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# PHARMACOGNOSTICAL AND PHYTOCHEMICAL EVALUATION OF *AAMRA* [*Mangifera indica* Linn.]

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## ABSTRACT

INTRODUCTION- *Amra* [*Mangifera indica Linn.] h*as been used in traditional medicine for the treatment of different disease. It contains tannins, pyrogallotannins, mangiferin and also contains vitamin A and C. Although several studies have been conducted on *Amra* [*Mangifera indica Linn.]*, there remains a lack of a comprehensive review focusing on its pharmacognostical, physicochemical, phytochemical, and chromatographic evaluations. AIM AND OBJECTIVE- To conduct a thorough pharmacognostical, physicochemical, phytochemical, and chromatographic evaluation of *Amra* [*Mangifera indica Linn.]*. MATERIAL AND METHODS- This study was designed to evaluate *Amra* [*Mangifera indica Linn.]* through pharmacognostical, physicochemical, phytochemical, and chromatographic analyses to assess its quality, purity, and safety. OBSERVATIONS AND RESULTS- The results obtained from the pharmacognostical, physicochemical, phytochemical, and chromatographic assessments were all found to be within the acceptable quality standards. CONCLUSION- The tested sample of *Amra* [*Mangifera indica Linn.]* demonstrated high quality and was confirmed to be pure, safe, and authentic.

**Keywords:** Pharmacognosy, Phytochemicals, *Amra*, *Mangifera indica Linn.* Pharmacognostical.

**INTRODUCTION:** *Mangifera indica,* also referred to as Mango or *Amra,* has been a significant herb in indigenous and Ayurvedic medicine for more than 4,000 years. The genus *Mangifera*, which includes roughly 30 species of tropical fruiting trees in the *Anacardiaceae* family of flowering plants, is where mangoes are found.[1]

*Ayurveda* attributes a variety of therapeutic qualities to the various sections of the mango tree. *Mangiferin* has potent antioxidant, anti-lipid peroxidation, immunomodulatory, cardiotonic, hypotensive, wound healing, antidegenerative, and antidiabetic properties since it is a polyphenolic antioxidant and a glucosyl xanthone.[2]

**Taxonomy** *[3]*

**Kingdom:** *Plantae plants*
**Subkingdom:** *Viridaeplantae* – green plants
**Division:** *Angiospermae*
**Subdivision:** *Spermatophyta* (Seed plant)
**Class:** M*agnoliopsida*
**Subclass:** Rosidae
**Order:** Sapindales
**Family:** Anacardiaceae
**Genus:***Mangifera*
**Species:** M. Indica L
**English name:** *Mango*

**Pharmacological characters**

Table-01: *Raspanchaka of Aamra* [4-5-6]

| | Seed | Bark | Unripe fruit | Ripe fruit |
|---|---|---|---|---|
| ***Rasa*** | *Kashaya, Madhura, Amla* | *Kashaya* | *Amla* | *Madhura* |
| ***Guna*** | *Laghu, Ruksha* | *Laghu,Ruksha* | *Laghu,Ruksha* | *Guru, Snigdha* |
| ***Veerya*** | *Sheeta* | *Sheeta* | *Ushna.* | *Sheeta* |
| ***Vipaka*** | *Katu* | *Katu* | *Amla* | *Madhura* |

Despite the availability of a few reviews on this plant, there is a lack of comprehensive studies that encompass the pharmacognostical, physicochemical, phytochemical, and chromatographic evaluation of *Amra* [*Mangifera indica Linn.].*

**AIM AND OBJECTIVE** To perform pharmacognostical, physio-chemical, phytochemical and chromatography evaluation of *Amra* [*Mangifera indica Linn.].*

## MATERIAL AND METHODS

**Sample Preparation**- The powdered form of Beej of *Amra* [*Mangifera indica* Linn.] was processed using a vibro sifter to achieve a uniform particle size of 80 mesh for testing. The evaluation procedures included the following:

**Pharmacognostical Study**

The organoleptic properties of the sample, such as color, odor, taste, and texture, were assessed using naked eye observation and a magnifying lens.

**Powder Microscopy-** Powder microscopy is used to identify diagnostic features in medicinal plants, including trichomes, fibers, vessels, starch grains, and crystals. For this, the powdered sample was treated with various chemical reagents, such as phloroglucinol + HCl for lignified tissues, iodine for starch, and Sudan III for oils and fats, to make specific structures more visible.

**Physicochemical Analysis**

The physicochemical properties of *Amra* [*Mangifera indica* Linn.] were evaluated, with the following tests:

**Determination of Moisture Content / Total Soluble Solids**

Moisture content was determined by placing 5 grams of the sample in an oven at 105°C for 5 hours. The weight of the sample was checked every 30 minutes until it stopped changing. The sample was cooled in a desiccator to room temperature before being weighed again.

**Calculations:**

Weight of empty petridish = W1gm

Weight of the drug sample = X gm

Weight of petridish with drug before drying (W3) = (W1 + X)

Weight of petridish with drug after drying = W2gm

Loss on drying in % = [(W3 - W2) × 100]/X

**Determination of pH**

The pH of an aqueous solution of *Amra* [*Mangifera indica* Linn.] was measured using a digital pH meter. This provides a quantitative indication of the solution's acidity or alkalinity. A digital pH meter was used to measure the pH of a specific solution.

The device was first calibrated using standard solutions with pH values of 6, 7, and 8.

Each solution was prepared by diluting a tablet of the respective pH in 100 ml of distilled water. Before measurements, the device was turned on and allowed to warm up. The electrode was dipped into the buffer solution in a beaker. A 10% aqueous solution of the sample was prepared, and the electrode was immersed in it to record the pH value.

**Determination of Extractive Values**

- **Determination of Water-Soluble Extractive**

5 grams of *Amra* [*Mangifera indica* Linn.] was soaked in 100 ml of distilled water in a closed flask. It was left for 24 hours, then shaken continuously for 6 hours using a rotary shaker. After that, it was allowed to

stand for 18 hours. The mixture was filtered using filter paper. The filtrate was transferred to a pre-weighed flat-bottomed dish and evaporated to dryness on a water bath. The dish was then placed in an oven at 105°C until a constant weight was reached, and the final weight was recorded.

**Calculations:**

Weight of the drug material = X gm
Weight of the empty petridish = W1gm
Weight of the petridish with dried extract = W2gm
Percentage of extractive value = [(W2 - W1) × 100]/X
The procedure was repeated three times, and the average was calculated.

- **Determination of Alcohol-Soluble Extractive**

The procedure for alcohol-soluble extractive is similar to that of water-soluble extractive, with the only difference being the use of alcohol instead of distilled water.

- **Determination of Petroleum Ether-Soluble Extractive**

5 grams of *Amra* [*Mangifera indica* Linn.] was mixed with 100 ml of petroleum ether in a closed flask and allowed to stand for 6 hours in a continuous extraction apparatus. The mixture was then filtered using filter paper. The filtrate was transferred to a pre-weighed flat-bottomed dish and evaporated to dryness on a water bath. The dish was then placed in an oven at 105°C until a constant weight was achieved, and the final weight was recorded.

**Calculations:**

Weight of the drug material = X gm
Weight of the empty petridish = W1gm
Weight of the petridish with dried extract = W2gm
Percentage of extractive value = [(W2 - W1) × 100]/X

**Determination of Ash Value**

- **Determination of Total Ash Value**

5 grams of powdered *Amra* [*Mangifera indica* Linn.] was placed in a silica crucible. The crucible was placed in a muffle furnace after spreading the sample in a thin layer. The furnace temperature was set to 450°C for about 6 hours or longer until the ash was completely free of carbon. The crucible with the ash was cooled in a desiccator and then weighed to a constant weight.

**Calculations:**

Weight of empty silica crucible = A1 gm
Weight of the sample (X) = X gm
Weight of crucible with ash = A2 gm
Percentage of total ash = [(A2 - A1)/X] × 100

- **Determination of Acid-Insoluble Ash**

The total ash was boiled with 25 ml of 2M hydrochloric acid for 5 minutes.
The insoluble residue was collected in a Gooch crucible, washed with hot water, and ignited at a temperature not exceeding 450°C for 15 minutes. The crucible was cooled to room temperature in a desiccator and weighed.

**Calculations:**

Weight of the drug sample = X gm
Weight of crucible = G1 gm
Weight of crucible with insoluble ash = G2 gm
Weight of insoluble ash (G3) = G2 - G1
Percentage of acid-insoluble ash = (G3/X) × 100

- **Determination of Water-Soluble Ash**

The total ash was boiled with 25 ml of water for 5 minutes. The insoluble residue was collected in a Gooch's crucible, washed with hot water, and ignited at a temperature not exceeding 450°C for 15 minutes. The weight of the insoluble residue was subtracted from the weight of the total ash. The difference represented the water-soluble ash.

**Calculations:**

Weight of the drug sample = X gm
Weight of total ash = A gm
Weight of crucible = G1 gm
Weight of crucible with insoluble ash = G2 gm
Weight of insoluble ash (G3) = G2 - G1
Water-soluble ash (G4) = Weight of total ash (A gm) - Weight of insoluble ash (G3)
Percentage of water-soluble ash = [(A - G3)/X] × 100

**Phytochemical Study[10]**

**Qualitative phytochemical analysis** was conducted on both aqueous and alcoholic extracts of *Amra* [*Mangifera indica Linn.]* to identify the presence of various bioactive constituents using the following tests:

**Tests for Carbohydrates**

- **Molisch's test-** 2ml of test solution taken in a test tube added with 2ml of the Molisch's reagent, shaken carefully and then about 1ml of conc. H2SO4 is poured from side of the test tube and allowed to stand for 1 minute. Formation of purple colour ring at the junction of the two layers will indicate the presence of Carbohydrate.
- **Benedict's test-**It is used for detecting reducing sugars and is mainly composed of Copper sulphate and sodium hydroxide. To 4ml of aqueous solution of drug, 1ml of Benedict's solution was added and heated almost to boiling. Formation of green, yellow, orange, red and brown colours in order of increasing concentrations of simple sugar due to formation of cuprous oxide.
- **Fehling solution test**-It is generally used for detecting reducing sugars and composed of two solutions, which are mixed in situ. Fehling solution A (0.5% of copper sulphate) and Fehling solution B (Sodium Potassium Tartrate). Equal volumes of Fehling A and Fehling B solutions were mixed (1ml each) and 2ml of aqueous solution of drug was added and then boiled for 5-10 minutes on water bath.

**Tests for Alkaloids**

- **Dragondroff's reagent test-** 2ml of test solution was taken in a test tube in which 2ml of the DragonDroff's reagent (mixture of Potassium Iodide and Bismuth sub nitrate solution) was added. Formation of orange precipitate indicates presence of alkaloids.
- **Wagner's Test-** Drug solution when added with few drops of Wagner's reagent (dilute Iodine solution) a formation of reddish-brown precipitate indicates presence of alkaloids.
- **Hager's Test-** A saturated aqueous solution of picric acid was used for this test. It was added to the test sample. The formation of an orange yellow precipitate will indicate the presence of alkaloids.

**Test for Amino acids å**

- **Ninhydrin test-** It is used to detect the presence of alpha-amino acids and proteins containing free amino groups. Protein solution when heated with ninhydrin molecules, it results in formation of complex between two ninhydrin molecule and nitrogen of free amino acid. This gives a characteristic deep blue or pale yellow colour.

**Tests for Proteins**

- **Biuret test-** A few mg of the residue was taken in water and 1ml of 4% sodium hydroxide solution was added to it, followed by a drop of 1% solution of copper sulphate. Development of violet or pink colour indicates the presence of proteins.
- **Xanthoprotic test-** 2ml of test sample in test tube is added with 0.5 ml of concentrated nitric acid. Development of yellow colour indicates presence of proteins.
- **Millons test-** A small quantity of test sample was taken and 2 to 3 ml of millons reagent was added. The white precipitate slowly turning to pink, indicate the presence of proteins.

**Test for saponin**

- **Foam test-** A small quantity of the test sample was taken in a test tube and shaken vigorously with a small amount of sodium bicarbonate and water. A stable, characteristic honeycomb like froth indicates the presence of saponins.

**Test for glycosides**

- **Borntragor's Test-** 1ml of Benzene and 0.5ml of dilute ammonia solution was added to the ethanolic extract and was observed for the formation of reddish pink colour.

**Test for Phenolic Compound**

- **Phenolic test-** The extract was taken in water and warmed; to this 2 ml of ferric chloride solution was added and observed for the formation of green and blue colour.

**Test for Steroids**

- **Salkoweski reaction-** Few mg of extract was taken in 2ml of chloroform and 2ml of concentrated sulphuric acid was added from the side of test tube. The test tube was shaken for few minutes. The development of red colour indicates the presence of steroids.

**Test for Tannins**

- **Ferric chloride solution-** A 5 percent solution of ferric chloride in 90% alcohol was prepared. Few drops of this solution were added to the test sample. Appearance of dark green or deep blue colour indicates the presence of tannins.
- **Lead acetate-** A 10 percent w/v solution of basic lead acetate in distilled water was added to the test filtrate. Development of precipitate indicates the presence of tannins.
- **Pot. Dichromate-** A solution of potassium dichromate was added to the filtrate. Appearance of dark colour indicates the presence of tannins.

**Chromatographic Study**[11]

Chromatography is a technique to separate mixture of substances into components on the basis of their molecular structure and molecular composition. TLC-thin layer chromatography, is used for separation of mixture and identification of its chemical constituent.

The plates utilized were T.L.C. plates covered with a 0.25 mm layer of silica gel 60 F254 with fluorescent indicator. (Each plate measures 10 cm in length and 2 cm in breadth.)"

**Activation of pre-coated Silica gel 60 F254** -Plates were dried for one and a half hours in a hot oven at $105^0$ C.

**Preparation of mobile solution-** n-Butanol: Acetic acid: water (4:1:5)

**Test solution:** Alcoholic Extract

**Visualization-** In Iodine vapours

**Rf Value**-The distance of each spot from the place of application was measured and recorded, and the Rf value was computed by dividing the distance travelled by the spots by the distance travelled by the front of the mobile phase.

**Calculation of Rf Value-** Distance travelled by solute from origin line /Distance travelled by solvent from origin line

## RESULTS AND OBSERVATION

The observations and the results of the present study are tabulated below.

**Pharmacognostical analysis**

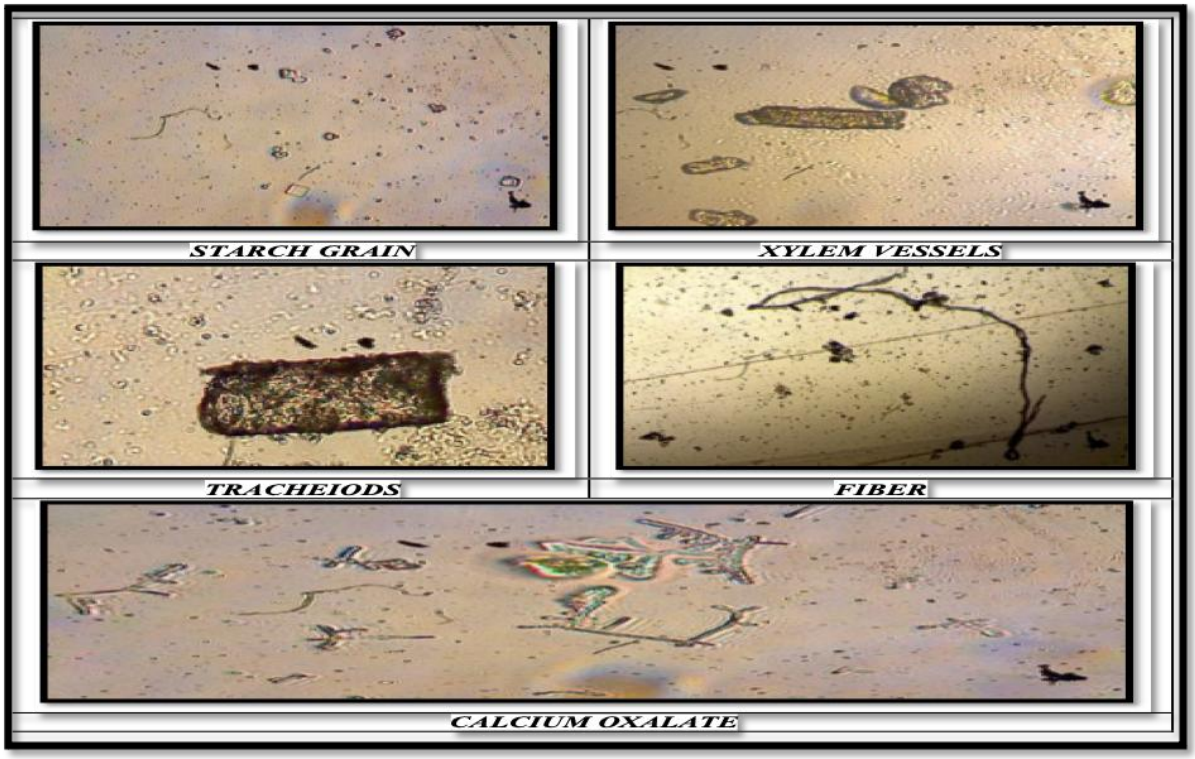
**Photo-01- Macroscopic study of *Amra [Mangifera indica Linn.]***

**Table-02: Organoleptic characters of dried *Amra* [*Mangifera indica Linn.]***

| S. No | Parameters | Observations |
|---|---|---|
| 1 | Color | Light Ivory |
| 2 | Odor | Characteristic |
| 3 | Taste | Bitter |
| 4 | Texture | smooth, Leathery |

**Powder microscopy**[12]: The presence of fibres, starch, crystals, oil glands and parenchyma were observed as shown in Figure.

**Photo-02- Powder Microscopy of *Amra* [*Mangifera indica Linn.]***




**Physiochemical analysis-**

**Moisture content of sample-**

**Table-03: Moisture content of sample of *Amra* [*Mangifera indica Linn.]***

| S.NO. | Weight of sample | Weight of container | Weight after drying with container | Weight after drying without container | Value% |
|---|---|---|---|---|---|
| 1. | 4.9803 gm | 60.8575 gm | 65.3273 gm | 4.4698 gm | **10.25%** |

**pH value of sample-**

**Photo-03: pH value of sample of *Amra* [*Mangifera indica Linn.]***

**Table-04: pH value** of sample of ***Amra [Mangifera indica Linn.]***

| S.NO. | Sample | pH |
|---|---|---|
| 1. | *Amra [Mangifera indica Linn.]* | 4.9 |

**Extractive value of sample-**

**Photo-04: Extractive value of sample of *Amra [Mangifera indica Linn.]***

**Table-05: Extractive value of sample of *Amra [Mangifera indica Linn.]***

| S.NO. | Extractive values | Sample weight | Beaker weight | Beaker +extract weight | Extract weight | Extract value (%) |
|---|---|---|---|---|---|---|
| 1. | Alcohol soluble extractive Value | 5.0119 gm | 138.21gm | 138.8605 gm | 0.6505 gm | **12.98%** |
| 2. | Water soluble extractive value | 5.0120 gm | 146.023 gm | 146.7583 gm | 0.7353 gm | **14.67%** |

**Ash value of sample-**

**Photo-05: Ash value of sample of *Amra [Mangifera indica Linn.]***

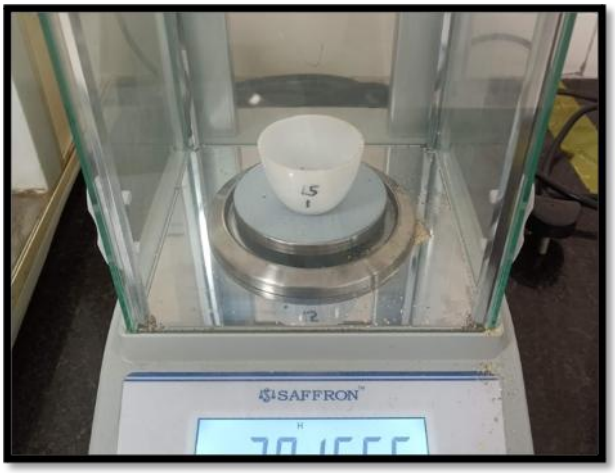


**Table-06: Total Ash value of sample of *Amra* [*Mangifera indica Linn.]***

| S.NO. | A1 | X | A2 | Total ash (%) |
|---|---|---|---|---|
| 1. | 39.7840 gm | 4.9770 gm | 39.8895 gm | **2.12%** |

**Table-07: Acid Insoluble Ash value of sample of *Amra* [*Mangifera indica Linn.]***

| S.NO. | X | G1 | G2 | G3 | Total ash (%) |
|---|---|---|---|---|---|
| 1. | 4.9770 gm | 39.7840 gm | 39.7989 gm | 0.0149 gm | **0.3%** |

**Table-08: Water Soluble Ash value of sample of *Amra* [*Mangifera indica Linn.]***

| S.NO. | X | A | G1 | G2 | G3 | Total ash (%) |
|---|---|---|---|---|---|---|
| 1. | 5.0058 gm | 0.1061 gm | 31.5600 gm | 31.5935 gm | 0.0335 gm | **1.45%** |

**Phytochemical study-**

**Photo-06: Phytochemical study of *Amra* [*Mangifera indica Linn.]***

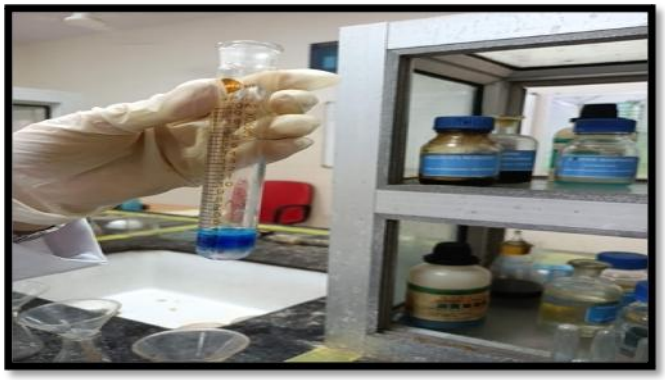


**Table-09: Observations of Phytochemical parameters of *Amra* [*Mangifera indica Linn.]***

| Phytochemicals | Tests | Aq. Ext of *Amra* | Al. Ext of *Amra* |
|---|---|---|---|
| **Carbohydrates** | 1.1-Molish test | + | + |
| | 1.2- Benedict test | + | - |
| | 1.3-Fehling test | + | + |
| **Alkaloids** | 2.1-Dragendorff test | + | + |
| | 2.2-Wagner test | + | + |
| | 2.3-Hager test | - | - |
| **Amino acids** | 3.1- Ninhydrine test | + | - |
| **Proteins** | 4.1-Biuret test | + | - |
| | 4.2-Xanthoprotic test | + | - |
| | 4.3- Millon test | + | + |
| **Saponin** | 5.1-Foam test | + | - |
| **Glycosides** | 6.1- Borntrager test | - | - |
| **Phenolic Compound** | 7.1- Phenolic test | - | + |
| **Steroids** | 8.1- Salkowaski test | + | - |
| **Tannins** | 9.1-$Fecl_3$ | - | + |
| | 9.2- Lead acetate | - | - |
| | 9.3-Pot. Dichromate | + | + |

**Chromatography study**

Visualization was done under normal light and Iodine.

**Table-10: Results of TLC of *Amra* [*Mangifera indica Linn.]***

| Distance of solvent | R F Value | IMAGE |
|---|---|---|
| 5.0 | 0.37 |  |
| | 0.63 | |
| | 0.82 | |
| | 0.89 | |

## DISCUSSION

- **Pharmacognostical study-**sample is organoleptically within the limits. Table-02 and Photo-01-02 shows the presence of fibers, starch, crystals and oil glands in the sample.
- **Physiochemical analysis-** sample is stable as it has normal moisture level. The ash value which is less than 5 percent is indicating the authenticity and purity of the present sample. Extractive values within the standards indicate the absence of exhausted or adulterated drugs in the sample.
- **Phytochemical study-** The water extract of sample had shown positive results for the presence of carbohydrates, alkaloids, amino acids, proteins, saponins, steroids and tannins. The alcohol extract shows presence of carbohydrates, alkaloids, proteins, phenolic compounds and tannins.
- **Chromatography study-**TLC of the Alcohol extract of sample shows bands at Rf-0.37, 0.63, 0.82 and 0.89.

**CONCLUSION:** On the basis of the observations, results and discussions it has been concluded that the present sample of *Amra* [*Mangifera indica Linn.]* is within all the standards of quality. All the Pharmacognostical, Physiochemical, Phytochemical and Thin Layer Chromatography study helped in identification and authentication of the sample of *Amra* [*Mangifera indica Linn.].*

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