



Detailed Pharmacognostical and Phytochemical Evaluation of *Quassia Amara* linn. Heart wood - Emerging Herb for Diabetes

Ankush Gunjal^{*1}, Manisha Walunj², Harisha CR³, Mandip Goyal⁴, VJ Shukla⁵

1. Ph.D Scholar, Department of Kayachikitsa.

2. Ph.D Scholar, Department of Rasashastra and Bhaishajya Kalpana.

3. Head, Pharmacognosy Laboratory.

4. Assistant Professor, Department of Kayachikitsa.

5. Head, Pharmaceutical Chemistry Laboratory.

ABSTRACT

In Ayurvedic texts the description of '*Apathyanimittaja Prameha*' which is generally seen in obese individuals have a clear cut resemblance with type-2 diabetes and physicians believe in treating diabetes with herbal dietary supplements among which *Quassia amara* Linn. is a small indigenous tree native of Suriname. Detailed investigation of Transverse section including its powder of heartwood of *Quassia amara* Linn. was carried out. Preliminary organoleptic features and results of powder microscopy reveals presence of tannin and large amount of fibres. The information generated by this particular study will provide relevant pharmacognostical and physiochemical data needed for proper identification and authentication of heartwood of this particular species.

Keywords: Heartwood, pharmacognosy, physiochemical analysis, *Quassia amara*.

*Corresponding Author Email sadiq.masud@udusok.edu.ng

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INTRODUCTION

Diabetes mellitus, a metabolic disorder, is a major global health concern with a projected rise in prevalence from 171 million in 2000 to 366 million in 2030¹. Around 3.2 million deaths every year are attributable to complication of diabetes. In top ten countries, India ranks first having 18% of the total diabetic population of the world with 31705000 patients. In *Ayurvedic* texts the given characteristic features of *Prameha* shows marked similarity with the syndrome of Diabetes mellitus. The description of '*Apathyanimitaja Prameha*'² which generally seen in obese individuals have a clear cut resemblance with type-2 diabetes (non insulin dependent diabetes mellitus). Type 2 diabetes comprises 90% of people with diabetes around the world, and is largely the result of excess body weight and physical inactivity. It is very difficult to gain victory over genetic factors but certainly by diet control and use of appropriate choices of drugs better quality of life of the patient can be ensured. In western countries physicians believe in treating diabetes with herbal dietary supplements³ among which *Quassia amara* Linn. is a small indigenous tree native of Suriname, 6 to 18 feet tall and contains the phytochemical quassin, the bitterest substance found in nature. The bitterness threshold is 1: 60.000. It stimulates the liver and among other properties, controls the blood sugar. In traditional medicine in Suriname, quassia is used for carcinoma, debility, dyspepsia, fever, heptos, hyperglycemia, malaria, snakebite and spasms and the bark is considered to be depurative, insecticidal, laxative, stomachic, tonic, and vermifuge.⁴ The present article deals with the standardization of *Quassia amara* Linn. through pharmacognostical and physicochemical standards.

MATERIALS AND METHOD

Collection and authentication of raw drug

The market sample of herb *Quassia amara* Linn. was procured from Mumbai. The pharmacognostical authentication of *Quassia amara* Linn. was based on the morphological features, organoleptic characters and powder microscopy of the sample drug and was performed at pharmacognostical laboratory of IPGT and RA, GAU, Jamnagar.

Pharmacognostical evaluation

Pharmacognostical analysis of *Quassia amara* Linn. comprises of organoleptic characters i.e., color, odor, taste, and texture, was recorded, along with TS of heartwood and powder microscopy. Sections and powder were treated with and without stain to find out the lignified material along with other cellular components. The microphotographs were taken under Carl Zeiss Trinocular microscope attached with camera.⁵⁻⁶

Histochemical evaluation

Thick sections of sample were subjected to Histochemical tests to find starch grains, tannin, calcium etc. by treating various reagents.⁷

Physicochemical evaluation

Quassia amara Linn. were analyzed through relevant physicochemical parameters such as loss on drying, water soluble extract, alcohol soluble extract, and pH value.⁸⁻¹⁰ In qualitative analysis, presence of Sugar, Flavanoids, Tannins - Phenolic Compounds, Alkaloids, Carbohydrates, Mucilage, Protein, Steroid, Glycoside and Saponin were assessed. High performance thin layer chromatography (HPTLC) was carried out with methanolic extract of *Quassia amara* Linn. powder.¹¹⁻¹²

High performance thin layer chromatography

Methanolic extract of *Quassia amara* Linn. was spotted on pre-coated silica gel GF 60254 aluminum plate by means of Camag Linomat V sample applicator fitted with a 100 µL Hamilton syringe. Chloroform: MeOH (9:1) was used as the mobile phase. After development, densitometric scan was performed with a Camag TLC scanner III in reflectance absorbance mode at UV detection as 254 nm and 366 nm under the control of Win CATS Software (V 1.2.1. Camag).

RESULTS AND DISCUSSION




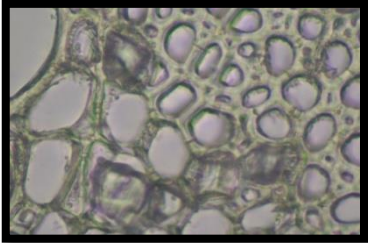
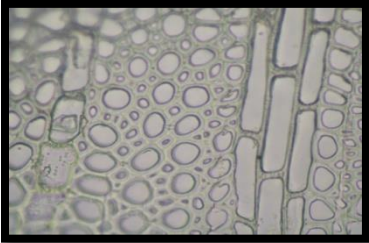
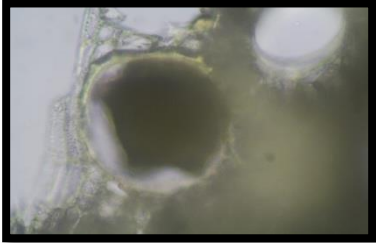
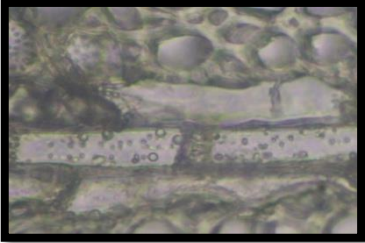

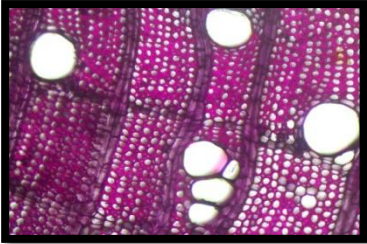
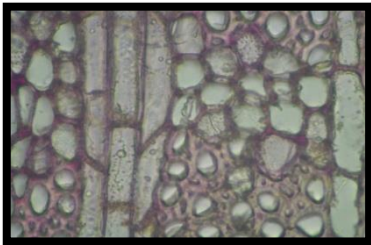
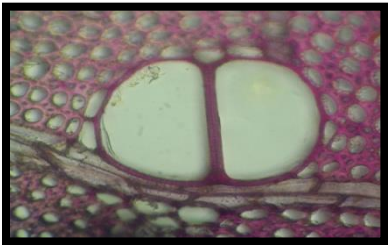
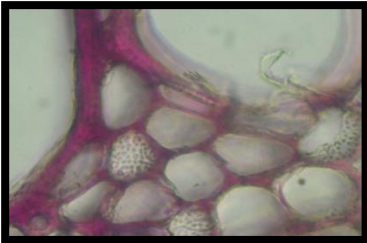
Macroscopic study of dried heartwood

Heartwood was creamish yellow, very hard, light in weight, even textured, odor characteristic, taste bitter.

Transverse section of Heartwood

Detailed TS showed discontinuous arranged fibres passing through the medullary rays were lignified fibres. Vascular bundle was radially arranged and 2-4 xylem vessels were radially arranging along with heavy xylem fibres and parenchyma. Some of the xylem vessels were filled with tannin and oil content. Some of the parenchyma cells contained prismatic crystals of calcium oxalate. Wood showed vessels which were solitary or arranged in small radial groups and often blocked with tylosis impregnated with tannin. Medullary rays uniserriate to triserriate started from tail region of xylem and was passing through all over the vascular bundle (Xylem). Medullary rays were rich in oil globules with rarely prismatic crystals of calcium oxalate.(plate 1 figure 1-12)

Plate 1. Microphotographs of heart wood


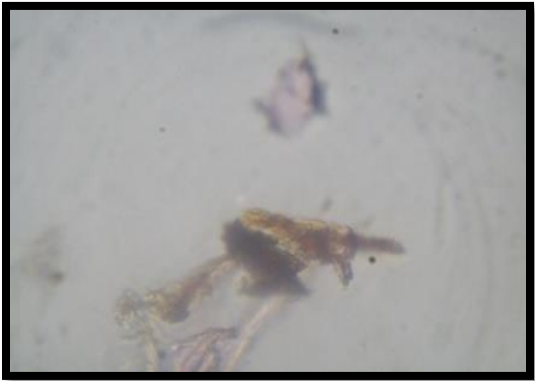

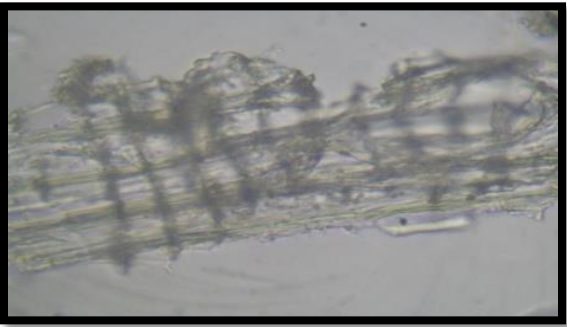


		
Figure 1. Quassia amara in natural habitat	Figure 2. Heart wood	Figure 3. Xylem with parenchyma
		
Figure 4. Calcium oxalate crystals	Figure 5. Triserriate medullary rays	Figure 6. Xylem filled with oil with tannin
		
Figure 7. Medullary rays with starch grains	Figure 8. Intravascular pitting xylem	Figure 9. Isolated 2-3 xylem vessels
		
Figure 10. Lignified parenchyma	Figure 11. Xylem fibres and parenchyma	Figure 12. Pitted parenchyma cells

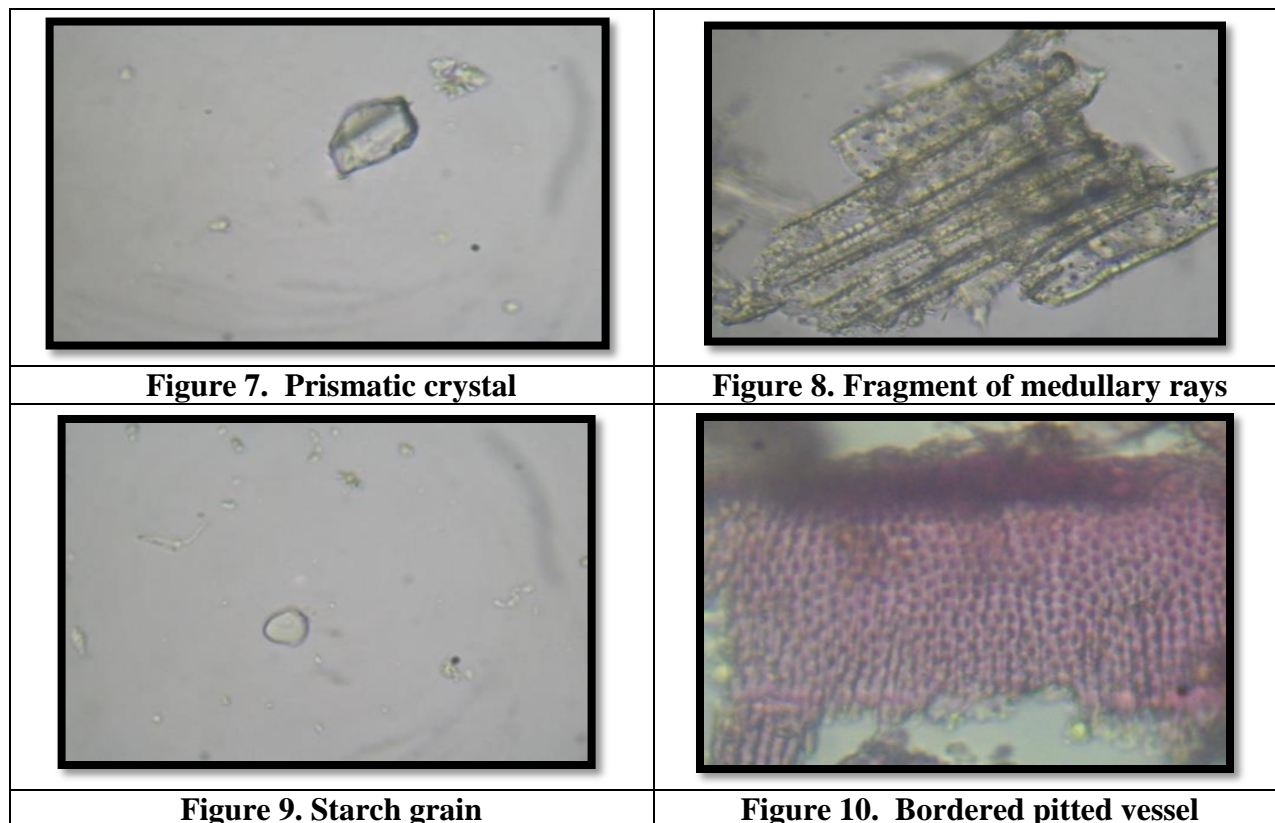
Powder microscopy of dried heartwood

Organoleptic characters

Color of the powder was light creamish yellow, characteristic odour, bitter in taste and rough in touch. The diagnostic characters of Powder microscopy of heartwood of *Quassia amara* Linn. showed presence of oil globules, simple starch grains, fibres passing through medullary rays, fragments of medullary rays, dark brown content, prismatic crystals crystal of Ca-oxalate, group of fibres, bordered pitted vessels and lignified fibres. (plate 2 fig 1-10)

Plate 2. Powder Microphotographs

	
Figure 1. Heart wood powder	Figure 2. Brown content
	
Figure 3. Simple fibre	Figure 4. Fibres passing through medullary rays
	
Figure 5. Lignified fibres	Figure 6. Oil globule



Histochemical analysis

Quassia amara Linn. powder was treated with FeCl₃ solution tannin and its cellular constituent immediately changed into black color which confirmed presence of tannin content. Further, *Quassia amara* Linn. powder was treated with Iodine solution which confirmed presence of starch content by appearance of dark blue color.

Physicochemical analysis

Quassia amara Linn. was analyzed using relevant physicochemical parameters such as Aqueous Extractive, Alcohol Extractive (methanolic extract), pH, Loss on drying at the pharmaceutical chemistry lab. The observations are presented in Table 1.

Table 1: Physicochemical parameters

Sr. No.	Test	Result
1	Aqueous Extractive	7.394 % w/w
2	Alcohol Extractive (methanolic extract)	6.198 % w/w
3	pH	6.0
4	Loss on drying	7.775 % w/w

Preliminary phytochemical analysis

Presence of presence of Flavanoids, Tannins - Phenolic Compounds, Carbohydrates, Steroid and Glycoside, was confirmed through the suitable tests while Sugar, Alkaloids, Mucilage, Protein and Saponin could not be detected in *Quassia amara* Linn. [Table 2].

Table 2: Qualitative Test

Material	Functional Group	Result
Alcoholic Extract of powder of <i>Quassia amara</i> Linn.	Sugar	Absent
	Flavanoids	Present
	Tannins + Phenolic Compounds	Present
	Alkaloids	Absent
	Carbohydrates	Present
	Mucilage	Absent
	Protein	Absent
	Steroid	Present
	Glycoside	Present
Saponin	Absent	

HPTLC

On analyzing under densitometer at 254 nm, the chromatogram showed nine peaks. While at 366 nm, the chromatogram showed 5 peaks [Plate.3 and Table 3]. The pharmacognostical evaluation of heartwood showed that it consists of large amount of lignine, and presence of large amount of fibres in the transverse section, recent studies reports that more intake of fiber intake improves glycemic control and reduced absolute values of glycated hemoglobin and fasting plasma glucose.¹³ The possible mechanisms for metabolic improvements with dietary fiber include delay of glucose absorption, increase in hepatic extraction of insulin, increased insulin sensitivity at the cellular level, and binding of bile acids.¹⁴ Xylem vessels were filled with tannin content, tannic acid stimulates glucose transport and inhibits adipocyte differentiation which is effective in diabetic individuals.¹⁵ Starch and oil globules were present all over the section. Preliminary qualitative analysis proves the presence of Flavanoids, Tannins - Phenolic Compounds, Carbohydrates, Steroid and Glycosides. Flavanoids stimulate glucose uptake in peripheral tissues, regulate the activity and expression of the rate-limiting enzymes in the carbohydrate metabolism pathway and act per as insulin secretagogues or insulin mimetics, probably, by influencing the pleiotropic mechanisms of insulin signaling, to ameliorate the diabetes status.¹⁶ The presence of certain definite constituents in *Quassia amara* Linn. were observed in TLC and HPTLC in which maximum nine spots were distinguished and most of the Rf values were identical in the alcoholic extract.

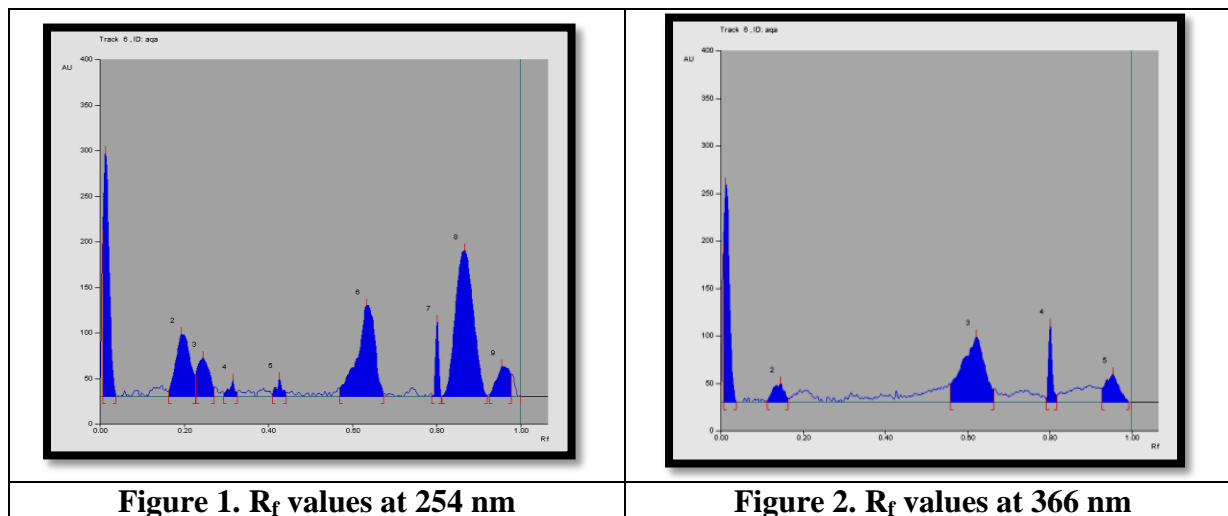
Figure 1. R_f values at 254 nmFigure 2. R_f values at 366 nm

Plate 3 HPTLC

Table 3: HPTLC

	Spots	R_f values at 254 nm
HPTLC	9	0.01, 0.19, 0.25, 0.32, 0.43, 0.63, 0.80, 0.87, 0.96
		R_f values at 366 nm
	5	0.01, 0.15, 0.62, 0.80, 0.95.

CONCLUSION

Preliminary organoleptic features and results of powder microscopy reveals presence of tannin and large amount of fibres. Preliminary qualitative analysis proves the presence of Flavanoids, Tannins - Phenolic Compounds. These identified phytochemical components support the intended action of the drug to manage Diabetes mellitus (Type II). As no published information is available on pharmacognostical and physico-chemical profile of heartwood of *Quassia amara* Linn. this preliminary information can be used for reference in future.

REFERENCES

1. S. Wild, G. Roglic, A. Green, R. Sicree, and H. King, "Global prevalence of diabetes: estimates for the year 2000 and projections for 2030," *Diabetes Care*, vol. 27, no. 5, pp. 1047–1053, 2004. View at Google Scholar [Last cited on 2014 Apr 24].
2. Sushruta Samhita with Dalhan's "Nibandhasamgraha"&Gayadas's "Nyayachandrika" Commentaries, Edited by Vaidya Yadavaji Trikamaji Acharya, Chaukhamba Surabharati Prakashan, Varanasi. (2003), Chikitsa Sthana 11/3
3. E. A. Ryan, M. E. Pick, and C. Marceau, "Use of alternative medicines in diabetes mellitus," *Diabetic Medicine*, 2001;18(3): 242–245, 2001.
4. <http://www.tropilab.com/applicationanddosagequassia.html>, cited on 16-10-2013 at 8pm.

5. Khandelwal KR. Examination of powdered drugs. In: Khandelwal KR, editor. Practical Pharmacognosy techniques and experiments. 19th ed. Pune: Nirali Prakashan; 2008. p. 162-6.
6. Anonymous. The Ayurvedic Pharmacopoeia of India, Part 2. Vol 1. 1st ed. New Delhi: Ministry of Health and Family welfare, Department of AYUSH Government of India; 2008. p. 136-9.
7. Jesse W, Li H, Vaderas JC. Drug discovery from natural products: End of an era or an endless frontier. *Science* 2009; 325:161-5.
8. Ibidem. The Ayurvedic Pharmacopoeia of India, Part 2. Vol 1; p. 140, 141, 147, 239.
9. Anonymous. Quality control methods for medicinal plant materials. Geneva: World Health Organization; 1998.
10. Anonymous. Parameters for qualitative assessment of Ayurveda and Siddha drugs, Part A. New Delhi: CCRAS; 2005. p. 31
11. Anonymous. Basics of Analytical and Phytochemical Analysis of Medicinal plants, analytical methods for ASU Single/ Compound drugs sponsored by WHO. Chennai: CSMDRI for Ayurveda and Siddha CCRAS, Govt. of India; 2008. p.11.
12. Stahl E. Thin layer chromatography a laboratory hand book. Berlin: Springer-Verlag; 1969. p.125-41.
13. Silva FM1, Kramer CK, de Almeida JC, Steemburgo T, Gross JL, Azevedo MJ, Fiber intake and glycemic control in patients with type 2 diabetes mellitus: a systematic review with meta-analysis of randomized controlled trials. *Nutrition Reviews*. 2013 Dec;71(12):790-801. doi: 10.1111/nure.12076. Epub 2013 Nov 1. Available on <http://www.ncbi.nlm.nih.gov/pubmed/24180564> [Last cited on 2014 Apr 24].
14. Tabatabai A1, Li S. Dietary fiber and type 2 diabetes. *Clin Excell Nurse Pract*. 2000 Sep;4(5):272-6. PMID: 11858448 [PubMed - indexed for MEDLINE] available on <http://www.ncbi.nlm.nih.gov/pubmed/11858448#> [Last cited on 2014 Apr 24].
15. Liu X, Kim J-K, Li Y, Li J, Liu F, Chen X. Tannic acid stimulates glucose transport and inhibits adipocyte differentiation in 3T3-L1 cells. *J Nutr* 2005;135:165-171. Available form: <http://jn.nutrition.org/content/135/2/165.full> [Last cited on 2014 Apr 24].
16. Cazarolli, Luisa H.; Zanatta, Leila; Alberton, E Heloisa; RB Figueiredo, M Santos; Folador, Poliane; Damazio, R Guollo; Pizzolatti, M Geraldo; MB Silva, F Regina, Flavonoids: Cellular and Molecular Mechanism of Action in Glucose Homeostasis, Mini Reviews in Medicinal Chemistry, Bentham Science Publishers, Volume 8, Number 10, September

2008,pp.1032-1038(7) Available on: <http://www.ingentaconnect.com/content/ben/mrmc/2008/00000008/00000010/art00007> [Last cited on 2014 Apr 24].



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