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RESEARCH ARTICLE

PROCESS OF ISOLATION AND CHARACTERIZATION OF LYCOPENE FROM GRAPEFRUIT

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ABSTRACT

Lycopene is a natural, red-coloured, organic compound notable for its antioxidant properties. Red pigment with a simple molecular formula ($C_{40}H_{56}$), as a good antioxidant is beneficial for human health. This research represents the positive properties of lycopene and its protective effect on the cardiovascular system; it reduces blood pressure, prevents the oxidation of LDL cholesterol, lipids, etc. This red carotenoid has a number of health effects resulting from its antioxidant effect. In this experimental research, lycopene was isolated by a simple procedure from red grapefruit. The solvents used for the isolation procedure are acetone and petroleum ether. The identification and characterization of lycopene was confirmed by FTIR, UV/Vis and TLC methods.

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INTRODUCTION

Lycopene is a pigment that is primarily responsible for the characteristic dark red color of ripe fruits, vegetables, and is also present in algae and fungi. Tomatoes and their products are significant sources of this carotenoid. In addition to vegetables, fruits are also a significant source of natural lycopene (red grapefruit, watermelon, papaya, guava and apricots(Nguyen and Schwartz, 1999: 38-45).Grapefruit is a tropical plant of the citrus family that has a bitter taste. Figure 1 shows a grapefruit fruit. Natural pigment, a significant antioxidant that fights the damage of free radical, heart health, bones, high blood sugar levels and various types of cancer. Its molecular weight is 536. 89 and molecular formula is $C_{40}H_{56}$ with 89.45 % carbon and 10.51% hydrogen(Aghel,Ramezani,. Amirfakhrian, 2011: 9-15).Soluble in chloroform, hexane, benzene, carbon disulfide, acetone, petroleum ether and oil; and insoluble in water, ethanol and methanol (Roxana-Mădălina S., Tomulescu C., Căşărică A., Soare M.G., V. 2018, 2285-1364, 2285-5521, 2285-1372, 2285-1364). Due to its biological and physico-chemical characteristics, it attracted attention.Sensitivity of lycopene is light, oxygen, high temperature and acids (Naz, Butt, Sultan, Qayyum, Niaz, 2014: 650-666).Figure 2shows the chemical structure of lycopene. Lycopene, as a red pigment, consists of unsaturated hydrocarbons in whose molecule we have eleven conjugated and two non-conjugated bonds. Colorattraction in carotenoids is attributed to the presence of a long conjugated double bond system as it forms the light absorbing chromophore in these compounds(Rodriguez-Amaya, Kimura,2004). In general, linear hydrocarbon chains of carotenoids such as lycopene undergo various structural modifications (Telegina ,Yuliya, Vechtomova , Aybush , Buglak , Kritsky,2023).Carotenoid that is converted into cis and trans isomers under the influence of sunlight, thermal energy and chemical reactions (Zechmeister, LeRosen, Went, Pauling, 1941:468-474., Nguyen , Schwartz, 1999: 38-45).The structure of this compound is responsible for its red color and its antioxidant properties (Supatra, 2019: 08-14).Various researches show that lycopene can be used for the treatment of prostate cancer

(Matlaga, Hall, Stindt and Torti, 2001: 166,613., Giovannucci, 1999: 91, 317.). Lycopene, which is responsible for the red color found in fruits and vegetables, plays an important role in the biosynthesis of many carotenoids. The crude extracts containing lycopene typically include several components with similar physical and chemical properties to the desired target compound which can interfere with finally separating the target compound from the crude mixture (Horosanskaia et al., 2017).It is a compound that has attracted attention with other carotenoids because of its natural antioxidant activity.Because of its properties as an organic molecule, it was taken for research.Lycopene is also known for its use in the industrial, food and pharmaceutical industries. Its use in the food industry is as a natural colorant derived from fruits and vegetables. The importance of lycopene in the pharmaceutical industry is such that it plays an important role in the body to trap free radicals and as such can prevent unwanted reactions in the body.

Figure 1. Red grapefruit[1]

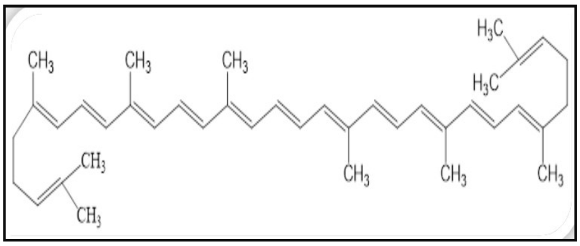

Figure 2.Chemical structure of lycopene

EXPERIMENTAL

MATERIAL AND METHODS

All reagents and solvents used were acetone (Sigma Aldrich), petroleum ether (Sigma Aldrich), hexane (Sigma Aldrich).

- FTIR spectroscopy: Analysis of isolated lycopene was performed on a Thermo Scientific Nicolet I S10 spectrometer.
- UV/ViS spectroscopy:The sample was analyzed on a UV/ViS spectrometer in the wavelength range of 200-600 nm.
- TLC analysis: The sample was analyzed on a Camag UV cabinet, where the mobile phase was hexane:acetone (9:1)(Smajlagić, Srabović, Huremović, Pehlić, Ademović, Kozarević , Ibišević and Siočić,2024: 2996-3003.), the stationary phase was silica gel.

Isolation method

- 30 grams of grapefruit was taken and transferred to a beaker containing 150 ml of acetone and stirred with a magnetic stirrer for 30 min. After that, the filtrate was taken and re-extracted with 75 ml of acetone and stirred for 15 min. After that, the filtrate was collected and a portion of the acetone extract was added to a funnel with 60 ml of petroleum ether. The sample was evaporated in a porcelain dish and placed in a desiccator until analysis.

RESULTS AND DISCUSSION

FTIR spectroscopy: The structural characterization of lycopene was confirmed by FTIR analysis and compared with pure lycopene in Figure 3. The results of the functional group analysis on isolated lycopene from grapefruit extract are presented. On the FTIR spectrum of isolated lycopene, we have a broad band at a wavelength of 3323.31 cm^{-1}, which is possible from the entry of steam into lycopene.The presence of a peak at 2929.29 cm^{-1} indicates that a C-H chain is present at that wavelength, which is consistent with literature data (Jeanne Dewi., Ririn, Zul, Nur, Ilham, Indo. 2021: 99-104).

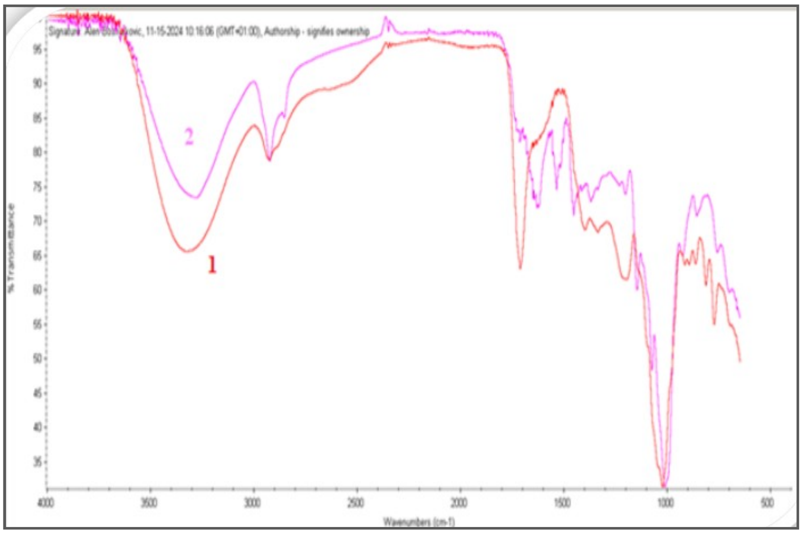

Figure 3. FTIR image of isolated lycopene (1) and pure lycopene (2)

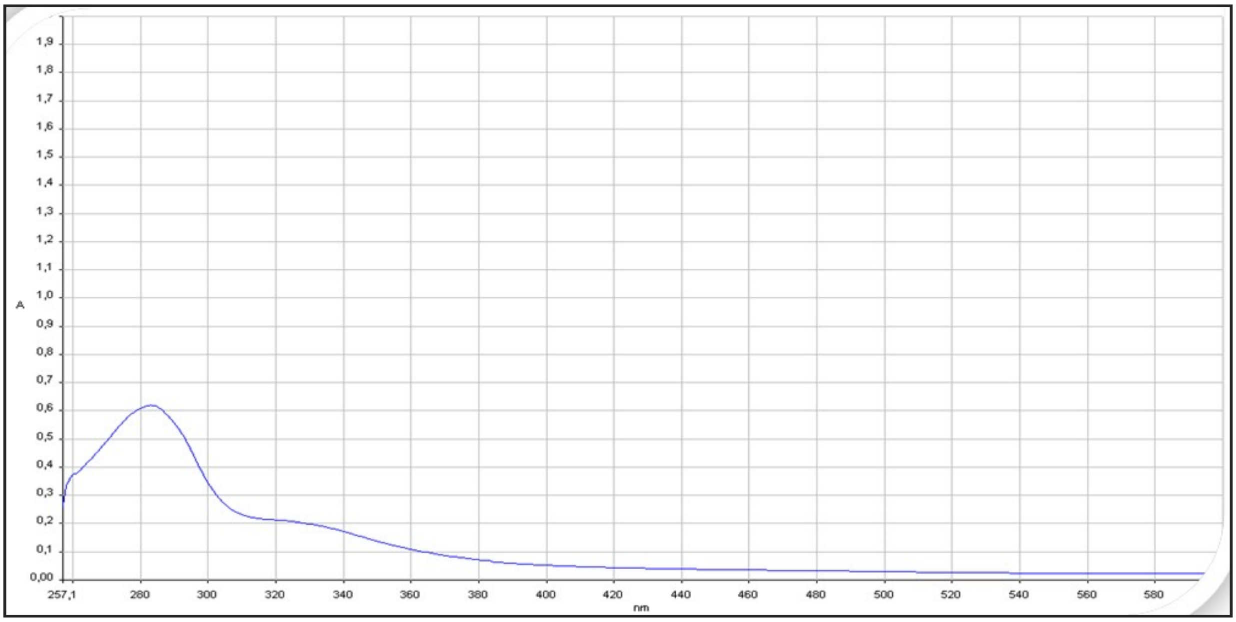

Figure 4. UV/Vis spectrum of isolated lycopene

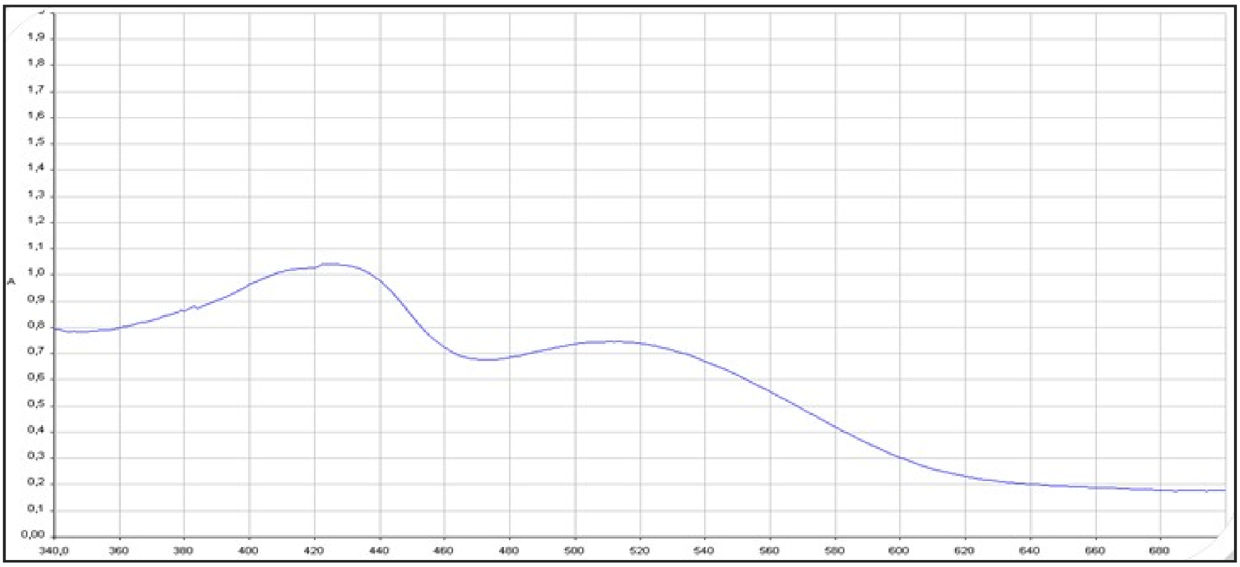

Figure 5. UV spectrum of pure lycopene

At the wavelength 1714,65 cm^{-1} we have a sharp peak that is slightly more expressive compared to the spectrum of pure lycopene. It refers to the presence of a C=C bond. The peak present at 1200 cm^{-1} and the peak at 1026.51 cm^{-1} refer to the stretching of C-C and C-C-H groups. The peak at 1026.51 cm^{-1} may also be present from a lipid or a C-O acid.The spectrum also shows a peak at a wavelength of 775.92 cm^{-1}, which also indicates deformations within the lycopene molecule.

UV/ViS spectrum: Figure 4 shows the UV spectrum of isolated lycopene from grapefruit with acetone solvent. As we can see on the spectrum, the absorption maximum at λ=283,25 nm and λ=320 nmis shown.

Comparing with literature data, there is a peak at this wavelength, consistent with the absorbance characteristics of lycopene with the presence of small impurities.Comparing

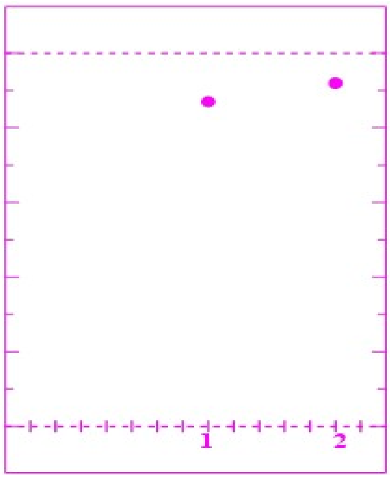

Figure 6. TLC plate of isolated (1) and pure lycopene (2)

with the spectrum of pure lycopene (Figure 5), it can be seen that the spectrum has peaks at λ=424.12 nm and λ=512.34 nm.Under the influence of the solvent, there is a shift of wavelengths to shorter wavelengths with a decrease in absorption intensity, as can be seen on the spectrum.

TLC analysis: Figure 6 shows a TLC plate showing an isolated sample of lycopene compared to the pure compound. After calculating the distance traveled by the substance, the results for isolated lycopene are Rf=0.87 and pure lycopene Rf=0.92. The solvent used is hexane:acetone (9:1) Comparing the results, it is assumed that the isolated compound has impurities present.

CONCLUSION

Lycopene, a natural compound of red color with recognized health benefits, was isolated from grapefruit and confirmed by appropriate methods. Its intake into the human body reduces the risk of many diseases such as diabetes, cardiovascular diseases, prevents the formation of cancer and many other diseases. FTIR, UV/Vis and TLC methods confirm the isolated compound with the presence of small impurities.The solvent acetone was the key solvent for this research.The main aim for future research is to purify the well-known carotenoid, lycopene.

Conflict of interest: Authors were cleared that no conflict of interest.

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