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## Comparative Study of Beta Carotene Determination by various Methods: A Review

**Naseem Zahra, Alim-un-Nisa, Farah Arshad, Sehrish Muhammad Malik, Imran Kalim, Sajila Hina, Ali Javed and Syed Manzar Inam**  
Food and Biotechnology Research Centre, PCSIR,  
Laboratories Complex, Ferozepur Road, Lahore-54600, Pakistan

(Corresponding author: Naseem Zahra)

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**ABSTRACT:** -Carotene, a precursor of vitamin A, possesses pronounced radical scavenging properties. It is an orange colored pigment, which is present in many fruits and vegetables. This review critically discusses different strategies for the determination and extraction of beta carotene. Methods are Gas chromatography (GC), high performance liquid chromatography HPLC, and capillary electro chromatography (CEC), Spectrophotometry and FTIR analysis. The extraction and determination of beta carotene in different food stuffs has been quantified. Beta carotene is a prime part of food and it has also nutritional importance. It is useful for the cure of many diseases due to its various positive effects. This has centered the attention on -carotene dietary supplementation in healthcare as well as in the therapy of degenerative disorders and several medical issues but overdosing can cause many health problems.

**Keywords:** Beta carotene, HPLC, FTIR, CEC, Gas chromatography, Spectrophotometry

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### INTRODUCTION

In our daily life, we are encountered by highly reactive species such as molecular oxygen and other reactive oxygen species (ROS) that are extremely destructive for the health of living beings (Halliwell & Gutteridge, 1984). As we are highly exposed to such reactive species, so we are at higher risk to develop oxidative stress (Thomson & Paton, 2014). Antioxidants, on the other hand, shield us against ROS and are very crucial for survival. Thus, nutrients that are rich in antioxidant property or antioxidant as supplements are administered on large scale to control ROS triggered damage (Seifried *et al.*, 2007). It was first believed that antioxidants tend to scavenge free radicals and absorb their reactivity. However, modern concepts revealed that the molecular

mechanism takes place in three major steps. First step involves scavenging, second step cause the free radical to get sheltered in the antioxidant radical. Finally in the last step, radical is transferred into antioxidant matrix.

-carotene is considered as one of the most powerful scavenger of singlet oxygen (Niki, 2014; Stahl & Sies, 2003) and its lower plasma levels can even lead to death (Lee *et al.*, 2001). -carotene is a tetra terpene, lipid soluble plant pigment with molecular weight of about 536 Da. It is widely distributed in nature as a plant pigment and reported to have a large number of beneficial biological effects (Sies, 1992). -carotene is also known as provitamin A because it is enzymatically transformed into retinal, and then finally into retinol (vitamin A) (Mayne, 1996).

It exhibits antioxidant property by scavenging radicals (peroxyl), in order to prevent lipid peroxidation (Sies, 1992; Mortensen *et al.*, 1997., Palozza & Krinsk, 1991). It also shows anti-mutagenic effects and induces apoptosis & cell cycle arrest (El-Habit, 2000). In addition -carotene has its important role in modulating immune function by stimulating the activity of neutrophils in the blood (Elliott, 2005). The increased uptake of carotenoids from fruits & vegetables lowers the incidence of various diseases such as cataract formation (Gale *et al.*, 2001; Jacques & Chylack, 1991), cardiovascular diseases (Gaziano *et al.*, 1995; Gey *et al.*, 1993), macular degeneration (Seddon *et al.*, 1994) and cancer (Ziegler, 1991). The hypothesis leads to the fact that -carotene supplementation may cause prevention of above mentioned diseases. With an appropriate consumption of carotenoids and other such nutrients (phytochemicals) can lower the mortality rate globally (Khoo *et al.*, 2011).

**Metabolism:** In nature -Carotene predominantly exist in its all-trans -carotene form. Two molecules of retinaldehyde is produced by the symmetrical cleavage of -Carotene by an enzyme known as -Carotene- 15,15'- oxygenase (BCO1). However, it can also be cleared by another enzyme -carotene-9, 10-oxygenase (BCO2) in an asymmetric manner to generate apocartenals and -ionone ring, which is further transformed into retinaldehyde (Von, 2013; Von, 2010; Von, 2012). The proper mechanism by which the second conversion takes place has not been completely understood.

After the generation of retinaldehyde from the provitamin A, it is further oxidized by the enzymes (belong to retinaldehyde dehydrogenase family) to form all-Trans retinoic acid. The product retinoic acid is actually the active form of vitamin A (Pares *et al.*, 2008). After the over-production of retinoic acid in tissues, the enzymes of cytochrome P450 family can cause its oxidative degeneration to produce more polar species which are transcriptionally inactive (Abu-Abed *et al.*, 2001). On the other hand, retinaldehyde can also be reduced reversibly to generate retinol that is commonly known as vitamin A (D'Ambrosio *et al.*, 2011). Furthermore, lecithin retinol acyltransferase (LRAT) act upon retinol to produce retinyl ester which is the important storage form of vitamin A found mainly in liver but also in heart, adipose tissue, kidney and lungs (O'Byrne *et al.*, 2005; Batten *et al.*, 2004).

**Functions:** Approximately 600 naturally occurring carotenoids are being known to man, out of which only 50 exhibit vitamin A activity (Bauernfeind, 1981). As whole carotenoids are extensively present in plants and photosynthetic bacteria, and perform two major functions: act as accessory pigments during photosynthesis and in photoreception. The conjoined polyene structure of carotenoids allows them to effectively carry out the above mentioned functions. Their conjugated organization permits the molecule to absorb light and to further inactivate free radicals. Different carotenoids are being ingested by humans on regular basis; including those exist naturally in fruits and vegetables & those which are used as food colorants (Mayne, 1996). Furthermore, it has been reported that the precursor of Vitamin A- -carotene affects myocardial tissue, cell, and organ functions which permits the build-out of systematic strategies for utilization of this novel compound in management and prevention of heart diseases (Csepanyi *et al.*, 2015). The medical welfare of -carotene is also well established. It can be used as adjuvant treatment in patients suffering from erythropoietic pro-toporphyria and is also known to reduce the chances of age related macular degradation. Additionally it also lowers the risk of breast cancer in pre-menopausal women. (Gandini *et al.*, 2000; Seddon *et al.*, 1994; Thomsen *et al.*, 1979).

The major property of -carotene is that it has antioxidant activity that quenches free radicals in human cell membranes, HDL, and as well as LDL (Ringer *et al.*, 1991). With the aid of in-vitro studies gathered by using radical generating systems show the potential of -carotene to scavenge free radicals by utilizing the process which includes the incorporation of radicals into carotenoids, hydrogen extraction, and electron transfer (Niki *et al.*, 1995; Krinsky, 2001). The most important function includes protection of lipid membranes by its synergistic action along with vitamin C and E (Niki *et al.*, 1995; Zhang & Omaye, 2000).

**Hydrophobicity:** The hydrophobic character of -carotene represents an evident challenge for the significant use in cell cultures and hence they demand efficient solubilizers. It is insoluble in water and even barely soluble in Acetonitrile, Methanol, or Dimethylsulfoxide (DMSO). In order to enhance the solubility DCM, Hexane, and Chloroform can be employed with the highest possible solubility in Tetrahydrofuran (Fellt *et al.*, 2005; Aust *et al.*, 2001; Craft & Soares, 1992).

This information proves to be helpful in choosing an appropriate solvent for chromatographic separation, extraction, and purification of such entities (Aust *et al.*, 2001; Craft & Soares, 1992).

**Handling & storage:** Due to the sensitive nature of  $\beta$ -carotene, it must be handled with great care. Its stock solutions must be prepared and stored under inert environment i.e., by employing certain gases such as nitrogen and argon. The prepared solutions must be stored at  $-20^{\circ}\text{C}$  to  $-80^{\circ}\text{C}$  under light protected atmosphere (Feltl *et al.*, 2005; Franssen-van Hal *et al.*, 2005; Oliver & Palou, 2000). During extraction of  $\beta$ -carotene, evaporation is recommended at the temperature below  $40^{\circ}\text{C}$  due to the thermal instability of the compound (Feltl *et al.*, 2005). The entire sample handling should be carried out under red, orange, or yellow light (Feltl *et al.*, 2005; Rodríguez, 2005; Middtun & Ueland, 2011). Furthermore, it should not be exposed to extreme pH conditions (Oliver & Palou, 2000).

**Toxicity:** The widespread applications of  $\beta$ -carotene in various fields such as in cosmetics, drug industries, and food etc, it was therefore very crucial to carry out authentic toxicity tests of the compound with the aid of wide range of techniques. No mutagenicity was observed after the substance has been passed through various examinations (Bagdon *et al.*, 1960; Heywood *et al.*, 1985). Even no signs of embryotoxicity were detected in rats and rabbits, and no interference in their reproductive function was noticed. After passing  $\beta$ -carotene through various trials, it was being placed in the US Food and Drug Administration (FDA) category of foods and is generally recognized as safe (GRAS) (Diplock, 1995). Furthermore,  $\beta$ -carotene has been used to treat the patients suffering from the genetically inherited photo-sensitivities for at least 20 y, and in this context even large doses of pure  $\beta$ -carotene do not show harmful effects (Mathews-Roth, 1986). However, hypercarotenemia may develop in those individuals who are taking supplement doses of about  $>30$  mg/d for longer period of time. The symptoms disappear gradually after the treatment is terminated and this condition is entirely non- malignant (Diplock, 1995).

Apart from favorable impacts of  $\beta$ -carotene, there are various clinical trials which report that their supplementations can even results in toxicity. Studies carried out in 1994 reveals that  $\beta$ -carotene supplementation is linked with greater chances of "lungs cancer" in smokers (Albanes *et al.*, 1994) In the following years, various studies verified this

alarming situation, and the entire blame was put on  $\beta$ -carotene (Tanvetyano & Bepler, 2008). Despite of that, the actual mechanism does not provide any evidence which blames  $\beta$ -carotene for inducing lung cancer in smokers (Vrolijk *et al.*, 2015).

**Sources:**  $\beta$ -carotene is an essential part of different fruits and vegetables. It is present in sweet potato, carrot, and tomato belonging to the genera *Ipomea*, *Daucus*, and *Solanum* respectively. The major source of  $\beta$ -carotene in the diet is carrot along with leafy green vegetables. Furthermore, yellow- orange colored vegetables (such as pumpkin) also serve as a rich source of  $\beta$ -carotene (Khoo *et al.*, 2011).  $\beta$ -carotene naturally exists with 11 double bonds in its structure and exhibits orange color (Rao & Rao, 2007). Fruits with Yellow color have relatively lower levels of  $\beta$ -carotene in them, for e.g., the fruits and vegetables belong to the genera Citrus, Ananas, Pyrus, Cucumis, Vigna, and Maranta. Additionally, Zea mays are also a great source of  $\beta$ -carotene (Muzhingji & Tang, 2008).

It is also responsible for being a multi-colored agent in various plant species and is extensively employed as food colorant (Khoo *et al.*, 2011). Furthermore, red palm oil can be used as an alternative source of  $\beta$ -carotene which is cultivated in the different areas of Southeast Asia & Africa and has positive effect on health (Oyewole & Amosu, 2010; Rice & Burns 2010).

#### **Methods for determination of $\beta$ -carotene**

There are some methods for the detection and determination of  $\beta$ -carotene.

##### *A. HPLC*

High pressure liquid chromatography is an efficient technique for the detection of  $\beta$ -carotene in various samples. It has the ability to discriminate between the similar geometrical forms of carotenoids. It is a rapid technique and the use of very minute quantity of sample makes it an acceptable method for analysis of different samples (Feltl *et al.*, 2005). The chemicals which are utilized in this technique must be of HPLC grade. Even the water used in it must be of ultra pure quality.

The preparation of standard solution is the requisite for the  $\beta$ -carotene analysis. Stock solutions are frequently prepared in n-hexane and their concentrations can be adjusted according to the need of experiment and solutions are prepared in ppm concentration.

First the working standard solutions are introduced into the HPLC system via an injecting port. HPLC is attached to an isocratic pump and has a column which is further connected to UV visible detector. Complete software is associated with an HPLC system which identifies the peaks and represents quantified results. Calibration is an important step of HPLC which is attained by a mobile phase such as methanol, dichloromethane, acetonitrile. The wavelength and pressure of the column is adjusted according to the practical requirements. After running standard solutions the peaks thus obtained were plotted to obtain a straight line. All the samples containing  $\beta$ -carotene is treated similarly to that of standard solutions. The resulting peaks are automatically analyzed and quantified by the software (Ahamad *et al.*, 2007).

#### B. Spectrophotometry

Spectrophotometry is for the quantification and determination of  $\beta$ -carotene. Spectrophotometry is a sharp way to analyze the concentration of  $\beta$ -carotene in vegetables and fruits samples (Arabshahi *et al.*, 2007). For the determination of  $\beta$ -carotene in samples the process of liquid-liquid extractions is considered with the UV absorbance of 461nm. Spectrophotometry is an accurate and precise detection method for the quantitative analysis of samples having presence of  $\beta$ -carotene. This method illustrates confined linearity with increased average recovery of carotenoids (Careri *et al.*, 1999). According to this, the average recovery is 100.2% and the limit of detection of spectrometer is 0.04% and the relative standard derivation is less than 6.4%, limit of detection of spectrophotometer is 0.11 $\mu$ g/ml. The results showed that corn has highest amount of  $\beta$ -carotene that is 31%, carrots 24%, pumpkin 20% and paprika 25% (Karnjanawipagul *et al.*, 2010). This method is inexpensive, linearized, and accurate and could be transferred to many quality control departments.

#### C. Gas Chromatography

Gas chromatography is a fast and preferred method for the analysis of  $\beta$ -carotene and its cleavage products. This focuses on the separation of many volatile mixtures. The temperature gradient is 50°C to 280°C with the presence of capillary columns (Wu *et al.*, 1999). Different type of columns is used for the analysis of  $\beta$ -carotene in various samples. PB-5MS TM column with 70eV of electron impact by applying 60°C-130°C is mostly used for detection purposes (Nonier *et al.*, 2004).

$\beta$ -carotene analyte fragmentation and detection is mainly done by the use of gas chromatography by the oxidation of  $\beta$ -carotene with copper or radical reaction at 70eV (Emenhiser *et al.*, 1996).

#### D. Capillary electrophoresis

Capillary electrophoresis is a useful technique for the separation and determination of many photosynthetic pigments. A number of capillary electrophoresis is also used for the analysis of  $\beta$ -carotene i.e. non aqueous capillary electrophoresis, electro kinetic electrophoresis and micellar capillary electrophoresis is preferred by using by coated and uncoated columns. The separation principle depends upon differences between the mobility of analyte also on the electric field which is produced by the high voltage among the electrodes present in the buffer solution (Jorgenson & DeArman, 1981). The optimal conditions are attained by setting up the separation conditions using diode assay detector. Non aqueous capillary electrophoresis (NACE) is an effective method for separation of  $\beta$ -carotene ranging from 1.11-2.45 ppm. The solvent mixture is methanol, acetonitrile, tetrahydrofuran having 5:4:1 ratio. The aliquot 336, tricaprylmethylammonium chloride is use as a cationic surfactant and to coat the capillaries. The recovery percentage of quantitative results is found between 96.7-102% (Riffe-Chalard *et al.*, 2000; Negro *et al.*, 2003).

By using the 30% THF, 30% methanol, 5% 5mmol/L tris, 30% CAN as substrate, the beta carotene can be separate in 6 minutes (Belin & Gulacar, 2007).

#### Atmospheric pressure chemical ionization

This process has the positive ionization mode for the identification of beta carotene and various products like retinoic acid or retinol. The negative ionization mode is more efficient and provides ~10-folds sensitivity 9 (Arnold *et al.*, 2012). The principle of beta carotene detection is based on the high mass accuracy which is detected in the form of radical cations (Matuszewski *et al.*, 2003). Atmospheric pressure chemical ionization, APCI-MS is useful for the analysis of  $\beta$ -carotene and  $\beta$ -carotene and also for the all Trans and 13-cis retinol forms of beta carotene (Heudi *et al.*, 2004). APCI-TSQ triplequadrupole MS is a type of APCI which is for the determination of beta carotene in tomatoes. Another type of APCI which is associated with HPLC is used to determine and characterize the cleavage products of beta carotene in vegetables at 110°C (Zeb & Murkovic, 2013).

### A. FTIR spectroscopy

This technique is used to structural and vibration properties of  $\beta$ -carotene. The data obtained from the Fourier transform infrared spectroscopy provides information about the angle obtained by the beta ionone rings comparable to the molecular plane. Furthermore it also gives data about the individual C-C bonds of beta carotene and its respective length and angles (Schlucker *et al.*, 2003). This technique also provides an additional advantage of differentiating between cis  $\beta$ -carotene and all Trans  $\beta$ -carotene. Particularly this technique proves to be useful in quality assurance of treatment and standard solutions. Determination of beta carotene is done by the FTIR in real samples, such as carrot roots and pumpkin (Schulz *et al.*, 2005; Bayerl C, 2008). FTIR is applicable in the quality control departments.

### Health impacts of $\beta$ -carotene

$\beta$ -carotene is a pigment which is converted into vitamin A in body. When one molecule of beta carotene is broken down it is converted into the formation of two molecules of vitamin A. Vitamin A is necessary for the eye health and good vision aspects. It has also useful effects on mucous membrane, healthy skin and many metabolic processes (Bjelakovic *et al.*, 2007; Gabriele *et al.*, 2000). The main shiny feature of beta carotene is its antioxidant activity. It protects the body from the free radicals which damages the tissues by the process of oxidation also secures the immune system; lessen the risk of cancer and many other heart diseases (Chan *et al.*, 2009). Another aspect is that, too much amount of beta carotene is dangerous for smokers and pregnant females. So it may become a cause of lung cancer, cataract and other health hazards (Hu & Cassano, 2000).

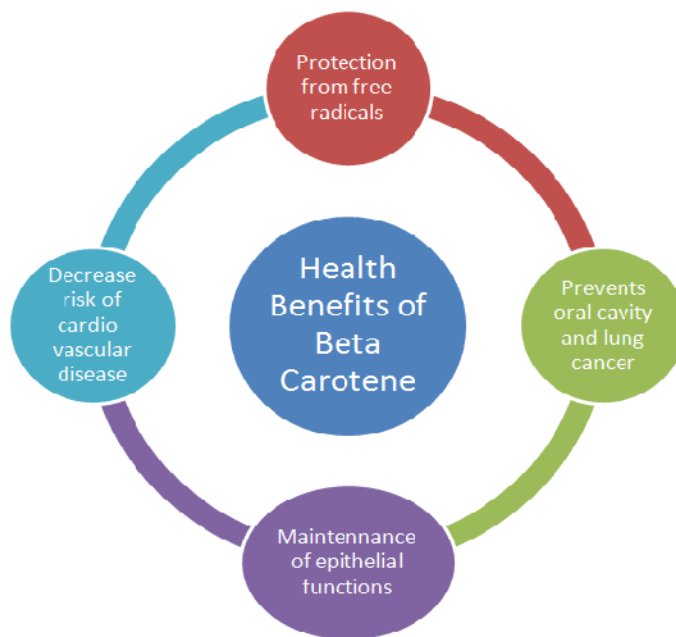


Fig. 1. Health Benefits of Beta carotene.

### Medical uses

There are following beneficial effects of beta carotene which is being used in medical area.

#### A. Erythropoetic protoporphyria

The supplementation of beta carotene has proven advantageous and beneficial effects to people having this disease because it has been an effective and diverse treatment for the Erythropoetic protoporphyria for many decades i.e. 45 years (Besur *et al.*, 2014; Burri, 1997; Mathews-Roth, 1993). It is an autosomal dominant disease (Burri, 1997).

The symptoms of this disease appear after birth or may be years later. In this disease the mutation of ferrochelatase enzyme which is responsible for the heme biosynthesis. Ferrochelatase starts to increase the insertion of ferrous into protoporphyrin which produce protoheme (Besur *et al.*, 2014; Mathews-Roth, 1993). So the activity of EPP is decrease from 10-50% which results in an increase concentration of protoporphyrin in tissues whose symptoms are pain, burning, erythema and puritis cause by even a small exposure to sunlight.

In this case the high dose of  $\beta$ -carotene is the main treatment. The doses may be 60mg, 180mg and 300mg per day. Beta carotene is also useful against the treatment of sunburn (Gollnick *et al.*, 1996; Micozzi *et al.*, 1998).

#### *B. Cystic fibrosis*

Cystic fibrosis is a genetic disease which is a main cause of lethal inflammation in children and lung infection in adults. In this disease the concentration of  $\beta$ -carotene is become half or one quarter of the normal amount (Homnic *et al.*, 1993; Bui, 1988; Portal *et al.*, 1995). So the oxidative damage seems to be abnormally high which increase the oxidative stress due to lung infection and intestinal malabsorption from diet (Winklhofer *et al.*, 1995). For the maintenance of beta carotene concentration in serum 0.5mg  $\beta$ -carotene per kg body weight is necessary for 3 months to reduce the chances of cystic fibrosis (Lepage *et al.*, 1996).

#### *C. AIDS/HIV infection*

Due to AIDS or HIV infection the amount of beta carotene is highly decreased in blood plasma (Phuapradit *et al.*, 1995; Tang *et al.*, 1996; Fryburg *et al.*, 1995). The short term supplementation of beta carotene leads to change in CD<sub>4</sub> cells. So the 180mg amount of  $\beta$ -carotene per day increase the white blood cells, T cells, B cells and lymphocytes which has an impact on healthy immune system (Tang *et al.*, 1996). Because  $\beta$ -carotene intake, starts the formation of vitamin A concentration which stimulate the macrophages, lymphocytes and bone marrow to produce white blood cells. In this way beta carotene is useful to minimize the probability of AIDS (Garewal *et al.*, 1992).

#### *D. Osteoporosis*

It is the most pervasive metabolic bone disease which is caused by the oxidant activity and oxidative stress (Fryburg *et al.*, 1995). Different studies shows that antioxidants, like beta carotene and lycopene from natural sources restrain the damaging effects of oxidative stress (Rao, 2006; Silverton, 1994). The  $\beta$ -carotene has challenging effect on cell proliferation (Kim *et al.*, 2003) due to its endogenous and synthetic ability (Suda *et al.*, 1993; Liu *et al.*, 1999; Key *et al.*, 1990), also mark the differentiation of alkaline phosphatase and inhibit the osteoclast formation (Park *et al.*, 2003). The oxidative stress is related to osteoporosis and antioxidants have also rousing effect on it. There is a reduced level of antioxidant enzymes and vitamin A in osteoporotic females which may cope up by the daily uptake of beta carotene as

suggested amount (Rao *et al.*, 2003; Ishimi *et al.*, 1999).

#### *E. Cardiovascular diseases*

Beta carotene has a supportive role in the prevention of cardio vascular diseases. Serum cholesterol level is used as biomarker for the risk of CHD (Arab *et al.*, 2000; Rao & Rao, 2007; Rissanen *et al.*, 2000). Oxidation of LDL is a key role in the pathogenesis of arteriosclerosis which leads to cardio vascular issues (Agarwal & Rao, 1998).  $\beta$ -carotene and lycopene was shown notably response to reduce the level of oxidize low density lipoprotein by the consumption and uptake of tomato sauce, juices and oleoresin capsule as a source of  $\beta$ -carotene which has ability to reduce serum cholesterol level and thereby lessening the risk of CVD (Fuhrman *et al.*, 1997).

Beta carotene is also used to reduce the Alzheimer disease, epilepsy, Parkinson's disease, schizophrenia, psoriasis, headache, asthma, macular degeneration, infertility, night blindness during pregnancy as well as fever after giving birth and diarrhea (Poh-Fitzpatrick, 1985).

#### **Side effects**

Excessive dose of beta carotene may leads to some health issues which are mentioned below:

##### *A. Carotenosis*

Beta carotene is a precursor of vitamin A which main source is carrots and other fruits as well as vegetables. Vitamin A is particularly converted into retinoid so the over consumption of  $\beta$ -carotene can leads to carotenosis, a disease in which skin color changes to turn into orange by the deposition of carotenoids in outer layer of epidermis. So the chances of carotenosis can reduce by the decreased dietary intake of  $\beta$ -carotene (Stahl *et al.*, 1998).

##### *B. Hypervitaminosis A*

This disease is caused by the increased amount of vitamin A in body. In intestine, an enzyme dioxygenase converts the beta carotene into vitamin A increase over the normal amount it leads to a disease called Hypervitaminosis A, which also infects the skin. When beta carotene supplements are put to stop or dietary intake is reduced the problem of skin disease can resolve (Mathew *et al.*, 2012).

##### *C. Lung cancer risk*

High dose of beta carotene diet or supplements have been assorted with chronic and increased rate of lung cancer in smokers.

### C. Lung cancer risk

High dose of beta carotene diet or supplements have been assorted with chronic and increased rate of lung cancer in smokers. It also increases the risk of intracerebral hemorrhage, prostate cancer and cardiovascular diseases by the cell proliferation in smokers who have a history of increase amount of asbestos (Fuhrman *et al.*, 1997). Various epidemiological studies showed that individuals who accumulate a high dose or large quantity of carotenoids rich food, fruits and vegetables have increased risk of lung cancer (Batieha *et al.*, 1993; Giovannucci *et al.*, 1995). So the people who smoke should not take beta carotene rich diet except under doctor's supervision.

### D. Effect on reproductive system

Beta carotene is a precursor of vitamin A which is a group of retinoic acid, retinyl esters, retinal and retinol. Excessive amount of vitamin A is linked with many birth defects like heart deformities, eye and lung diseases (Gaziano & Hennekens, 1993). That's why pregnant females are also recommended not to take high amount of vitamin A supplements. So the standard dose of beta carotene is necessary for metabolic reactions in body whether the high doses of carotenoids are accompanied with toxicity (Sluijs *et al.*, 2009).

### Fruits and vegetables highest in beta carotene

This chart shows the amount of beta carotene in following fruits and vegetables:

**Table 1: Amount of -carotene in some fruits and vegetables.**

Fruits	Amount of -carotene (mcg)	Vegetables	Amount of -carotene (mcg)
Melons	11883	Chilies	63455
Apricot	8665	Lettuce	59240
Grapefruit	3450	Spinach	48917
Cherries	3080	Turnip	43448
Plums	2449	Cabbage	41247
Peaches	2200	Carrot	40419
Watermelon	2020	Basil	27323
Apricot	1800	Pumpkin	23851
Papaya	1415	Potato	19790
Mango	1369	Tomato	4988
Guava	1100	Okra	1452
Peach	831	Peas	1109
Plum	826	Cucumber	600
Orange	355	Eggplant	133
Figs	230	Beet	93
Pineapple	140	Radish	50
Apple	71	Soya bean	30
Banana	58	Garlic	7
Strawberry	44	Onion	5
Lemon	24	Corn	2

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