IJNRD.ORG ISSN : 2456-4184
INTERNATIONAL JOURNAL OF NOVEL RESEARCH AND DEVELOPMENT (IJNRD) | IJNRD.ORG
An International Open Access, Peer-reviewed, Refereed Journal

A STUDY OF UV – VISIBLE AND IR SPECTRAL DATA OF LYCOPENE IN SELECTED SOLVENTS EXTRACTED FROM BIOLOGICAL SOURCES

Mr .[1]Srikanta A S *, Mr.[2]Ujwal S S *, Mrs.[3]Kusum B Y *, Mr.[4]Sunil M C *, Mrs.[5]Gouthami R*, Mr.[6]Sunil L N*, Mr.[7]Varun Manjunath

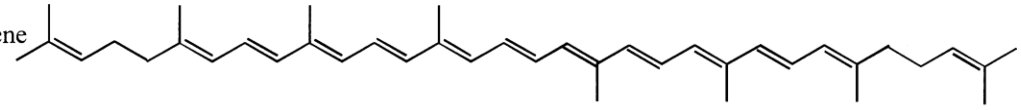
[1]Associate Professor, Department of Biochemistry
[2, 3, 4, 5, 6, 7] Undergraduate students, Department of Biochemistry
Vijaya college, R V road, Basavanagudi, Bangalore, India

Abstract: Lycopene is one of the carotenoid pigments naturally occurring in red fruits and vegetables. The most common source for scientific studies on lycopene is from the sources like tomato, watermelon and papaya. It is an antioxidant containing polyene chain with high chemical reactivity, when present in the human system and shows reaction with several oxidizing agents and free radicals generated during several detoxification reactions. The role of lycopene in removal of toxic molecules generated is well recorded. They find major role in prevention of human health disorders including heart diseases. It occurs in cis and tans isomers. Lycopene is synthesized only in plants and its molecular structure undergoes several modifications depending on the presence of solvents. The extraction method adopted includes obtaining the lycopene solid from the aqueous layer which is generally discarded in regular Benzene extraction method. The study of spectral data in selected solvents is carried out with extracted lycopene sample and an attempt is made to elaborate and interpret the spectral data using FT-IR and UV-Visible spectrophotometer. The structural studies help us in understanding possible mechanisms involved in the role of lycopene in alleviating life style disorders and mal-functions in several organ systems.

Key words: Antioxidants, solvent extraction, benzene extraction, UV – Visible spectroscopy and FT – IR spectral data.

I. INTRODUCTION

Fruits and vegetables are the main source of natural antioxidant components *(1).* One of the most efficient carotenoid antioxidants is lycopene *(2).*Studies have shown that Lycopene is a natural pigment which protects the body by neutralizing the negative effects of oxidants and free radicals in the body, regular intake of lycopene containing food reduces heart diseases and risk of body tumor especially prostate cancer*(3,4).*Some of natural sources of lycopene are tomato, watermelon, papaya, pink Guava and apricots*(5).*Lycopene is a Polyene which is tetra terpene hydrocarbon, an acyclic open chain, unsaturated carotenoid consisting of 13 double bonds of which 11 are conjugated double bonds arranged in linear array and the molecular formula of lycopene isC_{40} H_{56} having molecular weight of 536.85 Da. Lycopene in nature is of all trans forms since lycopene is unstable in heat, light and chemical reaction conditions, seven bonds in lycopene isomerizes into mono or poly cis *(6).* Lycopene is soluble in chloroform, benzene and other organic solvents but insoluble in water*(7,8).* In this work, lycopene is extracted from watermelon, papaya, guava by solvent extraction using benzene as the main solvent and checking the purity of the extracted lycopene by performing TLC, and spectral analysis of the extracted lycopene sample in UV – visible spectroscopy under selected solvents and FT-IR spectroscopy.

All *trans* lycopene 

`Fig 1.1: Structure of $C_{40}H_{56}$ Lycopene (

AIM AND OBJECTIVE

AIM – To extract lycopene by using benzene solvent extraction process and study UV – Visible and IR spectra in selected solvents.

OBJECTIVES

1. To extract pure lycopene and check its purity by using TLC.
2. To find out lycopene structural changes in different solvents using UV- visible spectra.
3. To study FT-IR spectral data and interpret lycopene structure.

II. MATEIAL AND METHOD

2.1: Material

Biological sources: Citrullus lanatus (Watermelon), Psidium guajava (pink guava), Carica papaya (papaya).
Chemical: Benzene, Na_2CO_3, Acetone, Chloroform, ethanol, toluene, xylene, THF, Silica gel-G.

2.2: Methods

2.2.1: Methodology: Lycopene extraction, most of the research papers were achieved by using tomato source. As the structure of lycopene practically remains same in all the remaining sources, three major sources were selected for this project namely Citrullus lanatus (watermelon), Psidium guajava (pink guava), Carica papaya (papaya). The selected sources are rich in lycopene content and few research papers were published from these sources *(9,10).*The method includes a common procedure in which 150g of pulp sample was taken and was finely blended by mixer to obtain a paste. 30g of paste is weighed and transferred into the separating funnel. 30ml of equal volume of Benzene is added successively into the separating funnel for every five times of each stage and stirred for 15 minutes. The aqueous layer containing the target lycopene in upper layer is formed by agitating the solvent in the funnel. Now the mixture is filtered into a beaker using funnel to remove insoluble fiber using a pinch of Sodium Carbonate (Na_2CO_3). The filtrate organic solution is kept for evaporation in the hot air oven by adding boiling chip. The organic liquid undergoes complete evaporation in 2-4hours leaving behind brownish red solid lycopene compound. The high yield in papaya and low yield in other two sources indicated the content of target compound in the respective sources. The method was employed for all the other sources in the research papers *(11, 12).*

The purity of sample extracted was checked using thin layer chromatography. The lycopene extracted from the three samples were analyzed in UV-Visible spectral data under different solvents and FT-IR spectral data, were studied and interpreted.

Fig 2.1: Lycopene extracted from watermelon

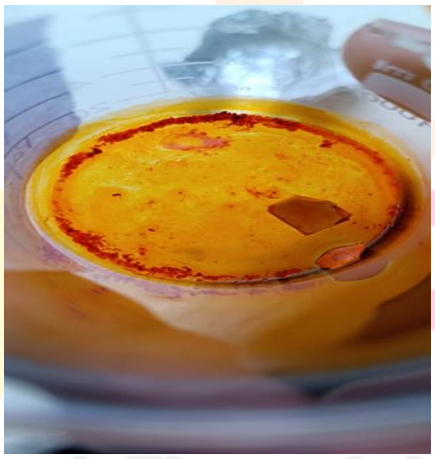


Fig 2.2: Lycopene extracted from papaya

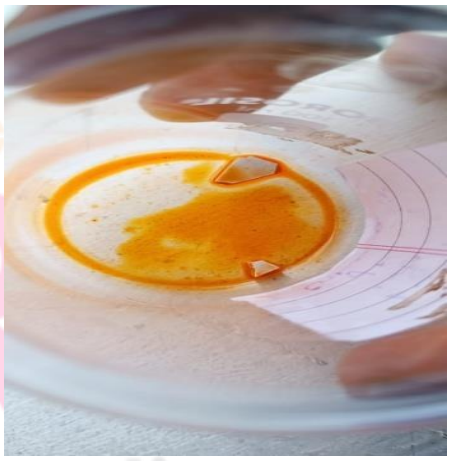


Fig 2.3: Lycopene extracted from Guava

III. RESULTS AND DISCUSSION

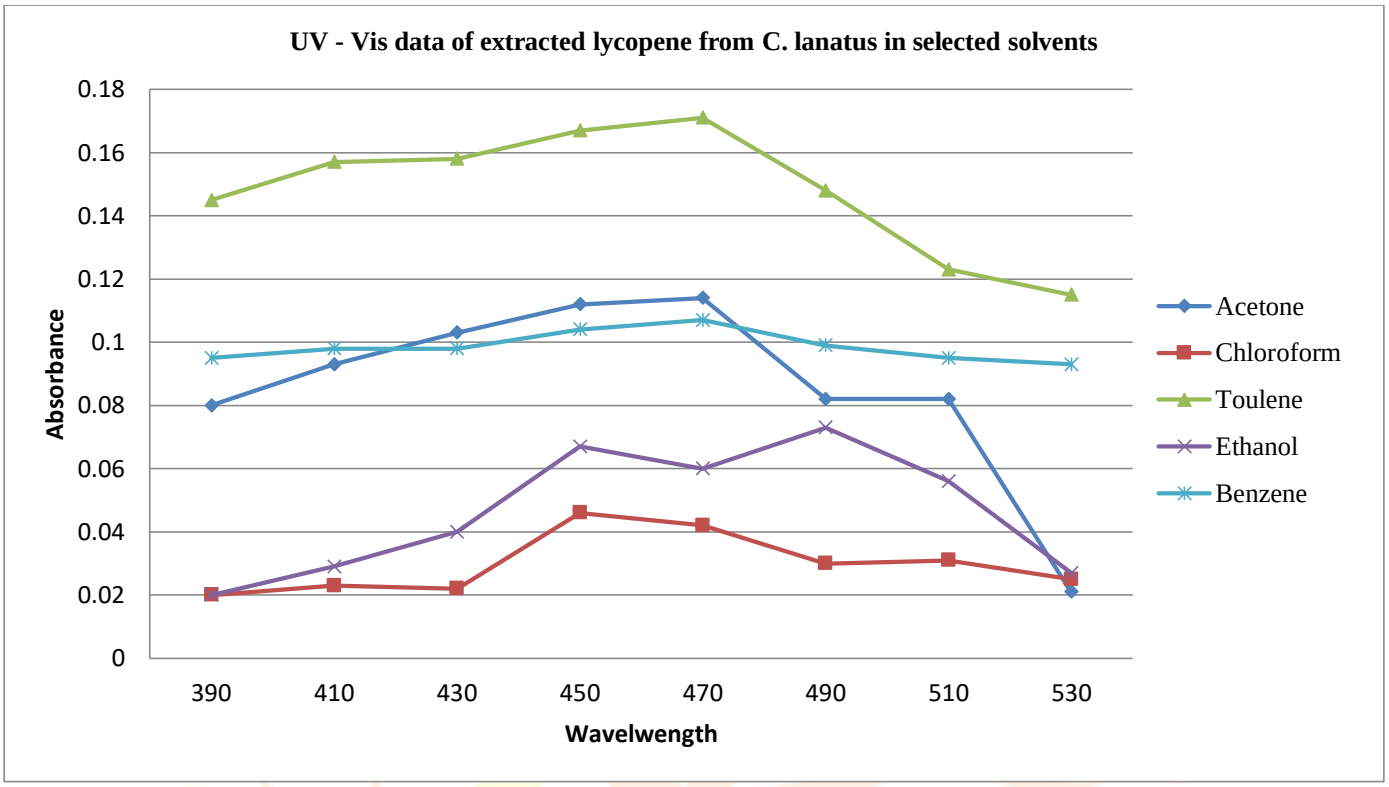
3.1: Thin Layer Chromatography Results: TLC was performed using Benzene, THF, Xylene. The reported Rf value for lycopene extract is 0.64 *(12)*.

Sources	Solvents	Distance of the solvent travelled on the TLC plate	Distance of the spot travelled on the TLC plate	R_f value
Citrullus lanatus (Watermelon)	Benzene	3.3	2.3	0.69
	THF (Tetrahydrofuran)	3.3	2.0	0.66
	Zylene	3.6	2.4	0.66
Carica papaya (papaya)	Benzene	4.3	2.9	0.67
	THF (Tetrahydrofuran)	4.5	3.0	0.66
	Zylene	3.6	2.4	0.66
Psidium guajava (Guava)	Benzene	3.7	2.5	0.6
	THF(Tetrahydrofuran)	3.9	2.5	0.64
	Zylene	3.5	2.2	0.62

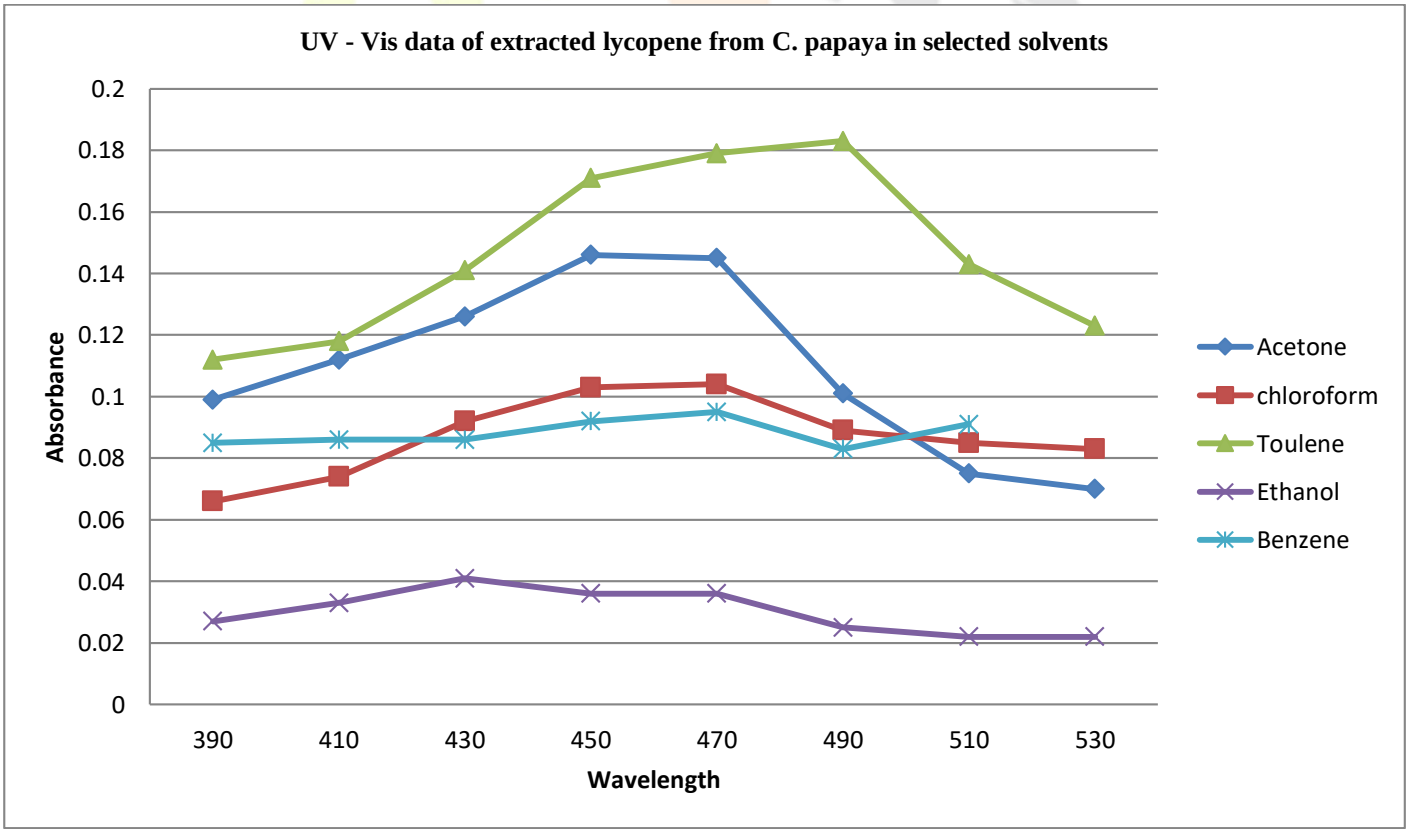
3.2: UV visible spectroscopy

UV visible spectroscopy was performed after TLC. Lycopene extracted from watermelon, papaya, and guava was subjected with selected solvents based on the solubility property of the lycopene. The solvents used in UV visible spectroscopy are acetone, chloroform, benzene, ethanol *(7, 8)* and toluene. Lycopene extracted from three fruits were dissolved into all the above mentioned solvents separately and the spectral data was noted and graphs were plotted accordingly.

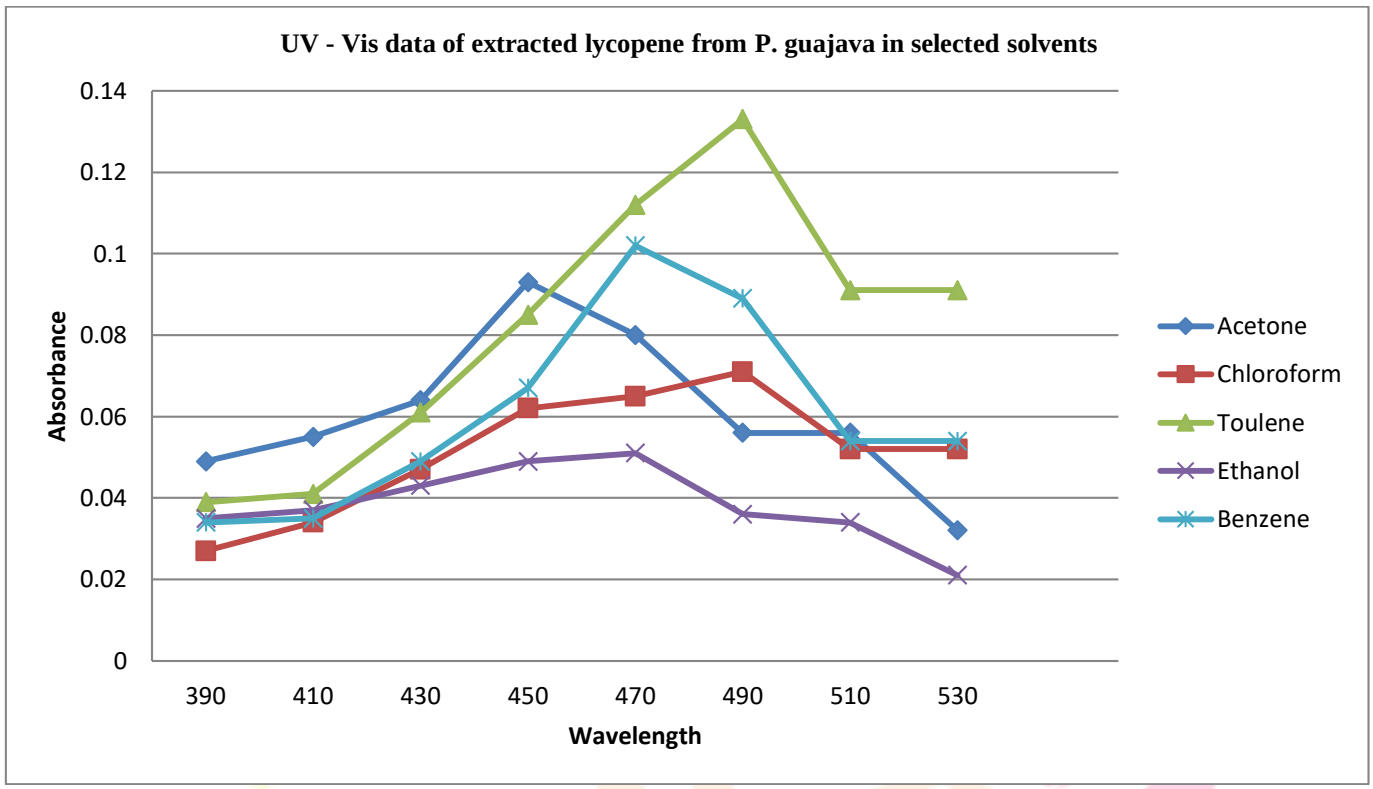
UV – Visible data of extracted lycopene from C.lanatus in selected solvents



UV – Visible data of extracted lycopene from C. papaya in selected solvents



UV – Visible data of extracted lycopene from P. guajava in selected solvents



The spectral data obtained in benzene extracted lycopene sample is checked using acetone solvent showed the peaks at 420nm, 454nm, 470nm and 510nm respectively are found almost typically same for the sample analyzed in the sample solvent *(11)*. This helps us to draw conclusion that the sample extracted is pure and methodology is correct. The 11 double bonds in the molecule are responsible for the prominent peaks in which the electronic transition gives the spectral output. The peak at 420nm is prominently absent in all the sources used in the project. It can be reasoned to solvent compound interaction during UV – Visible analysis, it requires further more studies. The research papers with the spectral data 420nm peak observed is due to use of acetone solvent extraction in which solvent compound interaction gives the peak. The project team has used the other solvents like ethanol and toluene and the results obtained require further study and interpretation.

3.3: Infrared spectroscopy

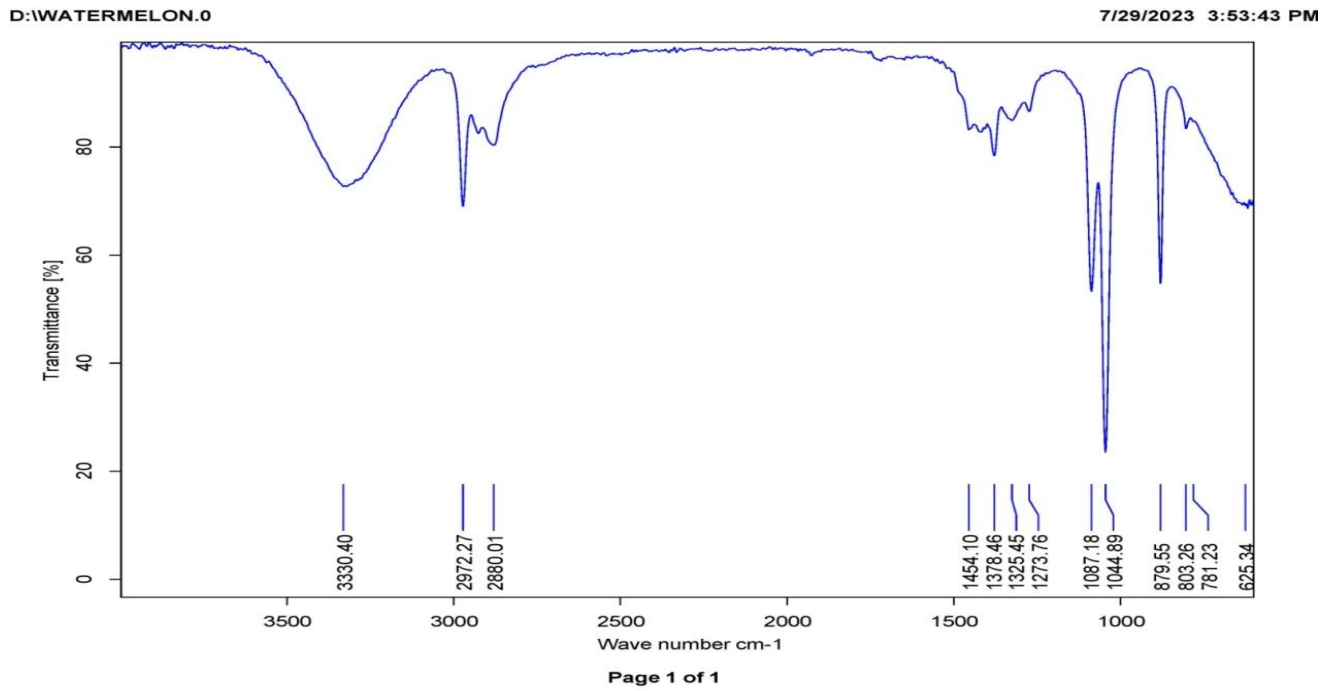


Fig 3.1: FT-IR graph of Watermelon

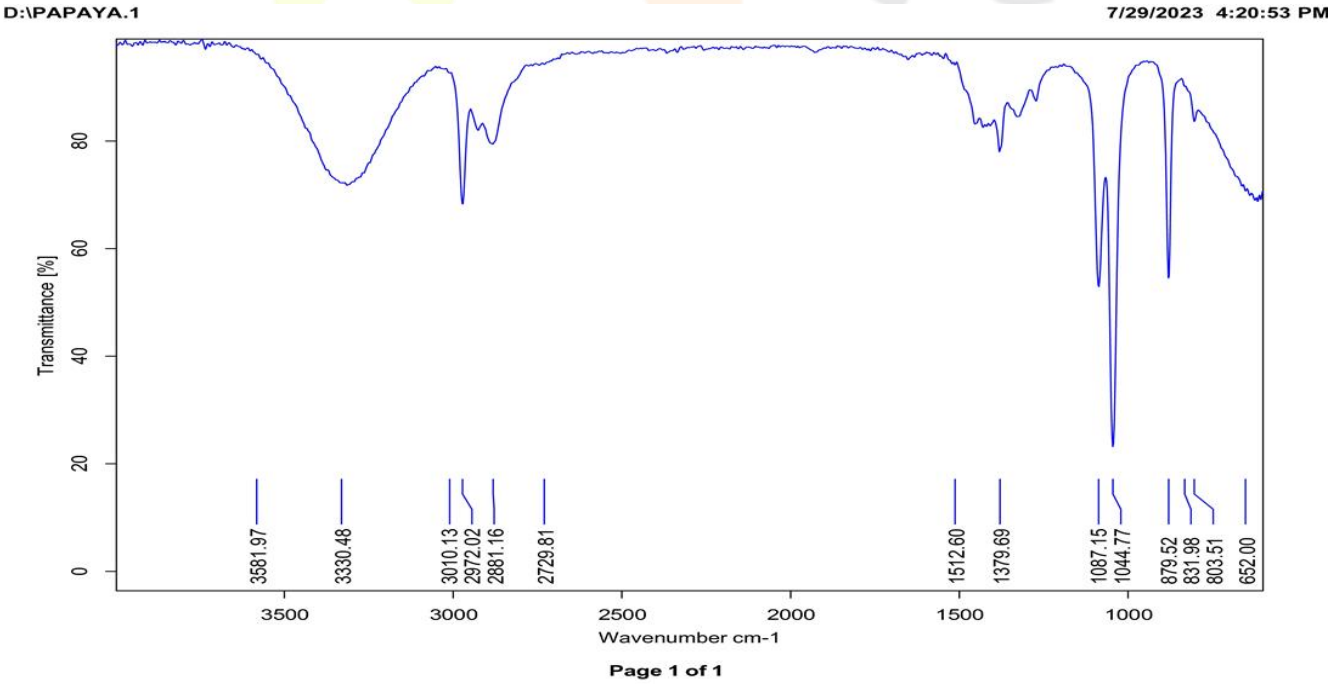


Fig 3.2: FT-IR graph of Papaya

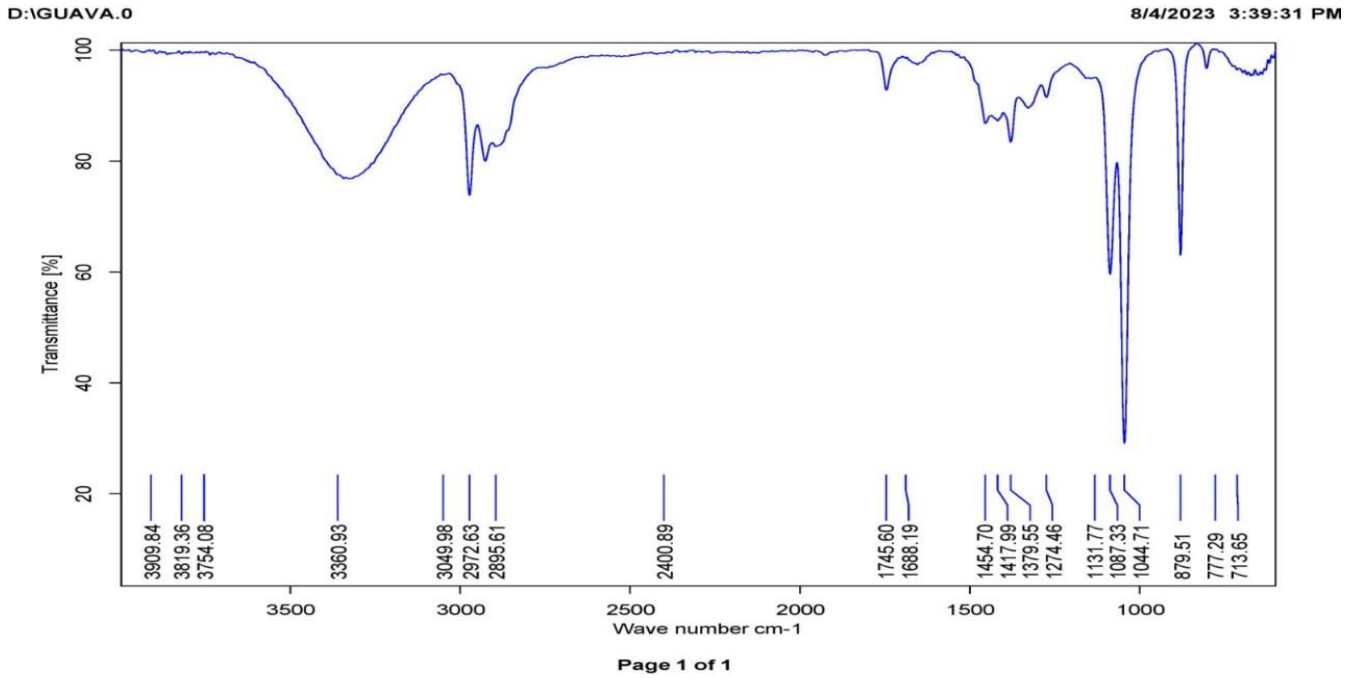


Fig 3.2: FT-IR graph of Guava

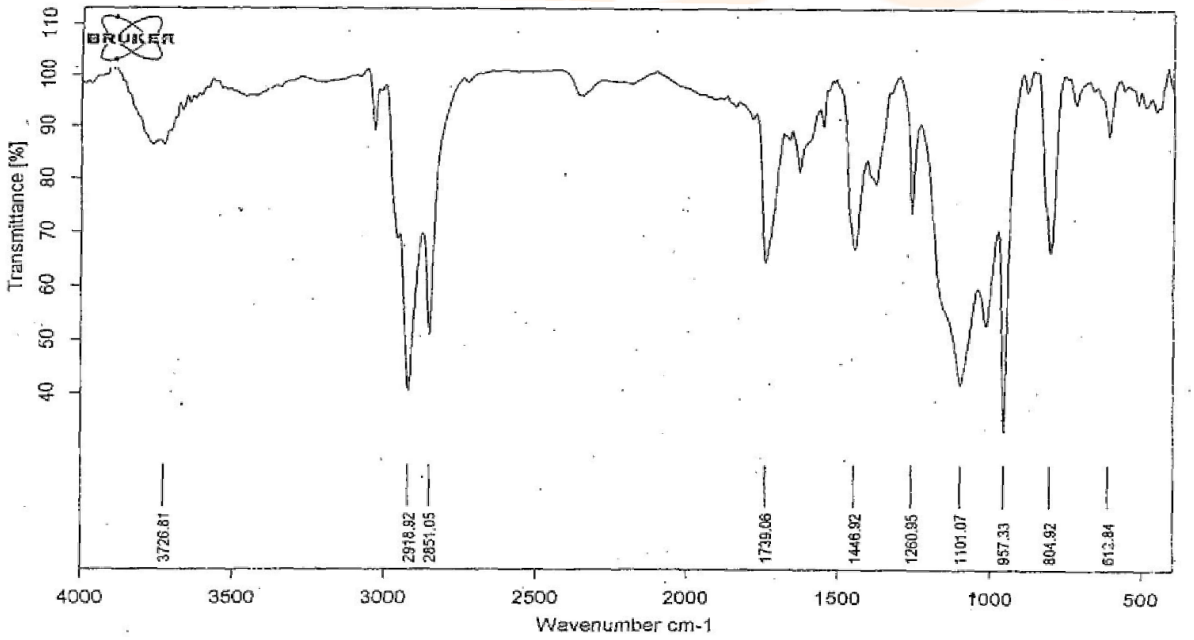


Fig 3.4: Standard FT-IR graph in KBr (Aghel N et, al., 6(1), 9-15, 2011)

The FT-IR spectral band obtained is typically similar to that of Aghel N et, al., 6(1), 9-15, 2011. The FT - IR graphs obtained from Bruker alpha in ATR of each from 3 different fruits samples indicated common peaks 1045 cm^{-1} region which is a common observation in lycopene indicating C-H and C-C bending and stretching. Other groups such as lipid groups in 2800 - 2900 cm^{-1} and C-C and C-H out the plane bending at 600-900 cm^{-1} and water molecules in 3000-3500 cm^{-1} is observed *(14)*. The present work on lycopene is carried out using three different sources and the graphs obtained for pure sample is unique and requires further study and interpretation. This project aimed to produce FT-IR data for the sources and the comparison to the standard KBr based IR data requires more studies.

Table 3.15: FT-IR of lycopene from Aghel N et,al., 6(1), 9-15, 2011

Wavelength	Phenomenon
3100 (cm^{-1})	CH str (SP2)
2918.92, 2851.05 (cm^{-1})	CH str (SP3)
1670, 1640 (cm^{-1})	C= C str (Trans)
1446.92, 1400 (cm^{-1})	CH2 (Bending)
1101.07, 1000, 957.33 (cm^{-1})	CH (Trans OOP)
612.84 (cm^{-1})	R2C=CR

IV. DISCUSSIONS AND CONCLUSION

Lycopene extracted from benzene - solvent extraction method is adopted for the sources namely, watermelon, papaya and guava are widely used. The benzene layer is usually discarded after the solidification of lycopene in aqueous phase. In normal procedure the benzene layer having substantial amount of lycopene is evaporated along with the solvent. Presently the Benzene layer was used for extraction of lycopene by evaporation method. The extraction of lycopene in this work is that benzene evaporation is too long 2-4 hours. In the available literature to get UV – Visible spectra, the solvents used are benzene, chloroform and acetone *(11)*. Apart from this the spectra are also obtained by using toluene and ethanol solvents. A total of five solvents were employed for every dilute solution of lycopene to get spectra. The absorbance graphs obtained shows minimum interference due to the presence of β carotene from the available sources. From the present work the ⋋max value obtained in all solvents shows peak at 450nm and 470nm except in toluene and ethanol solvents 480nm and 490nm were obtained in toluene and ethanol. This work helps to determine the concentrations and thereby quantifications. The organic solvents such as chloroform and ethyl alcohol help to study the lypophilic characteristics of β carotene. The structural analysis of lycopene in different solvents can be done with the spectral data. The reported vibrational wavelength of FT-IR spectrum is 3100 cm^{-1} in most reported works, it corresponds to C-H bond stretching (sp^2). There are several peaks in spectra between 2920 cm^{-1} and 2850 cm^{-1} indicating symmetric and asymmetric of C-H bond. From 3360 to 2800cm-1 all the graphs showed similar peaks due to the presence water and lipid. The bending stretching of C-H and C-C bond is observed between 1044 to 1445 cm^{-1}. Since the conjugated bond system involves absorption location between 1640 to 1680 cm^{-1} the lower value for C-C double bonds indicates the solvent interference. The molecule of lycopene is characterized by the presence of not only double bonds and also cyclohexene ring and cyclopentene, the IR graphs shows only stretching and bending frequencies of C-H and C-C bonds. Since lycopene is an isoprenoid lipid molecule which is nontoxic, it can be employed as antioxidant, anti-inflammatory medicine. The project on FT-IR spectrum gives input to study the structural relation with different organic solvents and thus can be used as potential drug in chemotherapy

V. References

1. Charanjit Kaur, Harish C. Kapoor. Antioxidants in fruits and vegetables the millennium's health, International Journal of Food Science and Technology 2001, 36, 703-725.
2. Paolo Di Marcio, Stephan Raiser, Helmut Sies. Lycopene as the Most Efficient Biological Carotenoid. Archives of biochemistry and Biophysics 1989, 274(2), 532-538.
3. A. V. Rao, Sanjiv Agarwal. Role of Antioxidant Lycopene in Cancer and Heart Disease. Journal of the American College of Nutrition 2000, 19(5), 563-569.
4. Anita Agarwal, Honglei Shen, Sanjiv Agarwal, A.V.Rao. Lycopene Content of Tomato Products: Its Stability, Bioavailability and In Vivo Antioxidant Properties. Journal of medicinal food 2001, 4(1), 9-15.
5. Tanak, T., Shimizu, M. and Moriwaki, H. (2012) Cancer chemoprevention by carotenoids.Molecules 17: 3202-3242.

6. Greory A. Chasse, Melody L. Mak, Eugen Deretey, Imre Farkas, Ladislaus L. Torday, Juslius G. Papp, Dittakavi S.R. Sarma, Anita Agarwal, Sujatha Chakaravati, Sanjiv Agarwal, A. Venket Rao. An ab initio computational study on selected lycopene isomers. Journal of molecular structure (theochem) 57 (2001) 27-37.
7. Shi, J. and Le Maguer, M., Lycopene in Tomatoes: Chemical and Physical Properties Affected by Food Processing. Critical Reviews in Food Science and Nutrition 2000, 40, 1-42.
8. Clinton SK, Emenhiser C, Schwartz SJ, Bostwick DG, Williams AW, Moore BJ, Erdman JW. Cis-trans lycopene isomers, carotenoids and retinol in the human prostate. Cancer Epidemiol Biomarkers and prevention vol 5, 1996, 823–833.
9. Neelu Malviya(2014), Isolation and Quantification of Lycopene from Watermelon, Tomato and Papaya, Vol 3, pp 2277-2502.
10. Kin-Weng Kong, Nor Fadilah Rajab, K. Nagendra Prasad, Amin Ismail, MasturahMarkom, Chin-Ping Tan. Lycopene-rich fractions derived from pink guava by-product and theirpotential activity towards hydrogen peroxide-induced cellular and DNA damage, Food Chemistry, 123 (2010), 1142–1148.
11. Vrinda Nair, Simran Lilwani, Extractionand isolation of Lycopene from Solanunlycopersicum and Citrullus Lazarus for bioplastic colouring (2016), 5, 2277-8616
12. V. Judia Harriet Sumathy. Identification of Lycopene extracted from Papaya Using Thin Layer Chromatography and FT-IRStudies. Int. J. Curnt. Tren. Pharm, Res., 2016, 4(6): 351-354.
13. Aghel N, Ramezani Z, Amirfakhrian S(2011), Isolationand qualification of lycopene from tomato cultivated in Dezfoul, Iran,Jundishapur Journal of Natural Pharmaceutical Products, 6(1),9-15.
14. Ramakrishna Shabaraya A, Viresh K Chandur. Spectrophotometric Characterization of lycopene Phytosomes from FruitPeels of Lycopersicum esculentum. Indian J Pharm Drug Studies,2023, 4(3).