



PHYTOCHEMICAL STUDIES OF BIOACTIVE COMPOUNDS FROM *Cucumis melo* LINN.

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Objective: The present study deals with the qualitative identification of phytochemical constituents from *Cucumis melo* Linn fruits and seeds

Method: The methanolic extract of both fruit and seed was subjected to phytochemical screening by qualitative and HPTLC methods.

Outcome: The analysis revealed that the extract of *Cucumis melo* Linn is rich in phytochemical compounds like Alkaloids, Essential oils, Flavonoids, Phenolic compound, Steroids, Tannins and Triterpenes. The qualitative HPTLC analyses of the methanolic extract of the *Cucumis melo* fruit and seed extract showed the presence of Alkaloids, Essential oils, Flavonoids, Phenolic compound, Steroids, Tannins and Triterpenes. Further research is warranted for screening the biological activities of these extracts.

ABSTRACT

1. INTRODUCTION

Cucumis melo Linn belonging to Cucurbitaceae family, commonly known as Musk melon or sweet melon (in English), Kharbuja (in Hindi), Ervaru (in Sanskrit). Leaves about 7.5 cm diameter, orbicular- reniform in outline, 5-angled or lobed, scabrous on both surfaces and also often with soft hairs; lobes not deep nor acute; petiole 5 cm; petals 1.6 cm. Female peduncle sometimes 5 cm. Fruit spherical ovoid elongated or contorted, glabrous or somewhat hairy, not spinous nor tuberculate¹.



Cucumis melo fruits

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The unripe fruit is bitter, sour; may cause skin eruptions and strangury. The ripe fruit is sweet, oily, and wholesome. The fruits are of different kinds; sweet, acrid, and sour. The fruits are tonic, laxative, galactagogue, diuretic, strengthens the heart, the brain, and the body in general cures ophthalmic, urinary discharges; causes congestion of the eyes in plethoric people gives headache; may cause indigestion. The oil from the seeds is said to be very nourishing. Not only the seeds but the pulp of the fruit is a powerful diuretic, very beneficial in acute and chronic eczema. In China and Japan, the stalks of the fruit are considered cooling and demulcent. It is widely used in cosmetics such as skin lotions which contain melon juice.

Phytochemical work by Mariod.A et al (2008)² indicated that isomultiflorenol is the major component, accompanied by its $\Delta 7$ -isomer, multiflorenol, in the triterpene alcoholic fractions of the unsaponifiable matter of *C. sativus* and *C. melo* seed lipids. Skin protecting cosmetic liposomes comprise Ca ion and/or Mg ion and superoxide dismutase from melon concentrates³. Fruits contain ferulic, caffeic and chlorogenic acids. Fruit stalk contains cucurbitacin B and E⁴. Methanolic extract of *Cucumis melo* fruit contains a saponin (C₄₀ H₆₄ O₁₆, mp.158-59°) which is identified as stigmasta-7-16-25(26) triene-3-O- β -D-glucopyranosyl (15)-O- β -D- Xylofuranoside. Presence of curcumin and leptodermin is also reported in the fruits⁵.

The seeds of melon contain multiflorenol, isomultiflorenol, 24-methylenecycloartenol, α - and β -amyrin, teraxerol, lupeol, euphol, 24-methyl-25(27)-dehydrocycloartanol, 24-methylene-24-dihydrolanosterol, 24-methylene-24-dihydroparkeol, tirucallol and cycloartenol⁶.The seeds contain myristic acid, phosphates, galactane, lysine, citrulline, histidine, tryptophan, cystine⁷. Secondary metabolites such as alkaloids, flavonoids, terpenoids, sterols, carbohydrates, saponins and phenolic compounds were detected to be present in the fruits of *Cucumis Melo* plant⁸.The present aim of the study is to identify the various phytochemical constituents present in the fruit and seed of *Cucumis melo* Linn.

2. MATERIALS AND METHODS

2.1. Collection and Extraction of Plant materials

The plant materials, fruit and seed for extraction was collected from Vadanerkulam part of Tindivanam area in Villuparam district of Tamilnadu and authenticated by Dr.KV.George at School of Environmental Science, M.G.University, Kottayam.

The collected materials were thoroughly washed, cleaned, chopped dried under shadow and powdered. The powdered sample (10 g of both fruit and seed) were taken for extraction in 250 ml 100% methanol by Soxhlet extraction method for 8 hrs. The extract was filtered and then concentrated using Rotary vacuum evaporator.The percentage yield of different extracts are presented in Table.1

Table 1: The percentage yield of different extracts

Solvent	% Yield
Methanol	22.8% w/v
Isopropyl Alcohol	14.8% w/v
Petroleum Ether	6.4% w/v
n-hexane	9.8% w/v

2.2. Preliminary Phytochemical Screening

In order to determine the presence of major classes of phytochemicals, different evaluation parameters were carried out and the presence of phytochemicals were confirmed by thin layer chromatography.

2.3. Screening of phytochemical groups using HPTLC

HPTLC is a flexible, reliable, and cost-efficient separation technique ideally suited for the analysis of botanicals and herbal drugs. The Camag HPTLC instrument consisting of Linomat V automatic spotter equipped with a 100 μ L syringe connected to a nitrogen cylinder, Scanner-III, twin-trough developing chambers, and viewing cabinet with dual wavelength UV lamps (Camag, Muttenz, Switzerland) were used. HPTLC plates used were of aluminium backed silica gel 60 F₂₅₄ with 0.2mm thickness. Before analysis, HPTLC plates were cleaned by predevelopment with methanol and activated at 110°C for 5min for solvent removal. Specific mobile phases were used for each phytochemical.

2.3.1. Sample application

10 μ l of sample was spotted on pre-coated TLC plate in the form of narrow bands (8 mm) with 10 mm from the bottom and at least 15 mm from left and right edges of the plate using Linomat V spotter. Samples were applied under continuous dry stream of nitrogen gas at constant application amount 10 μ l.

2.3.2. Mobile phase and migration

The spotted plates were developed using different mobile phases to detect the various classes of phytochemical. The proportion of the chemicals in the mobile phases^{9,10} is as follows:

Alkaloid - Toluene: Methanol: Diethyl amine (8:1:1)

Flavonoids - Toluene: Ethyl acetate: Formic acid (7:3:0.1)

Essential oils - Toluene: Ethyl acetate (8.5:1.5)

Phenolic compounds- THF: Toluene: Formic acid: Water (16:8:2:1)

Saponins- Chloroform: Acetic acid: Methanol: Water (6.4:3.2:1.2:0.8)

Steroids - Toluene: Methanol: Acetone (6:2:2)

Tannins- Ethyl acetate: Acetic acid: Ether: Hexane (4:2:2:2)

Triterpenes- Toluene: Chloroform: Ethanol (4:4:1)

Linear ascending development was carried out in 10 x 10 cm twin trough glass chamber equilibrated with mobile phase. The optimized chamber saturation time for mobile phase was 20minutes at 25 \pm 2°C with a relative humidity of 60 \pm 5%. Ten milliliters of the mobile phase (5 ml in trough containing the plate and 5 ml in other trough) was used for the development and allowed to migrate a distance of 85 mm from the point of sample application. After development, TLC plate was dried and the chromatogram was viewed at 254 nm and 366 nm to visualize and detect various phytochemical constituents.

2.3.3. TLC Plate development^{4,6}

The TLC plates were developed with the following reagents to detect the various classes of phytochemical compounds.

Alkaloids - Dragendorff reagent

Essential oils, Saponins, Steroids and Triterpenes–Anisaldehyde sulphuric acid

Flavonoids - NP/PEG Reagent

Phenolic compounds and Tannins- Fast blue salt B

2.3.4. Documentation

The various conditions for documentation were selected. The plates were photographed in various conditions under UV 254 nm, UV 366 nm and UV 366 nm after derivatization.

3. RESULTS AND DISCUSSION

3.1. Preliminary Phytochemical Screening

Methanolic extract of both fruit and seed were subjected to preliminary qualitative tests for the detection of major phytochemical groups using standard protocols⁽²³⁾. The analysis revealed the presence of alkaloids, Flavonoids, Essential oil, Phenolic compounds, Tannins, Steroids, and Triterpenoids. The results are presented in Table 2.

Table 2: Preliminary Phytochemical screening studies

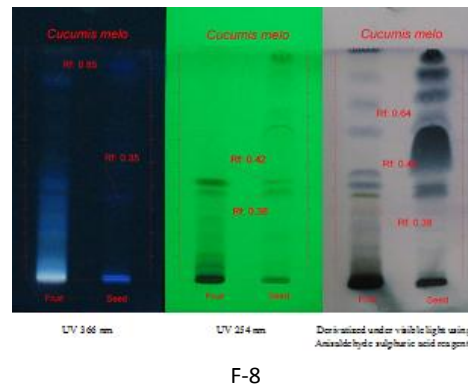
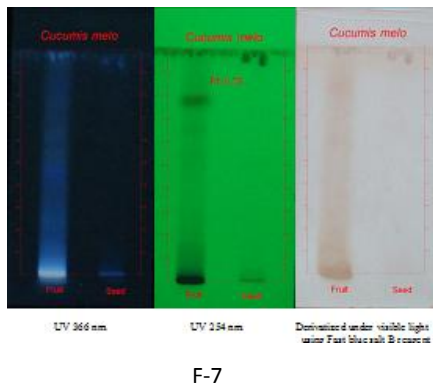
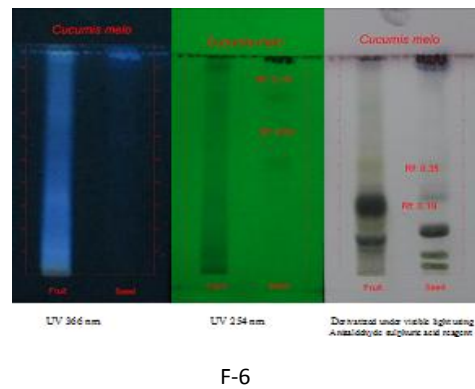
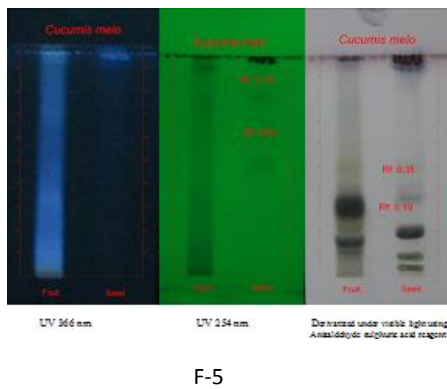
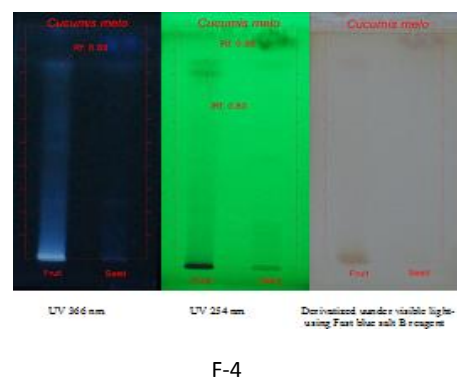
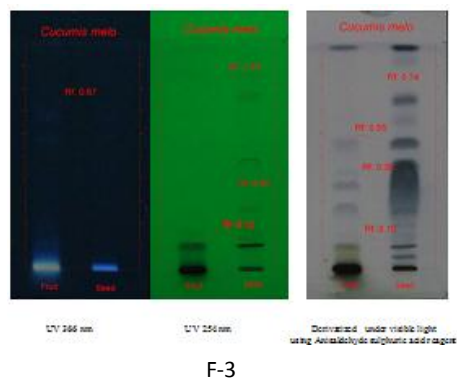
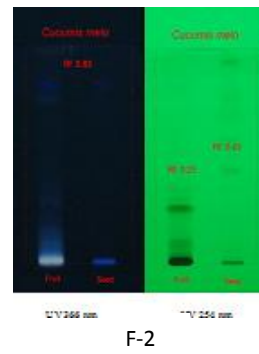
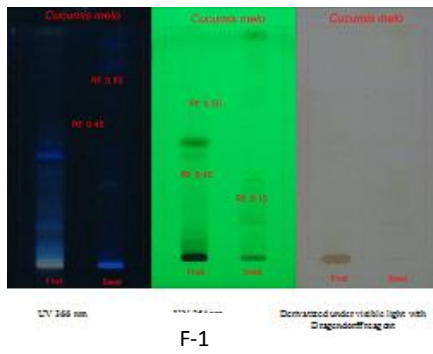
Sr. No.	Constituents	Test	Fruit Extract	Seed Extract
1.	Alkaloids	Dragendorff test	+	+
		Mayer's test	+	+
		Wagner's test	+	-
2.	Flavonoids	With 1% KOH	+	+
		With H ₂ SO ₄	+	-
		Lead acetate test	+	+
3.	Saponins	Foam test	-	-
4.	Steroids and Triterpenoids	Salkowski's test	+	+
		Liebermann Burchard test	+	+
5.	Tannins and Phenolic compounds	FeCl ₃ test	+	-

3.2. HPTLC analysis

The results obtained from HPTLC analysis of the methanolic extract of *Cucumis melo* with respect to Alkaloids, Essential oils, Flavonoids, Phenolic compounds, Saponins, Steroids, Tannins and Triterpenes are given in Table No:3 and corresponding figures are represented in Figure No.1 to 8

Table 3: HPTLC analysis of the methanolic extract of *Cucumis melo*

Sr. No.	Compounds	Rf Values	
		Fruit Extract	Seed Extract
1	Alkaloids	0.45, 0.50	0.15, 0.83
2	Essential oils	0.10, 0.36, 0.55, 0.67	0.10, 0.25, 0.55, 0.74
3	Flavonoids	0.25, 0.83	0.43, 0.83
4	Phenolic compounds	0.80, 0.83, 0.86	-
5	Saponins	-	0.19, 0.35, 0.50, 0.79
6	Steroids	0.56, 0.65, 0.73	0.26, 0.65, 0.73
7	Tannins	-	0.78
8	Triterpenes	0.38, 0.42, 0.64, 0.85	0.35, 0.38, 0.42, 0.64



F-1: Alkaloids, F-2: Flavonoids, F-3 Essential oil, F-4 Phenolic compounds, F-5 Saponins, F-6 Steroids, F-7 Tannins, F-8 Triterpenes

4. CONCLUSION

The present study was aimed to extract the phytochemical constituents from *Cucumis melo Linn.* Different extraction processes were carried out to get maximum yield. From the various extraction processes the methanolic extract of both fruit and seed showed good yield (Table.1). The obtained extract was subjected to phytochemical screening by qualitative and HPTLC methods. The phytochemical screening revealed that the extract of *Cucumis melo Linnis* are rich in phytochemical compounds like Alkaloids, Essential oils, Flavonoids, Phenolic compound, Steroids, Tannins and Triterpenes which can be further isolated and evaluated for potent biological activity.

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