



Supercritical fluid extraction of lycopene from tomatos by using CO₂ as a solvent: A review

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ABSTRACT

Supercritical fluid extraction by using CO₂ as a solvent, a relatively new separation technique, can be used as a very efficient in course of analysis or preparation of many useful substance from many of the plant materials. The extracts from these materials are a good basis for the new pharmaceutical products & ingredients in functional foods. This paper deals with supercritical CO₂ extraction of carotenoids which are present in natural plants. Lycopene is one of the most important & useful carotenoid found in vegetable or fruits. Due to its antioxidant capacity it can prevent almost all kind of cancers, coronary heart diseases & neurodegenerative diseases. The lycopene in dried and powdered tomato skins was extracted. In this study, process parameters in supercritical extraction such as pressure, temperature, solvent flow rate, particle size & time duration for extraction are presented and studied their learning effects on the yield & quality of extracts. Extracts were analyzed by HPLC (high performance liquid chromatography) technique to determine total lycopene & trans-lycopene from total amount of yield.

Key words: Supercritical extraction, Lycopene, process parameters, tomatoes, HPLC, global extraction yield.

INTRODUCTION

In recent decades, the supercritical fluid extraction (SFE) has received special attention in the fields of solid material extraction and fractionation of liquid mixtures. Supercritical fluid extraction using carbon dioxide (SC-CO₂) is a particularly suitable isolation method for isolation of the valuable components from plant materials. A natural plant extract, free from chemical alterations brought about by heat and water, and without solvent residues and other artifacts can be obtained by this method. Moreover, conventional methods are usually carried out at high temperatures, which can be responsible for the destruction of valuable substances. Additionally, the use of organic solvents can also lead to product contamination with solvent residues. SFE method is very advantageous and environmentally friendly over other conventional either solvent or enzyme extraction methods for recovering the natural oil or carotenoid. Use of SFE technology that offers suitable extraction and fractionation appears to be promising for the food and pharmaceutical industries[1].

All products characterized by their internal content are claimed by the market's consumers to have a reproducible and stable quality. Technological solutions are sought that ensure these properties of the products and satisfy the standards in a more reproducible way[2].

The SFE is a separation technology that uses supercritical fluid as the solvent. Every fluid is characterized by a critical point, which is defined in terms of the critical temperature and critical pressure. Fluids cannot be liquefied

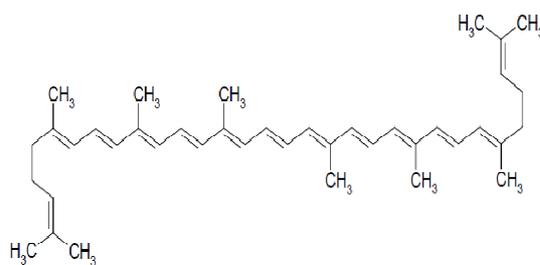
above the critical temperature regardless of the pressure applied, but may reach a density close to the liquid state. A substance is considered to be a supercritical fluid when it is above its critical temperature and critical pressure. Several compounds have been examined as SFE solvents. For example, hydrocarbons such as hexane, pentane but the main supercritical solvent used is carbon dioxide[3].

Carbon dioxide(critical conditions = 31.4°C and 73.8 bar) is cheap, environmentally friendly and generally recognized as safe. Carbon dioxide is non-toxic, non-explosive, readily available and easily removed from the extracted products. Supercritical CO₂ (SC-CO₂) is also attractive because of its high diffusivity and its easily tunable solvent strength. Another advantage is that CO₂ is gaseous at room temperature and ordinary pressure, which makes analyte recovery very simple and provides solvent-free analytes. Also important for the sample preparation of food and natural products, is the ability of SFE using CO₂ to be operated at low temperatures using a non-oxidant medium, which allows the extraction of thermally labile or easily oxidized compounds[1].

Moreover, supercritical CO₂ has zero surface tension, which allows easy penetration into most matrices. In addition, in the supercritical state, supercritical CO₂ is extremely sensitive to small changes in temperature and pressure such that a compound may be extracted from a matrix at one set of conditions and then separated from supercritical CO₂ in a downstream operation under a slightly different set of conditions. Some of the other advantages of supercritical CO₂ that it is available in high purity at relatively low cost, it can be easily removed from the matrix after the process, and it can be easily separated from the extracted compounds. It is well documented that CO₂, a nonpolar solvent, is best suited for the extraction of nonpolar organic compounds. Solubility of substance which is to be extracted, is a strong function of supercritical CO₂ density while using supercritical fluid extraction[4].

Lycopene is a red pigment that occurs naturally in certain fruits, vegetables, algae and fungi. It is a well-known carotenoid. Tomato and tomato based products are major sources of natural lycopene in the human diet. Other significant source include watermelon, pink grapefruit, pink guava and apricots. The importance of this natural carotenoid as a colouring and antioxidant agent in the food industry increased in recent years. It can be used as nutraceutical, due to its high antioxidant activity, reducing the risk of atherosclerosis and coronary heart disease. Recent epidemiological studies revealed that the intake of tomatoes and blood lycopene level are inversely associated with the risk of some diseases like cancers including the prostate gland, stomach, and lung[5].

Lycopene is one of the best biological suppressants of free radicals, especially those derived from oxygen. It has the highest singlet oxygen-quenching rate of all carotenoids in biological systems^[5]. Lycopene is an acyclic, open chain, unsaturated carotenoid having 13 double bonds, of which 11 are conjugated, arranged in a linear array, and has a molecular formula of C₄₀H₅₆ with molecular weight of 537. The structural formula of all *trans*-lycopene is shown below[6].



Lycopene present in red tomato fruits typically contains 94-96% of all-*trans*-lycopene. However, It may undergo *trans*-to-*cis* isomerization during tomato processing. It is unstable when exposed to light, heat, and oxygen. Exposure to light and heat starts isomerization from the *trans* to *cis* configurations. The *cis*-isomers of lycopene have different physical and chemical characteristics than all-*trans*-lycopene. Some of these differences include lower melting points, lower specific absorption, and a shift in the absorption maximum. Lycopene can also undergo oxidation. when exposed to oxygen with the formation of many different oxidation products. To prevent isomerization and oxidation, lycopene is kept under inert gas in lightproof containers and stored in a cool place[7].

2. SFE of lycopene from tomatoes

2.1 Preextraction

It was observed during review studies that researches should be careful in specifying methods and process parameters during sample preparation before extraction process by considering the factors, which may affect occurrence of various chemical changes in its structure, in addition to the factors, which affect lycopene recovery. To our knowledge, all the studies dealing with supercritical fluid extraction of lycopene from tomato or tomato industrial waste only used fresh, or well-dried samples (moisture content of 2.3–10%). It has been shown that the

supercritical extraction of compounds from fresh tomatoes or fresh tomato industrial wastes resulted in a low extraction recovery of lycopene. Also, it has been reported that the lycopene content of dried samples was lower than that of fresh tomato waste samples[8].

Most popular method for drying purpose was freeze-dryer. However, other methods like drying with sunlight or at room temperature drying under vacuum, drying in oven and drying with warm air (air-drier) were used. After drying, grinding was occurred. The smallest size particle gave maximum yield of extracts. Particle size was generally reduced below 2 mm. The smallest size was found as 0.3 mm while biggest size was found as above 3 mm. The highest trans-lycopene recovery was obtained at particle size of 0.36 mm (93% recovery). It was reported in some studies that sieving operation was carried out after size reduction operation[9].

2.2 Extraction temperature, pressure and time

It was reported from some studies that optimum extraction temperature varies within a wide range like 50-110^oc. Optimum temperature was 60-70^oc for recovery >80%. Extraction was also done at 80^oc. It was highlighted at the end of many studies that an increase in extraction temperature increase lycopene yield. Different opinion take place about optimum extraction temperature. One study reported that at 110^oc, 96% total lycopene was extracted after 40 min, and full 100% recovery was achieved in 50 min[9].

Increase in extraction pressure increase efficiency in lycopene recovery. Optimum pressure parameter in lycopene recovery were generally reported between 300&400 bar. Especially, 400 bar was set out as optimum process parameter with different extraction temperature ranging between 40^oC&100^oC. If it was required, pressure more than 400 bar was also used. Optimum extraction time ranges between 0.5&8 hrs. This parameter is related to type of equipment & flow rate of CO₂. It was reported that at 60^oC & 300 bar pressure, particle size 0.36 mm, extraction time was 5 hrs and efficiency of process was 93%[10,11].

2.3 Isomerization, storage conditions & stability

Extraction temperature higher than 60^oc may cause lycopene isomerisation from trans to cis configuration which is unstable form. While this value was determined as 70^oc & 80^oc by some researchers. Generally, The temperature above 100 or 110^oc makes lycopene unstable[5].

The concentration of lycopene decreases as a function of time depending also on matrix. The decrease of lycopene concentration was faster with smaller particle size (after 30 days 0.6% lycopene left in powder & 47% left in crushed tomatoes). Light did not seem to have much effect on decrease lycopene. Sharma's & Lemagure's observation of lycopene loss in tomato pulp even when the pulp was stored in frozen state under vacuum suggests an autocatalytic reaction. It was found that nitrogen atmosphere to have relatively little effect on lycopene stability. The stabilizer like (+) α -tocopherol or rosemary extract was added to improve stability of lycopene stored in solvent with the internal standard included, the amount of lycopene left was 90% for sample with no addition. These results suggests that no stabilizer is needed when lycopene samples are stored under non-solvent conditions[10].

The dissolved samples were left on the vial tray of HPLC instrument and their color was visually followed. The sample with no addition of stabilizer lost its color during night, but redish color of samples with addition of stabilizer persisted for weeks[10].

CONCLUSION

In this study, The influence of optimum parameters in SFE for examining lycopene level of tomatoes are reviewed. Drying, grinding and sieving operations have to be completed before extraction. Furthermore, Drying of tomato materials in preparation stage caused a decrease in trans-lycopene content of extract. The materials with lower moisture content is more preferable than material with higher moisture content. The studies reporting that if optimum process parameters are employed, pure lycopene may be obtained without using a co-solvent. Essential critical parameters were reported as temperature, pressure, extraction time and flow rate of SC-CO₂ in lycopene recovery. Although the significant effect of SC-CO₂ flow rate on efficiency increase is not clear. Optimum temperature seems as 60-70^oc. While thermal degradation may be observed at 80^oc. The lycopene having largest antioxidant activity was produced at 40^oc as extraction temperature. Isomerization due to heat, oxidization and due to effect of oxygen and solubility of lycopene in SC-CO₂ are the significant parameter.

In brief, 100% yield of lycopene extraction of tomatoes should not be targeted due to degradation of the product. There are the affecting factors like matrix type, particle size, lycopene content, flow rate of SC-CO₂, temperature and pressure on lycopene yield.

REFERENCES

1. Lili Xu, Xiaori Zhan, Zhaowu Zeng, Rong Chen, Haifeng Li, Tian Xie and Shuling Wang, *African Journal of Pharmacy and Pharmacology*, **2011**,5(9), 1196-1211.
2. Milan N. Sovilj, *APTEFF*, **2010**,41,105-120.
3. Brunner G., *Gas extraction: An introduction of fundamentals of supercritical fluids and the application to separation techniques*, 1st edition, Steinkopff publisher, Michigan, **1994**.
4. Lu-E Shi, Zhi-Liang Zhang, Liang-Ying Xing, Dan-Dan Yang, Yu-Peng Guo, Xiao Feng Guo, Li-Ming Zhao and Zhen-Xing Tang, *Journal of Medicinal Plants Research*, **2011**, 5(3), 300-308.
5. Nevzat Konar, Iraz Haspolat, Ender Sinan Poyrazoğlu, Köksal Demir, Nevzat Artık, *Karaelmas Science and Engineering Journal*, **2012**, 2(1), 69-75.
6. Nguyen, M. L. and Schwartz, S. J., *Food Technology*, **1999**, 53, 38-45.
7. Schierle J., Bretzel W., Buhler I., Faccin N., Hess D., Steiner K. and Schuep W., *Food Chem.*, **1997**, 59, 459-465.
8. Vasapollo, G., Longo, L., Rescio, L., Ciurlia, L., *J. Supercrit. Fluid.*, **2004**, 29(1,2), 87-96.
9. Nobre, B P., Palavra, A F., Pessoa, F L P., Mendes, R L., *Food Chem.*, **2009**, 116(3), 680-685.
10. Ollanketo, Hartonen, Riekkol, Holm, Hiltunen, *Eur Food Res Technol*, **2001**, 212, 561-565.
11. Vagi, E., Simandi B., Vasarhelyine, K P., Daood, H., Kery, A., Doleschall, F., Nagy, B., *J. Supercrit. Fluid.*, **2007**, 40(2), 218-226.
12. Danielski L, *Extraction & fractionation of natural organic components from plant material with supercritical carbon dioxide*, **2007**, Hamburg-Harburg.
13. Zaidul, Norulaini, Mohd Omar, Smith R, *Journal of food Engi*, **2007**, 79, 1007-1014.