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Ability of Potassium Factor in *Phyllanthus niruri* Infuse for Calcium Salt Solubility in Kidney Stones by *Ex Vivo*

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ABSTRACT

Phyllanthus niruri has large potassium content, this plant is used as a kidney stone crushers. The purpose of this study was to observe the ability of potassium in *phyllanthus niruri* infuse to assay the solubility of calcium kidney stones. Methodology of research conducted in form of dissolving kidney stones intact in *phyllanthus niruri* infuse before and after destruction and both measured with atomic absorption spectrophotometry. The results of research was exhibited that potassium factor has been increase intake calcium levels in kidney stones with treatment before and after destruction of *phyllanthus niruri* with various concentrations, The calcium content in the kidney stones after being given the 2% infuse *phyllanthus niruri* and after destruction of 2% *phyllanthus niruri* each grading 25.41% and 43.78%; *phyllanthus niruri* and after destruction of 4% *phyllanthus niruri* each grading 24.56% and 34.26%; and 10% the *phyllanthus niruri* infusion and after destruction of 10% *phyllanthus niruri* each grading 7.76% and 16.61%. This means the content of potassium in the *phyllanthus niruri* have a major role in the reduction in calcium content of kidney stones. The methods that was used had a good accuracy and precision with recovery test was 116.54% and relative standard deviation was 1.22%. The concluded that potassium factor in *phyllanthus niruri* has a major role in the dissolution of calcium in the kidney stones can be seen in terms of calcium content of the infusion before and after destruction.

Keywords: *Elephantopus scaber* L., Deoxyelephantopin, Isodeoxyelephantopin, RP-HPLC

INTRODUCTION

Indonesia which consists of many islands that inhabited by various tribes allow for differences in the use of plants as traditional medicine. This is due to each tribe had empirical experience and unique culture in accordance with their respective regions. The lives of ancestors that blend with nature growing awareness that nature is a provider of drugs for themselves and society [1].

One of these plants is *Phyllanthus niruri* from the Euphorbiaceae family. This plant grows wild in moist and rocky environment with green stem, the leaf blade rounded to complement the elongated egg and green fruit. *Phyllanthus niruri* tastes somewhat bitter, cool, and as astringent [2] Kidney stones frequent occur in tropical countries. Tropical weather causes dehydration so thickening urine sediment into rock easily formed. Another factor of kidney stones are content of salt in the urine and metabolic imbalance in the body that cause salts in the urine precipitate and form crystals [3,4].

P. niruri plants contain potassium which works to inhibit the formation of calcium crystals that can be used as an alternative medicine for healing kidney stones. High content of potassium can destroy calcium in kidney stones, because potassium will get rid of calcium to join carbonate, oxalate, phosphate, or uric forming potassium oxalate, potassium carbonate, potassium phosphate or potassium urate compounds that soluble in water [5,6].

Several researchers have conducted a study of some types of plants that can dissolve kidney stones. For example The study about ethanol extract of corn silk can dissolve kidney stones by complexometric titration method 70% ethanol extract of blue grapes have the ability to dissolve kidney stones of calcium as measured by atomic absorption spectrophotometer. According to Hidayati et al., the use of tempuyung dried tea leaves with certain frequencies can dissolve the calcium oxalate as measured using gravimetric method. Based on these studies, potassium and flavonoid involved in the decaying process of kidney stone [7-9].

Treatment of kidney stones with surgery, endoscopy or ultrasound requires relatively high costs so that the use of drugs that can prevent and decay kidney stones are chosen.

Kidney stones that are still small to medium possible to be dissolved with certain compounds. The aim of this study are be analyzed the ability of *P. niruri* infuse for dissolving of calcium in kidney stones and measure using atomic absorption spectrophotometry.

Recovery test

Then proceed with the wet destruction procedure as has been done previously [10-15]. Percentage recovery can be calculated by the following formula:

$$\text{Percentage recovery} = \frac{C_F - C_A}{C_A^*} \times 100\%$$

Explanation: C_A =Mineral levels in the sample before adding standard, C_F =Mineral levels in the sample before adding standard, C_A^* =Levels of standard solution that being added.

Determination of relative standard deviation

The formula for calculating the relative standard deviation is as follows [14]:

$$\text{RSD} = \frac{SD}{\bar{X}} \times 100\%$$

Explanation: \bar{X} =Average levels of sample, SD=Standard deviation, RSD=Relative standard deviation.

RESULTS AND DISCUSSION

Analysis of calibration curves of potassium and calcium

The calibration curve of standard solution of potassium and calcium can be seen in Figures 1 and 2.

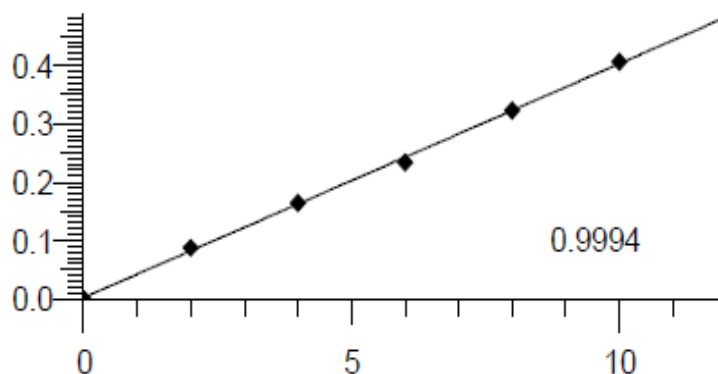


Figure 1: Calibration curve of standard solution of potassium

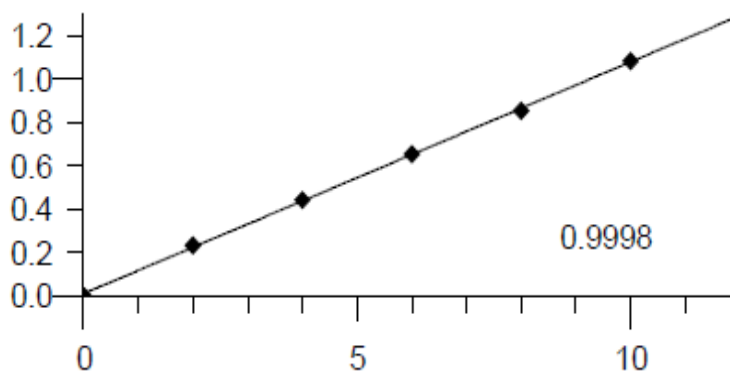


Figure 2: Calibration curve of standard solution of calcium

The calibration curve in atomic absorption spectrophotometre made in various concentrations with increasing concentration. From the measurement of the calibration curve obtained the regression equation is $Y=0.1071X-0.0083$ for potassium, and $Y=0.0401X-0.0017$ for calcium.

Analysis of initial calcium and potassium content before incubation with kidney stones without and with destruction

Based of Tables 1 and 2 can be seen that there is a difference between the average levels of calcium and potassium at various concentration of infuse.

Table 1: The Results of initial calcium and potassium levels before incubation with kidney stones without destruction

S. No.	Infuse sample	Calcium content (µg/ml)	Potassium content (µg/ml)
1	2%	13.48 ± 0.3238	211.21 ± 0.6614
2	4%	18.54 ± 0.2092	310.30 ± 0.7602
3	6%	23.44 ± 0.7944	326.14 ± 0.7268
4	8%	27.33 ± 0.8262	338.27 ± 0.6410
5	10%	29.60 ± 0.7491	379.50 ± 0.7592

Table 2: The Results of initial calcium and potassium levels before incubation with kidney stones with destruction

S. No.	Infuse sample	Calcium content (µg/ml)	Potassium content (µg/ml)
1.	2%	16.81 ± 0.5445	239.17 ± 0.9837
2.	4%	20.54 ± 0.1383	340.54 ± 1.8372
3.	6%	26.47 ± 0.9926	368.33 ± 1.3860
4.	8%	30.47 ± 0.3811	390.99 ± 1.2721
5.	10%	34.80 ± 0.5727	416.24 ± 1.2229

Table 3: Dissolved levels in intact kidney stones without destruction

S. No.	Infuse concentration	Initial K concentration (µg/ml)	Initial Ca concentration (µg/ml)	Ca concentration after incubation (µg/ml)	Dissolved Ca concentration (µg/ml)
1	2%	211.2104	13.4808	16.9057	3.4249
2	4%	310.3042	18.5370	23.0903	4.5533
3	6%	326.1354	23.4374	25.5071	2.0397
4	8%	338.3708	27.3297	29.5013	2.1716
5	10%	379.4958	29.5989	31.8953	2.2964

Table 4: Dissolved levels in crushed kidney stones without destruction

S. No.	Infuse concentration	Initial K concentration (µg/ml)	Initial Ca concentration (µg/ml)	Ca concentration after incubation (µg/ml)	Dissolved Ca concentration (µg/ml)
1	2%	211.2104	13.4808	19.3828	5.9020
2	4%	310.3042	18.5370	24.8878	6.3508
3	6%	326.1354	23.4374	28.3271	4.8897
4	8%	338.3708	27.3297	31.9098	4.5801
5	10%	379.4958	29.5989	34.5138	4.9149

Table 5: Dissolved levels in intact kidney stones with destruction

S. No.	Infuse concentration	Initial K concentration (µg/ml)	Initial Ca concentration (µg/ml)	Ca concentration after incubation (µg/ml)	Dissolved Ca concentration (µg/ml)
1	2%	239.1625	16.8123	21.8579	5.0456
2	4%	340.5333	20.5341	27.2111	6.6770
3	6%	368.3271	26.4693	30.8229	4.3536
4	8%	390.9917	30.4717	33.4975	3.0258
5	10%	416.2375	34.7985	37.3255	2.5270

Table 6: Dissolved levels in crushed kidney stones with destruction

S. No.	Infuse concentration	Initial K concentration (µg/ml)	Initial Ca concentration (µg/ml)	Ca concentration after incubation (µg/ml)	Dissolved Ca concentration (µg/ml)
1	2%	239.1625	16.8123	28.0508	11.2385
2	4%	340.5333	20.5341	33.909	13.3749
3	6%	368.3271	26.4693	39.5303	13.061
4	8%	390.9917	30.4717	45.7793	15.3076
5	10%	416.2375	34.7985	49.1272	14.3287

Based of Tables 3-6, can be seen that there is a difference between the initial calcium with that after incubation with intact kidney stones.

This shows that phyllanthus niruri infuse at various concentrations can dissolve kidney stones either without or with destruction and can be concluded that there is no linear correlation between the amount of potassium in the infusion with the amount of dissolved calcium. This is most likely because the factor of the calcium salt that are not homogeneous in the lining of the kidney stones, both in the type and number of compounds, so that the solubility of calcium salts concentrations infusion that was made ascending, the increase is not linear.

According to Winarto and Karyasari, potassium which makes kidney stones in the form of the calcium salt is scattered, because potassium will get rid of the calcium contained as a compound of calcium oxalate, carbonate, phosphate or uric which is forming kidney stones, and will form compounds of potassium salt which is more soluble in water, so the calcium salt in the kidney stones will dissolve slowly and come out with urine [6].

Levels of dissolved calcium is the increment calcium levels after incubation with kidney stones in phyllanthus niruri infuse at 37°C for 4 h and stirred every 10 min. Levels of dissolved Ca is the Ca levels after incubation minus the initial Ca levels. The ability of dissolving potassium ion towards calcium salt in the kidney stones caused by the posititon of potassium in the series volta is located in the left, so that potassium will get rid of calcium to join carbonate, oxalate, or urate and calcium become soluble [6]

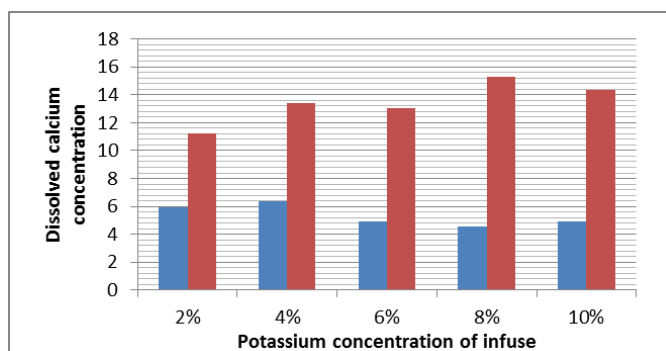


Figure 3: Dissolved calcium levels in crushed kidney stones without and with destruction graph

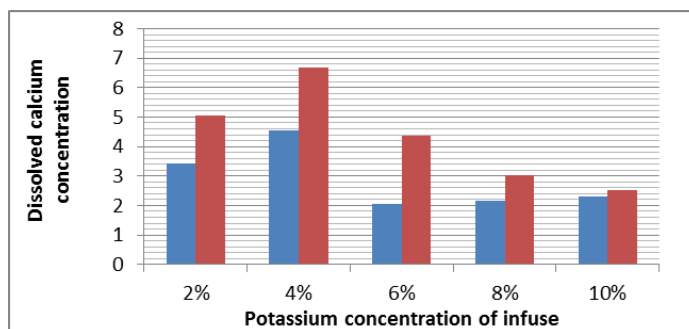


Figure 4: Dissolved calcium levels in intact kidney stones without and with destruction graph

Based on Figures 3 and 4 can be seen that the solubility of calcium salts in kidney stones either intact or crushed are greater in the state with destruction. This is caused by the state after the destruction, the contents left on infusion just the minerals contained in plants phyllanthus niruri, and if the less solute in a solution, the ability of dissolving another substance will be higher, whereas in the absence of destruction, some substances contained in plants phyllanthus niruri (filantin, saponins, flavonoids, polyphenols, potassium, and resin) in the infusion, so the ability of dissolving another substance will be smaller [6].

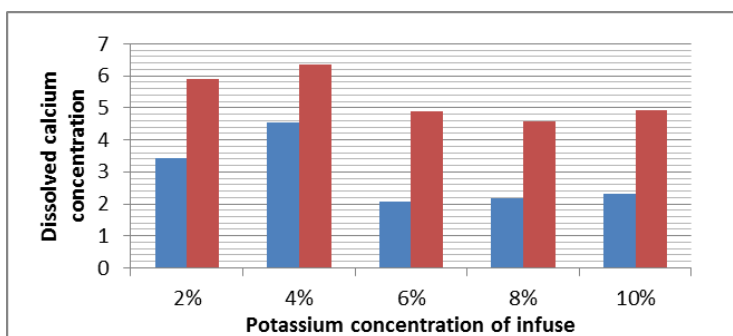


Figure 5: Dissolved calcium levels in intact kidney stones without destruction

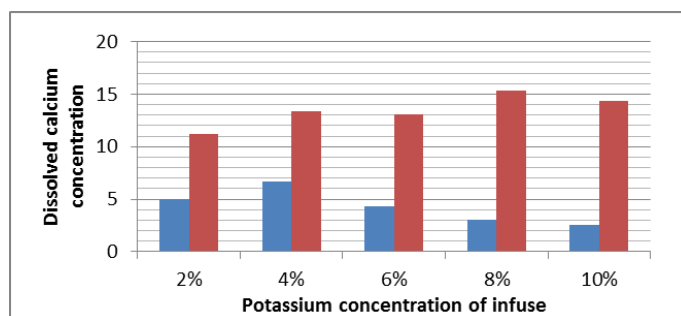


Figure 6: Dissolved calcium levels in intact kidney stones with destruction

Based of Figures 5 and 6 that can be seen the solubility of calcium salts in crushed kidney stone higher than in intact kidney stones. This is due to the effect of the increase of the surface area of the kidney stones. With a larger surface area will increase the solubility of a substance. The wider the surface resulting in more surface contact with the solvent, so that at the same time more and more particles are soluble.

LOD and LOQ

Based on the calibration curve obtained calcium and potassium limit of detection and limit of quantitation for both the minerals. The limit of detection and limit of quantitation calcium and potassium can be seen in Table 7.

Table 7: LOD and LOQ

S. No.	Mineral	Limit of detection ($\mu\text{g/ml}$)	Limit of quantitation ($\mu\text{g/ml}$)
1	Calcium	0.5623	1.7545
2	Potassium	0.626	2.0868

From the results of calculations can be seen that all the results obtained in the sample measurement is above the LOD and LOQ.

Recovery test

The recovery test of calcium level after the addition of standard solution of calcium in the sample can be seen in Table 8.

Table 8: Recovery percentage of calcium levels

S. No.	Analysis mineral	Recovery (%)	Requirement ranges of recovery (%)
I	Calcium	116.54	80-120

Based on Table 8. it can be seen that the average results of recovery test for the calcium content of 116.54%. The percent recovery showed satisfactory precision of the work at the time of examination levels of calcium. The recovery test is eligible accuracy that is determined, if the average results of recovery are in the range of 80-120% [10,14,15].

Relative standard deviation

From the results performed on the measured data of the minerals calcium and potassium levels in *Phyllanthus niruri* infuse without and with destruction, the value of relative standard deviation (RSD) is presented in Table 9.

Table 9: The value of standard deviation and relative standard deviation of calcium

S. No.	Mineral	Standard Deviation	Relative Standard Deviation
1	Calcium	1.4229	1.22%

Based on Table 9 above, it can be seen the value of standard deviation (SD) for calcium mineral is 1.42 while the value of relative standard deviation (RSD) is obtained at 1.22% for the calcium mineral.

According to Harmita, the value of relative standard deviation (RSD) for analytes with levels of parts per million ($\mu\text{g/ml}$) is not more than 16% and for analytes with levels of parts per billion (ppb) its RSD is not more than 32%. From the results obtained indicate that the methods do have a good precision [14].

CONCLUSION

That potassium contained in *Phyllanthus niruri* has a great influence to destroy and dissolve calcium; it is evident that with the increasing concentration of infusion means the concentration of potassium increases as well, so it has a major role in the destruction and dissolving the calcium in the kidney stones.

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