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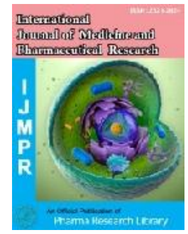

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Extraction and Application of Lycopene from Papaya

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A B S T R A C T

Lycopene a carotenoid in the same family as beta-carotene, is what gives tomatoes, pink grapefruit, apricots, red oranges, watermelon, and guava their red color. Lycopene is not merely a pigment, it is a powerful antioxidant that has been shown to neutralize free radicals, especially those derived from oxygen, thereby conferring protection against prostate cancer, breast cancer, atherosclerosis, and associated coronary artery disease. It reduces LDL (low-density lipoprotein) oxidation and helps reduce cholesterol levels in the blood. In addition, preliminary research suggests lycopene may reduce the risk of macular degenerative disease, serum lipid oxidation, and cancers of the lung, bladder, cervix, and skin. The chemical properties of lycopene responsible for these protective actions are well-documented. The present study is aimed at extracting Lycopene from Papaya and study its multidimensional applications.

Keywords: Lycopene, Fruits and Vegetables, Antimicrobial Property and Food Colorant.

A R T I C L E I N F O

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1. Introduction

Papaya (***Carica papaya* L.)** is a fruit crop widely grown in tropical and sub-tropical environments. There has recently been increased interest in the study of the genome of papaya due to its small genome size of 372 Mb and its short life cycle compared with many other tropical fruit tree crops (**Neelu Malviya, 2014).** The two major papaya fruit flesh colours, red and yellow, are controlled by a single genetic locus with yellow being dominant over red (**Simran**

Lilwani and Vrinda Nair, 2015). The fruit flesh colour of papaya is determined largely by the carotenoid content. Red-fleshed papaya fruit contain high levels of lycopene, whereas yellow-fleshed fruit contains minimal level (**Figure 1**).

Figure 1: Papaya

Lycopene belongs to the family of carotenoids. It has a structure that consists of a long chain of conjugated double bonds, with two open end rings (**M. Basuny *et. al.*, 2009**). The structure lycopene is the longest of all carotenoids. Lycopene ([C40H56], molecular weight 536.85) is an unsaturated hydrocarbon carotenoid containing 13 carbon-carbon double bonds, 11 of which are conjugated and arranged in a linear array (**Joseph Levy and Yoav Sharoni, 2004).** These conjugated double bonds are responsible for the vibrant red color of lycopene. Lycopene is a lipophilic compound that is insoluble in water, but soluble in organic solvents **(Sanjay Metkar *et. al.*, 2014)** (**Figure 2).**

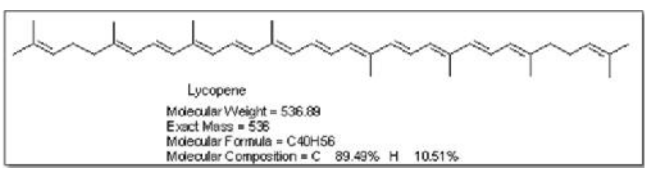

Figure 2: Structure of Lycopene

Tomatoes are widely known for their outstanding antioxidant content, including, of course, their often times-rich concentration of lycopene (**Naviglio D, *et. al.*, 2008**). Researchers have recently found an important connection between lycopene and its antioxidant and antimicrobial properties (**Suraj Ashok Bhagat, *et. al.*, 2012**). A study was designed in which tomato and other dietary sources of lycopene were removed from the diets of postmenopausal women for a period of 4 weeks, to see what effect lycopene restriction would have on bone health (**Zhao Yapinga, 2002)**. At the end of 4 weeks, women in the study started to show increased signs of oxidative stress in their bones and unwanted changes in their bone tissue (**Wayne W. Fish *et. al.*, 2002).** The study investigators concluded that removal of lycopene-containing foods (including tomatoes) from the diet was likely to put women at increased risk of osteoporosis (**R. Bunghez, 2011).** They also argued for the importance of tomatoes and other lycopene-containing foods in the diet. We don't always think about antioxidant protection as being important for bone health, but it is, and tomato lycopene (and other tomato antioxidants) may have a special role to play in this area (**P.D. Fraser and P.M., Bramley, 2004).** While most often associated with lycopene (a carotenoid phyto nutrient widely recognized for its antioxidant properties), tomatoes provide a unique variety of phyto nutrients. Include an additional carotenoids (including beta-carotene, lutein and zeaxanthin); flavonoids (including naringenin, chalconaringenin, rutin, kaempferol, and quercetin); hydroxycinnamic acids (including caffeic, ferulic, and coumaric acid); glycosides (including esculeoside A); and fatty acid derivatives (including 9-oxo octadecadienoic acid) (**Wee Sim Choo and Wai Yen Sin 2012).**

The FDA has approved generally Recognized as Safe (GRAS) status to lycopene. Recently the FDA has also given a limited health claim declaration for lycopene, stating "very limited and preliminary scientific research suggests that eating one cup of tomatoes and/or tomato sauce a week may reduce the risk of prostate Cancer". Consuming cooked tomato sauces, tomato ketchup, tomato soup, stewed tomatoes and other cooked tomato dishes are other excellent sources of lycopene besides papaya (**Sanjay Metkar *et. al.*, 2014**).

The present study is aimed at extracting lycopene from papaya and identifying its multidimensional applications.

2. Materials and Methods

Extraction of Lycopene by Acetone-Petroleum Ether Method

Materials Required

1. Red-fleshed papaya
2. Acetone
3. Petroleum Ether
4. Magnesium Sulphate
5. Whatman Filter Paper

100 gms of papaya paste was weighed and 125ml of acetone was mixed and was allowed to stand for 3-4 mins to remove water. The mixture was filtered by using Whatman Filter paper. The filtrate was collected and squeezed by using a filter paper to dehydrate the paste. Then 125 ml of petroleum ether and magnesium sulphate was added to the filtrate and the content was stirred well for 3-4mins. It was then filtered by using Whatman Filter paper. Finally the filtered lycopene extract was collected and allowed to evaporate. Evaporated sample was used by adding petroleum ether: acetone in the ratio (9:1).

Antimicrobial Activity

Materials Required

1. Bacterial culture ***(Bacillus, Staphylococcus, and Pseudomonas)***
2. Mueller Hinton Agar (**Hi-media)**
3. Nutrient broth
   a. Peptone - 0.05gm
   b. Sodium chloride - 0.05gm
   c. Beef extract - 0.015gm
   d. Yeast extract - 0.015gm
   e. Distilled water - 10ml
4. Crock Borer
5. Sterile swab

Bacterial inoculums were prepared by transferring a loopful of bacterial culture from fresh culture plates to tubes containing 10 ml of Nutrient Broth and incubated for 24 hours at 37°C. The tubes were shaken occasionally to aerate and promote growth. The two gram positive strains ***Staphylococcus spp***. and ***Bacillus spp***. and one gram negative strain ***Pseudomonas spp.*** were used for the study.

Agar Well Diffusion Method

The antibacterial activity of lycopene extract obtained with Petroleum ether: Acetone (9:1) was evaluated by the agar well diffusion method. The petriplates were prepared with 20 ml of sterile Mueller Hinton Agar (**MHA), (Hi-media**) and the strains *(**Bacillus, Staphylococcus**, and **Pseudomonas**)* that had been incubated for 24 hrs were used for the assay. A sterile cotton swab was dipped into the bacterial suspension and then evenly streaked over the entire surface of a sterile Mueller Hinton Agar plate to obtain uniform inoculums. The wells were punched on the seeded plates using a sterile borer (7 mm), and the plates were allowed to dry for 5 mins. The solvent extraction Petroleum Ether: Acetone (9:1) extracts were dispensed into the well using a sterile micropipette. The plates were incubated overnight for bacteria at 37°C. The antibacterial activity was determined by measuring the diameter of zone of inhibition (mm).

Food Colourant

Materials Required

1. Sugar cubes
2. Standard orange colour (caramoisine)
3. Lycopene extract
4. Aluminium foil paper
5. Dropper

The sugar cubes were made by using appropriate concentration of sugar and water. By using distilled water, the standard colour and lycopene samples were diluted according to standard concentrations that are usually used in foods. By using spreader, both sample standard and lycopene was spreaded on the sugar cubes. Then both sugar cubes were packaged by using aluminium foil and were put in polythene bags. Both were stored at room temperature. The colour efficiency was observed and the effect of light on lycopene colour sample was measured against the standard colour.

3. Results and Discussions

Extraction of Lycopene by Acetone-Petroleum Ether Method: A simple liquid-liquid extraction method was employed to extract lycopene in minimum organic solvent. The yield of lycopene from papaya is extracted by acetone-petroleum ether method (**Figure 3).**

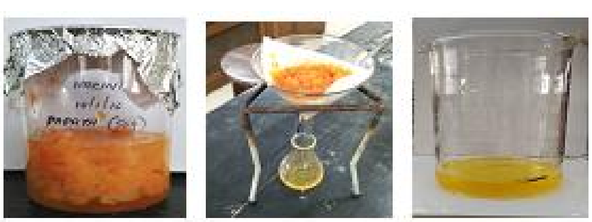

Figure 3: Extraction - Acetone-Petroleum ether method

Antimicrobial Activity

The agar well diffusion method was used to evaluate the antibacterial activity by measuring the zone of inhibition against the test microorganisms. The Petroleum Ether: Acetone (9:1) extract exhibited the prominent antibacterial activity against, *Pseudomonas spp.* and *Staphylococcus spp.* But no zone of inhibition was found against *Bacillus spp* **(Table 1 & Figure 4).**

Table 1: Zone of Inhibition

S.No	Test of organism	Zone of inhibition
1	*Staphylococcus* spp.	1.5mm
2	*Pseudomonas* spp.	1mm
3	*Bacillus* spp.	0mm

Figure 4: Anti- bacterial activity against (A) *Staphylococcus* spp. (B) *Bacillus* spp (C) Pseudomonas spp.

Food Colourant

Extracted Lycopene and standard orange colour was made to spread on sugar cubes. After 24hrs, the results were observed **(Figure 5)**.

Figure 5: Lycopene colour spreaded on sugar cubes

4. Conclusion

Lycopene sample was extracted by acetone – petroleum ether method. The antibacterial activity of lycopene was studied against *Bacillus spp. Staphylococcus spp. and Pseudomonas spp*. Due to its deep red color, lycopene can be used as a promising food colorant. Further work can be carried out to investigate anticancer activity in various cell lines. A modern life style keeps people away from healthy diet. For healthy dietary habits one should increase the consumption of food products which are helpful in the prevention of illness. Fruits and vegetables are main source of natural antioxidant and antimicrobial components. Antioxidants and Antimicrobial virtues give protection against harmful free radicals and reduce rate of cancer and heart disease and the most efficient carotenoid antioxidant is Lycopene. Thus the present study unravels two major applications of Lycopene extracted from Papaya and many more such applications by future research are yet to be unraveled.

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