Comparative in Vitro Antioxidant Efficiency and Physicochemical Study of *Cocos Nucifera* and *Palmyra Palm* Fruit Endosperm Tissues

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Abstract

Antioxidant activity of the plant to indicate the therapeutic activity of the plants, physicochemical studies in helpful in shaping dependability and clarity of the medicine and also these values are significant quantitative standards. This research is to investigate the antioxidant activity, physicochemical studies and identification of new theraputetic compounds in cocos nucifera endosperm and Palmyra fruit endosperm ethanolic solvents extracts. Antioxidant efficiency of proportion radical scavenging efficiency of the test sample calculates by using DPPH technique. Physico-chemical property of ethanol extracts of cocos nucifera endosperm and Palmyra fruit endosperm were determined by following WHO standards procedure and values are recorded. Identification of the new compounds by using TLC/HPTLC method. Antioxidant activity of the two test samples, cocos nucifera endosperm extracts shows more antioxidant activity than Palmyra fruit endosperm ethanolic extracts of DPPH radical scavenging activity. The present examination exposes standardization which contains acid insoluble ash, water soluble extractive value, total ash, and Moisture content of cocos nucifera endosperm and Palmyra fruit endosperm ethanolic solvent extracts. Total ash value is valuable in shaping dependability and purity of the medicine and also these values are significant quantitative standards. Total ash values of substance show the quantity of natural resources and earthy material present in the examination material. Diagnostic result shows the total ash higher values was 1.25%w/w in ethanol extract of cocos nucifera endosperm. Percent mass loss on drying or moisture substance of ethanol cocos nucifera endosperm and Palmyra fruit endosperm extracts is 11.52 and 7.72%. The smaller amount of moisture content could reduce fungal and bacterial improvement. Palmyra fruit endosperm ethanolic extracts values are associate cocos nucifera endosperm extracts. The high quantity of acid-insoluble siliceous substance in attendance in the extracts of cocos nucifera endosperm was 0.25% w/w. The extractive values water soluble indicates the occurrence of carbohydrate, inorganic and acids substance. The ethanolic cocos nucifera extract have high value of water soluble extractive values. Antioxidant activity of the two test samples, Cocos nucifera endosperm extract shows more antioxidant activity than palmyra fruit endosperm ethanolic extracts of DPPH radical scavenging activity. Identification of

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new compounds in Cocos nucifera endosperm tissues three major band observed between the Rf value of 0.35, 0.85 and 0.92. Palmyra fruit endosperm tissues two major band observed between the Rf value of 0.33 and 0.93 under TLC plate.

Keywords: Physiochemical analysis, Cocos nucifera endosperm, loss on drying, Palmyra fruit endosperm.
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Introduction

Cocos nucifera is a main kind tree belong to the own circle of relatives Arecaceae (palm)[1]. The acquainted call of Cocos nucifera is coconut or coconut palm. General names in exceptional area Burmese (on, mak-un); Bengali (narikel); Duthc English(coconut palm, coconut); Malay (kelapa); Thai (ma phrao); Trade call (coconut); Tamil (tennai-maram); Vietnamese (dua)[2].

The cocos nucifera fruit comprise expcarp is the skinny outmost layer (or pores and skin) of the fruit. The mesocarp is a impenetrable husk collected of coarse brown fibres (coir). The endocarp is the hard, however noticeably skinny woody internal layer of a fruit that comprise the endosperm. The endosperm is in component liquid ("coconut water") and in part steady (fibrous white coconut "flesh" or "meat", which adhere to the inner wall of the endocarp). The liquor part of the endosperm is rich in mineral, Lauric (fatty acid-based) oils, vitamins and carbohydrates[3,4]. The little embryo (or seed) is bounded with within the stable fleshy part of the endosperm simply inside one of the germination pores (referred to as "eyes") thru which the radicle (the embryonic root of the seedling) emerge whilst the embryo germinates. From the unconventional survey plant life of cocos nucifera has mighty healing fee at the regions of anti bacterial, antioxidant and anti caner[5,6].

Scientific name of Palmyra fruit is Borassus flabellifer, native from Indian subcontinent and southeast asia, along with Nepal, india, Thailand Vietnam, malayis, bangaladesh, Burma, Indonesia, sri lanka, combodia. Genus call comes from the Greek phrase borassos which means the immature spadix of date palm[7,8]. Specific epithet from the Latin phrase flabellatus which means like an open fan. Tree sap, referred to as toddy, is tapped to be used as a beverage, for this reason the not unusualplace call of toddy palm. The candy sap of the Palmyra tree is referred to as Toddy and is utilized in training of Palm jiggery[9]. Leaves have been previously used as paper in Indonesia, for this reason the not unusualplace call of lontar palm. Sap is fermented to make Arrack that is an alcoholic beverage. The fan fashioned tall tree bears end result like that of a coconut tree. Almost each a part of the tree is beneficial to mankind. The pulp is soft and the husk is fibrous just like that during Coconut. The fruit has a black husk and is four to inches in diameter. It is borne in clusters. The pinnacle segment of the fruit is reduce off to expose the candy jelly seed sockets[10,11].

Palmyra end result are suitable for eating in all of the degrees and having a few treasured bioactivity[12]. The male and lady plant life are constantly produced in assorted plants. The plant life are small, and faded yellow developing in cluster with a white string like inflorescences. Fruits are sub globose, and once more in clusters. Usually a unmarried tree will generate everywhere among fifty five to 350 end result. The length of the end result variety from four-eight inches diameter, and are black, greenish white and black whilst ripe. The higher a part of the fruit should be reduce off to expose the candy jelly seed

sockets to eat. There is one to a maximum of 5 jelly sockets in a unmarried fruit even though it is maximum commonly discovered to have 3 sockets. The kernel that is smooth as jelly and translucent like ice is accompany with sweetish water[13,14]. This liquid has healing belongings and is utilized by the area people to deal with pores and skin diseases. The ripened fruit of outer layer additionally may be eaten uncooked or boiled. The sparkling end result are used as wheels for gambling through children. The goal of the present day look at is to estimate the antioxidant pastime and physico-chemical evaluation of Cocos nucifera endosperm and palmyra fruit endosperm ethanolic extracts[15,16].

Materials and Methods

All the chemicals and necessary solvents are of analytical grade and received in SD fine, HI-MEDIA chemicals are used. The in good physical shape and disease free fruit of cocos nucifera endosperm and Palmyra fruit endosperm is composed from south region of Coimbatore season of March 2022. The fresh fruit endosperm parts are wash with valve water and then rinse with distil water. Wash endosperm matter was air dehydrated out at atmospheric temperature for 5-6 days. Once entirely dehydrated, fruit parts were converted into fine particle using an electric processor. Fruits endosperm was store in a stopped up box at normal temperature until necessary to use[17].

Preparation of flowers raw extract

The crushed cocos nucifera endosperm and Palmyra fruit endosperm of every substance had been blend with adequate amount of solvent ethanol. It becomes saved in rotating shaking at 110 RPM for forty eight hours. At the give up of forty eight hours, every extract become clear out thru whatman number 1 clear out filter paper and the filtrate had been concerted at room temperature in an effort to condense the quantity. The adhesive like extract had been keep in pre weigh cap bottles and then given extracts had been weighted. These twist cap bottles had been saving in fridge at 4ºC for further destiny use. Each extract be converted into independently reconstitute the usage of smallest number of quantities of the extract solvent previous to use[18,19].

Determination of Antioxidant efficiency

Radical scavenging action DPPH

The Radical scavenging action DPPH reaction in between the antioxidant (H - A) and DPPH can be put in writing as following

DPPH+H-A → DPPH-A + A
(Purple) (Yellow)

The anti-oxidants be active in response by (1,2-diphenyl-2-picrylhydrazyl), it is a continuous free radicals and is condensed to the DPPH-H and the outcome of the DPPH radical decrease in its absorbance. The quantities of decolouration indicate that scavenging activity is achievable in the compound or the plant extract in conditions of hydrogen donate capacity[20].

Procedure

The antioxidant activity of the test sample of cocos nucifera endosperm and Palmyra fruit endosperm ethanolic extract was concluding of hydrogen donate capability by means of the constant radical of DPPH. The cocos nucifera endosperm and Palmyra fruit endosperm test sample extracts at different concentrations like 100-500 µg/mL were occupied and the volume was used to to 100µl with ethanol. Methanolic solvent solution (5 mL of 0.1mM) of the DPPH● was additional and legitimate to stand up for 20 minutes at 27ºC. The the test extracts absorbance was determined at 517nm. Standard ascorbic acid different concentration like 10 to 50 µg/ml was used. Minor the absorbance of the free radical chemical reaction combination indicate upper radical free scavenging action[21,22]. The analysis was performing in triplicate and reported in table 1. Proportion radical scavenging action of the experiment test sample was calculating bye using the below maths formula.

%DPPH = (Control OD - Sample OD / Control OD) ×100

Physicochemical studies

Determination of Total Ash Value

Weigh an empty silica crucible along with lid, formerly ignited for one hour. Transfer 1 gm of test substance into the silica crucible and cover the crucible with cover and for a second time weigh it. Ignite the crucible for 3 hrs approximately 450^0C high temperature[23]. Ignite for one hour period where ignition to incessant weight is indicated. Upon the achievement of the each ignition cover up the silica crucible and permit it to cold in a desiccator to room temperature earlier than weigh.

$$\text{Ash Value} = \frac{\text{Residue weight x 100}}{\text{Sample weight}}$$

Determination of acid insoluble ash content

Ash sample are obtain from the above total ash then it was heated with 25 ml of 2N HCl boiled for a two minutes. Filtered passed through ashless Whatman ashless filter paper. The ashless filter paper was transfer in to a tarred silica crucible. Incinerate at 450ºC in a high temperature muffle furnace until remove all the carbon[24]. The silica crucible is reduce the temperature cooled and weighed. The following formula used to acid insoluble ash percentage was calculated.

$$\text{Determination of acid insoluble ash} = \frac{\text{Weight of the acid insoluble residue} \times 100}{\text{Sample weight}}$$

Water insoluble ash determination

Ash obtains from the above test total ash was boil through heating with 25 mL of purifying water for a two minutes and then filter an ashless filter paper. The ashless filter paper was transfer into silica crucible. Incinerate at 450ºC in a high temperature muffle furnace until remove all the carbon[25]. The silica crucible then reduces the heat then cooled to room temperature and weighed. Proportion was calculated for water soluble ash.

Water soluble extractive value determination

Test sample of 1 gm each air dehydrated extracts sample was taken with 110 mL of $CHCl_3$ water in a clogged flask for 24 hours trembling regularly through the first 6 hours and then legitimate to stand up for 18 hours. After that if was filter rapidly attractive protection next to loss of the water. The 25ml leftovers were evaporate to waterlessness in a flat bottomed dish dried at 105ºC weigh. The quantity of the water solvable extractive value was calculated following formula.

$$\text{\% of water soluble extractive} = \frac{\text{Weight of the extract} \times 100 \times 100}{25 \times \text{ Weight of the sample taken}}$$

Alcohol soluble Extractive value determination

Identical path of action was do again as that of determination of water soluble extractive value. The transform was made by replace water with absolute alcohol (aprox 95%)

Determination of Moisture content

1g of the test sample was positioned in a pre-weighed flat china dish; dry in the furnace at 100ºC±5 ºC till the stable weight was obtain. The loss of weight was calculated.

$$\text{Moisture Content} = \frac{\text{Weight loss x 100}}{\text{Sample weight}}$$

physicochemical studies like P^H, water soluble extractive value, acid insoluble ash, water insoluble ash, total ash, alcohol soluble extractive values and moisture content at 105ºC were carry out as per the WHO procedure for individual fruit alcoholic extract of cocos nucifera endosperm and palmyra fruit endosperm[26]. physico-chemical

property of ethanol extracts of cocos nucifera endosperm and palmyra fruit endosperm were determined by following WHO standards procedure and the values are recorded in the table 2.

Identification of New Compounds by TLC/HPTLC

Sample preparation

Transfer about 70 ml of the sample into a separating funnel. Add 150 ml of petroleum ether and shake well for about 10 minutes. Allow the upper and lower layers to divide. Slowly transfer the lower layer in to another separating funnel and add 70 ml of petroleum ether. Shake well for about ten minutes and permit the layer to separate. Drain off the lower layer. Remove the upper layer obtain from both separation into a hygienic and dry beaker. Evaporate the remains to waterlessness by placing on a water bathe[27]. Add 1 ml of methanol to the remains and dissolve.

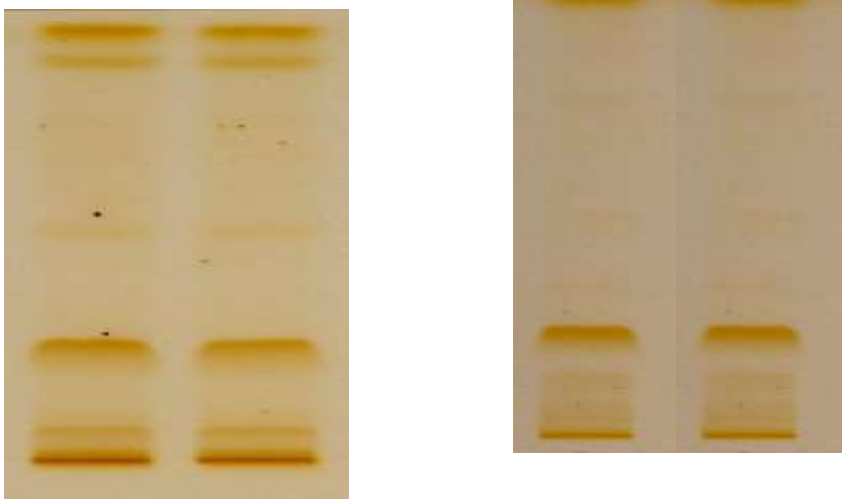
Chromatographic system

TLC plate type : Pre coated thin layer silica plate 60, F254 20×20 cm
Mobile phase : 90 ml Dichloromethane + 10 ml Methanol
Spotting volume : 20 µl

Procedure

Spot 20 µl of both Cocos nucifera endosperm tissues profiles and palmyra fruit endosperm tissues solution using suitable spotter on the TLC plate. Develop the plate up to 16 cm in the mobile phase, which is pre saturated for 2 hours. Air-dry the plate. Visualize the dried plate under TLC visualizer. Capture the image of the plate under light compare the Cocos nucifera endosperm Tissues profiles with the Palmyra fruit Endosperm Tissues spot figure 1.

Figure 1. a) Cocos nucifera Endosperm Tissues b) Palmyra fruit Endosperm Tissues



Results and Discussion

Antioxidant efficiency

The free radical scavenging capacity DPPH of cocos nucifera endosperm and Palmyra fruit endosperm values are tabulated in table 1, Figure2. Free radical scavenging is normally used to examination the scavenging action of more than a few natural products[28]. This free scavenging efficiency of extract could be connected to the usual of phenolic, thus contribute to their electron donates capability. The outcome shows that healing vegetation has considerable special property on free radical scavenging efficiency. The enhanced scavenging DPPH may be associated to the greater phenolic contents[29]. The decrease the DPPH radical associate with the hydroxyl groups on the antioxidant fragment, so the antioxidant efficiency might employ he delocalization of an electron into the para substitute hydroxyl group on the molecule previous to the contribution of a second

hydrogen decrease DPPH which also depend on the steadiness and response prospective of the molecular structure [30]. The ethanol extracts cocos nucifera endosperm and Palmyra fruit endosperm DPPH radical scavenging activity was originate to be equivalent and efficiency is similar to ascorbic acid pattern.

Table 1. R**adical scavenging competence Cocos nucifera endosperm and Palmyra fruit endosperm**

S.No	Sample Concentration (µg)	Inhibition of flowers extracts activity (%)		Concentration of Ascorbic acid standard (µg)	Ascorbic acid % inhibition Standard
		Cocos nucifera	palmyra fruit		
1	100	22.01±0.11	19.08±0.02	10	61.13±0.13
2	200	34.32±0.37	22.01±0.12	20	72.25±0.11
3	300	41.41±0.20	29.02±0.13	30	81.26±0.21
4	400	57.31±0.21	35.12±0.02	40	92.15±0.10
5	500	66.20±0.15	45.11±0.10	50	100.04±0.03
6	IC50 (µg/mL)	120.30±0.02	245.251±0.09	IC50 (µg/mL)	11.21±0.05

* Values are earnings of three independent investigation± standard deviation (n=3)

Radical scavenging efficiency DPPH of Cocos nucifera endosperm extracts

Radical scavenging efficiency DPPH of cocos nucifera endosperm ethanoic extracts are offered in table 1. The ethanoic cocos nucifera endosperm fruit extract at 100-500 µg/ml exhibit 22.01±0.11, 34.32±0.37, 41.41±0.20, 57.31±0.21 and 66.20±0.15%. DPPH action was found to be similar to standard ascorbic acid different concentring of 10,20,30,40, and 50 µg/ml exhibited 61.13±0.13, 72.25±0.11, 81.26±0.21, 92.15±0.10 and 100.04±0.03% inhibition shows figure 2.

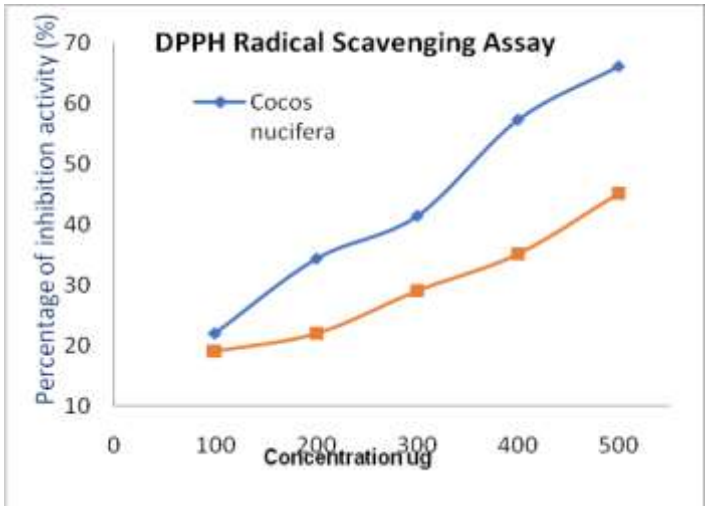


Figure 2. Comparision of DPPH Radical Scavenging Assay

Radical scavenging efficiency DPPH of palmrya fruit endosperm extracts

Radical scavenging efficiency DPPH of palmrya fruit endosperm ethanolic extracs are offered in table 1, figure 3. The sample ethanolic palmrya fruit endosperm fruit extract at 100-500 µg/ml shows 19.08±0.02, 22.01±0.12, 29.02±0.13, 31.12±0.02 and 43.11±0.10%.

DPPH efficiency was initiate to be similar to standard ascorbic acid concentration of 10-50 µg/ml exhibited 61.13±0.13, 72.25±0.11, 81.26±0.21, 92.15±0.10 and 100.04±0.03% inhibition. Comparison of the two extracts having less IC_{50} value it is representing that the extract has more antioxidant ability. Amongst the two samples cocos nucifera endosperm extracts having more antioxidant capacity then palmyra fruit endosperm extracts.

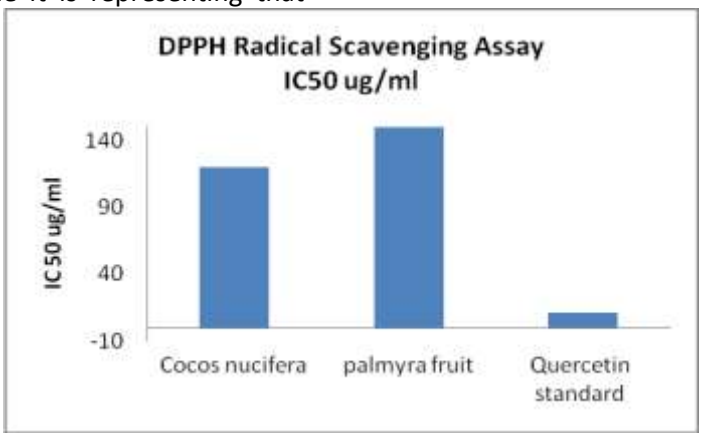


Figure 3. DPPH radical scavenging activity IC_{50} Values comparative

Physio-Chemical parameters

Results of quantitative investigation for water insoluble ash, acid insoluble ash, alcohol soluble extractive, water soluble extractive, total ash and humidity content at 105ºC values are tabulated in table 2.

Table 2. Physiochemical analysis of ethanolic extracts of Cocos nucifera endosperm and palmyra fruit endosperm

S.No	Test Parameters	Cocos nucifera endosperm %	palmyra fruit endosperm %
1	Odour	Pleasant	Pleasant
2	pH	5.7	6.32
3	Total ash value (w/w %)	1.25	0.27
4	Acid insoluble ash (w/w %)	0.26	0.18
5	Water insoluble extrative (w/w %)	0.58	0.43
6	Water soluble extractive (w/w %)	16.33	11.12
7	Alcohol soluble extrative (w/w %)	61.43	42.95
8	Loss on drying (w/w %)	11.52	7.72

Total ash value is beneficial in responsible and limpidness of medicine and also these value are significant quantitative standards. Total ash value of cocos nucifera endosperm and palmyra fruit endosperm value of 1.25% and 0.27%. The total ash value of plant subsance indicates the quantity of natural resources and earthy resources occurrence in the plant green materials. Investigative result show the total ash substances are in superior value was 1.25% w/w in extract of ethanolic cocos nucifera endosperm. Percent mass loss on

drying content of ethanoic extracts cocos nucifera endosperm and palmyra fruit endosperm is 11.52% and 7.72%. The moisture content values if less it could inhibit bacterial, fungal and micro-organism growth. Palmyra fruit endosperm of ethanoic extracts having less value associate cocos nucifera endosperm extracts. The elevated amount of values of acid-insoluble siliceous material in attendance in the cocos nucifera endosperm extracts was 0.26%w/w. The extractive values of water soluble indicate the occurrence of sugar, acids and inorganic compound. The ethanolic cocos nucifera endosperm extract have high value of water soluble extractive values.

Identification of New Compounds by TLC/HPTLC

Identification of new compounds in Cocos nucifera endosperm tissues three major band observed between the Rf value of 0.35, 0.85 and 0.92. Palmyra fruit endosperm tissues two major band observed between the Rf value of 0.33 and 0.93 under TLC plate.

Conclusion

The present examination exposes standardization of total ash value is beneficial in responsible and purity of medicine and also these values are significant quantitative standards. The total ash values of cocos nucifera endosperm and palmyra fruit endosperm value of 1.25% and 0.27%. Total ash value of plant subsance indicates the quantity of natural resources and earthy resources in attendance in the plant green materials. Analytical result show the total ash substance is in higher value was 1.25% w/w in ethanolic extract of cocos nucifera endosperm. Percent weight moisture content of ethanoic extracts cocos nucifera endosperm and palmyra fruit endosperm is 11.52% and 7.72%. The smaller amount of moisture content could inhibit micro-organism, bacterial and fungal growth. Palmyra fruit endosperm of ethanoic extracts having less value associate cocos nucifera endosperm extracts. The upper quantity of acid-insoluble siliceous material in attendance in the cocos nucifera endosperm extracts was 0.26% w/w. The extractive value of water soluble indicates the occurrence of sugar, acids and inorganic compounds. The ethnolic cocos nucifera endosperm extract have higher value of water soluble extractive values. Antioxidant activity of the two samples, Cocos nucifera endosperm extract shows good antioxidant activity than palmyra fruit endosperm ethanolic extracts of DPPH radical scavenging activity. Identification of new compounds in Cocos nucifera endosperm tissues three major band observed between the Rf value of 0.35, 0.85 and 0.92. Palmyra fruit endosperm tissues two major band observed between the Rf value of 0.33 and 0.93 under TLC plate.

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