

Minerals such as iron and copper are present in seaweeds at higher levels than in many well-known terrestrial sources of minerals, such as cheese, bananas, spinach. Iodine is an important nutrient in metabolic regulation and growth patterns and is abundant in most seaweed. The role of seaweed as a rich source of iodine is particularly relevant in the UK, where recent surveys have highlighted insufficiency in different groups of the population, with iodine being an essential component of thyroid hormones, which are essential for neuro development in utero and after birth (Paul *et al.*, 2007; and Maria and Emilie, 2015).

Vitamins

Seaweeds are a good source of some water-(B₁, B₂, B₁₂, C) and fat-soluble (β-carotene with vitamin A activity, vitamin E) vitamins. To ensure that the adequate intake of all vitamins is received in the diet, people (especially people on special

diet, strict vegetarians, and vegans) can consume foods enriched with vitamins, for example, in the form of functional foods with vitamins as nutraceuticals, extracted from natural sources such as seaweeds. Seaweed vitamins are important not only due to their biochemical functions and antioxidant activity but also due to other health benefits such as decreasing of blood pressure (vitamin C), prevention of cardiovascular diseases (β-carotene), or reducing the risk of cancer (vitamins E and C, carotenoids) (Skrovankova, 2011).

From the Table 6 *Laminaria digitata* and *Ulva spp* are good sources for vitamin B₃ and C than other terrestrial foods. Seaweeds are also one of the few vegetable sources of vitamin B₁₂. This may provide an alternate source of vitamin B₁₂ for vegetarians or vegans.

Antioxidant and Bioactive Compounds

Antioxidants play an important role in inhibiting and

Table 6: Vitamin Composition of Different Seaweeds and Some Terrestrial Foods

Seaweed	B1	B2	B3	B6	B9	C	E	B12†
(mg per 8 g Dry Portion)*								
<i>Laminaria digitata</i>	0.011	0.011	4.896	0.513	0	2.842	0.275	0.495
<i>Porphyra umbilicalis</i>	0.077	0.274	0.761	0.119	1.003	12.885	0.114	0.769
<i>Ascophyllum nodosum</i>	0.216	0.058	0	0.001	3.648	0.654	0.029	0.131
<i>Undaria pinnatifida</i>	0.403	0.936	7.198	0.259	0.528	14.779	1.392	0.345
<i>Palmaria palmata</i>	0.024	0.08	0.8	0.002	0.021	5.52	1.296	1.84
<i>Ulva spp.</i>	0.06	0.03	8	NA	0.012	10	NA	6.3
Whole Food [§] (mg/100 g Edible Portion)								
Bovine milk	0.04	0.18	0.09	0.04	0.08	1.7	0.07	0.4
Emmental cheese	0.02	0.04	0.17	0.01	0.08	6.5	0.28	0.05
Whole wheat	0.48	0.09	5.1	0.27	0.58	-	1.4	-
Carrots	0.07	0.05	0.6	0.27	0.17	7.1	0.47	-
Tomatos	0.06	0.04	0.5	0.1	0.33	19	0.81	-
Spinach	0.09	0.2	0.6	0.22	0.145	2.5	2.5	-
Apple	0.035	0.032	0.3	0.1	0.05	12	0.49	-
Orange	0.08	0.04	0.3	0.1	0.22	5	0.32	-

Note: * Values for seaweeds from the Institut de Phytonutrition (2004). [§] Values for whole foods from Belitz (2009). †: Values expressed in µg/100 g wet weight. Abbreviations: NA, no data available.

scavenging radicals and thus providing protection to humans against infections and degenerative diseases. A number of marine algae were reported to possess antioxidant properties (Hanan and Mohamed, 2015).

Antioxidant compounds act as free radical scavengers to protect living organisms from the systemic production of Reactive Oxygen Species (ROS), lipid peroxidation, protein damage and DNA breaking (Kokilam and Vasuki, 2014). Many seaweed species verified natural antioxidant capacity that can protect the human body from free radicals and retard the progress of many chronic diseases such as hypertension, heart diseases, diabetes and cancer (Gehan, 2017).

Phenolic and flavonoid compounds were broadly recognized in seaweeds confirming their potent role in chelating metal ions, preventing radical formation and improving the internal antioxidant system under stress environmental conditions. These activities protect the body from progressive diseases caused by the adverse effects of Reactive Oxygen Species (ROS) (Chakraborty *et al.*, 2013). Similar attitude was reported for carotenoid pigments of seaweeds, especially β carotene, for which activity against cancer diseases was postulated (Jane, 2013; and Gehan, 2017).

Extracts from macroalgae or seaweeds are rich in polyphenolic compounds. Which have well documented antioxidant properties. They also have antimicrobial activities against major food spoilage and food pathogenic micro-organisms (Hanan *et al.*, 2015).

CONCLUSION

Seaweeds as a new source of valuable nutrients, food additives nutraceuticals, and nutritional supplements for human and animal consumption. Microalgae and seaweeds have important amounts of macronutrients (proteins, carbohydrates and lipids), vitamins (fat soluble (A and E) and water soluble (C, B₁, B₃, nicotinate, panthotenic acid, biotin, folic acid, B₁₂) minerals, essential and non-essential aminoacids, chlorophylls, carotenoids, polyphenols, polysaccharides, and minerals, although the concentration varies widely among the different species. Specifically, trace elements and minerals are abundant in seaweeds compared to terrestrial foodstuffs, and their non animal nature lends them to use in many food products.

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PROBIOTICATION OF SWEET ORANGE JUICE USING LACTOBACILLUS STRAINS

L Hruyia^{1*}, H W Deshpande¹ and M A Bhate²

*Corresponding Author: L Hruyia, ✉ l.hruyia@gmail.com

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A study was carried out for the production of probiotic sweet orange juice using two lactobacillus strains viz., *Lactobacillus delbrueckii* ssp. bulgaricus and *Lactobacillus plantarum*. Encapsulated and free strains were used to prepare the probiotic juice. Prior to juice extraction sweet oranges were treated with activated charcoal solution and lye peeled to prevent delayed bitterness in the juice. Encapsulation of strains was done using a mixture of sodium alginate and guar gum by extrusion technique and the probiotic beads were added to the juice and incubated at 37 °C for 10 hrs. The probioticated juices were studied for sensory acceptability and cell viability for 4 weeks at 4 °C. Microencapsulation of probiotic strains was found to be suitable in maintaining the cell viability in the juice, since viable cells were approximately 9.00 log cfu/ml even after a storage period of 4 weeks. During storage it was found that the viable cell count and sensory score for juice containing encapsulated strains was higher than juice with free strains.

Keywords: Probiotic sweet orange juice, *Lactobacillus delbrueckii* ssp. bulgaricus, *Lactobacillus plantarum*, Encapsulation, Cell viability

INTRODUCTION

Increasing awareness of health and wellness among people and across the age spectrum in the past two decades is fuelling interests in functional foods. Foods are no longer considered only in terms of taste and immediate nutritional needs but also in terms of their ability to provide specific benefits above and beyond their basic nutritional value. Functional foods targeted towards improving the balance and activity of intestinal milieu provides the largest segment of functional food market (Saarela *et al.*, 2000).

Probiotics are live microbial food ingredients, which provide the consumer with numerous health benefits by improving the intestinal microbial balance. However, probiotic foods available in the markets today, are usually in the form of fermented dairy products such as milk and

yogurt and thus probiotication of fruit juices is beneficial as fruits and fruit juice based drinks are important components of the human diet. People choose fruit juices as a drink for many reasons, including relieving thirst, refreshment and nutritional benefits. Furthermore, fruits and vegetables do not contain any dairy allergens that might prevent usage by certain segments of the population (Luckow and Delahunty, 2004). Also, fruit and vegetable juices have an established market sector as a functional drink including calcium and vitamin fortified juices. It has been suggested that fruit juices could serve as a good medium for cultivating of probiotics (Mattila-Sandholm *et al.*, 2002). Different studies have been implemented to investigate the suitability of fruit juices such as tomato, orange, grape, carrot, pomegranate, beet and cabbage juices as raw vegetables and fruits for probiotic drink production.

¹ Department of Food & Industrial Microbiology, College of Food Technology, Vasantrao Naik Marathwada Krishi Vidhyapeeth, Parbhani 431402 (MS), India.

² Department of Microbiology, Shri Shivaji College, Parbhani 431402 (MS), India.

Lactobacillus plantarum, *L. delbrueckii*, *L. acidophilus*, *L. casei* and *L. paracasei* have been used as probiotic cultures. Results have shown that all the strains of probiotic bacteria are capable of growth in the mentioned juices. Moreover, *L. plantarum*, *L. acidophilus* and *L. delbrueckii* have shown to be resistant to high acidic and low pH conditions during storage periods at 4 °C for four weeks (Rasic, 2003; King *et al.*, 2007; Mousavi *et al.*, 2011; and Tamminen *et al.*, 2013).

The role of citrus fruits in providing nutrients and medicinal value has been recognized since ancient times. The fruits are known for their refreshing fragrance, thirst-quenching ability, and providing adequate vitamin C as per Recommended Dietary Allowance (RDA). In addition to ascorbic acid, these fruits contain several vitamins and phytochemicals. Sweet orange (*Citrus sinensis* L. Osbeck) is a widely grown sweet orange cultivar in India and most popular in Maharashtra, medium to large fruit, thin peeled, juicy nucellar selections available, acidity about 0.35-0.5 per cent when fully mature and hence sweet to taste when stored, slightly acidic fruit with higher acidity and greenish-yellow color rind are better, early maturity (Ladaniya, 2008).

Viability maintenance of probiotic cells throughout food-processing, storage and gastro-intestinal transit is important for the microorganisms to reach the intended site of action in sufficient numbers. International Dairy Federation (IDF) has suggested that a minimum of 10^7 cfu/ml probiotic bacterial cells should be alive at the time of consumption per gram of the product. Probiotic stability in fruit and vegetable juice products is difficult to maintain during cold storage however probiotic encapsulation might solve this problem. Providing probiotic living cells with a physical barrier against adverse external conditions by microencapsulation is an approach currently receiving considerable interest. Microencapsulation helps to separate a core material from its environment until it is released. It protects the unstable core from its environment, thereby improving its stability, extends the core's shelf life and provides a sustained and controlled release (Franjione and Vasishtha, 1995; and Gibbs *et al.*, 1999).

Numerous studies have been carried out on probiotic foods and its health benefits and it is also being increasingly promoted by health professionals. Thus the objective of this study is to determine the suitability of the sweet orange juice as a probiotic drink. Also the present work studies the

sensory quality and cell viability of juice with encapsulated strains and free strains during a storage period of 4 weeks.

MATERIALS AND METHODS

Probiotic Strains

The probiotic strains: *Lactobacillus delbrueckii* ssp. *bulgaricus* and *Lactobacillus plantarum* were isolated and identified in Department of Food and Industrial Microbiology, College of Food Technology, VNMKV, Parbhani, Maharashtra and further confirmation of the results were carried out by genotypic identification (16S rRNA multiplex PCR analysis) at ARI Pune.

Preparation of Probiotic Cultures

The lactobacilli strains viz. *Lactobacillus delbrueckii* ssp. *bulgaricus* and *Lactobacillus plantarum* were individually activated in MRS broth at 37 °C for 24 h and further subculturing was done. The starter culture was prepared by mixing equal amounts of cultivated broths and centrifuged at 4000 rpm for 10min. The harvested cells were washed twice with sterile water to remove the residual MRS media.

Sweet Orange Juice Extraction

Sweet oranges (cv. Mosambi) were procured from the local market of Parbhani, Maharashtra. The rind of the fruits were removed manually and the fruits were dipped in 1% activated charcoal solution and allowed to stand for 1hr. This was done to adsorb the bitter precursors from the fruit surface and the core. The fruits were then lye peeled by dipping it in boiling lye solution (1.5%) for 2 mins to remove the albedo section which is the major contributor of limonin precursors during juice extraction (Sheu and Marshall, 1993; and Teanpaisan *et al.*, 2015). Standardization of the concentration level and time of both the treatments were done based on the extent of broken segments after lye treatment, taste (bitterness reduction) and aroma of the juice. After lye treatment, the fruits were washed thoroughly in running tap water and the remaining alkali was neutralized by dipping in citric acid solution (1%) for 1min and washed again thoroughly. Juice was extracted without pressing the seeds and subsequently the juice was filtered using a strainer and the filtered juice is collected in a dispenser and pasteurized at 90 °C for 2 mins.

Chemical Analysis

Chemical parameters play a significant role in the quality of the juice and also survival of probiotic cultures. In the