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RESEARCH ARTICLE

Phytochemical and antioxidative potential of orange, red, yellow, rainbow and black coloured tropical carrots (*Daucus carota* subsp. *sativus* Schubl. & Martens)

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Abstract This study was executed to determine phytochemical content i.e. total carotenoids, phenolics and flavonoids, and antioxidant ability expressed in the form of FRAP, CUPRAC and ABTS activity among different coloured tropical carrots (orange, red, yellow, rainbow and black carrot) developed at ICAR-IIVR, Varanasi, Uttar Pradesh, India. Overall, within different colour group, the extent of variation for various phytochemical content and antioxidant potentiality is narrow i.e. ranged from 1.04- to 3.21-fold; but at the same time, the genotypic variability across genotypes is too wide which varied 20.90- to 57.92-fold for phytochemical and antioxidants is an indication of broad genetic base of carrot germplasm. Among all the carrots, black carrot had an exceptionally high content of total phenolics and flavonoids, and thereby led to the highest antioxidant ability in the terms of FRAP, CUPRAC and ABTS activity expressing about 76–83% relative potentiality followed by rainbow carrot, and least in orange, red and yellow carrot (black carrot > rainbow carrot > red carrot ≈ orange carrot ≈ yellow carrot). The content of phenolics and flavonoids were highly correlated with antioxidant activity (0.955** to 0.992**). However, the most cultivated and consumed carrots, orange and red one, possessed higher amount of carotenoids. The content of carotenoids negatively correlated with total phenolics, flavonoids and antioxidants activity (− 0.612** to − 0.627**). Broad genetic base and selection based on total phenolics content could be pivotal in the future breeding to harness the genetic wealth of carrot efficiently.

Keywords Tropical carrot · Antioxidant · Carotenoids · Phenols · Phytochemical

Introduction

Phytochemicals are biologically active compounds produced by plants through primary and/or secondary metabolism which have been identified as being beneficial to human as well as plant health in various ways. They are classified into the major categories such as carotenoids, polyphenols and organosulfides (glucosinolates, allicin and glutathione). The carotenoids are tetraterpenoids, lipid-soluble organic pigments that are mostly produced by plants, algae and several bacteria and fungi. Structurally, carotenoids take the form of a polyene hydrocarbon chain with or without additional oxygen and soluble in lipids (lipophilic), named as xanthophylls (lutein, zeaxanthin, cryptoxanthin, astaxanthin, etc.) and carotenes (α-carotene, β-carotene, γ-carotene, δ-carotene, β-zeacarotene, lycopene, phytoene, etc.), respectively. Carotenoids are responsible for the brilliant colours ranging from pale yellow through bright orange to deep red, and absorb wavelengths ranging from 400 to 550 nm (Eldahshan and Singab 2013; Anonymous 2017). Polyphenols are secondary metabolites of plants, soluble in water (hydrophilic), derived from phenylalanine or shikimic acid which generally contribute to the colour, bitterness, astringency, flavour, odour and oxidative stability. The main classes of polyphenols include phenolic acids, flavonoids (anthocyanins, flavones, flavanones, isoflavones, flavonols, etc.), stilbenes and lignans (Pandey and Rizvi

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2009). The majority of recent research trend focuses on carotenoids and anthocyanins, two bioactive pigments responsible for the colour of the vast majority of fruits and vegetables, including carrot.

Antioxidants are the molecules that inhibit the process of oxidation (the chemical reaction that produces free radicals) of other molecules. They are grouped into two broad categories, hydrophilic and lipophilic. Usually, water-soluble antioxidants react with oxidants in the cell cytosol and the blood plasma; while lipid-soluble antioxidants protect cell membranes from lipid peroxidation. Most of the disorders/illness in humans and plants are aroused through the production of deleterious reactive oxygen species (ROS) or free radicals which are invariably produced during metabolic processes as well as exposure to various stresses. The endogenous antioxidant enzymes such as superoxide dismutase, peroxidase and catalase counteract the detrimental effects of ROS. However, conditions when ROS production is excessive, or when the antioxidant system is insufficient may favour oxidative damage to aerobic life (Singh et al. 2009) responsible for various chronic diseases like cancer, cardiovascular diseases, rheumatoid arthritis and age related macular degeneration in human (Halliwell 1991); and biotic and abiotic disorders in plants. The effects of ROS can be balanced by natural dietary antioxidants and antioxidant enzymes in living organisms (Alezandro et al. 2013; Singh et al. 2009). Fruits and vegetables are rich sources of natural antioxidants like carotenoids, phenolics, flavonoids, vitamin C, tocopherol, organosulfides and thus they reduce the risk of chronic diseases (Kaur and Kapoor 2001; Boeing et al. 2012; Manchali et al. 2012; Koley et al. 2014).

Carrot (*Daucus carota* subsp. *sativus* Schubl. & Martens), a member of Apiaceae family, is an important nutritious salad vegetable grown and consumed throughout the world for fleshy roots. The roots are eaten as crunchy salad; used to prepare juice, sweet and halwa; cooked with mixed vegetables; and preserved by salting, pickling, canning and drying. Carrot has numerous categories varying in colour, shape, size and flavour of roots, and vernalization requirement to complete life cycle [(a) Tropical/Oriental/Eastern carrot does not require vernalization, annual in nature; and (b) Temperate/European/Western carrot requires vernalization, biennial in nature]. Afghanistan is recognized as the primary center of origin because of the existence of huge diversity in morphological characters. Purple carrots from the place of origin, together with a yellow variant, spread to the Mediterranean area and Western Europe in the 11th to 14th centuries; and to China, India and Japan in the 14th to 17th centuries. The orange types appeared as a chance mutation in the Netherlands during the 17th century, and typical to temperate climates only. The orange carrot; because of improved colour, flavour, cylindrical shape, smooth surface, uniformity and tolerant to bolting; became popular in Europe and thereafter all over the world, and dominates in production and consumption. While red carrot finds its place up to some extent in Asian countries, including India. Primarily, carrot is classified on the basis of colour of roots which varies from white to black colour due to presence/absence of different organic pigments; but the carotenoid pigments (yellow, orange and red colour) and anthocyanin pigments (purple and dark purple i.e. black colour) are most common and of commercial importance worldwide (Singh 2016). Each unique colour of carrots has different pigments and health benefits. The orange carrots contain β-carotene and α-carotene, important for healthy vision and immune system; red carrots possess lycopene, associated with the reduced risk of heart diseases and cancers; yellow carrots have xanthophylls and lutein, significantly reduces the risk of atherosclerosis, macular degeneration and cancers; black/purple carrots contain anthocyanins, possessing properties of anti-bacterial, anti-fungicidal and anti-inflammatory, and inhibit the bad cholesterol; and white carrots lack any pigmentation and are least healthy (Simon et al. 1989; Rubatzky et al. 1999; Zhang and Hamauzu 2004). Black carrot, also mentioned as solid purple in other literatures, is one of the richest and best sources of anthocyanins having very high anti-oxidative potential, nutraceutical values and colorant properties. Moreover, Rainbow carrot is a category of carrot selectively bred for multiple pigments in single root that reflects almost all colours of rainbow. Generally, rainbow carrot hued with purple pigmentation on root exterior (periderm), and orange, red, purple and yellow pigmentation in the tissues of root interior i.e. cortex and core (phloem, cambium and xylem) are the good source of organic pigments containing balanced amounts of xanthophylls, β-carotene, α-carotene, lycopene and anthocyanins. It is usually a good idea to eat a colorful assortment of fruits and vegetables every day to get a wide array of vitamins, antioxidants and minerals; rainbow carrots can certainly help in that regard.

Usually, orange carrots are used for table, frozen and powder purposes; red carrots for table, juice, halwa and pickle making; and black carrots for colour extraction, halwa and preparation of a beverage called kanji. The rising health awareness among the aging population, children and pregnant women are major drivers propelling the demand for dietary supplements; carrot is one of the best candidates among root vegetables because of its virtue for phytochemical and antioxidant properties. Among different coloured carrots; the orange carrots, typical to temperate conditions only, probably developed from mutations of yellow forms followed by human selection which thought to be originated in the Netherlands (Stolarczyk and Janick 2011). The present investigation includes twenty-five

genotypes of tropical carrot, including recently developed three genotypes of orange-type and categorized in five colour groups. There are reports available on quantification of few biochemical compounds and antioxidant ability among either temperate carrots or temperate and tropical carrots involving few to more genotypes (Simon et al. 1989; Alasalvar et al. 2001; Nicolle et al. 2004; Zhang and Hamauzu 2004; Grassmann et al. 2007; Leja et al. 2013; Koley et al. 2014); but very fragmented information is available on phytochemical and antioxidant potentiality of different coloured tropical carrots. Therefore, the present study convinced with the objective to assess the potential of phytochemicals and antioxidants in orange, red, yellow, rainbow and black coloured carrots developed exclusively in tropical conditions.

Materials and methods

Basic experimental materials

Twenty-five diverse genotypes of tropical carrot, annual in nature and possessing self-coloured core; including eleven of red colour, five of black colour, and three each of orange, rainbow and yellow carrot; comprised the basic experimental materials which were evaluated for this study. All the genotypes under study are developed and maintained at ICAR-Indian Institute of Vegetable Research (ICAR-IIVR), Shahanshahpur-221305, Varanasi, Uttar Pradesh, India.

Field trial

The crops were grown in open field during 2015–2016 and 2016–2017 at the Research Farm, ICAR-IIVR, Varanasi, Uttar Pradesh, India. The experimental site is located at 25°10′55″N latitude, 82°52′36″E longitude and 85 m altitude which receives an annual rainfall of 1050–1100 mm. The soil of the Farm was of silt-loam in texture, having pH 7.3 and electrical conductivity of 0.28 dSm^{-1}. During both year of experimentation, soil preparation, sowing and other agronomic practices were carried out unvaryingly as mentioned by Singh and Karmakar (2015) for better morphological expression and root development. Carrot seeds, botanically schizocarp, were sown at 1.0–1.5 cm interval in double row of 7–8 cm apart and 25–28 cm wide ridge with the spacing of 80 cm between each pair of ridges. Each genotype comprises four ridges of 5.50 m long and replicated three times following randomized block design. The crop was uniformly fed with doses of chemical fertilizers i.e. 80 kg N, 50 kg P_2O_5 and 40 kg K_2O per hectare; supplied in the form of urea, single super phosphate and muriate of potash, respectively. Half of the N, and full of the P_2O_5 and K_2O were applied at the time of ridges preparation, while remaining half dose of N was dressed in furrows at 45 days after sowing. To keep the plant to plant spacing about 4–5 cm apart, manual thinning was done after 15–18 days of seed germination for proper root development.

Root sampling and laboratory analyses

Each genotype in replicated trial was uprooted at marketable maturity i.e. 95–105 days after sowing. Eight roots of each genotype selected arbitrarily and the leaf-tops were cut with stainless steel knife. The roots were washed thoroughly with tap water to remove adhering soil, secondary roots and other extraneous substances. A representative sample of the edible root i.e. 6–7 cm long piece from middle portion from all selected roots were rapidly cut into thin slices, put into transparent polyethylene bag, mixed properly, and 5 g of composite root sample was taken for subsequent assay and estimation on fresh weight (FW) basis.

Methods to estimate phytochemical

Determination of total carotenoids

The carrot sample of 100 g was repeatedly extracted with acetone using a pestle and mortar until the residue was colourless. The pooled acetone extracts were transferred into separatory funnel and the entire carotenoids were transferred to petroleum ether phase. The concentration was measured using UV–visible spectrophotometer (SHIMADZU UV 1601) at 452 nm and calculated as mg/100 g FW.

Extraction of hydrophilic fraction

Water soluble phytochemicals of carrot were extracted according to the method reported previously (Chu et al. 2002) with slight modification. For the extraction of soluble nutraceuticals, 5 g sample was homogenized with 80% ethanol (1:2 w/v) using a chilled warring blender for 5 min. Extraction of the residue was repeated for two times using the same conditions. The mixture was then further homogenized using a polytron homogenizer for an additional 3 min to obtain a thoroughly homogenized sample. The homogenates were centrifuge at 13,000 rpm for 15 min. The solvent was then removed using a rotary evaporator. The supernatants were collected in 50-mL plastic centrifuge tubes and then stored at − 20 °C until analysis (within next few days) of total phenolics and flavonoids content; and activity of ferric reducing antioxidant power (FRAP), cupric reducing antioxidant capacity

(CUPRAC), and 2,2′-Azino-bis (3-ethylbenzthiazoline-6-sulfonic acid) diammonium salt (ABTS), a peroxidase substrate.

Determination of total phenolics content

Total phenolics content in carrot root was estimated spectrophotometrically using Folin–Ciocalteu reagent (Singleton et al. 1999). Aliquots (100 µL) of hydrophilic extract were mixed with 2.9 mL of deionized water, 0.5 mL of Folin–Ciocalteu reagent and 2.0 mL of 20% Na_2CO_3 solution. The mixture was allowed to stand for 90 min and absorption was measured at 760 nm against a reagent blank in UV–visible spectrophotometer. Results were expressed as gallic acid equivalent (mg GAE/100 g FW).

Determination of total flavonoid

The concentration of total flavonoids in carrot root sample was analysed using aluminium chloride method (Zhishen et al. 1999). An aliquot (1 mL) of hydrophilic extract in 10 mL of volumetric flask containing 4 mL of distilled water, 0.3 mL portion of 5% $NaNO_2$ and 0.3 mL portion of 10% $AlCl_3$, $6H_2O$. The mixture was allowed to stand for 6 min at room temperature. Two millilitres of 1 mol NaOH was added and the solution was diluted to 10 mL with distilled water. Immediately, the absorbance of the solution versus a blank was measured at 510 nm. The results were expressed as catechin equivalent (mg CE/100 g FW).

Methods to estimate antioxidant capacity

FRAP assay

The FRAP activity determine compounds able to reduce ferric ions, was performed according to the procedure described by Benzie and Strain (1996). In brief, initially FRAP reagent was prepared by mixing 300 mM acetate buffer (pH 3.6), 10 mM TPTZ in 40 mM HCl and 20 mM $FeCl_3$ in the ratio 10:1:1 (v/v/v). Thereafter, 100 µL aliquot of hydrophilic extract was thoroughly mixed with 3 mL of the FRAP reagent in a test tube and vortexed in the incubator at 37 °C for 30 min in a water bath. An intense blue colour was formed due to reduction of ferric-tripyridyltriazine to the ferrous complex. The absorbance was measured at 593 nm using UV–visible spectrophotometer. Results were expressed in terms of trolox equivalent (µmol TE/g FW).

CUPRAC assay

The CUPRAC assay, determine compounds able to reduce cupric ions, of root was analysed following the method of Apak et al. (2008). Briefly, according to the protocol, 100 µL of hydrophilic extract was mixed with 1 mL each of $CuCl_2$ solution (1.0×10^{-2} mol/L), neocuproine alcoholic solution (7.5×10^{-3} mol/L), and NH_4Ac (1 mol/L, pH 7.0) buffer solution and 1 mL of water to make the final volume 4.1 mL. Then the mixture was incubated at 50 °C in a water bath. After 30 min, the absorbance was recorded at 450 nm against the reagent blank. Standard curve was prepared using different concentration of Trolox and results were expressed in terms of µmol TE/g FW, using molar absorptivity of Trolox as 1.67×10^4 L mol^{-1} cm^{-1}.

ABTS assay

ABTS activity, a peroxidase substrate, as Trolox equivalent antioxidant capacity (TEAC) in carrot roots was determined through a modified procedure using ABTS as described by Re et al. (1999). The ABTS* + free radical (7 mmol) was prepared through reaction of 7 mmol ABTS and 2.45 mmol of potassium persulphate as the oxidant agent. Aliquots (10 µL) of extract was added to 90 µL of ABTS* + solution and absorbance readings at 734 nm were taken at 30 °C exactly 10 min after initial mixing. The percentage inhibition of ABTS of the test sample and known solutions of Trolox were calculated by the following formula:

$$\%Inhibition = 100 \times (A_0 - A)/A_0$$

where A_0 was the initial absorbance at 734 nm, obtained by measuring the same volume of solvent, and A was the final absorbance of the sample extract at 734 nm. The radical-scavenging activity of the test samples was expressed as TEAC µmol TE/g FW.

Statistical analyses

Data were collected from triplicate experiments during the both years for each genotype and were analysed statistically for analysis of variance as per Singh and Chaudhary (1977). To reveal the potentiality of different coloured carrots for various phytochemicals and antioxidants, relative content (%) was calculated by multiplying average quantified value of each colour group to 100 and divided by sum of quantified values of all five colour group (quantified value of each colour group*100/sum of quantified values of all five colour group). The pooled data of both years were used for making histograms to compare mean differences among coloured genotypes of carrot by using standard error bars with $p < 0.05$ through Microsoft Excel, and

correlation analysis was done as mentioned by Singh and Chaudhary (1977).

Results and Discussion

The data of years 2015–2016 and 2016–2017 for the content of total carotenoids, total phenolics and total flavonoids, and antioxidant ability in terms of FRAP, CUPRAC and ABTS value were analysed for analysis of variance to observe the effect of year and colour segments. Between the two experimental years, there were no considerable differences found among genotypes; but highly significant variations were comprehended among genotypes of different coloured carrots. Hence, the pooled data of both years and different colour segment are presented in various Figures. Since, the traits under this study were qualitative in nature, easily identifiable, and the crops were raised in almost similar climatic conditions during both years, it is obvious to get least differences over the year and lack any interactions between both years for various estimates. The significant variability among genotypes; and non-significant differences between the two experimental years for total phenols, phenylpropanoids, flavonoids, anthocyanins and antioxidant activity (DPPH) was also reported by Leja et al. (2013) in carrot. The genotypic variability for carotenoids, phenols, flavonoids, anthocyanins and antioxidants has been reported in carrots and other vegetables by various researchers (Nicolle et al. 2004; Zhang and Hamauzu 2004; Grassmann et al. 2007; Singh et al. 2011; Karmakar et al. 2013; Leja et al. 2013; Koley et al. 2014).

Total carotenoids content (mg/100 g FW) varied greatly i.e. 0.47–9.90 mg (20.90-fold) among genotypes of different coloured carrots with a general mean value of 4.78 mg (Fig. 1a); and it was estimated highest in red root (7.37 mg, 1.36-fold) followed by rainbow root (7.14 mg, 1.90-fold), orange root (3.94 mg, 1.09-fold), black root (0.66 mg, 2.65-fold), and least in yellow roots (0.64 mg, 1.42-fold). Total carotenoids was significantly at par in red and rainbow carrots, and very less in black and yellow carrots (Fig. 1a); which reveals that the carotenes, especially two pigments β-carotene and lycopene is the main constituent of respective orange and red carrot, contribute maximum towards total carotenoids and least by xanthophylls. This result is in concurrence with the finding of Surles et al. (2004) who estimated 9.8 ± 1.4 mg and 0.71 ± 0.38 mg of total carotenoids (per 100 g FW) in red and yellow carrots, respectively. The relative content (%) of total carotenoids (Fig. 2) was calculated to be maximum for red carrot (37.3) followed by rainbow carrot (36.2), orange carrot (19.9), and least in black carrot (3.4) and yellow carrot (3.2) which is following the trend as red carrot ≈ rainbow carrot > orange carrot > black carrot ≈ yellow carrot. The present results are in accordance with the previous findings by Alasalvar et al. (2001) and Grassmann et al. (2007) who reported the lower content of carotenoids (< 2 mg/100 g FW) in yellow carrots and solid purple (black) carrots.

Total phenolics concentration (mg GAE/100 g FW) showed 40.84-times variation among the highest and lowest performing genotypes with a range of 7.80–318.37 mg and an average phenolics level of 68.72 mg (Fig. 1b). Moreover, among different colour roots investigated, phenolics content was maximum in black carrots (268.08 mg, 1.66-fold) followed by rainbow carrots (41.66 mg, 1.79-fold), significantly at par in red and orange carrots (16.06 mg, 2.04-fold and 14.52 mg, 1.46-fold) and minimum in yellow carrots (10.82 mg, 1.83-fold). Further, the trend of the relative content of total phenolics (Fig. 2) are as follows: black carrot (76.3%) > rainbow carrot (11.9%) > red carrot (4.6%) ≈ orange carrot (4.1%) > yellow carrot (3.1%). Higher content of phenolics in black (solid purple) and/or rainbow (purple with orange/red core) carrots have been reported by Alasalvar et al. (2001), Grassmann et al. (2007), Leja et al. (2013) and Koley et al. (2014). Another polyphenol, flavonoids content (mg CE/100 g FW) among different coloured genotypes of carrot showed a large extent of variation ranging from 3.61 to 140.04 mg with a mean value of 29.17 mg (Fig. 1c). Like phenolic content, the content of flavonoids was also estimated maximum in black carrots (118.93 mg, 1.32-fold) followed by rainbow carrots (13.14 mg, 3.21-fold), significantly at par in red and orange carrots (6.06 mg, 1.69-fold and 5.33 mg, 1.04-fold) and minimum in yellow carrots (4.19 mg, 1.25-fold). Similar to total phenolics, the relative content of flavonoids (Fig. 2) too followed the same trend with different intensity such as black carrot (80.5%) > rainbow carrot (8.9%) > red carrot (4.1%) ≈ orange carrot (3.6%) > yellow carrot (2.8%). It is worthwhile to mention that the average phenolic content is higher (> two-folds) than the corresponding total flavonoids in different carrot groups. In the plant system, the most common and abundant flavonoids are the anthocyanins, flavonols and flavones. In carrot, anthocyanins are responsible for the purple and black colour of roots which is being reflected in the present analysis with higher values of phenolics and flavonoids in the roots of black and rainbow carrots. Leja et al. (2013) and Koley et al. (2014) estimated the highest amount of flavonoids in solid purple/black coloured carrots.

The dietary antioxidants are biochemical molecules that inhibit the process of oxidation. In nutritional research of food items, the evaluation of antioxidant activity is becoming more important as it provides useful information with regard to health promoting functional quality of food items. Various natural antioxidants work in a synergistical

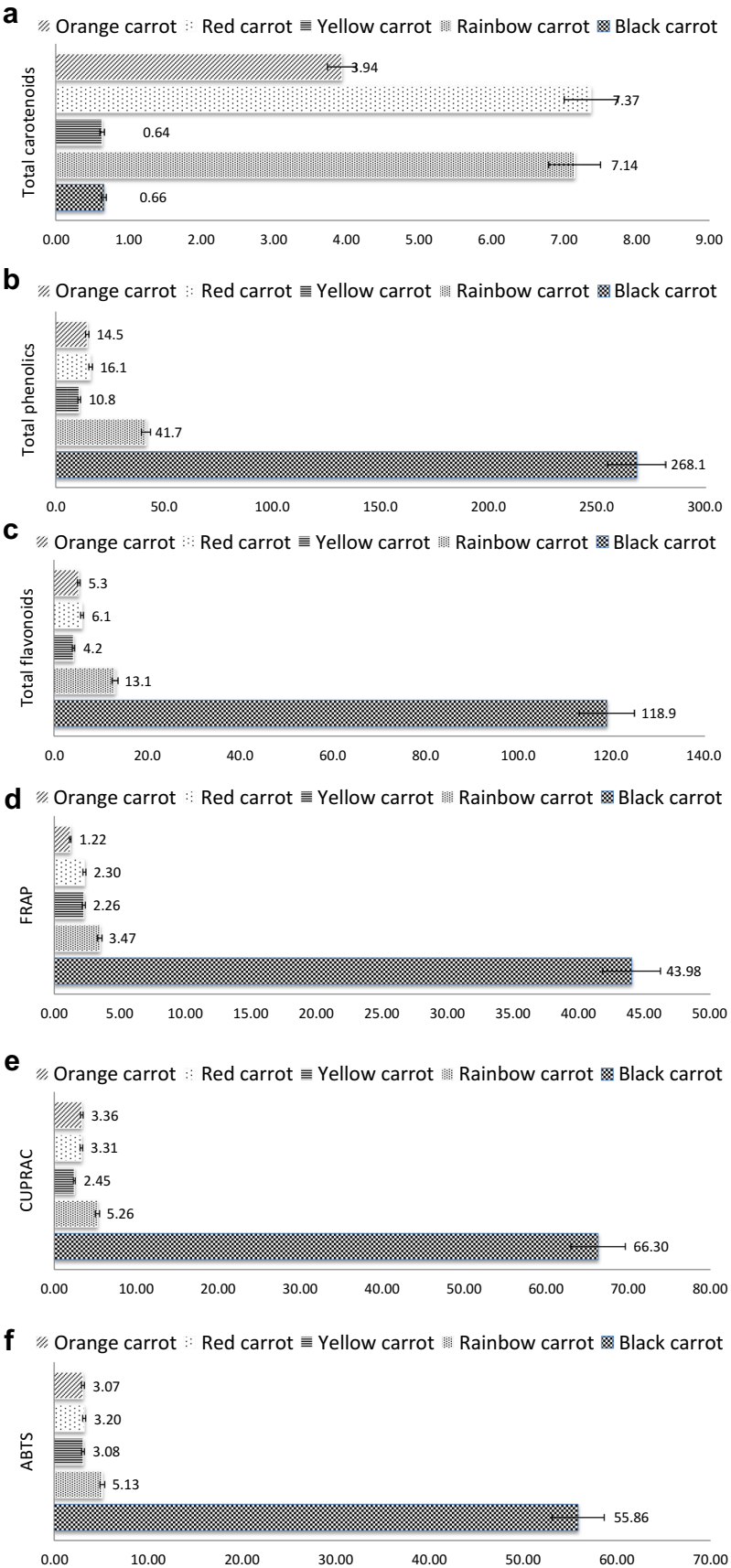

Fig. 1 **a** Total carotenoids content (mg/100 g FW) in orange, red, yellow, rainbow and black coloured tropical carrots (standard error bars with $p < 0.05$). **b** Total phenolics content (mg GAE/100 g FW) in orange, red, yellow, rainbow and black coloured tropical carrots (standard error bars with $p < 0.05$). **c** Total flavonoids content (mg CE/100 g FW) in orange, red, yellow, rainbow and black coloured tropical carrots (standard error bars with $p < 0.05$). **d** FRAP activity (μmol TE/g FW) in orange, red, yellow, rainbow and black coloured tropical carrots (standard error bars with $p < 0.05$). **e** CUPRAC activity (μmol TE/g FW) in orange, red, yellow, rainbow and black coloured tropical carrots (standard error bars with $p < 0.05$). **f** ABTS activity (μmol TE/g FW) in orange, red, yellow, rainbow and black coloured tropical carrots (standard error bars with $p < 0.05$)

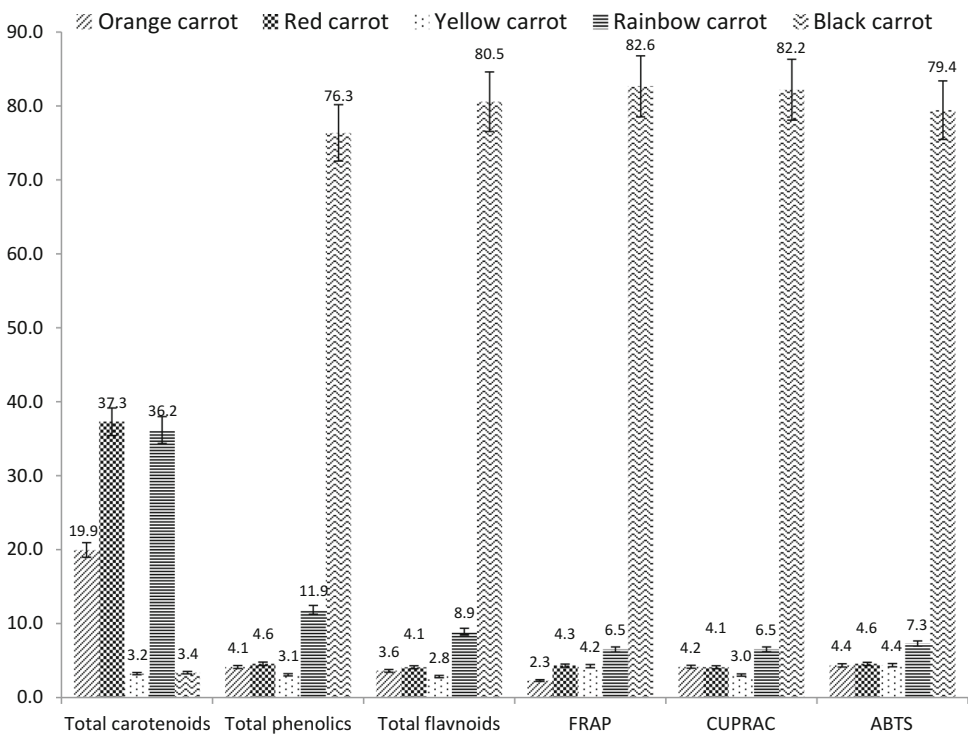

Fig. 2 Relative content and activity (%) of phytochemicals and antioxidants in orange, red, yellow, rainbow and black coloured tropical carrots (standard error bars with $p < 0.05$)

and/or antagonistic manner. In the present experimentation, the antioxidant activity of various carrots was estimated by three different in vitro methods i.e. FRAP, CUPRAC and ABTS assay to compare the potential of orange, red, yellow, rainbow and black carrots. FRAP value (µmol TE/g FW) in tropical carrots ranged from 1.03 to 59.40 µmol (57.92-fold difference) with a mean of 10.64 µmol for 25 genotypes having self-coloured core (Fig. 1d). It was analysed higher in black carrot (43.98 µmol, 2.32-fold) followed by rainbow carrot (3.47 µmol, 1.93-fold), significantly at par in red carrot (2.30 µmol, 2.93-fold) and yellow carrot (2.26 µmol, 1.09-fold), and minimum in widely consumed orange carrot (1.22 µmol, 1.19-fold). Additionally, the relative antioxidant potential (Fig. 2) in terms of FRAP activity is very high for black carrot which followed the pattern as black carrot (82.6%) > rainbow carrot (6.5%) > red carrot (4.3%) ≈ yellow carrot (4.2%) > orange carrot (2.3%). The higher value for black carrot in comparison to the other carrots is due to the presence of elevated content of polyphenols, especially anthocyanins. The trend of present result is in concurrence with those of previous studies (Bouayed et al. 2011; Koley et al. 2014). Further, the antioxidant potential in terms of CUPRAC value (µmol TE/g FW) among various tropical carrots varied 34.97-folds ranging from 2.38 to 83.23 µmol with an average value of 16.04 µmol (Fig. 1e). In five different colour groups, CUPRAC value was assayed highest in black carrot (66.30 µmol, 2.10-fold), followed by rainbow carrot (5.26 µmol, 2.40-fold), significantly equivalent in red carrot (2.30 µmol, 2.93-fold) and yellow carrot (2.26 µmol, 1.09-fold), and lowest in widely cultivated and consumed orange carrot (1.22 µmol, 1.19-fold). The higher average of CUPRAC values for black carrot in comparison to the other carrots is due to the presence of polyphenols, especially anthocyanins. The present results follow the trend of previous studies (Koley et al. 2014). The ability to reduce metal ions was measured using CUPRAC and FRAP assay. Among five different colour group of carrot, the overall values of mean and range for CUPRAC assays were considerably higher than the values of FRAP assay. Higher antioxidant ability in CUPRAC may be credited because of the presence of flavonoids such as quercetin and kaempferol in carrot. A similar trend was reported Koley et al. (2014) in carrot.

Similar to FRAP and CUPRAC activities, the radical scavenging ability in terms of ABTS value (µmol TE/g FW) among various genotypes of tropical carrots ranged from 2.14 to 73.14 µmol and varied 34.23-folds with a mean value of 13.93 µmol (Fig. 1f). Among five coloured group, the ability to scavenge free radicals was estimated too low in orange, red and yellow carrots i.e. 3.07, 3.20 and 3.08 µmol, respectively which is statistically at par. Moreover, it was quantified maximum in black carrot (55.86 µmol) followed by rainbow type (5.13 µmol). The relative antioxidant potentiality in terms of ABTS among different carrot types were as follows—black carrot

Table 1 Correlation coefficient between phytochemical/antioxidants in tropical carrot

Parameter	Total carotenoids	Total phenolics	Total flavonoids	FRAP activity	CUPRAC activity	ABTS activity
Total carotenoids	1.000					
Total phenolics	− 0.618**	1.000				
Total flavonoids	− 0.627**	0.981**	1.000			
FRAP activity	− 0.619**	0.990**	0.972**	1.000		
CUPRAC activity	− 0.619**	0.992**	0.974**	0.997**	1.000	
ABTS activity	− 0.612**	0.987**	0.955**	0.997**	0.996**	1.000

**Significant at 1% level

(79.4%) > rainbow carrot (7.3%) > red carrot (4.6%) ≈ orange carrot (4.4%) ≈ yellow carrot (4.4%). Overall, the antioxidant ability is higher in anthocyanins rich black carrot followed by rainbow carrot (contains blend of anthocyanins and carotenoids) and least in carotenoids rich, red, orange and yellow carrots. Purple carrots contained very high contents of phenolics and were characterized by a higher antioxidant capacity than orange, yellow or white varieties (Alasalvar et al. 2001). Moreover, Zhang and Hamauzu (2004) also found stronger radical scavenging ability in phenolic extracts than pure chlorogenic acid, vitamin C and β-carotene in carrot. In another study at Krakow, Poland; Leja et al. (2013) estimated radical scavenging ability (RSA%) among 35 genotypes, both temperate and tropical in nature, and five colour group of carrots which followed the trend as purple carrot (51.3) > red carrot (9.3) > orange carrot (6.6) ≈ red carrot (6.0) ≈ white carrot (6.0); the pattern is almost in agreement with the findings of present study at ICAR-IIVR, Varanasi, UP, India.

Red beetroot (*Beta vulgaris*), the main source of betalains, has been ranked among the 10 most potent antioxidant vegetables whose antioxidant potential in terms of FRAP and ABTS values on dry weight basis quantified as 40–125 mmol TE/kg DW (i.e. ≈ 4.0–12.5 µmol TE/g FW) and 30–110 mmol TE/kg DW (i.e. ≈ 3.0–11.0 µmol TE/g FW), respectively (Song et al. 2010; Carrillo et al. 2017). It is very interesting to note that the black carrots in the present investigation possessed > 500% higher antioxidant ability than red beetroot with respective FRAP, ABTS and CUPRAC values of 25.64–59.40 µmol TE/g FW, 26.39–73.14 µmol TE/g FW and 39.67–83.23 µmol TE/g FW. It reveals that among vegetables, the black carrots are one of the richest sources of phytochemicals and antioxidants that need to be promoted in the daily diet and utilized for the manufacturing of dietary food supplements.

The correlation coefficients (Table 1) between total carotenoids, and polyphenols (phenolics and flavonoids) and antioxidants (FRAP, CUPRAC and ABTS) were estimated significantly negative (− 0.612** to − 0.627**). Moreover, total phenolics and flavonoids showed very high estimates of correlation with all three dietary antioxidants i.e. 0.987** to 0.992** and 0.955** to 0.974**, respectively because of the presence of polyphenols in purple/black carrots. The activity of antioxidants FRAP, CUPRAC and ABTS were also highly associated with each other. Higher estimates of total phenolics over flavonoids indicate that phenolics could be used as a biochemical indicator while genetic improvement programmes pertaining to dietary antioxidants. The other researchers also observed a very good direct association between total phenolics/polyphenols and antioxidant ability (Leja et al. 2013; Koley et al. 2014; Singh et al. 2017; Carrillo et al. 2017) in root vegetables.

Conclusions

Overall, within the different colour group, the extent of variation for various phytochemical content and antioxidant ability is narrow i.e. ranged from 1.04- to 3.21-fold; but at the same time, the genotypic variability across genotypes is too wide that varied 20.90-fold to 57.92-fold for phytochemical and antioxidants. Remarkably, the phytochemicals such as total phenolics and flavonoids express very high association with all three dietary antioxidants i.e. FRAP, CUPRAC and ABTS. Wide genetic base and selection based on total phenolics content could be taken into consideration in the future breeding programmes to harness the genetic wealth of carrot for the benefit of mankind.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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