Microbiological and Functioning Quality of Organically Grown Fresh Vegetables

A DISSERTATION SUBMITTED FOR PARTIAL FULFILMENT OF

REQUIRNMENT FOR THE AWARD OF DEGREE OF

MASTER OF SCIENCE IN MICROBIOLOGY

BY

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CANDIDATE'S DECLARATION

I hereby declare that the work which is being presented in the dissertation entitled 'Microbiological And Functioning Quality Of Organically Grown Fresh Vegetables' in partial fulfillment of the requirements for the award of the degree of MASTERS OF SCIENCE IN MICROBIOLOGY, Department of Biotechnology, Thapar University, Patiala is an authentic record to my own work during a period of six month from January 2014 To June 2014,under the supervision of Dr. Abhijit Ganguli, Department of Biotechnology, Thapar University.

Harjinder Singh
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Place - Patiala

CERTIFICATE

This is to certify that the thesis entitled **"MICROBIOLOGICAL AND FUNCTIONING QUALITY OF ORGANICALLY GROWN FRESH VEGETABLES"** being submitted by Harjinder Singh, Registration number 301205002 in partial fulfillment of the requirements for the award of degree of Master of Science in Microbiology, Department of Biotechnology, Thapar University, Patiala, is a bonafide work carried out under my supervision and guidance. The thesis has not been submitted for award of any other degree or certificate in this or any other university

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Contents

1. Abstract

Based on health beneficial properties the consumer awareness of organically grown fresh vegetables has assumed significant importance across the globe. In this study we attempted to compare selected organic and conventional fresh vegetables in terms of nutritional value and microbiological safety. The results indicate that organically produced carrot, fenugreek, cabbage, coraínder contained higher levels of vitamin C, riboflavin, beta carotene, lutein than the inorganically grown counterparts, though significant enhancements were not observed except in case of lutein in fenugreek. Microbiological analysis indicated lack of high total aerobic counts and pathogens such as *Salmonella*, *Shigella* sp and *Staphylococcus aureus* could not be detected in the organically grown vegetables.

2. Introduction

Organic farming is form of agriculture that relies on techniques such as crop rotation, green manure, compost and biological pest control. Organic farming is a system which avoids use of synthetic inputs and to maximum extent feasible relies upon crop rotations, crop residues, animal manures, off farm organic waste, mineral grade rock additives and biological system of nutrients. The term "organically grown food' denotes products that have been produced in accordance with the principle and practices of organic agriculture. Organic agriculture and food processing practices are wide ranging and overall seek to foster the development of a food production system that is socially, ecologically, and economically sustainable. The key principle and practices of organic food production aim to encourage and enhance biological cycles within the farming system to maintain and increase long term fertility of soils, to minimize all forms of pollution, to avoid the use of synthetic fertilizers and pesticides, to maintain genetic diversity of the pollution system, to consider the wider social and ecological impact of the food production and processing system, and to produce food of high quality in sufficient quantity[Willer et al. 2013].

Organically grown vegetables

Organic foods provide a variety of benefits. Some studies show that organic foods have more beneficial nutrients, such as antioxidants, than their conventionally grown counterparts. In addition, people with allergies to foods, chemicals, or preservatives often find their symptoms lessen or go away when they eat only organic foods (Dye 1996).

Organic produce contains fewer pesticides. Pesticides are chemicals such as fungicides, herbicides, and insecticides. These chemicals are used in conventional agriculture.
Fenugreek rich in vitamins such as thiamin, folic acid, riboflavin, niacin, vitamins A, B6, and C, and are a store house of minerals such as copper, potassium, calcium, iron, selenium, zinc, manganese, and magnesium. Fenugreek leaves are a rich source of vitamin k as well.

Carrots are an excellent source of vitamin A, providing 210% of the average adult's needs for the day. They also provide 6% of vitamin C needs, 2% of calcium needs and 2% of iron needs per serving. It is the antioxidant beta carotene that gives carrots their bright orange color. Beta carotene is absorbed in the intestine and converted into vitamin A during digestion.

Tomatoes are a good source of calcium and iron. They also contain decent amounts of phosphorus, sulfur, and potassium. 100 grams of tomatoes contain only 20 calories. These calories are also easy to absorb by the body. The low calorie content of tomato makes it a favorite for healthy diet. Eating tomatoes is good recommended in weight loss programs as it provides various essential minerals and vitamins.

Coriander is also rich in many vital vitamins, including folic acid, riboflavin, niacin, vitamin A, beta carotene, vitamin C that is essential for optimum health.

Cabbage is one of the most popular vegetables in kitchen gardens, coming in color of red or green. Cabbage also contains antioxidant and fiber. Fresh organic vegetables contain substantial amounts of carbohydrates, vitamin, fibers and minerals. They can protect the body from such major diseases as cancer and heart diseases. Some are high in substance called anti-oxidants, such as beta-carotene, vitamin C and E and Selenium, which are nutrients that protect cell membranes from the damage of free radicals. Some are rich in other anti-cancer compounds, such as indoles. \

Comparisons of organic and inorganic production system

A wide range of factors has been investigated in studies comparing organic and conventional food production systems, including economics, crop yields, agronomic factors (soil chemical properties, soil physical properties, soil microbiological activity, pest and disease burdens etc), farm management practices, product quality (nutritional value, taste, shelf life), environmental impacts, biodiversity, farm nutrient inputs and social, trade, and political issues associated with food production (Worthington 2001). Food quality and what is meant by quality in the context of organic food production systems is one area that has received much attention in the debate on differences between organically and conventionally produced foods. Hence, a number of areas have been identified as important to consider, functional properties, biological factors, nutritional value, sensory characteristics, ethical issues, environmental issues, and social issues in produced food is going to be an important factor in ensuring a sustainable food supply and that environmental issues in food production and food 'quality' should not be the only factors to consider, in fact a number of studies have identified the reasons behind this considerable increase in consumer demand for organic foods, although the relative importance of factors influencing the purchase of organic foods may vary from country to country. For example, consumers in Germany have tended to be more concerned with environmental issues than those in the UK, although this may be changing with more recent reports suggesting that approximately 70% of organic consumers cite health as the primary reason for purchasing organic food. In the USA, consumers who considered organic foods to be better than conventional foods believed that the following characteristics were important when they purchased organic foods: safety, freshness general health benefits, nutritional value, effect on environment, flavor, and general product. A group of Norwegian organic consumers identified health and environmental reasons for purchasing organic foods.

Functional properties of organic vegetables

The antioxidant capacities were higher for most organic vegetables as compared to conventional vegetables. Regular intake of organic vegetables reduces the risk of disorders including cancers. Green and fresh organic vegetables are a rich source of antioxidants that help to clear the free radicals that are associated with cell damage. The daily diet of organic food reduces the incidence of disorders of the heart, nervous system and also cancer (Hemmelgarn 1999).

Carrot is major source of beta-carotene. The beta-carotene is converted into vitamin A in our body and act as antioxidant. Vitamin A prevents damage of the white blood cells that are helped to protect infection. The high level of the beta carotene acts an antioxidant to the cell damage done to body through the regular metabolism. It helps to slow down the aging of cell.

The tomato contains high level of lycopene, an effective antioxidant. Regular intake of lycopene protects the heart from heart failure and function normally. It is also beneficiary to the blood vessels around the heart and demonstrates protection to the vessels around the neck. The 10 mg of lycopene daily can be used as supplement for health maintenance.

The lutein a carotenoid plant pigment found in green leafy vegetables like fenugreek leaves. Lutein is absorbed best when it is taken with a high fat meal. Antioxidants have strongly linked to protection from heart diseases to cancer, eye diseases to regulation of the immune system. Many multivitamins contains Lutein. They usually produce small amount of the 0.25mg per tablet.

The coriander and cabbage is good sources of vitamin C, which is necessary to make and maintain collagen, the connective tissue that holds the body and organs in place. Vitamin C helps heal cuts and wounds and resist infection. Vitamin C maintain the immunity of human body to long time.

The coriander and fenugreek are the good sources of riboflavin. Riboflavin is used for preventing the low level of the riboflavin deficiency. It is also used for boosting immune system function, maintain healthy hair, skin, mucous membrane and slowing aging. Few works in India have actually analyzed the nutritional parameters of beneficial fresh vegetables. In this study selected vegetables with nutritional properties were grown organically and compared in terms of specific nutritional attributes with inorganic or synthetic counterparts. Their microbiological quality was also examined.

3. Review of Literature

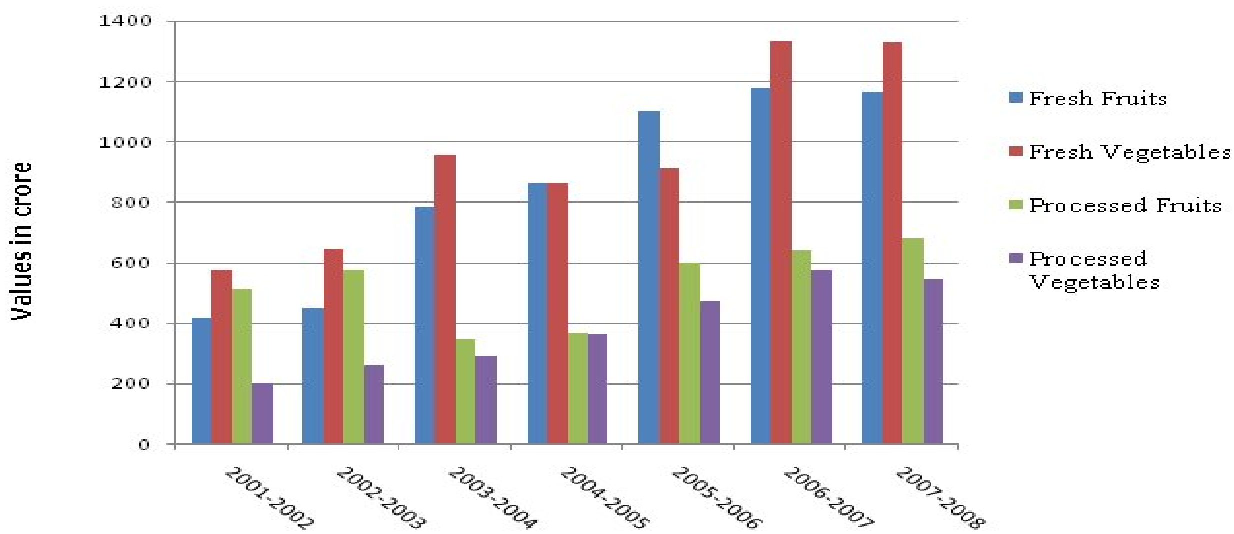
Vegetables crops have been well advocated in solving the problem of food security. They are rich source of minerals, vitamins, fiber and contain a low amount of proteins and carbohydrates. In addition to local market demand vegetables have the potential for both domestic and export market. The vegetable production of our country before independence was merely 15 million tones and now it is about 88.6 million tons during 2001-02, accounting 11.4% share of world vegetable production (Rai and Pandey et.al. 2005). Although India is the second largest producer of vegetables next only to china in world, the productivity of different vegetables in our country is comparatively lower than the world's average productivity. Again the per capita availability of vegetables is still behind the recommended quality. We demand by 2020 will be around 250 million tonnes. Thus due to the rapid growth of the population with reduction in land, in order to feed the population, the only solution is the vertical expansion or by increasing the productivity per unit area per unit time as the potential available land and water resources. A strategy is to used to be produce more vegetables from less land, less water with less pesticide and with less detrimental to soil and environmental as well. Organic vegetables fetch a premium price of 10-15% over conventional products. Market of organic products is growing at faster rate (20%) as compared to conventional ones (5%). The growth rate is highest in japan, USA, Australia and EU. Export preference of organic vegetables offers a great scope to a country like India, which has inoculated the skill of growing organically since time immemorial.

India is an exporting country and does not import any organic products. The main market for the export product is the European Union. Another growing market is USA which is equivalent to about 100kg NPK/ha/yr.

Market overview: Fresh vegetables

Consumption and consumer demand of organic fresh produce vegetables rose by more than 13% to $9.7 billion in 2012, according to the organic trade association survey. Total organic food sale rose 10.2% in 2012, topping more than $29 billion, according to the 2013 organic industry survey. That double digit organic growth compares favorably to conventional food sale, the repot said rose 3.7% in 2012 (F. Zang et al. 2006). Organic food account for the 4.35 of the total food sale, according to the survey, but the share of market is bigger for organic fruits and vegetables. The 2013 survey reported that organic food and vegetables account for just over 10% of consumer fruit and vegetable sale compare with 6% in the 2006 and 2% in 1998. The survey said that organic food produce account for the 90.8% of all organic fruits and vegetables sales. Organic food and vegetables are a substantial and rising component of the total organic food sales, according to the OTA survey. Organic food and vegetables sales account for the 34.8% of all organic food sales in 2012, up from 34% in 2011 and 33.2% in 2010. In general, the survey found that growth in organic food sale in 2012 reflects an improving economy.

Fig:1 The yearly growth of the fresh vegetables corresponding to their market value



In 2002 organic fresh vegetables sales account for the 4.5% of the total sales of total fresh vegetable sales (in fig 1). Natural food merchandisers reported that sales of packaged fresh produce had the highest growth rate among the sales of the organic product during the 2002-2003, expanding to 26% to $364 million. The conventional supermarkets account for the fourth part of this total. The number of new organic produce items introduced in the retail market has more than the doubled than the over a decade, from 14 in 1993 to 30 in 2003. In addition, organic produce has the largest market value among all organic products. Produce accounted for the 42% of US organic fruit sale in 2000, according the market research firm packaged fact. Even through the implementation of the organic standard and increasing the public awareness of organic food is helpful in promoting organic fresh produce sale, more affordable prices are important for the long term growth of the organic product market. This data rarely indicate the consumer preference of organically grown fresh vegetables.

Organic cultivation of vegetables

The process of growing vegetables was initiated in the month of December. First a land of area size 25×15 square meter was selected. This was cleaned and makes it to use for the purpose of organic farming. Seeds were kindly provided by Technocare Nursery and Garden centre, Punjab Agriculture University Ludhiana. Green manure and compositing to the soil after mulching was added and then add water to make the proper composition and mix the nutrient with soil. After that, the seeds were plowed in the field in the particular rows. We used the direct seeding method for plough seeds. Neem leaves was used as a biopesticide on the area to preserves the seeds from the diseases. Rock Phosphate also spread on the area of farming in three steps, in the starting, mid period of vegetable growth and at the end time. Watering was done after every 15 days in the periodic manner to the plants (depending upon the requirements of vegetables). The whole field of farming was covered with the paddy waste to save the vegetables from the disease due to fog in the month of December and January. Field was inspected after every 5 days to check the growth and other effects on the vegetables. All types of weeds were removed so the young vegetables plants get full nutrients. We analyzed the sample after one and half month to check that the vegetables growth is according to the requirements or not (Harwood, 1980). Watering the plants was stopped before 15 days of harvesting.

Chemicals, vegetable samples and Media

All chemicals, reagents were of highest analytical grade and obtained from SRL (Mumbai). Bacteriological media were from HiMedia(Mumbai) and standards from Sigma (USA).
Good quality Inorganic vegetable samples have been collected from local market and local contact farms resorting to synthetic farming practices.
The following key nutritional parameters are examined in inorganic and organic vegetables:

I. Determination of beta carotene.
II. Determination of vitamin C.
III. Determination of Riboflavin
IV. Analysis of lycopene in tomatoes
V. Analysis of lutein.
VI. Microbiological analysis of organically grown and inorganic vegetables.

1. **Determination of beta carotene**

The quantity of beta carotene in fresh organic vegetables was determined spectrophotometrically as described by (Ranganna et al. 2000). Briefly, 5gm fresh sample was crushed in 10-15ml acetone after adding a few a crystals of anhydrous sodium sulphate using pestle mortar, 10-15 ml petroleum ether was added and mixed thoroughly. Of the two layers which separate out on standing, the lower layer was discarded and upper layer collected in a 100ml volumetric flask. The volume was made up to 100ml with petroleum ether and optical density recorded at 452nm using petroleum ether as blank.

Calculations:

Beta carotene (ug/100g)= O.D.$\times 13.9\times 10^4 \times 100$/wt. of sample$\times 560\times 100$

2. **Determination of vitamin C**

Vitamin C is an essential nutrient for a large number of higher primate species and nearly all bats and a small number of other species. Standard solution of ascorbic acid of different concentrations was prepared (Tillman et al. 1932). The 10 ml of different standard solutions of ascorbic acid were prepared. 10 drops of 0.1% starch solution was added. The iodine solution added drop wise until a permanent color appears.

3. **Analysis of Lutein**

Free radical scavenging activity of lutein isolated from fenugreek leaves given by (Gowda et al. 2010). Extraction of carotenoids was carried out by taking 10g of the each fresh samples with 100 ml of the 5% ethanolic KOH. The mixture ground well for 5 minutes using pestle and mortar at room temperature in dim light. The extraction was repeated till the resultant extract was colorless. Total volume of the ethanol extract was 500ml and which was concentrate to 50ml. hexane was added to the ethanol extract in the ratio of 1:2 in a separating funnel, shaken well for 5 minutes and kept in the dark for 15 minutes. Two phase were separated funnel, shaken well for 5 minutes and kept in the dark for 15 minutes. Two phases were separated and the solvent partitioning was repeated till the hexane extract was colorless. All the hexane phases were pooled and flash evaporated at 30 ^{0}C and re-dissolved in known volume of hexane.

4. **Determination of riboflavin**

Standard solution of riboflavin of different concentrations was prepared (Hashmi et al. 1969). Take 100ml of different standard solutions of riboflavin and mix NaOH in test tubes. The preparation of stock by 2ml riboflavin added 18ml NaOH. Then take every test tube to 0.8 ml of solution and added 2 ml of 0.1M potassium phosphate buffer. Resultant solution was vortexed for 2 minutes and absorbance was measured at 444 nm.

5. **Analysis of lycopene**

Extraction method was performed according to given by (Lidia et al. 2008) Approximately 0.3 to 0.6g sample were weighed and 5ml of 0.05% BHT in acetone, 5ml of ethanol and 10ml of hexane were added. The recipient was introduced in ice and stirred on a magnetic stirring plate for 15 min. after shaking for 5 min. on ice. Sample was then left at room temperature for 5 min to follow the separation of both phases. The absorbance of the hexane layer was measured in a 1cm path length quartz cuvette at 503nm blanked with hexane.

6. **Microbiological analysis**

The determination of total aerobic bacteria was done by weighing a sample of 250g, then surface rinsing the vegetables in sterile peptone water (750ml) and subsequently performed graded dilution. A total of 0.1 ml solution of suspension, grown on TSA media for Petri dishes containing nutrient and stored at room temperature for 24-28 hours. For pathogenic analysis, dilutions were plated onto XLD agar for Salmonella, Baird Parker agar for *S.aureus* and SS agar for Shigella which were incubated for 24-48 hours at 37^0 C(US FDA, 2013).

5. Result and Discussion

1. Cabbage

Table: 1 Concentration of vitamin c in inorganic and organic cabbage in units of µg/gm corresponding to number of days.

Days	Inorganic conc.(µg/gm) (Result Value ±S.D)	Organic conc.(µg/gm) (Result value ±S.D)
10	5.67±0.0164	6.012±0.032
20	12.143±0.0153	13.143±0.0122
30	19.482±0.0242	21.001±0.0362
40	25.915±0.0125	27.652±0.0202
50	30.639±0.0386	33.973±0.0282
60	35.180±0.0254	39.898±0.0238
70	37.568±0.0259	43.658±0.0144
80	36.786±0.00147	42.477±0.0141

Table 1 shows that concentration of vitamin C in organic cabbage is greater than that of inorganic cabbage over 80 days.

2. Coriander

Table: 2 the concentration of vitamin C in inorganic and organic coriander in units of µg/gm corresponding to number of days.

Days	Inorganic conc.(µg/gm) (Result Value ±S.D)	Organic conc.(µg/gm) (Result Value ±S.D)
10	5.953±0.0516	6.738±0.057
20	11.728±0.012	13.961±0.072
30	17.162±0.077	19.731±0.0108
40	24.541±0.016	26.486±0.068
50	29.052±0.700	32.651±0.071
60	35.914±0.048	39.683±0.0214
70	36.688±0.082	40.678±0.053
80	35.6486±0.011	38.659±0.0174

Table 2 shows that concentration of vitamin C in organic coriander is greater than that of inorganic Coriander in 80 days.

3. Fenugreek

Table: 3 the concentration of vitamin C in inorganic and organic fenugreek (µg/gm) corresponding to number of days.

Days	Inorganic conc.(µg/gm) Result Value ±S.D)	Organic conc(µg/gm) Result Value ±S.D)
10	6.947±0.124	7.483±0.140
20	12.482±0.155	13.446±0.016
30	18.827±0.130	20.368±0.140
40	24.171±0.189	26.974±0.169
50	29.491±0.104	31.928±0.0507
60	34.587±0.087	35.180±0.083
70	35.655±0.247	37.987±0.196

Table 1 shows that concentration of vitamin C in organic fenugreek is greater than that of inorganic fenugreek in 70 days.

4. Carrot

Table: 4 the concentration of vitamin c in inorganic and organic carrot (µg/gm) corresponding to number of days.

Days	Inorganic conc.(µg/gm)	Organic conc(µg/gm)
10	4.234±0.0245	4.983±0.143
20	7.922±0.0546	8.498±0.0325
30	11.345±0.00659	15.294±0.218
40	16.454±0.118	21.986±0.249
50	21.543±0.0131	27.452±0.168
60	26.171±0.0096	33.434±0.050
70	30.642±0.0320	39.234±0.253
80	34.986±0.265	44.189±0.278

Table 4 shows that the highest concentration of vitamin C in organic carrot in 80 days is larger than that of inorganic carrot.

5 Tomato

Table: 5 the concentration of vitamin c in inorganic and organic tomato in units of µg/gm corresponding to number of days.

Days	Inorganic conc.(µg/gm)	Organic conc(µg/gm)
10	9.734±0.020	12.342±0.146
20	17.876±0.074	21.345±0.191
30	25.876±0.029	31.425±0.182
40	33.687±0.081	43.325±0.176
50	40.678±0.207	52.234±0.0311
60	47.879±0.159	64.345±0.029
70	55.422±0.030	71.345±0.193
80	63.324±0.129	80.234±0.252
90	70.207±0.207	89.234±0.201
100	78.243±0.240	98.868±0.276

Table 5 shows that concentration of vitamin C in organic tomato is much greater than that of inorganic tomato in 80 days.

Determination of Riboflavin

1 Coriander

Table: 6 concentration of riboflavin in organic coriander and inorganic coriander in units of µg/gm corresponding to number of days.

Days	Inrganic conc(µg/g) (Result +S.D.)	Organic conc(µg/g) (Result+S.D.)
10	0.376±0.0007	0.469±0.045
20	0.601±0.009	0.876±0.031
30	0.923±0.0007	1.436±0.019
40	1.78±0.028	1.543±0.016
50	1.781±0.021	1.97±0.007
60	1.789±0.003	1.979±0.005
70	1.79±0.021	1.98±0.016
80	1.801±0.011	1.99±0.014

Table 6 shows that concentration of riboflavin in organic coriander is greater than that of inorganic coriander in 80 days.

2 Fenugreek

Table: 6 concentration of riboflavin in organic fenugreek and inorganic fenugreek in units of µg/gm corresponding to number of days.

Days	Inorganic conc(µg/g) (Result+S.D.)	Organic conc(µg/g) (Result+S.D.)
10	0.247±0.033	0.375±0.0007
20	0.584±0.0098	0.599±0.002
30	0.865±0.041	0.904±0.0021
40	1.148±0.0021	1.152±0.021
50	1.178±0.0014	1.199±0.14
60	1.2±0.14	1.229±0.0035
70	1.258±0.0041	1.273±0.0028
80	1.293±0.0021	1.302±00.0035

Table 7 shows that concentration of riboflavin in organic fenugreek is greater than that of inorganic fenugreek in 80 days. Samples in each case were obtained after indicated days of harvesting wherever possible and analyzed. Inorganic vegetables were collected from a local contact farmer resorting to syntheticfarming

Estimation of beta carotene

Table: 6 concentration of Beta-carotene in organic and inorganic vegetables in units of µg/gm

Organic Vegetables	Beta-carotene Conc.(gm)	Inorganic Vegetables	Beta-carotene(gm) Conc.
Carrot	0.021824	Carrot	0.0203
Coriander	0.01106	Coriander	0.0107
Tomato	0.004488	Tomato	0.004315

Table 6 shows that the concentration of beta-carotene in organic carrot is greater than organic coriander and tomato and also from inorganic carrot, coriander and tomato.

Analysis of Lycopene in tomatoes

Organic tomato =1.345
Lycopene content (mg/kg) = (1.345 – 0.0007) ×30.3/g tissue
= 40. 732mg/kg
Inorganic tomato= 1.050
Lycopene content (mg/kg) = (1.050– 0.0007)×30.3/g tissue
= 31.793mg/kg

The above results indicate that lycopene in organic tomato is greater than that of inorganic tomato concentration.

Estimation of Lutein in Fenugreek leaves

$$\text{Total carotenoids (\%)} = \frac{0.245 \times 10000}{10 \times 2550}$$

$$\text{Total carotenoids (\%)} = 0.0960$$
$$= 9.6\%$$

From above calculation we conclude that fenugreek leaves showed high content of lutein and prevents free radical formation more effectively than the standard, available synthetic antioxidants. and lutein is about 10% in organic fenugreek leaves; however in inorganic fenugreek it is around 6.5 %.

The method adopted for the isolation of lutein from fenugreek leaves is simple. The extraction with saponofication was performed which does not allowed the chlorophyll and other water soluble contaminants expects lutein with negligible amount of the beta-carotene. Carotenoids isolated from the fenugreek leaves was measured spectrophotometrically, the absorbance spectra of the fenugreek leaves extract indicated the presence of lutein and beta-carotene. The concentration of lutein was calculated on the basis of the molar absorption of lutein indicated notably high yield of lutein in organic fenugreek. Few work have actually emphasized on nutritional benefits of organically grown vegetables and fruits especially in lines of those components mentioned in this work. In fact a critical review of 15 journal articles articles indicate 14 articles focused specifically on 15 varieties of fruit, 3 articles focused specifically on 6 varieties of vegetables and 1 article focused on 2 varieties of fruit as well as 1 vegetable.Many of the articles focused on both antioxidants and polyphenols, while others mentioned either antioxidants or polyphenols.Twelve of the 17 fruits studied (71%) and 2 of the 7 vegetables studied(29%) reported higher levels of antioxidants, polyphenols, or both in the organically grown produce. Four of the 17 fruits studied (24%) and 5 of the 7 vegetables studied (71%) reported no differences in antioxidants or polyphenols between organically grown and conventionally grown produce. Only 1 of 17 fruits studied (6%) found higher levels of antioxidants and polyphenols in the conventionally grown produce. No support was found for higher levels of total antioxidants and polyphenols in conventionally grown vegetables.Organic phosphorous may play an important role in providing higher amounts of organic phosphorous unavailable from synthetic fertilizers and alleviating abiotic stresses and production of phytochemicals of nutritional value. Our result with fenugreek seems interesting and deserves further studies. However, extensive systematic studies are required before arriving at any definite conclusion on a case-by case basis.

Microbiological analysis

Table7: Total aerobic plate counts of organically and inorganically grown vegetables

sample vegetables	Inorganic/synthetically grown vegetables (CFU/gm)	organic vegetables(CFU/gm)
Carrot	8.50	4.01
Cabbage	8.41	5.92
Coriander	8.39	6.09
Fenugreek	8.51	5.31
Tomato	8.47	4.2

Salmonella , *Shigella*, *S.aureus* were not noted. Total aerobic counts were also observed to be much lower in organic vegetables. The application of neem based pesticides might have been responsible for this observation.

6. Conclusion

We concluded that the nutrition quality of organic vegetables is higher than inorganic vegetables. From the above analysis discussed above we observe the Vitamin C and riboflavin in organic vegetables. We also observed that the beta-carotene is highest in carrot as compared to coriander and tomato. Lycopene is obtained in tomatoes with high concentration value. Lutein is powerful antioxidant and mainly presents in the fenugreek leaves. The value of luteinin organic fenugreek was much larger than the synthetically grown fenugreek leaves. High aerobic plate counts were not observed in organic grown vegetables; neither did *Salmonella*, *Shigella* and S.*aureus* exist indicating acceptable microbiological quality for consumers.

1 Eosin ethylene blue agar (1ltr)
Stock solution
EMB = 27.1gm
Agar = 5gm
Distilled water = 1ltr

Autoclave sterilize for 15 min at a temperature of 121 and pressure of 15 psi, store in refrigerator.

2 Macconkey agar (1ltr)
Stock solution
Macconkey broth = 40.1gm
Agar = 15gm
Distilled water = 1ltr

Autoclave sterilize for 15 min at a temperature of 121 and pressure of 15 psi, store in refrigerator.

3 Nutrient agar (1ltr)
Stock solution
Nutrient broth = 13gm
Agar = 15gm
Distilled water = 1ltr

Autoclave sterilize for 15 min at a temperature of 121 and pressure of 15 psi, store in refrigerator.

4 Xylose lysine deoxycholate agar (1ltr)
Stock solution
Xylose lysine deoxycholate broth = 56 gm
Agar = 2.5gm
Distilled water = 1ltr

Do not autoclave it. Just heat for 5 mints.

5 Potassium Phosphate Buffer (0.1 M)
Stock solution
K_2HPO_4 = 61.5ml
KH_2 PO_4=38.5ml
Temp. 25^0C

6 DNS reagent
Stock solution
DNS=1gm
NaOH =16ml
Rochelle salt=30gm
Distilled water=50ml
Rochelle salt = sodium potassium tartrate
Bring to 100ml distilled water. Storage in brown water.

7 Saline solution
Stock solution for 100ml
NaCl=0.85gm
Distilled water =100ml

Autoclave sterilize for 15 min at a temperature of 121 and pressure of 15 psi, store in refrigerator.

8 Iodine solution
Stock solution 1ltr
Iodine =3.3gm/L
Potassium Iodide=6.6gm/L
Specific temp 20^0C
Storage at room temperature, protection from light.

9 $$beta\ carotene \left(\frac{\mu g}{100gm}\right) of\ sample = OD \times 13.9 \times 10^4 \times \frac{\text{volume of aqueous layer}}{\text{wt.of sample} \times 560 \times 1000}$$

10 $$beta\ carotene \left(\frac{\mu g}{100gm}\right) of\ sample = OD \times 13.9 \times 10^4 \times \frac{10}{100 \times 560 \times 1000}$$

11 $$beta\ carotene \left(\frac{\mu g}{100gm}\right) of\ sample = OD \times 0.0248$$

12 Standard deviation

$$S = \sqrt{\frac{1}{N}\sum_{i=1}^{N}(x_i - \bar{x})^2}$$

13 Lycopene content (mg/kg) = (A_{503} – 0.0007) ×30.3/g tissue.

14 $\text{Total carotenoids (\%)} = \frac{\text{Absorbance at 446nm}\times 10000}{\text{Sample mass in g}\times 2550}$

Lutein (%) = total carotenoids× lutein

Zeaxanthin(%)= total carotenoids × area 5 zeaxanthin

15 $colony\ forming\ unit(per\ gram) = \frac{no\ of\ colonies}{volume\ of\ plates\ (in\ ml)} \times dilution\ factor$

16 Standard of vitamin C

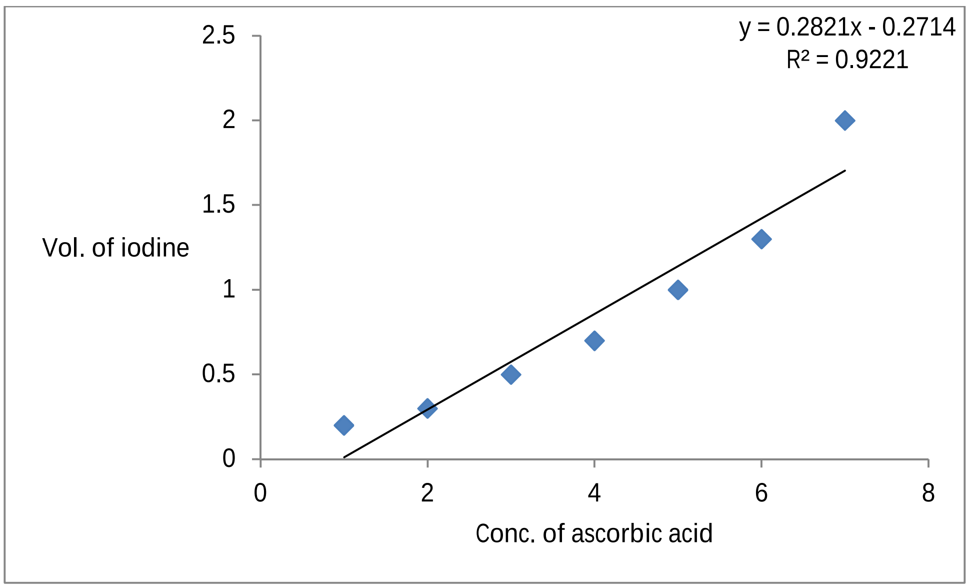


Fig 2 Concentration of ascorbic acid with the iodine

17 Standard of lycopene

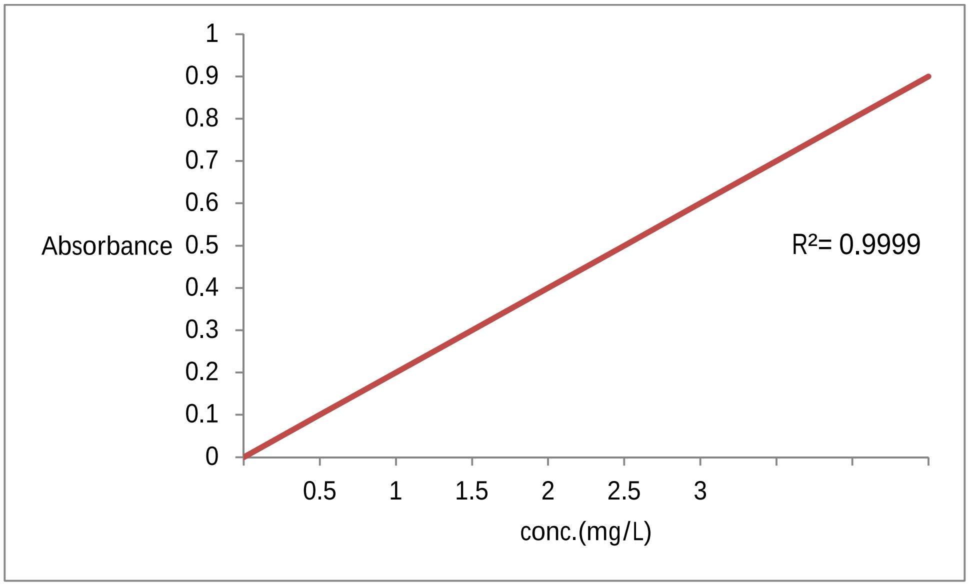


Fig: 3 Absorption vs Lycopene Conc.(mg/L) in Hexane

18 **Bibliography**

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