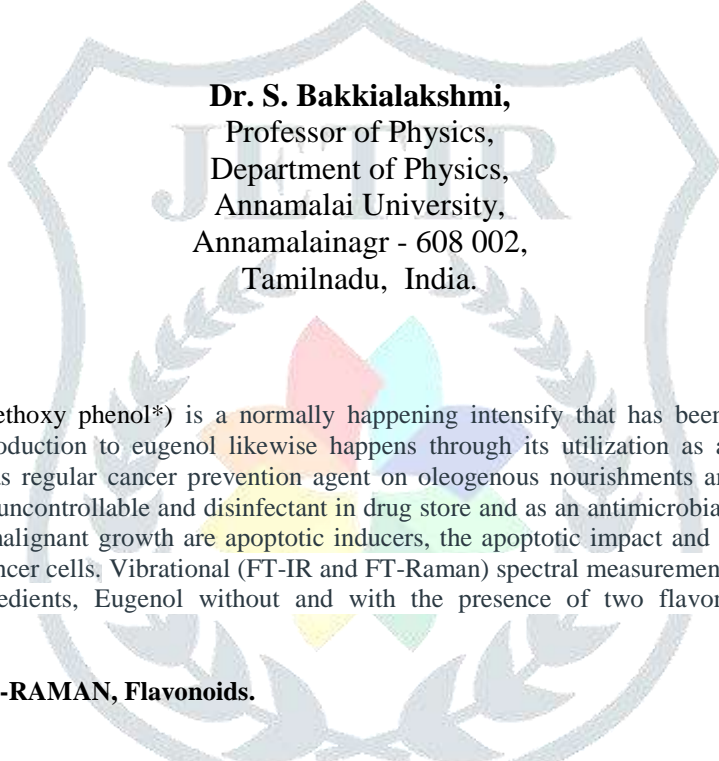


CHARACTERISATION OF EUGENOL WITH TWO FLAVONOIDS BY VIBRATIONAL SPECTROSCOPY MEASUREMENTS

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Abstract: Eugenol (4-allyl-2-methoxy phenol*) is a normally happening intensify that has been utilized widely as a seasoning operator and scent. Human introduction to eugenol likewise happens through its utilization as a pain relieving and from clove cigarettes. Eugenol goes about as regular cancer prevention agent on oleogenous nourishments and furthermore goes about as an enemy of carminative, hostile to uncontrollable and disinfectant in drug store and as an antimicrobial specialist. Since the majority of the medications utilized in the malignant growth are apoptotic inducers, the apoptotic impact and anticancer instrument of eugenol were researched against colon cancer cells. Vibrational (FT-IR and FT-Raman) spectral measurements were performed for an analysis of one of the Tulasi leaf ingredients, Eugenol without and with the presence of two flavonoids Quercetin and Hesperidin nanoparticles.

Keywords: Eugenol, FT-IR, FT-RAMAN, Flavonoids.

1.Introduction

Essential oils and extracts of various species of healthy to be eaten and medicinal plant life, herbs, and spices constitute very strong biologically active dealers. They have a complex composition, containing from a few to several hundred components, specifically hydrocarbons and oxygenated compounds. Both, hydrocarbons and oxygenated compounds are responsible for the characteristic odours and flavours, which is probably fashioned through fragrant vegetation as secondary metabolites¹. Clove oil (CO) is an vital oil from the dried flower buds, leaves and stem of the tree *Syzygium aromaticum* (Eastern Hemisphere) or *Eugenia caryophyllata* and *Eugenia aromaticum* (Western hemisphere)². It has been used for loads of years as anesthetic for toothaches, headaches and joint pain³⁻⁴. Clove has received attention as an excellent fish anaesthetic⁵⁻⁸ and it's been used as a aromatic and flavouring agent in a selection of food and beauty merchandise⁹⁻¹¹. However, irritation towards the mucosa and pores and skin, smelly taste, volatility, moderate sensitivity and terrible water solubility make it flawed to apply as such. Eugenol (EG) (four-allyl-2-methoxyphenol) is the most constituent of the essential CO, being 90% - ninety five% of the general oil amount¹²⁻¹³. It has a robust phenolic smell and sharp acrid taste¹⁴⁻¹⁵ and its chemical shape is represented in Figure 1.

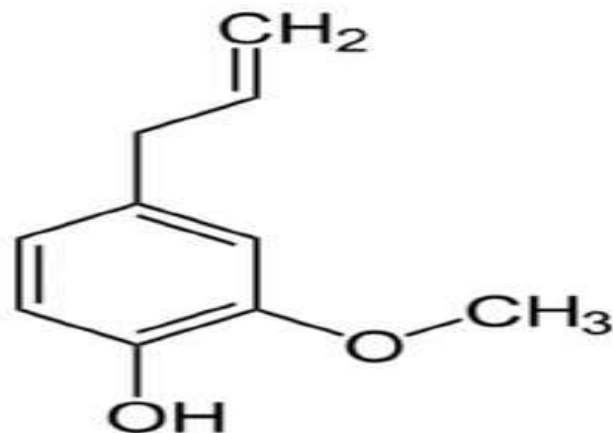


Fig 1 Chemical structure of Eugenol (EG).

Flavonoids are polyphenolic compounds which may be ubiquitous in nature and are categorized, consistent with chemical shape, into flavonols, flavones, flavanones, isoflavones, catechins, anthocyanidins and chalcones. More than 4000 flavonoids have been recognized, a considerable lot of which happen in organic products, vegetables and refreshments, for example, tea, espresso, brew, wine and natural product drinks. Flavonoids have stirred significant intrigue as of late in view of their likely valuable impacts on human wellbeing - they have been accounted for to have antiallergic, antiviral, calming, antiplatelet, antitumor and cell antioxidant activities¹⁶⁻¹⁸. The antioxidant activity of flavonoids is related to their ability to chelate metallic ions and to scavenge singlet oxygen, superoxide anions, peroxy radicals, hydroxyl radicals, and peroxy nitrite¹⁹⁻²⁰. Another thrilling detail of flavonoids is their use as an innovative pigment. In truth, flavonoids also can be located in cultural historical past works of artwork²¹⁻²⁵. Quercetin and Hesperidin are the two flavonoids taken for this work.

2. Experimental

Eugenol and the flavonoids, (quercetin and hesperidin) were bought in its most purity from Sigma-Aldrich and used without similarly purification. Stock solutions of the Eugenol had been prepared at a consciousness of 10^{-3} M and saved in dark with a view to shield them from light. For the same cause, to limit a possible photodegradation of flavonoid molecules, the examined solutions had been include from light moderate during the out of measurement times. All solutions had been freshly organized with triple distilled water before experiments and used straight away.

The vibrational infrared spectra of samples of Eugenol and flavonoids were recorded 4000 at room temperature, with an AGILENT CARY 630 FTIR Spectrometer. The normal Raman spectra were obtained directly from FT-Raman measurements. The FT-Raman spectra were received by using a BRUKER RFS - 27 STAND alone FT-Raman Spectrometer.

3. Results and Discussion

Eugenol is the most plentiful ingredient inside clove essential oil²⁷⁻³⁴. It changed into mentioned that the oil extricated from clove buds incorporates 36-90% of eugenol, even as the oil extricated from leafs and stems include 75-90% and 85-5% of eugenol, respectively²⁸⁻³¹. The second fundamental factor clove essential oil is probably each eugenyl acetate (for oils extricated from clove buds and stems) or β -caryophyllene (for oils remove from clove leaves)²⁸⁻³¹. Eugenyl acetate is not always found in the chromatograms in the essential oil solutions, likely as long as it was immediate in very low concentration in the analyzed samples. At this the point of these chromatographic estimations was not the measurement of the segments, however the distinguishing proof of the principle segments which are the dependable to shape the Raman spectra of the fundamental oils. Both eugenol and β -caryophyllene are normal segments in the ethereal oils removed from Clove departs, however benzyl liquor ought not be available. In prior reports on the creation of Clove basic oil, benzyl liquor was missing or just follows were found²⁶⁻³². Quercetin and Hesperidin nanoparticles were prepared using ionic gelation method.

FTIR combination of eugenol and flavonoids have been in comparison with the spectra of separate components. The FTIR Spectrum of eugenol is shown in Fig 2a. Eugenol confirmed its signature peaks in 720-1250 cm^{-1} location similar to $\text{C}=\text{C}$ region. In addition, the peaks at 1649 cm^{-1} and 1461 cm^{-1} were also found from eugenol, which could be because of $\text{C}-\text{O}$ stretching. In the case of (Eugenol + Hesperidin) mixture, the sharp peak at 879 cm^{-1} shows the $\text{C}-\text{C}$ stretching. The sharp peaks at 1044 cm^{-1} and 1085 cm^{-1} were also found which denotes $\text{C}-\text{H}$ bending vibrations.

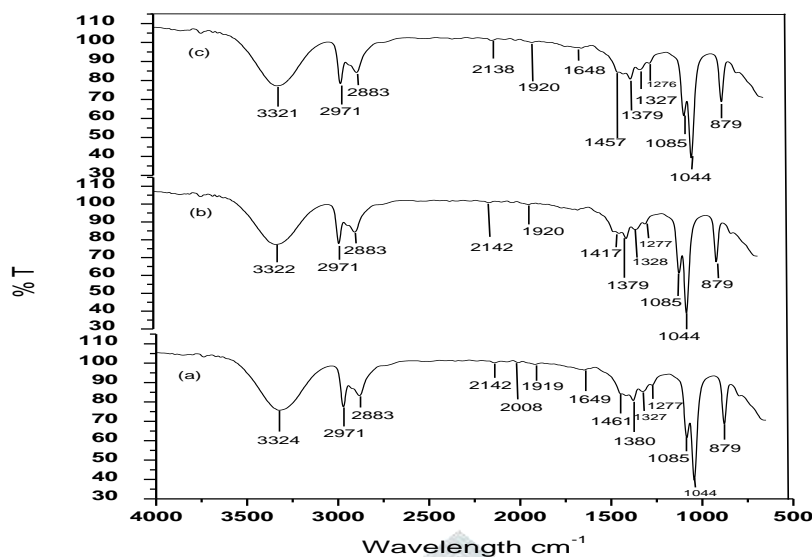


Fig 2: FTIR Spectra of (a) Eugenol (b) Eugenol + (Quercetin nanoparticle) mixture and (c) Eugenol + (Hesperidin nanoparticle) mixture

Fig 2b, shows the FTIR Spectra of Eugenol + quercetin nanoparticle mixture. Fig.2c shows the (Eugenol + Hesperidin nanoparticle) mixture of FTIR Spectrum. The peaks at 879 cm^{-1} and 1085 cm^{-1} denotes C-C stretching. The sharp peaks at 1379 cm^{-1} and 1328 cm^{-1} also stated within the FTIR Region of (Eugenol + quercetin nanoparticle) mixture these peaks display the CH_3 Symmetric bending. The results of the IR Spectroscopy showed the similarity of eugenol with the flavonoids, hesperidin nanoparticle and quercetin nanoparticle.

Table 1: FTIR peak assignments of Eugenol, Quercetin nanoparticle and Hesperidin nanoparticle complex

EG (cm^{-1}) FTIR	EG+ QUR nanoparticle (cm^{-1}) FTIR	EG+HES nanoparticle (cm^{-1}) FTIR	Peak Assingments
3324.262	3322.789	3321.775	OH bond
2291.777	2971.687	2971.964	C- H Stretching
2883.602	2883.401	2883.311	CH_3 Symmetric Strecthing
1649.476		1648.653	C- O asymmetric stretching
1461.116		1457.389	C - O stretching
1380.016	1379.908	1379.527	CH_3 asymmetric Strecthing
1327.625	1328.197	1327.846	CH_3 Symmetric Strecthing
1277.739	1277.575	1276.481	OH in plane bending
1085.386	1085.386	1085.725	C- C stretching
1044.467	1044.507		$\text{CH}_3 - \text{O} - \text{C}$ bending
879.501	879.574	879.538	C - C ring stretching
656.956		655.593	Hydrogen bonded out of plane OH bend

Experiments band positions for eugenol and mixtures of eugenol with nanoparticles of flavonoids are listed in Table 1. The Raman Spectra of Eugenol is shown in 3a. The mixtures of (Eugenol + Quercetin nanoparticle) and (Eugenol + Hesperidin nanoparticle) Raman Spectra are shown in Fig 3b and 3c respectively.

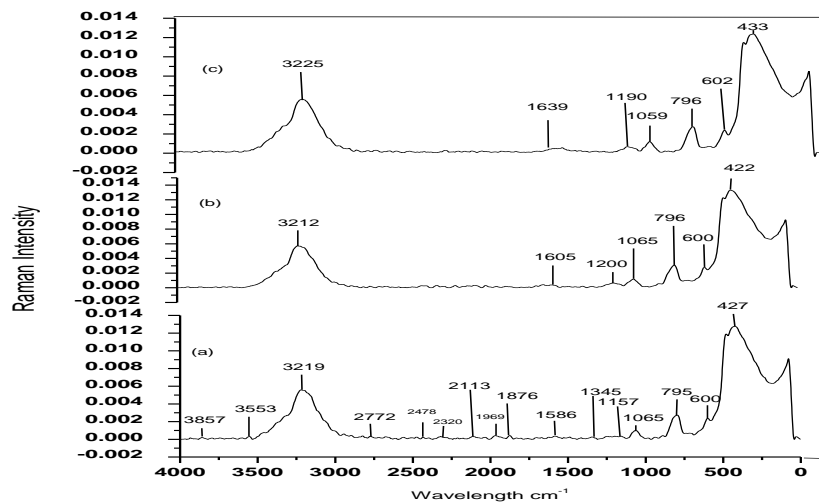


Fig 3: FT-Raman Spectra of (a) Eugenol (b) Eugenol +(Quercetin nanoparticle) mixture and (c) Eugenol+(Hesperidin nanoparticle) mixture

For mono-, di - and trisubstituted benzenes, the frequency of two groups in the stages 1620 - 1585 cm^{-1} and 1590 - 1565 cm^{-1} are expected, each group being relegated to the ring quadrant extending modes.

Table 2: RAMAN peak assignments of Eugenol, Hesperidin nanoparticle and Quercetin nanoparticle complex

EG (cm^{-1}) RAMAN	EG+ QUR nanoparticle (cm^{-1}) RAMAN	EG+ HES nanoparticle (cm^{-1}) RAMAN	Peak Assignments
3857			O-H Stretching
3553			O-H Stretching
3219	3212	3225	O-H Stretching
2772			O-H Stretching
2478			Hydroxyl Stretching
2320			Hydroxyl Stretching
2113			Hydroxyl Stretching
1969			C=O Stretching
1876			C=O Stretching
1586	1605	1639	C-H Stretching
1345	1200		C-H bending
1157		1190	C-O Stretching
1065	1065	1059	C-C stretching

An exemption are identically substituted 1,3,5 - trisubstituted benzenes; they have rings with a three - overlap hub of symmetry, along these lines the two quadrant extending modes are doubly degenerate and appear at the equivalent wavenumber.

The band discovered at 1345 cm^{-1} in the normal Raman Spectrum of Eugenol was relegated to the O - CH_3 wagging mode. The normal range for this mode is 1390 - 1340 cm^{-1} . Table 2 display the experiment band positions for eugenol and mixtures nanoparticle of eugenol with flavonoids.

4. Conclusion

The use of the spectroscopic methods FT-IR and FT-Raman are recommended for quantitative analysis of Eugenol with the flavonoids. The consequences clearly demonstrate that the interaction of Eugenol with the nanoparticle of flavonoids, quercetin and hesperidin can result in the emergence of complexes.

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