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PHYTOCHEMICAL AND PHYSICOCHEMICAL PROPERTIES OF *HIBISCUS ROSA SINENSIS* LEAVES EXTRACT: A COMPARISON BETWEEN CONVENTIONAL AND MICROWAVE ASSISTED EXTRACTION

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ABSTRACT
The extraction of the herbal drugs are need to be analysed to give the documentary prove of the herbal drugs against many diseases. *Hibiscus rosa sinensis* is one of the famous herbal medicine used for the treatment of bleeding soother irrited tissue and relxes spams. Various extract of *Hibiscus rosa sinensis* is also used for the hair growth. *Hibiscus rosa sinensis* belongs to the Malvaceae family. In the present research work the comparersion of conventional extraction (CE) and microwave assisted extraction (MAE) was done. It is observed that both are hydro extract but gives different results for phytochemicals and physicochemical properties. Carbohydrate, glycosides proteins, amino acid, phenolic compounds and tannins were present in both the extracts. The CE is found to be active against *E.coli, B.Subtilis, S.Aureus* bacteria while MAE is found to be inactive.

KEYWORDS: Phytochemical, physicochemical, microwave extract, aqueous extract, antibacterial, ant-tuberculosis, anti-malarial.

INTRODUCTION
Beautiful flowers of *Hibiscus rosa sinensis* is well known in India. It belongs to the Malvacacae and commonly known as jasvant in India.[1] The leaves of the plant is known for the treatment of antidiarrhetic and are antipertensive and antiphlogestic activities.[2] In ayurvedic system of medicine *Hibiscus rosa sinensis* is widely used in India as a demulcent refrigerant drink in fever and decoction is given in bronchial catarsh. Sikawar Mukesh and et al[3] also reported the antihyperlipidemic activity from ethanolic extract of *Hibiscus rosa sinensis*. Anti-implantation and antispermatogenic activities are possess by flowers of *Hibiscus rosa sinensis*. Leaves and flowers are also possess hypoglycemic activity. The petroleum ether extracts of the leaves and flowers have been shown to increase the hair growth. Antifungal, insect-repelling and toxic activities[4] bleeding, soother irritated tissues and relaxes spasms. It contains anodyne, operient, emollient and laxatine leaves. The flowers are hermaphrodite and are pollinated by insects.[5] Flowers of *Hibiscus rosa sinensis* are found to be more effective against arterial hypertension. India intended for hair growth in the market by the herbal products included various extract of *Hibiscus rosa sinensis* Linn. N Adhirajan[6] found the leaf extract is more effective to increase the length of hair significantly higher as compare to flower extract.

MATERIAL AND METHODS
Collection of plant leaves
Plant leaves were collected from the local area near to Maulana Azad College Aurangabad. The leaves were washed gently and dried under shade. It was grind and used for the analysis.

Fluorescent test
0.5 gm of samples were added in different solvents and fluorescent behavior was observed. Normal light florescent behavior was different in different solvent.

Ash analysis
Accurately weighted 10 gm of sample was taken in finely clean silica crucible and ignited for 4 hrs with gradually increasing in temperature up to 300^{o}C. After ignition of leaves of plant, the residue was remain is designated as ash. The residue was again ignited with the interval of 10 min, till to get the constant weight. This ash was used to determine the three parameters called as total ash, acid insoluble ash and water soluble ash.

Acid insoluble ash
Acid insoluble ash, ash which is insoluble in dilute HCl. 1 gm of total ash was dissolved in 2 N hydrochloric acid. Stirred well for the digestion of ash and filtered through wattman filter paper no. 41. The residue remain after

filtration is ignited in clean silica crucible by gradually increase in temperature up to 300°C. The residue was cooled and weighted and again kept for ignition till to get the constant weight. The residue is remain after ignition is acid insoluble ash. The percentage was calculated for the acid insoluble ash.

Water soluble ash

1 gm of total ash was boiled with20ml of double distilled water. The residue was collected by filtered through wattman filter paper no 41. Residue was washed with hot water and kept for ignition not more than 400°C. The weight of residue was subtracted from total ash. This difference between residue and total ash represent the water soluble ash. The percentage was calculated.

Bulk density

Bulk density and tab density were determined by densitometer (Bio Technics India, serial No. 40195, Model BTI-09). 50 cm^3 of powder was introduced into the 50 ml graduated cylinder. The dropping interval of the cylinder was two sec at the height of 2 cm three times on the hard wooden surface. The bulk density was calculated by dividing the weight of the sample in grams by the final volume in cm^3 of the sample contained in the cylinder.

Tapped density

50cm^3 of powder was introduced into the 50 ml graduated cylinder. The dropping interval of the cylinder was two sec at the height of 2 cm 100 times on the hard wooden surface. The tab density was calculated by dividing the weight of the sample in grams by the final volume in cm^3 of the sample contained in the cylinder.

The compressibility of the powder was evaluated using the HR (Housner Ratio). The Housner ratio may be defined as ratio of tap density and bulk density.

$$HR = \frac{TD}{BD}$$

Carr's Index (Compressibility Index- CI): This was calculated by using the formula:

$$CI = \frac{Dt - Db}{Dt} \times 100$$

Extraction procedure

Conventional Extraction

Accurately weight 30 gms of sample was introduced into the 500 ml round bottom flask (which was first clean by very dilute hydrochloric acid and then distilled water) with 300 ml double distilled water. The pourciline pieces were add to avoid bumping of the sample. The condenser was fitted with circulation of water. The sample was refluxed on flame for six hrs. The sample was cooled and filtered by the suction pump. The excessive water was evaporated for the preservation of the sample and it was kept at 4°C for 12 hrs. The percentage of the extract was calculated.

Microwave Assisted Extraction

30 gms of the sample was kept in the clean round bottom flask. 300ml of double distilled water was used as the solvent. The porciline pieces were added to avoid bumping of the sample. The condenser was fitted with circulation of water. The sample was refluxed by microwave radiations using microwave oven (Catalyst microwave synthesizer Sr. No. 130602954) for 30 min at 60% power, 420 watt and 120°C. The sample was cooled and filtered by the suction pump. The excessive water was evaporated for the preservation of the sample and it was kept at 4°C for 12 hrs. The percentage of the extract was calculated.

Physicochemical test

Physicochemical parameters like relative density, viscosity, surface tension and refractive index were measured of the solutions of different ppm.

Relative density

Clean and dry empty density bottle with stopper weighted accurately. The density bottle was filled with double distilled water up to it fall from the bottle and stopper was fitted and the bottle was cleaned from outside. The bottle was weighted.

The procedure was repeated for the samples. The density was measured by taking difference between bottle with sample and empty bottle. Relative density was calculated by the formula,

$$\text{Relative density} = \frac{\rho_2}{\rho_1}$$

Where ρ_1 is density of the water and ρ_2 is the density of the sample.

Viscosity

The different concentration in ppm of the samples were prepared by using double distilled water. The Ostwald's viscometer was cleaned by NaOH to remove greasy impurities than with chromic acid and finally with the distilled water. The 10 ml of double distilled water was inserted in viscometer from large diameter tube. And the sample was sucked through second tube of the same viscometer till it rises with 2-3 cm above the mark. By keeping stop watch ready the liquid was allowed to decent down the time required to flow of the liquid between two points was noted. The same procedure was repeated for the samples which have to study.

Viscosity was measured by sing formula,

$$\eta_2 = \frac{t_2}{t_1}\frac{\rho_2}{\rho_1}\eta_1$$

Surface tension

The different concentration in ppm of the samples were prepared by using double distilled water. The stalagmometer was cleaned by NaOH to remove greasy impurities than with chromic acid and finally with the distilled water. The rubber tubing with the with a screw

clip was attached to the top of the stalgmometer. The flat end of the stalagmometer was dipped into the standard solution (double distilled water) suck through the water tubing until the water level rises above the mark. The screw was adjust the pressure until the rate of the drop was 10 to 15 per minute. The number of drops were counted for double distilled water when passes from upper mark to the lower mark.

The stalagmometer was removed and rinse with alcohol and dried. Stalagmometer was filled with the test sample and number of drops were determine. Same procedure was repeated for every concentration three times and mean was taken.

The surface tension was than calculated by the formula,

$$\gamma_2 = \frac{n_1}{n_2}\frac{\rho_2}{\rho_1}\gamma_1$$

Where γ_1 and γ_2 are the surface tension of the double distilled water and the sample respectively.

And n_1, ρ_1 and n_2, ρ_2 are the number of drops and relative densities of the double distilled water and samples under study respectively.

Refractive index

Refractive index depends upon temperature and concentration. However the specific refraction is independent of temperature and it is characteristic property of the liquid. The refractive index was measured by Abbe's refractometer. The refractometer was placed on the table near to the window so that sufficient light should reached to the prism. The prism box was opened b turning the lock nut and both the phases of the prism was cleaned with the help of cotton wool by the acetone and prism box was closed after drying. Few drops of the liquid were pumped through the small hole on the prism box with the help of the dropper. The crosswire of the telescope was focus by rotating the eye piece and the mirror was adjust for the reflection of maximum light towards the prism box. The prism box was moved forward and backward until the clear boundary between the light and dark region was appear. The scale reading was noted down. And refractive index were calculated by using the formula,

$$\text{Specific refraction (r)} = \frac{n^2-1}{n^2+2}\mathbf{X}\ \frac{1}{\rho}$$

Where ρ is the density of the samples.

Qualitative Phytochemical Screening

The different qualitative chemical tests were performed for establishing profile of given extract for its chemical composition. Qualitative Phytochemical analysis was done using the following procedures.

Detection of alkaloids

Solvent free extract, 50 mg was stirred with 5 mL of dilute hydrochloric acid and filtered. The filtrate was tested carefully with various alkaloidal reagents as follows.

i) Mayer's test: To a few mL of filtrate, two drop of Mayer's reagent added by the side of test tube. A white or creamy precipitate indicated the test as positive.

Mayer's reagent: Mercuric chloride (1.36 g) was dissolved in 60 mL of water and potassium iodide (5.0 g) was dissolved in 10 mL of water. The two solutions were mixed and made up to 100 mL with water.

ii) Wagner's test: To a few ml of filtrate, few drops of Wager's reagents were added by the side of the test tube. A reddish-brown precipitate confirmed the test as positive.

Wagner's reagent

Iodine (1.27 g) and potassium iodide (2 g) was dissolved in 5 ml of water and made up to the 100 ml with distilled water**.**

iii) Hager's test: To a few ml of filtrate 1 or 2 of Hager's reagent (saturated aqueous solution of picric acid) were added. A prominent yellow precipitate indicated the test as positive.

Detection of Carbohydrate

The extract 100 mg was dissolved in 5 ml of water and filtered. The filtrate was subjected to the following tests.

i) **Molisch's tests:** To 2 ml of filtrate, two drops of alcoholic solution of α-naphthol were added, the mixture was shaken well and 1 ml of concentrated sulphuric acid was added slowly along the sides of test tube and allwed to stand. A violet ring indicates the presence of carbohydrates.
ii) **Fehling's test:** One ml of filtrate was boiled on water bath with 1 ml each Fehling's solutions A and B. Red precipitate indicates the presence of suger.

Fehling's solution

Solution A; copper sulphate (34.66 g) was dissolved in distilled water made up to 500 ml with distilled water.

Solution B: potassium sodium tartarate (173 g) and sodium hydroxide (50 g) was dissolved in water and made up to 500 ml.

iii) **Benedict's test:** To 0.5 ml of filtrate, 0.5 ml of Benedict's reagent was added. The mixture was heated on a boiling water bath for 2 min. A characteristics colored precipitate indicates the presence of sugar.

Benedict's reagent

Sodium citrate (173 g) and sodium carbonate (100 g) were dissolved in 800 ml distilled water and boiled to

make it clear solution. Copper sulphate (17.3 g) dissolved in 100 ml distilled water.

iv) **Barfoed's test:** To 1 ml of filtrate, 1 ml of Barfoed's reagent was added and heated on a boiling water bath for 2 min. red precipitate indicates the presence of suger.

Barfoed's reagents: copper acetate 30.5 g was dissolved in 1.8 ml of glacial acetic acid.

Detection of glycosides

For the detection of glycosides, 50 mg of extract was hydrolysed with concentrated hydrochloric acid for 2 hrs on water bath, filtered and the hydrolysate was subjected to the following test.

i) **Borntrager's test:** To 2 ml of the filtrate hydrolysate, 3 ml of chloroform was added and shaken, chloroform layer was seprated and 10% ammonia solution was added to it. Pink color indicates the presence of glycosides.
ii) **Legal's test:** Fifty mg of extract was dissolved in pyridine, sodium nitroprusside solution was added and made alkaline using 10% sodium hydroxide. Presence of glycoside was indicated the pink color**.**

Detection of Saponins by Foam test

The 50 mg was diluted with distilled water and made up to 20 mL. The suspension was shaken in a graduated cylinder for 15 min. A two cm layer of foam indicated the presence of saponins.

Detection of proteins and Amino acids

The extract (100 mg) was dissolved in 10 ml of distilled water and filtered through Whatman filter paper no. 41 and filtrate was subjected to test of proteins and amino acids.

i) **Millon's test:** To 2 mL of filtrate, few drops of Millon's reagent were added. A white precipitate indicated the presence of proteins.

Millon's reagent

Mercury (1 g) was dissolved in 9 mL of fuming nitric acid. When the reaction was completed, equal volume of distilled water was added.

ii) **Biuret test:** An aliqot of 2 ml of filtrate was treated withone drop of 2% copper sulphate solution. To this 1 ml of ethanol (95%) was added, followed by excess of potassium hydroxide pellets. Pink color in the ethanolic layer indicates the presence of proteins.
iii) **Ninhydrine test:** two drops of ninhydrine solution (10 mg of ninhydrine in 200 ml of acetone) were added to two ml of aqueous filtrate. A characteristic purple color indicates the presence of amino acids.

Detection of phenolic compounds and tannins

i) **Ferric chloride test:** The extract (50 mg) was dissolved in 5 ml distilled water. To this few drops of neutral 5% ferric chloride solutions were added. A dark green color indicates the presence of phenolic compounds.
ii) **Gelatin test:** The extract (50 mg) was dissolved in 5 ml of distilled water and 2 ml of 1% solutions of gelatin containing 10% sodium chloride was added to it. White precipitate indicates the presence of tannins.
iii) **Lead acetate tests:** the extract (50 mg) was dissolved in distilled water and 3 ml of 10% lead acetate solution was added. A bulky white precipitate indicates the presence of flavonoids compounds.
iv) **Alkaline reagent test:** an aqueous solution of the extract was treated with the 10% ammonium hydroxide solution. Yellow fluorescence indicates the presence of flavonoids.

Spectral analysis: UV-Visible

The powder of extract was dried and dissolved in distilled water to prepared solution of approximately 50 ppm. The spectra was recorded in the range from 190 to 800 nm by using double beam spectrophotometer of Model Elico-159 and λmax is determine from the software Spectra treat**.**

IR Spectra

The FTIR instrument IRT3000, JASCO, having serial no. B051061016, and the spectra were recorded using spectra manager. IR instrument is calibrated by using polystyrene. Spectra were recorded by using potassium bromide (KBr) of IR grade manufactured by Marck life sciences. KBr was kept in hot oven at 50^{o}C for half hour to free it from moisture. Spectra of that dry KBr is measured within IR range. The samples of leave extract were crushed to make it fine powder and mixed with dry KBr, and spectra of the samples were measured.

Anti-microbial study

Antibacterial activity was investigated using disc method. With the help of sterile wire loop, the test was inoculated into a test tube containing Mueller Hinton broth. The O.D of the inoculums was adjusted in between 0.08-0.1.

As per the composition, Mueller Hinton Agar was prepared by using sterile distilled water and was sterilized at 121oC at 15lb pressure for 15 min in an autoclave. The medium was cooled at room temperature and poured in sterile petri plates and were allowed to solidify. Bacterial inoculums was swabbed over the medium using sterile cotton swab. Sterile disc was placed on medium, on which 20 µl of complex suspension was added. Zone of inhibition were observed and measured after incubation at 30oC for 18-20 hrs.

RESULT AND DISCUSSION

Fluorescent test

The powdered samples were treated with different chemicals and observed with naked eyes. The results were summarized in table 1. It is observed that the dark green color of powder is retaining its color in benzene, faint in presence of butanol, ethanol, chloroform, acetic acid etc. It becomes blackish brown in strong inorganic acids or base. This can attributed to oxidation or reduction or incomplete decomposition of compounds and formation of carbon particles.

Table 1: Florescent test for leave powder.

Sr. No.	Solutions	Observation
1	Powder as such (P)	Dark green
2	P + n-butanol	Whitish green
3	P + Conc. HCl	Brown
4	P + Conc. HNO_3	Reddish orange
5	P + Conc. H_2SO_4	Blackish brown
6	P + Chloroform	Light whitish green
7	P + Ethanol	Whitish green
8	P + Glacial acetic acid	Light whitish green
9	P + 1N HCl	Almond caream
10	P +1N NaOH	Yellowish green
11	P + 5% HCl	Cream
12	P + 5% NaOH	Chocaloty
13	P + benzene	Dark green

Ash value

The total ash content of the powder is formed to be 6.3%. The acid insoluble ash was found to be 39%. Whereas water soluble as 18%. The ash is mostly consist of transition elements i.e. cobalt or magnesium. The tapped density and bulk density of the powdered material is found to be 0.5535 and 0.4169 respectively. Carr's index and housner's ratio is found to be 24.68% and 1.3277 respectively. B.V Basavaraj et al[7] also report the bulk density, tapped density, Carr's index and housner's ratio of *Hibiscus rosasinensis Linn*, from that they get the result as 0.42, 0.44, 4.54% and 1.047 respectively.

The results of present study are given in table 2.

Table 2: Ash analysis and densities of leave powder.

Sr. No.	Ash	Results
1.	Total Ash	6.3 %
2.	Acid Insoluble	39 %
3.	Water Soluble	18 %
4.	Bulk density	0.4169
5.	Tapped density	0.5535
6.	Housner Ratio	1.3277
7.	Carr's index	24.68%

Percent Extraction

The extractive values are given in table 3. It is less when conventional method is used. When the extraction is carried out by microwaves, the extractive value is more as well as the time of extraction required is also less. Mehran Moradalizadeh et al[8] report that percent extraction of microwave assisted hydro extraction (MAHE) and conventional hydro extraction (CHE) was compared and it was found that MAHE gives more percentage as compared to CHE with significantly shorter time of extraction (15 min against 3 hrs for hydro Extraction). Microwave assisted extraction has many advantages such as shorter extraction time, lesser solvent consumption, higher extraction rate and better products with lower cost.[9,10]

Table 3: Extractive value of *Hibiscus rosa sinensis* plant leaves.

Sr. No.	Solvent	Percentage
1.	Conventional Hydro Extraction	12.26
2.	Microwave Assisted Hydro Extraction	15.16

Physico-chemical properties

Physicochemical parameters determine were Relative density, viscosity, surface tension, Refractive index in table 4 and 5. The relative density and surface tension of CE was found to be higher. The refractive index was found to be less in MAE. The surface tension for the MAE found to be approximately constant over the range of concentration (fig1 to 4). The extract contains saponins which are surface active agents. They may get disperse into water molecules, reducing the surface

tension of the solutions. Sometimes these molecules may get aggragate to form micelle. The polar group present in the molecules may get oriented at particular concentration, known as critical micelle concentration (CMC). Such decrease in surface tension is reported in the form 72 mNm^{-1} (pure water) to 25mNm^{-1} for extract of *Pfaffia glomcrata* and *Hebanthe erintha* plant extract.[11] There are different complex macromolecules present in the plant extract. Some of them like polysaccharides are rich in hydroxyl groups. Hence they bind with each other and take up water and become rich in cis-OH group. They allow aggregation from chain to chain via hydrogen bonding. The hydration becomes more complicated due to the interchange cross linking.[12] Hence hydration affects the physic-chemical properties of extract.

Table 4: Physicochemical properties of CE of *Hibiscus rosa sinensis* Leaves.

Sr. No.	Solution in ppm	Relative density	Viscosity (Pascal sec)	Surface tension (Newton/meter)	Refractive index
1.	5	1.00192	0.83739	69.8346	0.99556
2.	10	0.99697	0.83326	67.0941	0.99898
3.	20	0.99849	0.83447	60.8967	0.99897
4.	40	0.99828	0.83436	62.8473	0.99919
5.	60	0.99707	0.83334	60.8101	1.00040
6.	80	0.99919	0.83512	60.9393	1.00056
7.	100	1.00282	0.83814	61.1610	0.99466

Table 5: Physicochemical properties of MAE of *Hibiscus rosa sinensis* Leaves.

Sr. No.	Solution in ppm	Relative density	Viscosity (Pascal sec)	Surface tension (Newton/meter)	Refractive index
1.	5	0.9988	0.9029	57.2031	0.9997
2.	10	1.0019	0.9057	60.9731	0.9947
3.	20	1.0032	0.8720	63.0109	0.9953
4.	40	0.9990	0.8684	57.2314	0.9995
5.	60	1.0002	0.8694	59.0553	0.9983
6.	80	1.0007	0.8698	57.3514	0.9978
7.	100	1.0009	0.8739	57.6200	0.9976

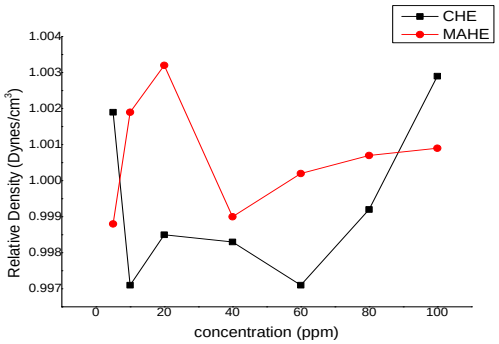

Fig. 2: Relative density of CE and MAE.

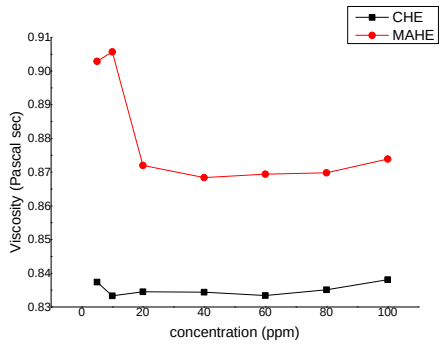

Fig. 1: Viscosity of CE and MAE.

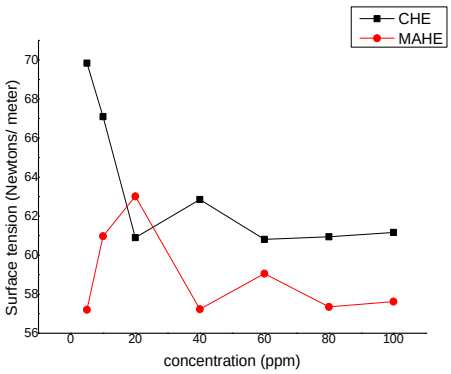

Fig. 3: Surface tension of CE and MA.

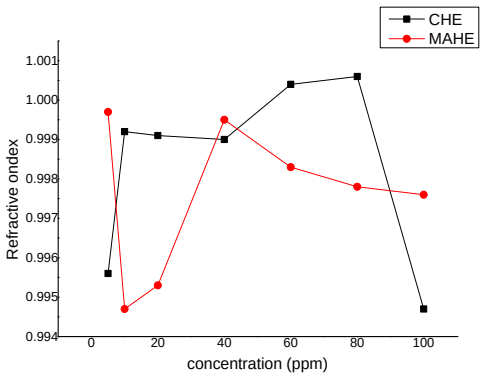

Fig. 4: Refractive index of CE and MAE.

Phytochemical test

Both extract shows almost identical Phytochemicals (table 6). The alkaloid gives negative test, except wagner's test in MAHE. The carbohydrate test positive, foam test also positive indicating presence of saponins. The proteins, amino acids, phenolic compounds and tannins are present in both the extracts. Faten R and et al found that 70% ethanol extract give positive test for carbohydrate glycosides, steroids/ terpenoids, flavonoids and tannins.[13] Uzama Danlami et al[14] shows that hexane wxtract of securinega virosa leaf shows maximum positive results for phytochemcal like saponins, alkaloids, flavonoids etc.

Table 6: Qualitative test for *Hibiscus rosa sinensis* leaves extract.

Sr. No.	Reagent	CE	MAE
1.	**Detection of Alkaloids**		
A.	Mayer's test	-ve	-ve
B.	Wagner's test	-ve	+ve
C.	Hager's test	-ve	-ve
2.	**Detection of carbohydrate**		
A.	Molish test	+ve	+ve
B.	Fehling's test	+ve	+ve
C.	Benedic test	-ve	-ve
D.	Barfoad's test	+ve	+ve
3.	**Detection of Glycosides**		
A.	Borntrager's test	-ve	-ve
B.	Legal's test	-ve	-ve
4.	**Test for saponins**	+ve	+ve
5.	**Detection of proteins and amino acid**		
A.	Millon's test	**-ve**	-ve
B.	Nitric acid test	+ve	+ve
C.	Biuret test	+ve	+ve
D.	Ninhydrine test	+ve	+ve
6.	**Detection of phenolic compound and tannins**		
A.	Ferricchlorid test	+ve	+ve
B.	Gelatin test	-ve	-ve
C.	Lead acetate test	+ve	-ve
D.	Alkaline reagent test	+ve	+ve

FTIR spectrum

The IR spectrum of extract of *Hibiscus rosa sinensis* was recorded from FTIR instrument IRT3000, JASCO, having serial no. B051061016. (fig5) Though it contains a mixture of compounds but still in order to find out various functional groups and a general finger print of samples, it will help. There are various IR bands observed. Which are represented in table 7.

Table 7: IR bands of *Hibiscus rosa sinensis* plant leaves extract.

Band (cm^{-1})	Intensity	Functional group
3382	Very broad	Inter molecular hydrogen bonding
1583	Sharp	Aldehydic ketone
1417	Sharp	C-H bending vibrations
1121	Broad	C-O-C asymmetrical stretching
770	Broad	Four adjacent hydrogen
657	Broad	Aromatic hydrocarbon
604	Broad	C=C bend

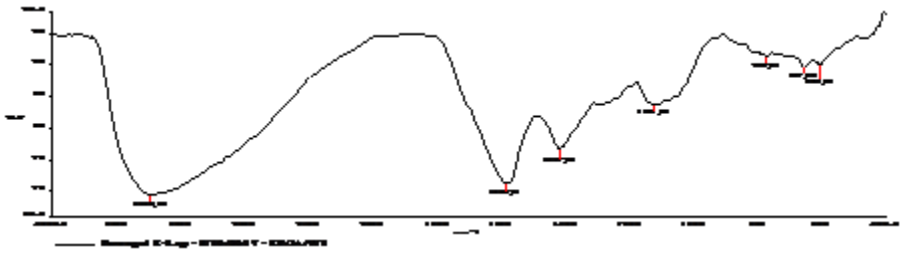

Fig. 5: FTIR spectrum of *Hibiscus rosa sinensis leaves extract.*

UV spectrum

The UV spectra of both the extract shows λmax at 196 nm and 206nm for conventional and microwave assisted extract respectively. The spectral trend in conventional and microwave assisted extract is somewhat similar (Fig 6 & 7).

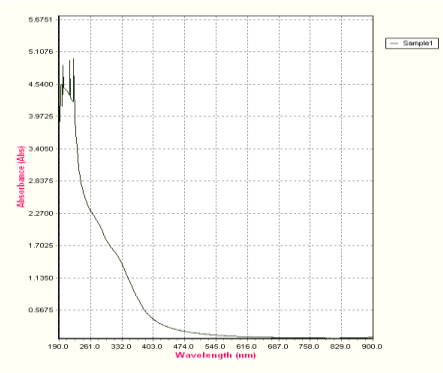

Fig. 6: UV-visible spectra of CE.

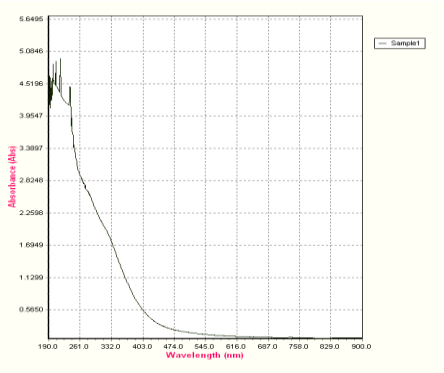

Fig. 7: UV-visible spectra of MAE.

Antibacterial study

The extracts are screened for the biological activity particularly for antibacterial activity. We observed that MAE is totally inactive against all the tested bacteria table 8. In MAE, sudden energy is supplied to the molecule, due to that some molecule may get decomposed by the heat generated inside the vessel.

Table 8: Antibacterial, anti tuberculosis and anti malarial properties of *Leasves extract.*

Sr. No.	Bacteria	MAE	CE
1.	***E. coli***	- -	Active
2.	**B. Subtillis**	- -	Active
3.	**S. Typhi**	- -	Inactive
4.	**S. Aureus**	- -	Active
5.	**T.B**	- -	-----
6.	**Maleria**	--	-----

CONCLUSION

Although the extractive value of microwave assisted hydro extraction is higher but the Phytochemical ingrideint and physicochemical properties differs compared to conventional hydro extraction. Even the biological activity is also changes with the type of extraction methods.

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