

Evaluation of phytochemical profile and antioxidant status of laboratory mode *Dasamoolarishtam* health tonic and a marketed product

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ABSTRACT

The mankind has been using the medicinal herbs in all the aspects of their life since time immemorial. Ayurvedic formulations are as old as the Vedic age (1500 BCE–500 BCE), recently a surge in preference of Ayurvedic medicines for their health care properties. *Dasamoolarishtam* is an Ayurvedic preparation which contains both plant and non-plant ingredients. The aim of the current study is to prepare plant only *Dasamoolarishtam* through the extraction with ethanol solvent (DET). This preparation was evaluated for phytochemical constituents and antioxidant analysis and is compared with commercially available market sample/traditional formulation (MKT). The DET showed better results in all the studies such as qualitative and quantitative analysis of secondary metabolites as well as *in vitro* antioxidant parameters such as 2,2-diphenyl-1-picrylhydrazyl, 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid, ferric reducing antioxidant power, superoxide radical scavenging, and phosphomolybdenum assays. The results of the present study open up a new approach in the scientific preparation/modification of traditional medicines. The overall findings of the current research corroborate and validate the phytochemical and antioxidant properties of DET and MKT samples and also propose the scope for modification of Ayurvedic drug for more efficient and quick response during the treatment.

Keywords: Antioxidant, Ayurveda, *Dasamoolarishtam*, Preparation, Secondary metabolites

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INTRODUCTION

India is a treasure house for traditional systems of medicines such as Ayurveda, Siddha, Unani, and Homeopathy which are being practiced since many hundreds of years. Ayurveda generally focuses on plant based medicines for the management of various diseases.^[1] Scientific validation of the traditional medicines are being attempted to trace clues regarding contributing factors in ameliorating debilitating disease conditions.^[2] Ayurvedic Formulary of India mentioned that there are 21 types of drug formulations which have been used in Ayurveda. Out of them 16 are mainly plant based and rest of them are mineral/metal product based formulations.^[3] Ayurvedic medicines are available in the form of powder, tablet, pill, liquid, and semi-solid which are classified into arishta, asava, rasa rasayan, lauha, bati, churna, avaleha, ghrita, parpati, taila, goggu, etc.^[4]

Dasamoolarishtam also spelled with *Dashamoolarishta*, *Dashmoolarishta*, *Dashmularishta*, *Dasamularista*,^[5-8] etc. *Dasamoolarishtam* is an Ayurvedic polyherbal formulation prepared in the form of fermented decoction. It consists of around 67 plants and non-plant ingredients such as water, jaggery (brown sugar), ghrita (ghee), honey, and kasthuri (gland of musk deer).^[9] This formulation contains 5–10% of self-generated alcohol.^[10] *Dasamoolarishtam* is commonly used for post-natal care to avoid secondary complications.^[11] It is used as digestant to treat all kind of indigestion problems and is a curative aid for stomach-ache.^[8,12,13] This drug is much useful in the treatment of long period Alzheimer's disease^[14] and hemiplegia.^[15] It helps to restore health and vitality and recover the normal physiological state from inflammatory conditions.^[5,6] This drug is a rejuvenator and revitalizer and commonly used as general health tonic.^[16,17] It decrease the effect of anemia and cardiac disorders^[18,19] and also a remedy to cure loss of sense of taste, emesis, malabsorption

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syndrome, abdominal lump, piles, excessive flow of urine and gravel in urine.^[20]

Dasamoolarishtam is known to possess various therapeutic actions against certain common ailments. Self-preparation and market samples of *Dasamoolarishtam* showed strong antiinflammatory, antioxidant, and antimicrobial properties.^[5,6,17,21-23] It also tested for oxidative stress, analgesic activity and postpartum evaluation.^[7,17,24,25] *Dasamoolarishtam* is a fermented decoction, which probably possess various active phytochemicals and pharmacological actions without queries. However, the modern phytomedicine research has proven that organic solvents have great potential in the extraction of many bioactive phytochemicals. In the past few decades, research findings of the medicinal plant research concluded that ethanol/methanol solvent has great ability to yield many phytochemicals

and that kind of extracts possess significant results in bioactivity studies. Addition of non-plant materials into herbal products is an ancient holistic approach. Now a days, sugar, honey and ghee are not used in novel medicinal preparations due to sociocultural preferences. At this outset of the present study is framed to analyze the phytochemical constituents and evaluate the antioxidant potential of ethanol extract of *Dasamoolarishtam* (DET) plants (mixture of powders). Further, the study compares the results with traditional market sample (MKT).

MATERIALS AND METHODS

Collection and Identification of *Dasamoolarishtam* Plants and Their Parts

Dasamoolarishtam preparation is generally based on *Sharangdhara Samhita Madhyama Khanda, Bhaishajyaratnavali*.^[9] The list of plants and the quantity of plant parts used in DET formulation was selected [Table 1] by the reference of various literatures.^[9,26-28] The plant materials were collected from different parts of Tamil Nadu (Siruvani, Thondamuthur, Coonoor, Kotagiri, Kil kotagiri, Sholur, etc.) and some of them are procured from local market (herbal shops) in Coimbatore city. The plant materials are identified in our laboratory with the help of various references.^[29-33] Traditionally using market sample (MKT) was procured from local herbal shops.

Extraction and Preparation of *Dasamoolarishtam*

The collected plant materials were washed under running tap water to remove all the surface pollutants. The washed samples were air dried under shade condition, then coarse powdered and stored in sterile container for extraction. Required quantity of the plant part powders [Table 1] were mixed together and extracted with sufficient volume of ethanol solvent using Soxhlet apparatus for 72 h. Non-plant ingredients namely water, honey, jiggery

(sugar), ghritam (ghee), and kasturi (gland of Musk Deer) are not included in the direct ethanol extraction of *Dasamoolarishtam* (DET). Contrastingly, the market preparation (MKT) contain both plant and non-plant ingredients.

The liquid preparations of DET and MKT samples were studied for secondary metabolites screening. Further, both the samples were concentrated using rotary evaporator until the solvent was completely removed. Then, the residue was collected and stored in sterile containers for quantification and antioxidant studies.

Secondary Metabolite Screening

The DET and MKT samples were screened for alkaloids,^[34-36] flavonoids,^[37] tannins,^[38] steroids,^[39] triterpenoids,^[39,40] saponins,^[41] glycosides,^[42] gum and mucilages,^[43] fixed oils^[41] and anthraquinones^[44] with standard procedures.

Quantification of Antioxidant Constituents

Quantification of total phenolics

The total phenolic content was determined using of Folin–Ciocalteu method.^[45] The formation of blue color in the incubated test tubes indicated the presence of phenolics. Soon after, the incubation absorbance was read at 725 nm against the reagent blank. Total phenolic content was expressed as milligrams of gallic acid equivalent (mg/GAE) per gram of extract using the following equation based on the calibration curve: $y = 0.0003x + 0.0716$, $R^2 = 0.9365$, where x was the absorbance and y was the gallic acid equivalent (mg GAE/g extract).

Quantification of total tannins

The total phenolics contain both tannin and non-tannin phenolics. The amount of tannins was calculated by subtracting

Table 1: List of plants and quantity of parts used in the ethanol extraction of *Dasamoolarishtam* (DET)

Contents of each 100 mL of DET	Quantity (g)
<i>Desmodium gangeticum</i> (Wh. Pl.), <i>Uraria picta</i> (Wh. Pl.), <i>Solanum indicum</i> (Wh. Pl.), <i>Solanum xanthocarpum</i> (Wh. Pl.), <i>Tribulus terrestris</i> (Wh. Pl.), <i>Aegle marmelos</i> (St. Bk.), <i>Clerodendrum phlomidis</i> (St. Bk.), <i>Oroxylum indicum</i> (St. Bk.), <i>Stereospermum suaveolens</i> (St. Bk.), <i>Gmelina arborea</i> (St. Bk.)	1.20 each
<i>Plumbago zeylanica</i> (Rt.), <i>Inula racemosa</i> (Rt.)	5.85 each
<i>Symplocos racemosa</i> (St. Bk.), <i>Tinospora cordifolia</i> (St.)	4.70 each
<i>Phyllanthus emblica</i> (Ft.)	3.75
<i>Fagonia cretica</i> (Wh. Pl.)	2.825
<i>Acacia catechu</i> (Ht. Wd.), <i>Pterocarpus marsupium</i> (Ht. Wd.), <i>Terminalia chebula</i> (Pe.)	1.875 each
<i>Saussurea lappa</i> (Rt.), <i>Rubia cordifolia</i> (Rt.), <i>Cedrus deodara</i> (Ht. Wd.), <i>Embelia ribes</i> (Ft.), <i>Glycyrrhiza glabra</i> (Rt.), <i>Clerodendrum serratum</i> (Rt.), <i>Feronia limonia</i> (Ft. Pl.), <i>Terminalia bellirica</i> (Pe.), <i>Boerhavia diffusa</i> (Rt.), <i>Piper chaba</i> (St.), <i>Nardostachys jatamansi</i> (Rz.), <i>Callicarpa macrophylla</i> (Fl.), <i>Hemidesmus indicus</i> (Rt.), <i>Carum carvi</i> (Ft.), <i>Operculina turpethum</i> (Rt.), <i>Vitex negundo</i> (Sd.), <i>Pluchea lanceolata</i> (Lf.), <i>Piper longum</i> (Ft.), <i>Areca catechu</i> (Sd.), <i>Hedychium spicatum</i> (Rz.), <i>Curcuma longa</i> (Rz.), <i>Anethum sowa</i> (Ft.), <i>Prunus cerasoides</i> (St.), <i>Mesua ferrea</i> (Sm.), <i>Cyperus rotundus</i> (Rz.), <i>Holarrhena antidysenterica</i> (Sd.), <i>Pistacia integerrima</i> (Gl.), <i>Pueraria tuberosa</i> (Rt. Tr.) as <i>Jivaka</i> (official substitute), <i>Pueraria tuberosa</i> (Rt. Tr.) as <i>Rishbhaka</i> (official substitute), <i>Asparagus racemosus</i> (Rt.) as <i>Meda</i> (official substitute), <i>Asparagus racemosus</i> (Rt.) as <i>Mahameda</i> (official substitute), <i>Withania somnifera</i> (Rt.) as <i>Kakoli</i> (official substitute), <i>Withania somnifera</i> (Rt.) as <i>Ksirakakoli</i> (official substitute), <i>Dioscorea bulbifera</i> (Rt. Tr.) as <i>Riddhi</i> (official substitute), <i>Dioscorea bulbifera</i> (Rt. Tr.) as <i>Vridhhi</i> (official substitute), <i>Piper cubeba</i> (Ft.), <i>Coleus vettiveroides</i> (Rt.), <i>Santalum album</i> (Ht. Wd.), <i>Myristica fragrans</i> (Sd.), <i>Syzygium aromaticum</i> (Fl. Bd.), <i>Cinnamomum zeylanicum</i> (St. Bk.), <i>Elettaria cardamomum</i> (Ft.), <i>Cinnamomum tamala</i> (Lf.), <i>Piper nigrum</i> (Ft.)	14.065
<i>Vitis vinifera</i> (Dr. Ft.)	7.035
<i>Woodfordia fruticosa</i> (Fl.)	0.23
<i>Strychnos potatorum</i> (Sd.)	

Parts used: Wh. Pl.: Whole plant, Lf.: Leaf, St.: Stem, St. Bk.: Stem bark, Ht. Wd.: Heart wood, Rt.: Root, Rt. Tr.: Root tuber, Rz.: Rhizome, Fl.: Flower, Fl. Bd.: Flower bud, Sm.: Stamen, Pe.: Pericarp, Ft.: Fruit, Ft. Pl.: Fruit pulp, Dr. Ft.: Dried fruit, Sd.: Seed, Gl.: Gall

the non-tannin phenolics from total phenolics. The same method (quantification of total phenolics method) was used for determination of non-tannin phenolics.^[45] The results were expressed in gallic acid equivalents (mg GAE/g extract).

Quantification of total flavonoids

The total flavonoid content of the samples was quantified by the standard method.^[46] The pink color developed due to the presence of flavonoids was read by spectrophotometer at 510 nm. Total flavonoid content was calculated as quercetin equivalents (QE mg/g) using the following equation based on the calibration curve: $y = 0.0255x$, $R^2 = 0.9812$, where x was the absorbance and y was the quercetin equivalent (mg QE/g extract).

In Vitro Antioxidant Activity

2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activity

The antioxidant activity of the samples was determined in terms of hydrogen donating or radical scavenging ability using the stable radical DPPH[•] method.^[47] The absorbance of the samples and control were measured at 517 nm against the methanol blank. The radical scavenging activity of the samples was expressed as IC₅₀ value which is the concentration of the sample required to inhibit 50% of DPPH[•] concentration.

2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid (ABTS))⁺⁺ scavenging activity

The total antioxidant activity was determined by ABTS⁺⁺ radical cation scavenging method.^[48] After incubation the absorbance of samples and standards (Butylated hydroxytoluene [BHT] and Rutin) were measured at 734 nm against the ethanol blank. The results were expressed as the concentration of Trolox having equivalent (TE) antioxidant activity expressed as $\mu\text{M/g}$ sample from the standard curve graph.

Ferric reducing antioxidant power (FRAP) assay

The antioxidant capacities of the samples were determined by the standard procedure.^[49] At the end of incubation period, the absorbance of the blue colour developed was read immediately at 593 nm against the reagent blank. Methanol solutions of known $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ (ferrous sulfate) concentration ranging from 100 to 2000 μM were used for the preparation of the calibration curve. The parameter equivalent concentration was expressed as the concentration of antioxidant having a ferric-TPTZ (ferric tripyridyltriazine) reducing ability equivalent to that of 1 mM $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$. The FRAP value is expressed as mM Fe (II) equivalent/mg extract.

Superoxide radical scavenging activity

Superoxide radical scavenging activity was followed by the improved assay.^[50] This assay was based on the capacity of various extracts to inhibit formazan formation by scavenging the superoxide radicals generated in riboflavin/nitro blue tetrazolium (B2/NBT) system. The absorbance was measured at 590 nm against the blank (unilluminated reaction mixture without plant sample).

The scavenging activity on superoxide anion generation was calculated as:

$$\text{Scavenging activity (\%)} = \frac{[(\text{Control OD} - \text{Sample OD}) / \text{Control OD}] \times 100}{1}$$

Phosphomolybdenum assay

The antioxidant activity of the samples was determined by the green phosphomolybdenum complex formation method.^[51] The absorbance of the mixture was measured at 765 nm against the reagent blank. Ascorbic acid (AA) was used as the reference standard, and the results were expressed as milligrams of AA equivalents per gram extract (AAE/g).

Statistical Analysis

All the quantification and antioxidant assays were done in triplicates and the mean value was calculated, and it was denoted as mean \pm standard deviation.

RESULTS AND DISCUSSION

Secondary Metabolite Screening

The results of secondary metabolite screening of DET and MKT samples are presented in Table 2. Alkaloids, flavonoids, tannins, triterpenoids, saponins, gum and mucilages, and anthraquinones showed positive results in DET and MKT samples. Glycosides and fixed oils were present only in DET which are not observed in MKT. The compound steroids couldn't be detected in both the samples. In addition, alkaloids, tannins and triterpenoids showed very high colour formation/precipitation (+++) during the tests indicating abundant occurrence. Flavonoids, saponins and gum and mucilages showed high colour formation (++) during the tests. This indirectly denotes these components may highly present in the samples. The DET extract showed positive results in 9 compounds, whereas MKT showed positive results in 7 compounds.

Secondary metabolites screening showed that DET extract is rich in phytoconstituents than market sample. Among the various secondary metabolites alkaloids, flavonoids, triterpenoids, and gum & mucilages showed elevated level in DET when compared

Table 2: Secondary metabolite screening of *Dasamoolarishtam* samples

Chemical constituent	Tests	DET	MKT
Alkaloids	Dragendorff's test	++	++
	Mayer's test	++	++
	Hager's test	+++	++
Flavonoids	5% NaOH and 10% HCl test	++	++
Tannins	5% FeCl_3 test	+++	++
Steroids	Liebermann-Burchard's test	-	-
Triterpenoids	Liebermann-Burchard's test	+++	++
	Salkowski's test	-	+
Saponins	Foam test	+	++
Glycosides	Keller - Kiliani test	+	-
Gum and Mucilages	Whistler and BeMiller test	++	+
Fixed oils	Spot test	+	-
Anthraquinones	Sanker and Nahar test	+	+

DET: Direct ethanol sample, MKT: Market sample. "-" denotes absence of compounds; "+" denotes presence of compounds; "++" denotes presence of high colour formation of compounds; "+++ denotes presence of very high colour formation of compounds

to market sample. In 2003, Santosh *et al.*^[52] detected the presence of phenols, terpenoids, glycosides and alkaloids in *Dasamularista* preparation, but flavonoids and steroids couldn't detected in their study. However, the flavonoids were detected in our present study of both DET and MKT samples.

A lot of research reports suggested that secondary metabolites render curative aid for several diseases because of its versatile pharmacological actions such as anti-inflammatory,^[53] antioxidant,^[54] anticancer,^[55] antidiarrheal,^[56] antiallergic,^[57] antifungal,^[58] antiviral,^[59] and allelopathic properties^[60]. The presence of several bioactive compounds in the current study concluded that the therapeutic uses of *Dasamoolarishtam* could be valuable. However, screening of bioactive metabolites of the current research will be helpful in future for isolation and characterization of the phytochemicals that are present in the samples.

Quantification of Antioxidant Constituents

Quantification results of total phenolics, tannins and flavonoids content was statistically analyzed and tabulated [Table 3]. Folin-Ciocalteu reagent was used to determine the total phenolics content. The total phenolics value was determined from the standard graph equitation as gallic acid equivalents per gram extract (mg GAE/g extract). In the present study, the DET sample have 1079.89 mg GAE/g extract in total phenolics, 979.05 mg GAE/g extract in total tannins, and 221.45 mg QE/g extract in total flavonoids, whereas MKT also registered considerable amount of secondary metabolites, but which are lower than DET. However, the phenolic content of the present study was found to be higher than the previous study.^[5] In that study, they obtained 21.67 mg/g of dry mass of GAE in *Dasamoolarishtam* market samples. In similar way, there is no findings have been published previously for estimation of total tannins and total flavonoids of *Dasamoolarishtam* samples.

Phenolic compounds of medicinal plants have been reported to possess antimicrobial,^[61] anti-inflammatory,^[62] anticancer,^[63] cardioprotective^[64] activities. Excessive amount of tannins have been reported to suppress the absorption of minerals such as irons, it may lead to anemia. Therefore, the occurrence of fewer amount of tannins in plant parts is always preferred.^[65]

On the other hand, tannins have been reported to possess many biological activities such as anti-tumor,^[66] antiulcer,^[67] antidiabetic,^[68] and anti-inflammatory^[69] activities. The compound flavonoids are universally distributed in plants, they having hydroxyl groups in their basic structure which mediated for their antioxidant effect. Flavonoids which exist as phenolic compounds that display antioxidant activity particularly by quenching free radicals.^[70,71] In addition, flavonoids has been reported to have various biological activities such as anticarcinogenic and anti-inflammation activities.^[72,73] Therefore, rich amount of secondary

metabolites present in the DET sample is identified to be a good indication of higher antioxidant activity.

In Vitro Antioxidant Activity

DPPH[•] radical scavenging activity

DPPH[•] radical scavenging activity (IC₅₀) was measured by determining the concentration required to remove 50% of the free radical DPPH[•] after 30 min of reaction. In the present study, the DET extract showed high radical scavenging activity (5.16 ± 1.45 µg/mL) when compared to standards rutin and BHT [Figure 1]. Meanwhile MKT registered very low radical scavenging activity (30.86 ± 3.37 µg/mL) in this assay. The IC₅₀ value of rutin and BHT used as positive control in this study which is 7.73 ± 0.71 and 6.87 ± 0.57 µg/mL, respectively. Antioxidant studies indicated that DPPH[•] possess the ability to identify the radical scavenging capacity of the selected samples. During the oxidative stress, excessive amount of free radicals have been produced which is root cause damage for biomolecules in health system.^[74,75] Free radicals are reactive and short-lived molecules which employ fast kinetic methods.^[74] However, the result of DPPH[•] radical scavenging activity suggested that DET extract expressed more radical scavenging effect than other samples.

ABTS^{•+} scavenging activity

ABTS^{•+} scavenging activity of the present study is showed in Table 4. The DET extract showed high radical scavenging activity (20013.89 µM TE/g extract) when compared to that of MKT sample. While standards rutin and BHT showed 30375.0 and 32736.11 µM TE/g extract, respectively. The results showed that the DET and MKT samples possess significant level of total antioxidant capacity. In evaluating the total antioxidant potential of antioxidants agents, the ABTS scavenging assay play an important role where it helps in measuring the reduction of the cation radical as the percentage of inhibition.^[76] ABTS^{•+} assay has been widely studied in *in vitro* models as it is an easy method to evaluate antioxidant potential of the plant extracts.^[77] Antioxidants are source of protectors to the human body, the plant extracts reacts with stable free radical molecules which donate electrons and convert nonradical molecules with discoloration of reaction mixture.^[78] However, the DET sample showed highest ABTS^{•+} scavenging activity than other samples.

FRAP assay

The natural antioxidants possess ability to donate electrons to reactive radical cation, reducing them to more stable and un-reactive compound which is said to be the basic of ferric reducing power assay. The results of FRAP analysis [Table 4]

Table 3: Total phenolics, tannins and flavonoids content of DET and MKT samples

Extracts	Total phenolics (mg GAE/g extract)	Total tannins (mg GAE/g extract)	Total flavonoids (mg QE/g extract)
DET	1079.89±1.15	979.05±2.41	221.45±8.14
MKT	267.02±3.49	254.25±3.33	22.96±1.32

Values are mean of triplicate determination (n=3) ±standard deviation. DET: Direct ethanol sample, MKT: Market sample, GAE: Gallic acid equivalents; QE: Quercetin equivalents

Table 4: ABTS scavenging activity, FRAP and superoxide radical scavenging activity of DET and MKT samples

Samples	ABTS scavenging activity ($\mu\text{M TE/g extract}$)	FRAP activity (mM Fe (II) E/mg extract)	Superoxide radical scavenging activity (% of inhibition)
DET	20013.89 \pm 610.36	689.25 \pm 4.17	22.71 \pm 2.30
MKT	19284.72 \pm 262.14	679.38 \pm 3.60	17.59 \pm 1.60
Rutin	30375.0 \pm 416.67	814.81 \pm 1.20	63.4 \pm 0.10
BHT	32736.11 \pm 433.68	826.17 \pm 2.30	64.7 \pm 0.25

Values are mean of triplicate determination ($n=3$) \pm standard deviation. DET: Direct ethanol sample, MKT: Market sample, BHT: Butylated hydroxytoluene, TE: Trolox equivalents, Fe (II) E: Ferrous ion equivalents. FRAP: Ferric reducing antioxidant power

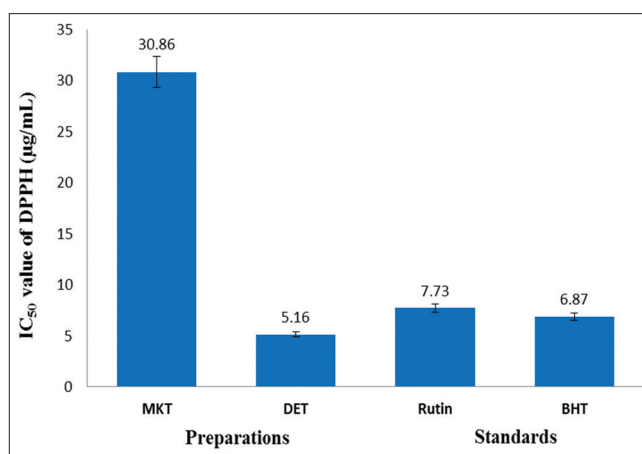
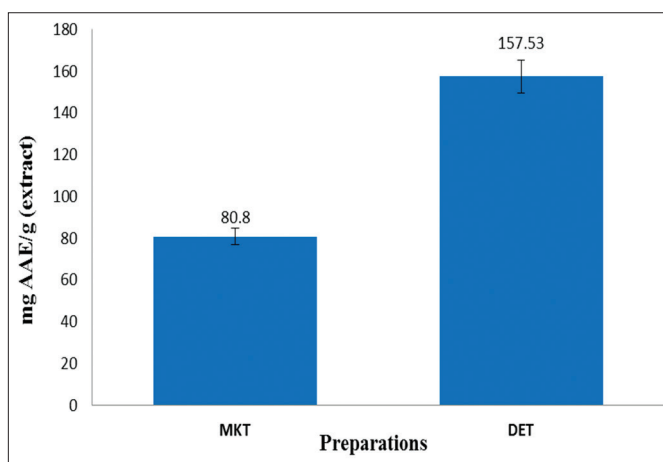
revealed that the ferric reducing capacity of DET extract was higher (689.25 mM Fe (II) E/mg extract) than MKT sample (679.38 mM Fe (II) E/mg extract), which is less than standards BHT (826.17 mM Fe (II) E/mg extract) and Rutin (814.81 mM Fe (II) E/mg extract). Plants have the ability to reduce ferric ions which plays an important role in determining FRAP assay. Antioxidants are capable of donating a single electron to the ferric-TPTZ (Fe (III)-TPTZ) complex which would cause the reduction of this complex into the blue ferrous TPTZ (Fe (II)-TPTZ) complex which absorbs strongly at 593 nm.^[79] DET extract showed significant FRAP antioxidant activity in a concentration-dependent manner. The reducing power of the DET sample is associated with the phenolic compounds which donate hydrogen atom and it plays a key role in breaking the free radical chains. Therefore, phenolics and flavonoids presence in the DET extract exhibited the ability to have this kind of antioxidant potential.

Superoxide radical scavenging activity

The superoxide radicals generated from dissolved oxygen can be measured by their ability to reduce riboflavin/nitro blue tetrazolium (B2/NBT). The secondary metabolites present in plants possess a wide range of biologically active compounds which protect the cells from colossal oxidative damage.^[80] The decrease in absorbance at 590 nm with DET and MKT samples and also the reference compounds indicate their ability to quench the superoxide radicals in the reaction mixture. As shown in Table 4, the DET extract has significant inhibition in superoxide radical scavenging activity (22.71%) than MKT (17.59%) sample, whereas the standards rutin and BHT showed very high activity in this assay.

Phosphomolybdenum assay

The phosphomolybdenum assay was used to determine the total antioxidant capacity of DET and MKT samples, and the results are presented in Figure 2. The DET sample showed high reducing ability (157.53 \pm 1.0 mg AAE/g extract). While, MKT sample also exhibited significant amount of reduction ability (80.8 \pm 1.83 mg AAE/g extract). Qualitative determination of antioxidant capacity can be determined by spectrophotometric method; it was done based on the principle in which the reduction of molybdenum (VI) to molybdenum (V), which leads to the subsequent formation of a green phosphate/molybdenum (V) complex in the maximum absorbance at 695 nm.^[51] From the results, antioxidant capacity of the DET was able to inhibit the molybdenum complex. Apart from this, the total antioxidant ability of the selected samples may be due to the occurrence of phenolic compounds.

**Figure 1:** DPPH radical scavenging activity**Figure 2:** Phosphomolybdenum assay

CONCLUSION

Ayurvedic plant-based formulations play an important role in the general healthcare system of Indian population and recently gaining popularity in worldwide also. However, there is an urgent need for standardization of Ayurvedic preparations by the evaluation of phytochemical and pharmacological standards through modern scientific methods. Such studies enhance the acceptability and commercialization of these vital plant-based drugs at worldwide. In the current study, ethanol extract obtained from *Dasamoolarishtam* plants (DET) and widely used marketed *Dasamoolarishtam* formulation (MKT) were studied for their phytochemical and antioxidant potentials. Both the samples

showed significant results. However, the data presented in this study revealed that the DET is rich in phytochemicals by means of qualitative and quantitative analysis of secondary metabolites. The results also concluded that the DET sample exhibited strong antioxidant activity. Further, studies are warranted for the isolation and identification of individual bioactive compounds and also *in vivo* studies for understanding their mechanism of actions.

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