

Development of Standardization Parameters of Leaves of *Trianthema portulacastrum* Linn.

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Abstract

As per estimate of WHO aAbout 80% of the Indian population depends on traditional system of medicine for the treatment of several diseases. Increasing population relying on herbal plants and the product prepared from the herbs and because of the same plant need to be scientifically screened out. *Trianthema portulacastrum* Linn. belongs to the family Aizoaceae, is one of the common weed, which has enormous traditional uses against diseases and some bioactive compounds have been isolated from this weed. In the present investigation standardization parameters of leaves of *Trianthema portulacastrum* Linn. were developed and reported.

Keywords: *Trianthema portulacastrum* Linn., Leaves, Standardization Parameters

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Introduction

World Health Organization (WHO) has recommended that traditional health and folk medicine systems are proved to be more effective in health problems worldwide. [1] *Trianthema portulacastrum* Linn. (Sabuni in Hindi) is a herb used in Ayurvedic medicine. The active and main phyto-constituents are ecdysterone, trianthenol, 3-acetylaleuritic acid, 5,2'-dihydroxy-7-methoxy-6,8-dimethylflavone, leptorumol, 3,4-dimethoxy cinnamic acid, 5-hydroxy-2-methoxybenzaldehyde, p-methoxybenzoic acid, and beta cyanin. Different parts of *Trianthema portulacastrum* Linn. are traditionally used as analgesic, stomachic, laxative, treatment of blood disease, anemia, inflammation, and night blindness. The plant is alexiteric, analgesic, stomachic, laxative, alterative; cures Kapha, bronchitis, heart diseases of the blood, anemia, inflammations, Vata, piles and ascites. The plant has been used in the indigenous system of medicine for the obstruction of the liver asthma, amenorrhea, dropsy, edema, ascites, and beri-beri. A decoction of the herb is used as a vermifuge and is useful in rheumatism; it is also an antidote to alcoholic person. [2-4] So, far no any systematic study was carried out in evaluating the standardization parameters of leaves of selected plant, therefore, the present work was undertaken.

Material and Methods

Collection of herbs and their authentication

The leaves of *Trianthema portulacastrum* Linn. was collected in the months of July-September 2021 from the various local sites of Malwa region of Madhya Pradesh and identified & authenticated by Dr. S. N. Dwivedi, Retd. Prof. and Head, Department of Botany, Janata PG College, A.P.S. University, Rewa, (M.P.) and was deposited in our Laboratory.

Development of Standardization parameters of *Trianthema portulacastrum* Linn. [5-8]

Pharmacognostical Evaluation

Macroscopic studies

The macroscopy of different parts of the plant such as color, odor, size, shape, taste, surface characters and fractures were carried out.

Physicochemical Evaluation

The dried parts were subjected to standard procedure for the determination of various physicochemical parameters.

Determination of foreign organic matter (FOM)

Accurately weighed 100 g of the drug sample and spread it out in a thin layer. The foreign matter should be detected by inspection with the unaided eye or by the use of a lens (6X). Separate and weigh it and the percentage present was calculate.

Determination of moisture content (LOD)

Place about 10 g of drug (without preliminary drying) after accurately weighing in a tared evaporating dish and kept in oven at 105⁰ C for 5 hours and weigh. The percentage loss on drying with reference to the air dried drug was calculated.

Determination of ash value

The determination of ash values is meant for detecting low-grade products, exhausted drugs and sandy or earthy matter. It can also be utilized as a mean of detecting the chemical constituents by making use of water-soluble ash and acid insoluble ash.

Total ash

Accurately about 3 gms of air dried powder was weighed in a tared silica crucible and incinerated at a temperature not exceeding 450⁰C until free from carbon, cooled and weighed and then the percentage of total ash with reference to the air dried powdered drug was calculated. The percentage of total ash with reference to the air-dried drug was calculated.

Acid insoluble ash

The ash obtained in the above method was boiled for 5 minutes with 25ml of dilute HCl. The residue was collected on ash less filter paper and washed with hot water, ignited and weighed. The percentage of acid insoluble ash was calculated with reference to the air dried drug.

Water soluble ash

The ash obtained in total ash was boiled for 5 minutes with 25 ml of water. The insoluble matter was collected on an ash less filter paper, washed with hot water and ignited to constant weight at a low temperature. The weight of insoluble matter was subtracted from the weight of the ash. The difference in weights represents the water soluble ash. The percentage of water soluble ash with reference to the air dried drug was calculated.

Determination of swelling index

Swelling index is determined for the presence of mucilage in the seeds. Accurately weigh 1 g of the seed and placed in 150 ml measuring cylinder, add 50 ml of distilled water and kept aside for 24 hours with occasional shaking. The volume occupied by the seeds after 24 hours of wetting was measured.

Determination of extractive value

This method determines the amount of active constituents extracted with solvents from a given amount of medicinal plant material. It is employed for materials for which as yet no suitable chemical or biological assay exists.

Cold maceration

Place about 4.0g of coarsely powdered air-dried material, accurately weighed, in a glass-stoppered conical flask. Macerate with 100ml of the solvent specified for the plant material concerned for 6 hours, shaking frequently, then allow to stand for 18 hours. Filter rapidly taking care not to lose any solvent, transfer 25 ml of the filtrate to a tared flat-bottomed dish and

evaporate to dryness on a water bath. Dry at 105°C for 6 hours, cool in a desiccator for 30 minutes and weigh without delay. Calculate the content of extractable matter in mg per g of air dried material. For ethanol-soluble extractable matter, use the concentration of solvent specified in the test procedure for the plant material concerned; for water-soluble extractable matter, use water as the solvent.

Successive Extraction of selected herbs

Sample were shattered and screened with 40 mesh. The shade dried coarsely powdered plant material (250gms) were loaded in Soxhlet apparatus and was extracted with petroleum ether (60-62°C), Chloroform, ethanol and water until the extraction was completed. After completion of extraction, the solvent was removed by distillation. The extracts were dried using rotator evaporator. The residue was then stored in dessicator and percentage yield were determined.

Preliminary phytochemical screening of extracts

The various extract obtained after extraction were subjected for phytochemical screening to determine the presence of various phytochemical present in the extracts. The standard procedure were adopted to perform the study.

Tests for carbohydrates

Molisch's test

To the Sample 2-3 drops of 1% alcoholic - naphthol solution and 2 ml of conc. sulphuric acid was added along the sides of the test tube. Appearance of purple to violet ring at the junction of two liquids shows the presence of carbohydrates.

Fehling test

To the sample add fehling reagent, appearance of brick red precipitate shows presence of carbohydrates.

Test for glycosides

Legal's test

To the sample add 1 ml of pyridine and few drops of sodium nitropruside solutions and then it was made alkaline with sodium hydroxide solution. Appearance of pink to red colour shows the presence of glycosides.

Borntrager's test

Sample was treated with chloroform and then the chloroform layer was separated. To this equal quantity of dilute ammonia solution was added. Ammonia layer acquires pink color, showing the presence of glycosides.

Baljet's test

To the sample add picric acid, orange color shows presence of glycosides.

Test for alkaloids

A small portion of the sample was stirred separately with few drops of dilute hydrochloric acid and was tested with various reagents for the presence of alkaloids. The reagents are

- Dragendroff's reagent - Reddish brown precipitates
- Wagner's reagent - Reddish brown precipitates
- Mayer's reagent - Cream color precipitates
- Hager's reagent - Yellow color precipitate

Test for proteins and free amino acids

Small quantities of the sample was dissolved in few ml of water and treated with following reagents.

- Million's reagent: Appearance of red color shows the Presence of protein and free amino acid.
- Ninhydrin reagent: Appearance of purple color shows the Presence of Proteins and free amino acids.
- Biuret's test: Equal volumes of 5% sodium hydroxide solution & 1% copper sulphate solution was added. Appearance of pink or purple color shows the presence of proteins and amino acids.

Test for tannins and phenolic compounds

A small quantity of the sample was taken separately in water and test for the presence of phenol compounds and tannins was carried out with the following reagents.

- Dilute Ferric chloride solution (5%) - Blue color or green color
- 10% lead acetate solution - White precipitates

Test for flavonoids

Alkaline reagent test

To the test solution add few drops of magnesium hydroxide solution, intense yellow colour is formed which turns to colourless on addition of few drops of dilute acid indicates presence of flavonoids.

Shinoda's test

Small quantities of the sample was dissolved in alcohol, to them piece of magnesium followed by conc. hydrochloric acid drop wise added and heated. Appearance of pink, crimson red, green to blue color shows the presence of flavonoids.

Tests for fixed oils and fats

Spot test

A small quantity of sample was separately pressed between two filter papers. Appearance of oil stain on the paper indicates the presence of fixed oil.

Saponification test

Few drops of 0.5 N alcoholic potassium hydroxide were added to a small quantity of sample along with a drop of phenolphthlein, the mixture was heated on a water bath for 1-2 hours, formation of soap or partial neutralization of alkali indicates the presence of fixed oils and fats.

Tests for steroids and triterpenoids

Libermann-burchard test

Treat the sample with few drops of acetic anhydride, boil and cool. Then add con. sulphuric acid from the side of test tube, brown ring is formed at the junction two layers and upper layer turns green which shows presence of steroids and formation of deep red colour indicates presence of triterpenoid.

Salkowski test

Treat the sample with few drop of conc. sulphuric acid, red colour at lower layer indicates presence of steroids and formation of yellow coloured lower layer indicates presence of triterpenoids.

Test for mucilage and gums

- Small quantities of sample was added separately to 25 ml of absolute alcohol with constant stirring and filtered. The precipitates was dried in oil and examined for its swelling property for the presence of gum and mucilage.
- To the sample add ruthenium red solution, pink color shows presence of mucilage.

Test for waxes

To the test solution add alcoholic alkali solution, waxes get saponified.

Results and Discussion

The leaves of *Trianthema portulacastrum* Linn. was collected from local sites of Malwa region of Madhya Pradesh, India and identified morphologically and compared with standard pharmacopoeial monograph. The sample of drug was also identified & authenticated by Dr. S. N. Dwivedi. The macroscopy of different parts of the plant such as color, odor, size, shape, taste, surface characters and fractures were carried out. The results were presented in table 1. The dried leaves of *Trianthema portulacastrum* Linn. were subjected to standard procedure for the determination of various physicochemical parameters. The results were presented in table 2. The shade dried coarsely powdered plant material of *Trianthema portulacastrum* Linn.: Leaves was extracted with petroleum ether, Chloroform, ethanol and water. The extracts obtained were evaluated for pH, color and % yield. The results are presented in table 3. The various extract obtained after extraction were subjected for phytochemical screening to determine the presence of various phytochemical present in the extracts. The standard procedure was adopted to perform the study. (Table 4)



Fig. 1: *Trianthema portulacastrum* Linn.: Leaves

Table 1: Morphological characters of *Trianthema portulacastrum* Linn.: Leaves

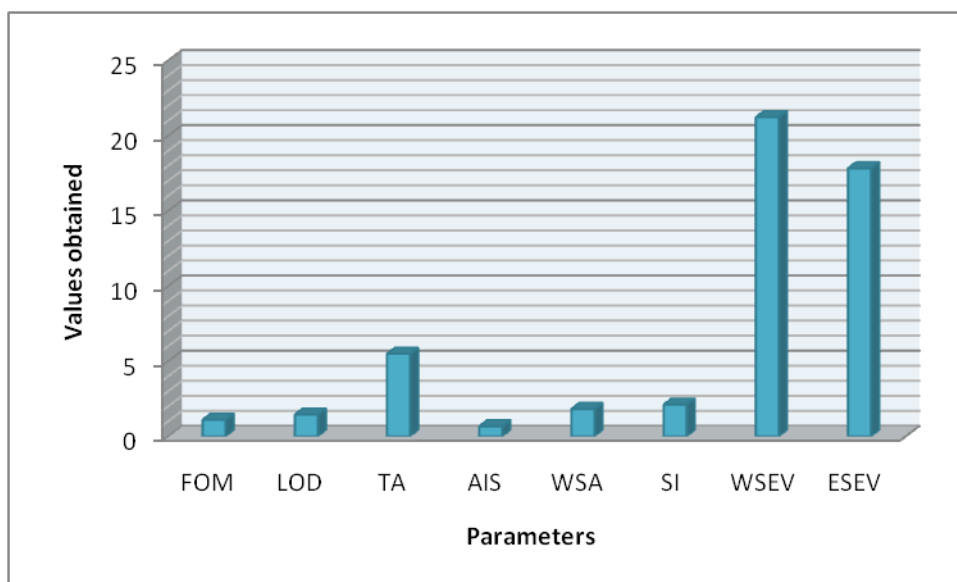
S/No.	Parameters	TPL
1.	Color	Green
2.	Odor	Characteristics
3.	Taste	Sweet
4.	Shape	oval
5.	Size	Variable
6.	Surface character	Smooth
7.	Fractures	Absent

Abbr.: TPL: *Trianthema portulacastrum* Linn.: Leaves

Table 2: Physicochemical Evaluation of *Trianthema portulacastrum* Linn.: Leaves

S/No.	Parameters	TPL
1.	FOM	1.10±0.23
2.	LOD	1.45±0.43
3.	TA	5.49±0.11
4.	AIS	0.67±0.15
5.	WSA	1.82±0.02
6.	SI	2.11±0.01
7.	WSEV	21.23±1.24
8.	ESEV	17.83±1.19

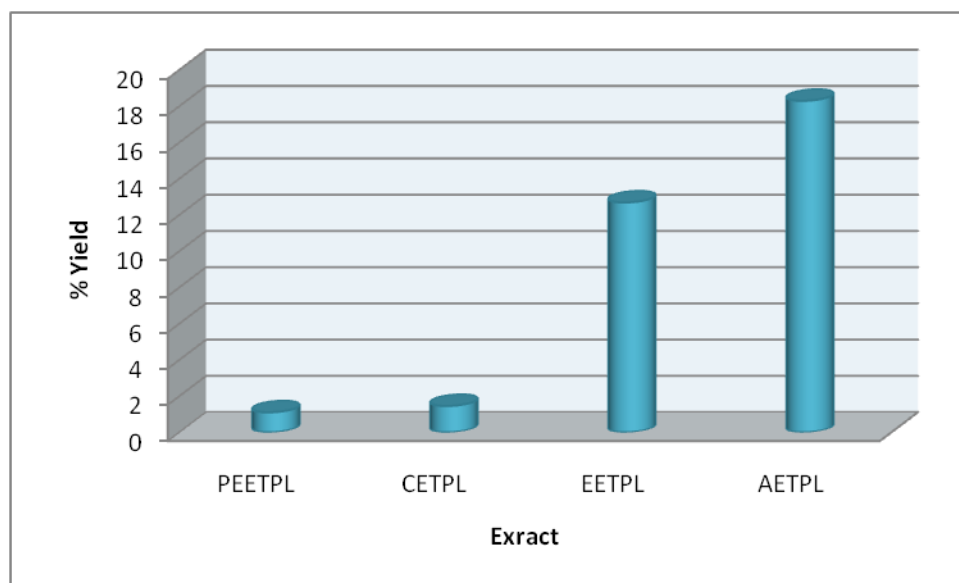
Note: All values are expressed as Mean±SEM, n=3



Graph 1: Physicochemical Evaluation of *Trianthema portulacastrum* Linn.: Leaves

Table 5.3: Estimation of % yield of various extract of selected plant material

S/No.	Extract	Parameters			
		Nature of Extract	Color	pH	% Yield (w/w)
1.	PEETPL	Semi Solid	Creamish	6.99	1.10
2.	CETPL	Semi solid	Light cream	7.01	1.45
3.	EETPL	Solid Powder	Light green	7.01	12.68
4.	AETPL	Solid Powder	Green	7.02	18.29



Graph 2: % yield of extract

Table 5.7: Preliminary phytochemical screening of *Trianthema portulacastrum* Linn.:
Leaves

S/No.	Constituents	Leaves Extract			
		PEETPL	CETPL	EETPL	AETPL
1.	Carbohydrates	-	-	-	-
2.	Glycosides	-	-	+	+
3.	Alkaloids	-	-	+	+
4.	Protein & Amino acid	-	-	-	-
5.	Tannins & Phenolic compounds	+	+	+	+
6.	Flavonoids	+	+	+	+
7.	Fixed oil and Fats	-	-	-	-
8.	Steroids & Triterpenoids	+	+	+	+
9.	Waxes	-	-	-	-
10.	Mucilage & Gums	-	-	-	-

Abbr.: +=Present; -=Absent

Conclusion

Traditional system of medicine healers treats diseases using the plants which have immense medicinal potentiality. But due to lack of standardization parameters correct identification of the plant is lacking, therefore development of standardization parameters is of great interest. In this study physicochemical, extraction and preliminary phytochemical screening of the selected plant material was done and reported.

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