PROJECT REPORT ON

ISOLATION AND CHARACTERIZATION OF LYCOPENE PIGMENT FROM TOMATO

SUBMITTED BY,

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SUBMITTED TO,

VIVEKANAND COLLEGE, KOLHAPUR (AUTONOMOUS)

DEPARTMENT OF BIOTECNOLOGY.

FOR PARTIAL FULFILMENT OF BACHELOR OF

SCIENCE IN BIOTECHNOLOGY

THE YEAR

2021-2022

UNDER THE GUIDANCE OF

Miss. Srushti Sarnaik

Assistant Professor, Department Biotechnology



"EDUCATION FOR KNOWLEDGE SCIENCE AND CULTURE"



- DR BAPUJI SALUNKHE

SHRI SWAMI VIVEKANAND SHIKSHAN SANSTHA'S

VIVEKANAND COLLEGE, KOLHAPUR, (AUTONOMOUS)

Department of Biotechnology

Certificate

This	is	to	certify	that	Pathan	Alim
Alatif						

Exam Number 9325 has satisfactorily carried out his project report as per the syllabus prescribed by Bo'S Department of Biotechnology, Vivekanand College (Autonomous) for B.Sc- III Biotechnology (Entire). This project report represents his bonafied work during academic year 2021-2022.

Place: Kolhapur

Date: 201722.

amair Project Guide



Examiner

Head Department of Biotechnology (Entire) Vivekanand College, Kolhapur (Autonomous)

DECLARATION

I hereby declare that the project work entitled "Isolation & Charaterization of Lycopene Pigment From Tomato" submitted to the Vivekanand College. Kolhapur for the award of the degree of "Bachelor of Science in Biotechnolgy is the result of bonafied work carried out by me under the guidance of Asst/Prof Miss. Srushti Sarnaik.

1 further declare that the results presented here have not been the basis for the reward of any other degree

Place:- Kolhapur

Date: 26/05/2022

A. A. Pothon. Mr. Alim Alatif Pathan. Acknowledgment

This project work is a successful outcome of the contribution and guidance of other person which express my deep gratitude.

I also express our thanks to Prof. Mr. S.G. Kulkarni Head of Department of Biotechnolgy, Vivekanand College, Kolhapur for availing me with the laboratory facilities to the Biotechnology Department to carry experiment work.

I also express our gratitude towards Prof. Miss. Srushti Sarnaik my project guide for his guidance and who gave me encouragement and support throughout the course of study so that could complete my project work.

I also wish to express my gratitude to the laboratory staff for completing the project work. Lastly, express my gratitude to my parents, all our friends and classmates for their support and co operation. I am also grateful to all those who have directly or indirectly supported me in completion of this work.

A.A.Pothun. Mr. Alim Alatif Pathan

INTRODUCTION:-

Fruits and vegetables are main source of natural antioxidant components. Antioxidants give protection against harmful free radicals and reduce rate of cancer and heart disease. The most efficient carotenoid antioxidant is lycopene. Lycopene is a natural pigment which protects the body by neutralizing the negative effects of oxidants. In the synthesis of vitamin A lycopene plays an important role as an intermediate and carotenoid like ß-carotene and ß cryptoxent in, influences its development. Lycopene is soluble in fat and synthesized by plants and microorganisms. Regular intake of lycopene containing food reduces the risk of body tumor especially prostate cancer, studies have shown that the antioxidants vitamin E, selenium, and lycopene all reduces risk of prostate cancer. Therefore, it would say that lycopene is very important for cancer prevention; it also reduces LDL cholesterol and cardiovascular diseases. It is a carotenoid and gives red colour to vegetables and fruits. Lycopene in processed foods is mainly in the form of the isomers. Its molecular formula is C40 H56 and 536.88 is its molecular weight. Lycopene is highly unsaturated hydrocarbon with 13 double bonds, it has been reported that 11 unsaturated bonds are conjugated. Conjugated bonds of lycopene molecule gives ability to act as an antioxidant and make it more efficient for the use of human health. Natural food sources of lycopene are tomatoes, watermelon, pink guava, pink grapes, papaya and apricots Lycopene has been extracted from the several different fruits and berries. It was first isolated from Tamuscommunis by Harsten in 1873. When consumption of lycopene from different products up to 150 mg daily shows no side effects Recent studies have shown that ingested lycopene is metabolized in the body. Several metabolites have now been identified and characterized.

AIM AND OBJECTIVES

■AIM :- Isolation of carotenoid pigment lycopene from tomato and its biochemical characterization

.OBJECTIVES :-

- To isolate carotenoid pigment from tomato
- Characterization with help of Thin Layer Chromatography
- FTIR Analysis of purified sample

MATERIALS AND METHODS

.MATERIALS :-

Tomato paste, Distilled water.

Glasswares:- Separating funnel ,funnel ,glass rod, measuring cylinder ,flasks, test tubes.

Chemicals :- Acetone ,Petroleum ether,NaCl, aqu.Pottasium Carbonate.

Equipments:- Test tube stand, Compound microscope, micropipette, Aluminium foil.

.METHODS:-

Tomato paste:- Cut the tomatoes and make paste of it .Wight about 50 gm of paste into the beaker

1]Acetone-petroleum ether extraction method-

Sample (1.0-1.5 g powder) was extracted with $10 \, \text{mL}$ acetone-petroleum ether (50% v/v). The upper lycopene-containing organic layer was removed by means of a pipette and collected in test tube. Extraction was repeated. The extracts were combined, washed with $15 \, \text{mL}$ saturated aqueous sodium chloride (NaCl) and removed the aqueous wash with a micropipette. The extract was washed with $10 \, \text{mL}$ of $10 \, \text{m}$ aqueous potassium carbonate (K2CO3) and removed the aqueous wash. The lycopene-containing organic layer was dried with a drying agent (calcium chloride). The excess solvent was allowed to evaporate at room temperature for a few minutes in the dark. The tubes containing lycopene extracts were covered with aluminium foil and stored in freezer until further analysis.

2] After isolation of lycopene pigment it was further processed for its characterization with the help of thin layer chromatography. Silica gel plate was prepared ,the mobile phase(solvent system) used Toluene&nHexane in ratio 1:9 respectively. Solvent system kept in closed conditionin for about 15-20 minutes in beaker. Purified sample was loaded on silica gel plate with help of capillary on marked line and dried in oven for 10 minutes.

Further plate was kept in solvent system for the development. Sample was eluted with solvent. After reaching solvent upto marked position plate was taken out of the solvent system.

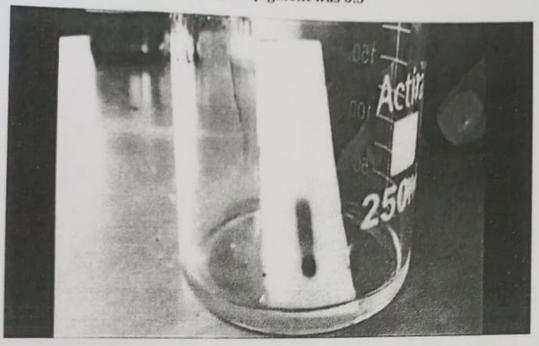
Calculation:-

Rf= Distance travelled by solute

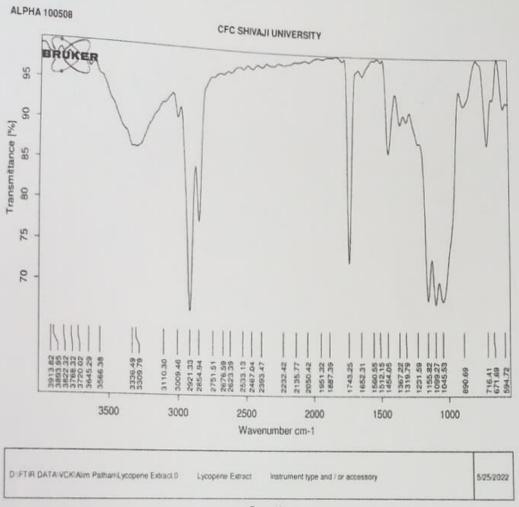
Distance travelled by solvent

i.e,
$$= 1.95 = 0.3$$

1] Reported Rf value for lycopene pigment was 0.3



2] FTIR analysis OF Sample was performed



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CONCLUSION:-

Thus we conclude that, the isolation of carotenoid pigment from tomato and its biochemical characterization was performed with the help of Thin Layer Chromatography .

Lycopene in the sample was analysed with the help Fourier transform infrared spectroscopy(FTIR). Presence of acyclic groups ,alkyl groups confirms presence of carotenoid pigment in sample.