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Red beetroot: A source of natural colourant and antioxidants: A review

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Abstract
Beetroot (*Beta vulgaris*) is a root vegetable also known as red beet, garden beet, table beet, or just beet. It is rich in essential nutrients as fibre, folate (vitamin B9), manganese, potassium, iron, and vitamin C. It is widely used as additives in the food industry because of its natural and harmless pigments and colorant properties and absence of toxicity. It is important from the medicinal point of view also due to its antiviral, antimicrobial and antioxidant properties. Beetroot is associated with numerous health benefits, including healthy blood pressure, improved athletic performance, digestive health, brain health, fights inflammation and also contains anti-cancer properties. The present paper reviews the potential application of Beetroot as an antioxidant and natural colorant in food products.

Keywords: Red beetroot, beet, food colourant, betalains, antioxidants

1. Introduction
Beet root, scientifically known as *Beta vulgaris* (L) is one of the well-known plants belonging to the family Chenopodiaceae. Members of this family are dicotyledonous (Gokhale and Lele, 2014) [19]. Apart from their use as natural and harmless pigments in the food industry, betalains are important from the medicinal point of view. In a recent study, the antiviral and antimicrobial effects of betalain pigments were reported (Strack *et al*. 2003) [48]. Furthermore, the antioxidant properties of betalains were demonstrated in a wide range of assays (Zakharova & Petrova 1998; Gentile *et al*. 2004; Pavlov *et al*. 2005; Číž *et al*. 2010) [62, 16, 38, 11], and it was reported that the enrichment of human low-density lipoproteins by betalains effectively increased the resistance to oxidation (Tesorie- re *et al*. 2003) [51].
Betalains are water – soluble, nitrogen containing pigment, found in high concentration in red beet (Beta vulgaris). There are red and yellow beet varieties that purely display betacyanin (reddish purple pigment) or betaxanthin (yellow pigment) respectively. In the common commercially available red beets, the level of betacyanin among red beet varieties varies from 0.44 to 0.60 g betalain/ kg and the yellow pigment ranges from 0.32 to 0.42 g vulgaxanthine/ kg. Betanin is a natural pigment with anti-oxidant properties used as a food colourant.
Betalains are attracting increasing attention because of their use for food coloring and their antioxidant and radical scavenging properties against certain oxidative stress-related disorders, anticancer, antiviral and antiparasitosis properties [1, 9-21].

2. Different varieties of Beetroot
There are three types of beet (Beta vulgaris ssp. vulgaris), of which the common or red beetroot (conditive variety) is consumed as a vegetable. Within this type we distinguish three more types according to their shape and size: spherical, elongate and intermediate. The other varieties are sugar beet (var. altissima), of white colour and chiefly intended for the sugar industry and mangelwurzel (var. alba) that is widely used as cattle food. Maroto classifies table beets (common or red beet) in two commercial groups according to the shaoe of the " roots':

- Elongate: "Larga Roja Virtudes", "Larga de Covent-Garden", "Cylindra', "Crapaudine', "Cheltenham" (up to 30-40 cm long).
- Rounded or flat-topped: "Roja de Egipto', "Roja Globo', "Detroit Mejorada', "Bikores", "Globe-Rondarka', "Dwegina', "Boltardy", "Redpack", "Globe-Faro', "Detroit-Precoz", "Detroit-Nero', "Detroit Dark-Red", "Negra de Egipto raza Emir", " Monopoly", "Aplastada de Egiptó.

This second type of table beets (rounded and flat-topped) is the most cultivated and the one preferred for exportation. Table beet is original from the Mediterranean Europe and North Africa. Later on it spread all over Europe to western India, forming a secondary diversity centre in the Near East.

3. Health benefits of Beet

Beetroot is a good tonic food for health. The main markets for beet greens and bunched beets are roadside, farmers markets and deliveries to wholesalers. The market for beetroot is not a large market but it is significant. With storage the marketing season may be extended for roots (Boswell, 1967) [8]. Beta vulgaris var. rubra revealed significant tumour inhibitory effects in skin and lung cancer (Kapadia *et al*., 1996) [26]. These findings suggest that beetroot ingestion can be a useful means to prevent development and progression of cancer. But extracts of beetroot also showed some antimicrobial activity on *Staphylococcus aureus* and on *Escherichia coli* and also antiviral effect was observed (Rauha *et al*., 2000; Prahoveanu *et al*., 1986) [43, 41]. It ranks among the ten most potent vegetables with respect to antioxidant property. It makes an excellent dietary supplement being not only rich in minerals, nutrients and vitamins but also has unique phytoconstituents, which have several medicinal properties (Odoh and Okoro, 2013) [35]. Recent studies have provided that beetroot ingestion lead to beneficial physiological effects that may improve the treatments such as; hypertension, atherosclerosis, type 2 diabetes and dementia (Gilchrist *et al*., 2014; Beard *et al*., 2011; Vanhatalo *et al*., 2010) [18, 7, 55]. Hypertension in particular has been the target of many therapeutic interventions and there are numerous studies that show beetroot, delivered acutely as a juice supplement (Bailey *et al*., 2009; Jajja *et al*., 2014; Webb *et al*., 2008) [4, 24, 60] or in bread (Hobbs *et al*., 2013; Hobbs *et al*., 2012) [21, 22] significantly reduce systolic and diastolic blood pressure.

4. Beetroot Nutrition

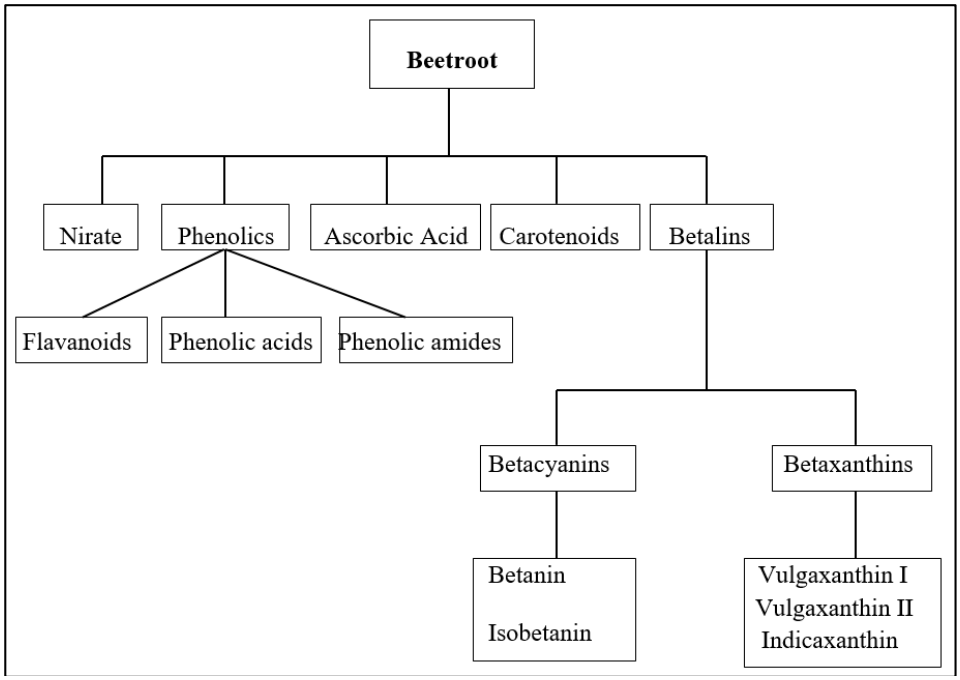

Fig 1: Overview of potentially bioactive compounds in beetroot based on data from (Ninfali and Angelino, 2013; Georgiev *et al*., 2010; Kujala *et al*., 2002) [34, 17, 27]

Beetroot is a rich source of phytochemical compounds that includes ascorbic acid, carotenoids, phenolic acids and flavonoids. Beetroot is also one of the few vegetables that contain a group of highly bioactive pigments known as betalains (Lee *et al*., 2005; Vulic *et al*., 2014) [29, 59]. Members of the betalain family are categorised as either betacyanin pigments that are red-violet in colour or betaxanthin pigments that are yellow-orange in colour. They are largely used as food colorants in food products like yogurts, ice cream and other products. A number of investigations have reported betalains to have high antioxidant and anti-inflammatory capabilities in vitro. (Przyjemska *et al*., 2009; Tesoriere *et al*., 2004; Vidal *et al*., 2014) [42, 52, 57]. This has generated interest to use beetroot in clinical pathologies characterised by oxidative stress and chronic inflammation such as liver disease, arthritis and even cancer.

5. Betalain as an anti- oxidant

When our body cells use oxygen, they produce free radicals as byproducts of normal cellular oxidative metabolism. Excessive free radical concentration in the body cause damage to DNA, lipids, proteins, and natural defense enzymes. Nutraceiticals such as antioxidants and food colorants act as free radical scavengers and prevent oxidative damage to our body. Betalain is considered as a potential nutraceuticals.

The betalain pigments has been shown by several in vitro studies to protect cellular components from oxidative injury (Kanner *et al*., 2001; Reddy *et al*., 2005; Tesoriere *et al*., 2008) [25, 44, 53]. For example, in the study by Kanner *et al*., (2001) [25] two betalain metabolites (betanin and betanidin) were shown to reduce linoleate damage induced by cytochrome C oxidase and lipid membrane oxidation induced by H_2O_2-activated metmyoglobin and free iron (AA-Fe). The authors also reported that betanin, the most abundant betalain found in beetroot (300-600 $mg \cdot kg^{-1}$), was the most effective inhibitor of lipid peroxidation. Betanin's high antioxidant activity appeared to stem from its exceptional electron donating capacity and ability to defuse highly reactive radicals targeting cell membranes. However, as alluded to earlier, betalains are not the only antioxidant compounds present in beetroot. Beetroot contains several highly bioactive phenolics, such as rutin, epicatechin and caffeic acid which are also known to be excellent antioxidants (Manach *et al*., 2005; Frank *et al*., 2005) [33, 15].

Furthermore, nitrite and other NO donors a kin to beetroot have been shown to suppress radical formation and directly scavenge potentially damaging free radicals such as superoxide and hydrogen peroxide, suggesting nitrate may also exhibit antioxidant effects (Lundberg *et al*., 2011; Wink *et al*., 2001) [31, 61].

6. Betalains Extraction

Betanine can be obtained from beetroot by milling followed by pressing, filtration and evaporation of the resulted juice (Dobre and Floarea, 1997) [12]. The product of this process is a red powder. Solid liquid extraction, carried out under conditions which lead to a maximum extraction yield and minimum pigment degradation, continues to be a useful method for obtaining beetroot juice. Extraction techniques such as continuous diffusion (counter current extraction) with subsequent enzyme treatment are currently used. Betalains extracted using these methods contain large amounts of salts, sugars and other impurities such as proteins. Even though fermentation is carried out to remove sugars, it is a time consuming process (3-7 days) and a considerable loss of pigments occurred (Pourrat *et al*., 1983; Thakur & Gupta, 2006) [40, 54]. Pulsed electric field technique is also used in the extraction which is mainly helpful in increasing juice extraction efficiency and shelf life of the juice. However, a drawback with this method is when it combines with dielectric breakdown non-conductive molecules within the structure become conductive (Fincan *et al*., 2004; Takhistov, 2006) [14, 50].

Aqueous membrane technology offers potential for selective separation of betalain compounds in a more efficient and acceptable form. There are only two reported studies on the clarification and filtration of beetroot juice using enzyme treatment followed by ultrafiltration/reverse osmosis (Lee *et al*., 1982; Thakur & Gupta, 2006) [30, 54].

Membrane technology successfully separated and concentrated betalains. Microfiltration of beet juice helped to remove 99% turbidity and achieve stable flux with subsequent filtrations (Mereddy *et al*., 2017). The LRO (loose reverse osmosis) membrane effectively separated betalains with 98% rejection. Up to 96% salts and 47% of dissolved solids were separated from betalains on concentration and defiltration. They reported on the use of membrane technology for complete removal of salts, nitrate and reduction of other dissolved solids to a significant level in addition to obtaining a betalains rich concentrate from beetroot juice. Moreover, membrane filtration can be the potential green technology for beetroot juice industries and has been widely used for recovery of bioactive compounds in various large-scale industrial operations such as dairy, wine and waste water (Avula *et al*., 2009; Antuono *et al*., 2014; Kumar *et al*., 2013; Rektor *et al*., 2004) [3, 1, 28, 45].

Sturzoiu *et al*., (2011) [49] used solvent extraction method and tested week acid solutions for use as solvents for betanin extraction from dried beetroot and reported that aqueous solution of citric acid 0.2% and ascorbic acid 0.1% and aqueous solution of ethanol 20% and ascorbic acid 0.5% are best suited for betanin extraction and also reported good stability and extraction yield of betanin in both solvents.

7. Impact of processing on betalains

Extracted betalains are susceptible to pH, water activity, and exposure to light, oxygen, metalions, temperature and enzymatic activities However temperature is the most decisive factor for betalain decomposition within the optimal pH. However, processing changes the content of betalains and consequently food color as well as the antioxidant activity. The susceptibility of betalains to the above mentioned factors restricts their use as food colorants. Betalains exhibit broad pH stability which are suited for low-acid foods where coloring with anthocyanins usally not possible (Stintzing & Carle, 2004) [47].

Processing of foods include thermal process such as microwaving, boiling, roasting or non-thermal process such as vacuum treatment. The main disadvantage in thermal processing includes betalain degradation under high temperatures. Studies reported that increasing betalain degradation rates resulting from increasing temperatures (Barrera *et al*., 1998) [5]. A previous enzyme inactivation by a short heat treatment of 70 °C for 2 min of the extract is desirable, to avoid betalain enzymatic degradation (Vargas *et al*., 2000) [56]. According to Strack *et al*., (2003) [48] acidification improves betacyanin stability and avoids oxidation by polyphenoloxidases. Red beets have several endogenous enzymes such as β-glucosidases, polyphenoloxidases and peroxidases, which if not properly inactivated may result in betalain degradation and colour losses (Escribano *et al*., 2002) [13].

Vacuum treatment includes removal of available oxygen or under low oxygen levels results in decreased pigment degradation than under air atmosphere. Low oxygen levels favor the pigment to be partially recovered after degradation (Von *et al*., 1981) [58]. Betalains are known to be sensitive to oxidation, which has an impact on their color stability (Herbach *et al*., 2006) [20]. Temperature is the important factor influencing betalain stability. Previous studies done by Barrera *et al*., (1998) [6] have reported betalain degradation with increase in temperatures.

Roy *et al*., (2004) [46] have shown that the extraction of betalains from red beet was optimal at 40°C. Boiling and roasting led to decrease in the yield of the obtained betacyanins and betaxanthins. Herbach *et al*., (2006) [20] explained that thermal treatments decreased betalains stability.

8. Conclusion

Betalains have both aesthetic values and positive health effects in food, unlike synthetic colorants which may trigger adverse effects in humans. Betalains are also water-soluble which facilitates their incorporation into aqueous food system. The pharmacological properties of betalains-rich foods such as red beetroot show their great potential as functional foods.

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