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Screening the Antioxidant activity of Thamarai Magarantha Podi Karpam

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Abstract

The Siddha System of Medicine is a Traditional Medical System, which Provides “Holistic Health”. In which one of the entity is *Kayakarpam*. *Kayakarpam* plays a major role in Preventing the ageing process and by treating various types of communicable and Non-communicable diseases. Thamarai magarandha podi is a herbal Kayakarpam indicated for treating infertility and impotency. It is formulated by collecting the stamens from the flower of plant of *Nelumbo nucifera*. The main Aim of the study is to screen the Antioxidant Activity of Thamaraimagarantha podi karpam by DPPH, ABTS, Nitric oxide radical, Superoxide radical scavenging activity. The result shows, Thamarai magarantha podi karpam has significant free radicals scavenging properties.

Keywords: *Kayakarpam*, *Thamarai magaranthapodi*, *Nelumbu nucifera*, Antioxidant activity, Radical scavenging property.

Introduction

The Siddha science is a traditional curative as well as preventive system of medicine and was developed in the flourished culture of Dravidians. It was invented by the Siddhar’sthrough their high intellectual, supernatural and spiritual powers. The Siddhar’s achieved spiritual power (siddhi). This achievement was related to their discipline of mind and its superiority over the body, and was accomplished through both yoga and “Karpam” medicine (rejuvenating medicines). Thus Siddhar’s became the symbols of psychosomatic perfection and so the Siddha system of medicine became a combination of medicine and yoga.

There are so many Siddhar's, out of them eighteen Siddhar's are the most important and they all have more knowledge about the universe and its contents. Siddhar's believes that, there is a connection between the celestial bodies of our universe to the living beings of earth. Any changes in the external world, brings changes to human beings. So they believe a healthy body is essential to attain eternal life!!!

Apart from that, Siddhar's invented the Siddha pharmacopeia; they describe the plants, minerals, metals, animal products and methodology of medicinal preparations with these materials.

The Siddha System of Medicine is a Traditional Medical System, which provides "Holistic Health". The system provides preventive, promotive, curative, rejuvenative, rehabilitative health care with scientific and holistic approach. The word Siddha is derived from the root word "Siddhi", which means attaining perfection, eternal bliss and accomplishment. The Siddha system comprises essentially of philosophical concepts including the three main components: Medical practice, Yogic practice and wisdom.

Siddha medicine (Marunthu) is classified into two types. They are 32 types of Internal Medicine and 32 types of External medicines. Of these 32 types of Internal Medicine KarpamMarunthu is one among them.

"மறுப்பதுடல்நோய்மருந்தெனலாகும்
மறுப்பதுளநோய்மருந்தெனல்சசாலும்
மறுப்பதிணிநோய்வாராதிருக்க
மறுப்பதுசாவையுமருந்தெனலாமே"
-திருமூலர்திருமந்திரம்.

The word kaya Karpam means (Kayam- body, Karpam–Kallpolakkuthal) to make our body competent and youthful. There are two ways is available to prevent ageing, disease and death i.e., Yogam and Kaya Karpam.

"கற்பத்தையுண்டால்காயம்அழியாது
கற்பத்தினாலேகாணலாம்கைலையை
கற்பத்தினாலேகாணலாம்சோதியை
கற்பத்தினாலேகாலையுங்கட்டிடே".
-திருமூலர்திருமந்திரம்.

Karpam Medicine is formulated to prevent ageing, disease and to cure disease. Karpam Marunthu (Rejuvenation) has rich antioxidant activity.

"அஞ்சுயுகத்திலழியாமற்காய்ந்தான்
மிஞ்சியகற்பம்விளம்பினோம்நூற்றெட்டுத்
தஞ்சமுறவேதான்தின்னவல்லோர்க்குப்
பஞ்சுநரைபோய்ப்பதிந்தோங்கிவாழ்வரே."
-திருமூலர்(திருமூலர் வைத்தியபகுதி)

According to siddha system of medicine, the human body is sustained by the appropriate ratio of Vatham, Pitham, Kabam. These three humors are made of five elements of nature. They are Earth, Water, Fire, Air and space. Whenever there is a alteration in the ratio of Vatham, Pitham and Kabam the body gets diseased.

Kayakarpam plays major role in preventing ageing process and by treating various types of communicable and non-communicable diseases. Therefore, to achieve eternal life, both body and mind must be nourished. Kayakarpam such as Pranayamam and Yogam helps in rejuvenating ting the body along with our mind and soul. Henceforth, Kayakarpam plays a vital role in combating degenerative disease that occurring due to senility and also helps in treating and preventing many lively diseases.

Kayakarpam Medicine (Rejuvenation Therapy) has rich antioxidant activity[4]. (Antioxidant compounds are capable of protecting against oxidative damage by decreasing the number of free radicals).

Aim and objectives

Aim

- To screen the antioxidant activity of *"Thamarai magarantha podi karpam"* in different methods.

Objective

- To prove Siddha Karpamarunthu has Antioxidant activity.
- To prove Karpamarunthu / Drug rich in antioxidant activity has rejuvenates and delays ageing.

Literature review

தாமரைமகரந்த பொடி

"சண்டனையுஞ்சண்டனையுந்தள்ள
மலருள்ளுறையுண்
சண்டனையுஞ்சண்டனையுஞ்சார்ம
திபால் - சண்டனையும்
வையாராயுண்மறையவைகுவார்
வாகடத்தை
வையாராயுண்மறையவா."
-தேரன்வெண்பா

Gunapadam review

வேறுபெயர்:-அரவிந்தம், எல்லிமனை, சூரியநட்பு,பொன்மனை, விந்தம், புண்டரீகம், பதுமம், கமலம், நளினம், முளரி, முண்டகம், மாலுத்தி, சரோகம், கோகனகம், இண்டை, கஞ்சம், அப்புசம், அம்போருகம், சலசம், வனசம், வாரிசம், சரசீருகம், பங்கேருகம், சரோருகம், பங்கசம்.

Botanical name: *Nelumbu nucifera*

Vernacular names:

- Eng : The sacred Lotus
- Tel : Tamara
- Mal : Aravindam
- Kan : Tavare
- Sans : Pankaja
- Arab : Nilufer
- Pers : Nifure

இதுநீரில் வளருங்கொடி, இந்தியாவில் குளங்குட்டைகளில் செழித்து வளருகின்றது. இப்பூவின் நிறவேற்றுமையைக் கொண்டு இதை வெண்மை,செம்மை, நீலம், மஞ்சள் என, நான்கு இனங்களாகப் பிரிக்கலாம். இதன் பூவிற்கு நறுமணமுண்டு, இப்பூவில் இலட்சுமிவாசஞ்செய்கிறாள் என்னும் நம்பிக்கை இந்துக்களுக்கிருப்பதால், இப்பூ மிகவும் போற்றப்படுகிறது.
ப-உ: பூ, விரை, பூத்தாள், கிழங்கு.

Organoleptic actions:

- சுவை (Taste) –இனிப்பு(Sweet), துவர்ப்பு (Astringent)
- தன்மை (Character) –சீதம்(Cold)
- பிரிவு (Potency)–இனிப்பு(Sweet)

செய்கை (Therapeutic actions):

- குளிர்ச்சியுண்டாக்கி-சீதளகாரி-Cooling
- துவர்ப்பி-ஸங்கோசனகாரி-Astringent
- கோழையகற்றி-கபஹாகாரி-Expectorant
- தாதுவெப்பகற்றி-சமனகாரி-Sedative

கிழங்கு

- உள்ளழலாற்றி-அந்தர்ஸ்நிக்தகாரி-Demulcent

விதை

- உரமாக்கி-வன்மைஉண்டாக்கி-Tonic
- உடலுரமாக்கி-போஷணகாரி-Nutrient

Botanical Aspect - Literature Review

Taxonomy:

Kingdom: Plantae -Plants
Subkingdom: Tracheobionta – Vascular plants
Superdivision: Spermatophyta – Seed plants
Division; Magnoliophyta – Flowering plants
Class: Magnoliopsida – Dicotyledons
Subclass: Magnoliidae
Order: Proteales
Family: Nymphaeaceae /Nelumbonaceae – Lotus-lily
Genus: Nelumbo Adans. – Lotus
Species: nucifera Gaertn. – Sacred lotus

Ecology:

Although historically the genus Nelumbo was consideredto be closely related to Nymphaeales, new systematic work has allied Nelumbo with the lower eudicots, particularly Platanus. Worldwide, there are only two species of Nelumbo. *N. Lutea Willd*. (Synonyms: *N. Pentapetala* (Walter) Fernald and Nelumbiumluteum Willd.) And *N.Nucifera* (synonyms: *N.Speciosa* Willd, Nelumbium speciosum Willd, Nelumbium *N. Druce* and Nymphaea N. L*) N.Nucifera* Gaertn , theIndian or sacred lotus, is found throughout Asia and Australia, whereas N. Lutea, the American lotus or water chinquapin, occurs in eastern and southern North America. *N. Luteais* considered to be a subspecies of *N. Nucifera*. In India, *N. Nucifera*, commonly known as lotus, kamala or padma, isan aquatic species, requiring plenty of space and full sun in order to thrive. It has stout, creeping, yellow rhizomes and green fruits. The leaves are enormous, reaching 2 feet India meter. There are two varieties of 'kamala': one has white flowers and is commonly called 'Pundarika' or 'Svetakamala'; the other has pink or reddish-pink flowers and is called 'Raktakamala'. The whole plant with flowers is known as 'Padmini', the rhizomes as 'Kamalkand', the tender leaves as 'Sambartika', the peduncle as 'Mrinal' or 'visa', the stamens as 'Kirijalaka', the torus as 'Padmakosa', the seed as 'Karnika' or 'Padmaksya', and the honey formed in the flowers by the bees feeding upon Padma is known as 'Makaranda' or 'Padma- Madhu'.

The plant is often cultivated for its elegant sweet scented flowers, which are the national flower of India. Almost all parts of lotus are eaten as a vegetable, consumed all over the world, especially in South-east Asia, Russia and some countries in Africa. It is used not only as an ornamental plant and dietary staple, but also as a medicinal herb in Eastern Asia, particularly in China. *N. Nucifera* has been cultivated as a crop in Far-East Asia for more than 3000 years, where it was used for food and medicine and played a significant role in religious and cultural activities. Almost all parts of *N.Nucifera* are marketed; the rhizome holds the largest share.

Fruit and seeds

The fruit of this plant is an aggregate of indehiscent nutlets. Ripenutlets are ovoid, roundish or oblongs, up to 1.0cmlong and 1.5 cm broad, with a hard, smooth, brownish or greyish black pericarp which is faintly longitudinally striated, pedunculated and single seeded. Seeds fill in the ripe carpel. The seeds are sold as a vegetable in Indian markets, under the name of 'Kamal Gatta'.

Leaves

The leaves are large and orbicular, 20–90 cm in diameter and non-wettable. Leaves are of two types: aerial and floating, and are petiolated and entirely glaucous. The aerial leaves arecup shaped whereas the floating leaves are flat. The petioles of the aerial leaves are erect, smooth, greenish or greenish brown in color with small brown dots and are sometimes rough. The aerial leaves are usually 24–33 cm in length, and the floating leaves 23–30 cm. Odor is distinct; fractures are fibrous. The young leaves are eaten as vegetables and used in traditional medicine.

Flowers

The flowers are solitary, large, 10–25 cm in diameter, white, pink or pinkish white, fragrant and have peduncles arising from the nodes of the rhizome, and 1–2 cm long sheathing at the base. The sepals, petals and stamens are spirally arranged, passing gradually one into another.

Rhizome

The rhizomes are 60–140 cm long, 0.5–2.5 cm in diameter, yellowish white to yellowish brown, smooth, with longitudinal striations and brown patches, and with nodes and internodes. Transverse section of the rhizome shows an outer layer of epidermis, surrounded by cuticle followed by a dense sub-epidermal layer, a spongy layer and an inner dense layer, continuous with the parenchyma cells. When freshly cut, the rhizome exudes mucilaginous juice and shows a few large cavities surrounded by several larger ones. Fracture is tough and fibrous, and the odor is indistinct.

Calyx

Sepals 4, polysepalous, first and second antero-posterior and other two are lateral. Often considered as perianth; small, triangular waxy, greenish pink, imbricate.

Corolla

Petals numerous, polypetalous, ovate, waxy, pink, spirally arranged.

Androecium

Stamens indefinite, spirally arranged, filament long, slender, anther long, basifixed, erect, introrse, connective protudes beyond the anther as an appendage.

Gynoecium

Polycarpellary, apocarpous, superior, carpels embedded at the top of a flattened spongy receptacle, each carpel unilocular, one ovuled, ovule pendulous and style very short stigma flat.

Materials and Methods

Drug profile:

A. Drug selection and reference:

Thamarai magaranda podi karpam is a classical siddha herbal medicine mentioned in *Theraiyar Yamaga Venba.*

B. Ingredient:

Thamarai magaranda podi (*Nelumbo nucifera*) - Required Quantity (350gm used in this study)

C. Raw drug collection and authentication:

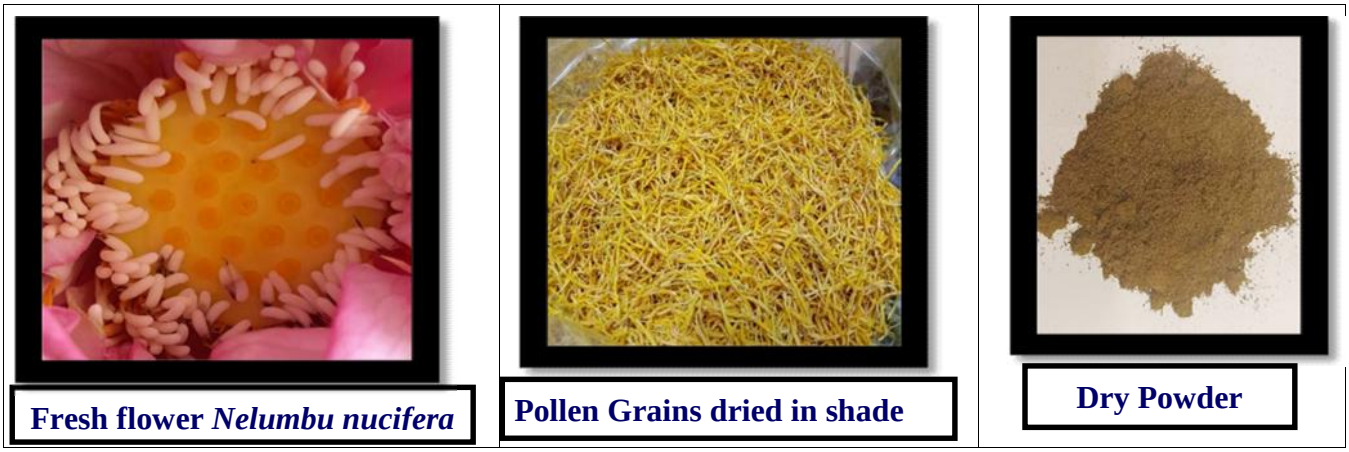
The raw drug were collected from the local market, Koyambedu in Chennai. The drug were identified and authenticated from the Gunapadam Department experts, Govt Siddha Medical College, Chennai.

Purification of herb

After collection, it would be cleaned thoroughly and washed well with fresh water. Then it was allowed to dry completely in the shade for 2 weeks.

Preparation of Thamarai magaranda podi

The cleaned and dried *Thamarai Magaranda Podi* would be made into a fine powder (Chooranam) and sieved with a white cloth (Vasthirakayam).



Fresh flower *Nelumbu nucifera*

Pollen Grains dried in shade

Dry Powder

Evaluation of bioactive constituents

1. Estimation of total phenolic content
2. Determination of total flavonoid content

Estimation of total phenolic content

Total phenolic content was determined using gallic acid as the reference standard. One milliliter of the sample (0.2-1 mg/mL) was added with 0.5 mL of FolinCiocalteaue reagent and incubated at room temperature for 10 minutes. Furthermore, 2.5 mL of saturated Na2CO3 solution was added and incubated at room temperature for 30 minutes. The resultant color was measured at 750 nm versus a blank containing distilled water and FolinCiocalteau reagent. CONTROL as 2.316.

Total phenolic content was expressed in gallic acid equivalents using the equation

T = C X V / M

(T = Total Phenolic Content (mg/g) of extract as gallic acid equivalents,
C =Concentration of Gallic acid established from the calibration curve,
V = Volume of the extract solution in ml,
M = weight of extract in grams).

Determination of total flavonoid content:

Total flavonoid content was determined with slight modifications using quercetin as the reference standard. One milliliter of sample (0.2-1 mg/mL) was added with 0.5 mL of 1.2% aluminum chloride in 10% methanol, 0.5 mL of 1M potassium acetate, and made up to 3 mL with distilled water. The mixture was incubated for 30 minutes in the dark, and the absorbance was read at 415 nm. Aluminum chloride without the sample alone served as the blank. Total flavonoid content was expressed in gram quercetin equivalents. CONTROL AS 1.145

Antioxidant assay

The aqueous flower's pollen extracts of *Nelumbo nucifera* were subjected to the following methods of antioxidant assay.

1. DPPH radical scavenging assay
2. Nitric acid radical inhibition assay
3. Total iron reducing power assay
4. Superoxide radical scavenging activity by alkaline DSMO method.

DPPH radical scavenging assay

0.2 mg/ml of Drugwas used for diphenyl-picrylhydrazyl (DPPH) radical assay, with some modifications, and a final concentration range of 20-100 µg/ml was used for the assay. The sample was made up to 1 ml with 95% methanol followed by which 1 ml of 0.2 mM DPPH was added and incubated in the dark for 30 min. The purple color developed was read spectrophotometrically at 515 nm. Ascorbic acid (20 µg-100 µg/ml) was used as standard and 95% methanol alone was used as blank. A reaction mixture with 95% methanol and DPPH alone was used as a control(1.591). The % Inhibition was calculated using the formula:

% I = (C – E) / C x 100

Whereas % I=% Inhibition; C=Absorbance of control; E=Absorbance of extract.

Nitric oxide radical inhibition assay

Nitric oxide was generated from sodium nitroprusside and measured by the Griess reaction. Sodium nitroprusside in aqueous solution at physiological pH spontaneously generates nitric oxide which interacts with oxygen to produce nitric ions that can be estimated using the Griess reagent. Scavengers of nitric oxide compete with oxygen leading to reduced production of nitric oxide. The reaction mixture (6 mL) contained sodium nitroprusside (10 mm, 4 mL), phosphate buffer saline (PBS, pH 7.4, 1 mL), and extract or standard (1 mL) in DMSO at various concentrations and it was incubated at 25 for 150 min. After incubation, 0.5 mL of the reaction mixture containing nitrite ion was removed, sulphanilic acid reagent was added (0.33% w/v, 1 mL), mixed well, and allowed to stand for 5 min for completion of diazotization.

Then, 1 mL of NEDD was added, mixed, and allowed to stand for 30 min in diffused light. A pink-colored chromophore was formed. The absorbance was measured at 540 nm. (CONTROL -1.323)

Total iron reducing power assay

A volume of 1 mL of the plant extract was mixed with 2.5 mL of phosphate buffer (0.2 M, pH 6.6) and 2.5 mL of 1% potassium ferricyanide. The reaction mixture was incubated at 50 for 20 min. After incubation, 2.5 mL of 10% trichloroacetic acid was added and the reaction mixture was centrifuged at 1000 rpm for 10 min. The upper 2.5 mL layer was mixed with 2.5 mL of deionized water and 0.5 mL of ferric chloride and thoroughly mixed. The absorbance was measured spectrophotometrically at 700 nm. A higher absorbance indicates a higher reducing power. (CONTROL-1.964)

Superoxide radical scavenging activity by alkaline DMSO method

In this method, superoxide radical is generated by the addition of sodium hydroxide to air saturated DMSO. The generated superoxide remains stable in solution and reduces nitroblue tetrazolium (NBT) into formazan dye at room temperature which can be measured at 560 nm. Briefly, 0.1ml of NBT (1 mg/ml) was added to the reaction mixture containing 1 ml of alkaline DMSO (1ml DMSO containing 5mM of NaoH in 0.1 ml water) and 0.3 ml of the extract in freshly distilled DMSO at various concentration, to give a final volume of 1.4mL. The absorbance was measured at 560nm.

Results

Evaluation of bioactive constituents

1. Estimation of total phenolic content

Table 1

S.No	Drug/Std concentration (µg/ml)	Std	Test 1	Test 2	Test 3
1	10	0.091	0.665	0.671	0.804
2	20	0.152	1.071	1.121	1.189
3	30	0.191	1.541	1.509	1.542
4	40	0.275	1.938	2.041	2.299
5	50	0.238	2.169	2.184	2.754

Total Phenolic Content expressed as 6.458 ±0.851mg/g gallic acid equivalents

2. Determination of Total flavonoid content:

Table 2:

S.No	Drug/Std concentration (µg/ml)	Std	Test 1	Test 2	Test 3
1	10	0.071	0.149	0.14	0.143
2	20	0.085	0.191	0.197	0.212
3	30	0.068	0.301	0.313	0.281
4	40	0.071	0.428	0.357	0.364
5	50	0.073	0.461	0.418	0.431

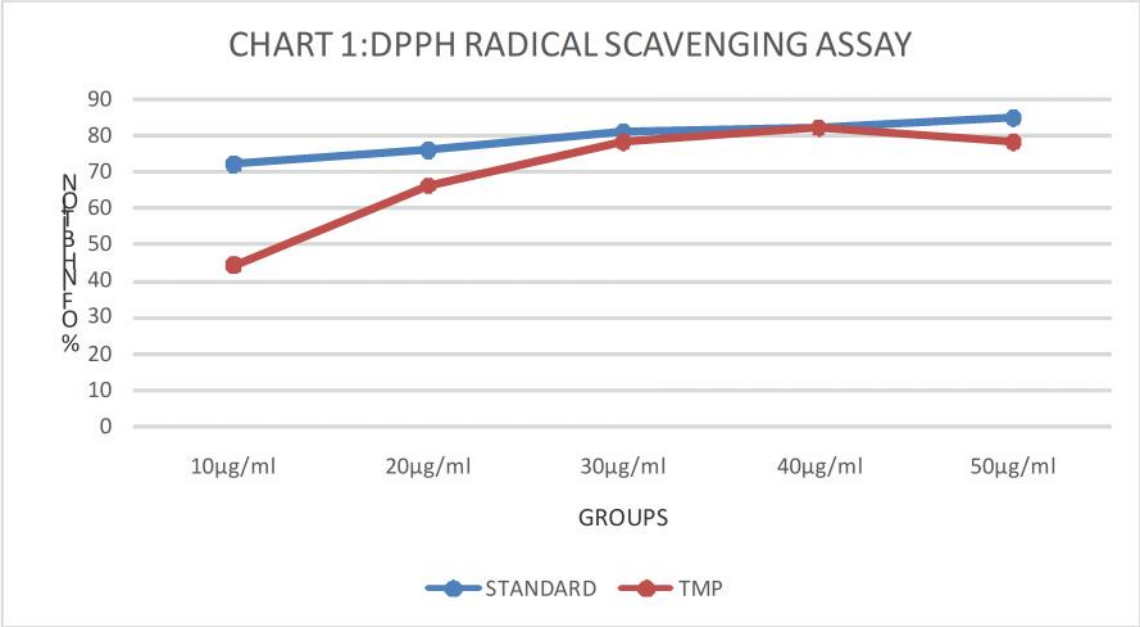
The total Flavonoid Content is expressed as 5.695±0.663mg/g quercetin equivalents

Antioxidant assay

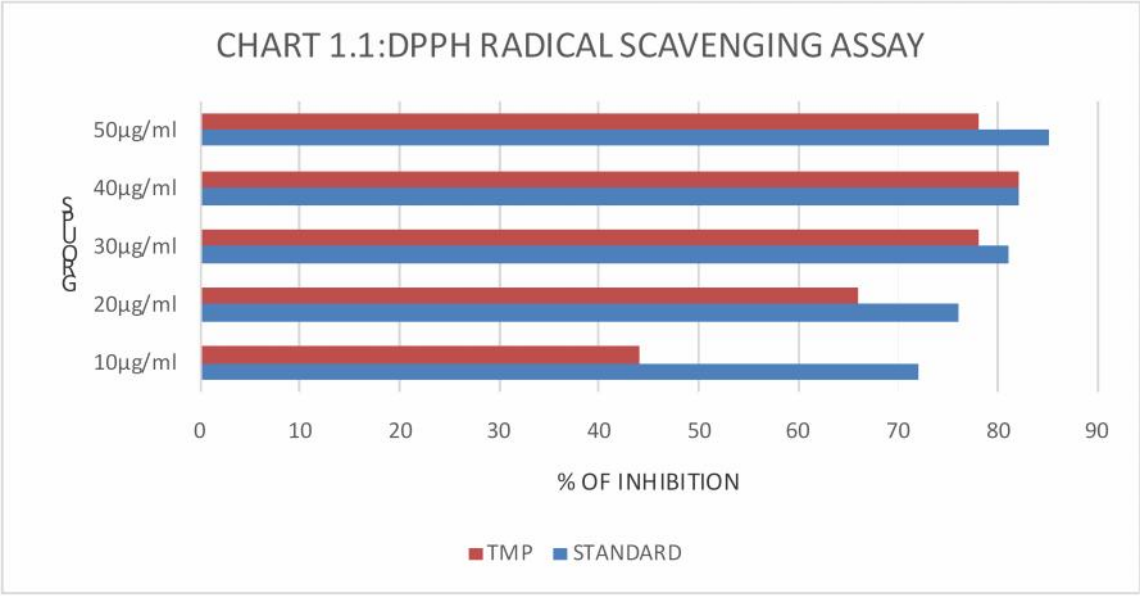
1) Percentage inhibition of test drug Thamarai magarantha podi on DPPH radical scavenging assay:

Table 3:

S.No	Drug conc (µg/ml)	Std	% of inhibition	Test 1	% of inhibition
1	10	0.327	72	0.888	44
2	20	0.285	76	0.529	66
3	30	0.228	81	0.349	78
4	40	0.219	82	0.285	82
5	50	0.179	85	0.342	78



***TMP-** Thamarai Magarantha Podi

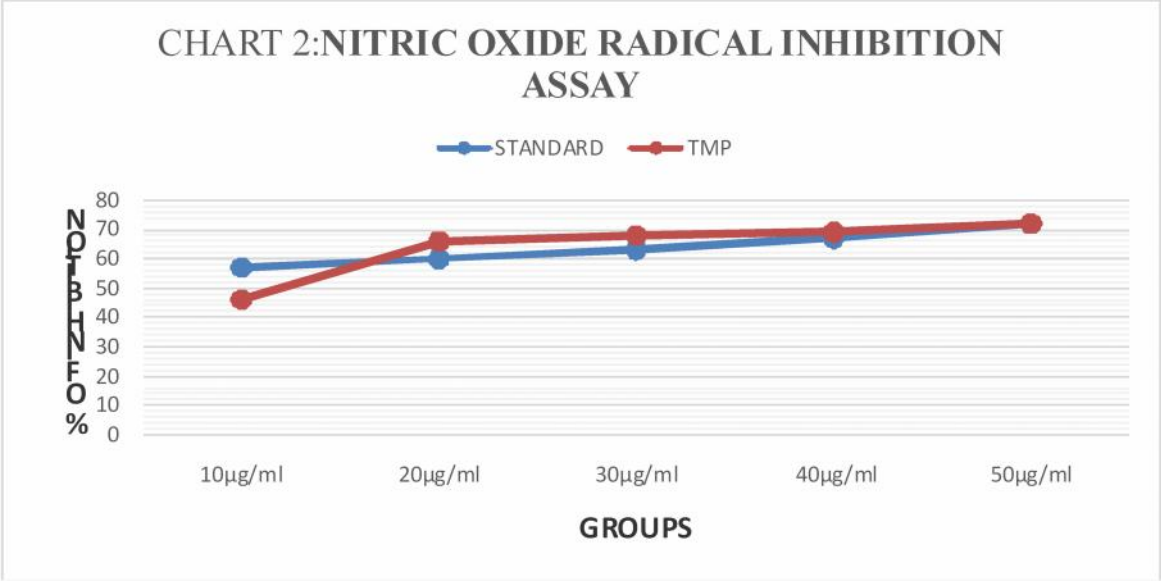


***TMP-** Thamarai Magarantha Podi

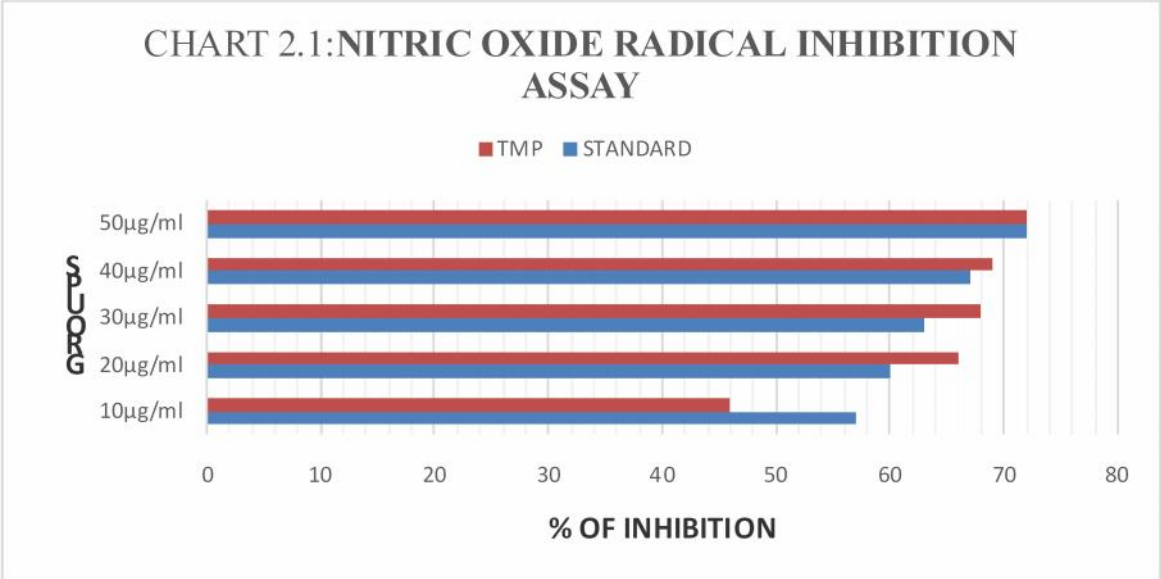
2) Percentage inhibition of test drug Thamarai magarantha podi on nitric oxide radical inhibition assay

Table 4

S.No	Drug conc (µg/ml)	Std	% of inhibition	Test 1	% of inhibition
1	10	0.825	57%	0.706	46
2	20	0.751	60%	0.448	66
3	30	0.694	63%	0.431	68
4	40	0.624	67%	0.407	69
5	50	0.526	72%	0.361	72



***TMP** – Thamarai Magarantha Podi

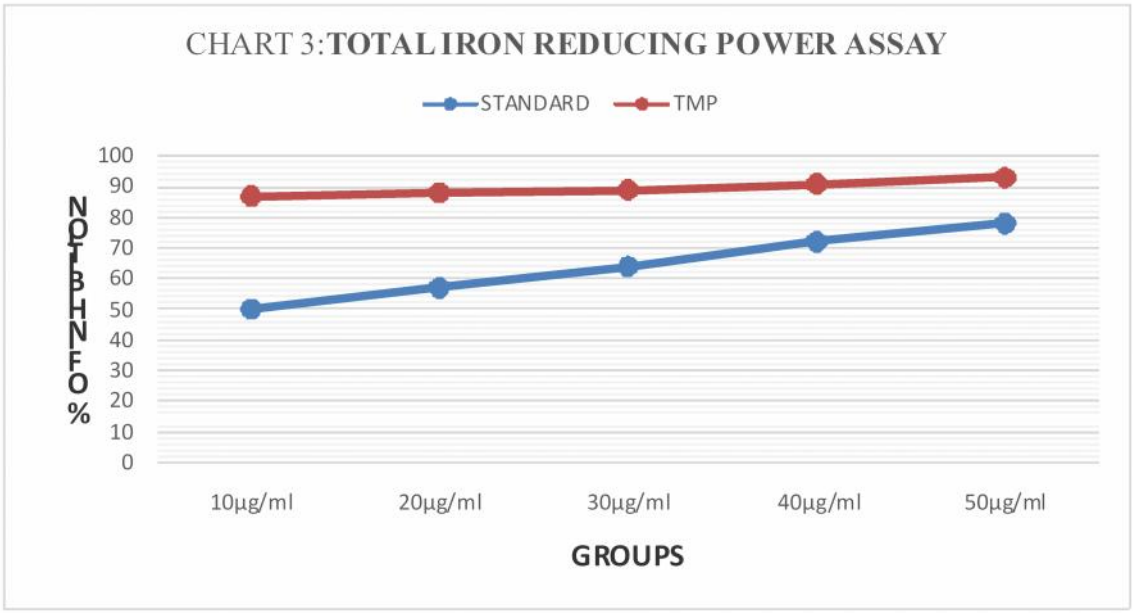


***TMP** – Thamarai Magarantha Podi

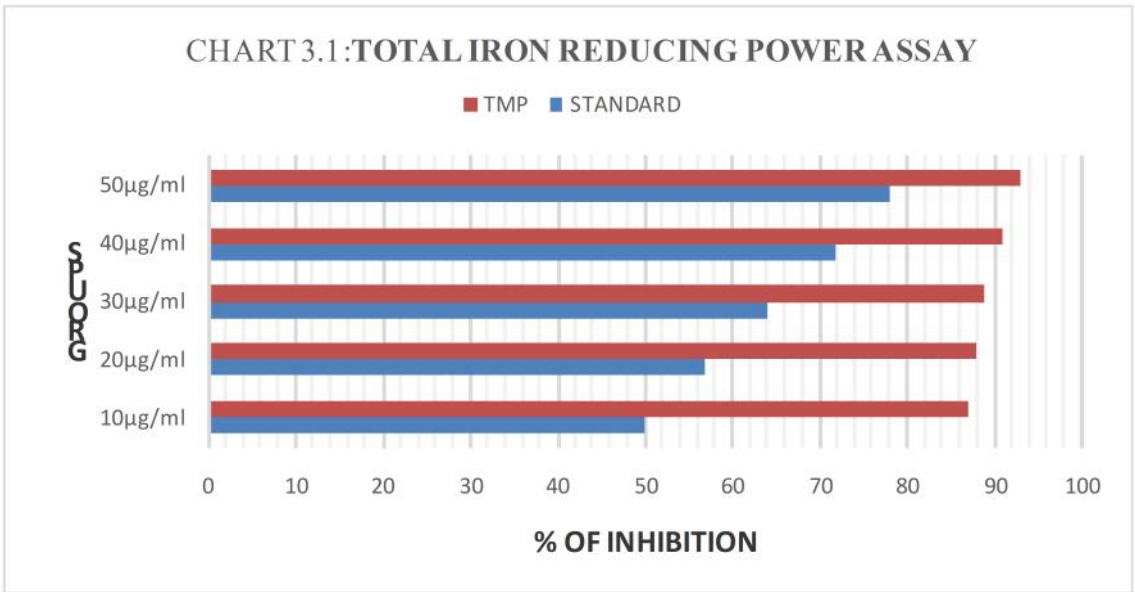
3) Percentage inhibition of test drug Thamarai magarantha podi on Total iron reducing power assay:

Table 5:

S.No	Drug conc (µg/ml)	Std	% of inhibition	Test 1	% of inhibition
1	10	0.977	50.25458248	0.244	87
2	20	0.843	57.07739308	0.231	88
3	30	0.689	64.9185336	0.202	89
4	40	0.539	72.55600815	0.18	91
5	50	0.421	78.56415479	0.151	93



***TMP** – Thamarai Magarantha Podi

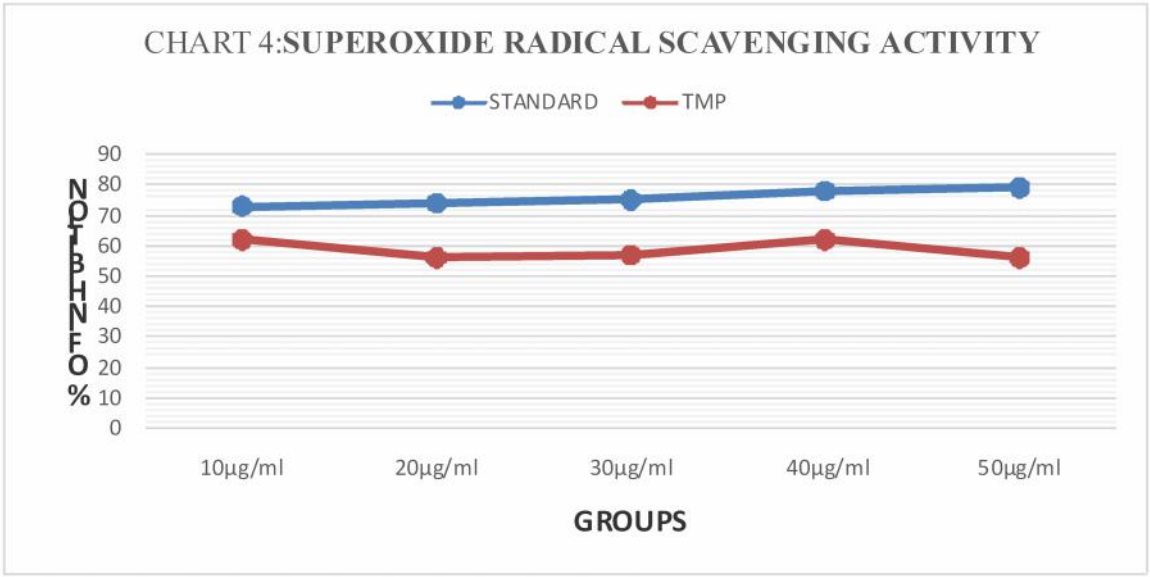


***TMP** – Thamarai Magarantha Podi

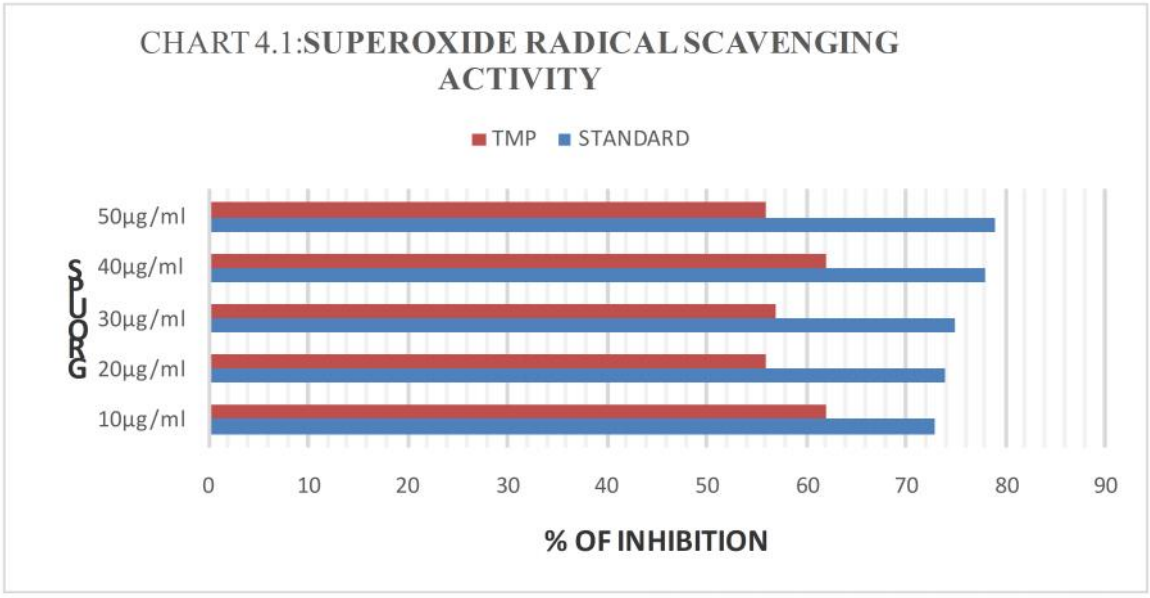
4) Percentage inhibition of test drug Thamarai magarantha podi on superoxide radical scavenging activity by alkaline DMSO method:

Table 6:

S.No	Drug conc (µg/ml)	Std	% of inhibition	Test 1	% of inhibition
1	25	0.857	73%	1.204	62
2	50	0.816	74%	1.406	56
3	75	0.782	75%	1.37	57
4	100	0.692	78%	1.204	62
5	125	0.667	79%	1.406	56



***TMP** – Thamarai Magarantha Podi



***TMP** – Thamarai Magarantha Podi

Discussion

The trial drug Thamarai Magarantha PodiKarpam was selected from the text Theraiyar yemaha venba for screening the antioxidant activity of the drug (In vitro Assay). The drug was prepared as per the procedure and subjected to various studies to reveal potency and effectiveness against the disease. Literary review about the ingredients of Thamarai Magarantha Podi Karpam from various text book gave hope about its activity. Literary review, which consists Botanical Aspect, Gunapadam Aspect and Pharmacological review which support this study.

DPPH Radical scavenging assay

In the present study, the extract of TMP was found to possess concentration dependent scavenging activity on DPPH radicals. The values of DPPH free radical scavenging activity of the TMP extract was given in (Table 3). The extract of TMP showed the highest DPPH scavenging activity (82%) at 40µg/ml and the lowest percentage of inhibition (44%) at 10µg/ml. Ascorbic acid (Standard) showed highest percentage of inhibition (85%) at 50µg/ml and the lowest percentage of inhibition (72%) at 10µg/ml. This indicated that % of inhibition increased with increase in concentration of both the standard and TMP extract. The TMP extract has more or less equal DPPH scavenging activity when compared to the standard. From the present study, it was concluded that the TMP extract has a marked antioxidant activity at higher concentrations.

Nitric oxide radical inhibition assay

The extract of TMP was found to possess concentration dependent scavenging activity onnitric oxideradicals. The values of NITRIC OXIDE radical scavenging activity of the TMP extract was given in (Table 4). The extract of TMP showed the highest NITRIC OXIDEs cavenging activity (72%) at 50µg/ml and the lowest percentage of inhibition (46%) at 10µg/ml. DMSO (Standard) showed highest percentage of inhibition (72%) at 50µg/ml and the lowest percentage of inhibition (57%) at 10µg/ml. This indicated that % of inhibition increased with increase in concentration of both the standard and TMP extract. The TMP extract has more or less equal NITRIC OXIDE scavenging activity when compared to the standard. From the present study, it was concluded that the TMP extract has a marked antioxidant activity at higher concentrations.

Total iron reducing power assay

The extract of TMP was found to possess concentration dependent total iron reducing power assay. The values of total iron reducing power assay of the TMP extract was given in (Table 5). The extract of TMP showed the highest total iron reducing power assay (93%) at 50µg/ml and the lowest percentage of inhibition (87%) at 10µg/ml. Standard showed highest percentage of inhibition (78%) at 50µg/ml and the lowest percentage of inhibition (50%) at 10µg/ml. This indicated that % of inhibition increased with increase in concentration of both the standard and TMP extract. The TMP extract has more or less equal total iron reducing power assay when compared to the standard. From the present study, it was concluded that the TMP extract has a marked antioxidant activity at higher concentrations.

Superoxide radical scavenging activity by alkaline DMSO method

The extract of Thamarai Magarantha Podi karpam was found to possess concentration dependent scavenging activity on nitric oxideradicals. The values of SUPEROXIDE radical scavenging activity of the TMP extract was given in (Table 6). The extract of TMP showed the highest SUPEROXIDE scavenging activity (62%) at 100µg/ml and the lowest percentage of inhibition (56%) at 50µg/ml. DMSO (Standard) showed highest percentage of inhibition (79%) at 125µg/ml and the lowest percentage of inhibition (73%) at 25µg/ml. This indicated that % of inhibition increased with increase in concentration of both the standard and TMP extract. The TMP extract has more or less equal SUPEROXIDE scavenging activity when compared to the standard. From the present study, it was concluded that the TMP extract has a marked antioxidant activity at lowest concentrations.

Antioxidant activity of trial drug Thamarai Magarantha Podi karpamwas found to be 44% to 82%, 46% to 72%, 87% to 93% for DPPH assay, Nitrous oxide assay and Total iron reducing

power assay respectively. When compared to Standard drug Ascorbic acid and DMSO they had more Antioxidant activity i.e., 72% to 85%, 57% to 72% and 50% to 78% respectively. Thus, trial drug had High antioxidant activity when compared to standard synthetic compound but it sure the trial drug also had antioxidant activity.

Thus, the drug had high antioxidant activity when compared to standard drug. Standard drug is single trial raw drugs. And dosage of the trial drug is approximately 5 – 10gm but here 10mg – 50mg of trial drug is tested but these amounts also had enough amount of antioxidant activity.

Conclusion

Based on the results obtained from the In-vitro anti-oxidant assay for the sample Thamarai Magarantha Podi karpamit was concluded that the siddha formulation Thamarai Magarantha Podi karpam has promising phenolic,flavonoid content andanti-oxidant activity in the estimated DPPH, Nitrous oxide and Total iron reducing power assay, Superoxide radical scavenging activity. Antioxidant substance prevent ageing and cell death which is similar to Karpamarunthu action as mentioned classical Siddha text. From the above results siddha text are correct based on scientific parameters, In future Pre-clinical and Clinical studies are done to evaluate their effectiveness and make strong evidence of Siddhar's science.

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