

Evaluation of Phytochemicals in polar and non polar solvent extracts of leaves of *Aegle marmelos* (L.)

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Abstract: *Aegle marmelos* (L.) Correa, commonly known as Bael, is a sacred tree for Hindu Religion and is native to northern India. The various phytochemicals from different parts of *Aegle marmelos* tree have already been investigated and isolated. Different phytochemicals have been isolated from the stem bark of *Aegle marmelos* such as marmesin-1''- α -L - rhamnopyranoside and 1, 5 -dihydroxy - 6 - methoxy -2 - methyl anthraquinone along with lupeol and β -sitosterol. The leaves of *Aegle marmelos* also yielded other chemical compounds such as Aegeline, Lupeol, Cineol, Citral and Eugenol. Moreover, Marmelosin, Luvangetin and Marmelide have been reported and isolated from fruits of *Aegle marmelos*.

In the present investigation the dried leaves of *Aegle marmelos* were powdered using a mixture grinder and extracted using six different solvents (five non polar viz. Acetone, Petroleum ether, Ethanol, Benzene and Methanol and one polar solvent, the Distilled water). All the six solvent extracts were assayed for qualitative and quantitative screening of their phytochemicals. All the six solvent extracts revealed the presence of various phytochemicals such as tannins, phlobatannins saponins, , terpinoids, diterpinoids, emodins, flavonoids, cardiac glycosides, anthraquinones, carotenoids, reducing sugars, alkaloids, anthocyanin, coumarins, steroids, phytosterols, phenol, fatty acids, proteins and amino acids. Of these 20 phytochemicals tannin, saponin, terpenoid, diterpinoid, emodin, flavonoid, cardiac glycoside, anthraquinone, carotenoid, reducing sugar, alkaloids, anthocyanin, coumarin, phenol, fatty acid, protein and amino acids were detected in all the six solvent extracts.

The leaves of *Aegle marmelos* contained a significant amount of alkaloid, flavonoids, phenolic, saponins and tannin content. The amount of flavonoids was maximum (42.75mg/gm) followed by phenols (23.85mg/gm), alkaloids (15.75mg/gm), saponins (14.65mg/gm) and tannins (12.35mg/gm).

The leaf extracts of *Aegle marmelos* (L.) Correa. and showed the presence of terpenoids, steroids and phytosterols, tannins, alkaloids, glycosides, saponins, reducing sugars, phenols and flavonoids. The extraction of various phytochemicals was seen to be more effectively done in polar solvents (ethanol, methanol and distilled water) than the non polar (Acetone, petroleum ether, benzene) solvents. Especially, ethanolic, methanolic and distilled water leaf extracts showed presence of most of the tested phytochemicals. Hence, it can be reported that alcoholic and aqueous extracts were the best for extracting the active principle than others.

Key words: *Aegle marmelos*, leaf extracts, polar solvents, non polar solvents, phytochemicals

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I. Introduction

Aegle marmelos (L.) is a spinous deciduous and aromatic tree of family Rutaceae with long, strong and axillary spines. This tree grows up to 18mt in height and thickness of tree is about 3- 4ft. Leaves are tri- to pentafoliate, leaflets are ovate and have typical aroma. Flowers are greenish white in colour and sweet scented. Fruits are large, woody, grayish yellow, 8- 15 celled and have sweet gummy orange coloured pulp. Seeds are compressed, oblong and numerous found in aromatic pulp.

Aegle marmelos (L.) Correa, commonly known as Bael, is a sacred tree for Hindu Religion, native to northern India, but is found widely throughout the Indian peninsula and in Ceylon, Burma, Thailand and Indo-China [1] (Bailey 1963). All parts of the tree viz. root, leaf, trunk, fruit and seed are used for treatment of many different diseases.

The constituents of *Aegle* are used in heart diseases [2] (Kakiuchi et al. 1991), inflammatory and wound healing [3] (Udupa et al. 1994). Leaves of *A. marmelos* have been reported as hypoglycemic effect [4,5] (Santhoshkumari and Devi 1990; Sharma et al. 1996). The essential oil from the leaves of *A. marmelos* is known

to exhibited antifungal properties [6,7] (Renu et al. 1986; Rana et al. 1997). Besides the medicinal uses, this plant was also studied for their antimicrobial, antifungal and insecticidal properties [8, 9] (Satyal et al., 2012; Kumar et al., 2008). The effect of leaf extracts of *Aegle marmelos* was also studied against *Anopheles subpictus* in their oviposition deterrent, ovicidal and repellent activities [10, 11] (Elango et al., 2009; Vineetha et al. 2009).

Higher and aromatics plants have been used traditionally in folk medicine as well as to extend the shelf life of foods, showing inhibition against bacteria, fungi and yeasts [12] (Hulin, et al. 1998). Biologically active compounds from natural sources have always been a great interest for scientists working on infectious diseases [13] (Perumal and Ignacimuthu, 2000). Today a substantial number of drugs are developed from plants which are active against a number of diseases. The majority of these involve the isolation of the active ingredient found in a particular medicinal plant and its subsequent modification. In the developed countries 25 percent of the medical drugs are based on plants and their derivatives [14] (Devi and Manoharan, 2011) and the use of medicinal plants is well known among the indigenous people in rural areas of many developing countries. In the past our ancestors have made new discoveries on the healing power of plants through trial and error. The medicinal plant therapy is based on the empirical findings of hundreds and thousands of years [15] (Fakim, 2006). *Aegle marmelos* Correa is commonly called as Bael in Hindi, and Bilva in Sanskrit. It belongs to the family Rutaceae. It is indigenous to India and is used in folk medicines. The Ayurvedic practitioners use almost all of their parts but the greatest medicinal value of its fruits [16] (Ariharan and Prasad, 2013). The leaves are used as astringent, laxative, febrifuge and expectorant. The leaves are useful in ophthalmia, inflammations, catarrh, diabetic and asthmatic complaints [17] (Chakraborty, 2012). The leaves are used for the heart and brain disorders.

The various phytochemicals from different parts of *Aegle marmelos* tree have already been investigated and isolated. Different phytochemicals have been isolated from the stem bark of *Aegle marmelos* such as marmesin-1st- α -L-rhamnopyranoside and 1, 5-dihydroxy-6-methoxy-2-methyl anthraquinone along with lupeol and β -sitosterol. The leaves of *Aegle marmelos* also yielded other chemical compounds such as Aegeline, Lupeol, Cineol, Citral and Eugenol. Moreover, Marmelosin, Luvangetin and Marmelide have been reported and isolated from fruits of *Aegle marmelos*.

II. Materials and Methods

In the present investigation the dried leaves of *Aegle marmelos* were powdered using a mixture grinder and stored in air-tight container for future use. Six different solvents (five non polar viz. Acetone, Petroleum ether, Ethanol, Benzene and Methanol and one polar solvent, the Distilled water) were used for preparation of solvent extracts. The dried plant sample was soaked separately with acetone, petroleum ether, ethanol, benzene, methanol and distilled water under reflux condition for the solvent extract preparation. About 1 gm of the dried sample of leaves was added respectively into the test tubes containing 5 ml of solvents, and was extracted at room temperature. In the present investigation the important phytochemicals of leaves of *Aegle marmelos* have been quantitatively analyzed for alkaloids, flavonoids, tannins, saponins and total phenols.

Phytochemical Analysis: Phytochemicals in leaves of *Aegle marmelos* were analyzed qualitatively and quantitatively in all the six solvent extracts

Qualitative Phytochemical Analysis

The extracts in all the six solvents of leaves of *Aegle marmelos* were tested for the presence of biological compounds by using following standard methods.

Test for Carbohydrates

Fehling's test

Equal volume of Fehling A and Fehling B reagents were mixed together and 2ml of it was added to crude extract and gently boiled. A brick red precipitate appeared at the bottom of the test tube indicated the presence of reducing sugars.

Benedict's test

Crude extract when mixed with 2ml of Benedict's reagent and boiled, a reddish brown precipitate formed which indicated the presence of the carbohydrates.

Iodine test

Crude extract was mixed with 2ml of iodine solution. A dark blue or purple coloration indicated the presence of the carbohydrate.

Test for Phenols and Tannins

Crude extracts were mixed with 2ml of 2% solution of FeCl_3 . A blue-green or black coloration indicated the presence of phenols and tannins.

Test for Flavonoid

Alkaline reagent test

Crude extracts were mixed with 2ml of 2% solution of NaOH. An intense yellow color was formed which turned colorless on addition of few drops of diluted acid which indicated the presence of flavonoids.

Test for Saponins (Frothing test)

Crude extracts were mixed with 5ml of distilled water in a test tube and it was shaken vigorously. The formation of stable foam was taken as an indication for the presence of saponin.

Test for Glycosides

Liebermann's test

Crude extracts were mixed with each of 2ml of chloroform and 2ml of acetic acid. The mixture was cooled in ice. Carefully concentrated H_2SO_4 was added. A colour change from violet to blue to green indicated the presence of steroidal nucleus, i.e., glycone portion of glycoside.

Salkowski's test

Crude extracts were mixed with 2ml of chloroform. Then 2ml of concentrated H_2SO_4 was added carefully and shaken gently. A reddish brown color indicated the presence of steroidal ring, i.e., glycone portion of the glycoside.

Keller-kilani test (Cardiac Glycosides)

Crude extracts were mixed with 2ml of glacial acetic acid containing 1-2 drops of 2% solution of FeCl_3 . The mixture was then poured into another test tube containing 2ml of concentrated H_2SO_4 . A brown ring at the inter phase indicated the presence of cardiac glycoside.

Test for Alkaloids:

The crude extract of all the six solvents was boiled in 10 ml methanol and filtered separately. 1% HCl was added followed by 6 drops of Dragendorff reagent, and the brownish-red precipitate was taken as evidence for the presence of alkaloids.

Phlobatannins:

The deposition of a red precipitate denoted the presence of phlobatannins when crude extract of all the six solvent of plant material was dissolved in 10 ml of aqueous extract and few drops of 1% HCl were added in the boiling tube.

Anthraquinones: All the six solvent extracts of leaves were boiled in 10% HCl for 5 mins separately and the filtrate was allowed to cool. An equal volume of CHCl_3 with few drops of 10% NH_3 was added to the 2ml filtrate. The formation of rose-pink colour implies the presence of anthraquinones.

Quantitative estimation of phytochemicals

Determination of Alkaloids

Alkaloids content was measured by method suggested by Harborne (Harborne, 1973) [28]. A suspension was prepared by dispersing 5 gm of the dried leaves in 10% acetic acid solution in ethanol and kept at 28°C for 4hrs which was further filtered through Whatman No. 42. Thereafter alkaloid was precipitated by concentrating the filtrate to one-quarter of its original volume and drops of conc. aqueous NH_4OH were added. Finally, the precipitate was washed with 1% ammonia solution and dried at 80°C in the oven. The content of alkaloid was calculated and expressed as mg/g of sample.

Determination of Flavonoids

The flavonoids content was also determined by Harborne (Harborne, 1973) method. 5 gm of leaves were boiled in 2M HCl for 30 min under reflux condition and filtered after cooling. An equal volume of ethyl acetate was then added drop wise to the filtrate. The weight of precipitated flavonoid was determined and recorded as mg/g.

Determination of Tannins

The finely powdered leaves of *Aegle marmelos* were kept in a beaker containing 20 ml of 50% methanol covered with parafilm and then heated at 80°C in a water bath for 1 hr with continuous stirring. The extract was quantitatively filtered using a double layered Whatman No.1 filter paper and rinsed with 50% methanol. 1 ml of sample extract was treated with 20 ml distilled water, 2.5 ml Folin-Denis reagent and 10 ml of 17% Na₂CO₃ for the development of a bluish-green colour and was allowed to stand for 20 mins. The absorbance was measured at 760 nm and the amount of tannin was calculated by comparing it with a standard curve prepared in the range of 0-10 ppm.

Determination of Saponins

100 ml Isobutyl alcohol was added to 1 gm of the finely powdered sample and stirred for 5 hrs. 20 ml of 40% saturated solution of Magnesium carbonate was added to the mixture and filtered. 2 ml of 5% FeCl₃ solution and 50ml volume of distilled water was added to 1ml of colourless solution and kept for 30 mins for colour (blood red) development. The absorbance of the samples as along with the standard were read at 380 nm and calculated in mg/g. Standard saponin solution was prepared in the reference range of 0-10 ppm.

Determination of total phenols

Five gms of the powdered leaves were boiled with 50 ml of ether for 15 mins and distributed in the ratio 1:2 (extract: distilled water). 2ml of ammonium hydroxide followed with 5ml of pentanol was added to it and incubated at the room temperature for 30mins. The absorbance was read at 505 nm wavelength.

For measuring alkaloids a suspension was prepared by dispersing 5 gm of the dried leaves in 10% acetic acid solution in ethanol and kept at 28°C for 4hrs which was further filtered through Whatman No. 42. Thereafter alkaloid was precipitated by concentrating the filtrate to one-quarter of its original volume and drops of conc. aqueous NH₄OH were added. Finally, the precipitate was washed with 1% ammonia solution and dried at 80°C in the oven. The content of alkaloid was calculated and expressed as mg/g of sample.

For determining flavonoids 5 gm of leaves were boiled in 2M HCl for 30 min under reflux and filtered after cooling. An equal volume of ethyl acetate was then added drop wise to the filtrate. The weight of precipitated flavonoid was determined and reported as mg/g.

For measuring tannin the finely powdered leaves of *Aegle marmelos* were kept in a beaker containing 20 ml of 50% methanol covered with parafilm and then heated at 80°C in a water bath for 1 hr with continuous stirring. The extract was quantitatively filtered using a double layered Whatman No.1 filter paper and rinsed with 50% methanol. 1 ml of sample extract was treated with 20 ml distilled water, 2.5 ml Folin-Denis reagent and 10 ml of 17% Na₂CO₃ for the development of a bluish-green colour and was allowed to stand for 20 mins. The absorbance was measured at 760 nm and the amount of tannin was calculated by comparing it with a standard curve prepared in the range of 0-10 ppm.

For determining saponin content 100 ml Isobutyl alcohol was added to 1 gm of the finely powdered sample and stirred for 5 hrs. 20 ml of 40% saturated solution of Magnesium carbonate was added to the mixture and filtered. 2 ml of 5% FeCl₃ solution and 50ml volume of distilled water was added to 1ml of colourless solution and kept for 30 mins for colour (blood red) development. The absorbance of the samples as along with the standard were read at 380 nm and calculated in mg/g. Standard saponin solution was prepared in the reference range of 0-10 ppm.

For determining total phenolic content five gms of the powdered leaves were boiled with 50 ml of ether for 15 mins and distributed in the ratio 1:2 (extract: distilled water). 2ml of ammonium hydroxide followed with 5ml of pentanol was added to it and incubated at the room temperature for 30mins. The absorbance was read at 505 nm wavelength.

Quantitative analysis of phytochemical constituents in six different solvent extracts

Six solvent extract of leaves of *Aegle marmelos* viz. acetone, petroleum ether, ethanol, methanol, benzene and distilled water were prepared by soaking 10gm of the powdered sample in 200 ml of each of the solvent separately for 12 hrs. The extracts were then filtered using filter paper. The extracts were then concentrated to ¼ of the original extracts i.e. 50 ml.

The amount of total phenolics in extracts was determined by the Folin-Ciocalteu method. Gallic acid was used as a standard by using different concentrations of (20-200µg) from which the total phenol content in the extract was expressed in terms of gallic acid equivalent (mg GAE /gm) extract. Different aliquots of 0.1 to 1.0 ml of plant extract were also prepared in methanol and 0.5 ml of each sample were introduced into test tubes and mixed with 2.5 ml of a 10-fold dilute Folin- Ciocalteu reagent and 2 ml of 7.5% sodium carbonate. The mixture was allowed to stand for 30 mins at room temperature. Phenols react with the phosphomolybdic acid in Folin- Ciocalteu reagent in alkaline medium and produce blue coloured complex (Molybdenum blue). The absorbance of the resulting solutions was measured at 760 nm against reagent blank. A standard calibration

curve was prepared by plotting absorbance against concentration and it was found to be linear over this concentration range. The concentration of total phenol in the test sample was determined from the calibration graph. The assay was carried out in triplicate and the mean values with \pm SD are presented.

The aluminium chloride colorimetric method was used for flavonoids determination. Each solvent extract (0.5 ml of 1:10 gm ml⁻¹) was separately mixed with 1.5 ml of methanol, 0.1 ml of 10% aluminium chloride, 0.1 ml of 1 M potassium acetate and 2.8 ml of distilled water. It was kept at room temperature for 30 min; the absorbance of the reaction mixture was measured at 418 nm. The percentage of total flavonoids were calculated from the calibration curve of Quercetin (200-1000 μ g) plotted by using the same procedure and total flavonoids was expressed as Quercetin equivalents (QE) in mg per gm sample.

The results of qualitative phytochemical analysis of *Aegle marmelos* extracts have been presented in Table- 1, 2 and 3; Figure- 1 and 2.

Table- 1: Phytochemicals of *Aegle marmelos* leaf in six different solvent extracts

Solvent extracts of leaves	Ta	Phl	Sap	Terp	Dter	Emd	Fla	Car	Anth	Crt	Res	Alk	Anc	Cou	Str	Pst	Phe	Fa	Prt	Aa
Acetone	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Petroleum ether	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Ethanol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+
Methanol	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+
Benzene	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Distilled water	+	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-	+	+	+	+

Ta= Tannin; Phl= Phlobatannin; Sap= Saponin; Ter= Terpinoid; Dtr = Diterpinoid ; Emd= Emodin ; Fla= Flavonoid; Car= Cardiac glycoside; Anth= Anthraquinones; Crt= Carotenoids; Res= Reducing sugar; Alk= Alkaloid; Anc= Anthocyanin; Cou= Coumarin; Str= Steroids; Pstr= Phytosterol; Phe= Phenol; FA= Fatty acids; Prt= Protein; Aa= Aminoacids

Table- 2: Quantitative estimation of Phytochemicals in leaves of *Aegle marmelos*

Phytochemicals	Amount in mg/gm
Alkaloids	15.75 \pm 0.25
Flavonoids	42.75 \pm 0.45
Phenols	23.85 \pm 0.05
Saponins	14.65 \pm 0.35
Tannins	12.35 \pm 0.25

Mean \pm SD of five measurements

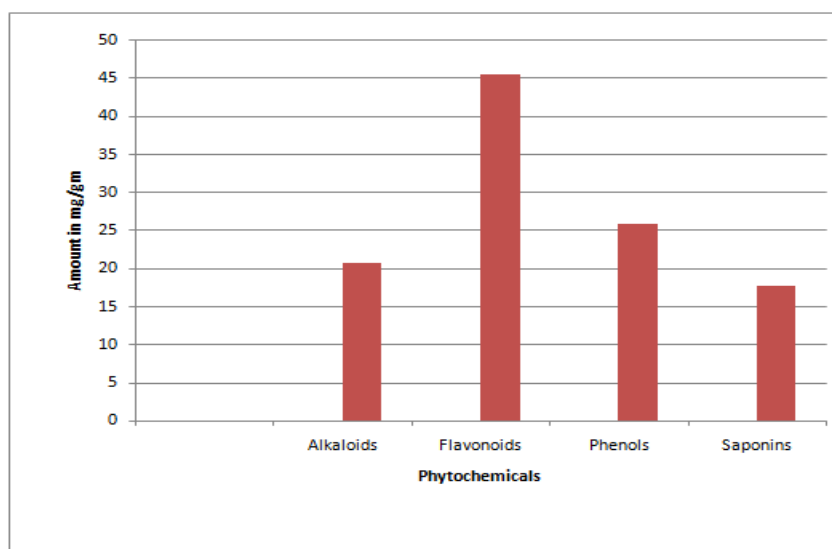


Fig- 1: Phytochemicals in leaves of *Aegle marmelos*

Table- 3: Comparative analysis of total Alkaloids, Flavonoids, Phenol, Saponins and Tannins in six different solvent extracts of leaves of *Aegle marmelos* (amount in mg/gm)

Solvent Extracts	Total Alkaloids	Total Flavonoids	Total Phenol	Total Saponins	Total Tannin
Acetone	11.35±0.35	12.45±0.42	8.75±0.75	9.45±0.35	10.55±0.17
Petroleum ether	12.45±0.45	13.65±0.18	10.67±0.65	10.35±0.21	12.53±0.16
Ethanol	13.75±0.31	40.45±0.12	14.25±0.24	15.35±0.25	11.35±0.27
Methanol	16.65±0.27	41.65±0.15	14.45±0.23	12.37±0.31	9.75±0.42
Benzene	10.75±0.16	14.45±0.70	10.85±0.09	16.35±0.81	11.37±0.26
Distilled water	34.75±0.07	26.85±0.24	11.38±0.40	14.35±0.80	10.43±0.08

Mean ± SD of five measurements

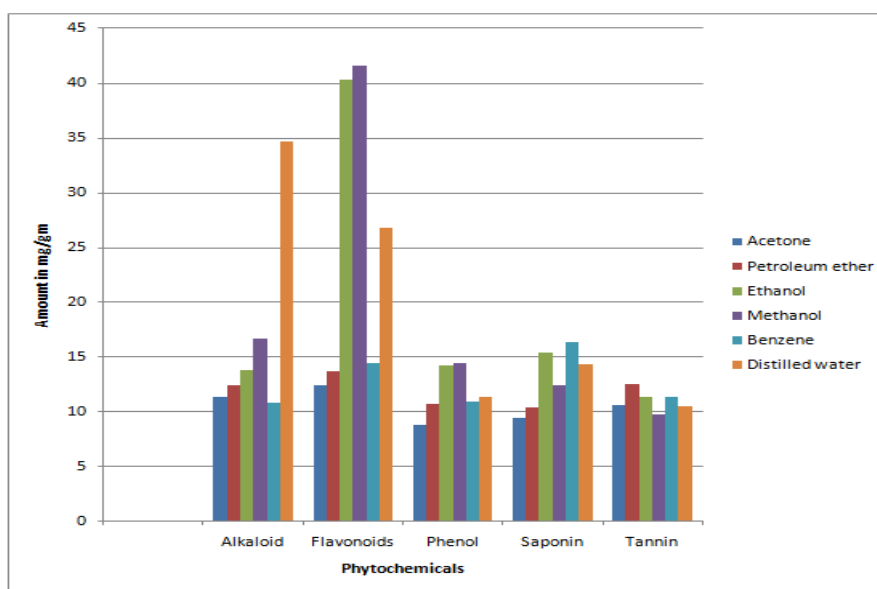


Fig- 2: Comparative analysis of phytochemicals in six different solvent extracts of leaves *Aegle marmelos*

III. Results and Discussion

The results of qualitative phytochemical analysis of *Aegle marmelos* is shown in Table- 1. The present study was carried out on six solvent extracts of *Aegle marmelos* to investigate the presence of medicinally important phytochemicals in their leaves. All the six extracts revealed the presence of various phytochemicals such as tannins, phlobatannins saponins, , terpinoids, diterpinoids, emodins, flavonoids, cardiac glycosides, anthraquinones, carotenoids, reducing sugars, alkaloids, anthocyanin, coumarins, steroids, phytosterols, phenol, fatty acids, proteins and amino acids. Of these 20 phytochemicals tannin, saponin, terpenoid, diterpinoid, emodin, flavonoid, cardiac glycoside, anthraquinone, carotenoid, reducing sugar, alkaloids, anthocyanin, coumarin, phenol, fatty acid, protein and amino acids were detected in all the six solvent extracts. Phlobatannin, steroid and phytosterol were not detected in ethanol, methanol and distilled water extracts. Emodin was detected in all extracts except petroleum ether and benzene. The presence of different phytochemicals and the antimicrobial activity of ethanolic, petroleum ether, chloroform and methanolic extract of a of *Aegle marmelos* has been previously reported [18] (Venkatesan *et al.*, 2009). These phytochemicals are active bioreactive chemicals and effectively used to cure Tridosha,- Joundice, Asthma and Inflammation. A more or less similar phytochemicals were screened by [19] Sonu and Neeta (2016) in the chloroform leaf extract of *Aegle marmelos*.

From the results (Table- 2; Fig- 1) it is evident that the leaves of *Aegle marmelos* contained a significant amount of alkaloid, flavonoids, phenolic, saponins and tannin content. The amount of flavonoids was maximum (42.75mg/gm) followed by phenols (23.85mg/gm), alkaloids (15.75mg/gm), saponins (14.65mg/gm) and tannins (12.35mg/gm) (Table- 2; Fig- 1).

The comparative analysis of phytochemicals viz. total alkaloids, flavonoids, phenols, saponins and tannins in six different solvent extracts from leaves of *Aegle marmelos* has been presented in Table- 3 and Fig- 2. From the results it is evident that the concentration of total alkaloids was maximum in distilled water extract (34.75mg/gm), followed by methanol extract (16.65mg/gm), ethanol extract (13.75mg/gm), petroleum ether extract (12.45mg/gm), acetone extract (11.35mg/gm) and benzene extract (10.75mg/gm). The concentration of total flavonoids was maximum in ethanol and methanol extracts (40.45mg/gm and 41.65mg/gm respectively), followed by distilled water extract (26.58mg/gm), benzene extract (14.45mg/gm), petroleum ether extract (13.65mg/gm) and acetone extract (12.45mg/gm). The amount of total phenol was maximum in ethanol and methanol extracts (14.25- 14.45mg/gm), followed by distilled water extract (11.38mg/gm), benzene and

petroleum ether extracts (10.67- 10.85mg/gm) and acetone extract (8.75mg/gm), least amount. Saponin concentration was maximum in ethanol (15.35mg/gm), benzene extract (16.35mg/gm) and distilled water extract (14.35mg/gm). Acetone and petroleum ether extracts contained relatively least amount of saponins (9.45-10.35mg/gm). The total tannin concentration was maximum in petroleum ether extract (12.53mg/gm), followed by ethanol and benzene extracts (11.35- 11.37mg/gm), acetone and distilled water extracts (10.43- 10.55mg/gm) and methanol extract (9.75mg/gm) (Table- 3; Fig- 2).

The leaf extracts of *Aegle marmelos* (L.) Correa. and showed the presence of terpenoids, steroids and phytosterols, tannins, alkaloids, glycosides, saponins, reducing sugars, phenols and flavonoids. The extraction of various phytochemicals was seen to be more effectively done in polar solvents (ethanol, methanol and distilled water) than the non polar (Acetone, petroleum ether, benzene) solvents. Especially, ethanolic, methanolic and distilled water leaf extracts showed presence of most of the tested phytochemicals. Hence, it can be reported that alcoholic extract was the best one for extracting the active principle than others. This may possibly be one of the reasons for highest antibacterial activity shown by the ethanolic leaf extracts of these plants. Flavonoids are water-soluble polyphenolic compounds which are extremely common and widespread in the plant kingdom as their glycosides. The flavonoids are known to act through scavenging or chelating process.

Ulahannan, *et al.*, (2008) [20], in their study have indicated presence of phenol and alkaloids in acetone and methanol extracts. Flavonoids were detected by them only in methanol extract of the plant and terpenes not detected in any of the tested extracts. However, in the present study the presence of phenols, saponins, tannins, alkaloids and flavonoids was detected in all the six solvent extracts of the leaves of *Aegle marmelos* (L.) Correa. Phytochemical analysis of the petroleum ether, benzene, chloroform, ethanol and water extracts of *Aegle marmelos*, was also done by Rajeshwari Sivaraj, *et al.* (2011) [21] and the absence of terpenoids in all the extracts was reported by them. The present findings gain support from the work of Samidha *et al.* (2017) [22] who have identified a more or less similar phytochemicals in polar and non polar solvent extracts of leaves of *Aegle marmelos*.

The results of the quantitative phytochemicals analysis showed that the leaf extracts of *Aegle marmelos* (L.) Correa. have appreciable amount of terpenoids, bitter substances, phytosterols, tannins, alkaloids, glycosides and saponins. Quaternary Alkaloids & N-oxides and glycosides were found to be the most abundant one followed by saponins and bitter substances. While, fats and waxes, terpenoids and phytosterols were comparatively low in concentration. The phytochemicals are known to have antimicrobial activity [23, 24, and 25] (Ebana *et al.* 1995; Hassan *et al.* 2004; Usman and Osuji, 2007). The presence of phenolic compounds indicates that the plants are antimicrobial agent [26, 27] (Okwu and Okwu, 2004; Rasool Rafia, 2010). It can be suggested that the presence of phenols, alkaloids, flavonoids, saponins in six solvent extracts viz, acetone, petroleum ether, ethanol, methanol, benzene and distilled water of leaves of *Aegle marmelos* (L.) Correa. may be considered as one of the reason for a good antibacterial property shown by the leaf extract.

IV. Conclusions

The results obtained in the present investigation are encouraging and can be used as reference data for the standardization of leaves of *Aegle marmelos* (L.) Correa. and the formulations containing these plant leaves as a main ingredient. Though this plant is very common plants having less possibility of adulteration, but to get highest efficacy of an herbal drug or its finished product, cent percent genuine plant material should be the source. All these above said characters reflect the diagnostic features of the leaves of *Aegle marmelos* (L.) Correa. and hence can be used to check adulteration. The evaluation of the various proximate parameters for the leaves of *Aegle marmelos* (L.) Correa. has given a clear idea about the specific characteristics of these crude drugs under examination, in their powder form. The preliminary screening tests may be useful in the detection of the bioactive principles and subsequently may lead to the drug discovery and development. Such screening experiments form a primary platform for further phytochemicals and pharmacological studies that may open the possibility of finding new clinically effective compounds. Thus, the present study has authenticated the usefulness of the *Aegle marmelos* (L.) Correa. Plants for medicinal purposes. These species could also be seen as potential sources of useful drugs due to their rich contents of phytochemicals. The data obtained in the present study is expected to serve as valuable tool for identification, authentication and detection of adulterants, standardization and quality control of the drugs. It can hence be concluded that the results of the present study have given qualitative information about the purity standards of the leaf powders of *Aegle marmelos* (L.) Correa.

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