	Aquades	+
Steroids	Liebermann Burchard	-
Terpenoids	Liebermann Burchard	-
Anthraquinone	10% KOH	+

Table 4. Biological evaluation of aloe vera instant powder preparation

Parameters	Methods	Results			Negative Control
		R1	R2	R3	
Coliform contamination	Most Probable Number (MPN)	Negative	Negative	Negative	Positive
Aerobic Microbial Contaminants	Total Plate Count (ALT)	Negative	Negative	Negative	Positive

A biological evaluation of the aloe vera instant powder preparation showed no coliform. This result was characterized by negative results in the three replicate tests. The test against the negative control as a comparison showed a positive result (Andersen, 2007; Elferjane et al., 2023; Rauwel et al., 2024). These results indicate that the powder is free from coliform contamination. The aerobic microbial contamination test showed negative results in three replications. In the negative control, it is known that the results show positive. Based on the two biological tests, it was found that the aloe vera instant powder was free of microbial contamination.

Stability testing of aloe vera instant powder preparations was carried out. The test was carried out for two months to ensure the identity, strength, and quality of products passed and circulated in the market so that they are safe for use by consumers. The stability test was conducted on the 0^{th}, 1^{st}, and 2^{nd} months. The results of the stability test are presented in Table 5.

The results of the preparation stability test on aloe vera instant powder showed that the flow time in months 0, 1, and 2 was consistent at the 8th second. This indicates that the flow time of the instant powder is stable. There is no flow time change during storage, so it can be declared stable (Saputro & Wardhana, 2021). In the stationary angle parameter, it was found that in the 0^{th}, 1^{st}, and 2^{nd} months, it was consistent at 34°. These results indicate that the instant powder preparation is stable in the stability test. In the compressibility index parameter, it was known that in the 0^{th}, 1^{st}, and 2^{nd} months, it was consistent at 7%. The compressibility

index of the powder preparation is stable. The stability test results based on the three physical parameters of the powder preparation showed that the aloe vera instant powder was stable.

Table 5. Stability test results of aloe vera instant powder preparations

Parameters	Month			Conclusion
	0	1	2	
Flow Time (seconds)	8.63	8.61	8.62	Stable
Angle of Silence (°)	34.33	34.41	34.56	Stable
Compressibility Index (%)	7.70	7.68	7.69	Stable

Stability testing is the ability of a product to maintain its quality based on qualitative and quantitative specifications over time. The stability test aims to ensure the identity, quality, purity, and strength of a product that consumers or patients will use. The stability test will later become one of the parameters for calculating the expiration date of preparation (Basar et al., 2013; Kholodenko, 2013; Rismana et al., 2015). The results of this study showed that the instant powder preparation was stable for two months based on the evaluation of physical properties. The results showed the consistency of the flow time, angle of repose, and compressibility index of the instant powder preparation. This can illustrate the durability of the instant powder (Deglas & Apriliani, 2022). The durability of aloe vera instant powder will make it easier to market the product and make production efficient.

The clinical trial was conducted on pregnant women with gestational age above the first trimester. The test was performed on the treatment group of 5 patients who were given aloe vera instant powder, while the control group consisted of 5 patients who were given calcium supplements. The study results are presented as average test results in each group. The results of the hematological analysis of patients' blood are presented in Table 6.

The clinical trial results showed that in the red blood cell parameters of the treatment group, patients experienced an increase in red blood cells, while the rise in the positive control was only slight. The positive control used calcium supplements, so they did not significantly influence the increase in red blood cells. There was also an increase in white blood cell parameters in the treatment group and control group, with the highest increase in the treatment group. Hemoglobin parameters also increased before and after treatment, while the use of calcium supplements did not increase. The potassium, calcium, phosphorus, and magnesium parameters also increased before and after giving aloe vera instant powder or calcium supplements.

Red blood cells function to transport and circulate oxygen throughout the body. Based on the literature, it is known that giving aloe vera can increase red blood cells. This suggests aloe vera instant powder can increase red blood cells (Arif et al., 2022; Prasetio & Fakhrudin, 2017). White blood cells function to protect and handle infections against foreign molecules that enter the body of Aloe vera. In this study, aloe vera instant powder increased white blood cells. Based on the literature, it is known that aloe vera administration can indeed increase white blood cells (Arif et al., 2022). Hemoglobin transports oxygen and carbon dioxide. In this study, aloe vera increased hemoglobin. In the control group, there was no increase in hemoglobin because the control used was a supplement. Lack of hemoglobin can cause anemia, which results in decreased growth, including bone and tooth mass. In this study, aloe vera powder increased hemoglobin. Aloe vera contains high calcium so that it can increase calcium levels in the blood (Sari et al., 2022).

Table 6. Results of blood hematology analysis of patients in the treatment and control groups positive

Parameters	Treatment		Positive Control		Normal	Unit
	Before	After	Before	After		
Red Blood Cells	4.6	4.9	4.5	4.6	3.5–5.2	million/CML
White Blood Cells	5.432	5.531	5.472	5.482	4.000–10.000	per µL of blood
Hemoglobin	13.4	14.7	13.3	13.4	12–16	g/dL
Potassium	4.4	4.9	4.3	5.0	3.5–5.2	mmol/L
Calcium	8.8	9.5	8.7	9.6	8.5–10.2	mg/dL
Phosphorus	3.4	3.9	3.2	3.4	2.8–4.5	mg/dL
Magnesium	1.9	2.1	1.8	1.9	1.8–2.2	mg/dL

There was an increase in potassium, calcium, phosphorus, and magnesium before and after giving aloe vera instant powder. Aloe vera contains potassium, calcium, phosphorus, and magnesium, so giving aloe vera powder can increase these mineral levels (Sari et al., 2022). A reasonably high increase in calcium levels occurred in the positive control group given calcium supplements. In other mineral contents such as potassium, phosphorus, and magnesium, the increase in

the positive control was not too high because the supplements given were only calcium supplements.

The results indicate that aloe vera instant powder is stable and retains key phytochemical compounds after formulation, making it a practical preparation for use. The powder's physical properties, including its flow time and angle of repose, meet industrial standards, ensuring it is easy to handle and package for large-scale production. The presence of active compounds such as flavonoids, tannins, and saponins suggests that the powder maintains its therapeutic potential, which aligns with previous studies on aloe vera's health benefits. The absence of microbial contamination further confirms its safety for consumption.

The clinical trial outcomes are promising, showing that aloe vera instant powder supports an increase in key blood and mineral parameters in pregnant women, which are essential for bone and tooth growth. This suggests that aloe vera can be a beneficial supplement for pregnant women, particularly in improving red blood cell count and overall mineral balance. Further studies could explore flavor improvements and longer-term stability.

Conclusion

The results of the physical properties evaluation of the instant powder preparation obtained a flow time of 8.63 seconds, an angle of repose of 34.33°, and a compressibility index of 7.70%. Chemical evaluation showed flavonoids, phenolics, tannins, saponins, steroids, and interquinones. The biological assessment showed no contamination of coliform or aerobic bacteria. The stability test showed that aloe vera instant powder was stable at two months of storage. Aloe vera instant powder tested on pregnant women increased red blood cells, white blood cells, hemoglobin, potassium, calcium, phosphorus, and magnesium in the blood.

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Author Contributions

All authors participated in drafting and revising the article, and jointly take responsibility for all aspects of this study.

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Conflicts of Interest

No conflicts of interest pertinent to this article were disclosed by the authors.

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The results of the preparation stability test on aloe vera instant powder showed that the flow time in months 0, 1, and 2 was consistent at the 8th second. This indicates that the flow time of the instant powder is stable. There is no flow time change during storage, so it can be declared stable (Saputro & Wardhana, 2021). In the stationary angle parameter, it was found that in the 0^{th}, 1^{st}, and 2^{nd} months, it was consistent at 34°. These results indicate that the instant powder preparation is stable in the stability test. In the compressibility index parameter, it was known that in the 0^{th}, 1^{st}, and 2^{nd} months, it was consistent at 7%. The compressibility index of the powder preparation is stable. The stability test results based on the three physical parameters of the powder preparation showed that the aloe vera instant powder was stable.

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