Journal of Phytonanotechnology and Pharmaceutical Sciences 2(3): 14-18, 2022

Online ISSN:2583-0376

http://jpps.ukaazpublications.com

DOI: http://dx.doi.org/10.54085/jpps.2022.2.3.3

Journal of Phytonanotechnology and Pharmaceutical Sciences

Original Article : Open Access

Physicochemical evaluation of endocarp of *Cocos nucifera* L.

Babita Aggarwal, Ajeet*, Shabnam Ain◆, Qurratul Ain*, R. K. Garg**, Pankaj Sharma***, H. S. Lamba**** and Archana Adhana*****

R. V. Northland Institute, Dadri, Greater Noida-II, G. B. Nagar-203202, Uttar Pradesh, India
** Sanskar College of Pharmacy and Research, Ghaziabad-201302, Uttar Pradesh, India*
*** K.D. Medical College, Mathura-281006, Uttar Pradesh, India*
**** Apex University, Jaipur-222001, Rajasthan, India*
***** H. R. Institute of Pharmacy, Ghaziabad-201001, Uttar Pradesh, India*
****** Bharat Institute of Technology, Meerut-250103, Uttar Pradesh, India*

Article Info

Article history
Received 15 July 2022
Revised 22 August 2022
Accepted 23 August 2022
Published Online 30 September 2022

Keywords
Cocos nucifera L.
Flavonoid
Phenolic
Physicochemical
Quality control

Abstract

The present study aimed to establish the quality control parameters of *Cocos nucifera* L. endocarp. Various parameters, which include, ash value, extractive values, foaming index, florescence analysis, preliminary phytochemical investigation, determination of total phenolic, tannin content and flavonoid content were carried out. Ash value, cold extractive value, hot extractive value, moisture content, foaming index and swelling index were estimated. The coarsely powdered air-dried material showed fluorescence when observed in different solvents. Preliminary phytochemical screening showed the presence of tannins, flavonoids, glycosides and steroids. Total phenolic and flavonoid content in alcoholic (A) and hydroalcoholic (HA) extract were found to be (A= 8.10 ± 0.011; HA = 8.19 ± 0.013) and (A = 3.45 ± 0.04; HA = 3.86 ± 0.05), respectively. This study provides the scientific data for the proper identification and establishment of standards for the use of *C. nucifera* endocarp which shall be extracted for whatever pharmacological activity, a scientist is working for; no pharmacological activity has been evaluated in this work.

1. Introduction

From ancient time, plants are used as medicine to maintain the human health as well as to treat the various diseases (Upadhya *et al, 2012*). It is estimated that about 25% of all modern medicines are directly or indirectly derived from higher plants (Calixto *et al*., 2000). In Ayurveda, either single drug or polyherbal formulations of natural origin are extensively used for the safe and effective treatment of diseases (Mukharjee, 2003). Plants, animals and microorganisms are also major natural source of medicinal compounds in current pharmacopoeias (Kingston *et al*., 2011). However, there are large number of plants, which have not been mentioned in these reports, in spite of their usage in the traditional and folk medicinal systems and required to be standardized in order to maintain the quality and safety of polyherbal formulations to attain the desired therapeutic effect (Sharma *et al*., 2009). The standardization includes the external (macroscopy/microscopy) as well as internal examination/ash values, extractive values and many other parameters to identify, authentify and study its chemical composition (WHO, 1998). Standardization also assures safety, efficacy, quality, and acceptability of the polyherbal formulations by reducing batch to batch variation (Ahmad *et al.*, 2006). These standards are based on pharmacognostical, physicochemical, phytochemical, and other biological parameters.

Corresponding author: Dr. Shabnam Ain
Professor and Head, Sanskar College of Pharmacy and Research (SCPR) Ghaziabad-201302, Uttar Pradesh, India
E-mail: shabnam.ain@sanskar.org
Tel.: +91-9310807567

Copyright © 2022 Ukaaz Publications. All rights reserved.
Email: ukaaz@yahoo.com; Website: www.ukaazpublications.com

Cocos nucifera L. (family Arecaceae), commonly known as coconut, is considered as an important fruit crop in tropical countries. Fruits of *C. nucifera* have long been used in the traditional medicine for the treatment of metabolic disorders.

Coconuts are unique in terms of their fruit (a drupe) morphology. The most interesting feature of the fruit is its wall. The fruit wall comprises of three layers, exocarp, mesocarp and endocarp. Due to extensive cross linking between phenolics, lignin and polysaccharides, the mesocarp becomes hard and fibrous (Chatterjee *et al*., 2003). The mesocarp, called husk is processed into rope, carpets, geotextiless and growing media. The endocarp, hard brown shell can be processed into very high quality activated charcoal (Pardesh *et al*., 2012).

Endocarp of *C. nucifera* was supposed to be the hardest part of the its fruit, but richest source of phenolic and flavonoid content and possess vasorelaxant, antihypertensive, antimicrobial, antidiabetic and inhibitory effect on oral microflora. On heating, the coconut shell gives oil that is used against ringworm infections in the popular medicine of India (Venkataraman *et al.*, 1980).

2. Materials and Methods

2.1 Plant material

The plant part endocarp (shell) of *C. nucifera* was collected in April 2016 from local area, Delhi. The plant material is authenticated by Principal Scientist, Dr. Roshni, National Bureau of Plant Genetic Resources (NBPGR), Pusa, Delhi. The voucher specimen (NHCP/NBPGR/2016-5/5634) was preserved in NBPGR and the crude drug

sample was deposited in the Department of Pharmacognosy, School of Pharmaceutical Sciences, Jaipur National University, Jaipur, Rajasthan, India for future reference. Endocarp was dried under shade, reduced to moderate coarse powder and stored in air tight container.

2.2 Physicochemical analysis

2.2.1 Foreign matter

The drug sample to be examined was weighed (2 g) and spread on a white tile uniformly without overlapping. The foreign matter was separated manually and examined in daylight with unaided eye. The suspected particles were transferred into a petri dish. After complete separation, the weight of the foreign matter was taken, and the percentage (%) (w/w) was determined.

2.2.2 Moisture content

About 5 g of drug after accurately weighing was placed in a tared (The weight of a container or wrapper that is deducted from the gross weight to obtain net weight) evaporating dish and was dried at 105°C. The drying and weighing were continued at 1 h intervals until the difference between two successive weighing was not more than 0.25%. A constant weight was supposed to have reached when two consecutive weighing after drying for 30 min and cooling for 30 min in a desiccator, showed not more than 0.01 g difference (Khandelwal, 2002; Trease *et al*., 2002).

2.2.3 Ash values

2.2.3.1 Total ash

About 2.0 g of the crude drug was accurately weighed and incinerated in a silica crucible at a temperature not exceeding 450°C (Sintering furnace :Model 3K) until free from carbon. The resulting ash was then cooled and weighed. The procedure was repeated to obtain a constant weight. The percentage of total ash with reference to the air-dried drug was finally calculated.

2.2.3.2 Acid insoluble ash

To the crucible containing total ash, 25 ml of dilute hydrochloric acid was added. The insoluble matter was collected on an ash less filter paper. It was then washed with hot water until it became neutral and ignited to a constant weight. The residue was allowed to cool in a suitable desiccator for 30 min, and it was immediately weighed. The procedure was repeated to obtain a constant weight. The percentage of acid-insoluble ash with reference to the air-dried drug was finally calculated.

2.2.3.3 Water soluble ash

To the crucible containing total ash, 25 ml of water was added and boiled for 5 min. The insoluble matter was collected on an ashless filter paper. It was then washed with hot water and ignited for 15 min at a temperature not exceeding 450°C. The procedure was repeated to obtain a constant weight. The difference in the weight of ash and weight of insoluble matter was calculated. The percentage of water-soluble ash with reference to the air-dried drug was finally determined (Khandelwal, 2002; Trease *et al*., 2002).

2.2.4 Cold extractive values

Extractive values for the plant drug sample were determined using different solvents, that is, petroleum ether, chloroform, methanol, water and 1:1 mixture of water and ethanol. For determination of cold extractive value, the air-dried coarse drug powder (4 g), accurately weighed in a glass-stoppered conical flask, was macerated with 100 ml of solvent shaken frequently for 6 h, and then allowed to stand for 18 h. It was then filtered rapidly taking precautions against loss of the solvent. 25 ml of the filtrate was evaporated to dryness in a tarred flat bottomed swallowed dish, dried at 105°C and weighed. The percentage of extractive values was calculated with reference to the air-dried drug.

2.2.5 Hot extractive values

The coarsely powdered air-dried material, accurately weighed (4 g), was placed in a glass stoppered conical flask. 100 ml of specified solvent (petroleum ether, chloroform, methanol, water and 1:1 mixture of water and ethanol individually) was added. After shaking well, it was allowed to stand for 1h, which was then gently heated in a reflux condenser for 1h, cooled and weighed. Shaken well and filtered rapidly. 25 ml of the filtrate was evaporated to dryness in a tarred flat bottomed swallowed dish, dried at 105°C and weighed. The percentage of extractive values was calculated with reference to the air-dried drug (Khandelwal, 2002; Trease *et al*., 2002).

2.2.6 Foaming index

About 1 g of the plant material was reduced to a coarse powder, weighed accurately, and transferred to a 500 ml conical flask containing 100 ml of boiling water. It was maintained at moderate boiling for 30 min, cooled, and filtered into a 100 ml volumetric flask. Sufficient water was added through the filter to dilute the filtrate to make up the volume. The decoction was poured into 10 stoppered test tubes in successive portions, that is, 1 ml, 2 ml, 3 ml, *etc*., and the volume of liquid in each test tube was adjusted with water to 10 ml. The tubes were stoppered and shaken in a lengthwise motion for 15 s at the rate of two shakes per second and then allowed to stand for 15 min. The height of the foam was measured, and the result was calculated (Mukherjee, 2005).

2.2.7 Fluorescence analysis

A small quantity of dried and finely powdered crude drug was placed on a grease-free clean microscopic slide and the same was treated with 1-2 drops of the freshly prepared reagent solutions separately, that is, 5% sodium hydroxide in water, ammonia, conc. H_2SO_4, conc. HCl and conc. HNO_3, *etc*. The added reagents were mixed by gentle tilting the slides and waited for 1-2 min. Then, each slide was placed inside the UV chamber and viewed in natural and ultraviolet (254 nm and 365 nm) lights. Powdered drug was subjected to fluorescence analysis. The colors observed by application of different reagents were recorded (Kumar *et al*., 2012).

2.3 Preliminary phytochemical investigation

Preliminary phytochemical screening was performed using standard procedures. The extracts obtained from different solvents were subjected to identification tests for the detection of various organic phytoconstituents such as alkaloids, glycosides, saponins, flavonoids, tannins, and steroids. All the concern tests were performed as per the standard procedures and their results were within the stipulated limits which can be accessed as per the requirement on personal request.

2.4 Total Phenolic content

Total phenolic content (TPC) in alcoholic and hydroalcoholic extract was determined by the Folin-Ciocalteu method (Kaur *et al.*, 2002) and gallic acid as standard. Brieûy, 0.5 ml of crude extract (1 mg/ml) were made up to 10 ml with distilled water, mixed thoroughly with 1.5 ml of Folin-Ciocalteu reagent for 5 min, followed by the addition 4 ml of 20% (w/v) sodium carbonate. The mixture was allowed to stand for further 30 min at room temperature. The absorbance was measured at 765 nm using a UV-VIS spectrophotometer (UNICO SQ4802E double beam). The total phenolic content is expressed as milligrams of gallic acid equivalent (GAE) to per gram of dry extract.

2.5 Tannin content

The tannins were determined by Folin-Ciocalteu method (Tambe *et al.*, 2014). About 0.1 ml of the sample extract was added to a volumetric flask (10 ml) containing 7.5 ml of distilled water and 0.5 ml of Folin-Ciocalteu phenol reagent, 1 ml of 35 % Na_2CO_3 solution and dilute to 10 ml with distilled water. The mixture was shaken well and kept at room temperature for 30 min. A set of reference standard solutions of tannic acid (20, 40, 60, 80 and 100 µg/ml) were prepared in the same manner as described earlier. Absorbance for test and standard solutions were measured against the blank at 725 nm with an UV/Visible spectrophoto meter (UNICO SQ4802E double beam). The tannin content was expressed in terms of mg of tannic acid equivalents per gram of dry extract.

2.6 Total flavonoid content

Flavonoid content in the examined plant extracts (alcoholic and hydroalcoholic) was determined using spectrophotometric method using auercetin (gift sample from S. R. Labs, Jaipur, Rajasthan) as standard (Quettier *et al.*, 2000). The sample contained 0.5 ml of methanolic solution of the extracts in the concentration of 1 mg/ml and 0.5 ml of 2% $AlCl_3$ solution dissolved in methanol. The samples were incubated for an hour at room temperature. The absorbance was determined using spectrophotometer at λmax=415 nm. The samples were prepared in triplicate for each analysis and the mean value of absorbance was obtained. The same procedure was repeated for the standard solution of quercetin and the calibration line was constructed. The content of flavonoids in extracts was expressed in terms of quercetin equivalent (mg of QU/g of extract).

3. Results

3.1 Physicochemical analysis

The results for physicochemical parameters such as moisture content, total ash value, cold and hot extractive values are summarized in Table 1. Total ash value found to be 0.513 ± 0.018, which is much greater than the acid insoluble ash (0.019 ± 0.011). Further, moisture content and foaming index were found to be 9.192 ± 0.128% and less than 100. Cold extractive values for solvents, that is, petroleum ether, chloroform, methanol, water and 1:1 mixture of ethanol and water were found to be 0.239 ± 0.029, 0.288 ± 0.009, 2.939 ± 0.030, 3.861 ± 0.366 and 2.053 ± 0.039, respectively. Hot extractive values using reflux method was also determined for solvents, that is, petroleum ether, chloroform, methanol, water and 1:1 mixture of ethanol and water. The values were found to be 1.408 ± 0.221, 2.405 ± 0.037, 4.454 ± 0.054, 5.188 ± 0.116 and 3.085 ± 0.015, respectively.

Table 1: Physicochemical values of *C. nucifera* endocarp

Parameter		Results
Foreign matter (%)		0.1
Moisture content (%)		9.192 ± 0.128
Ash values (% w/w)	Total Ash	0.513 ± 0.018
	Acid insoluble ash	0.019 ± 0.011
	Water soluble ash	0.347 ± 0.011
Cold extractive value (% w/w)	Per ether soluble extractive	0.239 ± 0.029
	Chloroform Soluble extractive	0.288 ± 0.009
	Alcohol soluble extractive	2.939 ± 0.030
	Water soluble extractive	3.861 ± 0.366
	Hydroalcoholic soluble extractive	2.053 ± 0.039
Hot extractive value (% w/w)	Per ether soluble extractive	1.408 ± 0.221
	Chloroform Soluble extractive	2.405 ± 0.037
	Alcohol soluble extractive	4.454 ± 0.054
	Water soluble extractive	5.188 ± 0.116
	Hydroalcoholic soluble extractive	3.085 ± 0.015
Foaming index	Less than 100	

Each value represents the mean ± SEM, N = 3.

3.2 Florescence Analysis

The result for florescence analysis was given in Table 2.

Table 2: Florescence analysis of *C. nucifera* endocarp

S. No.	Powdered drug/treatment	Observation under UV light		
		Ordinary light	UV short WL (254 nm)	UV long WL (365 nm)
1.	Powdered drug as such	Brown	Brown	Brown
2.	Powdered drug + distilled water	Brown	Brown	Dark brown
3.	Powdered drug + 5% NaOH	Black	Green	Dark Green
4.	Powdered drug + NH_3	Brownish red	Brownish red	Black
5.	Powdered drug + conc. H_2SO_4	Black	Dark greenish black	Dark green
6.	Powdered drug + conc. HCl	Light green	Green	Dark green
7.	Powdered drug + conc. HNO_3	Brown	Florescent yellow	Green
8.	Powdered drug + 5% Iodine	Light red	Red	Dark red
9.	Powdered drug + 5% $Fecl_3$	Reddish green	Red	Dark red
10.	Powdered drug + Picric acid	Yellowish green	Green	Dark green

Table 3: Phytochemical analysis of *C. nucifera* endocarp

Chemical group	Name of the extract				
	Petroleum ether	Chloroform	Ethanol	Water	Hydroalcohol
Alkaloids	-	-	-	-	-
Flavonoids	-	-	-	-	+
Tannins	-	-	+	-	+
Glycosides	-	-	-	+	+
Steroids	-	-	-	-	+
Carbohydrates	-	-	+	+	+
Proteins	-	-	+	-	-

3.3 Phytochemical analysis

Preliminary phytochemical screening of petroleum ether, chloroform, methanol, water and hydroalcoholic extracts showed the presence of flavonoids, tannins, glycosides and steroids (Table 3).

3.4 Total phenolic content

Total phenolic content for hydroalcoholic extract was found to be 8.19 ± 0.13 which is slightly greater than alcoholic extract, which was found to be 8.10 ± 0.011. Results are expressed as gallic acid equivalents per dry gram of extract (Table 4).

3.5 Tannin content

Tannin content for alcoholic and hydroalcoholic extract of endocarp was determined using tannic acid as standard. Hydroalcoholic extract showed much greater value 4.74 ± 0.016 as compared to alcoholic extract 4.47 ± 0.07 (Table 4).

3.6 Total flavonoid content

Flavonoid content of alcoholic and hydroalcoholic extract was found to be 3.45 ± 0.04 and 3.86 ± 0.05, respectively (Table 4).

Table 4: Total phenolic, tannin and flavonoid content of endocarp extract

S. No.	Name of the extract	Phenolic content	Tannin content	Flavonoid content
1	Alcoholic (A) extract	8.10 ± 0.011	4.47 ± 0.07	3.45 ± 0.04
2.	Hydroalcoholic (HA) extract	8.19 ± 0.13	4.74 ± 0.016	3.86 ± 0.05

Each value represents the mean ± SEM, N=3.

4. Discussion

In the present study, preliminary phytochemical screening and various physicochemical parameters such as ash values, extractive values, and moisture content of endocarp (shell) of *C. nucifera* were established. These parameters are important tools for determination of the identity, quality, and purity of the drug. Total ash indicates the presence of inorganic salts such as phosphates, carbonates, and silicates of sodium, potassium, magnesium, calcium, *etc*., and found

to be highest 0.513 ± 0.0118, followed by water soluble and acid insoluble ash 0.019 ± 0.011 and 0.347 ± 0.011, respectively. Moisture content is a major factor responsible for drug deterioration as moisture in conjunction with a suitable temperature causes activation of enzymes and provides a suitable condition to the proliferation of micro-organisms. Moisture content was within the specified limits; which was not observed for more than 0.01 gm as difference. Extractive values and florescence analysis indicates the presence of secondary metabolites (major secondary metabolites found were phenolic compounds in both the extracts) and various chemical entities which was further ascertained by phytochemical investigation. Considerable amount of total phenolic and flavonoid content was also detected which may contribute for the antioxidant activity of the plant part. Various parameters used are important for drug evaluation and helps in assuring the purity and quality of the drug and also provide a simple, cheap and reliable method for authenticating the drug.

Conflict of interest

The authors declare no conflicts of interest relevant to this article.

References

Ahmad, I.; Aqil, F. and Owais, M. (2006). Herbal medicine: Prospects and constraints in modern phytomedicine: Turning Medicinal Plants into Drugs, **384**:59-77.

Anonymous. (1998). Quality Control Methods for Medicinal Plant materials. Geneva: Office of the Publications, World Health Organization, pp:8-46.

Calixto, J.B., (2000). Efficacy, safety, quality control, marketing and regulatory guidelines for herbal medicines (Phytotherapeutic agents). Braz. J. Med. Biol. Res., **33**:179-89.

Chatterjee, A. and Pakrashi, S. (2003). The Treatise of Indian Medicinal Plants. New Delhi: NISCAIR.

Kaur, C. and Kapoor, H.C. (2002). Antioxidant activity and total phenolic content of some Asian vegetables. Int. J. Food Sci. Technol., **37**:153-161.

Khandelwal, K.R. (2002). Practical Pharmacognosy Techniques and Experiments. New Delhi: Nirali Prakashan, pp:15-163.

Kingston, D.G. (2011). Modern natural products drug discovery and its relevance to biodiversity conservation. J. Nat. Prod., **74**:496-511.

Kokate, C.K. (20005). Practical Pharmacognosy. 1st ed. New Delhi: Vallabh Prakashan, pp:111.

Kumar, D.; Gupta, J.; Kumar, S.; Arya, R.; Kumar, T. and Gupta, A. (2012). Asian Pac. J. Trop. Biomed., **2**:6-10.

Mukharjee, P.K. (2003). GMP for Botanicals, Regulatory and Quality Issues on Phytomedicines. 1st ed. New Delhi: Business Horizons, pp:219.

Mukherjee, P.K. (2005). Quality Control of Herbal Drugs. New Delhi: Business Horizons, pp:247.

Prades, A.; Dorneir, M.; Diop, N. and Pain, J.P. (2012). Coconut water uses, composition and properties: A review. Fruits, **67(2):**87-107.

Quettier, D.C.; Gressier, B.; Vasseur, J.; Dine, T.; Brunet, C. and Luyckx, M.C. (2000). J. Ethnopharmacol., **72**:35-42.

Tambe, V.D. and Bhambar, R.S. (2014). Res. and Rev. J. Pharmacog. Phytochem., **2(4)**,41-47.

Trease, G.E. and Evans, W.C. (2002). Pharmacognosy. 15th ed. New Delhi: A Division of Reed Elsevier India Private Limited, pp:95-105.

Upadhya, V.; Hegde, H.V.; Bhat, S.; Hurkadale, P.J.; Kholkute, S.D. and Hegde, G.R. (2012). Ethnomedicinal plants used to treat bone fracture from North-Central Western Ghats of India. J. Ethnopharmacol.,**14**:557-562.

Venkataraman, S.; Ramanujam, T.R. and Venkatasubbu, V.S. (1980). Antifungal activity of the alcoholic extract of coconut shell - *Cocos nucifera* Linn. J. Ethnopharmacol., **2**:291-293.

WHO. (1998). Quality Control Methods for Medicinal Plant Materials, World Health Organization, Geneva.

Citation **Babita Aggarwal, Ajeet, Shabnam Ain, Qurratul Ain, R. K. Garg, Pankaj Sharma, H. S. Lamba and Archana Adhana (2022). Physicochemical evaluation of endocarp of *Cocos nucifera* L. J. Phytonanotech. Pharmaceut. Sci., 2(3):14-18. http://dx.doi.org/10.54085/jpps.2022.2.3.3**