OPTIMIZATION OF NUTRITIONAL & BIOCHEMICAL COMPOSITION AND DEVELOPMENT OF HEALTH FOODS-A STUDY OF MICROGREENS

Dissertation

Submitted to the Punjab Agricultural University
in partial fulfillment of the requirements
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in
FOOD AND NUTRITION
(Minor Subject: Food Science and Technology)

By

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(L-2016-HSc-98-D)

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CERTIFICATE I

This is to certify that the dissertation entitled **"Optimization of nutritional & biochemical composition and development of health foods – A study of microgreens"** submitted for the degree of **Ph.D.** in the subject of **Food and Nutrition** (Minor Subject: **Food Science and Technology**) of the Punjab Agricultural University, Ludhiana, is a bonafide research work carried out by **Pooja Bhatt (L-2016-HSc-98-D)** under my supervision and that no part of the dissertation has been submitted for any other degree.

The assistance and help received during the course of investigation have been fully acknowledged.

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CERTIFICATE II

This is to certify that the dissertation entitled **"Optimization of nutritional & biochemical composition and development of health foods – A study of microgreens"** submitted by **Pooja Bhatt (L-2016-HSc-98-D)** to the Punjab Agricultural University, Ludhiana, in partial fulfillment of requirement for the degree of **Ph.D.,** in the subject of **Food and Nutrition** (Minor Subject: **Food Science and Technology**) has been approved by the student's Advisory Committee along with the Head of the Department after an oral examination on the same.

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ABSTRACT

Microgreens (seedling of vegetable and herbs) are gaining popularity as a new culinary ingredient due to their high nutrient content. The present study optimizes the best combination of cultivation and harvesting to attain nutrient quality in four culinary microgreens belonging to different families. Results revealed that the best technique for optimization of nutrient composition was soaking the seeds overnight, outdoor cultivation and harvesting at an early stage (10^{th} day). Based on NQS 7.1 broccoli microgreens were most nutrient dense microgreen followed by spinach microgreens. Microgreens contain significantly ($p<0.05$) higher nutrients and bioactive compounds as compared to their sprouts and mature counterparts. Just 100g of microgreens fulfilled more than 100% of Estimated Average Requirements (EAR) of minerals among Indian adults. Drying methods influenced the nutritional composition of the broccoli microgreen powder. β-carotene content (400.54ug/100g), total phenolic content (2645.88 mg GAE/100g) and flavonoids (673.82 mg QE/100g) of shade dried microgreens was significantly higher. A significant higher content of ascorbic acid (16.80 mg/100g), chlorophyll (3.63g/100g), antioxidant activity (67.55%) and all the minerals was reported in microwave dried microgreens. Further fresh microgreens, microgreen juice and powdered microgreens were incorporated in commonly consumed health foods in order to enhance their taste and nutritional composition. All the health products i.e. missi roti, cooked vegetable and dal, salads, juice blend, flavoured milk and food sprinkler supplemented with broccoli microgreens were organoleptic highly acceptable. In term of fresh broccoli microgreens supplementation, 25% in missi roti, 5 and 7.5% in cooked vegetable and dal, 30 and 40% in russian and tofu salad was highly accepted. Incorporation of fresh broccoli microgreen juice at 30% in juice blend and flavoured milk was highly acceptable. Powdered broccoli microgreens supplemented at 40 % of in regular spice mix was highly accepted. Overall nutritional composition in broccoli microgreens supplemented health foods had significantly ($p<0.05$) high content of vitamins C, β-carotene content and minerals namely calcium, zinc, iron, potassium and magnesium when compared to the control product.

Keywords: Microgreens, bioactive compounds, drying technique, health foods, nutrient quality score

Signature of Major Advisor **Signature of the Student**

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ਪੌਸ਼ਿਟਕ ਤੱਤਾਂ ਦੀ ਬਹੁਤਾਤ ਹੋਣ ਕਾਰਨ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ (ਸਬਜ਼ੀਆਂ ਅਤੇ ਹਰਬਸ ਦੇ ਅੰਕੁਰ) ਨੂੰ ਰਸੋਈ ਵਿੱਚ ਵਰਤੀ ਜਾਣ ਵਾਲੀ ਨਵੀਂ ਸਮੱਗਰੀ ਦੇ ਤੌਰ ਤੇ ਬਹੁਤ ਜ਼ਿਆਦਾ ਪਸੰਦ ਕੀਤਾ ਜਾ ਰਿਹਾ ਹੈ। ਵੱਖੋ-ਵੱਖਰੀਆਂ ਪ੍ਰਜਾਤੀਆਂ ਨਾਲ ਸਬੰਧਤ ਪਕਾਉਣ ਯੋਗ ਚਾਰ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਵਿੱਚ ਪੌਸ਼ਟਿਕ ਗੁਣਵਤਾ ਪ੍ਰਾਪਤ ਕਰਨ ਲਈ ਮੌਜੂਦਾ ਅਧਿਐਨ ਦੌਰਾਨ ਕਾਸ਼ਤ ਅਤੇ ਵਾਢੀ ਦੇ ਵਧੀਆ ਸੰਯੋਜਕਾਂ ਦਾ ਅਨੁਕੂਲਨ ਕੀਤਾ ਗਿਆ। ਅਧਿਐਨ ਦੇ ਨਤੀਜਿਆਂ ਤੋਂ ਪਤਾ ਚੱਲਿਆ ਕਿ ਬੀਜਾਂ ਨੂੰ ਰਾਤ ਭਰ ਭਿਉਂ ਕੇ ਰੱਖਣ, ਖੁਲ੍ਹੇ ਵਿੱਚ ਉਗਾਉਣ ਅਤੇ ਜਲਦੀ (10ਵੇਂ ਦਿਨ) ਤੋੜਨ ਨਾਲ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਤੋਂ ਵਧੇਰੇ ਪੌਸ਼ਟਿਕ ਤੱਤ ਪ੍ਰਾਪਤ ਹੁੰਦੇ ਹਨ। NQS 7.1 ਦੇ ਅਧਾਰ ਤੇ ਦੇਖਿਆ ਗਿਆ ਕਿ ਪੌਸ਼ਟਿਕਤਾ ਦੇ ਅਧਾਰ ਤੇ ਬਰੌਕਲੀ ਸਭ ਤੋਂ ਪਹਿਲੇ ਨੰਬਰ ਅਤੇ ਇਸ ਮਗਰੋਂ ਪਾਲਕ ਦਾ ਨੰਬਰ ਸੀ। ਅੰਕੁਰਾਂ ਅਤੇ ਪੱਕੇ ਹੋਏ ਅੰਸ਼ਾਂ ਦੇ ਮੁਕਾਬਲੇ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਵਿੱਚ ਪੌਸ਼ਟਿਕ ਤੱਤਾਂ ਅਤੇ ਜੀਵ ਕਿਰਿਆਤਮਕ ਸੰਘਕਟਾਂ ਦੀ ਮਾਤਰਾ ($p<0.05$) ਅਰਥਪੂਰਨ ਤੌਰ ਤੇ ਜ਼ਿਆਦਾ ਸੀ। ਸਿਰਫ 100 ਗ੍ਰਾਮ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਨਾਲ ਭਾਰਤੀ ਬਾਲਕਾਂ ਦੀ ਖਣਿਜਾਂ ਦੀ ਅਨੁਮਾਨਤ ਔਸਤਨ ਲੋੜ (EAR) ਦੀ ਸੌ ਫੀਸਦੀ ਪੂਰਤੀ ਹੋ ਜਾਂਦੀ ਹੈ। ਸੁਕਾਉਣ ਦੀਆਂ ਵਿਧੀਆਂ ਨੇ ਬਰੌਕਲੀ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨ ਪਾਊਡਰ ਦੀ ਪੌਸ਼ਟਿਕ ਸੰਰਚਨਾ ਨੂੰ ਪ੍ਰਭਾਵਿਤ ਕੀਤਾ। ਛਾਂ ਵਿੱਚ ਸੁਕਾਏ ਗਏ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨ ਵਿੱਚ ਬੀਟਾ ਕੈਰੋਟੀਨ (400.54ug/100g), ਕੁੱਲ ਫਿਨੋਲਿਕਸ (2645.88 mg GAE/100g) ਅਤੇ ਫਲੇਵੋਨਾਇਡਸ (673.82 mg QE/100g) ਦੀ ਮਾਤਰਾ ਅਰਥਪੂਰਨ ਤੌਰ ਤੇ ਜ਼ਿਆਦਾ ਸੀ। ਮਾਈਕ੍ਰੋਵੇਵ ਵਿੱਚ ਸੁਕਾਏ ਗਏ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਵਿੱਚ ਐਸਕਾਰਬਿਕ ਐਸਿਡ (16.80 mg/100g), ਕਲੋਰੋਫਿਲ (3.63g/100g), ਐਂਟੀਆਕਸੀਡੇਂਟ ਗਤੀਵਿਧੀ (67.55%) ਅਤੇ ਸਾਰੇ ਦੇ ਸਾਰੇ ਖਣਿਜਾਂ ਦੀ ਮਾਤਰਾ ਅਰਥਪੂਰਨ ਤੌਰ ਤੇ ਜ਼ਿਆਦਾ ਸੀ। ਆਮ ਖਾਧੇ ਜਾਣ ਵਾਲੇ ਭੋਜਨ ਉਤਪਾਦਾਂ ਦੇ ਸੁਆਦ ਅਤੇ ਪੌਸ਼ਟਿਕਤਾ ਨੂੰ ਵਧਾਉਣ ਲਈ ਉਹਨਾਂ ਵਿੱਚ ਤਾਜ਼ਾ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ, ਮਾਈਕ੍ਰੋਗ੍ਰੀਨ ਜੂਸ ਅਤੇ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਦੇ ਪਾਊਡਰ ਦੀ ਵਰਤੋਂ ਕੀਤੀ ਗਈ। ਬਰੌਕਲੀ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨ ਦੀ ਵਰਤੋਂ ਕਰਕੇ ਬਣਾਏ ਗਏ ਭੋਜਨ ਉਤਪਾਦ ਜਿਵੇਂ ਕਿ ਮਿੱਸੀ ਰੋਟੀ, ਪਕਾਈ ਹੋਈ ਸਬਜ਼ੀ ਅਤੇ ਦਾਲ, ਸਲਾਦ, ਜੂਸ, ਫਲੇਵਰਡ ਦੁੱਧ ਅਤੇ ਫੂਡ ਸਪਰਿੰਕਲਰ ਨੂੰ ਬਹੁਤ ਜ਼ਿਆਦਾ ਪਸੰਦ ਕੀਤਾ ਗਿਆ। ਮਿੱਸੀ ਰੋਟੀ ਵਿੱਚ 25%, ਸਬਜ਼ੀ ਅਤੇ ਦਾਲ ਵਿੱਚ ਕ੍ਰਮਵਾਰ 5 ਅਤੇ 7.5%, ਰਸ਼ੀਅਨ ਅਤੇ ਟੋਫੂ ਸਲਾਦ ਵਿੱਚ ਕ੍ਰਮਵਾਰ 30 ਅਤੇ 40% ਪੱਧਰ ਤੱਕ ਤਾਜ਼ਾ ਬਰੌਕਲੀ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਦੀ ਵਰਤੋਂ ਨਾਲ ਸਭ ਤੋਂ ਵਧੀਆ ਨਤੀਜੇ ਪ੍ਰਾਪਤ ਹੋਏ। ਜੂਸ ਅਤੇ ਦੁੱਧ ਵਿੱਚ ਤਾਜ਼ਾ ਬਰੌਕਲੀ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਦੀ 30% ਪੱਧਰ ਤੱਕ ਵਰਤੋਂ ਕਰਨਾ ਸਭ ਤੋਂ ਵਧੀਆ ਸੀ। ਆਮ ਵਰਤੇ ਜਾਂਦੇ ਮਸਾਲਿਆਂ ਵਿੱਚ ਬਰੌਕਲੀ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਦੇ ਪਾਊਡਰ ਦੀ 40% ਤੱਕ ਵਰਤੋਂ ਸਭ ਤੋਂ ਵਧੀਆ ਸੀ। ਦੂਜੇ ਉਤਪਾਦਾਂ ਦੇ ਮੁਕਾਬਲੇ ਬਰੌਕਲੀ ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ ਦੀ ਵਰਤੋਂ ਕਰਕੇ ਤਿਆਰ ਕੀਤੇ ਗਏ ਉਤਪਾਦਾਂ ਵਿੱਚ ਵਿਟਾਮਿਨ ਸੀ, ਬੀਟਾ-ਕੈਰੋਟੀਨ ਅਤੇ ਖਣਿਜਾਂ ਜਿਵੇਂ ਕਿ ਕੈਲਸ਼ੀਅਮ, ਜ਼ਿੰਕ, ਆਇਰਨ, ਪੋਟਾਸ਼ੀਅਮ ਅਤੇ ਮੈਗਨੀਸ਼ੀਅਮ ਦੀ ਮਾਤਰਾ ਅਰਥਪੂਰਨ ($p<0.05$) ਤੌਰ ਤੇ ਜ਼ਿਆਦਾ ਸੀ।

ਮੁੱਖ ਸ਼ਬਦ: ਮਾਈਕ੍ਰੋਗ੍ਰੀਨਸ, ਜੀਵ-ਕਿਰਿਆਤਮਕ ਸੰਘਟਕ, ਸੁਕਾਉਣ ਦੀ ਵਿਧੀ, ਸਿਹਤਯਾਬ ਭੋਜਨ ਪਦਾਰਥ, ਪੌਸ਼ਟਿਕ ਗੁਣਵੱਤਾ ਅੰਕ

ਮੁੱਖ ਸਲਾਹਕਾਰ ਦੇ ਹਸਤਾਖਰ | **ਵਿਦਿਆਰਥੀ ਦੇ ਹਸਤਾਖਰ**

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CHAPTER I

INTRODUCTION

In India most of the deaths are caused by diet related diseases such as obesity, cardiovascular, respiratory, hypertension, stroke, diabetes & cancers etc., due to imbalanced food consumption patterns and in next 25 years their burden will expand as an outcome of the rapidly ageing population which is major burden for public health sector (Patel *et al* 2011). Many epidemiological and clinical studies have persuading evidence that increasing the consumption of fruits and vegetables tend to have lower occurrence of diseases like hypertension, cancer, chronic heart disease and stroke (Boeing *et al* 2012). Vegetables are known as protective food in view of nutritive and medicinal value as well as the important components of nutritional security. Fruits and vegetables rich diet supply large quantity of human bioactive compounds, such as vitamin C, vitamin E, carotenoids which includes lutein, zeaxanthin and β-carotene, and pigments like anthocyanins and isoflavones, which have long been recognized as the most important prerequisites for preventing cancers and cardiovascular disease and reduce the occurrence of age related diseases (Craig 1997 and Rice-Evans *et al* 1997). Consumption of at least 400 grams of fruits and vegetables per day is recommended by World Health Organization (WHO/FAO 2005), while the World Cancer Research Fund recommends the consumption of fruit and non-starchy vegetables consumption of 600 grams per day (WCRF/AICR 2007). However, the average intake of vegetables is below the recommended levels. Vegetables with abundance of bioactive compounds not only supply necessary nutrients but also have effect on human health. The non-availability of fresh and pesticide residue-free vegetables for consumption are becoming major concern for the vegetarian population of India. In this respect, in the same genre microgreens are an emerging functional food crop that sustainably diversifying global food systems, promoting human health and make possible adaption to urbanization & global climate changes (Michell *et al* 2020, Choe *et al* 2018).

Microgreens are fascinating genre of consumable greens, also known as 'vegetable confetti'. This is a new special category crop, defined as tenuous immature greens produced from the seeds of herbs and vegetables, including wild species (Xiao *et al* 2012). A report of 2013 published by the USDA's Agricultural Research Service, called microgreens "the new substance that is a powerhouse of human bio nutrients". Microgreens are usually harvested at the first true leaf stage of growth and belong to the class of 'functional foods' and regardless of their small in size they have superior levels of bioactive compounds (Sharma *et al* 2012). Microgreens contain higher content of phytonutrients and secondary metabolites like microelements, carotenoids and phenolic compounds than their mature leaf counterparts. These new and tiny vegetables are nutritive, possess distinctive organoleptic properties, versatile and sustainable crop from cultivation to consumption.

Microgreens can be harvested 7–21 days afterward germination relying upon their species that has been used when cotyledonary leaves are fully expanded and still turgid and the first true leaves are formed (Gioia and Santamaria 2015). For microgreens, the crop will be suitable which germinate easily and grow quickly. Their size ranges from 1 to 3 inches (2.5 to 7.6 cm), including the stem and leaves. Microgreens has a single central stem which is trimmed just above the soil line during harvesting. Microgreens are diminutive but they can support a large array of brilliant colours, intense flavours and tender textures.

Now a day, microgreens are not only used as garnish, but also used as basic ingredients in the preparation of many dishes like salads, smoothies, soups, breads, juices and sandwiches (Renna *et al* 2016,Klopsch *et al* 2018,Ghoora and Srividya 2017, Sharma et al 2021). Over couple of years, they have attained immense popularity in upscale markets and restaurants as a new culinary trend because of its intrinsic characteristics (delicate texture, vivid colour and variety of flavor i.e. spicy, earthy, sweet and savoury) and mainly due to its nutraceutical value & reputed beneficial effects on human health. Even the eating quality of microgreens were best compared with flavor score (Xiao *et al* 2015). Its growing, harvesting and postharvest handling conditions have a huge effect on the accumulation and degradation of microgreens phytonutrients (Xiao *et al* 2012).

Microgreens are poles apart from sprouts even if both greens are consumed in an immature state (Treadwell *et al* 2010). They are older than sprouts and much younger than baby greens. Sprouts are generally produce under the cover of darkness, wet conditions which are good for microbial growth and their consumption, unlike that of micro and baby-greens has been implicated in outbreaks of food borne epidemics (Ebert 2012 and Xiao *et al* 2014a). Whereas, microgreens are grown in soil media and they take full use of light conditions which improves its nutritional quality than sprouts. Also, microgreens have a stronger flavour upgrading qualities over sprouts and a wide range of leaf colour, variety and shape (Ebert 2012). Microgreens are recommending by chefs and consumers in high-end restaurants for their attractive colours, fresh textures and most importantly there compelling flavours (Xiao et al 2013). Compared to sprouts, radish microgreens were 3-4 logs lower population of *E.coli* which showed that they had low safety risks (Xiao *et al* 2014b).

Microgreens are cultivated in a different environment specifically indoor and outdoor, relying upon the scale of production. Its production take very small area and can be grown in plastic trays, pots and benches or channels, so it is versatile both to micro-scale urban and large scale commercial business, allows its commercialization while it is still growing, so that it will be harvested or trimmed directly by the final consumer. This innovation of selling the product bypasses harvesting and many postharvest handling issues and may guarantee freshness and high quality in terms of nutritional value (Gioia and Santamaria 2015). Microgreens crop require no pesticide and fertilizer (Xiao *et al* 2015), still some organic manure and peat used to increase the yield of crop (Murphy *et al* 2010). The accretion of nutrients and bioactive compounds of microgreens is dependent upon a number of pre harvesting conditions like soaking of seeds, growth media's and light conditions and harvesting stages. In the regulation of vegetable growth and phytonutrient content light is the most important environmental factor which affects it and therefore light play major role in accumulation of nutrients. Microgreen production under natural light (sunlight) had the fastest growth rate and yield better quality in terms of carotenoids, antioxidants content, mass and height than those under artificial light (Lau *et al* 2019). The soaked seeds of chia microgreens led to higher microgreens yield than the unsoaked seeds (Junpatiw and Sangpituk 2019). The peak concentration of plant pigments, total phenols and vitamin C were on second week and antioxidant capacities and plant pigments tends to decrease with increase in etiolation period (Patras 2021). Easy maintenance and their phytochemical composition increased popularity among consumers, urban farmers, food technologists and nutritionists (Kyriacou *et al* 2020). With the projected increasing global urban population which tend to increase the needs of arable fields to meet food requirements by the population to avoid food insecurity. Microgreens commercially grown in vertical racks which tend to provide global food and nutrition security and safety. The suitable and economical production setting of microgreens will facilitate their implementation in the area of commercial microgreen cultivation on a large scale, adverse climate, war crisis and famine (Rajan *et al* 2019). Also containerized cultivation of microgreens in home scale or in an industrial level provides an additional benefit of harvesting them freshly before consumption with less waste than mature counterparts (Kyriacou *et al* 2016).

Microgreens farming is becoming popular among urban farmers these days because it is high value crop which requires minimal horticultural inputs. It has fast turnaround time as it can be harvested at 7-21 day after germination. It is year round growing thus the income generation will be whole year. Less space occupancy makes it a better option when compared to traditional crop farming. It is best for urban agriculture as the greens are not grown until maturity, harvested at infancy which reduces the risk and cost of investment. Many plant crops can be grown together in small area which makes microgreen farming high yield to space ratio. Alongside the possibility of conserving natural resources and chemical free crop, the production and consumption of microgreens have extra advantages, transform these products into a new, healthy and environmental-friendly vegetable option**.** Also, production of microgreens offers the opportunity to increase the sustainability of vegetable production. The requirement for water is 158-236 times lesser than the mature broccoli crop cultivated in field when compared to broccoli microgreens (Weber *et al* 2017) whereas the radish vegetables cultivated at field required 7-8 times more water than radish microgreens (Singh *et al* 2018).

Commonly cultivated microgreens are spinach (Allende *et al* 2004, Bergquist *et al* 2006) table beet (Murphy *et al* 2010 and Pill *et al* 2011), mustard (Kopsell *et al* 2012), buckwheat (Janovska *et al* 2010 and Kou *et al* 2013), amaranth, arugula, bull's blood beet, basil, celery, cilantro, golden pea, mizuna, peppercress, popcorn shoots, mustard, red mustard, red beet, red cabbage, red orach, radish, sorrel, red sorrel and wasabi (Xiao *et al* 2012), cabbage (Chandra *et al* 2012 and Sun *et al* 2013), broccoli (Kou *et al* 2014, Ghoora et al 2020), radish (Xiao *et al* 2014b), lettuce (Pinto *et al* 2015), fenugreek (Wadhawan *et al* 2017),Chicory (Reena *et al* 2018), carrot, fennel, spinach and onion (Ghoora *et al* 2020).

Different species of microgreens contains different amounts of functional compounds, e.g. red cabbage microgreens had the highest concentration of ascorbic acid, while green daikon radish microgreens was rich in tocopherol. Popcorn shoots and Golden pea tendrils had low concentration of vitamins and carotenoids as compared to other microgreens, but were still as high as some common mature vegetables (Xiao *et al* 2012). The antioxidant and phenolic content of three stages i.e. seeds, sprouts, and microgreens shows higher content in microgreens with 4873 mg GA equivalents/100 g dry weight of sesame microgreens and 4339 µg/ mL highest antioxidant activity of finger millet (Senevirathne *et al* 2019).

The shelf life of microgreens vary depending on the variety but there are certain factors during storage which can increase the shelf life i.e. temperature and packaging. The most favourable temperature for storage is 1 ^{o}C with no chilling injury and maintained acceptable quality for 28 days (Xiao *et al* 2014a). During storage, microgreens were found to have better retention of the phytochemical compounds i.e. Fenugreek microgreens retain acceptable sensory properties (freshness, aroma, colour, wiltness and tenderness) better than mature leaves. It is recommended to utilize the fenugreek microgreens within a fortnight of storage under refrigerated conditions and appropriate packaging (Ghoora *and* Srividya 2017).

Oxalic acid, an anti-nutrient known to interfere with divalent cations such as calcium, forming insoluble salts and they are widely distributed in green leafy vegetables. Fenugreek microgreens has 1.5 fold lower oxalic acid content than mature one where as spinach microgreens has 16-fold lower oxalate content than mature one (Ghoora *et al* 2020). Green leafy vegetables contain highest concentrations of nitrate whereas microgreen has lower content of nitrate (Pinto *et al* 2014).

Alpha-glucosidase and alpha-amylase inhibitors are used in antidaibetic medication for regulating postprandial glucose levels in diabetic patients (Sales *et al* 2012). Fenugreek microgreens extract found to significantly inhibit porcine pancreatic alpha-amylase. It is suggested to have 4-5 tablespoons of fenugreek microgreens in daily diet for promising dietary intervention **(**Wadhawan *et al* 2017**).** It is reported that microgreens are likely has effects in cancer prevention due to their high content of vitamins, carotenoids, polyphenols, and glucosinolates (Choe *et al* 2018). It is recommended that the consumption of microgreens especially from brassica species in daily diet could be an effective nutritional strategy in prevention of cancer as it showed a statistically significant anti proliferative effect (de la Fuente *et al* 2020).

Pea and lupin microgreens were supplemented in a bread to enhance nutritional value. There were low losses of bioactive compounds (carotenoids, chlorophylls and flavonoid) and significant formation of pheophytin. Genistein is highly retained in the lupin microgreen bread, which has anti carcinogenic properties, especially in prostate cancer, bladder cancer and breast cancer (Klopsch *et al* 2018). Microgreens should be supplemented to the diet of astronauts who cannot get the regular supply of fresh produce from earth for long duration spaceflight missions. Production of microgreens for spaceflight will give access to nutrient dense crop designed to fulfill the nutritional needs of space crew (Kyriacou *et al* 2017). The supply of perishable goods like fruit and vegetables to remote locations is a serious concern and a large number of Indian army of high altitude areas suffer from scanty supply of vegetables. For this situation, micro scale production of microgreen is excellent choice which can diversify and enrich the diet in remote areas where fresh food availability is serious problem. Due to the conditions and less availability of resources, a multilayer unit of microgreens production is adopted by the Defence Institute of High Altitude Research, Leh Ladakh (Singh *et al* 2019).

Microgreens are characterized by a higher nutrient concentration as compared to vegetables harvested at the standard growth stage (Ebert *et al* 2014 and Xiao *et al* 2012). The concentration of vitamins (ascorbic acid, tocopherol and phylloquinone) and carotenoids (lutein, β-carotene and zeaxanthin) in 25 varieties of microgreens revealed that compared to mature vegetables, microgreens have 10 times higher content of antioxidant (Xiao *et al* 2012). It demonstrated that few grams of microgreens can satisfy the recommended daily intake of these vitamins. The consumption of 41g of red cabbage microgreens and 17 g of garnet amaranth microgreens would satisfied the daily need of vitamin - C and vitamin - K respectively for a sedentary adult man. Also, 13 microgreens were reported to excellent sources of β-carotene, vitamin C and Vitamin E, meeting 24-72 %, 28-116 % and 28-332 %

of reference daily intake of the vitamins, respectively (Ghoora *et al* 2020). Therefore, microgreens could represent a nutrient rich food source that can enrich and diversify the diet of categories of particularly demanding consumers, like vegetarians and vegans (Gioia and Santamaria 2015). They are also rich in minerals, both macro-mineral i.e. potassium & calcium and micro-minerals i.e. iron & zinc (Xiao *et al* 2016). Microgreens consumption could be concerted strategy to promote health and prevent chronic diseases.

The cultivation and harvesting techniques in the pre harvest stages of microgreens were optimized to obtain better nutritional, functional and sensorial quality. Further, nutrient richness of microgreens can be translated into development of health foods well within reach of middle income consumers. Development of these health foods can be instrumental in sharing a paradigm shift in terms of perception about microgreens from upscale consumption to mass consumption. Thus, the present study was planned in this direction.

OBJECTIVE

1. To optimize nutritional and biochemical composition of microgreens under different cultivation and harvesting conditions.
2. To optimize nutritional and biochemical composition of microgreens under different processing conditions.
3. To compare microgreens with sprouts and mature leaves in term of nutritional and biochemical composition.
4. To develop health foods from microgreens obtained from optimized production and processing conditions.
5. Organoleptic & nutritional evaluation of various health foods supplemented with microgreens.

CHAPTER II

REVIEW OF LITERATURE

In the recent past, urbanization, economic development, industrialization and market globalization have accelerated which caused rapid changes in the diet and lifestyle of the population of developing countries, where the standard of living has increased but there are inappropriate dietary pattern and decreased in physical activity too and a corresponding increase in diet related chronic diseases. The adverse dietary shift in the diet of population is towards a high energy dense diet with more part of fat and sugar in food, more saturated fat intake, decreased consumption of complex carbohydrates and dietary fiber and decreased intake of fruit and vegetables. Vegetables and fruits play major role in a healthy diet. The consumption of vegetables and fruits in appropriate amounts linked to greater lifestyle which could help in prevention of diet related degenerative diseases and micronutrient deficiency.

World Health Organization estimated that 1.7 million deaths worldwide are due to low intake of fruit and vegetables and it's among the top ten selected risk factors for global mortality also. Worldwide, 14 percent deaths due to gastrointestinal cancer, 9 percent due to stroke and 11 percent deaths due to ischemic heart disease are caused by insufficient intake of fruit and vegetable. Vegetables are good source of vitamin, minerals, fiber with high amount of various antioxidant bioactive compounds such as tocopherols, polyphenols, carotenoids, ascorbic acid and glucosinolates which have been considered to prevent chronic diseases such as cancer, heart disease, hypertension, obesity and diabetes. Especially, green leafy vegetables are found to be a natural supplement of iron and beta carotene and its consumption doubled the vitamin A pool size. The meal containing adequate amount of green leafy vegetables found to contain higher bioavailable densities of micronutrients relative to cereal legume based meal. Therefore, natural antioxidant has lift considerable interest among dietitians, nutritionists, food technologist, food manufacturers and consumer due to their expected safety and possible therapeutic value.

Furthermore to the increasing scientific interest in the relationship between degenerative diseases and diet, the people's concerns about the effect of food in degenerative disease prevention have led to seek for healthy food products by both food industry and consumers. Thus the novel consumption of microgreens, which are considered functional food due to their high bioactive compounds, which are high in respective to their mature one. Microgreens may be diminutive, but they are punch well above their weight when it comes to nutrition. Xiao *et al* (2012) claimed that the microgreens were 4 to 40 fold more concentrated with nutrients than their mature one.

An effort was made to scan the available literature regarding the proposed research problem. A brief resume of the studies related to present investigation has been presented below under sub headings:

2.1. Prevalence of degenerative diseases

2.2. Production of microgreens

2.3. Chemical composition of microgreens

2.4. Microgreens consumption related to prevention of degenerative diseases

2.5. Microgreens shelf life and packaging

2.6 Sensory evaluation of Microgreens

2.1 Prevalence of degenerative diseases

The epidemiological transition in 21^{st} century with the rapid urbanization and growing megacities around the world there is huge change substantially in population's eating habit and lifestyle with two acquired characteristics that is over and inadequate nutrition, the consequence of which increase the prevalence of degenerative diseases such as diabetes, cardiovascular diseases, stroke, cancer, hypertension etc. The cardiovascular disease is the main cause of death globally. The prevalence of CVD accounts more than 80 percent deaths in low and middle income countries (Harikrishnan *et al* 2014). The World Health Report 2009 introduce the term " Risk Transition" to mention the changes in the pattern of life style, nutrition and alcohol consumption mostly in middle and low income countries.

In India more than 135 million people were affected by obesity. The prevalence of obesity in backward states (socioeconomically) Bihar, Madhya Pardesh, Jharkhand and Chhattisgarh were at low rate whereas the states like Punjab, Goa, Delhi, Andhra Pardesh, Puducherry and Telangana (high socioeconomic) were at high rate of obesity. The prevalence of obesity in South India was higher with 27.2%, followed by 23.8% in North India and with low rate 15% in West India (Ahirwar and Mondal 2019).

Goyal *et al* (2017) studied the risk factors associated with coronary artery disease and trend in prevalence of the disease from 1994 to 2014 in rural Punjab. The prevalence of risk factors associated with CAD was low physical activity (8.2% vs 41.3%), obesity (16.6% vs 35.4%), hypertension (14.5% vs 26.5%), diabetes (4.7% vs 9.7%) and hypercholesterolemia (7% vs 9.6) escalate over 20 years. The age standardized prevalence of coronary artery disease was 2.79% in 1994 and elevated to 4.06% in 2014.

India is diabetic capital with 74 million people, second highest after China. According to WHO (2010), India loses 1.5 percent of GDP as a result of diabetes, stroke and heart disease from 2005 -2015. The prevalence of adult onset diabetes and their correlation with the socio demographic profile among the Karnataka adults studied by Rao *et al* (2010). For the study 1,239 participants (male and female), aged 30 years and above, interviewed face to face and the blood sugar was estimated by glucometer. The high socioeconomic class had 32 percent of the subjects who had diabetes. Among the total respondent, the prevalence of adult onset diabetes was 16%. While the study was conducted 11.2 % were self-reported diabetes where as 4.8 % had high blood glucose during the study been conducted. The prevalence of adult onset diabetes was 18.8 % for males and 14.4 % in females.

Sanjeevaiah *et al* (2019) conducted a cross sectional study to find out the prevalence of diabetes and its related factors in 250 participants for 4 months of period. The age, Basal Metabolic rate, waist circumference, hypertension, smoking and total cholesterol were significant in prevalence of diabetes. The prevalence of diabetes mellitus was 6.4% and 32% out of 250 participants were frequent alcoholic while 6.4 percent out of 250 had ≥126 mg/dl fasting blood glucose.

Tripathy *et al* (2017) conducted a community based study to evaluate the prevalence of diabetes mellitus and its associated risk factors in Punjab. The survey was carried out in a multistage stratified sample of 5127 participants and followed WHO STEPS questionnaire, anthropometric and blood pressure measurements. The prevalence of diabetes among the total participants was 8.3%. The significant associated risk factors were family history of diabetes, hypertension, obesity and marital status. The author reported the high prevalence of diabetes in Punjab with most undiagnosed cases among the adult population. They emphasis need for systematic screening and awareness program to recognize the undiagnosed cases in the community.

The incidence of cancer in India is increasing day by day affecting large number of population. The burden of cancer, globally contributing 3rd highest number of cancer cases after China and USA. The report by NCRP (2016) revealed that 1.4 million cases of cancer were estimated for the year 2015 that consist 6, 92,704 males and 6, 95,693 females. It was estimated that the figure would grow to 1.74 million cases without any control measure for the year 2020.

The increasing trend of cancer in especially at Punjab region is alaraming. Bal *et al* (2015) studied the pattern of cancer patients registered in Mukh Mantri Punjab Cancer Rahat Kosh Scheme (MMPCRKS), registered at Rajindra Hospital Patiala from the different different districts of Punjab. Out of 500 cancer registry, 35 % were male and 65% were female patients. The major cancers found in females were breast cancer, next was cervix and ovary while in males the major were colon, next was esophagus and tongue.

Aggarwal *et al* (2015) conducted five year population based epidemiological study in the malwa region of Punjab to study the pattern and extent of cancer. The total 1328 cancer cases were there, out of which 60.9% (809) were females and 39.1% (519) were males. The peak age for male and female both for cancer found to be 50-54 years. The major cancer found in males were 9.6% lung cancer, followed by 8.3% myeloid leukemia,6.8% prostate,6.1% mouth and 6% gall bladder while top 5 leading sites in female were 35.7% breast , followed by 19.1% cervix,5.1% esophagus 4.7% myeloid leukemia and 3.9% gall bladder.

Thakur *et al* (2017) studied the difference in urban and rural cancer incidence and pattern of cancer disease in Punjab and Chandigarh on the basis of population based cancer registries (PBCRs). The population based cancer registry were setup in Punjab (rural) and Chandigarh (urban) covering the population of 2.6 million and 2.9 million, respectively. The rate of breast cancer were four to five times higher in urban registries when compared to the rural while the rate of lung cancer was four to seven times higher in urban when compared with rural. In the urban registries, the leading cancer site was lung among males and among females it was breast in urban registries while the leading site was esophagus among males and cervical cancer in females at rural registries. The author reported that the high incidence of cancer was due to lifestyle change, environmental factors and urbanization.

2.2 Production of microgreens

Bergquist *et al* (2006) studied the variation in nutritional quality of spinach with growth stage and postharvest storage sown on three different occasions. For each occasion, the spinach was harvested at three growth stages at 6-day intervals. The second stage corresponded to a growth period used for baby spinach by commercial growers. The harvested leaves were stored in polypropylene bags at 2ºC and 10ºC. The highest ascorbic acid content in fresh material was found at the stage first. However, during storage period ascorbic acid content decreased significantly, while as the dehydroascorbic acid ratio increased. The baby leaves stored at 2ºC gave a smaller reduction in ascorbic acid content than at 10ºC. The total carotenoid content increased or remained stable during storage. Lutein was the major carotenoid, making up about 39% of the total carotenoid content followed by violaxanthin, b-carotene, and neoxanthin. Visual quality decreased during storage in most cases and was correlated to initial ascorbic acid and dry matter contents. Results also indicated that harvesting the baby spinach a few days earlier than commercial stage of harvest improved the postharvest visual and nutritional quality.

Nadhira *et al* (2021) conducted a study to optimise the best harvesting stage of herbal microgreens species to get maximum nutrients and bioactive compounds. In the study three herbal species i.e. coriander, basil and fenugreek were cultivated and harvesting was done at 4 stages of growth that is sprouts, microgreens, baby greens and mature greens. The coriander microgreens had highest vitamin –E (40mg/g), vitamin- K (4.53 µg/g) and carotenoids (11.2 mg/g). The vitamin C content was highest in fenugreek microgreens whereas the phenol (76.35 mg/g), iron (42.44 mg/g) and antioxidant activity (1020.69 µg/g) was higher in mature coriander leaves. The basil microgreens had high content of calcium i.e 198.49 mg/g. The study suggested to utilize coriander microgreens to enhance the nutritional availability in the diet or meal.

The duration of harvesting influences the nutrient content of plant. Patras (2021) investigated the development stage of white cabbage microgreens and optimises harvesting stage at the most appropriate stage of their growth in order to stimulate the biosynthesis of phenolic compounds of *Brassicaceaes*. The effect of development stages i.e. 5, 7 and 9 days and 2 sodium salts were administrated in two concentrations i.e. 0.01 M and 0.1M) on the phenolic and antioxidant activity of white cabbage microgreens. The development stage of 5 days had highest antioxidant properties and 0.01 M NaCl and 0.1 M Na_2SO_4 had highest antioxidant properties.

Muchjajib *et al* (2015a) optimise the best organic media alternatives for the production of microgreens in Thailand. In the study various organic media were compared for maximum yield, nutrient content and the microbial population of microgreens grown in it. The organic media were sand, peat, coconut coir dust, sugarcane filter cake, vermicompost which were taken in pair of 1:1 that is - i)coconut coir dust + peat, ii) coconut coir dust + sugarcane filter cake and iii)coconut coir dust + vermicompost. The utmost microgreens yield of vine spinach (5.17 kg m^{-2}) & kangkong (2.26 kg m^{-2}), krathin (1.69 kg m^{-2}), leaf mustard (4.17 kg m^{-2}) and rat-tailed radish (3.90 kg m^{-2}) were obtained with coconut coir dust + peat, Coconut coir dust + sugarcane filter cake respectively. For the production of microgreens, the nearby natural bio-materials were observed to be powerful. The nutrient content per 100 g of microgreens from vine spinach, rat –tailed radish, leaf mustard, krathin and kangkong were protein 7.05, 6.83, 6.55, 6.72 & 6.67 g ; fiber: 5.33, 3.70, 3.94, 2.54 & 4.28 g ; iron:0.60, 0.65, 0.71, 0.99 & 2.67 mg ; calcium 18.1,19.8,31.8, 34.0 & 20.6 mg. Microgreens were stored at 5°C after 7 days and microbial populations were determined to be at safe levels.

In another study Muchjajib *et al* (2015b) studied the production of microgreens from local plant species in Thailand. The local plant species selected were: rat-tailed radish (*Raphanus sativus Linn. var. caudatus Alef*), leaf mustard (*Brassica juncea (L.) Czern*.) and sano (*Sesbania javanica Miq*). 7 days seedling heights were 6.18, 11.58, and 10.36 cm whereas 100-seedling fresh weights were 5.49, 16.34 and 11.47 g; and microgreens fresh weight yield were 2.21, 1.84 and 1.30 kg m^{-2} for *B. juncea, R. sativus var. caudatus* and *S. javanica*, respectively. During the distribution of fresh microgreens at a local supermarket on a trial basis revealed the consumer acceptance which was ranging from 4.4 to 4.9 on a 1 to 5 scale. The antioxidant capacity was highest in leaf mustard. The respective nutritional contents per 100 g of edible portion of microgreens from B. juncea, R. sativus var. caudatus and *S. javanica* were: protein 6.55, 6.83 and 6.97 g; fiber 3.94, 3.70 and 3.12 g; vitamin C 31, 28 and 3 mg; beta-carotene 1540, 569 and 366 µg. Chlorophyll contents were 0.324, 0.445 and 0.159 mg g^{-1} fresh weight, respectively. Following seven days of storage at 5°C, microbial populations were evaluated: populations of yeast, mould, *E. coli* and *Salmonella* were all determined to be at a safe level.

There are many factors which play role in accumulation of nutrients in plant, the utmost factors are light and soil conditions. The effect of light and soil condition on the accretion of antioxidant content in red amaranth microgreens studied by Lau *et al* (2019). Three types of soils were used: yellow soil, orange soil and black soil for the cultivation of microgreens and the light source was sunlight. The best growth rate of red amaranth microgreens was observed in black soil followed by orange and then yellow soil. The high yield of red amaranth microgreens were grown in orange soil i.e. increased by 38.24% and for black soil grown was increased by 161.76% when compared with yellow soil grown red amaranth microgreens. The fastest growth rate and yield of microgreens was cultivated under sunlight was better as compared to artificial lighting. The red amaranth had highest levels of carotenoids cultivated under red+ blue and white light whereas ascorbic acid was high in red light cultivated red amaranth microgreens.

Weber (2016) conducted comparative study on cultivation conditions of lettuce and cabbage microgreens, their nutritional quality of hydroponically grown (HP) and on vermicomposting (C) and compared to each other as well as to the nutritional quality of market purchased cabbage & lettuce (fully grown vegetables). High concentrations of nutrients were found in hydroponic and vermicomposting grown microgreens than market purchased mature vegetables. The microgreens grown on vermicompost had greater nutrient contents than those grown hydroponically and thus it was reported that microgreens can be grown easily in one’s home which may provide a means for consumer access to greater quantities of nutrients per gram plant biomass relative to store-bought mature vegetables, which had lower nutrient contents than microgreens with respect to most nutrients examined. Compared to the mature vegetable, high quantities of all the nutrients were found in cabbage microgreens, likewise lettuce microgreens had all the nutrients excluding calcium and sodium.

Tan et al (2020) investigated the sensory and nutritional content of broccoli microgreens cultivated in a local farms and commercial setting. The author evaluated the sensory and nutritional content of broccoli microgreens from market grown hydroponically, local farm grown hydroponically and local farm grown in soil. The overall liking (n=150) based on sensory property (taste, appearance and smell) of broccoli microgreens was from local farm grown in soil (in the range of 4.54-5.38 out of 7 which was fait to very good). Also author explored the correlation among the sensory attributes to overall liking for broccoli microgreens and found that score of overall liking was most strongly correlated with the taste (r=0.83****) of broccoli microgreens. The total chlorophyll concentration was higher among broccoli microgreens (0.33 mg/g) from local farm settings than commercial setting grown. The phenolic concentration and total antioxidant capacity was similar in both cultivation settings sample. The local farm grown microgreens had high vitamin c content (0.56 mg/g) than commercial settings.

Meas *et al* (2020) studied the effect of light irradiation on the enhancement of growth and phytochemical of amaranth microgreens. The red and blue spectra in combination increase the growth and phytochemicals whereas the blue spectra alone enhanced the ascorbic acid content of amaranth microgreens.

Polash *et al* (2019) optimized the growing media for the cultivation of microgreens and assess their nutritional content. The growing media were soil, soil + cow dung, tissue media and foam media. The germination percentage was higher in soil + cow dung with 99.67% (mustard microgreens), 97.33% (leaf mustard), 87.00% (radish) and 96.33% (cabbage). The vigor index and the maximum seedling height were higher in soil + cow dung. The moisture content was higher in radish microgreens 92.5 g per 100g of dry weight of microgreens whereas all the microgreens contain low levels of ash, fiber, carbohydrates, fat and proteins. Mustard microgreens contained high amount of beta carotene (0.24mg/100g of fresh weight) whereas leaf mustard contained high amount of total chlorophyll content (0.92 mg/100g of fresh weight) and lycopene (0.50 mg/100g of fresh weight) compared to radish and cabbage microgreens. The sodium content of leaf mustard was 0.23 g per 100 g dry weight and potassium was 0.52 g per 100 g dry weight whereas radish microgreens had high sulphur content (0.57 g per 100 g dry weight). The phosphorus content of cabbage microgreen was 10.53 g per 100 g dry weight. The leaf mustard contained high content of calcium (1.80 g per 100 g of dry weight) and magnesium (0.78 g per 100g of dry weight) as compared to rest of microgreens. The content of trace minerals was higher in leaf mustard i.e. zinc (748.6 mg per 100 g of dry weight) and iron (319.67 mg per 100 g of dry weight). The vitamin C content

(16 mg per 100 g of fresh weight) and the DPPH radical scavenging activity (IC_{50} value 0.75 µg) were higher in leaf mustard.

The optimization of sowing media, seed preparation, seed sowing rate and harvesting period on the cultivation of chia microgreens was carried out by Junpatiw and Sangpituk (2019). There were 5 treatment of seed preparation: i) unsoaked seeds ii) seeds were soaked for 6 hours at room temperature, iii) seeds were soaked for 12 hours at room temperature iv) seeds were soaked in 70-80˚ C water and then allowed to cool at room temperature for 6 hours and v) seeds were soaked in 70-80˚ C water and then allowed to cool at room temperature for 12 hours. The media's were used coconut coir, sand, rice husk ash, coconut peat + rice husk ash (1:1) and sand +coconut peat +rice husk ash (1:1:1). The seeds of chia were sowed at the rate of 56, 93,130,167 and 204 g per m^2. After sowing the seeds were harvested at 5, 6, 7, 8 and 9th day. The high germination percentage (96-98%), increased fresh weight of microgreens (1.63/100) and highest height (7.25 cm) were with the 5th treatment of seed preparation. The highest microgreen height i.e. 7.02cm, maximum fresh weight i.e. 1.12 g/100 microgreens and highest germination rate i.e. 96-98% were from the seeds grown in sand +coconut peat +rice husk ash (1:1:1). The seed sowing rate with the sowing rate of 204g per m^2 had the maximum fresh weight of microgreens whereas there was no significant difference ($P \leq 0.05$) in the height of microgreens. The crop harvested gives maximum height (7.5 cm) was observed after 9 days of sowing while the crop harvested at the period of 6-9 days of sowing gave the maximum fresh weight i.e. 1.43-1.45 g/ 100 microgreens.

Islam *et al* (2020) studied the effect of selenium bio fortification on the anti-oxidant activity and nutrient content of wheat microgreen extract. The wheat microgreens were cultivated with different concentration of sodium selenite 0, 0.125, 0.25, 0.50, and 1.00 mg/L of selenium in hydroponic system under controlled system for 10 days. The length and yield of wheat microgreens was decreased with the Se bio fortification whereas germination rate (94%) was increased as compared to control. The human bio active compounds such as chlorophyll, vitamin C, carotenoid, phenolic, flavonoids, anthocyanin and antioxidant activity remarkably increased in 0.25 to 0.50 mg/L of selenium bio fortified wheat microgreen extract.

Murphy *et al* (2010) studied the factors which affects the growth of table beet microgreens. The microgreens were cultivated at different sowing rate and compared with the commercially recommended rate that is 201 g per m^2. The shoot fresh weight per m^2 at 15 days after planting was high in 201 g per m^2 sowing rate. The seeds which were pre germinated before sowing had 26% increases in shoot fresh weight of microgreens as compared to unsoaked seeds.

The impact of soil salinity stress was studied by Islam et al (2019) on the bioactive compounds and antioxidant activity of wheat microgreen extract. The wheat microgreens were cultivated with organic media with different content of sodium (Na) that is 12.5,25,50 and 100mM from sodium chloride. The β – carotene was 1.21 µg/ mL, flavonoids was 165 .47 µg/mL, phenolic acid was 41.70 µg/mL and vitamin C was 29.51 µg /mL significantly increased with 12.5mM of sodium treatment in wheat microgreen extracts. The treatment with 25mM of sodium in wheat microgreen extract had increased level of anthocyanin and 2,2 –Diphenyl – 1- picrylhydrazyl radical scavenging activity.

Puccinelli *et al* (2019) cultivated the selenium enriched basil seeds for the production of selenium bio fortified microgreens. The basil plants were grown hydroponically with supplementation of sodium selenite in various concentrations i.e. 0 for control, 4 mg selenium per liter and 8 mg selenium per liter in nutrient solution to produce selenium fortified microgreen seeds. The selenium content of basil microgreens was highest in 8 mg selenium per liter in nutrient solution to produce selenium fortified microgreen seeds. The seeds from plant of selenium treated had high germination index than the seeds of control plants. The bio fortified basil microgreens had highest antioxidant capacity with increase the level of selenium content in the nutrient solution as compared to control.

Chen *et al* (2020) studied the consumer acceptability of hydroponically and farm grown broccoli microgreens and their perceptions towards the purchase and consumption of microgreens. The broccoli microgreens were locally soil grown, locally hydroponic grown and commercial hydroponic grown. The overall acceptability of locally grown (soil and hydroponic) was high as compared to commercial hydroponic broccoli microgreens. Thus, author found that the sensory testing had significant difference between commercially grown and local grown was due to growing condition, transportation and storage condition of microgreens in grocery store which affect appearance, texture and aroma. The perceived benefits (nutritional value, quality and environment friendly) and sensory evaluation had direct relation with the consumer willingness to buy microgreens.

Rocchetti *et al* (2020) investigated the in vitro gastrointestinal digestion of raw and digested sample of red beet and amaranth microgreens to assess antioxidant activity, polyphenols and betalains using untargeted metabolomics and effect of storage and digestion process on the phytochemical content of microgreens. The growing trays of the microgreens were stored for 10 days at 4˚C temperature and harvested at four growth stages: 1, 4, 7 and 10^{th} day of storage. The total phenolic content of red beet microgreens from the first day to tenth day of storage was in increasing trends i.e. 313.8 to 432.7 mg per 100 g of dry weight of microgreens ($p < 0.05$) and after in vitro digestion its was 116.3 to 219 mg/100 g DW. The TPC, total antioxidant capacity (CUPRAC and DPPH) and betacyanins (betaxanthins) content (BC) were significantly increased by 38%, 36% (CUPRAC), 45% (DPPH) and 10%. The

TPC, total antioxidant capacity (CUPRAC and DPPH) and betacyanins (betaxanthins) content (BC) range of amaranth microgreens showed different trends, they first increased on storage of 7 days and then start decreasing till 10th day of storage and was highest on 1st day of storage. The exception was with betacyanins content of amaranth microgreens, which was 478.4 mg/100 g DW on 1st day of storage and 615.8 mg/100 g DW on 10th day of storage. The in vitro digestion of TPC, TAC, and BC values of red beet and amaranth microgreen was significantly decreased from 1st day to 4th day and then increased from 4th day to 10th day of storage. The amaranth microgreens had maximum levels of BC, TPC, CUPRAC and DPPH as compared to red beet microgreens for all days of storage after in vitro digestion. Furthermore, the author studied the impact of storage and in vitro digestion process on the metabolomics profiles of red beet and amaranth microgreens. In the red beet, tyrosol equivalent and stilbenes had highest increasing trends with fold change value of 1.5 and 2.3,respectively, at the ends of storage period. The red beet and amaranth microgreens contained 9 common discriminant marker, encompasses dihydrochalcones (phloridzin), anthocyanins(malvidin hexosides), flavones(diosmin/neodiosmin), phenolic acids (feruloyl hexoside conjugates), hydroxybenzaldehydes(panisaldehyde), hydroxycoumarins(coumarin) and hydroxytyrosol.

2.3 Chemical composition of microgreens

Carotenoids, bound phenolic content, α-tocopherol, flavonoids and antioxidant activity were evaluated in common and tartary buckwheat microgreens by Janovska *et al* (2010). The antioxidant activity was found in both common and tartary buckwheat microgreens. The flavonoids (rutin, quercetine and kaempferol separately) was detected more in tartary buckwheat microgreens. No differences were identified amongst normal and tartary buckwheat microgreens in content of phenolic acids. Both the genotype of buckwheat microgreens represented as nutrient dense.

A comparative study is conducted by Marchioni *et al* (2021) among five Brassicaceae microgreens to analyse their nutrient and Phytochemical content. Broccoli had the highest polyphenol, carotenoid and chlorophyll contents as well as good antioxidant ability. Mustard was characterized by high ascorbic acid and total sugar contents. By contrast, rocket salad exhibited the lowest antioxidant content and activity.

Concentration of vitamins and carotenoids of commercially available microgreens were studied by Xiao *et al* (2012). Varying amounts of phytonutrients were present in different microgreens. The contents of vitamin and carotenoids of 25 commercially available microgreens were compared with the mature leaves. The content of identified vitamins and carotenoids /100g fresh weight (FW) were 20.4 to147.0 mg for total ascorbic acid , Phylloquinone content ranged from 0.6 to 4.1 μg/g FW (fresh weight), meanwhile, α-tocopherol and γ-tocopherol level varied from 4.9 to 87.4 and 3.0 to 39.4 mg/100 g FW (fresh weight), respectively. β-carotene, lutein/zeaxanthin and violaxanthin concentrations ranged from 0.6 to 12.1, 1.3 to 10.1 and 0.9 to 7.7 mg respectively. Among the 25 microgreens evaluated, red cabbage had highest level of ascorbic acid, cilantro had highest concentration of carotenoids, garnet amaranth had phylloquinone and green daikon radish had the highest concentrations of tocopherols. The study indicated that microgreens cotyledon leaves possessed higher nutritional densities than their mature counterparts.

Changes in macro-mineral, trace element & pigments content during lettuce (*Lactuca Sative L.*) growth was studied by Pinto *et al* (2014). In the study the macro-minerals, trace element and pigments were monitored at 5 stages of growth of lettuce. During the lettuce growth, the content of chlorophylls, carotenoids, macro-minerals and trace elements were decreased. At the younger stage (microgreens) high nutritional value was observed. Thus, it was concluded that nutritional value was strongly dependent on growth stage.

Finding of Cigić et al (2020) on accumulation of polyamines in microgreens and seeds of fenugreek, daikon radish, alfalfa and lentil revealed that alfalfa microgreens possessed high levels of agmatine i.e. 5392mg/kg and lentil microgreens had 579mg/kg spermidine. The putrescine and cadaverine content was high in sprouts as compared to microgreens but had 922 mg/kg spermine in fenugreek microgreens.

Pinto *et al* (2015) conducted comparative study of mineral profile and nitrate content of microgreens and mature lettuces. The moisture concentration had no significant difference in microgreens and mature lettuce. They observed that microgreens contain high amount of most of the minerals with lower content of NO_3^- than mature one and minerals were Calcium, Magnesium, Iron, Manganese, Zinc, Selenium and Molybdenum. Thus, microgreens can supply remarkable higher amount of essential minerals than mature lettuces. They can be considered a good source of minerals with very low content of NO_3^-. The mature lettuce was high in both NO_3^- and NH_4^+ concentration. Thus, microgreens can be safely used in the human diet which fulfil the everyday mineral requirements, while reducing their exposure to harmful NO_3^-.

In another study, Xiao *et al* (2015) assessed six microgreens varieties for sensory, chemical composition and nutritional quality characteristics. Among the six varieties of microgreens, the uppermost rated in terms of acceptability of appearance, texture, flavour and overall eating quality was Bull's blood beet (*Beta vulgaris L.)*. Though, as the undermost rated in term of acceptability of flavour and overall eating quality was peppercress (*Lepidium bonariense L.*). Red amaranth (*Amaranthus tricolor L.*) had the highest pH value and lowest total sugars whereas, China rose radish (*Raphanus sativus L.)* had the highest titratable acidity and total sugars. China rose radish, opal basil (*Ocimum basilicum L.*),red amaranth contained high content of total ascorbic acid, vitamin- K, carotenoids, vitamin- E and total phenolic were found. Overall eating quality of microgreens was best correlated with flavor scores.

Bulgari *et al* (2016) aimed to provide an insight into yield, mineral uptake, and quality of basil, Swiss chard, and rocket microgreens grown in a hydroponic system. With reference to data reported in literature for the same species hydroponically grown but harvested at adult stage, these microgreens yielded about half, with lower dry matter percentage, but higher shoot/root ratio. They showed high concentrations of some minerals, but their nutrient uptake was limited due to low yield. Nitrates content was lower if compared with that usually measured in baby leaf or adult vegetables of the same species, as well as the concentration of chlorophylls, carotenoids, phenols, and sugars. Therefore, microgreens seem to be interesting and innovative low-nitrate-salad crops requiring low fertilizer inputs. Nevertheless, an improvement in yield as well as in the content of nutraceutical compounds would be desirable.

Sun *et al* (2013) studied phenolic profiles in five *Brassica* species of microgreens. The study showed that *Brassica* species microgreens had more complex polyphenols profiles and contained more varieties of polyphenols compared to their mature one's. Five *Brassica* vegetables were red cabbage (*Brassica oleracea var. capitata*), purple kohlrabi (*B. oleracea var. gongylodes*), red and purple mustards (*Brassica juncea*), and mizuna (*Brassica rapa var. nipposinica or B. juncea var. japonica*). A total of 164 polyphenols including 30 anthocyanins, 105 flavonol glycosides, and 29 hydroxycinnamic acid and hydroxybenzoic acid derivatives were putatively identified. This study showed that these five *Brassica* species microgreens could be considered as good sources of food polyphenols.

Xiao *et al* (2016) reported that microgreens are good sources of both macro elements (K and Ca) and micro elements (Fe and Zn.). Consumption of microgreens could be a health-promoting strategy to meet dietary reference intake requirements for essential elements beneficial to human health. The mineral element composition was analyzed for 30 varieties of microgreens, representing 10 species within 6 genera of the Brassicaceae family. Brassicaceae microgreens were assayed for concentrations of macroelements, including calcium (Ca), magnesium (Mg), phosphorous (P), sodium (Na), potassium (K), and of microelements, including copper (Cu), iron (Fe), manganese (Mn), and zinc (Zn). Potassium was the most abundant macro element ranging from 176 to 387 mg/100 g fresh weight (FW), followed by P (52–86 mg/100 g FW), Ca (28–66 mg/100 g FW), Mg (28–66 mg/100 g FW), and Na (19–68 mg/100 g FW). Among the microelements, Fe tended to be most abundant (0.47–0.84 mg/100 g FW), followed by Zn (0.22–0.51 mg/100 g FW), Mn (0.17–0.48 mg/100 g FW), and Cu (0.041–0.13 mg/100 g FW).

Metabolomic assessment reveals an elevated level of glucosinolate content in $CaCl_2$ treated broccoli microgreens by Sun *et al* (2015). Preharvest calcium application has been shown to increase broccoli microgreens yield and extend shelf life. Chemical composition comparison shows that glucosinolates, a very important group of phytochemicals, are the major compounds enhanced by preharvest treatment with 10 mM calcium chloride (CaCl2). Aliphatic glucosinolates (glucoeruc in, glucoiberin, glucoiberverin, glucoraphanin, pentyl glucosinolate, and hexyl glucosinolate) and indolic glucosinolates (glucobrassicin, neoglucobrassicin, and 4-hydroxyglucobrassicin) were increased significantly in the CaCl2 treated microgreens using metabolomic approaches. Results indicate that glucosinolates can be considered as a class of compounds that are responsible for the difference between two groups and a higher glucosinolate level was found in CaCl2 treated groups at each time point after harvest in comparison with the control group.

Many local vegetable materials were underutilized due to inadequate scientific knowledge of their nutritional potential. Kaur and Kapoor (2002) reported that carrot leaves had higher phenolic content than carrots. Carrot leaves were used in development of many health foods by Joshi and Mathur (2015), due to its potential source of dietary antioxidant and act as antimicrobial agent, it should be utilized (Syama *et al* 2014).

Xiao *et al* (2012) showed that cilantro (*Coarinder sativum L.*) microgreens had excellent source of tocopherol and lutein/zeaxanthin,also revealed that it had 3 times more β-carotene and more than 5 times violaxanthin as compared to the mature one. It function as spice &herbal medicine both and it is known for its antioxidant, anti-diabetic, anti-convulsant, anti-inflammatory, anti-mutagenic, anti-anxiety, antimicrobial activity, hepatoprotective activity amongst others along with hormone balancing effect and analgesic properties thus promoting its use in foods.

Due to the unique flavor of microgreens they are highly used in culinary preparations as basic ingredients, not only as garnish (Renna *et al* 2016). Thus, Ghoora *et al* (2020), assess the overall nutritional quality and contribution to the daily diet of microgreens which furnish principal dietary guidelines to nutritionists, food industry, dietitians, other health care providers and consumers at large. The study reported the nutritionally ranking of ten culinary microgreens (spinach, carrot, mustard, radish, roselle, fenugreek, French basil, sunflower fennel and onion) based on the levels of twelve nutrients and oxalate (anti nutrient) and their relative importance to the diet. The nutrient quality score was compared to commonly consumed leafy vegetable i.e. spinach to find out the nutritional position of microgreens. The NQS showed that all the microgreens are 2-3.5 fold more nutrient dense than mature spinach with low level of oxalate in all the microgreens. Out of ten culinary microgreens selected, radish microgreen found to be most nutrients dense.

Khoja *et al* (2020) analyzed and compared the bioavailability of iron (in vitro iron solubility and iron uptake) and mineral content of microgreen and mature vegetables. The bioavailability of iron was estimated by human Caco-2 line. The iron uptake in Caco-2 cells was significantly more in case of fenugreek microgreens compared to their mature counterparts. The ratio of calcium to iron for fenugreek (247.21 for mature vs 93.04 for microgreens),rocket (280.94 for mature vs 302.04 for microgreens) and broccoli (71.94 for mature vs 457.95 for microgreens). Broccoli microgreens have high calcium content as compared to mature broccoli by which there is lower iron uptake by Caco-2 cells. Candia *et al* (2018) stated that Calcium exerts a dose-dependent inhibitory effect on iron absorption. The ratio of phytate to iron was 51.43, 173.62 and 55.18 for mature fenugreek, broccoli and rocket whereas 49.57,129.72 and 118.64 for microgreens respectively.

In another study de la Fuente et al (2019) reported the mineral content and bio accessibility of antioxidant compounds of four Brassicaceae microgreens (broccoli, mustard,kale and radish) grown hydroponically. Radish, kale and broccoli had high bioaccessable fraction value for total phenols while mustard and radish had highest bioaccessable fraction for total isothiocyanates, ascorbic acid and total carotenoids. Kale had the highest bioaccessable fraction value for calcium whereas broccoli had lowest BF value for potassium and mustard had highest BF values for magnesium.

Kyriacou *et al* (2019) studied the genotypic variation in phytochemical composition of coriander, cress, kohlrabi, komatsuna, mibuna, mustard, pak choi, radish, tatsoi, green & purple basil, jute and swiss chard microgreens. It was reported that green basil and coriander were excellent source of beta – carotene and total polyphenols whereas purple basil was high in ascorbic acid. The high content of potassium and magnesium were in swiss chard and basil. The harvesting of cress, komatsuna, mibuna, pak choi and tatsoi was at 16^{th} day after sowing while purple basil at 19^{th} day, coriander, green basil and mustard at 20^{th} day and jute, kohlrabi, radish and Swiss chard at 22th day. The highest yield was 5.97 kg fresh weight m^{-2} radish microgreens. The nitrate content was highest in mustard, jute and kohlrabi i.e. 4488, 5164 and 5386 mg per kg fresh weight.

Klopsch *et al* (2018) formulated Bread supplemented with lupin and pea microgreens and their mature leaves to enhance the nutritional value of bread and studied the changes in the bioactive compounds occurred during baking. The microgreens and leaves of pea and lupin contained high amount of carotenoid, flavonoids and chlorophylls but these metabolites change during ontogeny. It was reported that there were no low losses of flavonoids, carotenoids and chlorophyll, the levels of these bio active compounds maintained during baking. There was high retention of Genistein (anti carcinogenic property) in lupin microgreen enriched bread and there was significant formation of pheophytin. The author suggested lupin leaf bread (rich in chrysoeriol) could be a good substitute for simple bread to diabetic patients.

Senevirathne *et al* (2019) studied the phytochemical properties of the crops at different stages (seeds, sprouts and microgreens) and consumer acceptance of ten microgreens. The highest antioxidant activity was in sesame and green pea microgreens as compared to their seeds and sprouts. The microgreens of sesame show the highest total phenol content i.e. 4837 mg GA equivalent/100 g dry weight as compared to respective seeds and sprouts. Among the ten microgreens, lettuce and carrot microgreens were highly acceptable by the consumers with average hedonic value of 7.90 - like very much whereas finger millet, red amaranth and green pea microgreens were next in the consumer preference with average hedonic value of 6.24- like slightly.

Michell *et al* (2020) evaluated sensory perception of consumer and acceptability of six microgreens and studied the barriers and factors contributing to consumer acceptance of microgreens using consumer panel of (n=99). Consumers indicated that there are factors which impact the purchasing of microgreens were access and availability, shelf life, cost, freshness, familiarity of microgreens and knowledge. The highest appearance acceptability was in red cabbage, red garnet amaranth and bull's blood beet due to red color of the crop. The highest flavor acceptability was in tendril pea microgreen. The overall acceptability with highest mean liking score was tendril pea and lowest with arugula. The food neophobia (tendency to avoid or refuse unfamiliar foods) score was low (FNS < 25) among the consumer population.

The anti-nutrients (phenol, oxalate and saponin) and nutritional evaluation of ten selected microgreens were studied by Nair and Lekshmi (2019). All the microgreens were low in carbohydrates with lowest level 0.303g/100g of red amaranth microgreens to 1.250 g/100g of red cow pea microgreens. The carrot microgreens showed high levels of niacin relative to its mature leaves. All the microgreens contained safe levels of anti-nutrients saponin, oxalate and phenol.

Quantitative comparison of nutrient content of radish microgreen and the mature radish studied by Singh *et al* (2018). The water required for the cultivation of mature radish vegetable was 130 L water per square meter whereas radish microgreens required 18 L water per square meter. The yield of mature radish was 5.5 kg/square meter/90 days while the radish microgreens yield 6.075 kg/square meter/90 days. The comparison of nutrient content of dried samples of mature radish and radish microgreens was 2043 mg/100g vs 1716 mg/100g (fluoride), 214 mg/100 g vs 418mg/100g (magnesium), 1571 mg/100 g vs 947 mg /100g (sodium), 3447 mg/100g vs 4292 mg/100 g (potassium), 0.16 mg/kg vs 0.45 mg/kg (selenium), 364 mg/100 g vs 535 mg/100 g(calcium) and 286mg/100g vs 536 mg/100 g (total phosphorus), 497 µg/100g vs 486 µg/100 g (folic acid), 78.36 µg/100g vs 21.96 µg/100g and 0.13 % of total fat vs 0.47 % of total fat (omega 3 fatty acid). When the fresh radish microgreens and mature radish was compared for nutrient content, the radish microgreens were 1-3 times more nutrient dense in fluoride, magnesium, potassium, calcium, folic acid, total phosphorus, vitamin C and omega 3 fatty acid.

The antioxidant and genetic diversity of phytochemical of 30 Brassicaceae microgreens were assessed by Xiao et al (2019). The total ascorbic acid concentration was ranged from 32.9 -120.8 mg per 100 g fresh weight. The total ascorbic acid content was 120.8mg/100 g of fresh weight in cauliflower microgreens. The highest ascorbic acid was 89.3 mg per 100 g of fresh weight in broccoli and cauliflower microgreens while savoy cabbage microgreens had high content of dehydroascorbic acid i.e. 40.8 mg per 100 g fresh weight. The rapini and broccoli microgreens had highest phylloquinone content with 3.7 µg/g FW and 3.3 µg/g FW, respectively. The highest total glucosinolate content was found in china rose radish i.e. 535.5 µmol/100 g of fresh weight. The high phenol content and DPPH radical scavenging capacity was found in ruby radish i.e. 811.2 mg GAE per 100 g fresh weight and 806.3 µmol TE/100 g 363 Fresh weight, respectively. The author reported significant variation of composition and concentration phytochemicals within the 30 Brassicaceae microgreens species.

Rocchetti et al (2020) examine the effect of storage and gastrointestinal digestion (*in vitro*) of amaranth and red beet microgreens on the untargeted metabolomics profile. The total phenolic content was increased by 1.3 times for red beet and 1.1 fold for amaranth microgreens, maximum incrase at 10^{th} day storage. The *in vitro* gastrointestinal digestion of both the microgreens considerably increased the total phenolic content by 36-88 percent and DPPH by 6-43 percent at 10 days of storage.

Microgreens are rich source of nutrients and bioactive compounds but *invitro* gastrointestinal digestion determines its bio accessibility values. Tomas et al (2021) investigated the *in vitro* bio accessibility and bioactive properties of polyphenols and glucosinolates from *Brassicaceae* microgreens. The total phenolic content of kale, red cabbage, kohlrabi and purple radish was significantly decreased after *in vitro* digestion. Radish had the highest ($p < 0.05$) total phenolic content with 872.5 mg GAE/100 g DW whereas, kohlrabi microgreens had lowest with (660.5 mg GAE/100 g DW) after *in vitro* digestion. The radish microgreens were characterized by the highest values of total phenolic content, total antioxidant capacity and total monomeric anthocyanins, while kohlrabi microgreens presented the lowest values after in vitro gastrointestinal digestion.

2.4 Microgreens consumption related to prevention of degenerative diseases

Red cabbage microgreens contained more polyphenols and glucosinolates than mature red cabbage. Huang *et al* (2016) claimed that Red Cabbage Microgreens lower liver Cholesterol, Circulating Low-Density Lipoprotein (LDL) and Inflammatory Cytokines in Mice Fed a High-Fat Diet. The study used 5 week old 60 mice and they were randomly assigned to six feeding groups: (1) low-fat diet; (2) high-fat diet; (3) low-fat diet + 1.09% red cabbage microgreens; (4) low-fat diet + 1.66% mature red cabbage; (5) high-fat diet + 1.09% red cabbage microgreens; (6) high-fat diet + 1.66% mature red cabbage. The animals were on their particular diets for 8 weeks. After 8 weeks they found that supplementation with microgreens significantly lowered circulating LDL levels in animals fed the high-fat diet and reduced hepatic cholesterol ester, triacylglycerol levels, and expression of inflammatory cytokines in the liver. These data suggest that microgreens contained bioactive compounds which were beneficial cholesterol metabolism, weight loss and may protect against cardiovascular disease. The author reported that with the supplementation of red cabbage microgreens there was significant reduced LDL level i.e. 34%, cholesterol ester with 65.5% and hepatic triglycerides by 23% in animal fed a high fat diet.

Wadhawan *et al* (2017) was reported anti diabetic potential of fenugreek microgreen and mint leaf extract during in vitro assays comprise cell line based analysis. Fenugreek microgreen extract showed anti – amylase activity. Fenugreek microgreen extract significantly inhibit porcine pancreatic alpha amylase and 2 mg mL^{-1} of FME inhibited alpha-amylase by 70%. The fenugreek microgreen extract act as insulinomimetic property. The FME (10 mg mL^{-1}) enhanced the glucose uptake by 25% in L6 cells and then improved to 44% in the existence of insulin. The mint leaf extract and fenugreek microgreen extract both inhibited the non-enzymatic glycation of protein. The fenugreek microgreen extract (2mg mL^{-1}) decreased non-enzymatic glycation of protein by 70%. Fenugreek microgreens extract had high antioxidant activity i.e. 83%, further the total phenolic content was 727 µg GAE g^{-1} in FME and also had high flavonoid content of 154 µg CE g^{-1}.

de la Fuente *et al* (2020) studied the antiproliferative effect of bio accessible fractions of broccoli, mustard, kale and radish microgreens on human colon cancer Caco-2 cells related to microgreens phytochemical composition. The bio accessible fractions were obtained using standardized static in vitro gastrointestinal digestion method. The non-tumoral and tumoral human colon cells were treated with microgreen bio accessible fractions for 24 hours. All the microgreens bio accessible fraction showed higher cell viability reduction i.e. 20-42% and highest in kale followed by radish microgreens. The cell viability was 10-12.8% in mitochondrial enzyme activity assay (MTT test) and 41.9 % in Trypan blue test. There was reduction in the proliferation of human tumoral cells due to high antioxidant bioactive compounds in the four *Brassicaceae* microgreens.

Renna *et al* (2018) produce lettuce and chicory microgreens using hydroponic system with low potassium levels for patients with impaired kidney disease. The reduction of potassium in the nutrient solution for the production of microgreens does not affect the yield and proximate composition of microgreens. The different levels of potassium used for nutrient solution were 0, 29.1, 58.4, and 117 mg L^{-1}. The potassium content of the microgreens was reduced by 50% with a potassium concentration ≥58.4 mg $^{-1}$ of nutrient solution. The microgreen produce using potassium concentration of ≥58.4 mg $^{-1}$ supply 225 to 250 mg of potassium per 100 grams of microgreens. The patients of impaired kidney disease are recommended with low potassium diet, thus microgreens produced from low potassium nutrient solution can be used without reducing the vegetable serving.

2.5 Microgreens shelf life and packaging

Allende *et al* (2004) evaluated the quality changes in minimally processed baby spinach leaves under modified atmospheric conditions. Packages prepared with the barrier film of an initial oxygen level at 21% accumulated carbon dioxide during storage exhibited a significant reduction in aerobic mesophilic bacterial growth compared to the perforated film package, also induced off-odor and loss of tissue integrity. The addition of super atmospheric oxygen to packages decrease tissue injury in addition to reducing microbial growth and was beneficial in maintaining quality of fresh cut baby spinach.

Xiao *et al* (2014b) studied the postharvest quality of radish microgreens. The 3 major postharvest treatment i.e. i) chlorine wash ii) storage temperature and iii) packing film OTR affect the quality and shelf life of radish microgreens. Shelf life, package atmosphere and product quality were affected by storage temperature. Initial microbial counts were reduced by 0.5 log cfu/g when treated with chlorine wash. The optimum temperature for the storage of radish microgreens was found to be 1°C without chill injury. Head space gas composition was affected by OTR packaging film.

Dalal *et al* (2020) evaluated the sensory properties of sunflower microgreens treated with different chemicals to increase the shelf life and changes in organoleptic quality with storage period. The treatment were ethanol vapour treatment, ascorbic acid spray, citric acid spray, citric acid+ ethanol and citric acid+ ascorbic acid spray against distilled water as a control for the duration period 0 day,4 days,8 days,12 days and 16 days. The sensory properties were significantly decreased with increase in storage time. The mean overall acceptability scores at 0 day storage was 8.28 and which progressively decreased to 6.288 at 16th day of storage of sunflower microgreens. The lowest average overall acceptability score (6.288 on 16th day of storage) was with the treatment citric acid and ascorbic acid spray during storage. The high scores of sensory properties was sunflower microgreen treated with citric acid spray and by the 12th day of storage period microgreens were under acceptable scores for all the treatments.

Similar study on the quality of Chinese cabbage microgreens treated with different chemicals and packaging films and changes in microbial population was carried out by Chandra *et al* (2012). The microgreens was treated with chlorinated water (CW), citric acid + ascorbic acid (CA+AA) ,citric acid+ethanol (CA+E) and the packaging films were polypropylene (PP) and polyethylene (PE) stored for 9 days of storage at 5˚C. The sensory quality and production of off odour was with the samples treated with chlorine, CA+AA, CA+E and water washed samples. The polypropylene film packaging maintained the quality of microgreen till 5 days of storage. The microbial population and quality of microgreens, it was found that citric acid + ethanol appeared to be the best method to sanitize the microgreens to replace chlorine.

Polash *et al* (2018) studied the retention of bio active compounds and nutrients of microgreens after harvest to optimize best period to consume microgreens after harvesting. After harvesting the microgreens were stored in air tight polybag at 4-6˚C in the refrigerator. The chlorophyll content of microgreens at 1 day of harvest was highest when compared to the 5th day of harvest. The total chlorophyll content of mustard, leaf mustard, radish and cabbage was 8.22, 10.28, 7.62 and 7.63 mg/100g, respectively which reduced to 6.77, 6.67, 5.66 and 5.64 mg/100 g on 5th day of storage. The beta carotene content of mustard, leaf mustard, radish and cabbage was 2.41, 2.88, 2.11 and 2.06 mg/100g, respectively at 1st day and when compared the percentage reduction on 5th day it was 22.40, 49.65, 28.91 and 16.50%. The reduction percentage of lycopene was 14.17, 41.03, 30.55 and 13.51% in mustard, leaf mustard, radish and cabbage, respectively on 5th day of harvest as compared to 1st day of harvest. The reduction of vitamin c content was 31.05, 31.13, 44.34 and 41.84 percent on 5th day in mustard, leaf mustard, radish and cabbage, respectively whereas the DPPH radical scavenging showed maximum reduction i.e. 136, 141.67, 110.35 and 98.88% on 5th day in mustard, leaf mustard, radish and cabbage when compared to the 1st day of harvest.

A comparative study of fenugreek microgreens and mature leaves on the phytochemical, antioxidant activity and sensory quality on storage studied by Ghoora and Srividya (2017). The phytochemical and antioxidant activity was assessed for storage period of 14 days at the temperature 10 °C. The analysis was analyzed on 0 day, 7th day and 14 th day of storage. The fenugreek microgreens had greater retention of the nutrients than their mature counterparts on storage. The ascorbic acid reduction on storage was 17% on 7^{th} day and 25% on 14^{th} day of storage for fenugreek microgreens whereas mature fenugreek leaves had high reduction i.e. 35% and 72% were noted on the 7th and 14th day of storage, respectively. The reduction of total phenols was high during storage i.e. 51% on 7^{th} day and 68% on 14^{th} day in fenugreek microgreens while 47% on 7^{th} day and 71% on 14^{th} day of storage in mature fenugreek leaves. The antioxidant activity was higher in fenugreek microgreens as compared to their mature leaves. Fenugreek microgreens were supplemented to the traditional recipes, which showed high acceptability with highest in methi aloo curry i.e. 8.64 followed by methi pulao (8.54). The fenugreek microgreens (80%) retained better aroma, texture, freshness, colour and wiltness while fenugreek mature leaves retained only 40% acceptable sensory properties.

CHAPTER III

MATERIAL AND METHODS

The present research study was designed to optimize germination, cultivation and harvesting conditions of microgreens to attain highest nutrient content at different growth period. The most significant method of different processing conditions was carried out to determine maximum nutrient and bioactive compound retention in microgreens. Further, microgreens supplemented health products were developed from optimized production and processing condition and evaluated for organoleptic quality. The material and methods utilized for conducting research study have been discoursed under the following sub-headings.

3.1. Procurement of vegetable seeds

3.2. Optimize different cultivation and harvesting conditions

- 3.2.1. Germination condition
- 3.2.2. Cultivation and harvesting condition

3.3. Comparison of microgreen with sprouts and mature leaves in term of nutritional and biochemical composition

3.4. Optimize processing condition

- 3.4.1. Tray drying
- 3.4.2. Shade drying
- 3.4.3. Microwave drying

3.5. Nutritional and biochemical evaluation

- 3.5.1. Ascorbic acid
- 3.5.2. Beta-carotene
- 3.5.3. Chlorophyll
- 3.5.4. Flavonoids
- 3.5.5. Total phenols
- 3.5.6. Minerals
- 3.5.7. Antioxidant activity by DPPH method
- 3.5.8 Estimation of percent adequacy of microgreens
- 3.5.9 Nutrient quality Score

3.6. Formulation of health products supplemented with microgreens

3.7. Organoleptic and nutritional evaluation of health products

3.8. Statistical analysis

3.1. Procurement of vegetable seeds

Two commonly consumed vegetable greens seeds: Spinach (*Spinacia oleraceae* L. var. Punjab Green) & Coriander (*Coriandrum sativum* L. var. Punjab Sugandh) and two underutilized vegetable greens seeds i.e. Broccoli (*Brassica oleracea* L. var. Palam Samriddhi) & Carrot (*Daucus carota* L. var. Punjab Carrot Red) were procured from Department of Vegetable Science, PAU, Ludhiana.

3.2.1. Germination condition

The vegetable seeds were separated from the chaff (the seed casing and depris) and any infested seeds were removed. The half samples of microgreen seeds were soaked in distilled water for 12 hours at room temperature. The soaked seeds were rinsed thoroughly with distilled water and seeds were spread on paper towel.

3.2.2. Cultivation and harvesting condition

The microgreen seeds were cultivated under indoor and outdoor setting (Plate 1). The soil media was same for both the cultivation. The soil beds were prepared in field and under shade. For the microgreens cultivation no manure and fertilizer were used. The seeds were evenly spread over the soil and moist with water. Further, a thin layer of the soil sprinkled over the seeds to cover them. The soil beds were watered with tap water twice a day. The microgreens were harvested on two growing stages i.e. 10^{th} and 20^{th} day, after germination. The microgreens were harvested just above the soil line by cutting the cotyledon stems with ethanol cleaned scissor. The harvested biomass was cleaned for any wiltness and rinsed under tap water. The fresh microgreens placed in paper towel to absorb extra moisture and then samples were taken for nutritional and biochemical investigation.

3.3. Optimise processing condition

Microgreens powder preparation-The best variants of microgreens from ‘cultivation and harvesting condition’ will be dried by three methods viz. microwave, shade and tray drying to optimize the processing techniques for best nutritional and biochemical composition.

3.3.1. Microwave Drying

The microgreens were placed in a glass tray. The microgreens were dried to a constant weight in microwaves for 8-10 minutes at 1020 watt. The dried microgreen samples were ground into a fine powder with the help of grinder and packed in airtight high-density polyethene (HDPE) pouches. The samples were stored in a deep freezer at -18˚C until further use.

3.3.2. Shade Drying

The microgreens were spread over butter paper and placed in a tray. The microgreens were dried till constant weight under fan at room temperature. The dried samples were ground

Indoor

Outdoor

Plate 1: Indoor and outdoor cultivation of microgreens

into a fine powder with the help of grinder and packed in airtight high-density polyethene (HDPE) pouches. The samples were stored in a deep freezer at -18˚C until further use.

3.3.3 Tray Drying

A pilot scale convective tray dryer was used for drying. The microgreens were spread over butter paper in a single layer and placed over tray. The microgreens were dried to a constant weight in an oven at 50˚C for 8 hours. The fan speed was set to 850rpm and an air velocity to 230m/minute. The dried samples were ground into a fine powder with the help of grinder and packed in airtight high-density polyethene (HDPE) pouches. The samples were stored in a deep freezer at -18˚C until further use.

3.4 Comparison of microgreen with sprouts and mature leaves in term of nutritional and biochemical composition

The comparison between the different growth stages of the vegetable was carried out in terms of minerals (calcium, iron, magnesium, sodium, potassium and zinc), beta-carotene, vitamin-C, antioxidant activity, chlorophyll, total phenolic content and total flavonoids.

3.5 Nutritional and biochemical evaluation

3.5.1 Estimation of Ascorbic Acid (AOAC 2002)

Principle

The reduction of 2, 6-dichlorophenolindophenol dye by ascorbic acid makes a product blue in colour which is estimated colorimetrically.

Reagents

1. Acetate buffer, pH 4.0: 300 g of anhydrous sodium acetate dissolved in 700 ml of water and 100 ml of glacial acetic acid added to it.
2. 2,6-dichlorophenolindophenol dye solution: 25 mg of the sodium salt of 2,6-dichlorophenolindophenol dissolved in distilled water and marked up to 200 ml.
3. Metaphosphoric acid (HPO_3) 6%: 6% of metaphosphoric acid dissolved in 1 L of distilled water.
4. Ascorbic acid standard (1 mg/ml): 100 mg of pure ascorbic acid dissolved in 100 ml of 6% metaphosphoric acid.
5. Xylene

Procedure

For the procedure, two g of microgreen powder was weighted in an analytical weighing balance and placed in a mortar and pestle. To this 20 ml of 6% of metaphosphoric acid was added slowly and the sample was ground to a slurry. The slurry was filtered through Whatsman No. 1 filter paper. 30ml of metaphosphoric acid was added to the residue and filtered again. The volume was made up to 50 ml using 6% of metaphosphoric acid. Three Separating funnel (50 ml) were fixed and labelled as A, B and C. Where A funnel was for sample, B for dye and C for standard. In separating funnel A, 5 ml of filtrate was pipetted and in funnel C, 0.1 ml of standard ascorbic acid solution was pipetted. 5 ml acetate buffer was added to all three funnels followed by 2 ml of dye solution. 10 ml of xylene solution was quickly added and the contents were shaken for 5 to 10 seconds. The contents were allowed to separate into two layers. The bottom layer was discarded. The xylene layer was transferred into a test tube and the optical density was read in a spectrophotometer at wavelength 500 nm.

Calculation

$$X \text{ mg} = 0.1 (b-a)/(b-c)$$

Where b= OD of blank

a=OD of sample

c= OD of standard

3.5.2 Estimation of β-carotene (Ranganna 2002)

Reagents

1. Potassium hydroxide: prepared 60% KOH in distilled water and combined with absolute alcohol in the ratio 1:5
2. Ethanol
3. Calcium carbonate
4. Petroleum ether (60˚C -80˚C)
5. 3% acetone in petroleum ether
6. Calcium hydroxide or aluminium oxide (active)
7. Anhydrous sodium sulphate
8. Glass wool

Procedure

Five grams of powder sample was ground and mixed with 25 ml of KOH (12%) and allowed to stand overnight for saponification. To this, a spatula of calcium carbonate was added and vortexed. 30 ml of petroleum ether was added slowly and vortexed for another five minutes. The petroleum ether layer was allowed to separate and the top layer was removed into a separating funnel. 50 ml water was added in the mixture, shaken and the water layer was discarded. Washing was repeated till the solution was free from KOH. The petroleum ether extract was filtered through a funnel plugged with the glass wool. Anhydrous sodium sulphate was added in the funnel to remove excess water. The filtered ether extract was pulled together and the volume was measured. 10-50 ml of ether extract was taken and concentrated to 5 ml.

The glass column (30×1 cm) was packed with 6 g of activated alumina and 2 g of anhydrous sodium sulphate on top of the column under light suction.

With the help of gentle suction, the column was charged with 5 ml of extract (containing about 40-100 µg of the total carotene). 3% acetone in petroleum ether was added to the column which resulted in the formation of colour bands. After the band dropped 10 cm from the bottom of the column 3ml of each fraction was collected in test tubes. Eluted individually separated bands were studied for their spectra spectrophotometrically at 450 nm.

Calculation

1 OD is equivalent to 4 µg/ml of β-carotene when measured in 1 cm cell. The sum of the values of all the fractions present in the chromatogram of particular foodstuff gave its β-carotene value.

Amount charged on the column = Total volume $\times \frac{OD}{ml} \times 4$ µg (A)

Total OD× 3 ml ×4 µg = β-carotene eluted (B)

For A (total carotenoids) units= β units of β-carotene eluted

3.5.3 Chlorophyll (Thimmaiah 1999)

Reagent

80% aqueous acetone (pre chilled)

Method

The chlorophyll concentration was evaluated using direct spectrophotometric method followed by extraction**.** About 1 g of each finely chopped microgreen was grinded with 20 ml of chilled 80 % chilled aqueous acetone solution. The sample was homogenised for 5 minutes at a rate of 5000 rate per minute. The resulting mixture was filtered through Whatman filter paper and shifted into a 100 ml volumetric flask. The residue was rinsed with another 20 ml of 80 % chilled aqueous acetone solution, homogenised and shifted the supernatant. The process was performed again and again until the residue was left colourless. All the filterate were collected into a volumetric flask and the volume was made to 100 ml with 80% chilled aqueous acetone. The absorbance was read at 645,663 and 652 nm using a spectrophotometer. The 80 % acetone was utilised as the blank. Chlorophyll concentration was calculated using the following equations.

Calculation

Chlorophyll concentration present in samples was calculated using the following equations.

$$\text{mg of chlorophyll } \frac{a}{g} \text{ tissue} = 12.7(A_{663}) - 2.69(A_{645}) \times \frac{V}{1000 \times W}$$

$$\text{mg of chlorophyll } \frac{a}{g} \text{ tissue} = 22.9\ (A_{645}) - 4.29\ (A_{663}) \times \frac{V}{1000 \times W}$$

$$\text{mg total chlorophyll (per g tissue)} = 20.2(A_{645}) - 8.02\ (A_{663}) \times \frac{V}{1000 \times W}$$

Where,

A indicate absorbance at specific wavelength

V indicate final volume of the chlorophyll extract in 80% acetone

W indicate fresh weight of tissue extracted

3.5.4 Estimation of total flavonoids (Mathur and Vijayvergia 2017)

Principle

The C-4 keto group and either the C-3 or C-5 hydroxyl group of flavones and flavonols from acid-stable complexes with aluminium chloride. Besides, it forms acid-labile complexes with the orthodihydroxyl groups in the A- and B- ring of flavonoids.

Reagents

1. Methanol solution of the extract
2. 10% Aluminium chloride (AlCl3) solution in methanol
3. 5% sodium nitrite (NaNO2)
4. 4% sodium hydroxide (NaOH)
5. Quercetin
6. Methanol

Procedure

0.5 ml of sample extract was taken in a test tube, to this, 2 ml distilled water and 150 µl of sodium nitrite were added. After 6 min, 150 µl of 10% aluminium chloride was added. Later again after 6 min, 2 ml of 4% sodium hydroxide was added. The tubes were allowed to stand for 15 minutes and absorbance of the mixture was read at 510 nm against the blank. The blank was prepared from reagents and distilled water only. The total flavonoids content in samples was expressed as mg QE (Quercetin Equivalent)/ /100g for powder samples.

Standard curve

Quercetin was used to make the calibration curve. 10 mg of quercetin was dissolved in 100 ml of 80% methanol (100µg/ml) and then diluted to 20, 40, 60, 80 and 100 µg/ml using methanol. The same procedure was followed using different concentration of standard. The standard curve was plotted and the corresponding concentration was determined as the concentration of flavonoids (Fig. 3.1)

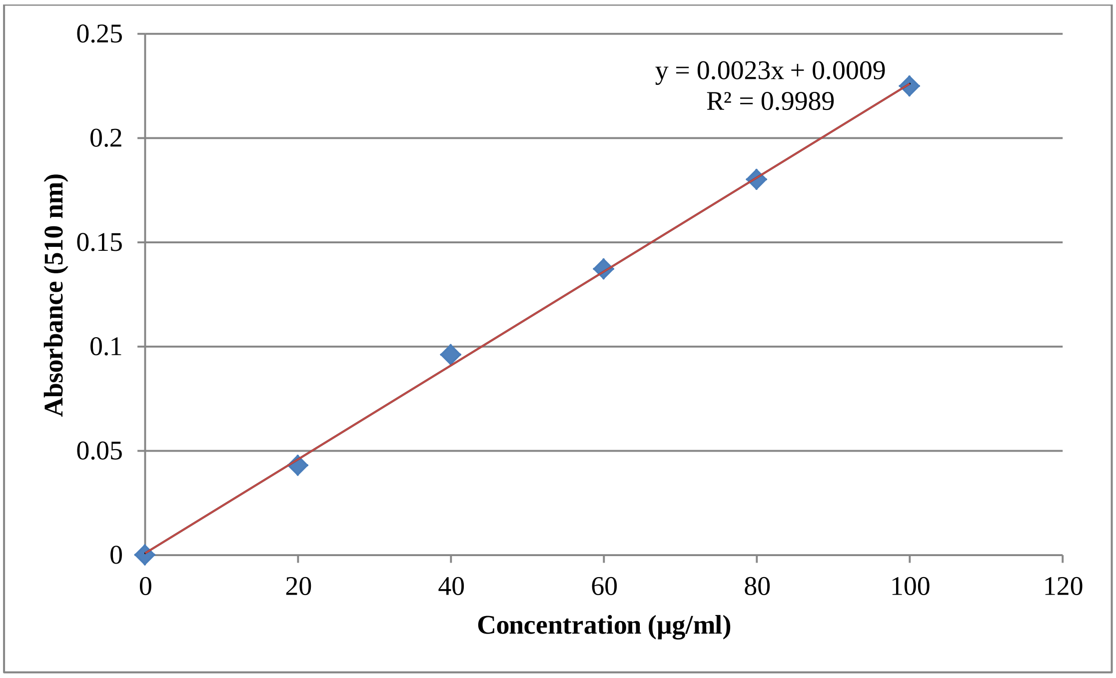

Fig 3.1 Standard curve for Total Flavonoid Content (TFC)

3.5.5 Estimation of Total Phenolic Content (TPC) (Mathur and Vijayvergia 2017)

Reagents

1. Methanol solution of an extract
2. 10% of Folin-Ciocalteu's reagent in distilled water
3. Saturated sodium carbonate
4. Gallic acid
5. Methanol

Procedure

Methanolic extract (0.2 ml) was taken in a test tube. To this, 5 ml Folin-Ciocalteu's reagent was added and shaken thoroughly. After 5 minutes, 2 ml of saturated sodium carbonate was added. The blue colour developed was read after 1 hour at 760 nm against the blank. The blank was prepared from reagents and distilled water only. The total phenols in the sample were expressed as mg GAE (Gallic Acid Equivalent) /100g for powder sample.

Standard curve

Gallic acid was used to make the calibration curve**.** 10 mg of gallic acid was dissolved in 100 ml of 80% methanol (100µg/ml) and then diluted to 20, 40, 60, 80 and 100 µg/ml using methanol. The same procedure was followed using different concentration of standard. The standard curve was plotted and the corresponding concentration was determined as the concentration of total phenols (Fig. 3.2)

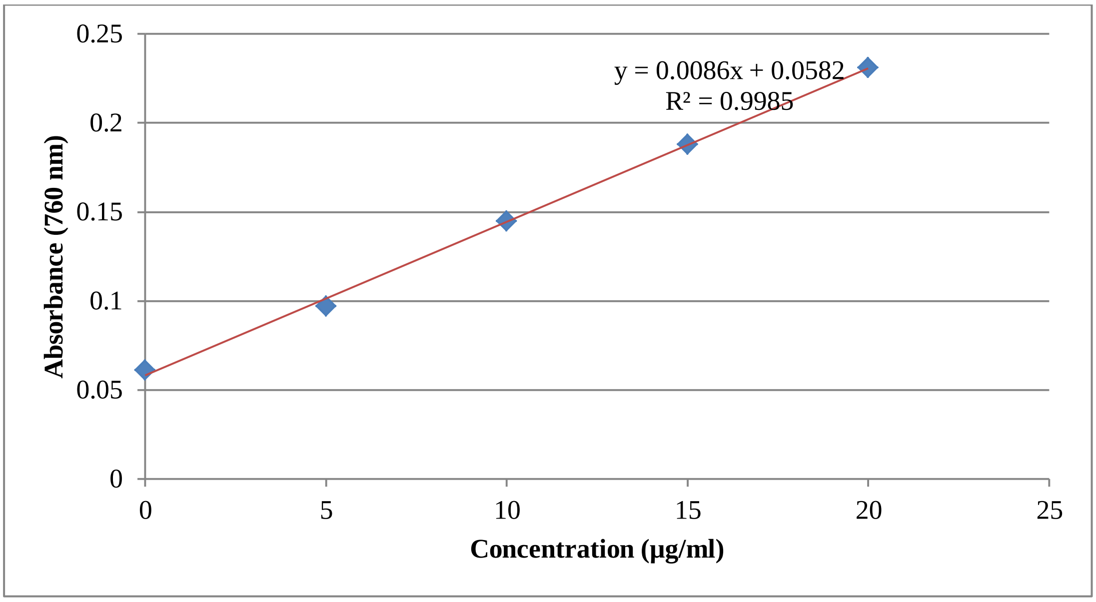

Fig 3.2 Standard curve for total phenolic content (Absorbance 760 nm)

3.5.6 Estimation of Minerals

Elements namely iron, calcium, zinc, magnesium, potassium and sodium were estimated using inductively coupled plasma-optical emission spectrometry (ICP-OES), after wet digestion (AOAC 2002).

Decontamination of the equipment

All the plastic bottles and glassware required for mineral estimation were washed, cleaned, and were soaked overnight in 10 per cent concentrated HCl followed by thorough rinsing with deionised water followed by drying and marking.

Reagents

The triacid mixture was used for digesting the juice sample consisting of nitric acid (AR), perchloric acid and sulphuric acid in the ratio 9:3:1 respectively. The freshly prepared triacid mixture was used every time analysis was conducted.

Procedure

Weighed 0.5 g sample of microgreens and added in 250 ml conical flask. To this 25 ml of the triacid mixture was added. The contents were kept overnight on a stand for slow digestion and then next day was heated at low temperature on a hot plate till the content remained was about 1 ml clear and colourless liquid. Later remaining contents were allowed to cool and then transferred into with 50 ml volumetric flask with repeated washing with deionized water and volume was made up to the mark. The digests were filtered through Whatsman No. 42 filter paper and stored in the dried, decontaminated, and labelled airtight polythene bottles for mineral determination by ICP-OES. For blank 25 ml of the triacid mixture was digested the same as samples and volume was made to 50 ml with deionized water.

Standard Curve

Standard solution of each element was used to make the standard graph. The solutions of 100 ppm concentration of each mineral were prepared. These were diluted to various concentrations with distilled water,1 ml of concentrated sulphuric acid was added and volume was made to 50 ml. The absorbance was recorded in the form of standard curve by the automated recorder in the ICP-OES. The concentration of the samples was also recorded automatically.

$$\text{Mineral content} = \frac{\text{Sample conc (ppm)} - \text{Blank conc (ppm)}}{\text{Weight of sample (g or ml)}} * \text{total dilution}$$

Mineral content is expressed as mg/100 g on dry matter basis of microgreen powder and mg.

3.5.7. Antioxidant activity (DPPH method by Dehshahri et al 2012)

Reagent

- 80% Methanol
- DPPH solution (0.1nM): 0.039 g of DPPH was dissolved in 100 ml methanol solution. 5ml of this solution was again diluted to 100 ml of methanol.

Procedure

2 grams of powdered microgreens sample was extracted with 20 ml of 80% methanol by vortex mixer for two hours. The Extraction procedure was repeated twice. The extract was pulled together and centrifuged at 10000 rpm for 15 minutes. The supernatant was stored at 20˚C till analysis. 100 µl of the aliquot of extract was taken in test tubes and added 2.9 ml of 0.1 mM DPPH reagent. The mixture was vortexed for 1 minute and incubated in dark for 30 minutes at room temperature. When DPPH reacts with an antioxidant compound, which can donate hydrogen, it is reduced. Discolouration (change in colour from deep violet to light yellow) of DPPH was measured against blank at 517nm.

Calculation

The DPPH scavenging effect was measured using the following formula

$$\textbf{\% DPPH inhibition} = \frac{A_B - A_A}{A_B} \times 100$$

Where

A_A = Absorbance of sample

A_B = Absorbance of blank

3.5.8 Estimation of percent adequacy of microgreens

The percent adequacy was calculated using Estimated Average Requirement (ICMR, NIN 2020) of the nutrients (calcium, zinc, iron, magnesium, potassium, vitamin C and β-carotene) against nutrient content of broccoli microgreens determined in the present study. The equation used to compute percent adequacy is presented below:

$$\textbf{Percent Adequacy (\%)} = \left(\frac{\text{Nutrient content of microgreens (mg/100g)}}{\text{EAR of the nutrient } (\frac{\text{mg}}{\text{day}})}\right) \times 100$$

3.5.9 Nutrient Quality Score

The NQS 7.1 was computed based on the sum of percent adequacy for 7 nutrients to encourage (calcium, zinc, iron, magnesium, potassium, vitamin C and β-carotene) minus the sum of percent adequacy of sodium (the nutrient to be limited). The calculation of NQS 7.1 was composed of the Estimated Average Requirements (EAR) for adult Indians with some modifications according to Ghoora et al (2020). The percent adequacy of the nutrients to encourage was capped at 100 % so that contribution of single nutrient would not result in disproportionately higher nutrient score. The percent adequacy of each nutrient, per serving size (100g), was calculated by referring to the data from ICMR-NIN (2020). The equation used to calculate NQS is reported below:

$$\text{NQS} = \Sigma_{1\text{–}7}\ (\text{percent adequacy}_i) - \text{percent adequacy}_{\text{Na}}$$

3.6 Formulation of health products supplemented with microgreens

Various health foods were developed from the best variant in terms of nutritional and biochemical composition of microgreens (Fresh, Juice and Powder).

The ingredients used in the formation of health products were wheat flour, dal, raw vegetables, fruit juice, sugar, spices, milk and additives purchased from the local market of Ludhiana city. The microgreens used for the supplementation of health products were cultivated and harvested at Punjab Agricultural University, Ludhiana.

Seven products were chosen for the formulation of health products from the microgreens namely, *salad, missi roti, dal, cooked vegetable, juice, flavoured milk* and *sprinkler*. For the formation of traditional recipes fresh raw microgreens were supplemented in appropriate ratio's in yellow split moong dal, missi roti and aloo gobhi vegetable. The fresh microgreens were added to Russian salad and tofu salad in different ratios. The different ratios of microgreens juice was blended with orange juice. The microgreen juice was prepared with water incorporation in the ratio of 1:1 of microgreens and water. The different ratio of microgreen juice was supplemented to the flavoured milk.

The *missi roti* was supplemented with 15%, 20% and 25%. The *dal and cooked veg* was enriched with 2.5%, 5%, 7.5% and 10% of microgreens. The *flavoured milk* was prepared by incorporation of microgreen juice with 20%, 30%, 40% and 50%. The different blends were prepared with orange juice and microgreen juice with 20%, 30%, 40% and 50%

supplementation. The *russian and tofu salad* was incorporated with 20%, 30% and 40% of microgreens. The *sprinkler* was formulated by mixing various spices enriched with 30% and 40% of microgreens powder. The microgreens used in the food products of high consumption frequency so, that consumer will be benefitted maximum of microgreen. The detailed product development has been shown in Table 3.1 to 3.5 and the recipes of preparations in Annexure II.

3.7 Organoleptic and nutritional evaluation of health products

The developed health products were evaluated for the sensory properties by a panel of semi trained judges (n=10) from the Department of Food and Nutrition, Punjab Agricultural University, Ludhiana. The products were evaluated for appearance, consistency, colour, aroma, taste and overall acceptability on a nine-point hedonic scale (Wichchukit and O'Mahony 2015). The judges were asked to score the products using a scale where 9 indicated "like extremely" and 1 indicated "dislike extremely". Score sheet used for the evaluation of products is given in Annexure-I. Each panelist was provided with enough privacy to avoid biased assessment. The most acceptable health product further nutritionally evaluated.

3.8 Statistical analysis

The data was tabulated keeping in view the objectives of the study. For data analysis, JAM 10.0.1 software was used. Analysis of variance (ANOVA) was done for all the parameters. Tukey-Post Hoc test was applied for determining the significant difference. Paired t-test was applied for experimental and control samples of health foods. Data from this study was reported as mean±SD for at least three replicates for each sample in product analysis. The significant difference was defined as $p \leq 0.05$ and $p \leq 0.001$.

Table 3.1 Standardized recipe supplemented missi roti with fresh microgreens

Recipe	Ingredients (g)	Control	15%	20%	25%
Missi Roti	Wheat flour	30	25.5	24	22.5
	Besan	30	25.5	24	22.5
	Methi	30	25.5	24	22.5
	Coriander	5	4.25	4	3.75
	Green chilles	5	4.25	4	3.75
	Microgreens	-	15	20	25

Table 3.2 Standardized recipes supplemented dal and cooked vegetable with fresh microgreens

Recipe	Ingredients (g)	Control	2.5%	5%	7.5%	10%
Dal	Split moong dal	30	29.25	28.5	27.75	27
	Onion	25	24.37	23.75	23.12	22.5
	Tomato	25	24.37	23.75	23.12	22.5
	Garlic	10	9.75	9.5	9.25	9
	Green chillies	10	9.75	9.5	9.25	9
	Microgreens	-	2.5	5	7.5	10
Cooked veg	Potato	30g	29.25	28.5	27.75	27
	Cauliflower	30g	29.25	28.5	27.75	27
	Onion	10g	9.75	9.5	9.25	9
	Tomato	10g	9.75	9.5	9.25	9
	Microgreens		2	4	6	8

Table 3.3 Standardized recipes supplemented fruit blend and flavoured milk with fresh microgreen juice

Recipe	Ingredients (ml)	Control	20%	30%	40%	50%
Fruit blend	Orange juice	100	80	70	60	50
	Microgreen Juice	-	20	30	40	50
Flavoured milk	Milk	100	80	70	60	50
	Cardamom powder	5g	5g	5g	5g	5g
	Cardamom essence	2 drops	2drops	2drops	2drops	2drops
	Sugar	10g	10g	10g	10g	10g
	Microgreens juice	-	20	30	40	50

Table 3.4 Standardized recipe supplemented *food sprinkler* with powdered microgreen

Recipe	Ingredients (g)	Control	30%	40%
Sprinkler	Coriander seeds powder	15	10.5	9
	Mango powder	10	7	6
	Anardana powder	10	7	6
	Pink salt	5	3.5	3
	Table salt	5	3.5	3
	Black pepper	10	7	6
	Microgreen powder	-	16.5	22

Table 3.5 Standardized recipes supplemented salad with fresh microgreens

Recipe	Ingredients	Control	20%	30%	40%
Russian salad	Tomato	15g	12	10.5	9
	Potato	20g	16	14	12
	Peas	10g	8	7	6
	Beans	15g	12	10.5	9
	Cucumber	15g	12	10.5	9
	Capsicum	10g	8	7	6
	Hung curd	15g	12	10.5	9
	Salt	3g	3	3	3
	Black pepper	3g	3	3	3
	Microgreens	-	20	30	40
Tofu salad	Tofu	20g	16	14	12
	Tomato	20g	16	14	12
	Cucumber	20g	16	14	12
	Onion	20g	16	14	12
	Salt	3g	3	3	3
	Black pepper	3g	3	3	3
	Microgreens	-	16	24	32

CHAPTER – IV

RESULTS AND DISCUSSION

Microgreens are potential source of vitamins, minerals and bioactive compounds but the nutrient content is affected by their cultivation and harvesting condition. Considering this, the present study was carried to optimise the cultivation, harvesting and processing of microgreens in terms of nutritional and biochemical parameters. The best variant of microgreens further supplemented in various traditional recipes and sensory quality was assessed. The results of the study have been discussed under following sub-headings.

4.1 Nutritional and biochemical composition of microgreens under different cultivation and harvesting condition.

4.2 Nutritional and biochemical composition of microgreens under different processing conditions.

4.3 Compare microgreens with sprouts and mature leaves in term of nutritional and biochemical composition

4.4 Development of health foods from microgreens obtained from optimized production and processing conditions.

4.5 Organoleptic & nutritional quality of various health foods supplemented with microgreens.

4.1 Nutritional and biochemical composition of microgreens under different cultivation and harvesting conditions.

4.1.1 Broccoli

The β-carotene content of broccoli microgreens under different cultivation and harvesting conditions is presented in Table 4.1 on dry matter basis. The β-carotene content of broccoli microgreens in the present study is in the range of 272.20 to 698.97 µg/100g DW. Broccoli microgreens cultivated indoor with soaked seeds and harvested at 10th day had high level of β-carotene i.e. 698.97 µg /100 g DW. Just 100 g of broccoli microgreens were fulfilling the daily requirements of β-carotene by 12.49% and 14.73 % for an adult male and female, respectively (Table 4.3). Broccoli microgreens compared with the mature counterparts were found to be high in β-carotene content. Pandey *et al* (2018) reported the β-carotene content of mature broccoli as 34.12 µg /100 g DW which is significantly lower than the broccoli microgreens reported in the present research. The underutilized leaves of mature broccoli had low β-carotene content as compared to the tiny broccoli microgreens. Liu *et al* (2018) reported that mature broccoli leaves had 248.4 µg /100 g DW of β-carotene content whereas broccoli microgreens had 698.97 µg /100 g DW, which are 2.81 folds more than the mature leaves. Polash *et al* (2019) reported the high content of β-carotene (0.24mg/100g of FW) was found in mustard microgreens as compared to mature one, but the value was lower than β-carotene of broccoli microgreens i.e. 698.97 µg /100 g DW. These finding suggest that broccoli microgreens possess high β-carotene concentrations than the mature counterparts.

Table 4.1 Vitamin, bioactive content and antioxidant activity of broccoli microgreens

Position	Seeds	10th day harvesting	20th day harvesting
β- carotene (µg /100 g)			
Indoor	Soaked	698.97±0.61	521.13±0.31
	Unsoaked	696.74±1.22	562.41±9.84
Outdoor	Soaked	344.75±0.51	335.27±2.31
	Unsoaked	303.13±0.31	272.20±0.30
Vitamin C (mg/100g)			
Indoor	Soaked	25.28±0.22	15.29±0.30
	Unsoaked	23.01±0.51	15.63±0.24
Outdoor	Soaked	21.69±0.36	15.63±0.24
	Unsoaked	21.48±0.18	13.69±0.36
Chlorophyll (g /100 g)			
Indoor	Soaked	3.61±0.03	2.87±0.05
	Unsoaked	3.47±0.02	3.51±0.05
Outdoor	Soaked	3.90±0.12	3.32±0.03
	Unsoaked	3.82±0.01	3.63±0.12
Total Phenolic content (mg GAE/100g)			
Indoor	Soaked	2518.5±28.46	2161.34±6.29
	Unsoaked	2513.26±1.93	2015.67±1.35
Outdoor	Soaked	2594.78±3.88	2135.40±0.69
	Unsoaked	2521.35±3.04	2106.98±5.49
Total Flavonoids (mg QE/100g)			
Indoor	Soaked	341.64±0.88	283.14±2.74
	Unsoaked	334.39±3.64	249.37±1.01
Outdoor	Soaked	382.32±2.21	241.33±0.55
	Unsoaked	304.40±0.84	224.51±1.26
Antioxidant activity (%)			
Indoor	Soaked	80.62±0.05	71.99±1.64
	Unsoaked	79.53±0.65	64.76±0.10
Outdoor	Soaked	81.90±00	60.94±1.04
	Unsoaked	81.53±00	67.56±1.78

Values are expressed as mean ±SD

The ascorbic acid content of broccoli microgreens cultivated and harvested at different location and duration respectively is in the range of 13.69 to 25.28 mg/100g DW. The ascorbic acid is significantly ($p<0.01$) higher i.e. 25.28 mg/100g of dry weight when cultivated indoor with soaked seeds and harvested at 10^{th} day (Table 4.1). The ascorbic acid is water soluble vitamin and must be supplied daily by the diet. The broccoli microgreens of 100g were fulfilling the daily requirements of ascorbic acid by 33.36% and 39.43 % for an adult male and female, respectively presented in Table 4.3. Tan *et al* (2020) reported the ascorbic acid content of broccoli microgreens samples were in the range of 0.33–0.56 mg per g, which is less than the present study results. The results of ascorbic acid content were high in recent published research data for microgreens of broccoli (89 mg per 100 g FW), kale (28–66 mg per 100 g FW), mustard (19–44 mg per 100 g FW) and radish (25–68 mg per 100 g FW) than our present data i.e. 25.28 mg per 100g DW (Kyriacou *et al* 2019, Xiao *et al* 2019 and Xiao *et al* 2015). Singh *et al* (2007) reported the ascorbic acid content of mature broccoli ranged from 25.5 to 82.3 mg per 100 g FW, which is high than the present study results as ascorbic acid being heat liable.

The bioactive component and antioxidant activity is presented in Table 4.1. The chlorophyll content of broccoli microgreens cultivated at different locations and harvested at different growth stages is in range from 2.87 to 3.90 g/100g DW. A significantly ($p<0.01$) higher chlorophyll content (3.90 g/100g DW) is observed in broccoli microgreens cultivated with soaked seeds at outdoor setting and harvested at 10^{th} day. The chlorophyll content of broccoli microgreens was influenced by light condition (sunlight) in the present study which showed high content of chlorophyll when cultivated outdoor (3.90 g/100g) than indoor (3.61g/100g). In the same manner, Andrejiová *et al* (2017) reported the chlorophyll content of broccoli microgreens 45.84g/100g and after supplementation of artificial light the content of chlorophyll increased i.e. 76.97g/100g which showed the influence of light on the chlorophyll content. Tan *et al* (2020) reported lower chlorophyll content i.e. 0.33 mg/g of broccoli microgreens, when compared with the present study. It was reported that mature broccoli contained about 0.02 mg/g of chlorophyll by Bohn *et al* (2004), while present study showed that chlorophyll in broccoli microgreens had 3.90 g/100 g, which shows higher chlorophyll content in broccoli microgreens than the mature broccoli. Similar results was found by Pandey *et al* (2018) which showed that mature broccoli contained 0.84g/100g of chlorophyll where broccoli microgreens range from 2.91 to 3.90 g/100 g in present study.

The average total phenolic content in broccoli microgreens in present study is in the range from 2015.67 to 2594.78mg GAE/100g DW. The broccoli microgreens had higher content of total phenols i.e. 2594.78 mg GAE/100g DW, when cultivated with soaked seeds at outdoor setting and harvested at 10th day (Table 4.1). Tan *et al* (2020) reported the TPC in broccoli microgreen range from 10.71 to 11.88 mg GAE/g, which is quite less than the present results. The total phenolic content (TPC) of present results was higher than the mature vegetables which are known to be excellent source of phenolic compounds reported by Li *et al* (2018) of broccoli, brussel sprout and kalian which ranges from 0.16 to 1.93 mg GAE/g FW. The previous studies (Xiao *et al* 2014 and Tan *et al* 2020) showed that the total phenolic content of microgreens did not get affected with the light and harvesting conditions whereas in the present study TPC was higher when cultivated outdoor and harvested at 10th day. The underutilized mature broccoli leaves had 15.2 mg/g DW of total phenolic content reported by Liu *et al* (2018) which is significantly less than the broccoli microgreens in the current study (2594.78 mg GAE/100g). Kaur and Kapoor (2002) reported mature broccoli had 87.5 mg/100g of total phenolic content whereas Hwang *et al* (2015) reported slight higher content of total phenolic i.e. 335.3 mg GAE/100 g DW of mature broccoli and 537.6 mg GAE/100 g DW of broccoli mature leaf, which is significantly lower than the broccoli microgreens.

The total flavonoid content of broccoli microgreens cultivated at different positions and harvested at different durations is at a range from 224.51 to 382.32 mg QE/100g DW (Table 4.1). The results showed that flavonoid content of broccoli microgreens cultivated with soaked seeds at outdoor settings were significantly higher ($p < 0.01$) when harvested at 10th day with value of 382.32 mg QE/100g DW. Thomas *et al* (2018) reported 5.4 mg QE/100g DW total flavonoid content of mature broccoli floret whereas the by-product of broccoli stalk had 2.4 mg QE/100g DW, which is significantly lower than the total flavonoid content of broccoli microgreens presented in the present study.

The antioxidant activity of broccoli microgreens cultivated indoor and outdoor setting with soaked and unsoaked seeds and harvested at different duration is in the range of 60.94 to 81.90 percent. On the basis of the results, it is clearly observed from Table 4.1 that the broccoli microgreens harvested on 10th day (81.90 percent) have higher antioxidant activity than the one harvested at 20th day (60.94 percent). The finding of Kaur and Kapoor (2002) observed that broccoli floret contained 78.4 percent antioxidant activity which is less than the antioxidant activity of broccoli microgreens. It was reported in many literatures that broccoli leaves were high in antioxidants when compared to broccoli floret. The antioxidant activity of broccoli leaves were significantly higher i.e. 30.7% than the broccoli floret i.e. 14.7% reported by Liu *et al* (2018). Paradiso *et al* (2018) reported highest antioxidant activity levels for broccoli microgreens while lowest antioxidant activity was reported in the chichory microgreens.

The mineral content of broccoli microgreens is presented in Table 4.2 on dry weight basis. The zinc content of broccoli microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting stages was in the range of 8.23 to 11.33 mg/100g DW. The highest zinc content of broccoli microgreens cultivated outdoor with soaked seeds was 11.33 mg/100g DW 10th day of harvesting. In the present study it was seen that few grams (100g) of broccoli microgreens would satisfy the daily requirements of zinc by 80.93% and 103% for an adult male and female, respectively presented in Table 4.3. The zinc content of broccoli microgreen by Weber (2017) was 7.32 mg/100g FW, which is quite lower when compared with the present results. The reason for this variation could be that the zinc content of different crop is dependent on the zinc content of the soil which is used for cultivation. So cultivation at different locations could result in different concentration of zinc. Whereas Paradiso *et al* (2018) reported 4.36 µg/100g very low content of zinc in broccoli microgreens. Liu *et al* (2018) reported the zinc content of mature broccoli 54 µg/100g DW which is less than the broccoli microgreens showed in the present study.

The iron content of broccoli microgreens cultivated and harvested at different position and duration was in the range of 35.12 to 43.13 mg/100g DW. The high content of iron was observed in broccoli microgreens cultivated with soaked seeds at indoor setting and harvested at 10th day with value of 43.13 mg/100g DW. From the present study it was seen that just 100g of broccoli microgreens would fulfills more than 100 percent requirements of iron for both male and female. Xiao *et al* (2016) reported the iron content was 0.67 mg/100g FW of broccoli microgreens contrary to the present study showed high content of iron i.e. 43.13 mg/100g DW. The similar result was of iron content 6.5µg/100g reported by Paradiso *et al* (2018). In another study Liu *et al* (2018) reported that the mature broccoli had 45.83 µg/100g (0.045mg) DW which is less than the present study values.

The calcium content of broccoli microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time was in the range of 1357.88 to 2381.22 mg/100g. The high content of calcium was observed in broccoli microgreens cultivated with unsoaked seeds at indoor setting and harvested at 10th day with value of 2381.22 mg/100g DW. The broccoli microgreens of 100g were satisfying the daily requirements of calcium by 283.90% for an adult male and female (Table 4.3), which shows that few grams of broccoli microgreens are good option for vegetarians to mitigate the deficiency of calcium. Broccoli microgreens in the present study had high content of calcium content as compared to the results of Weber (2017) who reported 0.59 mg/g for compost grown broccoli microgreens and for hydroponically grown 0.29 mg/g. Similar findings by Xiao *et al* (2016) showed lower content of calcium (88 mg/100g FW) whereas present study had high content (2381.22 mg/100g). The mature broccoli had 30.4mg/100g of calcium reported by Farnham *et al* (2011), lower value than the present study.

Table 4.2 Mineral compositions of broccoli microgreens

Position	Seeds	10th day harvesting	20th day harvesting
Zinc (mg /100 g)			
Indoor	Soaked	11.21±0.31	8.84±0.04
	Unsoaked	10.74±0.16	9.24±0.08
Outdoor	Soaked	11.33±0.04	8.60±0.04
	Unsoaked	9.53±0.01	8.23±0.02
Iron (mg /100 g)			
Indoor	Soaked	43.13±0.09	37.10±0.02
	Unsoaked	41.94±0.03	35.12±0.06
Outdoor	Soaked	41.62±0.17	36.42±0.03
	Unsoaked	40.67±0.19	35.62±0.03
Calcium (mg /100 g)			
Indoor	Soaked	2295.96±1.47	1857.98±0.03
	Unsoaked	2381.22±0.17	1357.92±0.02
Outdoor	Soaked	2271.16±0.20	1852.97±0.03
	Unsoaked	2257.75±6.59	1357.88±0.01
Potassium (mg /100 g)			
Indoor	Soaked	470.01±45.30	440.23±0.08
	Unsoaked	476.63±1.37	348.96±0.09
Outdoor	Soaked	511.79±0.79	404.07±0.74
	Unsoaked	482.32±0.40	334.23±0.10
Magnesium (mg /100 g)			
Indoor	Soaked	571.73±1.08	565.33±0.04
	Unsoaked	690.30±0.13	584.50±0.07
Outdoor	Soaked	698.43±0.04	591.45±0.68
	Unsoaked	679.40±0.04	586.38±0.34
Sodium (mg /100 g)			
Indoor	Soaked	22.30±0.15	25.77±0.30
	Unsoaked	21.66±00	26.96±0.32
Outdoor	Soaked	21.34±0.05	28.17±0.32
	Unsoaked	22.46±0.03	24.69±1.39

Values are expressed as mean ±SD

The potassium content of broccoli microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time is in the range of 334.23 to 511.79 mg/100g (Table 4.2). The high content of potassium is observed in broccoli microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day with value of 511.79mg/100g DW. The broccoli microgreens of 100g were satisfying the daily requirements of potassium by 14.62 percent for an adult male and female. Xiao *et al* (2016) reported potassium content of broccoli microgreens was 69mg/100g of FW whereas the present study it was 511.79mg/100g DW. Recent researches also reported that potassium is the main mineral found in the microgreens of Brassicaceae (Xiao *et al* 2016). Farnham *et al* (2011) reported the potassium content of mature broccoli was 354.4 mg/100g which is less than the value for broccoli microgreens i.e. 511.79 mg/100g.

The magnesium content of broccoli microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 565.33 to 698.43 mg/100 g DW. The high content of magnesium was observed in broccoli microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day. The broccoli microgreens of 100g were satisfying the daily requirements of magnesium by 218.26 and 258.68 percent for an adult male and female, respectively (Table 4.3). Magnesium is the most important cation essential for many enzymatic reactions in the body and deficiency of magnesium lead to many degenerative diseases like diabetes, hypertension and atherosclerosis etc. (Fox et al 2001). Xiao *et al* (2016) reported magnesium content of broccoli microgreens 326mg/100g contrary to present study contained high amount i.e. 698.43 mg/100g of DW. Finding of Farnham *et al* (2011) reported the magnesium content of mature broccoli was 28.2 mg/100g that is too lower than the present study.

The sodium content of broccoli microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 21.34 to 28.17 mg/100 g DW. The high content of sodium was observed in broccoli microgreens cultivated with soaked seeds at outdoor setting and harvested at 20th day. Xiao et al (2016) reported the sodium content of broccoli microgreens was 52mg/100g. The sodium content of broccoli microgreens is quite high but the estimated average requirement for sodium is 2000 mg/d and percent adequacy of sodium, calculated for broccoli microgreens comes out to be very less i.e. 1 percent only.

Factorial ANOVA analysis of the results obtained by broccoli microgreens cultivated with soaked and unsoaked seeds at different positions (indoor and outdoor) and harvested at 10^{th} and 20^{th} day after germination. Statistical results showed that there was significant effect of soaking ($F(1, 16) = 36.20$, $p<0.001$), position ($F(1, 16) = 18.00$, $p \leq 0.001$), duration ($F(1, 16) = 4440.92$, $p<0.001$), soaking*duration ($F(1, 16) = 26.75$, $p<0.001$), position*duration ($F(1, 16) = 33.39$, $p<0.001$) and soaking*position*duration ($F(1, 16) = 44.50$, $p<0.001$) on

Table 4.3 Percent adequacy of vitamins and minerals of broccoli microgreens

Position	Seeds	10th day harvesting			20th day harvesting		
β-carotene (µg /100 g DW)		**β-carotene**	**% adequacy**		**β-carotene**	**% adequacy**	
			Male (2760 µg /d)*	**Female (2340µg /d)***		**Male (2760 µg /d)***	**Female (2340 µg /d)***
Indoor	Soaked	698.97	25.33	29.87	521.13	18.88	22.27
	Unsoaked	696.74	25.24	29.78	562.41	20.38	24.03
Outdoor	Soaked	344.75	12.49	14.73	335.27	12.15	14.33
	Unsoaked	303.13	10.98	12.95	272.20	9.86	11.63
Ascorbic acid (mg /100 g DW)		**Ascorbic acid**	**% adequacy**		**Ascorbic acid**	**% adequacy**	
			Male (65 mg/d)*	**Female (55mg/d)***		**Male (65 mg/d)***	**Female (55mg/d)***
Indoor	Soaked	25.28	38.88	45.95	15.29	23.52	27.70
	Unsoaked	23.01	35.39	41.83	13.66	21.01	24.83
Outdoor	Soaked	21.69	33.36	39.43	15.63	24.04	28.41
	Unsoaked	21.48	33.04	39.05	13.69	21.06	24.89
Zinc (mg /100 g DW)		**Zinc**	**% adequacy**		**Zinc**	**% adequacy**	
			Male (14 mg/d)*	**Female (11 mg/d)***		Male (14 mg/d)*	Female (11 mg/d)*
Indoor	Soaked	11.21	80.07	101.91	8.84	63.14	80.36
	Unsoaked	10.74	76.71	97.64	9.24	66.00	84.00
Outdoor	Soaked	11.33	80.93	103.00	8.60	61.43	78.18
	Unsoaked	9.53	68.07	86.64	8.23	58.79	74.82

Position	Seeds	10th day harvesting			20th day harvesting		
Iron (mg /100 g DW)		**Iron**	**% adequacy**		**Iron**	**% adequacy**	
			Male (11 mg/d)*	**Female (15 mg/d)***		**Male (11 mg/d)***	**Female (15 mg/d)***
Indoor	Soaked	43.13	392.06	287.51	37.10	337.27	247.33
	Unsoaked	41.94	381.24	279.58	35.12	319.27	234.13
Outdoor	Soaked	41.62	378.36	277.47	36.42	331.12	242.82
	Unsoaked	40.67	369.73	271.13	35.62	323.79	237.44
Calcium (mg/100g DW)		**Calcium**	**% adequacy**		**Calcium**	**% adequacy**	
			Male (800 mg/d)*	**Female (800 mg/d)***		**Male (800 mg/d)***	**Female (800 mg/d)***
Indoor	Soaked	2295.96	287.00	287.00	1857.98	232.25	232.25
	Unsoaked	2381.22	297.65	297.65	1357.92	169.74	169.74
Outdoor	Soaked	2271.16	283.90	283.90	1852.97	231.62	231.62
	Unsoaked	2257.75	282.22	282.22	1357.88	169.74	169.74
Potassium (mg/100g DW)		**Potassium**	**% adequacy**		**Potassium**	**% adequacy**	
			Male (3500mg/d)*	**Female (3500mg/d)***		**Male (3500mg/d)***	**Female (3500mg/d)***
Indoor	Soaked	470.01	13.43	13.43	440.23	12.58	12.58
	Unsoaked	476.63	13.62	13.62	348.96	9.97	9.97
Outdoor	Soaked	511.79	14.62	14.62	404.07	11.54	11.54
	Unsoaked	482.32	13.78	13.78	334.23	9.55	9.55
Magnesium (mg/100g DW)		**Magnesium**	**% adequacy**		**Magnesium**	**% adequacy**	
			Male (320mg/d)*	**Female (270mg/d)***		**Male (320mg/d)***	**Female (270mg/d)***
Indoor	Soaked	571.73	178.67	211.75	565.33	176.67	209.38
	Unsoaked	690.30	215.72	255.67	584.50	182.66	216.48
Outdoor	Soaked	698.43	218.26	258.68	591.45	182.66	216.48
	Unsoaked	679.4	212.31	251.63	586.38	183.24	217.18

Values are given in Mean ±SD

*Estimated Average Requirements for Indians, (EAR), ICMR-NIN 2020

nutrient content estimated among the indoor and outdoor cultivation with soaked and nsoaked seeds harvested at 2 stages i.e. 10th and 20th day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in broccoli microgreens is by soaking the broccoli microgreen seeds, cultivating them in outdoor location and harvesting at an early stage (10th day).

Nutrient Quality Score

The NQS used for evaluating the overall nutritional quality of microgreens, instead of simply quantifying nutrient content to evaluate the variations of a single nutrient among different crops.

Table 4.4 Nutrient Quality Score of broccoli microgreens

Position	Seeds	10th day harvesting	20th day harvesting
Nutrient Quality Score			
Indoor	Soaked	1014.53±3.52	862±0.36
	Unsoaked	1020.58±2.95	787.23±1.45
Outdoor	Soaked	1044.74±1.26	855.36±1.00
	Unsoaked	989.12±1.13	774.46±1.31

Values are expressed as mean ±SD

Furthermore, for the first time, a NQS was used assess the effects of cultivation and harvesting conditions on nutritional quality. It is clearly seen from the Table 4.4 that NQS is highest at outdoor cultivation with soaked seeds and harvested at 10th day to get maximum overall nutritional quality. Ghoora et al (2020) compared NQS of ten microgreens with NQS of mature spinach and found 2-3.5 times more NQS than mature spinach. The findings are at par with current findings as 20th harvested broccoli microgreens had lower NQS as compared to the 10th harvested.

4.1.2 Carrot

The β-carotene content of carrot microgreens cultivated under different cultivation and harvesting conditions is discussed in Table 4.5. The β-carotene content of carrot microgreens grown under different conditions is in the range of 357.03 to 635.77 µg /100 g DW. The carrot microgreens cultivated outdoor with soaked seeds and harvested at10th day had the highest level of β-carotene. Just 100 g of carrot microgreens were fulfilling the daily requirements of β-carotene by 23.04% and 27.17 % for an adult male and female, respectively (Table 4.7).The content reported in this study is contrary with literature (Ghoora *et al* 2020) report where carrot microgreens had 5.8mg/100g FW with high levels of β-carotene compare to the present study results as in the present study β-carotene was assessed on dry matter basis. Yoo *et al* (2020) reported the β-carotene of carrot greens i.e. 24.5 µg /g whereas the β-carotene of carrot roots was 27.6 µg /g. Also, Singh *et al* (2001) reported β-carotene content of mature carrot was 2.2 mg/100g.

The ascorbic acid content of carrot microgreens cultivated at different location and harvested at different growth stages is in the range of 13.26 to 17.38 mg/100g DW (Table 4.5). The ascorbic acid content was 17.38 mg/100g, significantly higher ($p<0.001$) when grown with unsoaked seeds at outdoor setting and harvested on 10th day. Just 100 g of carrot microgreens were fulfilling the daily requirements of ascorbic acid by 25.29% and 29.88 % for an adult male and female, respectively (Table 4.7).Ascorbic acid content reported on fresh matter basis revealed that carrot microgreens contained 94.7 mg per 100 g of ascorbic acid (Ghoora *et al* 2020). Xiao *et al* (2012) revealed that the ascorbic acid content of twenty-five microgreens ranged from 20.4 to 147.0 mg/100 g FW. Carrot microgreens have a high content of ascorbic acid (17.38 mg/100g) as compared to mature leaves of carrot (10 mg/100g) reported by Mishra and Gupta (2016)

The bioactive component and antioxidant activity in carrot microgreens is presented in Table 4.5. The chlorophyll content of carrot microgreens cultivated at different locations and harvested at different growth stages is in a range from 2.89 to 4.80 g/100g DW. A significantly ($p<0.01$) higher chlorophyll content (4.80 g/100g DW) was observed in carrot microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day. The chlorophyll content of carrot microgreens was 100.32 g per 100 g reported by Andrejiová *et al* (2017) and the carrot microgreens cultivated under artificial lighting had high chlorophyll content (113.5g/100g) as compared to without artificial lighting. These results are in accordance with the results reported in the present study where outdoor cultivation (using natural light) had significantly higher chlorophyll content as compared to indoor cultivation. The stage of harvesting also affected the chlorophyll content of microgreens as in the present study harvesting at an early stage (10th day) resulted in higher chlorophyll content (4.80g/100g DW) as compared to harvesting at 20th day (3.65g/100g DW).

The average total phenolic content in carrot microgreens in the present study is in the range from 1240.53 to 1872.37 mg GAE/100g DW. The carrot microgreens had a higher content of total phenols, when cultivated with soaked seeds at outdoor setting and harvested at 10th day. Syama *et al* (2014) and Yamada *et al* (2011) reported 34.2 mg GAE/g and 45.5 mg GAE/100g of total phenolic content in mature underutilized leaves of carrot which were significantly lower than the total phenolic content of carrot microgreens (1872.37 mg GAE/100g). Also, Singh *et al* (2018) reported that the mature carrot root had 16.06 mg/ 100g FW, of total phenolic content, which is also lower than the total phenolic content reported in the present research.

Table 4.5 Vitamin, bioactive content and antioxidant Activity of carrot microgreens

Position	Seeds	10^{th} day harvesting	20^{th} day harvesting
β- carotene (µg /100 g)			
Indoor	Soaked	630.34±0.24	491.56±1.70
	Unsoaked	528.78±0.99	368.96±1.14
Outdoor	Soaked	635.77±10.94	357.03±0.10
	Unsoaked	520.85±0.77	381.23±0.81
Vitamin C (mg/100g)			
Indoor	Soaked	13.40±0.18	13.51±0.20
	Unsoaked	14.63±0.33	13.26±0.13
Outdoor	Soaked	16.44±0.11	15.38±0.22
	Unsoaked	17.38±0.23	16.22±0.11
Chlorophyll (g /100 g)			
Indoor	Soaked	4.74±0.04	3.34±0.10
	Unsoaked	2.93±0.07	2.89±0.07
Outdoor	Soaked	4.80±0.17	3.65±0.05
	Unsoaked	3.45±0.07	3.31±0.01
TPC (mg/100g)			
Indoor	Soaked	1379.82±6.89	1240.53±5.47
	Unsoaked	1773.76±16.84	1640.83±10.27
Outdoor	Soaked	1872.37±5.55	1333.77±2.13
	Unsoaked	1724.39±0.83	1651.64±28.29
Total Flavonoids (mg/100g)			
Indoor	Soaked	443.80±2.30	411.27±1.37
	Unsoaked	393.62±2.33	320.46±1.96
Outdoor	Soaked	492.32±0.68	439.86±8.31
	Unsoaked	405.51±1.82	351.68±0.33
Antioxidant activity (%)			
Indoor	Soaked	76.00±0.05	73.75±00
	Unsoaked	79.53±0.75	55.47±0.55
Outdoor	Soaked	81.51±0.03	71.39±0.44
	Unsoaked	77.22±0.55	70.23±0.69

Values are expressed as mean±SD

The total flavonoid content of carrot microgreens cultivated at different positions and harvested at different durations is at a range from 320.46 to 492.32 mg QE/100g DW (Table 4.5). The results showed that flavonoid content of carrot microgreens cultivated with soaked seeds at outdoor settings were significantly higher ($p < 0.01$) when harvested at 10th day. The carrot microgreens had a high content of total flavonoids when compared to the carrot mature leaves and carrot roots. According to Smaya *et al* (2014) carrot mature leaves had 48.4 mg / g of flavonoids which is significantly lower than the flavonoid content present in carrot microgreens. Also, the total flavonoid content reported by Singh *et al* (2018) was 6.06 mg/100g for a mature carrot.

The antioxidant activity of carrot microgreens cultivated at indoor and outdoor setting with soaked and unsoaked seeds and harvested at different duration is in the range of 55.47 to 81.51 percent (Table 4.5). Based on the results, it is observed from Table 4.5 that the carrot microgreens harvested on 10th day (81.51 percent) have higher antioxidant activity than the one harvested at 20th day (55.47 percent). Joshi *et al* (2019) reported carrot underutilised leaves powder had a high antioxidant activity (82.64%). Also, Ghoora *et al* 2020 reported exceptionally high DPPH RSA with an IC50 value of 97.6 μg/ml of carrot microgreens. Thus it is well established through the present study that these carrot microgreens are a potent source of bioactive and antioxidant compounds. These bioactive compounds are significantly higher in carrot microgreens when compared with the mature leaves of carrot crop.

The mineral content of carrot microgreens is presented in Table 4.6 on dry weight basis. The zinc content of carrot microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting stages was in the range of 16.43 to 20.82 mg/100g DW. The highest zinc content in carrot microgreens was reported when these were cultivated indoor with soaked seeds and harvested on 10th day. Just 100 g of carrot microgreens were fulfilling the daily requirements of zinc by 132.48% and 168.61 % for an adult male and female, respectively (Table 4.7). The carrot microgreens are superior to mature carrots in term of zinc content. Goneim *et al* (2011) reported the zinc content of dried carrot leaves i.e. 8.8 mg/100g which is quite lower than the present study results i.e. 20.82 mg/100g DW. Also, Singh *et al* (2001) reported the zinc content of mature carrot root was 3.2 mg/100g, dry weight basis.

The iron content of carrot microgreens cultivated and harvested at different position and duration was in the range of 15.59 to 20.58 mg/100g DW. The highest content of iron was observed in carrot microgreens cultivated with unsoaked seeds at indoor setting and harvested at 10th day (Table 4.6). Just 100 g of carrot microgreens were fulfilling the daily requirements of iron by more than 100% for an adult male and female (Table 4.7).

Table 4.6 Mineral compositions of carrot microgreens

Position	Seeds	10th day harvesting	20th day harvesting
Zinc (mg /100 g)			
Indoor	Soaked	20.82±0.11	19.12±0.10
	Unsoaked	17.60±0.01	17.21±0.02
Outdoor	Soaked	18.55±0.01	17.65±0.05
	Unsoaked	16.56±0.27	16.43±0.03
Iron (mg /100 g)			
Indoor	Soaked	18.14±0.03	15.59±0.06
	Unsoaked	20.58±0.23	16.60±0.01
Outdoor	Soaked	19.53±0.25	16.56±0.10
	Unsoaked	20.35±0.03	16.78±0.13
Calcium (mg /100 g)			
Indoor	Soaked	1659.12±26.23	1221.21±0.29
	Unsoaked	1711.92±0.11	1577.08±3.38
Outdoor	Soaked	1533.38±0.02	1246.41±0.02
	Unsoaked	1825.36±0.01	1384.92±0.02
Potassium (mg /100 g)			
Indoor	Soaked	324.13±2.23	231.26±0.95
	Unsoaked	308.84±0.02	188.25±0.01
Outdoor	Soaked	319.50±0.38	217.99±0.02
	Unsoaked	341.20±0.01	195.03±0.03
Magnesium (mg /100 g)			
Indoor	Soaked	683.96±9.10	585.79±0.02
	Unsoaked	637.87±0.03	605.28±0.02
Outdoor	Soaked	671.37±0.04	566.79±0.02
	Unsoaked	640.68±1.27	605.80±0.01
Sodium (mg /100 g)			
Indoor	Soaked	53.01±0.03	58.89±0.10
	Unsoaked	53.03±0.09	55.06±0.04
Outdoor	Soaked	53.12±0.48	57.31±0.03
	Unsoaked	49.46±0.55	53.11±0.17

Values are expressed as mean±SD

Singh *et al* (2001) reported the iron content of mature carrot 7.7mg/100g DW which is lower than the present study. Similarly, the mature leaves of carrot had low levels of iron as compared to the present study results. Mishra and Gupta (2016) reported iron content of carrot mature leaves i.e. 4.08 mg/100g which is five times less than the present study (20.58 mg/100g). Carrot microgreens could be used on regular basis by anaemic population to elevate their haemoglobin levels.

The calcium content of carrot microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time was in the range of 1221.21 to 1825.36 mg/100g (Table 4.6). A significantly high content of calcium was observed in carrot microgreens cultivated with unsoaked seeds at outdoor setting and harvested at 10th day. 100 g of broccoli microgreens were meeting the daily requirements of calcium by 191.67% for an adult male and female (Table 4.7). Leite *et al* (2011) reported carrot mature leaves had 0.05 mg/100g DW of calcium content whereas a significant high content of calcium i.e. 1825.36 mg/100g DW is reported in the present study. The calcium content of carrot microgreens is higher as compared to the mature leaves and roots of carrot. Mishra and Gupta (2016) reported the calcium content of carrot mature leaves was 77.34mg/100. Thus these microgreens as a very good source of calcium are highly recommended as a food based approach to reduce the incidence of osteoporosis or low bone density in adults. The potassium content of carrot microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time is in the range of 188.25 to 341.20 mg/100g (Table 4.6). The high content of potassium is observed in carrot microgreens cultivated with unsoaked seeds at an outdoor setting and harvested at 10th day with a value of 341.20 mg/100g DW. Just 100 g of carrot microgreens were fulfilling the daily requirements of potassium by 9.13 % for an adult male and female (Table 4.7).The carrot microgreens have high potassium content as compared to the mature leaves of carrot. Leite *et al* (2011) reported potassium content of 467.55 mg/100g in dehydrated carrot mature leaves. In the present study, the potassium content of carrot microgreens was high 341.20 mg/100g as compared to the potassium content of other microgreens like radish microgreen (176 mg/100g) given by Xiao *et al* (2016).

The magnesium content of carrot microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 566.79 to 683.96 mg/100 g DW. The high content of magnesium was observed in carrot microgreens cultivated with soaked seeds at indoor setting and harvested at the 10th day. 100 g of carrot microgreens were meeting the daily requirements of magnesium by 209.80 % and 248.65 % for an adult male and female, respectively (Table 4.7).

Table 4.7 Percent adequacy of vitamins and minerals of carrot microgreens

Position	**Seeds**	**10th day harvesting**			**20th day harvesting**		
β-carotene (μg /100 g DW)		**β-carotene**	**% adequacy**		**β-carotene**	**% adequacy**	
			Male (2760 μg /d)*	**Female (2340μg /d)***		**Male (2760 μg /d)***	**Female (2340 μg /d)***
Indoor	Soaked	630.34	22.84	26.94	491.56	17.81	21.01
	Unsoaked	528.78	19.16	22.60	368.96	13.37	15.77
Outdoor	Soaked	635.77	23.04	27.17	357.03	12.94	15.26
	Unsoaked	520.85	18.87	22.26	381.23	13.81	16.29
Ascorbic acid (mg /100 g DW)		**Ascorbic acid**	**% adequacy**		**Ascorbic acid**	**% adequacy**	
			Male (65 mg/d)*	**Female (55mg/d)***		**Male (65 mg/d)***	**Female (55mg/d)***
Indoor	Soaked	13.40	20.61	24.36	13.51	20.78	24.56
	Unsoaked	14.63	22.50	26.59	13.26	20.39	24.10
Outdoor	Soaked	16.44	25.29	29.88	15.38	23.67	27.97
	Unsoaked	17.38	26.73	31.59	16.22	24.95	29.49
Zinc (mg /100 g DW)		**Zinc**	**% adequacy**		**Zinc**	**% adequacy**	
			Male (14 mg/d)*	**Female (11 mg/d)***		Male (14 mg/d)*	Female (11 mg/d)*
Indoor	Soaked	20.82	148.71	189.27	19.12	136.60	173.85
	Unsoaked	17.60	125.74	160.03	17.21	122.93	156.45
Outdoor	Soaked	18.55	132.48	168.61	17.65	126.10	160.48
	Unsoaked	16.56	118.29	150.55	16.43	117.36	149.36

Position	Seeds	10th day harvesting			20th day harvesting		
Iron (mg /100 g DW)		**Iron**	**% adequacy**		**Iron**	**% adequacy**	
			Male (11 mg/d)*	**Female (15 mg/d)***		**Male (11 mg/d)***	**Female (15 mg/d)***
Indoor	Soaked	18.14	164.88	120.91	15.59	141.70	103.91
	Unsoaked	20.58	187.09	137.20	16.60	150.88	110.64
Outdoor	Soaked	19.53	177.52	130.18	16.56	150.58	110.42
	Unsoaked	20.35	185.00	135.67	16.78	152.55	111.87
Calcium (mg/100g DW)		**Calcium**	**% adequacy**		**Calcium**	**% adequacy**	
			Male (800 mg/d)*	**Female (800 mg/d)***		**Male (800 mg/d)***	**Female (800 mg/d)***
Indoor	Soaked	1659.12	207.39	207.39	152.65	**19.12**	19.12
	Unsoaked	1711.92	213.99	213.99	197.14	17.21	17.21
Outdoor	Soaked	1533.38	191.67	191.67	155.80	17.65	17.65
	Unsoaked	1825.36	228.17	228.17	173.12	16.43	16.43
Potassium (mg/100g DW)		**Potassium**	**% adequacy**		**Potassium**	**% adequacy**	
			Male (3500mg/d)*	**Female (3500mg/d)***		**Male (3500mg/d)***	**Female (3500mg/d)***
Indoor	Soaked	324.13	9.26	9.26	231.26	6.61	6.61
	Unsoaked	308.84	8.82	8.82	188.25	5.38	5.38
Outdoor	Soaked	319.50	9.13	9.13	217.99	6.23	6.23
	Unsoaked	341.20	9.75	9.75	195.03	5.57	5.57
Magnesium (mg/100g DW)		**Magnesium**	**% adequacy**		**Magnesium**	**% adequacy**	
			Male (320mg/d)*	**Female (270mg/d)***		**Male (320mg/d)***	**Female (270mg/d)***
Indoor	Soaked	683.96	213.74	253.32	585.79	183.06	216.96
	Unsoaked	671.37	199.34	236.25	605.28	189.15	224.18
Outdoor	Soaked	637.87	209.80	248.65	566.79	177.12	209.92
	Unsoaked	640.68	200.21	237.29	605.80	189.31	224.37

Values are given in Mean ±SD
*Estimated Average Requirements for Indians, (EAR), ICMR-NIN 2020

The carrot microgreens have higher magnesium content when compared to the root of carrot. Singh *et al* (2001) reported the magnesium content of carrot was 1.8 mg/100g DW whereas carrot microgreens contained 683.96 mg/100 DW. Similarly, Leite *et al* (2011) reported magnesium content of mature leaves of carrot was 0.28 mg/100g DW whereas the present study has significantly higher ($p<0.001$) magnesium content (683.96 mg/100g DW) than the mature leaves of carrot. These microgreens being an excellent source of minerals like iron, calcium, zinc and magnesium could be used as a food-based approach in combating various micronutrient deficiencies arising due to the deficiencies of these minerals.

The sodium content of carrot microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 49.46 to 58.89 mg/100 g DW. The high content of magnesium was observed in carrot microgreens cultivated with soaked seeds at indoor setting and harvested at the 20th day. Contrary to our results Ghoora *et al* (2020) reported sodium content of carrot microgreens was 123mg/100g FW.

Factorial ANOVA analysis of the results obtained by carrot microgreens cultivated with soaked and unsoaked seeds at different positions (indoor and outdoor) and harvested at 10th and 20th day after germination. Statistical results showed that there was significant effect of soaking ($F(1, 16)=176.50$, $p<0.001$), position ($F(1, 16)=133.77$, $p<0.001$), duration ($F(1, 16)=5521.02$, $p<0.001$), soaking*position ($F(1,16)=14.18$, $p=0.002$, soaking*duration ($F(1, 16)=473.31$, $p<0.001$), position*duration ($F(1, 16)=187.45$, $p<0.001$) and soaking* position*duration ($F(1, 16)=203.98$, $p<0.001$) on nutrient content estimated among the indoor and outdoor cultivation with soaked and unsoaked seeds harvested at 2 stages i.e. 10th and 20th day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in carrot microgreens is by soaking the carrot microgreen seeds, cultivating them in outdoor location and harvesting at 10th day.

Table 4.8 Nutrient Quality Score of carrot microgreens

Position	Seeds	10th day harvesting	20th day harvesting
Nutrient Quality Score			
Indoor	Soaked	766.98±2.31	656.26±1.50
	Unsoaked	773.98±2.29	696.48±0.39
Outdoor	Soaked	784.78±8.70	649.55±1.27
	Unsoaked	784.55±3.64	674.01±1.35

Values are expressed as mean ±SD

Nutrient Quality Score

The NQS used for evaluating the overall nutritional quality of microgreens, instead of simply quantifying nutrient content to evaluate the variations of a single nutrient among different crops. Furthermore, for the first time, a NQS was used assess the effects of cultivation and harvesting conditions on nutritional quality. It is clearly seen from the Table 4.8 that NQS is highest at outdoor cultivation with soaked seeds and harvested at 10th day to get maximum overall nutritional quality.

4.1.3 Coriander

The β-carotene content of coriander microgreens cultivated under different cultivation and harvesting conditions is discussed in Table 4.9. The β-carotene content of coriander microgreens in the present study is in the range of 317.64 to 629.53 µg /100 g DW. The coriander microgreens cultivated indoor with soaked seeds and harvested at10th had high level of β-carotene. Just 100 g of coriander microgreens were meeting the daily requirements of β-carotene by 19.12% and 22.55 % for an adult male and female, respectively (Table 4.11). Dietary supplementation with β-carotene (precursor of vitamin A) has been associated with protection against oxidative damage and degeneration, immune functions and visual impairment. The β-carotene content obtained in this study is contrary with literature (Kyriacou *et al* 2020) report where coriander microgreens had 325.1 mg/kg DW as compare to the results of the present study. Whereas the mature leaves of coriander had 208.29 µg/100g DW of β-carotene reported by Nambiar and Sharma (2014). The coriander microgreens have more β-carotene content as compared to the mature leaves of the coriander. The ascorbic acid content of coriander microgreens cultivated and harvested at different location and duration respectively is in the range of 14.56 to 18.85 mg/100g DW. The ascorbic acid content was 18.85 mg/100g, highest when grown with soaked seeds at indoor setting and harvested on 10th day. Just 100 g of coriander microgreens were fulfilling the daily requirements of vitamin C by 26.91% and 31.80 % for an adult male and female, respectively (Table 4.11). Comparable values have also been reported in other studies (Kyriacou *et al* 2020) showed that coriander microgreens contained 121.40 mg per kg of DW (12.14 mg/100g) of ascorbic acid whereas present study reported 18.85 mg/100g. The mature leaves of coriander had 98.1 mg/100g FW of ascorbic acid reported by Singh *et al* (2001). In the present study the estimation of ascorbic acid was done in dried samples whereas fresh samples were used by the Singh *et al* (2001).

The bioactive component and antioxidant activity is presented in Table 4.9. The chlorophyll content of coriander microgreens cultivated at different locations and harvested at

Table 4.9 Vitamin, Bioactive content and Antioxidant Activity of coriander microgreens

Position	Seeds	10th day harvesting	20th day harvesting
β- carotene (μg /100 g)			
Indoor	Soaked	629.53±1.06	385.09±4.31
	Unsoaked	426.88±4.52	366.72±2.60
Outdoor	Soaked	527.65±2.05	331.27±17.56
	Unsoaked	519.93±0.25	317.64±5.19
Vitamin C (mg/100g)			
Indoor	Soaked	18.85±0.12	15.70±0.18
	Unsoaked	15.65±0.08	14.58±0.26
Outdoor	Soaked	17.49±0.22	15.33±0.13
	Unsoaked	16.37±0.33	14.56±0.25
Chlorophyll (g /100 g)			
Indoor	Soaked	4.79±0.15	3.49±0.22
	Unsoaked	3.73±0.03	3.87±0.01
Outdoor	Soaked	4.59±0.01	3.68±0.02
	Unsoaked	3.89±0.09	3.51±0.03
Total phenolic content (mg GAE/100g)			
Indoor	Soaked	1387.93±2.11	1246.66±1.98
	Unsoaked	1774.07±25.73	1624.70±2.60
Outdoor	Soaked	1467.07±40.01	1455.70±14.57
	Unsoaked	1671.40±22.31	1585.19±3.06
Total Flavonoids (mg QE/100g)			
Indoor	Soaked	444.37±2.08	414.19±3.43
	Unsoaked	394.83±2.87	348.63±17.25
Outdoor	Soaked	473.37±23.16	325.23±3.85
	Unsoaked	350.63±47.14	349.09±2.26
Antioxidant activity (%)			
Indoor	Soaked	80.74±0.55	73.71±1.36
	Unsoaked	80.68±0.20	73.17±0.99
Outdoor	Soaked	81.35±0.05	78.13±0.69
	Unsoaked	81.11±0.05	74.23±1.59

Values are expressed as mean ±SD

different growth stages is in range from 3.49 to 4.79 g/100g DW. A significantly ($p<0.01$) higher chlorophyll content (4.79 g/100g DW) is observed in coriander microgreens cultivated with soaked seeds at indoor setting and harvested at 10th day. The chlorophyll content of coriander microgreens was 13.63 mg/ kg FW (0.01g/kg) reported by Kyriacou *et al* (2020). Kathirvel *et al* (2006) reported the mature coriander leaves contained 1.30mg/g FW of chlorophyll content. The average total phenolic content in coriander microgreens in present study is in the range from 1246.66 to 1774.07 mg GAE/100g DW. The coriander microgreens had higher content of total phenols i.e. 1774.07 mg GAE/100g DW, when cultivated with unsoaked seeds at indoor setting and harvested at 10^{th} day. The phenolic compounds act as natural antioxidant mainly due to their redox properties, which allow them to act as reducing agents and singlet oxygen quenchers. Also, they have metal-chelating potential (Rice-Evans *et al* 1995). Moreover, phenolic compounds show different biological activities as anti-inflammatory, antibacterial, anti-carcinogenic, anti-viral, anti-allergic and immune-stimulating agents (Larson, 1988). The coriander microgreens contained significantly higher amount of total phenolic content as compared to the mature counterparts. Nair and Lekshmi (2019) reported the total phenolic content of coriander microgreens was 0.131 mg/100g. Whereas the mature leaves of coriander had 4.95 mg GAE/g FW of total phenolic content reported by Agrawal et al (2016). Also Kaur and Kapoor (2002) reported total phenolic content of coriander 82.5 mg catechol/100 g of fresh weight whereas present study reported 1774.07 mg GAE/100g DW.

The total flavonoid content of coriander microgreens cultivated at different positions and harvested at different durations is at a range from 325.23 to 473.37 mg QE/100g DW. The results showed that flavonoid content of coriander microgreens cultivated with unsoaked seeds at outdoor settings were significantly higher ($p< 0.01$) when harvested at 10th day with value of 473.37 mg QE/100g DW. The mature leaves of coriander had 93.42 mg/100g FW of total flavonoids reported by Ji *et al* (2011). The tiny leaves of coriander microgreens are significantly high in flavonoids than the mature leaves of coriander reported by Ji *et al* (2011).

The antioxidant activity of coriander microgreens cultivated indoor and outdoor setting with soaked and unsoaked seeds and harvested at different duration is in the range of 73.17 to 81.35 percent. On the basis of the results, it is clearly observed from Table 4.9 that the coriander microgreens harvested on 10^{th} day (81.35 percent) have higher antioxidant activity than the one harvested at 20^{th} day (78.13 percent). Al-Juhaimi and Ghafoor (2011) reported the antioxidant activity of mature coriander leaves was 26.82% which is quite low then the values of the present study (81.35%). Also Kaur and Kapoor (2002) reported the antioxidant activity of coriander 71.8 %.

The mineral content of coriander microgreens is presented in Table 4.10 on dry weight basis. The zinc content of coriander microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting stages was in the range of 16.66 to 20.73 mg/100g DW. The highest zinc content of coriander microgreens cultivated indoor with soaked seeds was 20.73 mg/100g DW 10th day of harvesting. Just 100 g of coriander microgreens were fulfilling the daily requirements of zinc by 132.45% and 168.58 % for an adult male and female, respectively (Table 4.11).The coriander microgreens are superior to mature coriander in term of zinc content. Singh *et al* (2001) reported the zinc content of mature coriander was 2.7 mg/100g, dry weight basis. The iron content of coriander microgreens cultivated and harvested at different position and duration was in the range of 15.31 to 21.76 mg/100g DW. The high content of iron was observed in coriander microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day with value of 21.76 mg/100g DW. 100 g of coriander microgreens were fulfilling the daily requirements of iron more than 100% for an adult male and female, respectively (Table 4.11). Singh *et al* (2001) reported the iron content of mature coriander 22.3 mg/100g DW which is lower than the present study. Similarly Pushplata (2018) reported the iron content of leaves and stem of coriander 24.50 and 23.50 mg/100g, respectively.

The calcium content of coriander microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time was in the range of 982.06 to 2058.16 mg/100g. The high content of calcium was observed in coriander microgreens cultivated with unsoaked seeds at indoor setting and harvested at 10th day with value of 2058.16 mg/100g DW. Just 100 g of coriander microgreens were fulfilling the daily requirements of calcium by 189.83 % for an adult male and female, respectively (Table 4.11). Kyriacou *et al* (2019) reported coriander microgreens had 19.58 g/kg DW of calcium content whereas higher content of calcium i.e. 2058.16 mg/100g DW in the present study. Whereas the mature leaves of coriander had 305.50mg/100g calcium content reported by Pushplata (2018). Thus, these microgreens as a very good source of calcium are highly recommended as a food based approach to reduce the incidence of osteoporosis or low bone density in adults.

The potassium content of coriander microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time is in the range of 335.41 to 404.52 mg/100g (Table 4.10). The high content of potassium is observed in coriander microgreens cultivated with soaked seeds at indoor setting and harvested at 20th day with value of 404 mg/100g DW. Just 100 g of coriander microgreens were meeting the daily requirements of potassium by 10.89% for an adult male and female, respectively (Table 4.11). The coriander microgreens have high potassium content as compared to the mature counterparts.

Table 4.10 Mineral composition of coriander microgreens

Position	Seeds	10^{th} day harvesting	20^{th} day harvesting
Zinc (mg /100 g)			
Indoor	Soaked	20.73±0.16	19.44±0.18
	Unsoaked	17.23±0.04	17.22±0.04
Outdoor	Soaked	18.54±0.38	17.76±0.02
	Unsoaked	17.38±0.12	16.66±0.07
Iron (mg /100 g)			
Indoor	Soaked	18.64±0.27	15.31±0.14
	Unsoaked	20.34±0.42	19.53±0.35
Outdoor	Soaked	21.76±0.10	16.37±0.37
	Unsoaked	18.32±0.05	16.66±0.07
Calcium (mg /100 g)			
Indoor	Soaked	1614.36±0.01	982.06±2.31
	Unsoaked	2058.16±47.15	1019.74±5.23
Outdoor	Soaked	1518.65±4.67	1043.44±4.27
	Unsoaked	1872.86±32.86	1056.80±19.66
Potassium (mg /100 g)			
Indoor	Soaked	340.88±0.10	404.52±0.03
	Unsoaked	352.68±0.09	363.55±0.03
Outdoor	Soaked	381.05±0.22	396.45±0.05
	Unsoaked	382.78±0.08	335.41±0.05
Magnesium (mg /100 g)			
Indoor	Soaked	689.05±1.24	613.79±38.19
	Unsoaked	637.85±2.50	609.48±5.76
Outdoor	Soaked	671.23±0.47	511.11±40.90
	Unsoaked	620.37±0.79	504.94±0.47
Sodium (mg /100 g)			
Indoor	Soaked	28.37±0.33	30.36±0.29
	Unsoaked	28.37±0.10	32.46±0.35
Outdoor	Soaked	28.01±0.08	30.73±0.40
	Unsoaked	22.98±0.23	31.83±0.17

Values are expressed as mean ±SD

Table 4.11 Percent adequacy of vitamins and minerals of coriander microgreens

Position	Seeds	10th day harvesting			20th day harvesting		
β-carotene (µg /100 g DW)		**β-carotene**	**% adequacy**		**β-carotene**	**% adequacy**	
			Male (2760 µg /d)*	**Female (2340µg /d)***		**Male (2760 µg /d)***	**Female (2340 µg /d)***
Indoor	Soaked	629.53	22.81	26.90	385.09	13.95	16.46
	Unsoaked	426.88	15.47	18.24	366.72	13.29	15.67
Outdoor	Soaked	527.65	19.12	22.55	331.27	12.00	14.16
	Unsoaked	519.93	18.84	22.22	317.64	11.51	13.57
Ascorbic acid (mg /100 g DW)		**Ascorbic acid**	**% adequacy**		**Ascorbic acid**	**% adequacy**	
			Male (65 mg/d)*	**Female (55mg/d)***		**Male (65 mg/d)***	**Female (55mg/d)***
Indoor	Soaked	18.85	29.00	34.27	15.70	24.16	28.55
	Unsoaked	15.65	24.08	28.46	14.58	22.44	26.52
Outdoor	Soaked	17.49	26.91	31.80	15.33	23.58	27.87
	Unsoaked	16.37	25.18	29.76	14.56	22.40	26.47
Zinc (mg /100 g DW)		**Zinc**	**% adequacy**		**Zinc**	**% adequacy**	
			Male (14 mg/d)*	**Female (11 mg/d)***		Male (14 mg/d)*	Female (11 mg/d)*
Indoor	Soaked	20.73	148.07	188.45	19.44	138.86	176.73
	Unsoaked	17.23	123.07	156.64	17.22	123.02	156.58
Outdoor	Soaked	18.54	132.45	168.58	17.76	126.83	161.42
	Unsoaked	17.38	124.14	158.00	16.66	119.00	151.46

Position	Seeds	10^{th} day harvesting			20^{th} day harvesting		
Iron (mg /100 g DW)		**Iron**	**% adequacy**		**Iron**	**% adequacy**	
			Male (11 mg/d)*	**Female (15 mg/d)***		**Male (11 mg/d)***	**Female (15 mg/d)***
Indoor	Soaked	18.64	169.42	124.24	15.31	139.15	102.04
	Unsoaked	20.34	184.94	135.62	19.53	177.57	130.22
Outdoor	Soaked	21.76	197.85	145.09	16.37	148.79	109.11
	Unsoaked	18.32	166.52	122.11	16.66	151.45	111.07
Calcium (mg/100g DW)		**Calcium**	**% adequacy**		**Calcium**	**% adequacy**	
			Male (800 mg/d)*	**Female (800 mg/d)***		**Male (800 mg/d)***	**Female (800 mg/d)***
Indoor	Soaked	1614.36	201.79	201.79	982.06	122.75	122.75
	Unsoaked	2058.16	257.27	257.27	1043.44	127.46	127.46
Outdoor	Soaked	1518.65	189.83	189.83	1019.74	130.43	130.43
	Unsoaked	1872.86	234.10	234.10	1056.80	132.10	132.10
Potassium (mg/100g DW)		**Potassium**	**% adequacy**		**Potassium**	**% adequacy**	
			Male (3500mg/d)*	**Female (3500mg/d)***		**Male (3500mg/d)***	**Female (3500mg/d)***
Indoor	Soaked	340.88	9.74	9.74	404.52	11.56	11.56
	Unsoaked	352.68	10.08	10.08	363.55	10.39	10.39
Outdoor	Soaked	381.05	10.89	10.89	396.45	11.33	11.33
	Unsoaked	382.78	10.94	10.94	335.41	9.58	9.58
Magnesium (mg/100g DW)		**Magnesium**	**% adequacy**		**Magnesium**	**% adequacy**	
			Male (320mg/d)*	**Female (270mg/d)***		**Male (320mg/d)***	**Female (270mg/d)***
Indoor	Soaked	689.05	215.33	255.20	613.79	191.81	227.33
	Unsoaked	637.85	199.33	236.24	609.48	190.46	225.73
Outdoor	Soaked	671.23	209.76	248.60	511.11	159.72	189.30
	Unsoaked	620.37	193.87	229.77	504.94	157.79	187.01

Values are given in Mean ±SD

*Estimated Average Requirements for Indians, (EAR), ICMR-NIN 2020

In the present study, the potassium content is low when compared with Kyriacou et al (2019) research report of potassium content 41.39g/kg DW of coriander microgreens. The potassium content of coriander microgreens was high 404 mg/100g as compared to the potassium content of other microgreens also, like radish microgreen (176 mg/100g) given by Xiao *et al* (2016). The magnesium content of coriander microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 504.94 to 689.05 mg/100 g DW. The high content of magnesium was observed in coriander microgreens cultivated with soaked seeds at indoor setting and harvested at 10th day. Just 100 g of coriander microgreens were fulfilling the daily requirements of magnesium by 209.76 % and 248.60 % for an adult male and female, respectively (Table 4.11).The coriander microgreens have higher magnesium content when compared to mature leaves of coriander. Kyriacou *et al* (2019) reported the magnesium content of coriander had 6.19 g/kg DW whereas coriander microgreens contained 689.05 mg/100 g DW.

The sodium content of coriander microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 22.98 to 32.46 mg/100 g DW. The high content of sodium was observed in coriander microgreens cultivated with unsoaked seeds at indoor setting and harvested at the 20th day. Kyriacou et al (2020) reported sodium content of coriander microgreens was 0.48g/kg DW.

Factorial ANOVA analysis of the results obtained by coriander microgreens cultivated with soaked and unsoaked seeds at different positions (indoor and outdoor) and harvested at 10th and 20th day after germination. Statistical results showed that there was significant effect duration ($F (1, 16) = 616.63$, $p<0.001$), soaking*position ($F(1,16)=17.99$, $p\leq0.001$ and soaking*duration ($F (1, 16) = 22.46$ on nutrient content estimated among the indoor and outdoor cultivation with soaked and unsoaked seeds harvested at 2 stages i.e. 10th and 20th day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in coriander microgreens is by soaking the carrot microgreen seeds, cultivating them in indoor location and harvesting at 10th day.

Table 4.12 Nutrient Quality Score of coriander microgreens

Position	**Seeds**	**10th day harvesting**	**20th day harvesting**
Nutrient Quality Score			
Indoor	Soaked	813.33±5.70	641.19±16.30
	Unsoaked	795.51±4.56	663.45±2.95
Outdoor	Soaked	786.04±2.49	611.55±13.52
	Unsoaked	773.07±4.31	602.63±2.42

Values are expressed as mean ±SD

Nutrient Quality Score

The NQS used for evaluating the overall nutritional quality of microgreens, instead of simply quantifying nutrient content to evaluate the variations of a single nutrient among different crops. Furthermore, for the first time, a NQS was used assess the effects of cultivation and harvesting conditions on nutritional quality of coriander microgreens. It is clearly seen from the table 4.12 that NQS is highest at indoor cultivation with soaked seeds and harvested at 10th day to get maximum overall nutritional quality.

4.1.4 Spinach

The β-carotene content of spinach microgreens cultivated under different cultivation and harvesting conditions was discussed in Table 4.13. The β-carotene content of spinach microgreens in the present study is in the range of 357.03 to 696.37 μg /100 g DW. The spinach microgreens cultivated indoor with unsoaked seeds and harvested at 10th had high level of β-carotene i.e. 696.37 μg /100g DW. Just 100 g of spinach microgreens were meeting the daily requirements of β-carotene by 14.16 % and 16.70 % for an adult male and female, respectively (Table 4.15). Ghoora *et al* (2020) reported a significantly higher β-carotene content of spinach microgreen than the mature one. The β-carotene content of spinach microgreens was 6.1 mg/100g.

The ascorbic acid content of spinach microgreens cultivated and harvested at different location and duration respectively is in the range of 16.50 to 20.43 mg/100g DW. The ascorbic acid content was 20.43 mg/100g, highest when grown with soaked seeds at outdoor setting and harvested on 10th day. Just 100 g of spinach microgreens fulfills the daily requirements of vitamin C by 31.43 % and 37.14 % for an adult male and female, respectively (Table 4.15). Sharma *et al* (2021) reported the high ascorbic acid content of spinach microgreens (11.8 mg /100g) as compared to the mature counterparts (9.06 mg/100g)**.** Similar trend was observed by Ghoora *et al* (2020) that the ascorbic acid content of spinach microgreens was significantly higher as compared to mature spinach. The ascorbic acid content of spinach micogreens was 71.2 mg/100g. The bioactive component and antioxidant activity is presented in Table 4.13. The chlorophyll content of spinach microgreens cultivated at different locations and harvested at different growth stages is in range from 2 to 3.69 g/100g DW. A significantly ($p<0.01$) higher chlorophyll content (3.69 g/100g DW) is observed in spinach microgreens cultivated with unsoaked seeds at outdoor setting and harvested at 10th day. Contatrty to present results Ghoora *et al* (2020b) reported that spinach microgreen had 35.3 mg/100g of chlorophyll content.

Table 4.13 Vitamin, Bioactive content and Antioxidant Activity of spinach microgreens

Position	Seeds	10^{th} day harvesting	20^{th} day harvesting
β- carotene (μg /100 g)			
Indoor	Soaked	677.38±0.20	548.15±6.22
	Unsoaked	696.37±1.52	506.36±10.13
Outdoor	Soaked	390.83±7.26	357.03±4.87
	Unsoaked	496.68±2.03	346.76±2.94
Vitamin C (mg/100g)			
Indoor	Soaked	18.58±0.81	16.50±0.19
	Unsoaked	19.58±0.69	18.93±0.29
Outdoor	Soaked	20.43±0.33	19.67±0.24
	Unsoaked	19.96±0.56	19.29±0.31
Chlorophyll (g /100 g)			
Indoor	Soaked	2.71±0.01	2.00±0.01
	Unsoaked	2.80±0.02	2.80±0.03
Outdoor	Soaked	3.57±0.04	3.43±0.27
	Unsoaked	3.69±0.03	2.26±0.04
TPC (mg GAE/100g)			
Indoor	Soaked	2197.57±0.88	1270.81±0.15
	Unsoaked	2118.11±4.21	1468.33±1.27
Outdoor	Soaked	2017.06±1.65	1200.66±2.76
	Unsoaked	2012.14±1.55	1421.24±1.40
Total Flavonoids (mg QE/100g)			
Indoor	Soaked	357.52±0.05	312.86±2.91
	Unsoaked	410.85±0.87	336.80±1.35
Outdoor	Soaked	546.47±0.74	508.87±3.10
	Unsoaked	610.57±0.74	485.89±94.73
Antioxidant activity (%)			
Indoor	Soaked	74.48±0.01	68.68±0.13
	Unsoaked	71.39±0.05	67.56±0.44
Outdoor	Soaked	80.19±00	68.80±0.10
	Unsoaked	76.97±0.02	67.88±0.01

Values are expressed as mean ±SD

The average total phenolic content in spinach microgreens in present study is in the range from 1200.66 to 2197.57 mg GAE/100g DW. The spinach microgreens had higher content of total phenols i.e. 2197.57 mg GAE/100g DW, when cultivated with soaked seeds at indoor setting and harvested at 10^{th} day. Sharma *et al* (2021) reported the high total phenolic content of spinach microgreens (0.95 mg GAE/g) as compared to the mature counterparts (0.59 mg GAE/g)**.** Ghoora *et al* (2020b) explored the total phenolic content of spinach microgreens (14.6 mg/100g FW) which was very low as compared to the present results.

The total flavonoid content of spinach microgreens cultivated at different positions and harvested at different durations is at a range from 312.86 to 610.57 mg QE/100g DW. The results showed that flavonoid content of spinach microgreens cultivated with unsoaked seeds at outdoor settings were significantly higher ($p< 0.01$) when harvested at 10^{th} day with value of 610.57 mg QE/100g DW. The flavonoid content of spinach microgreens was reported by Ghoora *et al* (2020b) with 2.4 mg/100g FW.

The antioxidant activity of spinach microgreens cultivated indoor and outdoor setting with soaked and unsoaked seeds and harvested at different duration is in the range of 67.56 to 80.19 percent. On the basis of the results, it is clearly observed from Table 4.13 that the spinach microgreens harvested on 10^{th} day (80.19 percent) have higher antioxidant activity than the one harvested at 20^{th} day (68.80 percent) Sharma *et al* (2021) reported higher antioxidant activity in spinach microgreens (43.07%) than mature spinach (36.95%).

The mineral content of spinach microgreens is presented in Table 4.14 on dry weight basis. The zinc content of spinach microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting stages was in the range of 22.41 to 38.53mg/100g DW. The highest zinc content of spinach microgreens cultivated outdoor with soaked seeds was 38.53 mg/100g DW 10^{th} day of harvesting. Just 100 g of spinach microgreens were meeting the daily requirements of zinc more than 100 % for an adult male and female, respectively (Table 4.15). Ghoora *et al* (2020) reported zinc content of spinach microgreens 1.33 mg/100g.

The iron content of spinach microgreens cultivated and harvested at different position and duration was in the range of 19.78to 28.60 mg/100g DW. The high content of iron was observed in spinach microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day with value of 28.60 mg/100g DW. Just 100 g of spinach microgreens were meeting the daily requirements of iron by 260.03 % and 190.69 % for an adult male and female, respectively (Table 4.15). Sharma *et al* (2021) reported high content of iron in spinach microgreens (4.03mg/100g) than mature spinach (3.59 mg/100g). Ghoora *et al* (2020) reported iron content of spinach microgreens 1.88 mg/100g.

Table 4.14 Mineral composition of spinach microgreens

Position	Seeds	10th day harvesting	20th day harvesting
Zinc (mg /100 g)			
Indoor	Soaked	35.71±0.30	27.18±0.02
	Unsoaked	35.03±0.02	27.05±0.04
Outdoor	Soaked	38.53±0.03	22.41±0.01
	Unsoaked	38.14±0.02	23.46±0.23
Iron (mg /100 g)			
Indoor	Soaked	23.76±0.17	22.25±0.03
	Unsoaked	21.60±0.13	19.78±0.20
Outdoor	Soaked	28.60±0.05	21.38±0.02
	Unsoaked	23.57±0.04	22.34±0.08
Calcium (mg /100 g)			
Indoor	Soaked	797.81±0.04	723.91±0.03
	Unsoaked	819.92±0.30	851.38±0.05
Outdoor	Soaked	1111.02±0.46	1031.39±0.04
	Unsoaked	1082.30±0.37	1031.21±0.02
Potassium (mg /100 g)			
Indoor	Soaked	110.23±0.11	92.18±0.06
	Unsoaked	100.64±0.05	90.10±0.06
Outdoor	Soaked	221.44±0.05	105.33±0.02
	Unsoaked	140.08±0.02	105.20±0.16
Magnesium (mg /100 g)			
Indoor	Soaked	656.71±0.44	639.79±0.02
	Unsoaked	683.30±0.07	646.41±1.63
Outdoor	Soaked	645.17±0.49	615.13±0.47
	Unsoaked	695.27±0.20	674.18±4.43
Sodium (mg /100 g)			
Indoor	Soaked	36.49±0.03	30.52±0.01
	Unsoaked	34.92±0.04	41.66±0.16
Outdoor	Soaked	38.84±0.02	35.83±0.04
	Unsoaked	35.92±0.02	38.96±0.01

Values are expressed as mean ±SD

The calcium content of spinach microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time was in the range of 723.91 to 1111.02 mg/100g. The high content of calcium was observed in spinach microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day with value of 1111.02 mg/100g DW. Just 100 g of spinach microgreens were meeting the daily requirements of calcium by 138.88% for an adult male and female, respectively (Table 4.15). Ghoora *et al* (2020) reported calcium content of spinach microgreens 20.1 mg/100g.

The potassium content of spinach microgreens cultivated indoor and outdoor with soaked and unsoaked seeds at different harvesting time is in the range of 90.10 to 221.44 mg/100g (Table 4.14). The high content of potassium is observed in spinach microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day with value of 221.44mg/100g DW. Just 100 g of spinach microgreens were meeting the daily requirements of potassium by 6.33 % for an adult male and female (Table 4.15). Sharma *et al* (2021) reported high iron content in spinach microgreens (161 mg/100g) than mature spinach (132 mg/100g). Ghoora *et al* (2020) reported potassium content of spinach microgreens was 69.7 mg/100g.

The magnesium, content of spinach microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 615.13 to 695.27 mg/100 g DW. The high content of magnesium was observed in spinach microgreens cultivated with unsoaked seeds at outdoor setting and harvested at 10th. Just 100 g of spinach microgreens were meeting the daily requirements of magnesium by 201.61% and 238.95 % for an adult male and female, respectively (Table 4.15). Ghoora *et al* (2020) reported magnesium content of spinach microgreens 71.2 mg/100g. Spinach microgreens reported as good source of magnesium contributing 14.4 percent to the reference daily intake (RDI).

The sodium content of spinach microgreens cultivated indoor and outdoor with soaked and unsoaked seeds and harvested at different growth stages was in the range of 30.52 to 38.96 mg/100 g DW. The high content of sodium was observed in spinach microgreens cultivated with unsoaked seeds at outdoor setting and harvested at 20th day i.e. 38.96 mg/100g. Ghoora *et al* (2020a) reported sodium content of spinach microgreens 20.8 mg/100g. Sharma *et al* (2021) reported 70.02 mg/100 sodium content in mature spinach whereas the spinach microgreens had 78.02 mg/100g.

Factorial ANOVA analysis of the results obtained by spinach microgreens cultivated with soaked and unsoaked seeds at different positions (indoor and outdoor) and harvested at 10^{th} and 20^{th} day after germination. Statistical results showed that there was significant effect

Table 4.15 Percent adequacy of vitamins and minerals of spinach microgreens

Position	Seeds	10th day harvesting			20th day harvesting		
β-carotene (µg /100 g DW)		**β-carotene**	**% adequacy**		**β-carotene**	**% adequacy**	
			Male (2760 µg /d)*	**Female (2340µg /d)***		**Male (2760 µg /d)***	**Female (2340 µg /d)***
Indoor	Soaked	677.38	24.54	28.95	548.15	19.86	23.43
	Unsoaked	696.37	25.23	29.76	506.36	18.35	21.64
Outdoor	Soaked	390.83	14.16	16.70	357.03	12.94	15.26
	Unsoaked	496.68	18.00	21.23	346.76	12.56	14.82
Ascorbic acid (mg /100 g DW)		**Ascorbic acid**	**% adequacy**		**Ascorbic acid**	**% adequacy**	
			Male (65 mg/d)*	**Female (55mg/d)***		**Male (65 mg/d)***	**Female (55mg/d)***
Indoor	Soaked	18.58	28.58	33.78	16.50	25.38	29.99
	Unsoaked	19.58	30.12	35.60	18.93	29.12	34.41
Outdoor	Soaked	20.43	31.43	37.14	19.67	30.26	35.76
	Unsoaked	19.96	30.71	36.29	19.29	29.67	35.07
Zinc (mg /100 g DW)		**Zinc**	**% adequacy**		**Zinc**	**% adequacy**	
			Male (14 mg/d)*	**Female (11 mg/d)***		Male (14 mg/d)*	Female (11 mg/d)*
Indoor	Soaked	35.71	255.10	324.67	27.18	194.12	247.06
	Unsoaked	35.03	250.21	318.45	27.05	193.21	245.91
Outdoor	Soaked	38.53	275.19	350.24	22.41	160.05	203.70
	Unsoaked	38.14	272.45	346.76	23.46	167.55	213.24
Iron (mg /100 g DW)		**Iron**	**% adequacy**		**Iron**	**% adequacy**	
			Male (11 mg/d)*	**Female (15 mg/d)***		**Male (11 mg/d)***	**Female (15 mg/d)***
Indoor	Soaked	23.76	216.03	158.42	22.25	202.24	148.31
	Unsoaked	21.60	196.39	144.02	19.78	179.79	131.84
Outdoor	Soaked	28.60	260.03	190.69	21.38	194.33	142.51
	Unsoaked	23.57	214.27	157.13	22.34	203.09	148.93

Position	Seeds	10th day harvesting			20th day harvesting		
Calcium (mg/100g DW)		Calcium	% adequacy		Calcium	% adequacy	
			Male (800 mg/d)*	Female (800 mg/d)*		Male (800 mg/d)*	Female (800 mg/d)*
Indoor	Soaked	797.81	99.73	99.73	723.91	90.49	90.49
	Unsoaked	819.92	102.49	102.49	851.38	106.42	106.42
Outdoor	Soaked	1111.02	138.88	138.88	1031.39	128.92	128.92
	Unsoaked	1082.30	135.29	135.29	1031.21	128.90	128.90
Potassium (mg/100g DW)		Potassium	% adequacy		Potassium	% adequacy	
			Male (3500mg/d)*	Female (3500mg/d)*		Male (3500mg/d)*	Female (3500mg/d)*
Indoor	Soaked	110.23	3.15	3.15	92.18	2.63	2.63
	Unsoaked	100.64	2.88	2.88	90.10	2.57	2.57
Outdoor	Soaked	221.44	6.33	6.33	105.33	3.01	3.01
	Unsoaked	140.08	4.00	4.00	105.20	3.01	3.01
Magnesium (mg/100g DW)		Magnesium	% adequacy		Magnesium	% adequacy	
			Male (320mg/d)*	Female (270mg/d)*		Male (320mg/d)*	Female (270mg/d)*
Indoor	Soaked	656.71	205.22	243.22	639.79	199.93	236.95
	Unsoaked	683.30	213.53	253.07	646.41	202	239.41
Outdoor	Soaked	645.17	201.61	238.95	615.13	166.25	227.82
	Unsoaked	695.27	217.27	257.50	674.18	210.68	249.69

Values are given in Mean ±SD

*Estimated Average Requirements for Indians, (EAR) ICMR-NIN2020 position ($F(1,16)=488.15, p<0.001$), duration ($F(1, 16)=1646.56$, $p<0.001$), soaking*position ($F(1,16)=21.61$, $p<0.001$), position*duration ($F(1, 16)=74.02$ $p<0.001$)and soaking* position* duration ($F(1,16)=21.0, p<0.001$) on nutrient content estimated among the indoor and outdoor cultivation with soaked and unsoaked seeds harvested at 2 stages i.e. 10th and 20th day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in spinach microgreens is by soaking the spinach microgreen seeds, cultivating them in outdoor location and harvesting at 10th day.

Table 4.16 Nutrient Quality Score of spinach microgreens

Position	Seeds	10th day harvesting	20th day harvesting
Nutrient Quality Score			
Indoor	Soaked	819.11±0.51	733.13±0.84
	Unsoaked	830.52±2.31	729.38±1.40
Outdoor	Soaked	925.68±0.61	719.94±0.65
	Unsoaked	890.19±0.59	753.51±3.35

Values are expressed as mean ±SD

Nutrient Quality Score

The NQS used for evaluating the overall nutritional quality of microgreens, instead of simply quantifying nutrient content to evaluate the variations of a single nutrient among different crops. Furthermore, for the first time, a NQS was used assess the effects of cultivation and harvesting conditions on nutritional quality of spinach microgreens. It is clearly seen from the Table 4.16 that NQS is highest at outdoor cultivation with soaked seeds and harvested at 10th day to get maximum overall nutritional quality.

4.2 Nutritional and biochemical composition of microgreens under different processing conditions

Harvested microgreens have high respiration rate, which affects the shelf life of fresh microgreens. Microgreens can be stored at ≤5°C temperature, for over 14 days (Kou *et al* 2013, Chandra *et al* 2012 and Xiao *et al* 2014b). Dehydration of microgreens may help in increase the shelf life of microgreens. Also dehydration reduces microbiological activity through reduced moisture content in the vegetable and fruits. In order to optimize best method of drying, harvested microgreens was dried via shade drying, Tray drying and microwave drying for removing moisture and improving the shelf life. Further retention of the nutrients which were most affected by the drying processing presented in figure 4.1-4.4.

The vitamins and bioactive compounds of broccoli microgreens under different processing conditions have been presented in Table 4.17. The statistical analysis showed that the β-carotene content of shade dried broccoli microgreens was significantly ($p<0.05$) higher than the oven and microwave dried broccoli microgreens. Park (1987) reported a significant positive correlation between high temperature and β-carotene degradation. Many studies had shown that the carotene losses were directly dependent on the method chosen for drying. The degradation of β-carotene from green leafy vegetables dried by oven (6-255%) was significantly higher as compared to the sun drying method (24-40%) (Aletor and Adeogun, 1995). A significant reduction in β-carotene content of drumstick and coriander leaves dried by sun and oven drying as compared to shade drying was reported by Joshi and Mehta (2010) and Vyankatrao (2014). The β-carotene content of fresh broccoli microgreens was 554.95 μg/100g that significantly reduced after drying. The percentage retention in β-carotene content was 72, 71 and 62 percent for shade, oven and microwave dried broccoli microgreens (Fig 4.1). Low-temperature drying of leafy vegetables retained more β-carotene content than high drying temperature i.e. sun, solar or cabinet drying (Negi and Roy 2000). At high temperature and oxidation, β-carotene lead to degradation and more precisely to isomerization (Karabulut *et al* 2007, Penicaud *et al* 2011, Coşkun *et al* 2013) which explains the higher β-carotene content of broccoli microgreens in shade drying as compared to oven and microwave drying in the present study .

Table 4.17 Vitamins, bioactive compounds and antioxidant activity of broccoli microgreens under different processing conditions

Parameters	Fresh microgreens	Processing conditions		
		Shade	Oven	Microwave
β-carotene (μg/100g)	554.95^{d}±6.30	400.54^{c}±0.76	392.22^{b}±0.20	344.75^{a}±0.51
Ascorbic acid (mg/100g)	25.16^{d}±0.03	10.88^{a}±0.56	13.16^{b}±13.16	21.69^{c}±0.36
Chlorophyll (g/100g)	10.85^{d}±0.57	1.47^{b}±0.17	0.48^{a}±0.01	3.90^{c}±0.12
Total phenolic content (mg GAE/100g)	2346.46^{a}±0.36	2645.88^{b}±112.41	2538.22^{b}±67.36	2594.78^{b}±3.88
Flavonoid (mg QE/100g)	820.24^{c}±0.78	673.82^{b}±0.81	671.46^{b}±1.14	382.32^{a}±2.21
Antioxidant activity (%)	90.68^{d}±0.99	41.54^{b}±1.42	33.03^{a}±1.06	81.90^{c}±00

Values are mean ± SD of triplicates
Values in rows followed by different superscript differ significantly ($p < 0.05$)

The statistical analysis showed that the ascorbic acid content of microwave dried broccoli microgreens was significantly ($p<0.05$) higher followed by shade and oven dried broccoli microgreens (Table 4.17). Microwave drying retained maximum ascorbic acid as compared to shade and tray drying. This is associated with high sensitivity of ascorbic acid to most assessable atmospheric constitutes like oxygen, light and temperature. Similar results were reported by Managa *et al* (2020) that there was more retention of ascorbic acid in microwave dried Chinese cabbage (30.5 mg/100g) than oven dried (23.6 mg/100g). Contrary to the present results, Khatoniar *et al* (2019) reported that the shade dried amaranth leaves had 11.47mg/100g of ascorbic acid content whereas microwave dried amaranth leaves had 10.68 mg/100g. Ascorbic acid was shown to degrade maximum during tray drying as high heat treatment led to maximum deterioration of ascorbic acid. Microwave drying retained maximum ascorbic acid as compared to shade and tray drying (Fig 4.2). The percentage retention in ascorbic acid was 43, 52 and 86 percent for shade, oven and microwave dried broccoli microgreens. This is associated with high sensitivity of ascorbic acid to most assessable atmospheric constitutes like oxygen, light and temperature. Similar results were reported by Managa et al (2020) that there was more retention of ascorbic acid in microwave dried Chinese cabbage (30.5 mg/100g) than oven dried (23.6 mg/100g). Contrary to the present results, Khatoniar et al (2019) reported that the shade dried amaranth leaves had 11.47mg/100g of ascorbic acid content whereas microwave dried amaranth leaves had 10.68 mg/100g. Ascorbic acid is the main vitamin contributing to antioxidant activities of green leafy vegetable and also most heat labile of all the vitamins present in the food (Sagar and Kumar 2007). The primary reason of reduction in vitamin C is the degradation of ascorbic acid to diketoglulonic acid on the application of heat treatment (Maharaj and Sankat 1990). The vitamin may also degrade due to prolonged duration of drying and presence of oxygen (Maharaj and Sankat 1990, Kiremire *et al* 2010) that explains more deterioration of ascorbic acid in shade drying as compared to tray drying in the present study.

The chlorophyll content of microwave dried broccoli microgreens was significantly ($p<0.05$) higher than shade and oven dried microgreens. Microwave drying enables to shorten dehydration time and controls undesirable biological transformations (Bondaruk *et al* 2007). Studies reported in literature have shown the method of drying like microwave and freeze drying with short duration led to maximum retention in chlorophyll (Karva 2008, Kamel *et al* 2013). Chlorophyll is sensitive to heat and its retention is dependent on temperature and duration of heat treatment (Negi and Roy 2000). Higher losses in shade drying in the present study may be due to longer time of drying. Microwave drying had higher retention of chlorophyll than shade and tray drying techniques as compared to fresh microgreens (Fig 4.3).

The chlorophyll content of fresh broccoli microgreens was 10.85 g/100g. The percent retention in chlorophyll was 14, 4 and 36 percent for shade, oven and microwave dried broccoli microgreens. Zheng-Wei et al (2004) reported the high retention of chlorophyll in leaves of chive with by freeze drying (97.1%) as compared to hot air drying (38.3%). Cesar *et al* (2003) reported a larger retention of chlorophyll pigments in basil leaves by microwave drying as compared to air-drying and freeze-drying (with or without blanching). Microwave drying evaporate the moisture quickly using a shorter time span that reduces the oxidation reaction thus, retaining the nutrients and bioactive compounds (Danso-Boateng 2013).

The total phenolic content of shade dried broccoli microgreens was higher as compared to the oven and microwave dried sample. High temperature treatment which led to decomposition of phenolic compounds rapidly (Chism and Haard, 1996; Chang *et al* 2006). Similar to the present results Kumar *et al* (2014) reported higher total phenolic content of shade dried roselle leaves than microwave and oven dried roselle leaves. The statistical analysis showed that the flavonoid content of shade dried broccoli microgreens was significantly ($p<0.05$) higher than the microwave dried broccoli microgreens. Kumar *et al* (2014) reported roselle leaves had high content of flavonoid dried in shade drying 19.09 g RE/kg whereas oven dried roselle leaves had 3.21 g RE/kg.

The antioxidant activity of microwave dried broccoli microgreens was significantly ($p< 0.05$) higher than the shade and oven dried broccoli microgreens. Similar results were found by Managa *et al* (2020) that antioxidant capacity (FRAP) of oven dried Chinese cabbage (1.38 mg of TEAC/100g) was lower than the microwave dried Chinese cabbage (2.94 mg of TEAC/100g). Kumar *et al* (2014) reported the antioxidant activity of roselle leaves was highest in freeze drying (91.47%) followed by shade drying (89.45%) and microwave drying (86.58%). Okmen and Bayindirli (1999) observed high antioxidant activity (DPPH) of microwave dried moringa leaves than oven dried moringa leaves. The antioxidant activity of fresh broccoli microgreens was significantly ($p <0.05$) higher than dried broccoli microgreens. The percent retention in antioxidant activity was 46, 37 and 90 percent in shade, oven and microwave drying (Fig 4.4), thus clearly indicating that microwave drying retained the maximum antioxidants. Retention of antioxidant activity decreases with increase in temperature and duration of drying (Demarchi *et al* 2013). Similar results were reported by Ozcan *et al* (2019) that microwave dried kiwi and pepino fruits had higher antioxidant activity than oven dried. The antioxidant activity was higher in dried samples due to the concentrated antioxidant compounds during drying process (Nunes *et al* 2016).

Table 4.18 Mineral content of broccoli microgreens under different processing conditions

Parameters (mg/100g)	Fresh microgreens	Processing conditions		
		Shade	Oven	Microwave
Zinc	37.89^{d}±0.09	8.40^{a}±0.20	9.87^{b}±0.71	11.33^{c}±0.04
Iron	59.97^{d}±0.11	31.32^{a}±0.35	36.55^{b}±0.50	41.62^{c}±0.17
Calcium	2990.16^{d}±0.04	2031.72^{b}±35.39	2038.00^{b}±17.65	2271.16^{c}±0.20
Potassium	341.53^{d}±0.54	249.02^{b}±0.90	201.73^{a}±1.12	511.79^{c}±0.79
Magnesium	713.81^{d}±1.54	664.88^{a}±0.34	687.74^{b}±0.42	698.43^{a}±0.04
Sodium	25.46^{d}±0.08	19.79^{a}±0.52	18.16^{a}±0.57	21.34^{c}±0.05

Values are expressed as mean ± SD

Values in rows followed by different superscript differ significantly ($p < 0.05$)

The mineral content like iron, calcium, potassium, magnesium and sodium were significantly ($p<0.05$) higher in microwave drying as compared to shade and tray drying except zinc was highest in tray drying (Table 4.18). Similar results were reported in literature were maximum retention of minerals was done through microwave or shade drying as compared to tray drying. The results of the present study were in agreement with the finding of Danso-Boateng (2013) as it was reported that microwave dried basil leaves preserve (22%) more iron as compared to shade (8%) and sun drying. In another study, Khatoniar *et al* (2019) reported the iron content of shade dried amaranth leaves (84.78 mg/100g) were lower than the microwave dried amaranth leaves(86.26 mg/100g). Joshi and Mehta (2010) also reported a maximum content of iron in shade dried drumstick leaves (24mg/100g) and minimum retention in oven dried (19 mg/100g).Khanum *et al* (2013) reported the zinc content of coriander leaves dried by sun drying and oven drying was almost same with value of 10.2 and 10.3 µg /100g, respectively.

Khatoniar *et al* (2019) reported high concentration of calcium in microwave dried amaranth leaves (2906.21mg/100g) whereas significantly lower concentration in shade dried amaranth leaves (2886.43 mg/100g). Similarly, Sonkamble and Pandhure (2017) reported maximum retention of calcium was in shade dried spinach leaves i.e. 26.05 mg/100g than oven dried spinach leaves i.e. 17.31 mg/100g. Microwave drying was considered as a best method for retention of other minerals like potassium, magnesium and sodium. Oven dried broccoli microgreens had lower content of potassium which is in line with Liman *et al* (2014) who reported the maximum reduction of potassium content of spinach in oven drying (13.08 mg/100g) as compared to sun (18.41 mg/100g) and moisture analyzer drying method (18.16

mg/100g). Khanum *et al* (2013) reported the magnesium content was minimum in sun dried (148.7 µg / g) coriander leaves than oven dried one (390.1 µg / g). Liman *et al* (2014) reported the sodium content of sun dried spinach was 21mg/100g where it reduced to 17.91 mg/100g in oven dried spinach microgreens.

Thus, it could be concluded that microwave drying method was the best method for the drying of broccoli microgreens. Short period of heat treatment by microwave drying led to higher nutrient retention and also better quality attributes such as flavour, colour and texture.

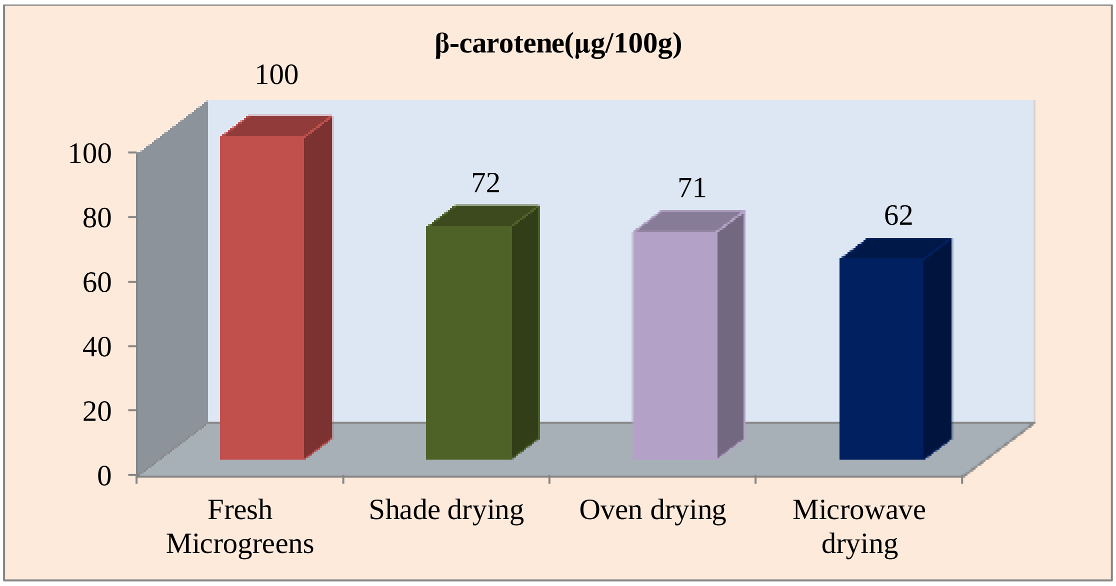

Fig 4.1 Retention of β-carotene content of fresh, shade, oven and microwave dried broccoli microgreens

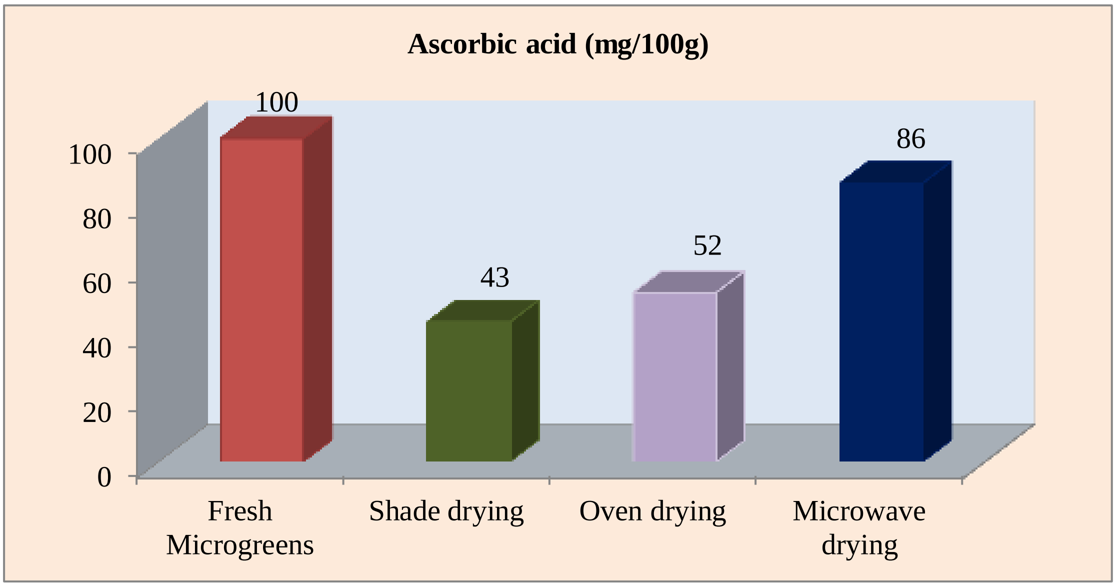

Fig 4.2 Retention of Ascorbic content of fresh, shade, oven and microwave dried broccoli microgreens

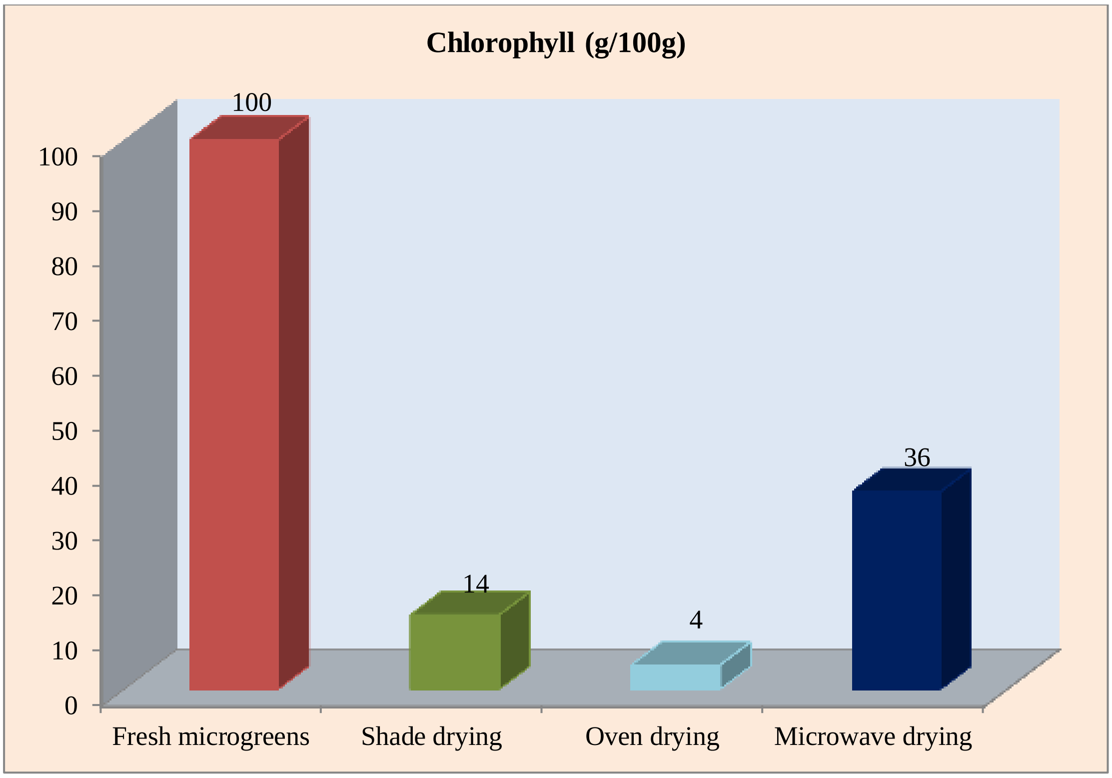

Fig. 4.3 Retention of chlorophyll content of fresh, shade, oven and microwave dried broccoli microgreens

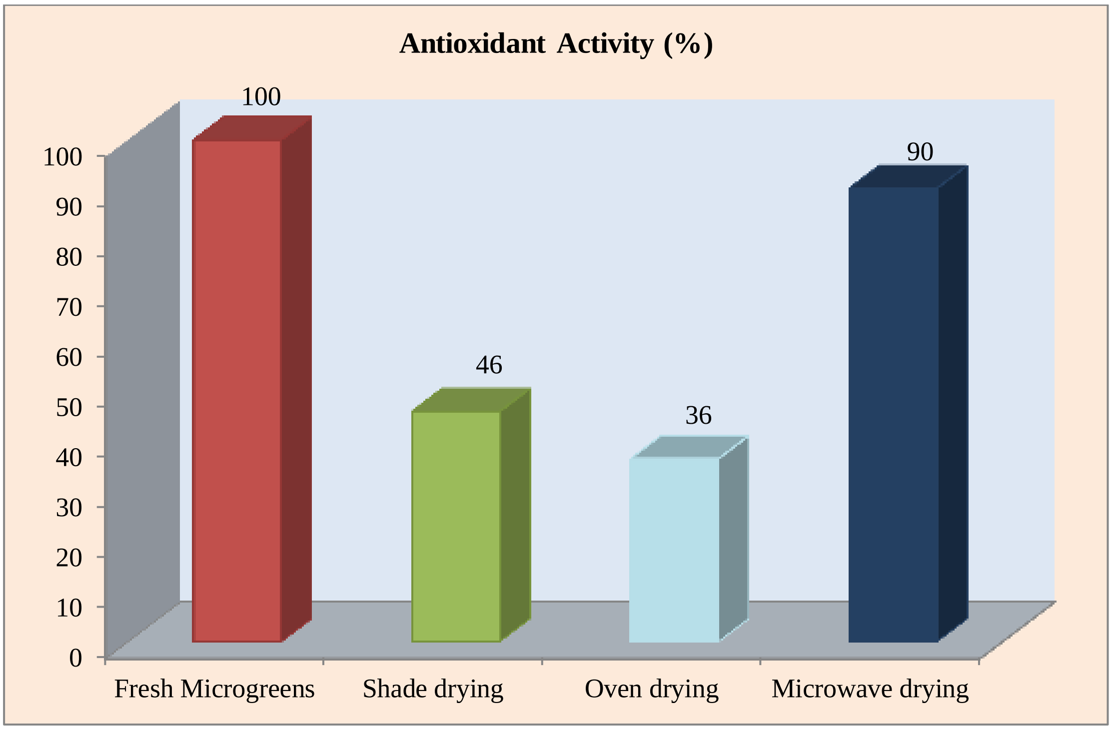

Fig 4.4 Retention of Antioxidant activity of fresh, shade, oven and microwave dried broccoli microgreens

4.3 Compare broccoli microgreens with sprouts and mature leaves in terms of nutritional and biochemical composition

Generally, vegetable leaves harvested at a young stage of development naturally possess a large portion of healthy compounds such as vitamin C and glucosinolates (Nordmark *et al* 2009). In the study conducted by Pandjaitan *et al* (2005), mid-maturity spinach leaves were observed with higher levels of total phenolics, total flavonoids and antioxidant capacity when compared with immature and mature leaves. Young baby spinach leaves have shown retention of total flavonoid concentrations higher than the baby spinach leaves harvested at 46 days (Bergquist *et al* 2005). On the contrary, it was reported that the total flavonoid levels decreased in response to cabbage maturity (Kim *et al* 2004). Thus, the growth stages of plant affect the nutrient content of the plant. In order to optimise the effect of different growth stages, microgreens were compared with sprouts and mature leaves of broccoli in terms of nutritional and biochemical composition.

Vitamins, Bioactive compounds and Antioxidant activity

Vitamins, bioactive compounds and antioxidant activity of broccoli under different growth stages have been presented in Table 4.19.

The β-carotene content of broccoli at different growth stages were 344.75, 110.63 and 248.42 μg/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the β-carotene content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Ebert et al (2014) reported the β-carotene content of amaranth microgreens was 1.87 mg/100g whereas amaranth sprouts had lower β-carotene content 0.10 mg/100g. Contrary to the results, Ghoora *et al* (2020) reported that the mature leaves of fenugreek and roselle had significantly higher ($p < 0.05$) β-carotene content than their respective microgreens.

The ascorbic acid content of broccoli at different growth stages were 21.69, 14.05 and 10 mg/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the ascorbic acid content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Ghoora *et al* (2020) compared microgreens of fenugreek, spinach and roselle with their respective mature counterparts and found significant ($p<0.05$) higher ascorbic acid in microgreens. Bergquist *et al* (2005) reported that harvesting of spinach at younger (17-18 days after sowing) stage had increased ascorbic acid content 460 mg/kg than baby spinach 140 mg/kg (29-30 days after sowing). As ascorbic acid is thermolabile in nature and leafy vegetable at mature stages are generally consumed after cooking, the human body is unable to harness the benefits of ascorbic acid from them. On the other hand, the human body can fully access the ascorbic acid present in microgreens as they generally consumed in fresh.

Table 4.19 Vitamins, bioactive compounds and antioxidant activity of microgreens under different growth stages

Parameters	Growth stage		
	Microgreens	**Sprouts**	**Mature leaves**
β-carotene (μg/100g)	$344.75^{c}\pm0.51$	$110.63^{a}\pm0.45$	$248.42^{b}\pm3.23$
Ascorbic acid (mg/100g)	$21.69^{c}\pm0.36$	$14.05^{b}\pm13.16$	$10.00^{a}\pm1.21$
Chlorophyll (g/100g)	$3.90^{c}\pm0.12$	$0.31^{a}\pm0.37$	$1.69^{b}\pm0.92$
Total phenolic content (mg GAE/100g)	$2594.78^{c}\pm3.88$	$357.20^{a}\pm19.45$	$539.63^{b}\pm0.47$
Flavonoid (mg QE/100g)	$382.32^{c}\pm2.21$	$55.30^{a}\pm0.88$	$143.67^{b}\pm0.25$
Antioxidant activity (%)	$81.90^{c}\pm00$	$29.93^{a}\pm0.40$	$58.06^{b}\pm2.34$

Value expressed as mean±SD
Value in rows followed by different superscript differ significantly at 5% level

The chlorophyll content of broccoli at different growth stages were 3.90, 0.31 and 1.69 g/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the chlorophyll content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Pandey *et al* (2018) reported that mature broccoli contained 0.84g/100g of chlorophyll whereas Tan *et al* (2020) reported lower chlorophyll content i.e. 0.33 mg/g of broccoli microgreens.

The total phenolic content of broccoli at different growth stages were 2594.78, 357.20 and 539.63 mg GAE/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the total phenolic content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. López-Cervantes *et al* (2013) reported total phenolic content was significantly increased from seeds to sprouts. Similarly Hanlon and Barnes (2011) reported that radish sprouts had significantly greater concentration (6.9 folds) of total phenolic than mature one. Senevirathne *et al* (2019) reported the total phenolic content of green peas microgreens (1871 GAE mg/100g) was higher than the sprouts(686 GAE mg/100g) and seeds (469 GAE mg/100g) of green peas.

The flavonoid content of broccoli at different growth stages were 382.32, 55.30 and 143.67 mg QE/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the flavonoid content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Bergquist *et al* (2005) investigated that harvesting of spinach at younger (6 days) stage had increased flavonoid content 21.8 mg/100 DW than baby spinach 15.7 mg/100g DW(24 days). The antioxidant activity of broccoli at different growth stages were 81.90, 29.93 and 58.06 percent for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the antioxidant activity of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Hanlon and Barnes (2011) investigated antioxidant activity and the FRAP values for the sprouts (50.0 µmol AA Equiv/g) were significantly greater than for mature radishes (36.6 µmol AA Equiv/g).

The zinc content of broccoli at different growth stages was 11.33, 2.72 and 5.46 for microgreens, sprouts and mature one, respectively (Table 4.20). The statistical analysis showed that the zinc content of broccoli microgreens was significantly ($p<0.05$) higher than the sprout and mature broccoli. Yadav *et al* (2018) reported the higher concentration of microgreens as compared to the mature counterparts. Similar to the present results Pinto *et al* (2015) observed higher concentration of zinc in lettuce microgreens compared to the mature one. The iron content of broccoli at different growth stages were 41.62, 10.36 and 18.47 mg/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the iron content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Sharma *et al* 2021 reported higher iron content of spinach microgreens (4.03 mg/100g) than the mature spinach (3.59 mg/100g). Zieliński *et al* (2005) reported calcium content of Cruciferous sprouts had significantly higher content of iron as compare to the seeds.

Table 4.20 Mineral content of broccoli under different growth stages

Parameters	Growth stage		
	Microgreens	**Sprouts**	**Mature leaves**
Zinc	$11.33^{a}\pm0.04$	$2.72^{a}\pm0.39$	$5.46^{b}\pm0.11$
Iron	$41.62^{c}\pm0.17$	$10.36^{a}\pm0.06$	$18.47^{b}\pm0.15$
Calcium	$2271.16^{c}\pm0.20$	$30.57^{a}\pm0.16$	$87.90^{b}\pm0.19$
Potassium	$511.79^{c}\pm0.79$	$107.52^{a}\pm1.79$	$316.57^{b}\pm1.37$
Magnesium	$698.43^{c}\pm0.04$	$156.10^{a}\pm1.86$	$356.02^{b}\pm0.37$
Sodium	$21.34^{b}\pm0.05$	$17.92^{a}\pm0.20$	$27.44^{c}\pm2.34$

Value are expressed as mean±SD
Value in rows followed by different superscript differ significantly at 5% level

The calcium content of broccoli at different growth stages were 2271.16, 30.57 and 87.90 mg/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the calcium content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Khader and Rama (2003) studied the effect of maturity of Amaranthus blitum on macro minerals content. They observed as plant matured from stage I (15 days) to stage II (30 days), there is increased in calcium level but decreased from stage II (30 days) to stage III (45 days). Pinto *et al* (2015) reported that the calcium content was significantly higher in lettuce microgreens than the mature lettuce.

The potassium content of broccoli at different growth stages were 511.79, 107.52 and 316.52 mg/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the potassium content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. In line with this findings, a similar trend was observed by Waterland *et al* (2017) potassium content was significantly higher in kale microgreens than the mature one. Yadav *et al* (2018) reported the potassium content of microgreens was higher than the leafy vegetables. The concentration of potassium tends to be high in microgreens due of its active involvements in photosynthesis, respiration, water homeostasis (Kopsell *et al* 2012). Contrary to the present results, Pinto *et al* (2015) reported higher content of potassium in mature lettuce than the lettuce microgreens. The results vary may be due to the different plant type, climate condition and soil composition.

The magnesium content of broccoli at different growth stages were 698.43, 156.10 and 356.02/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the magnesium content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Singh *et al* (2018) reported significantly higher magnesium content in radish microgreens (418 mg/100g) as compared to mature radish (214 mg/100g). Whereas radish sprouts (3.75 mg/g) had very low content of magnesium as compared to radish microgreens (Zieliński *et al* 2005). The sodium content of broccoli at different growth stages were 21.34, 17.92 and 27.44 mg/100g for microgreens, sprouts and mature leaves, respectively. The statistical analysis showed that the sodium content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. Similar to our results, Pinto *et al* (2015) reported that sodium content was higher in mature lettuce than lettuce microgreens. Whereas Sharma *et al* (2021) reported higher sodium content of spinach microgreens (78.02 mg/100g) than the mature spinach (70.02 mg/100g).

The growth stages of plant have an effect on the nutrient content of the plant. The statistical analysis showed that the β-carotene, ascorbic acid, chlorophyll, total phenolic content, flavonoid content, minerals (Ca, Mg, Zn, Fe and K) and antioxidant activity of broccoli microgreens were significantly ($p<0.05$) higher than the broccoli mature leaves (Fig 4.5-4.8). The data given in this study indicated that broccoli microgreens were an excellent source of vitamin, mineral and bioactive compounds as compared to the mature and sprouts. Broccoli microgreens also represent a new vegetable crop which comes under ready to eat vegetables assigned to direct consumption in fresh form and also be used as components in human diet.

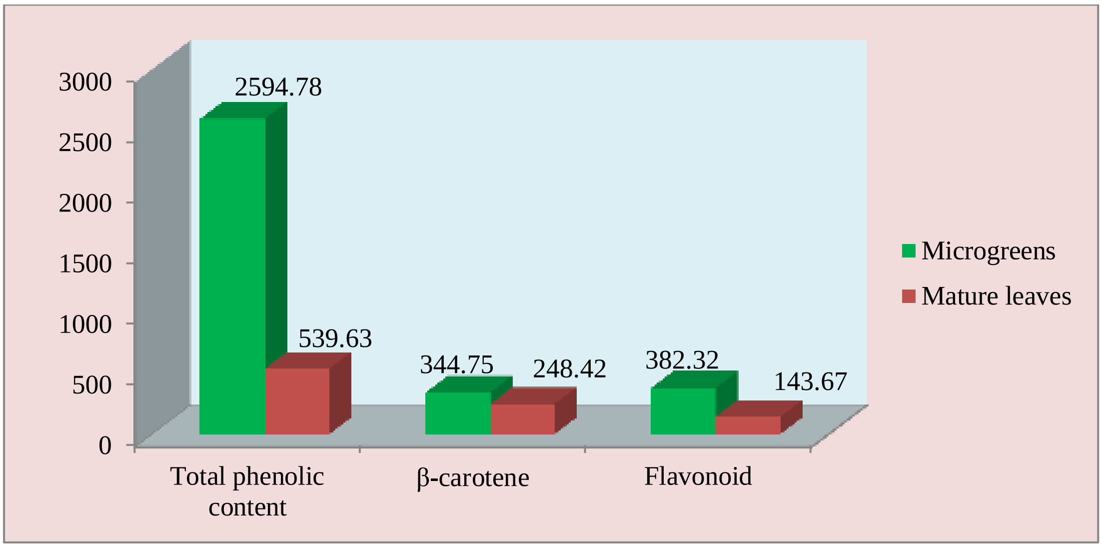

Fig 4.5 TPC, β-carotene and flavonoid of broccoli microgreens and mature leaves

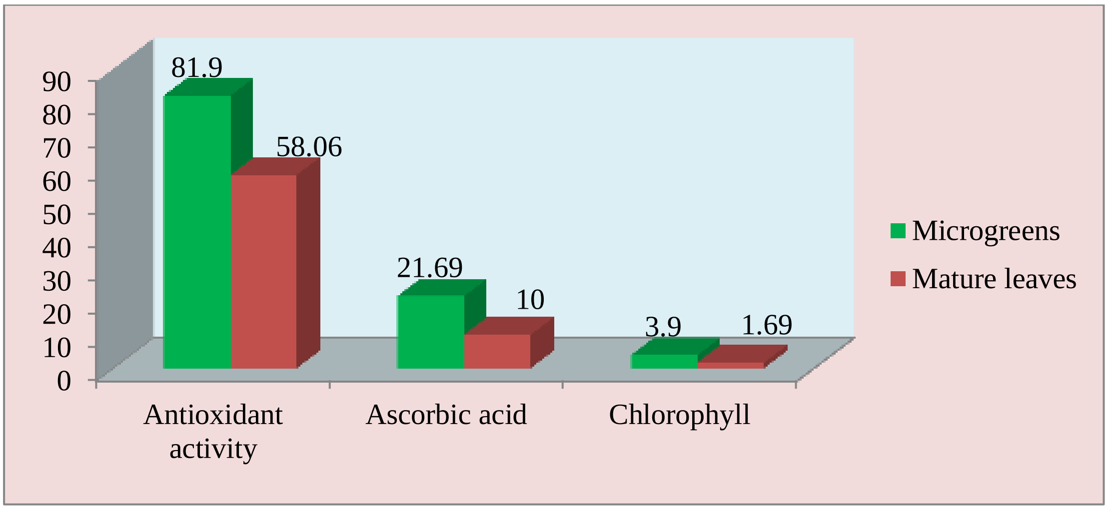

Fig 4.6 Antioxidant activity, ascorbic acid and chlorophyll content of broccoli microgreens and mature leaves

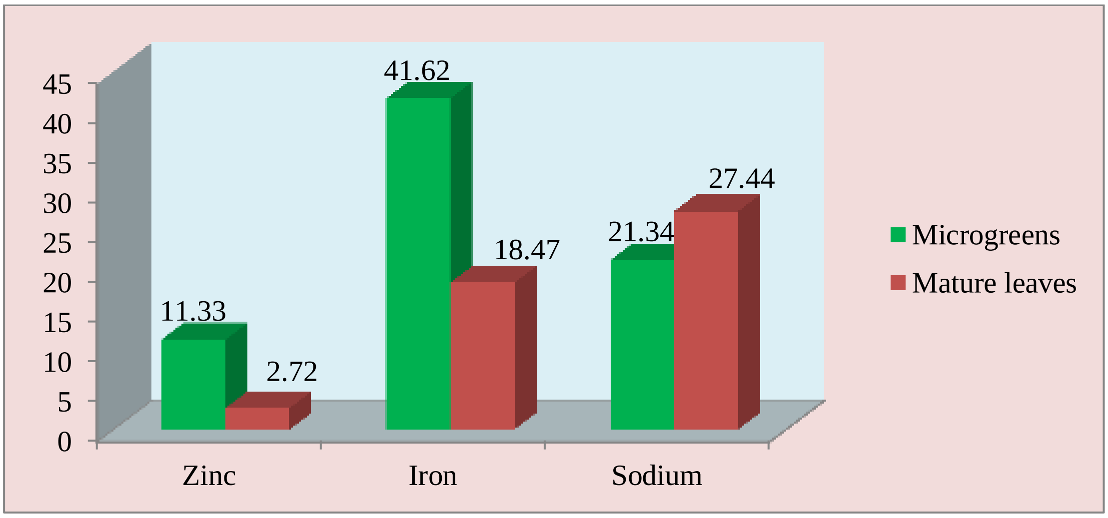

Fig 4.7 Zinc, iron and sodium content of broccoli microgreens and mature leaves

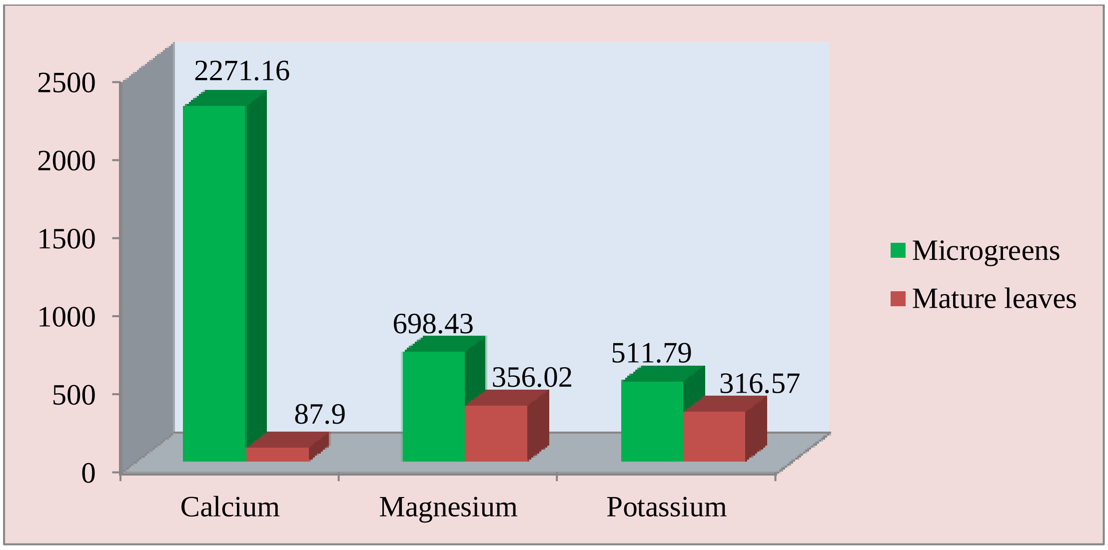

Fig 4.8 Calcium, magnesium and potassium content of broccoli microgreens and mature leaves

4.4 Development of health foods from microgreens obtained from optimized production and processing conditions

Development of various health foods from broccoli microgreens

Various health foods developed from the best variant (broccoli microgreens) in terms of nutritional and biochemical composition of microgreens (Fresh, Juice and Powder).Eight products including five from fresh microgreens, two from microgreens juice and one from powder of microgreens were developed. The developed products were organoleptically evaluated by semi expert panel of 10 judges from the Department of Food and Nutrition using 9-point hedonic rating scale. Organoleptic evaluations of developed products were discussed below.

4.4.1 Fresh microgreens

Five health products were developed from fresh microgreens i.e. missi roti, salad and incorporated in cooked dal and vegetable.

1. Missi roti

The Indian roti (flatten bread) is wholesome and healthy and often a staple in Indian household. It is the most commonly consumed cereal product in India and comes with large array of variety and shape. A recent development has been in the form of highly nutritious microgreens incorporated *missi roti*, which is not only preferred for its taste but also add colour and variety compiled with many nutrients. *Missi roti* prepared with whole wheat flour, Bengal gram flour, fenugreek leaves, coriander leaves, green chilies and salt was taken as control (C). Three samples of *missi roti* were prepared by supplementing fresh broccoli microgreens by 15%, 20% and 25% of control (Plate 2). The mean score for organoleptic

Table 4.21 Organoleptic scores for *Missi roti* supplemented with fresh broccoli microgreens

Missi roti		Colour	Appearance	Texture	Aroma	Taste	Overall acceptability
Control (C)		$7.70^{a} \pm 0.82$	$7.50^{a} \pm 0.85$	$7.50^{a} \pm 0.85$	$7.80^{a} \pm 0.78$	$8.0^{a} \pm 0.66$	$7.90^{ab} \pm 0.61$
Experimental	MGR1 (15%)	$7.4^{a} \pm 0.69$	$7.60^{a} \pm 0.51$	$7.50^{a} \pm 0.52$	$7.70^{a} \pm 0.48$	$7.90^{a} \pm 0.56$	$7.60^{a} \pm 0.47$
	MGR2 (20%)	$8.0^{ab} \pm 0.47$	$7.90^{ab} \pm 0.56$	$7.80^{a} \pm 0.42$	$7.80^{a} \pm 0.78$	$7.80^{a} \pm 0.91$	$8.0^{ab} \pm 0.47$
	MGR3 (25%)	$8.60^{b} \pm 0.51$	$8.50^{b} \pm 0.52$	$8.0^{a} \pm 0.94$	$8.20^{a} \pm 0.91$	$8.30^{a} \pm 0.94$	$8.50^{b} \pm 0.68$

Values are expressed as mean ± SD

Values in rows followed by different superscript differ significantly ($p < 0.05$)

Control- product developed with standard recipe

Experimental-product developed by incorporating fresh broccoli microgreen at different levels into standard recipe

Control (C)

MGR1 (15%)

MGR2 (20%)

MGR3 (25%)

Plate 2: Missi roti variants served for sensory evaluation

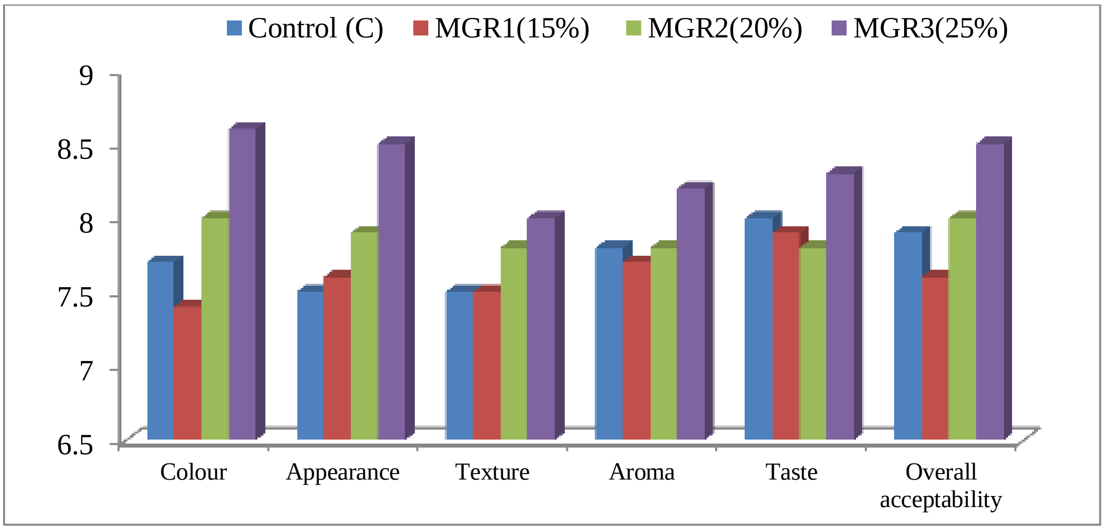

Fig 4.9 Organoleptic scores for *Missi roti* supplemented with fresh broccoli microgreens

evaluation of *missi roti* supplemented with fresh broccoli microgreens by semi trained panel of 10 judges using 9-point hedonic rating scale is presented in Table 4.21 and Fig. 4.9. The mean overall acceptability score of control and MGR 2 was at par. The MGR3 (25%) scored the highest point for all the parameters with 8.5 overall acceptability. The scores of MGR3 treatment found to be quite higher in the range 8 to8.60 than the scores of MGR2 (20%) in the range 7.80 to 8 for all the parameters followed by MGR1 (15%). Similar trend was found by Ghoora and Srividya (2017) who developed fenugreek flatbread (methi roti) incorporated fenugreek microgreens and reported that it was highly acceptable with overall acceptability score of 7.81 out of 9. Whereas mature broccoli powder was incorporated at 15% in missi roti was highly acceptable reported by Madhu and Kochhar (2014).

In term of colour, appearance and overall acceptability, a significant difference was observed in control (7.70, 7.50 and 7.90) and MGR3 (8.60, 8.50 and 8.50). Therefore, it can be concluded that the supplementation of broccoli microgreens have positive impact on traditional recipe of *missi roti*.

2. Dal* and *cooked vegetable

The organoleptic scores given for fresh broccoli microgreens incorporated *dal* samples are presented in Table 4.22. Five samples were prepared using split yellow lentil dal and cauliflower &potato (aloo gobi) as control, respectively and test samples were supplemented with fresh broccoli microgreens at 2.5%, 5%, 7.5% and 10% of levels (Plate 3-4). It was observed that organoleptic score for *dal* was increased with increased the level of microgreens for certain level (5%) and then starts decreasing with increase the level of microgreens. The highest score for all the sensory parameters were obtained by MGD2 with overall acceptability of 8.30, followed by MGD1 and MGD3 with same score of overall

Table 4.22 Organoleptic scores for cooked *dal* and *vegetable* supplemented with fresh broccoli microgreens

Product		Colour	Appearance	Texture	Aroma	Taste	Overall acceptability
Dal							
Control (C)		$6.80^{ab}\pm0.78$	$6.90^{ab}\pm0.87$	$7.0^{ab}\pm0.66$	$7.10^{ab}\pm0.73$	$7.10^{b}\pm0.73$	$6.90^{ab}\pm0.73$
Experimental	MGD1 (2.5%)	$7.70^{bc}\pm0.67$	$7.50^{bc}\pm0.85$	$7.70^{bc}\pm0.94$	$7.70^{bc}\pm0.82$	$7.90^{bc}\pm0.87$	$7.80^{bc}\pm0.78$
	MGD2 (5%)	$8.10^{c}\pm0.73$	$8.20^{c}\pm0.63$	$8.0^{c}\pm0.47$	$8.30^{c}\pm0.48$	$8.20^{c}\pm0.63$	$8.30^{c}\pm0.67$
	MGD3 (7.5)	$7.90^{c}\pm0.99$	$7.80^{bc}\pm0.91$	$7.60^{bc}\pm0.84$	$7.80^{bc}\pm0.78$	$7.70^{bc}\pm0.94$	$7.80^{bc}\pm1.03$
	MGD4 (10%)	$6.20^{a}\pm1.03$	$6.20^{a}\pm1.03$	$6.10^{a}\pm0.56$	$6.20^{a}\pm0.78$	$6.0^{a}\pm0.66$	$6.25^{a}\pm0.79$
Cooked vegetable							
Control (C)		$7.40^{a}\pm0.69$	$6.90^{a}\pm0.73$	$7.30^{a}\pm1.0$	$7.10^{ab}\pm0.56$	$7.10^{ab}\pm0.56$	$7.20^{a}\pm0.63$
Experimental	MGV1 (2.5%)	$7.50^{a}\pm0.85$	$7.0^{ab}\pm0.66$	$7.10^{a}\pm0.56$	$7.40^{ab}\pm0.96$	$7.30^{ab}\pm0.67$	$7.10^{a}\pm0.31$
	MGV2 (5%)	$7.60^{a}\pm0.51$	$7.80^{bc}\pm0.78$	$7.80^{ab}\pm0.42$	$7.90^{bc}\pm0.56$	$7.60^{bc}\pm0.69$	$7.80^{b}\pm0.42$
	MGV3 (7.5%)	$8.60^{b}\pm0.69$	$8.50^{c}\pm0.52$	$8.20^{b}\pm0.42$	$8.50^{c}\pm0.52$	$8.40^{c}\pm0.69$	$8.75^{c}\pm0.42$
	MGV4 (10%)	$6.90^{a}\pm0.73$	$7.10^{ab}\pm0.56$	$7.10^{a}\pm0.73$	$6.70^{a}\pm0.67$	$6.70^{a}\pm0.82$	$6.74^{a}\pm0.42$

Values are expressed as mean ± SD

Values in rows followed by different superscript differ significantly ($p < 0.05$)

Control- product developed with standard recipe

Experimental-product developed by incorporating fresh broccoli microgreens at different levels into standard recipe

Control (C)

MGD1 (2.5%)

MGD2 (5%)

MGD3 (7.5%)

MGD4 (10%)

Plate 3: Cooked dal variants served for sensory evaluation

Control (C)

MGV1 (2.5%)

MGV2 (5%)

MGV3 (7.5%)

MGV4 (10%)

Plate 4: Cooked veg variants served for sensory evaluation

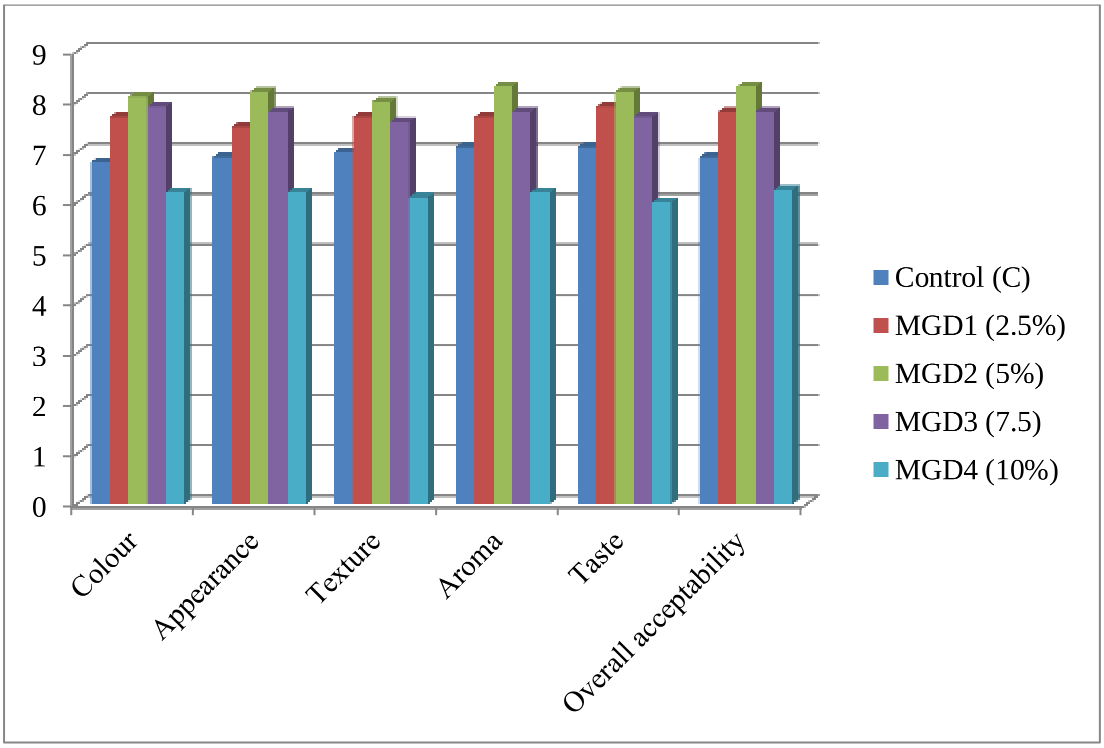

Fig 4.10 Organoleptic scores for *dal* supplemented with fresh broccoli microgreens

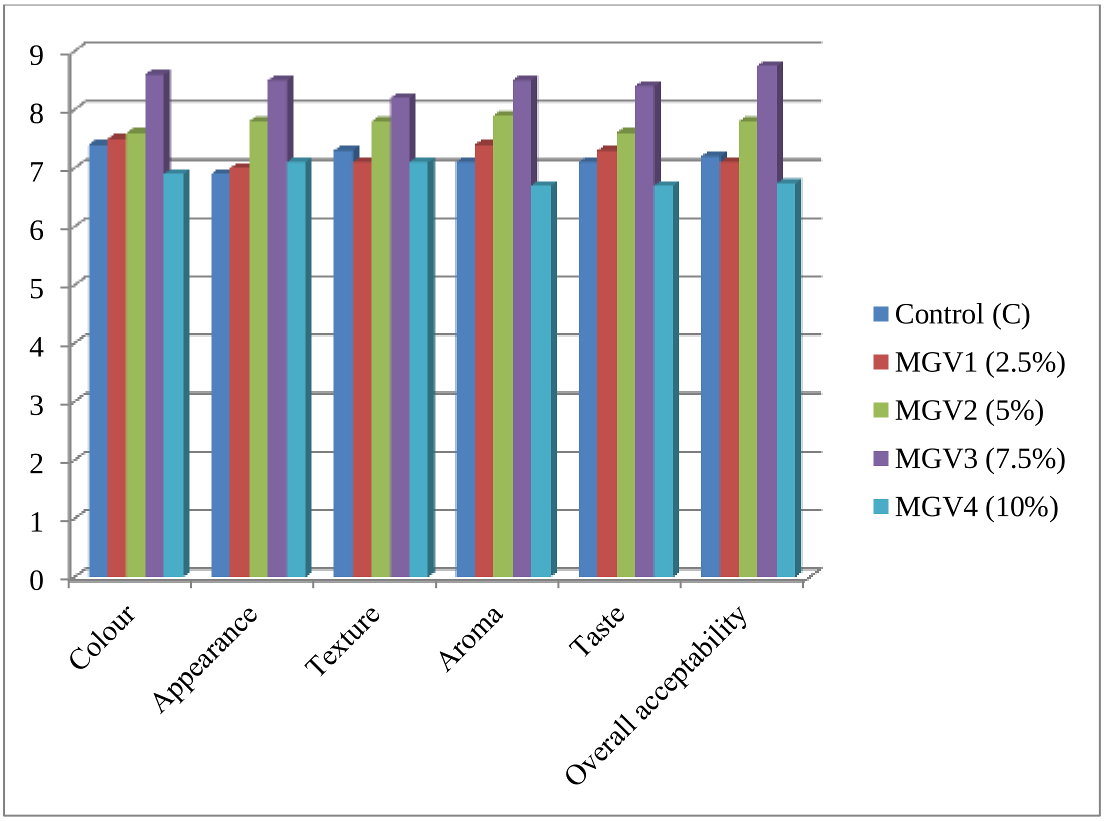

Fig 4.11 Organoleptic scores for *cooked vegetable* supplemented with fresh broccoli microgreens

acceptability (7.80). The mean scores for all the sensory parameters were in the range 6.80-7.10 for control and for significantly acceptable MGD2 in the range 8-8.30, which were liked very much (fig 4.10). It is clearly observed from the Table 4.22 that with increase in the percent of broccoli microgreens (10%) there was gradual decrease in all the sensory parameters. It might be because at 10 percent level, the prepared dal becomes more fibrous in texture than desired. For cooked vegetable, it was observed that organoleptic score for *cooked vegetable* was increased with increased the level of microgreens till 7.5 percent and then starts decreasing with increase the level of broccoli microgreens. The MGV3 (7.5%) scored the highest point for all the sensory parameters with 8.75 overall acceptability (Fig 4.11).

The mean scores for all the sensory parameters were in the range 6.90-7.40 for control and for significantly acceptable MGV3 in the range 8.20-8.75. Same trend was observed by Kachhawa and Chawla (2017) after the supplementation of drumstick leaves to the dal, with increase in percent of drumstick leaves, sensory parameters significantly decreased i.e. the mean sensory score for 30 percent incorporated drumstick leaves in dal ranges 7.65-8.05 and for 60 percent of drumstick leaves incorporated dal ranges 6.97-7. Nambiar and Parnami (2008) reported the overall composite score ranging from 3.06-3.53 out of 5 for kabuli channa, desi channa and mung dal incorporated with fresh leaves of drumstick. Similar results were reported by Kaur and Kochar (2009). Incorporation of underutilized carrot greens in saag (80%) and curry (40%) had mean overall acceptability of 6.20 and 6.80. Kushwaha and Chawla (2017) reported 25% drumstick leaves powder and amaranth leaves powder supplemented dal was liked very much with mean sensory score of 7.65 and 7.30, respectively**.** Also, Ghoora and Srividya (2017) prepared dal and cooked vegetable supplemented with fenugreek microgreens with overall acceptability of 7.92 and 8.64, respectively.

3. Salad

Two varieties of salads i.e. *tofu salad* and *russian salad* were prepared with control and test samples supplemented with fresh broccoli microgreens at 20%, 30% and 40% levels (Plate 5-6). It was observed that highest mean overall acceptability score of *tofu salad* was 8.30 for MGT3 (40%) (Table 4.23). The mean scores for all the sensory parameters of *tofu salad* were in the range 7.50-8 for control and for significantly acceptable MGT3 in the range 7.90-8.30, which were liked very much (fig. 4.12). In the present study, one of the main sensory attribute i.e. colour and texture was not affected upto the level of 40%. Michell *et al* (2020) reported consumer acceptability of raw broccoli microgreens in term of flavor, texture, and overall acceptability, broccoli microgreens had the highest, which may be due to their sweet and grassy sensory attributes and low bitter, astringent, sour, and heat sensory attributes.

Control (C)

MGT1 (20%)

MGT2 (30%)

MGT3 (40%)

Plate 5: Tofu salad variants served for sensory evaluation

Control (C)

MGS1 (20%)

MGS2 (30%)

MGS3 (40%)

Plate 6: Russian salad variants served for sensory evaluation

Table 4.23 Organoleptic scores for *salads* supplemented with fresh broccoli microgreens

Product		**Colour**	**Appearance**	**Texture**	**Aroma**	**Taste**	**Overall Acceptability**
Tofu salad							
Control (C)		$7.50^{a}\pm0.850$	$8.0^{a}\pm0.81$	$7.90^{a}\pm0.87$	$8.0^{a}\pm0.66$	$7.90^{a}\pm0.73$	$7.90^{a}\pm0.73$
Experimental	MGT1 (20%)	$7.90^{a}\pm0.31$	$7.60^{a}\pm0.69$	$7.70^{a}\pm0.67$	$7.30^{a}\pm0.82$	$7.50^{a}\pm0.85$	$7.90^{a}\pm0.73$
	MGT2 (30%)	$7.60^{a}\pm0.69$	$7.70^{a}\pm0.82$	$7.60^{a}\pm0.84$	$7.30^{a}\pm0.82$	$7.80^{a}\pm1.13$	$7.70^{a}\pm0.67$
	MGT3 (40%)	$8.20^{a}\pm1.13$	$7.90^{a}\pm0.99$	$7.90^{a}\pm0.87$	$8.0^{a}\pm1.15$	$8.0^{a}\pm1.15$	$8.30^{a}\pm0.94$
Russian salad							
Control (C)		$7.35^{a}\pm0.57$	$7.30^{a}\pm0.74$	$6.90^{a}\pm0.31$	$7.20^{a}\pm0.42$	$7.10^{a}\pm0.31$	$7.00^{a}\pm00$
Experimental	MGS1 (20%)	$8.0^{b}\pm0.66$	$7.65^{a}\pm0.66$	$7.35^{a}\pm0.47$	$7.65^{a}\pm0.47$	$7.55^{a}\pm0.68$	$7.60^{b}\pm0.45$
	MGS2 (30%)	$8.05^{b}\pm0.15$	$7.90^{a}\pm0.73$	$7.60^{a}\pm0.84$	$7.65^{a}\pm0.47$	$7.70^{a}\pm0.48$	$7.90^{b}\pm0.31$
	MGS3 (40%)	$7.55^{ab}\pm0.59$	$7.35^{a}\pm0.88$	$7.25^{a}\pm0.85$	$7.30^{a}\pm0.74$	$7.55^{a}\pm0.59$	$7.55^{b}\pm0.59$

Values are expressed as mean ± SD

Values in rows followed by different superscript differ significantly ($p < 0.05$)

Control- product developed with standard recipe

Experimental-product developed by incorporating fresh broccoli microgreens at different levels into standard recipe

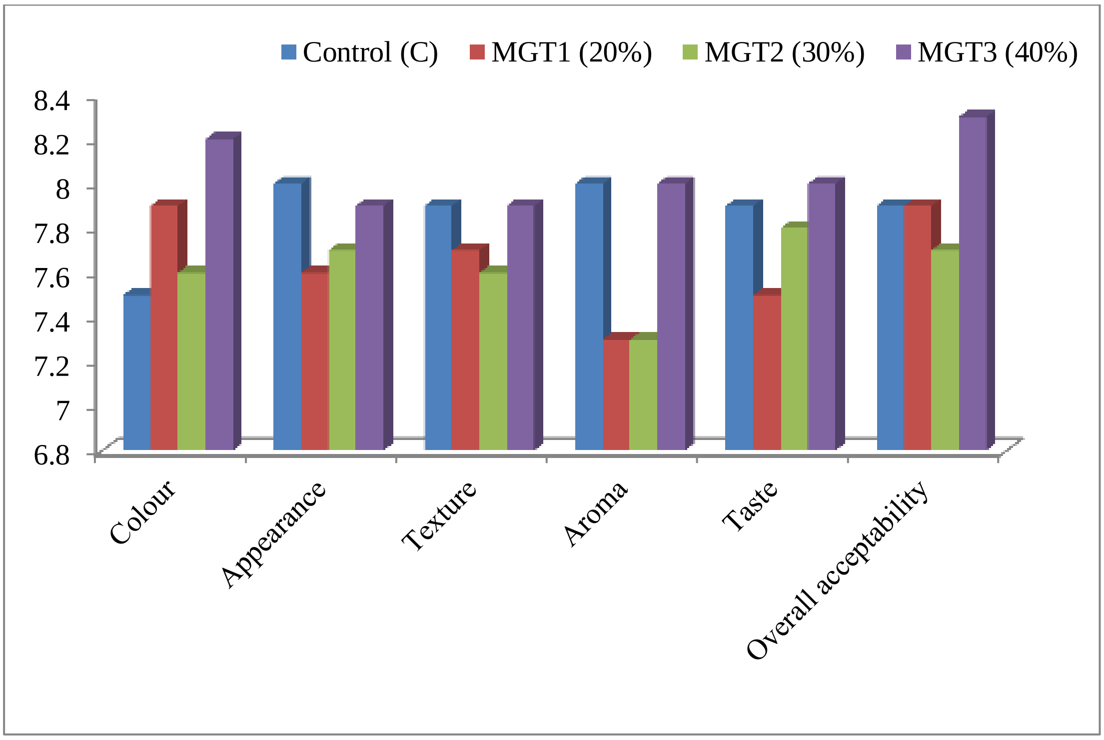

Fig 4.12 Organoleptic scores for tofu salad supplemented with fresh broccoli microgreens

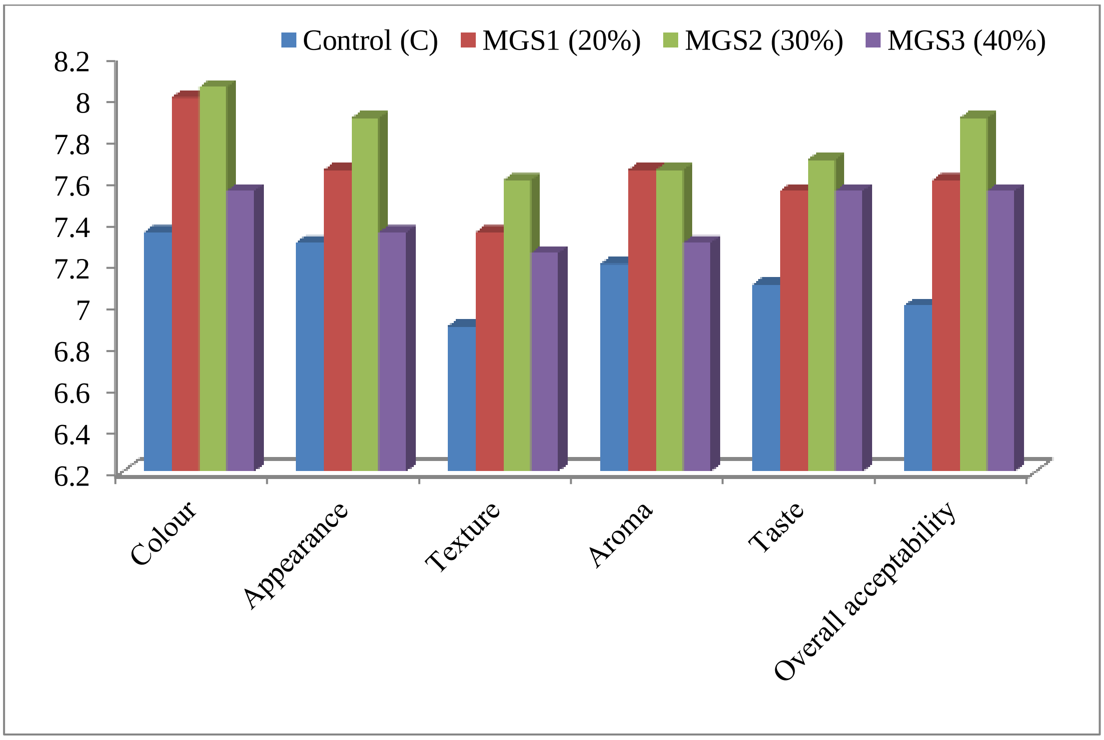

Fig 4.13 Organoleptic scores for Russian salad supplemented with fresh broccoli microgreens

The organoleptic properties for *russian salad* are described in Table 4.23. *Russian salad* supplemented with broccoli microgreens had highest mean over all acceptability score of 7.90 (fig 4.13). Among all the samples of *Russian salad*, the range were 6.90-7.35 for control and 7.60-8.05 for MGS2 (30%), which were liked very much followed by MGS2 and MGS3.With respect to colour and texture,MGS2 (30%) scored 8.05 and 7.60 respectively. Ghoora and Srividya (2017) prepared salad supplemented with fenugreek microgreens with overall acceptability of 8.08.

4.4.2 Microgreen juice

Two health products were developed i.e. juice blend and flavoured milk incorporated with microgreen juice.

1. Juice blend

The *juice blend* was formulated by mixing orange juice with fresh broccoli microgreens juice at 20%, 30%, 40% and 50% levels (Table 4.24). The control was orange juice. The *juice blend* imparted colour of orange to dark green (Plate 7). Overall acceptability score was highest for *juice blend* supplemented with 30% of broccoli microgreens juice (7.70), followed by juice supplemented with 40% of broccoli microgreens juice. The *juice blend* supplemented with 50% of broccoli microgreens juice had the lowest sensory score (6.65). The juice blend MGJ2 (30%) was superior in terms of colour, appearance, aroma, taste and overall acceptability and was decreased to 6.65 when increased the level to 50% of broccoli microgreens juice (Fig 4.14). Hence the *juice blend* incorporated with 30% of broccoli microgreens juice was considered as standard recipe. Sharma et al (2021) formulated fruit juice based juice blend supplemented with 17.26 ml/100ml spinach microgreen juice, 57.07 ml/100ml pomegranate juice, 24.66 ml/100 ml pineapple juice and 1.01 g/100 g sugar with overall acceptability score of 7.6.

2. *Flavored milk*

The high nutrient content of broccoli microgreens could be conveyed via certain foods as carriers. Milk is one of the important carrier which has been effectively used to deliver phytochemicals presents in microgreens for targeted health benefits in the traditional Indian system of medical science.

The *flavored milk* was formulated by mixing milk, sugar and flavoring agent with fresh broccoli microgreens juice at 20%, 30%, 40% and 50% levels (Table 4.24). The flavored milk imparted colour of light green to dark green (Plate 8). Overall acceptability score was highest for flavored milk supplemented with 30% of broccoli microgreens juice (7.70), followed by juice supplemented with 40% of broccoli microgreens juice (Fig 4.15).

Table 4.24 Organoleptic scores of broccoli microgreens juice incorporated *juice blend* and *flavoured milk*

Product		Colour	Apperance	Aroma	Taste	Overall acceptability
Juice blend						
Control (C)		$7.0^{a} \pm 0.66$	$6.80^{a} \pm 0.42$	$6.80^{a} \pm 0.42$	$7.0^{a} \pm 0.81$	$6.70^{ab} \pm 0.48$
Experimental	MGJ1 (20%)	$6.70^{a} \pm 1.05$	$6.90^{a} \pm 0.87$	$7.20^{a} \pm 0.91$	$7.30^{a} \pm 1.05$	$7.05^{ab} \pm 0.89$
	MGJ2 (30%)	$7.50^{a} \pm 0.70$	$7.50^{a} \pm 0.70$	$7.60^{a} \pm 0.69$	$7.50^{a} \pm 0.85$	$7.70^{b} \pm 0.82$
	MGJ3 (40%)	$7.20^{a} \pm 0.91$	$7.20^{a} \pm 0.91$	$7.30^{a} \pm 0.94$	$7.20^{a} \pm 0.91$	$7.20^{ab} \pm 0.91$
	MGJ4 (50%)	$6.80^{a} \pm 0.91$	$6.60^{a} \pm 1.07$	$6.60^{a} \pm 0.84$	$6.50^{a} \pm 0.85$	$6.65^{a} \pm 0.88$
Flavored milk						
Control (C)		$7.40^{a} \pm 0.84$	$7.30^{a} \pm 0.48$	$7.20^{a} \pm 0.63$	$7.50^{a} \pm 0.70$	$7.40^{a} \pm 0.84$
Experimental	MGM1 (20%)	$6.90^{a} \pm 0.73$	$6.90^{a} \pm 0.73$	$6.90^{a} \pm 0.56$	$6.90^{a} \pm 0.56$	$7.0^{a} \pm 0.81$
	MGM2 (30%)	$7.70^{a} \pm 0.48$	$7.60^{a} \pm 0.51$	$7.20^{a} \pm 0.63$	$7.30^{a} \pm 0.48$	$7.40^{a} \pm 0.51$
	MGM3 (40%)	$7.70^{a} \pm 0.48$	$7.60^{a} \pm 0.51$	$7.10^{a} \pm 0.73$	$7.00^{a} \pm 0.81$	$7.30^{a} \pm 0.67$
	MGM4 (50%)	$7.20^{a} \pm 0.91$	$6.80^{a} \pm 1.03$	$6.50^{a} \pm 0.85$	$6.60^{a} \pm 0.96$	$6.60^{a} \pm 0.84$

Values are expressed as mean ± SD
Values in rows followed by different superscript differ significantly ($p < 0.05$)
Control- product developed with standard recipe
Experimental-product developed by incorporating broccoli microgreens juice at different levels into standard recipe

Control (C)

MGJ1 (20%)

MGJ2 (30%)

MGJ3 (40%)

MGJ4 (50%)

Plate 7: Juice blend and flavored milk variants served for sensory evaluation

Control (C)

MGM1 (20%)

MGM2 (30%)

MGM3 (40%)

MGM4 (50%)

Plate 8: Flavored milk variants served for sensory evaluation

Table 4.25 Organoleptic scores for *food sprinkler* supplemented with fresh broccoli microgreens

Product		Colour	Appearance	Texture	Aroma	Taste	Overall acceptability
Food sprinkler							
Control (C)		$7.20^{a}\pm1.13$	$7.20^{a}\pm0.91$	$7.40^{a}\pm0.96$	$7.70^{a}\pm0.67$	$7.80^{a}\pm0.63$	$7.65^{a}\pm0.74$
Experimental	MGS1 (30%)	$7.90^{a}\pm0.56$	$7.90^{a}\pm0.59$	$7.90^{a}\pm0.73$	$8.0^{a}\pm0.47$	$8.0^{a}\pm0.47$	$8.0^{a}\pm0.52$
	MGS2 (40%)	$7.80^{a}\pm0.63$	$7.70^{a}\pm0.67$	$7.40^{a}\pm0.51$	$8.20^{a}\pm0.63$	$8.20^{a}\pm0.63$	$8.0^{a}\pm0.47$

Values are expressed as mean ± SD

Values in rows followed by different superscript differ significantly ($p < 0.05$)

Control- product developed with standard recipe

Experimental-product developed by incorporating broccoli microgreens powder at different levels into standard recipe

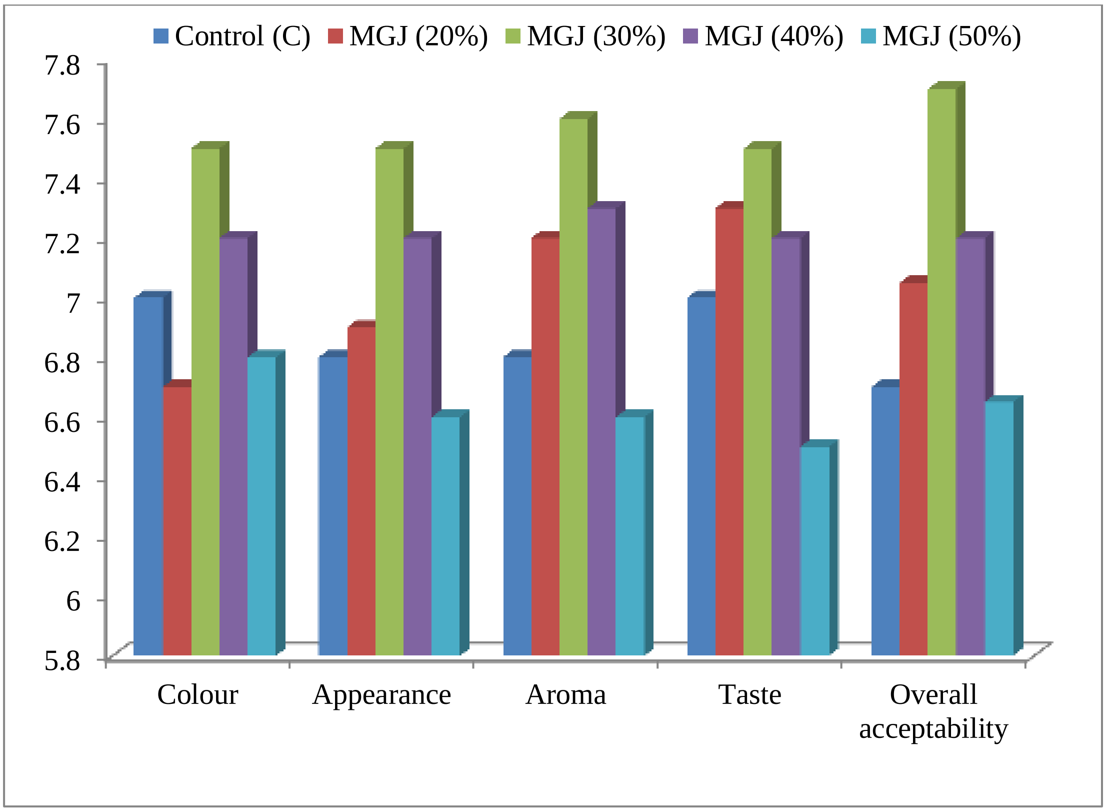

Fig 4.14 Organoleptic scores of broccoli microgreens juice incorporated *juice blend*

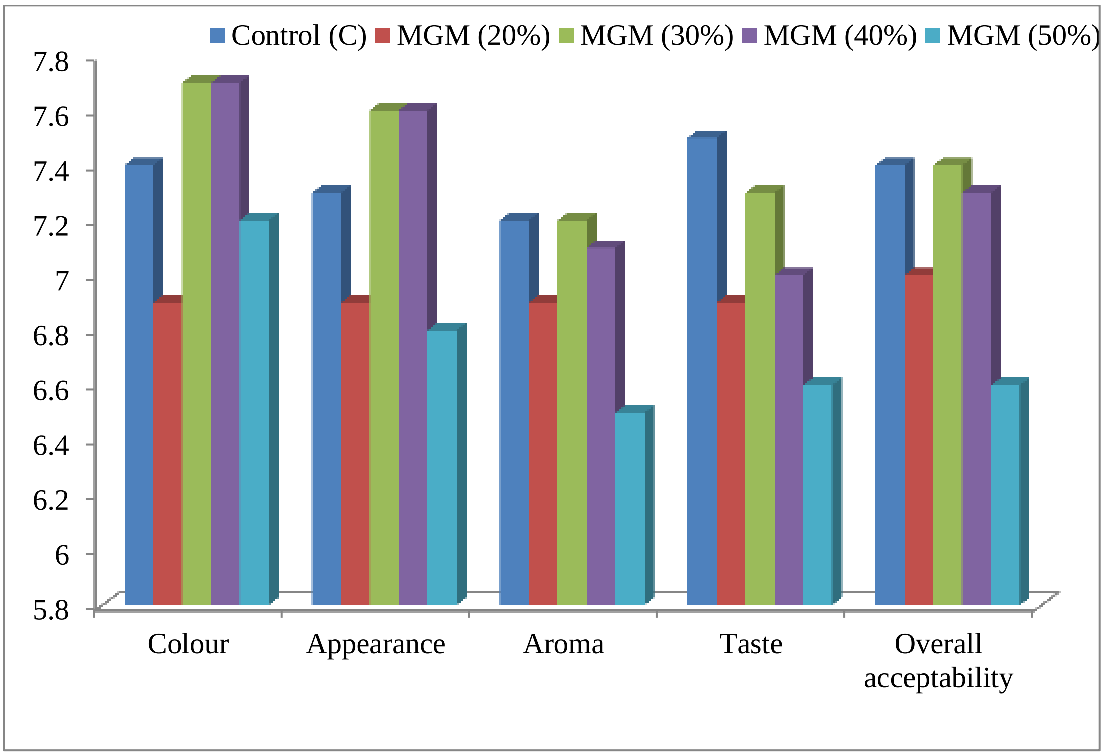

Fig 4.15 Organoleptic scores of broccoli microgreens juice incorporated *flavoured milk*

Control (C)

MGS1 (30%)

MGS2 (40%)

Plate 9: Microgreen food sprinkler variants served for sensory evaluation

The juice blend supplemented with 50% of broccoli microgreens juice had the lowest sensory score (6.65). Hingne et al (2020) formulated the flavoured milk by blending 3 percent tulsi juice and 6 percent aloe vera juice had good overall acceptability score of 7.68 .

4.4.3 Microgreen powder

Microgreen sprinkler was developed from powdered microgreens incorporated in regular spice mix.

1. Microgreens sprinkler

Microgreen *food sprinkler* (MGS) were prepared by supplementing different levels of broccoli microgreens powder prepared by microwave drying method by 30 and 40 percent incorporation to the regular spice mix without microgreen powder (control). The microgreen *food sprinkler* (MGS) with different levels of variants is presented in Plate 9. The mean sensory score of *food sprinkler* is presented in Table 4.25. Statistically there was no significant difference observed in mean score of all the sensory attributes. The regular spice mix supplemented with 40 percent of broccoli microgreens powder was well accepted organoleptically (Fig 4.16) and further evaluated for its nutritional composition.

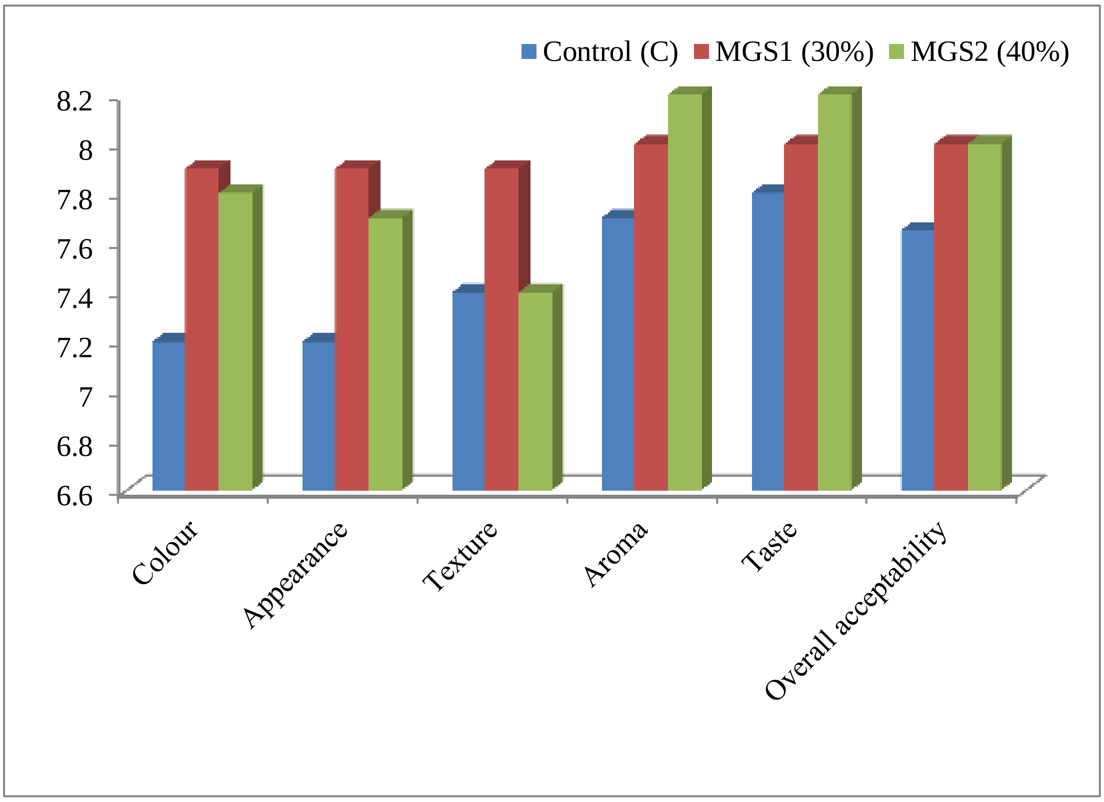

Fig 4.16 Organoleptic scores for *sprinkler* supplemented with fresh broccoli microgreens

Nutritional evaluation of various health foods supplemented with broccoli microgreens (fresh, juice and powdered)

Nutritional evaluation of heath foods

The results of nutritional composition of most acceptable health foods supplemented with broccoli microgreens (fresh, juice and powdered) have been discussed as under.

In the above Table 4.26 it is seen that the vitamin and mineral content of cooked dal supplemented with fresh broccoli microgreens was significantly ($p<0.05$) higher than the control cooked dal. The calcium, iron, potassium, magnesium, sodium and zinc content of supplemented cooked dal was significantly higher ($p<0.001$) than the control.

The vitamin and mineral content of cooked veg supplemented with fresh broccoli microgreens was significantly ($p<0.05$) higher than the control cooked veg (Table 4.27). The calcium, iron, potassium, magnesium and sodium content of supplemented cooked veg was significantly higher ($p<0.001$) than the control. The vitamin and mineral content of missi roti supplemented with fresh broccoli microgreens was significantly ($p<0.001$) higher than the control except sodium content (Table 4.28). Khan et al (2013) reported significantly increase content of calcium, iron, fibre, vitamin C and carotenoids in chapati supplemented with dehydrated spinach powder. Kachhawa and Chawla (2017) reported a significant increase in β-carotene, ascorbic acid and calcium content of missi roti, channa dal and potato leaves vegetable supplemented with drumstick leaves as compared to control.

The findings of Nambiar and Parnami (2007) supplemented drum stick leaves in mung dal enhanced the β-carotene, ascorbic acid and iron content. Each serving (30g) of supplemented dal provided 3955 μg of β-carotene, 46 mg ascorbic acid and 1.6 mg iron.

The overall nutritional composition of salad was increased with incorporation of broccoli microgreens. The vitamin and mineral content of russian salad supplemented with fresh broccoli microgreens was significantly ($p<0.001$) higher than the control (Table 4.29). The β-carotene, calcium, iron, potassium and zinc were high as compared to the control. The vitamin and mineral content of tofu salad supplemented with fresh broccoli microgreens was significantly ($p<0.001$) higher than the control except sodium content (Table 4.30).

The vitamin and mineral content of flavoured milk supplemented with fresh broccoli microgreens was significantly ($p<0.001$) higher than the control (Table 4.31). Gad and EL-Salam (2010) reported a high antioxidant and phenolic compound of milk supplemented with rosemary and green tea extract than control.

Table 4.26 Nutritional evaluation of *cooked dal* supplemented with fresh broccoli microgreens

Cooked dal	Vitamin C	β-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	9.36±0.33	1135.95±55.43	24.74±0.95	5.88±0.30	287.42±0.46	22.06±0.21	9.07±0.15	8.63±1.18
Acceptable (5%)	10.74±0.16	1225.38±0.02	175.17±0.19	9.19±0.00	304.90±0.02	57.98±0.02	10.44±0.02	11.23±0.03
Percentage Improvement	14.74	7.87	608.04	56.29	6.08	162.82	15.10	30.12
|t| value (p value)	6.373 (0.003)	2.794 (0.049)	268.456 (0.000)	19.005 (0.000)	65.635 (0.000)	293.064 (0.000)	15.543 (0.000)	3.787 (0.019)

Values are expressed as mean ± SD
t- values are absolute values

Table 4.27 Nutritional evaluation of *cooked veg* supplemented with fresh broccoli microgreens

Cooked veg	Vitamin C	β-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	24.02±0.48	569.96±1.0	33.10±2.08	0.52±0.10	298.51±0.15	17.34±0.41	11.92±0.51	0.22±0.10
Acceptable (7.5%)	27.02±1.45	586.82±5.97	182.05±0.16	3.54±0.37	315.57±0.15	53±0.27	13.47±0.02	2.14±0.03
Percentage Improvement	12.48	2.95	450.00	580.76	5.71	205.65	13.02	872.72
|t| value (p value)	3.403 (0.027)	4.819 (0.009)	123.481 (0.000)	134.051 (0.000)	1338.882 (0.000)	1244.071 (0.000)	48.818 (0.000)	85.304 (0.019)

Values are expressed as mean ± SD
t- values are absolute values

Table 4.28 Nutritional evaluation of *missi roti* supplemented with fresh broccoli microgreens

Missi roti	Vitamin C	β-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	17.55±0.29	2815.49±16.11	107.31±0.68	4.70±0.13	464.14±1.49	91.78±0.54	301.29±0.65	2.12±0.02
Acceptable (25%)	23.60±0.13	2962.28±0.59	855.14±0.64	19.74±0.75	550.46±0.34	271.53±0.20	306.52±3.07	11.95±0.33
Percentage Improvement	34.47	5.21	696.88	320.00	18.59	195.84	1.73	463.67
|t| value (p value)	31.847 (0.000)	15.768 (0.000)	1373.270 (0.000)	171.444 (0.000)	97.427 (0.000)	535.894 (0.000)	2.885 (0.045)	50.387 (0.000)

Values are expressed as mean ± SD
t- values are absolute values

Table 4.29 Nutritional evaluation of *tofu salad* supplemented with fresh broccoli microgreens

Tofu salad	Vitamin C	B-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	8.05±0.20	950.80±16.11	47.62±0.22	0.30±0.07	111.36±0.33	10.39±0	4.31±0.01	0.15±0.03
Acceptable (40%)	13.76±0.13	1061.05±0.03	645.42±0.02	12.25±0.03	179.92±0.03	153.16±0	9.42±0.01	7.70±0
Percentage Improvement	70.93	11.59	1255.35	3983.33	61.56	1374.10	118.56	5033.33
|t| value (p value)	80.61 (0.000)	1869.69 (0.000)	4650.83 (0.000)	255.92 (0.000)	97.427 (0.000)	535.894 (0.000)	2.885 (0.045)	50.387 (0.000)

Values are expressed as mean ± SD

t- values are absolute values

Table 4.30 Nutritional evaluation of *russian salad* supplemented with fresh broccoli microgreens

Russian salad	Vitamin C	β-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	23.21±0.01	858.04±0.11	36.06±0.07	0.57±0.15	256.55±0.20	20.21±0.01	10.00±0.05	0.25±0.01
Acceptable (30%)	30.77±0.15	1024.57±0.23	933.011±0.22	18.55±0.02	359.22±0.03	234.70±0.02	17.67±0.02	11.63±0.01
Percentage Improvement	32.57	19.40	2487.38	3154.38	40.01	1061.30	76.7	4552.00
|t| value (p value)	625.460 (0.000)	2402.204 (0.000)	6469.806 (0.000)	1056.838 (0.000)	871.402 (0.000)	11167.672 (0.000)	243.836 (0.000)	1005.400 (0.000)

Values are expressed as mean ± SD

t- values are absolute values

Table 4.31 Nutritional evaluation of *flavoured milk* supplemented with fresh broccoli microgreens juice

Flavoured milk	Vitamin C	β-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	2.34±0.02	8.40±0.02	121.23±0.03	0.15±0.01	109.03±0.07	10.01±0.05	30.09±0.01	0.33±0.02
Acceptable (30%)	7.31±0.02	170.58±0.55	955.64±0.03	18.13±0.02	211.45±0.04	220.35±0.02	32.73±0.04	11.64±0.03
Percentage Improvement	212.39	1930.71	688.28	11986.66	93.93	2101.29	8.77	3427.27
|t| value (p value)	256.04 (0.000)	509.09 (0.000)	32537.16 (0.000)	1212.40 (0.000)	1992.52 (0.000)	6423.49 (0.000)	105.83 (0.000)	439.132 (0.000)

Values are expressed as mean ± SD

t- values are absolute values

Table 4.32 Nutritional evaluation of *juice blend* supplemented with fresh broccoli microgreens juice

Juice blend	Vitamin C	β-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	64.30±0.02	15.05±0.04	5.12±0.02	0.73±0.02	9.31±0.02	8.99±0.05	4.49±0.02	0.03±0.01
Acceptable (30%)	71.85±0.364	165.53±0.02	896.06±0.02	18.83±0.12	111.74±0.05	223.14±0.15	12.15±0.02	11.42±0.03
Percentage Improvement	11.74	999.86	17401.17	2479.45	1100.21	2382.09	170.60	37966.66
|t| value (p value)	35.84 (0.000)	5828.06 (0.000)	46527.065 (0.000)	254.961 (0.000)	3228.39 (0.000)	6669.68 (0.000)	393.16 (0.000)	483.237 (0.000)

Values are expressed as mean ± SD
t- values are absolute values

Table 4.33 Nutritional evaluation of *food sprinkler* supplemented with powdered broccoli microgreens

Treatment	Vitamin C	β-carotene	Calcium	Iron	Potassium	Magnesium	Sodium	Zinc
Control	4.25±0.5	9.41±0.04	1.06±0.05	1.20±0.01	10.26±0.041	18.30±0.02	15.31±0.02	2.23±0.2
Acceptable (40%)	10.10±0.04	221.83±0.28	1196.06±0.12	23.99±0.50	136.61±0.04	285.51±0.01	10.18±0.00	15.17±0.02
Percentage Improvement	137.64	2257.38	112735.84	1899.16	1231.48	1460.16	-50.00	580.26
|t| -Value (p-value)	152.81 (0.000)	1146.23 (0.000)	15453.06 (0.000)	748.45 (0.000)	3523.48 (0.000)	18126.02 (0.000)	316.49 (0.000)	618.10 (0.000)

Values are expressed as mean ± SD
t- values are absolute values

The vitamin and mineral content of juice blend supplemented with broccoli microgreen juice was significantly ($p<0.001$) higher than the control (Table 4.32). The overall nutritional composition of supplemented microgreen juice blend was significantly increased as compared to control. Similar trend was observed by Sharma et al (2021) in juice blend (pomegranate and pineapple juice) supplemented with spinach microgreen juice had significantly enhanced protein, minerals (sodium, potassium and iron), vitamins (vitamin C), bioactive compounds (total phenols and total carotenoids) and had high antioxidant activity as compared with control.

Nutritional evaluation of *food sprinkler* supplemented with 40 percent of dried broccoli microgreens as compared to control (spice mix without broccoli microgreen) is presented in Table 4.33. Overall nutritional composition had significantly ($p<0.001$) improved in terms of vitamin C, β-carotene content and minerals namely calcium, iron, potassium, magnesium and zinc was significantly high when compared to the control product except sodium content. The high sodium content in the control sample was due to the presence of high salt content (10g) which was reduced (6g) with the addition of microgreens in the experimental sample. With the addition of 22g of broccoli microgreens powder the salt content of 4g reduced in the total amount of *food sprinkler*. Devi (2017) formulated *food sprinkler* with 50% incorporation of wheatgrass powder which enhanced its nutritional component and was highly acceptable to the sensory panellists.

CHAPTER V

SUMMARY

Microgreens are nutrient-dense, functional crops that are consumed fresh. They have a short growing span, do not require many inputs for growth, and have high nutrient, composition than their mature counterparts. The current food system revolves around the consumption of fresh fruit and vegetables due to their high phytochemical content. Microgreens can transform this and shift the focus to a new, product that can be grown in urban setups, even homes, with minimal input and maximum output. It is quick and delivers healthy produce that does not have to, be transported to great distances to be consumed. It saves transportation costs, reduce wastage, and spoilage that might take place in the transport chain and brings nutrient-dense food to the masses.

The present study on "Optimization of nutritional & biochemical composition and development of health foods-A study of microgreens" carried out to optimise the cultivation and harvesting stage of four vegetable crops to get maximum nutrients, bioactive compounds and antioxidant activity that have potential health benefits. Out of four microgreens, the microgreens with high nutrient quality score (NQS) was compared with sprouts and mature leaves. The most suitable technique of drying out of oven, shade and microwave drying of the microgreens was identified to study the retention of nutrients and bioactive compounds. Further, the fresh, juice and dehydrated microgreens were incorporated in the commonly consumed to optimised the maximum nutrient content and sensory attributes as well.

The β-carotene content of broccoli microgreens was highest for soaked seeds, indoor cultivation and harvested at 10th day. The ascorbic acid was significantly higher when cultivated indoor with soaked seeds and harvested at 10th day. A significantly higher chlorophyll content was observed in broccoli microgreens cultivated with soaked seeds at outdoor setting and harvested at 10th day. The broccoli microgreens had higher content of total phenols when cultivated with soaked seeds at outdoor setting and harvested at 10th day. The flavonoid content of broccoli microgreens cultivated with soaked seeds at outdoor settings were significantly higher when harvested at 10th day. The broccoli microgreens cultivated with soaked seeds at outdoor setting and harvested on 10th day had higher antioxidant activity than the one harvested at 20th day. The mineral content of namely zinc, potassium, and magnesium was significantly higher with soaked seeds, outdoor cultivation and harvested at 10th. The iron and calcium was higher at indoor cultivation and harvested on 10th day. The sodium content was higher with soaked seeds, outdoor cultivation and harvested on 20th day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in broccoli microgreens is

by soaking the broccoli microgreen seeds, cultivating them in outdoor setting and harvesting at an early stage (10th day). There was significant ($p \leq 0.001$) effect of soaking, position (indoor and outdoor cultivation) and duration of harvesting on broccoli microgreens nutritional composition. The Nutrient quality score of broccoli microgreens with soaked seeds, cultivated outdoor and harvested on 10^{th} day had highest score.

The β-carotene, ascorbic acid, chlorophyll, total phenolic content, total flavonoids and antioxidant activity was significantly higher with soaked seeds, cultivated at outdoor setting and harvested on 10^{th} day. The mineral content of namely calcium and potassium was significantly higher with unsoaked seeds, outdoor cultivation and harvested at 10^{th}. The zinc and magnesium content was higher with soaked seeds at indoor cultivation and harvested on 10^{th} day. The sodium content was higher with soaked seeds, indoor cultivation and harvested on 20^{th} day. The iron was highest with unsoaked seeds at indoor cultivation and harvested at 10^{th} day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in carrot microgreens is by soaking the carrot microgreen seeds, cultivating them in outdoor setting and harvesting at 10^{th} day. There was significant ($p \leq 0.001$) effect of soaking, position (indoor and outdoor cultivation) and duration of harvesting on carrot microgreens nutritional composition. The Nutrient quality score of carrot microgreens with soaked seeds, cultivated outdoor and harvested on 10^{th} day had highest score.

The β-carotene, ascorbic acid and chlorophyll content were higher with soaked seeds of coriander and cultivated indoor. The total flavonoids and antioxidant activity was significantly higher with soaked seeds and cultivated at outdoor setting whereas the total phenolic content was higher with soaked outdoor setting. The vitamin, bioactive and antioxidant activity was higher in coriander microgreens harvested on 10^{th} day. The mineral content of namely magnesium and zinc was significantly higher with soaked seeds, indoor cultivation and harvested at 10^{th}. The iron content was higher with soaked seeds at outdoor cultivation, whereas for calcium unsoaked seeds at indoor setting and harvested on 10^{th} day. The potassium content was higher with soaked seeds whereas sodium was with unsoaked seeds cultivated indoor and harvested on 20^{th} day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in carrot microgreens is by soaking the coriander microgreen seeds, cultivating them in outdoor setting and harvesting at 10^{th} day. There was significant ($p \leq 0.001$) effect of duration of harvesting and in combination, soaking* position and soaking*duration had significant ($p \leq 0.001$) effect on coriander microgreens nutritional composition. The Nutrient quality score of coriander microgreens with soaked seeds, cultivated outdoor and harvested on 10^{th} day had highest score.

The ascorbic acid and antioxidant activity were higher with soaked seeds and outdoor cultivation of spinach microgreens. The chlorophyll and total flavonoids were higher with unsoaked seeds cultivated outdoor. The β- carotene content was higher with unsoaked seeds whereas total phenols were with soaked seeds cultivated indoor. The vitamin, bioactive and antioxidant activity was higher in spinach microgreens harvested on 10th day. The mineral content namely zinc, iron, calcium and potassium were higher with soaked seeds whereas magnesium was higher with unsoaked seeds, cultivated outdoor setting. The sodium content was higher with unsoaked seeds cultivated indoor and harvested at 20th day. The present study optimizes the best combination of cultivation and harvesting techniques to attain maximum nutrients and bioactive compounds in carrot microgreens is by soaking the spinach microgreen seeds, cultivating them in outdoor setting and harvesting at 10th day. There was significant ($p \leq 0.001$) effect of position (indoor and outdoor cultivation), duration, soaking*position, position*duration and soaking*position*duration of harvesting on carrot microgreens nutritional composition. The Nutrient quality score of spinach microgreens with soaked seeds, cultivated outdoor and harvested on 10th day had highest score.

Microgreens are perishable in nature due to high moisture content. In order to optimize best technique of drying, harvested microgreens was dried via shade drying, tray drying and microwave drying for removing moisture and improving the shelf life. The β-carotene content of shade dried broccoli microgreens was significantly ($p<0.05$) higher than the oven and microwave dried broccoli microgreens. The ascorbic acid content of microwave dried broccoli microgreens was significantly ($p<0.05$) higher followed by shade and oven dried broccoli microgreens. The chlorophyll content of microwave dried broccoli microgreens was significantly ($p<0.05$) higher than shade and oven dried microgreens. The total phenolic content of shade dried broccoli microgreens was higher as compared to the oven and microwave dried sample. The flavonoid content of shade dried broccoli microgreens was significantly ($p<0.05$) higher than the microwave dried broccoli microgreens. The antioxidant activity of microwave dried broccoli microgreens was significantly ($p< 0.05$) higher than the shade and oven dried broccoli microgreens. The mineral content like zinc, iron, calcium, potassium, magnesium and sodium were significantly ($p<0.05$) higher in microwave drying as compared to shade and tray drying. Further the retention of ascorbic acid, β-carotene, chlorophyll and antioxidant activity of fresh and dried broccoli microgreens was studied. The ascorbic acid content was significantly ($p<0.05$) higher in fresh broccoli microgreens when compared with microwave dried microgreens followed by oven and shade. The β-carotene content of fresh broccoli microgreens was 5 significantly ($p<0.05$) reduced after drying. The fresh broccoli microgreens had significantly ($p<0.05$) higher content of chlorophyll than shade, oven and microwave drying technique. The antioxidant activity of fresh broccoli microgreens was significantly ($p < 0.05$) higher than dried broccoli microgreens. Thus, it was observed that retention of nutrients was maximum in microwave drying as compared to fresh microgreens, followed by shade and tray drying.

The microgreens are superior to the mature counterparts in terms of vitamins, minerals, bioactive compounds and antioxidant activity. The nutritional and bioactive composition of broccoli microgreens was compared with the sprouts and mature leaves of broccoli. The β-carotene content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts and mature leaves. The ascorbic acid content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli sprouts followed by mature leaves. The chlorophyll content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli mature leaves followed by sprouts. The total phenolic content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli mature leaves and sprouts. The statistical analysis showed that the flavonoid content of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli mature leaves and sprouts. The antioxidant activity of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli mature leaves and sprouts. The mineral content namely iron, zinc, potassium, calcium and magnesium of broccoli microgreens was significantly ($p<0.05$) higher than the broccoli mature leaves and sprouts. The sodium content of mature leaves of broccoli was significantly ($p<0.05$) higher than the broccoli microgreens followed by sprouts.

Fresh broccoli microgreens were incorporated in common traditional recipes like missi roti, cooked vegetable and dal at different levels. Missi roti with 25% fresh broccoli microgreens scored highest mean overall acceptability score whereas control and 20% were at par with the mean overall acceptability score. Overall nutritional composition of missi roti supplemented with 25% of fresh broccoli microgreens had significantly ($p<0.001$) improved in terms of vitamin C,β-carotene content and minerals namely calcium, iron, potassium and magnesium when compared to the control product. The cooked dal with 5% of fresh broccoli microgreens received the highest score of mean overall acceptability. It was observed that organoleptic score for dal was increased with increased the level of microgreens for certain level (5%) and then starts decreasing with increase the level of microgreens. The cooked vegetable supplemented with 7.5% of fresh broccoli microgreens scored the highest mean overall acceptability. The mineral content of cooked dal supplemented with 5% and cooked vegetable supplemented with 7.5 % of fresh broccoli microgreens was significantly ($p<0.001$) improved as compared to the control product in term of calcium, magnesium, iron and potassium and sodium. The Russian and tofu salad was supplemented with the fresh broccoli microgreens. The highest mean overall acceptability score was with 40% incorporation of fresh broccoli microgreens in tofu salad. Statically the values of different variants of tofu salad supplemented with fresh broccoli microgreens were at par with control product. Russian salad with 30% fresh broccoli microgreens scored highest mean overall acceptability score. The statistically analysis showed that the nutrient and mineral content was increased significantly ($p<0.001$) when salads were supplemented with fresh broccoli microgreens as compared to control product.

The juice blend was formulated by mixing orange juice with fresh broccoli microgreens juice at 20%, 30%, 40% and 50% levels. Overall acceptability was highest for juice blend supplemented with 30% broccoli microgreen juice followed by juice supplemented with 40% broccoli microgreen juice. The flavoured milk supplemented with 30% of broccoli microgreen juice had high overall acceptability score which was at par with the score of control product. The overall acceptability of juice blend and flavoured milk decreased with increased the amount of broccoli microgreen juice. The overall nutritional composition of microgreen juice blend and flavoured milk supplemented with broccoli microgreen juice was significantly ($p<0.001$) increased as compared to control product in term of vitamin C,β-carotene content and minerals namely calcium, zinc, iron, potassium, magnesium and sodium.

Microgreen *food sprinkler* (MGS) were prepared by supplementing different levels of broccoli microgreens powder prepared by microwave drying technique by 30 and 40 percent incorporation to the regular spice mix without microgreen powder (control). Statistically the mean overall acceptability of spice mix incorporated with 30 and 40% of broccoli microgreen powder score was at par with control product. The regular spice mix supplemented with 40 percent of broccoli microgreens powder was well accepted organoleptically and further evaluated for its nutritional composition. Overall nutritional composition had significantly ($p<0.001$) improved in terms of vitamin C,β-carotene content and minerals namely calcium, iron, potassium and magnesium when compared to the control product except sodium content.

Conclusions

- Out of four microgreens namely broccoli, carrot, coriander and spinach, the broccoli microgreens had the highest nutrient quality score in term of overall nutritional composition. Outdoor cultivation with soaked seeds and harvesting at 10^{th} day significantly ($p<0.001$) effect the nutritional composition of microgreens. The nutrient and bioactive compounds decreased with increase in harvesting stage.
- Broccoli microgreens dense source of vitamin (ascorbic acid and β-carotene), mineral (calcium, iron, potassium, zinc and magnesium), bioactive compounds (chlorophyll, TPC and TFC) and antioxidant activity as compared to the sprouts and mature leaves of broccoli.

- Drying method influence the nutritional composition and appearance of the broccoli microgreen powder. Microwave drying was the best method as it preserved the maximum amounts of vitamins, minerals, bioactive compounds and antioxidant activity of broccoli microgreens.
- All the health products i.e. missi roti, cooked vegetable and dal, salads, juice blend, flavoured milk and *food sprinkler* supplemented with broccoli microgreens were organoleptic highly acceptable.
- In term of fresh broccoli microgreens supplementation, 25% in missi roti, 5 and 7.5% in cooked vegetable and dal, 30 and 40% in Russian and tofu salad was highly accepted.
- In term of fresh broccoli microgreen juice, 30% incorporation of broccoli microgreen juice in juice blend and flavoured milk was highly acceptable.
- In term of powdered broccoli microgreens, 40 % supplementation of broccoli microgreen powder in regular spice mix was highly accepted.
- Overall nutritional composition had significantly ($p<0.05$) improved in terms of vitamin C,β-carotene content and minerals namely calcium, zinc, iron, potassium and magnesium when compared to the control product.

On the basis of the above conclusion drawn from the results of the study, the following recommendation can be given:

- Awareness needs to be created among masses pertaining to the cultivation of microgreens at household level, so that they could be included in daily diets.
- For commercial ventures best cultivation and harvesting stage can lead to optimum nutritional quality that can be helpful in increasing sales and profit.

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ANNEXURE-I

Score card for sensory evaluation Health foods

Name of judge: …………………   Date: ………..........

Taste these samples and check how much you like or dislike each one. Use the appropriate scale to show your attitude by checking at the point that best describes your feelings about the sample. Please give a reason for this attitude. An honest expression of your personal feeling will help me.

Nine point hedonic rating scale

9 = like extremely   6 = like slightly   3 = dislike moderately

8 = like very much   5 = neither like nor dislike   2 = dislike very much

7 = like moderately   4 = dislike slightly   1 = dislike extremely

Attributes	Colour	Consistency	Appearance	Aroma	Taste	Overall acceptability
Control						
Experimental 1						
Experimental 2						
Experimental 3						
Experimental 4						

Remarks:……………………………………………………   **Signature**

ANNEXURE-II

MISSI ROTI

INGREDIENTS

- Wheat flour-22.5g
- Besan-22.5g
- Methi-22.5g
- Microgreens-25g
- Coriander leaves-3.75g
- Green chilli-3.75g

METHOD

- Sieve the whole wheat flour and besan was mixed together and add chopped methi, coriander leaves and green chillies.
- Mix properly and knead smooth dough with water.
- For control sample it was followed as above without microgreens.
- Keep it aside and the dough was divided into equal portions and each portion was made into small balls. Balls were rolled out with the help of rolling pin.
- The missi roti was cooked and put it on the tava at constant temperature.
- After 45 seconds missi roti was turned over and cooked for 45 seconds and 1 tsp of oil was used to cook the missi roti and serve hot.

COOKED DAL

INGREDIENTS

- Split moong dal-27.75g
- Tomato-23g
- Onion -23g
- Garlic -9g
- Green chillies-9g
- Microgreens -7.5
- Oil-1tbsp
- Salt-to taste
- Turmeric powder-1/4tsp

METHOD

- Wash the dal and pressure cook for 15 minutes with salt and turmeric powder.
- Cut all the vegetables and cook by adding a little oil.
- Add vegetables (onion, tomato, garlic and green chillies) to dal for tadka and cook properly.
- Add coriander leaves and pour over cooked dal with chopped Fresh microgreens.
- Serve hot with chapati.

COOKED VEGETABLE

INGREDIENTS:

- Potato -28.5 g
- Cauliflower -28.5 g
- Onion -9.5 g
- Tomato -9.5 g
- Microgreens- 4g

METHOD:

- Heat a pan and pour oil into it.
- Once the oil heats add chopped vegetables and mix well.
- Now add pinch of salt, turmeric and spices.
- Stir well so the spices and vegetables infuse together.
- Cook all ingredients for about half an hour and then turn off the flame.
- Transfer the cooked vegetable into a serving bowl and add chopped microgreens .

JUICE BLEND

INGREDIENTS

- Orange juice- 70ml
- Microgreens juice- 30ml

METHOD

- Wash oranges and microgreens very well.
- Microgreens juice was prepared by taking water and microgreen in ratio 1:1.
- Remove the orange peel and white membrane as well.
- You will have to cut them to 2 halves and remove seeds.

- Add oranges to a juicer and sieve the juice and add Fresh microgreens juice.
- Mix it very well and serve chilled.

FOOD SPRINKLER

INGREDIENTS:

- Coriander seeds powder- 9g
- Mango powder-6g
- Anardana powder-6g
- Pink salt- 3g
- Table salt- 3g
- Black pepper-6g
- Microgreen powder-22 g

METHOD:

- Dry roast coriander seeds and black pepper for 1 minute.
- Allow the spices to cool and then grind them in a grinder jar.
- Remove from a jar and put it in a bowl and then add mango powder, anardana powder, microgreen powder, pink salt and table salt.
- Mix well with a dry clean spoon.
- Store it in a cool dry glass bottle and stays good without refrigeration for a couple of months.

FLAVOURED MILK

INGREDIENTS:

- Milk (SNF 8.5% and fat 4.5%)- 70ml
- Cardamon powder- 5g
- Cardamon essence- 2drops
- Sugar- 10g
- Microgreens juice-30ml

METHOD:

- Run an electric hand blender and add milk and microgreen juice, cardamon powder and essence and sugar.
- Blend it till the milk is frothy and light.
- Pour into the glass and chill till required.
- Serve chilled and immediately while still frothy.

RUSSIAN SALAD

INGREDIENTS:

- Tomato- 10.5g
- Potato- 14g
- Peas- 7g
- Beans- 10.5g
- Cucumber- 10.5g
- Capsicum- 7g
- Hung curd- 10.5g
- Salt- 3g
- Black pepper- 3g
- Microgreens -30g

METHOD:

- Wash and chop the all vegetables and transfer into deep bowl and add enough water.
- Steam the vegetables (peas, potato and beans) at least ½ hour.
- Drain using strainer.
- Transfer all vegetables to a bowl and add hung curd.
- Add microgreens, salt and pepper and mix everything very well.
- Refrigerate for at least 1 hour and serve Russian salad chilled.

TOFU SALAD

INGREDIENTS:

- Tofu- 12g
- Tomato- 12g
- Cucumber- 12g
- Onion- 12g
- Salt- 3g
- Black pepper- 3g
- Microgreen -32g

METHOD:

- Wash and chop all vegetables and cut tofu in diced form.
- Take deep bowl and transfer all vegetables in a deep bowl.
- Add microgreens, salt and pepper and mix properly.

VITA

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EDUCATIONAL QUALIFICATION

Bachelor degree	**:**	**B.Sc. (Nutrition & Dietetics)**
University	:	Punjab Agricultural University, Ludhiana
Year of award	:	2014
OCPA	:	7.15/10.00
Master's degree	**:**	**M.Sc. (Food and Nutrition)**
University	:	Punjabi University, Patiala
Year of award	:	2016
%age of marks	:	76.30%
Title of Master's Thesis	:	A study of dietary pattern and nutritional status of adolescence school going children (13-15 years) of urban area
Doctorate degree	:	Ph.D. (Food and Nutrition)
University	:	Punjab Agricultural University, Ludhiana
Year of award	:	2021
OCPA	:	8.02/10.00
Title of Ph.D. dissertation	:	Optimization of nutritional & biochemical composition and development of health foods – A study of microgreens
Awards/Distinctions/ Scholarships	:	Best oral presentation in Golden Jubilee International Seminar on 'Opportunities and Challenges in Food Processing Sector' held at Tamilnadu Agriculture University