International Journal of Engineering Science Invention (IJESI)
ISSN (Online): 2319 – 6734, ISSN (Print): 2319 – 6726
www.ijesi.org ||Volume 7 Issue 3 Ver. V|| March 2018 || PP30-37

Fluorimetric Study of Eugenol with Cyclodextrins

Rajendran.P[a], Devika P Nair[a],Pushpa.S[b],&Bakkialakshmi.S*
[a]*Departmentof Electronics and Communications, Amrita School Of Engineering, Amrita Vishwavidyapeetham, Coimbatore, Tamilnadu, India--641112.*
[*,b]*Department Of Physics, Annamalai University, Annamalainagar, Tamilnadu, India-608 002*
**Corresponding author:Bakkialakshmi.S*
**Corresponding author E-mail: bakkialakshmis@rocketmail.com*

Abstract:*Eugenol (4-Allyl-2-Methoxy Phenol) is a naturally occurringcompound that has been used extensivelyas aflavoring agent and fragrance. Human exposure to eugenol also occurs throughits use as an analgesic and from clove cigarettes. Steady State Time –Resolved Fluorescence UV / Absorption and FTIR spectroscopic techniques were used to study the complexation ofEugenol With β-CyclodextrinandHβ – Cyclodextrin. The Stiochiometry, binding constants and thermodynamic parameter upon complexation were obtained from the variation of fluorescence intensity and average lifetime withβcds.*

Keywords:*Eugenol, β - Cyclodextrin, Hβ -Cyclodextrin, Fluorescence,FTIR.*

Date of Submission: 15-03-2018 Date of acceptance 29-03-2018

I. Introduction:

Essential oils and extracts of various species of edible and medicinal plants, herbs, and spices constitute very potent biologically active agents. They have a complex composition, containing from a few to several hundred constituents, mainly hydrocarbons and oxygenated compounds. Both, hydrocarbons and oxygenated compounds are responsible for the characteristicodors and flavors, whichare formed by aromatic plants as secondary metabolites [1]. Clove Oil (CO) is an essential oil from the dried flower buds, leaves and stem of the tree syzygiumaromaticum (Eastern Hemisphere) or eugeniacaryophyllata and eugeniaaromaticum (Western Hemisphere) [2]. It has been used for centuries as anesthetic for toothaches, headaches and joint pain [3,4]. clove has received attention as an ideal fish anaesthesic [5-8] and it has been used as a fragrant and flavoring agent in a variety of food and cosmetic products [9-11]. However, irritation towards the mucosa and skin, pungent taste, volatility, light sensitivity and poor water solubility make it unsuitable to use as such.

Eugenol (EG) (4-Allyl-2-Methoxyphenol) is the principal constituent of the essential co, being 90% - 95% of the total oil amount [12]. It has a strong phenolic smell and sharp acrid taste [13].It is well established that cyclodextrins (CDs) form complexes with several molecules eliminating their unwanted effects. CDs are a group of naturally occurring cyclic oligosaccharides derived from starch with six, seven and eight glucose of residues linked by α (1-4) glycosidic bonds in a cylinder-shaped structure, and denominated α, β and γ - Cyclodextrins, respectively. The centralcavity of these molecules is hydrophobic, while the rims of the surrounding walls are hydrophilic. This hydrophobic cavity forms inclusion complexes with a wide range of organic and inorganic guest molecules, altering their physicochemical behaviour and reducing their undesirable effects.

II. Materials And Methods:

*β*cd, Hβcd andEugenol were purchased fromSigma –Aldrich Company, Bangalore.All the chemicals were used without further purification. UV/ Vis Absorption spectra were taken usingSHIMADZU 1800 PC UV-VISIBLE SPECTROPHOTOMETER,Steady State Fluorescence measurementswere made bySHIMADZU RF 5301 PCSPECTROFLUOROPHOTOMETER, Fluorescence Decay measurementswere recorded using HARIBA- JOBIN YVON [SPEX-SF B-III]SPECTROFLUORIMETERandFTIR Spectra were recorded using AGILENT CARY 630 FTIR SPECTROMETER.

III. Resultsand Discussion:

3.1: Uv/Absorption Study Of Eugenol

An attempt has been made to estimate the formation constant of 1:1 Eugenol and*β*-Cyclodextrin (*β*cd) andHβ-Cyclodextrin (Hβcd) complexes studying their absorption and fluorescence properties. The formation constant of the complexes was determined by studying their absorption and fluorescence spectral changes. To

prove the above, other spectroscopic techniques, (1) Time-Resolved Fluorescence and (2) Fourier Transform Infrared (FTIR) analyses were utilized.

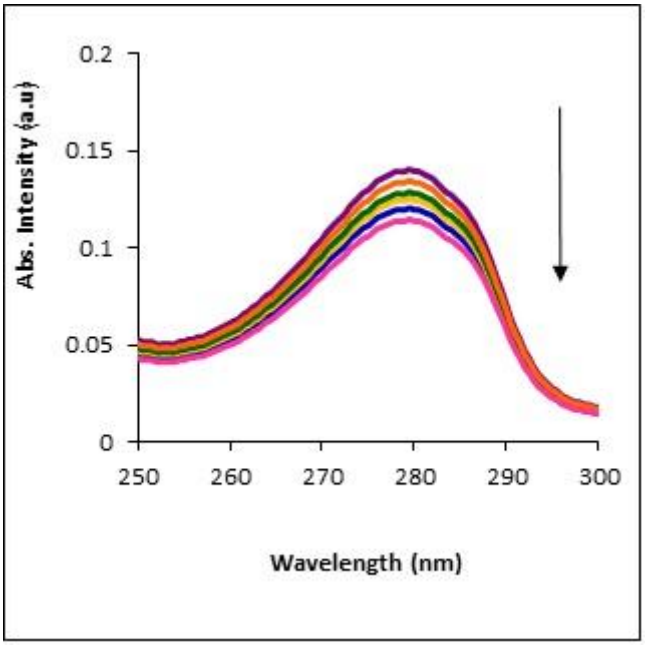

Figure 1: Absorption spectra of Eugenol indifferent concentrations ofβ-Cyclodextrin (mol dm^{-5}) (1) 0, (2)0.2 (3)0.4, (4)0.6, (5)0.8, (6)1.0

The decrease in absorbance with the addition of *β*cd and Hβcd can be attributed to interaction which indicates the formation of the inclusion complex ofEU-*β*cd as well asEU-Hβcd. Fig.3 shows the plot of absorbance versus concentrations of the*β*cds.

The Plot of $\left(\frac{1}{A_0\text{-}A}\right)$ versus $\left(\frac{1}{\beta CDs}\right)$ will result in a straight line as shown in Fig.4. From the slope values of this plot, K has been evaluated and tabulated. This is the ground stateformation constant K_g. Calculated values are shown in Table 1. Free energy change was calculated from the formation constant K_g.

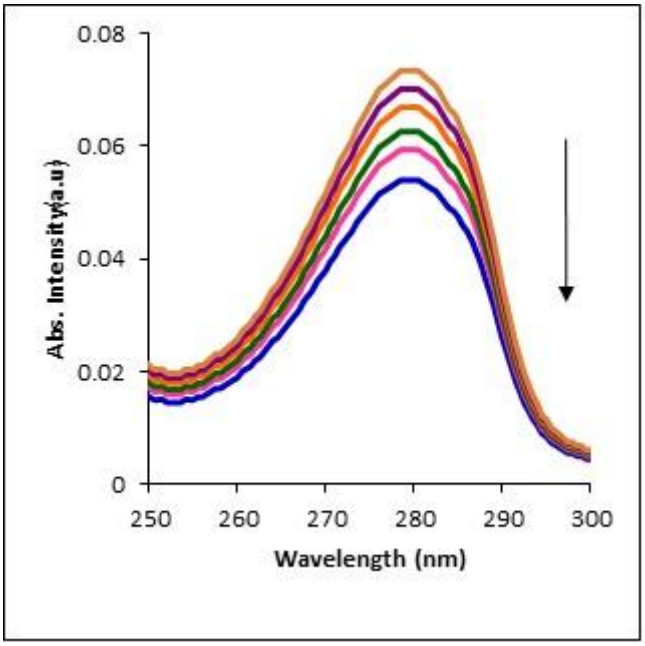

Figure 2: Absorptionspectra of Eugenol in different concentrations ofHβ-Cyclodextrin (mol dm^{-5}) (1) 0, (2) 0.2 (3) 0.4, (4) 0.6, (5) 0.8, (6) 1.0

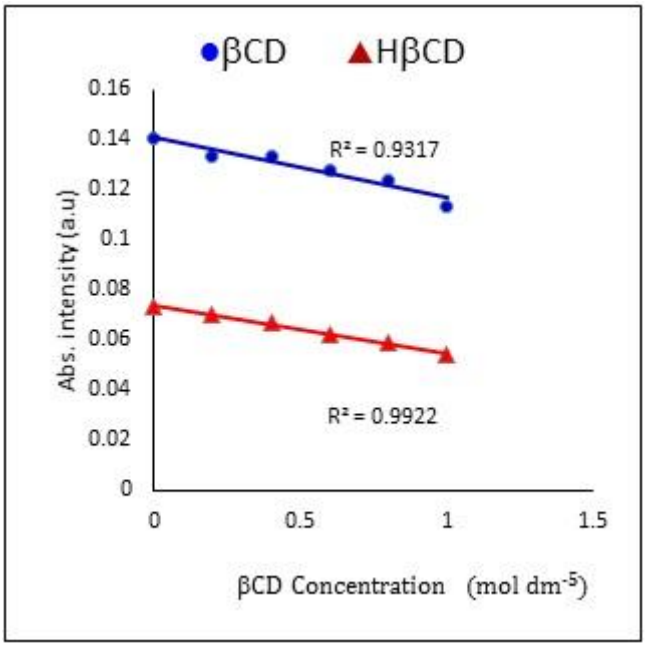

Figure 3: Absorbance of Eugenol versusβcd concentrations

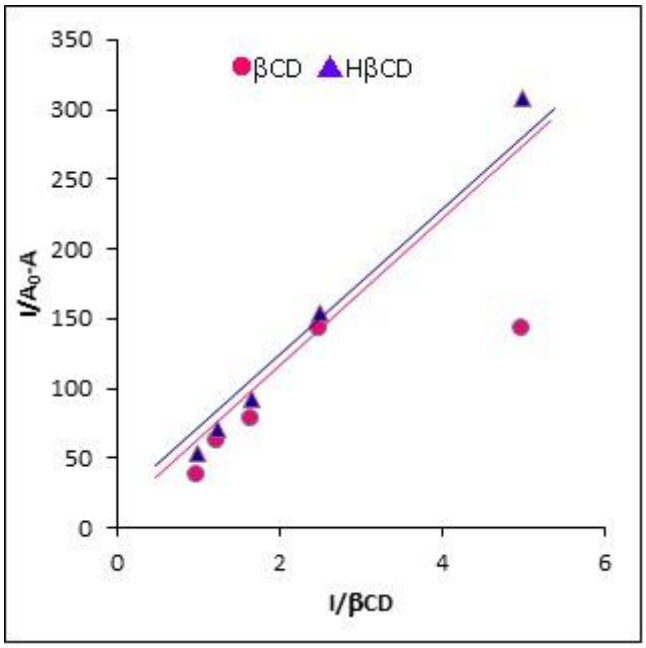

Figure 4: Plot of$\left[\frac{1}{A_0-A}\right]$versus$\left[\frac{1}{\beta cd}\right]$forEugenol

3.2: Fluorescence Spectral Study Of Eugenol

To investigate the interaction between eugenol and the βcds, fluorescence spectra were studied. It has been found in this study when free eugenol was excited at 280 nm in the absence of cyclodextrins, itshowed a broad fluorescence peak at around 515 nm in aqueous solution. Figs.5 and 6 depict the emission spectra of eugenol in various concentrations of βcd and Hβcd respectively. With the addition of βcd and Hβcd solutions to Eugenol, Emission intensities increase in thesamewavelength along with increase in the βcd and Hβcd concentrations, and its fluorescence intensity also increased. Fig.7 shows the plot of fluorescence intensity versus the βcd and Hβcd concentrations.

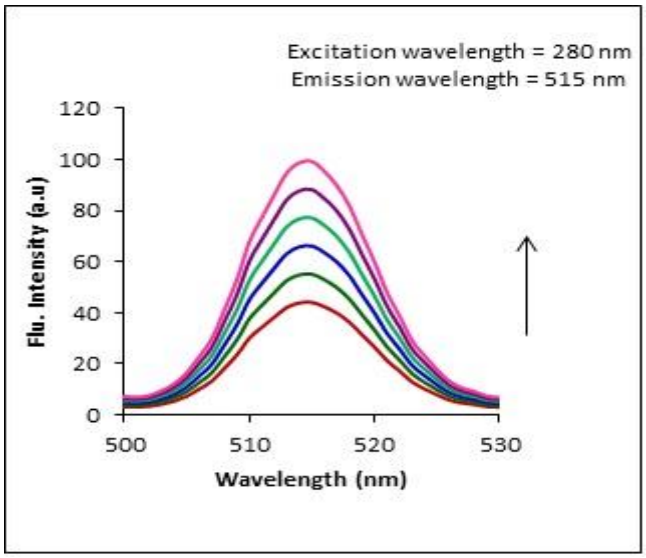

Figure 5: Fluorescence spectra of Eugenol in different concentrations ofβ-Cyclodextrin (mol dm^{-5}) (1)0, (2)0.2 (3)0.4, (4)0.6, (5)0.8, (6)1.0

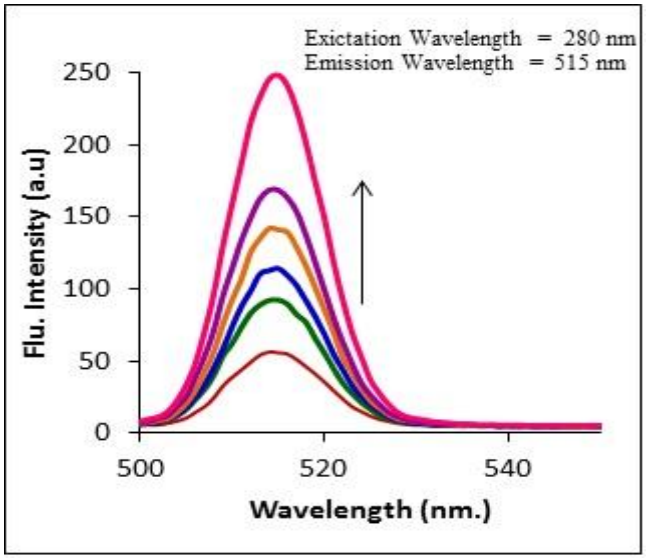

Figure 6: Fluorescence spectra of Eugenol in different concentrations ofHβ-Cyclodextrin (mol dm^{-5}) (1)0, (2)0.2 (3)0.4, (4)0.6, (5)0.8, (6)1.0

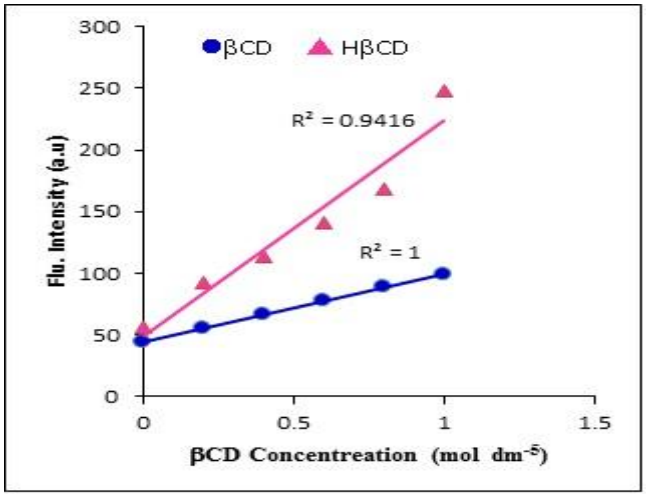

Figure 7: Plot of fluorescence intensity of Eugenol Versus βcdsconcentrations

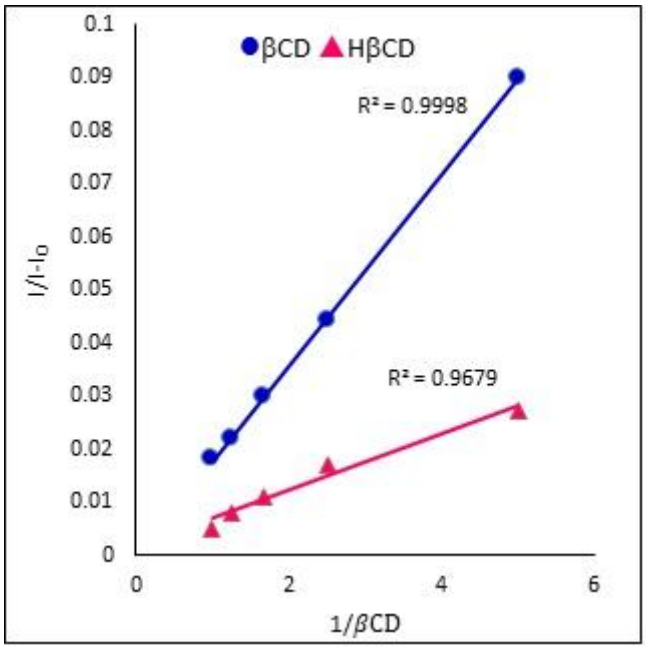

Figure8: Plot of$\left[\frac{1}{I-I_0}\right]$versus$\left[\frac{1}{\beta cd}\right]$forEugenol

The stoichiometry and the formation constants have been determined from the Benesi-Hildebrand plots shown inFig. 8 and the data are presented in Table 1

3.3: Time-Resolved Fluorescence Study Of Eugenol

Fluorescence lifetime measurement is a very useful technique for understanding the type of interaction between the donor and the acceptor systems. Figs. 9 And 10 depict fluorescence decay of eugenol with and without βcd and Hβcd respectively. The calculated average lifetime values have been given in Tables 2 and 3.

Table I: Formation constantK (M^{-1}) and free energy ΔG ($kJmol^{-1}$) of Eugenolwith βcd and Hβcd

βcd				**Hβcd**			
Kg (M^{-1})	**Ke (M^{-1})×10^{-2}**	**ΔG_g ($Kjmol^{-1}$)**	**ΔG_e ($Kjmol^{-1}$)**	**Kg (M^{-1})**	**Ke (M^{-1})×10^{-3}**	**ΔG_g ($Kjmol^{-1}$)×10^4**	**ΔG_e ($Kjmol^{-1}$)×10^4**
15.9	1.84	6990.5	100096.46	74.36	7.14	1.0888	1.2488

Table II: Fluorescence lifetime and amplitudes of eugenol with different concentrations of βcd

Concentration ofβcd (M)	**Lifetime (ns)**			**Average Life Time < T> 10^{-9}sec**	**Relative Amplitude**			**X^2**	**S.D 10^{-11}sec**		
	T_1	**T_2**	**T_3**		**B_1**	**B_2**	**B_3**		**T_1**	**T_2**	**T_3**
0	1.53	7.15	1.38	2.0185	25.25	10.41	64.34	1.37	5.0	1.95	8.03
0.6	1.25	1.27	7.17	1.7514	61.31	29.03	9.66	1.43	1.22	2.87	2.04
1.0	1.26	6.58	1.0	1.5862	23.53	9.41	67.06	1.32	4.18	1.86	9.74

Table III: Fluorescence lifetime and amplitudes of eugenol with different concentrations of Hβcd

Concentration of Hβcd (M)	**Lifetime (ns)**			**Average Life Time < T> 10^{-9}sec**	**Relative Amplitude**			**X^2**	**S.D 10^{-11}sec**		
	T_1	**T_2**	**T_3**		**B_1**	**B_2**	**B_3**		**T_1**	**T_2**	**T_3**
0	2.83	1.22	6.06	2.9929	94.45	3.26	2.29	1.22	5.10	5.38	1.91
0.6	1.25	1.27	7.17	1.7514	61.31	29.03	9.66	1.43	1.22	2.87	2.04
1.0	1.42	1.26	6.40	1.4242	99.76	0.15	0.09	1.42	1.42	5.21	2.19

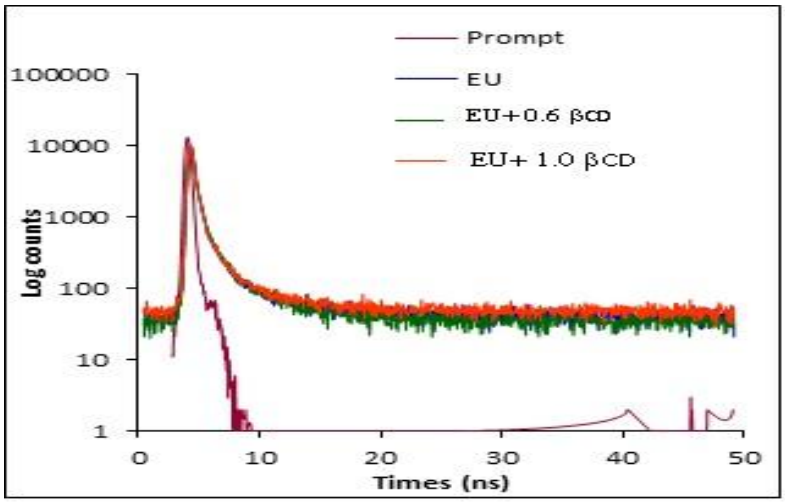

Figure 9: Time-resolved fluorescence spectra of eugenol with different concentrations ofβ-Cyclodextrin (mol dm^{-5}) (1)0, (2) 0.6, (3) 1.0

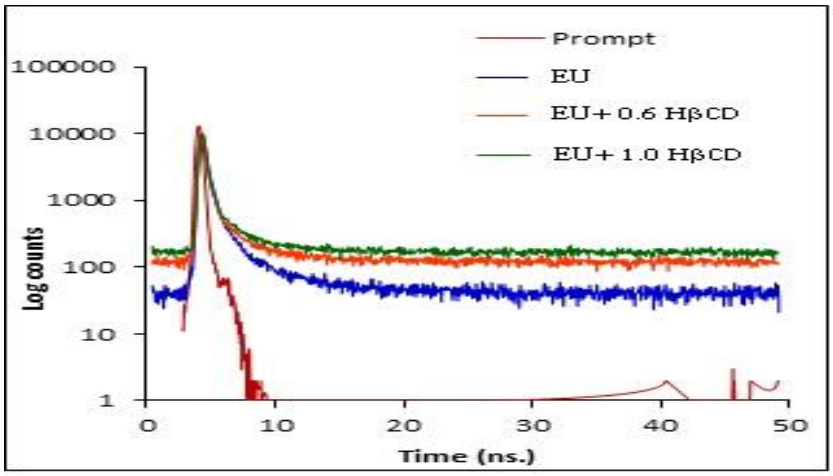

Figure10: Time-resolved fluorescence spectra of eugenol with different concentrations ofHβ-Cyclodextrin (mol dm^{-5}) (1)0, (2) 0.6, (3) 1.0

3.4: Fourier Transform Infrared (FTIR) Spectroscopic Study

FTIR spectra have been recorded to investigate the inclusion complex formation of eugenol – βcds.The FTIR spectra of Eugenol is shown in Fig. 11.The FTIR spectra ofβcd and Hβcd are given in Figs.12&13.

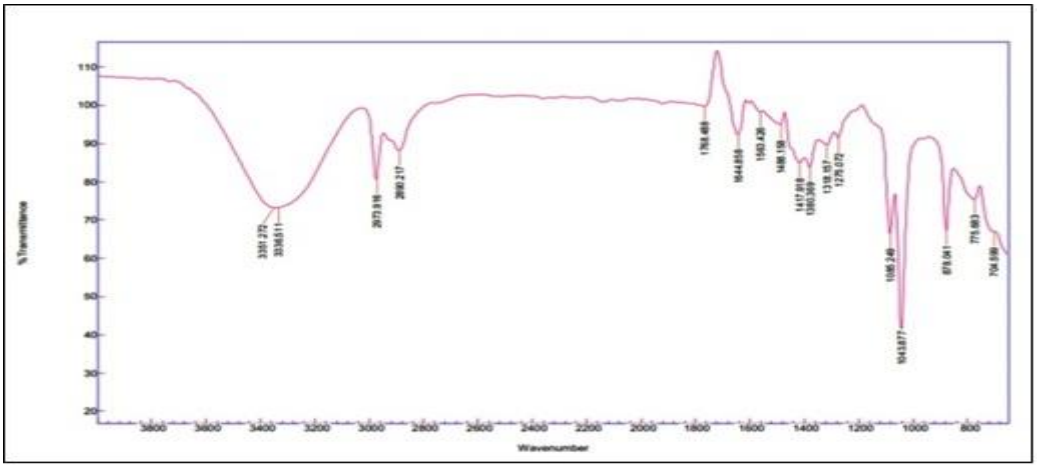

Figure 11: FTIR spectra of Eugenol

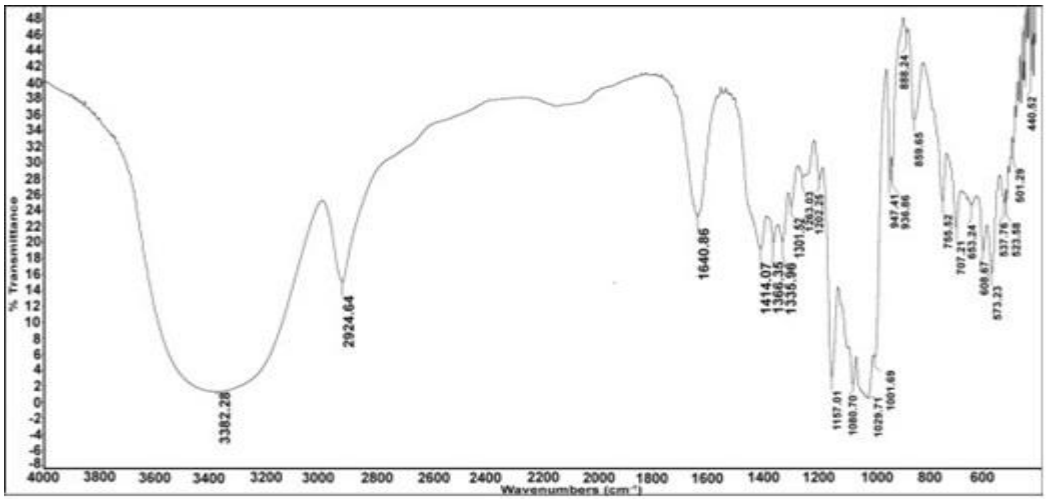

Figure 12:FTIRspectra of β-Cyclodextrin

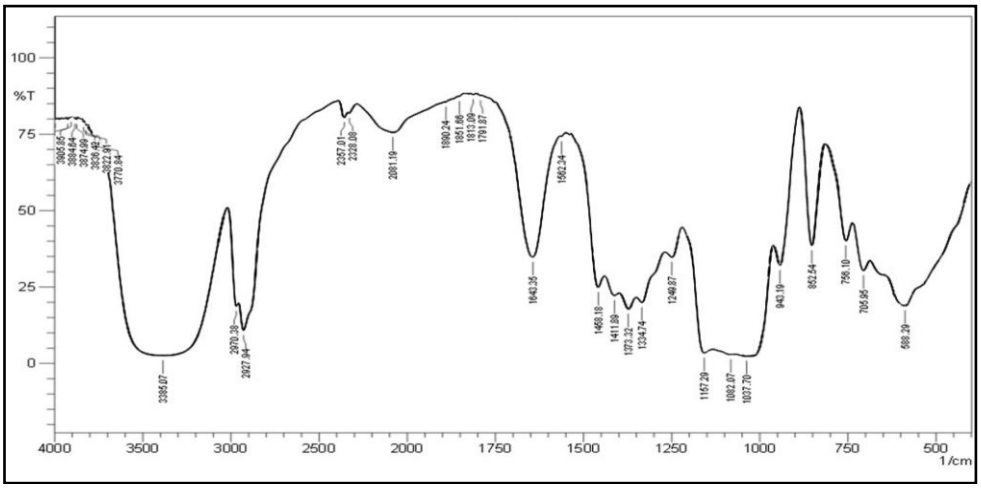

Figure13: FTIR spectra of Hβ-Cyclodextrin

IV. Conclusion:

Spectroscopic investigations have been carried out such as, UV/Vis, steady state and time resolved fluorescence and Fourier transform infra-red (FTIR) spectroscopy using one of the ingredients of tulasiherb, eugenol adding to it beta cyclodextrin (βcd) and hydroxyl propyl beta cyclodextrin (Hβcd). Hopefully these results will aid in judging their data during the evaluation of binding mode and designing a new rational herbal – interactive drugs.

References:

[1]. S. N. Ebrahimi, J. Hadian, M. H. Mirjalili, A. SonboliAnd M. Yousefzadi, Essential Oil Composition And Antibacterial Activity Of Thymes CaramanicusatDifferent Phonological Stages,Food Chemistry, 110 (2008) 927-931.
[2]. R. Schmid, A Resolution Of The Eugenia-Syzygiumcontroversy (Myrtaceae),American Journal Of Botany, 59 (1972) 423-436.
[3]. L. A.Shelef, Antimicrobial Effects Of Spices,Journal Food Safety, 6(1983) 29-44.
[4]. C. G. Soto And Burhanuddin, Clove Oil As Fish AnaesthesicFor Measuring Length And Weight Of Rabbit Fish (Siganuslineatus),Aquaculture, 136 (1995) 149-152.
[5]. J. L. Ackerman And D. R. Bellwood, Comparative Efficiency Of Clove Oil Vs Rotenone For Sampling Tropical Reef Fish Assemblages,Journal Fish Biology, 60 (2002) 893-901.
[6]. G. N. Wagner, T. D. Singer And M. Mckinley, The Ability Of Clove Oil And MS-222 To Minimize Handling Stress In Rainbow Trout (Oncohynchus Mykiss Walbaum),Aquaculture Research, 34 (2003) 1139-1146.
[7]. P.HoskonenAnd J.Pirhonen, The Effect Of Clove Oil Sedation On Oxygen Consumption Of Six Temperate- Zone Fish Species,Aquaculture Research, 35 (2004) 1002-1005.
[8]. R. Roubach, L. C. Gómez, F. A. FonescaAnd A. L. Val, Eugenol As An Efficacious AnaesthesicFor Tambaqui, Colosomamacropomum(Cuvier),Aquaculture Research, 36(2005) 1056-1061.
[9]. T. Atsumi, I. Iwaka, S. Fujisawa And T. Ueha, Reactive Oxygen Species Generation And Photo-Cytotoxicity Of Eugenol In Solutions Of Various Ph,Biomaterials, 22 (2001)
[10]. 1459-1466.
[11]. S. Fujisawa, T.Atsumi, Y.KadomaAnd H.Sakagami, Antioxidant And Prooxidant Action Of Eugenol Related Compounds And Their Toxicity,Toxicology, 177 (2002)39-54.

[12]. M.Ito, K.MurakamiAnd M.Yoshino, Antioxidant Action Of Eugenol Compound: Role Of Mental Ion In The Inhibition Of Lipid Peroxidation,Food Chemistry Toxicology, 43(2005) 461-466.
[13]. J. L. Briozzo, J. Chirife, L. HerzageAnd M. Dáquino, Antimicrobial Activity Of Clove Oil Dispersed In A Concentrated Sugar Solution, Journal Of Applied Bacteriology, 66(1989) 69-75.
[14]. A.OzturzAnd H.Ozbek, The Anti-Inflamatory Activity Of EugeniacaryophyllataessentialOils: An Animal Model Of Anti-Inflammatory Activity, European Journal Of General Medicine, 2(2005)159-163.

Figure Footnote

Figure 1: Absorption spectra of eugenol in different concentrations of β-Cyclodextrin (mol dm^{-5}) (1) 0, (2)0.2 (3)0.4, (4)0.6, (5)0.8, (6)1.0
Figure 2: Absorptionspectra of eugenol in different concentrations ofHβ-Cyclodextrin (mol dm^{-5}) (1) 0, (2) 0.2 (3) 0.4, (4) 0.6, (5) 0.8, (6) 1.0
Figure 3: Absorbance of eugenol versus βcd concentrations
Figure 4: Plot of $\left[\frac{1}{A_0-A}\right]$ versus $\left[\frac{1}{\beta cd}\right]$ for Eugenol
Figure 5: Fluorescence spectra of eugenol in different concentrations of
β-Cyclodextrin (mol dm^{-5}) (1)0, (2)0.2 (3)0.4, (4)0.6, (5)0.8, (6)1.0
Figure 6: Fluorescence spectra of eugenol in different concentrations of
Hβ-Cyclodextrin (mol dm^{-5}) (1)0, (2)0.2 (3)0.4, (4)0.6, (5)0.8, (6)1.0
Figure 7: Plot of fluorescence intensity of eugenol versus βcds concentrations
Figure 8: Plot of $\left[\frac{1}{I-I_0}\right]$ versus $\left[\frac{1}{\beta cd}\right]$ forEugenol
Figure 9: Time-resolved fluorescence spectra of eugenol with different concentrations of
β-Cyclodextrin (mol dm^{-5}) (1)0, (2) 0.6, (3) 1.0
Figure 10: Time-resolved fluorescence spectra of eugenol with different concentrations of
Hβ-Cyclodextrin (mol dm^{-5}) (1)0, (2) 0.6, (3) 1.0
Figure 11: FTIR spectra of Eugenol
Figure 12: FTIR spectra of β-Cyclodextrin
Figure13: FTIR spectra of Hβ-Cyclodextrin

Table Footnote

Table I: Formation constantK (M^{-1}) and free energy ΔG ($kJmol^{-1}$) ofEugenol withβcd and Hβcd
Table II: Fluorescence lifetime and amplitudes of Eugenol with different concentrations of βcd
Table III: Fluorescence lifetime and amplitudes of Eugenol with different concentrations of Hβcd

Bakkialakshmi.S"Fluorimetric Study of Eugenol with Cyclodextrins"International Journal of Engineering Science Invention (IJESI), vol. 07, no. 03, 2018, pp30-37