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Influence of leaf-cover on visual quality and health-promoting phytochemicals in loose-curd cauliflower florets

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ABSTRACT

Three cultivars were used to evaluate the influence of leaf-cover for three and six days on visual quality, health-promoting compounds and antioxidant capacity in loose-curd cauliflower. Leaf-cover significantly promoted the lightness and reduced chroma of the curd surface. Moreover, leaf-cover significantly reduced the contents of important antioxidants, including chlorophylls, carotenoids, and anthocyanins, as well as ascorbic acid in loose-curd cauliflower. Nevertheless, leaf-cover significantly induced the production of total phenolics in these loose-curd cauliflowers after three days. Furthermore, the changes of individual glucosinolates and ferric reducing antioxidant power were depended not only on the growing cover conditions but also on the genotypic characteristics. These results suggested that leaf-cover improved the visual quality but impaired the accumulation of some important health-promoting compounds in florets during the development of cauliflower curd.

1. Introduction

Loose-curd cauliflower (*Brassica oleracea* var. *botrytis*), typically characterized by loose-curds and long florets with green pedicels, is favored by Chinese consumers because its pedicels are tender and crispy with good flavor. In the past several years, the cultivation area and overall production of loose-curd cauliflower have surpassed that of common cauliflower in some major cauliflower production areas. This vegetable also has high nutritional value because of its high levels of major antioxidants and anticarcinogenic compounds, such as ascorbic acid (AA), carotenoids, and glucosinolates, compared with common cauliflower (Gu et al., 2014). The physiological functions of AA (Davey et al., 2000), phenolic compounds (Podsedek, 2007), carotenoids (Krinsky & Johnson, 2005), and chlorophyll derivatives (Ferruzzi, Böhm, Courtney, & Schwartz, 2002), as well as glucosinolates and their breakdown products (Traka & Mithen, 2009) in human beings have been reviewed previously. In addition, some glucosinolates and their myrosinase-hydrolysis products, extracted from cauliflower, notably glucobrassicin, glucoiberin, and gluconapin, contribute to the overall antioxidant capacity of cauliflower (Cabello-Hurtado, Gicquel, & Esnault, 2012). Epidemiological studies have shown that cruciferous vegetables could protect humans against various cancers, in particular bladder, colon, and lung cancer, and this effect may be attributable to these phytochemicals present in these vegetables.

Consumer choice and preference of fruits and vegetables are mainly influenced by several factors, such as convenience, culture, price, appearance, taste, and nutrition (Glanz, Basil, Maibach, Goldberg, & Snyder, 1998; Ragaert, Verbeke, Devlieghere, & Debevere, 2004), but the latter are difficult for consumers to determine. For a long time, many agrotechnical methods, including leaf-cover (LC) — notably a leaf broken to completely over the curd surface — have been used to improve the marketability of cauliflower curd, protecting it from sunscald and keeping it white (Forbes & Chapman, 1989; Naeve, 1997). It is important, however, to supply the consumer with products that are not only of good marketable quality but also with high nutritive value, properties that are undoubtedly affected by agrotechnical and technological factors. Previously, Thomas and Turner (1992) found that the content of flavonoids in the florets of common cauliflower covered by leaves or Tyvek was reduced significantly compared with uncovered samples. At present, self-wrapping cauliflower cultivars are widely grown, so these cultivars blanch very well and various covers are usually not required. However, self-wrapping loose-curd

Abbreviations: AA, Ascorbic acid; ANOVA, Analysis of variance; DW, Dry weight; FRAP, Ferric Reducing Antioxidant Power; FW, Fresh weight; GAE, Garlic acid equivalents; LC, Leaf-cover; ZAAS, Zhejiang Academy of Agricultural Science.

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cauliflower cultivars have not been used in production, hence LC, as the cheapest and one quick method for promoting the so called 'marketability' of cauliflower curds (Forbes & Chapman, 1989), is being widely used during the harvest period for loose-curd cauliflower. Exposure of growing loose-curds to sunlight may result in production of yellow-colored curds and green-colored pedicels, which implies some light-dependent pigments, such as carotenoids, chlorophylls, and flavonoids, might accumulate in the florets. In addition, other nutritive phytochemicals, including glucosinolates, AA and total phenolics, which are not visible may be affected by LC.

Some pre-harvest agricultural practices, including organic soil, choosing the correct harvest time and covering on curds, were designed to promote the phytochemical content or quality of cauliflower curds (Forbes & Chapman, 1989; Lo Scalzo, Bianchi, Genna, & Summa, 2007; Picchi et al., 2012). However, the effect of covering methods on quality of cauliflower, especially loose-curd cauliflower, has not been examined. In this study, three genotypes undergoing LC treatment for three and six days were used to determine the effect of LC on visual quality, antioxidant capacity, and health-promoting compounds in the florets of loose-curd cauliflower.

2. Material and methods

2.1. Plant materials

Three loose-curd cauliflower cultivars — 'ZHE047', 'ZHE091' and 'Qingnong65Tian' (QN65) — were analyzed in this study. The two former cultivars were developed and produced by the Zhejiang Academy of Agricultural Science (ZAAS), China, and the third was produced by Ching Long Seed Co., Ltd. In July 2011, seeds were germinated in growth matrix for plug seedlings in a greenhouse under natural day light. After four weeks of growth in the greenhouse, seedlings were transplanted into a field at the ZAAS experimental station in Hangzhou, China. At least 40 seedlings of each cultivar were planted in a ridge with 120 cm width as a plot with double rows. The plant spacing was about 50-cm. We used a randomized complete block experimental design with eight plots for each cultivar. All lines were cultivated under identical agronomic measurements until LC treatment.

After 20 days of floret appearance, more than 20 healthy cauliflower heads of uniform size in one plot were selected as control and for LC treatment, which involved a leaf being broken to completely cover the curd surface according to Forbes and Chapman's method (1989). At the third and six day, four mature and healthy cauliflower heads of uniform size per plot for each treatment were randomly cut, sent to the lab, and immediately stored in a refrigerator at 4 °C. Seven or eight lateral small balls 5–6 cm in length were selected and cut from each head of loose-curd cauliflower for analysis. The fresh samples were used for analyses of chlorophyll, AA and total carotenoid contents, while the other samples were frozen, lyophilized and stored at −20 °C for further analyses of total phenolics, antioxidant capacity, and glucosinolates.

2.2. Quality methods

2.2.1. Color

Color was measured at the surface of curd with a chromameter (CHROMA METER CR400, Osaka, Japan). The color at the same location, of five curds from each cultivar in one plot was recorded at the third and sixth day as L*, a* and b*. The meter was calibrated using the standard white plate provided by the manufacturer. The values were expressed in the CIE system and converted to $C^* = SQRT\ (a^{*2} + b^{*2})$ and hue angle, using the formula: $h^{\circ} = (ATAN\ (b^*/a^*)/6.2832) \times 360 + 180$, when $a^* < 0$ and $b^* > 0$ (McGuire, 1992). The photographs of curds were taken by a camera (Canon, Tokyo, Japan).

2.2.2. Chlorophylls and total carotenoids

The compounds were assayed according to the method of Lichtenthaler and Buschmann (2001). Small balls (4–5 g fresh weight, FW) were weighed and ground in 3 mL of ethanol:-water (96:4, v/v) solution, then washed with 17 mL of ethanol solution in a 50 mL tube. The extraction solution was centrifuged at 7000 × *g* at room temperature for 5 min. The residue was then re-extracted in 20 mL ethanol solution. The supernatants were collected and combined. Chlorophyll *a*, and *b* and total carotenoids were measured by reading the absorbance at 664, 649, and 470 nm, respectively, with a UV–Vis DU-800 spectrophotometer (Beckman Coulter Inc., Brea, USA). The concentrations of chlorophyll *a* (C*a*), chlorophyll *b* (C*b*), and the sum of the carotenoids C($x + c$) were calculated using the following equations: $Ca = 13.36A_{664} - 5.19A_{649}$, $Cb = 27.43A_{649} - 8.12A_{664}$, $C(x + c) = (1000A_{470} - 2.13Ca - 97.64Cb)/209$, and further expressed as micrograms per gram FW.

2.2.3. Ascorbic acid

AA content was analyzed as previously described with minor modifications (Gu et al., 2014). Fresh small balls (4–5 g) were ground in oxalic acid (Sangon, Shanghai, China): water (1 g:100 mL) solution on ice, then extracted twice with 40 mL oxalic acid solution and centrifuged at 7000 × *g* for 5 min at 4 °C. Each sample was filtered through a 0.45 μm cellulose acetate filter. HPLC analysis was carried out using a Waters 600 system with a 717 UV detector (Waters Inc., Milford, USA). Samples (20 μL) were separated at room temperature on an Elite Spherisorb C18 (5 μm, 250 mm × 4.6 mm I.D., Elite Analytical Instruments Co., Ltd., Dalian, China) using a solvent of oxalic acid:water (1:1000) at a flow rate of 1.0 mL/min. The amount of AA in each sample was calculated from the absorbance at 243 nm, using authentic AA (Sigma–Aldrich, Shanghai, China) as a standard. Results were expressed as milligrams per gram FW.

2.2.4. Anthocyanin

Anthocyanin contents were determined according to a published protocol with minor modifications (Mita, Murano, Akaike, & Nakamura, 1997). Frozen and dried samples (100 mg) were extracted at 4 °C in 5 mL hydrochloric acid:water (1:99, v/v) in methanol for 24 h. The mixture was centrifuged at 10,500 × *g* for 10 min and the absorbance of the supernatant was measured at 530 and 657 nm. Relative anthocyanin concentrations were calculated with the formula $[A_{530} - (1/4 \times A_{657})]$. The relative anthocyanin amount was defined as the product of relative anthocyanin concentration and extraction solution volume. The results were expressed as absorbance units $[A_{530} - (1/4 \times A_{657})]$ per gram dry weight (DW).

2.2.5. Total phenolics

Methanolic extraction for analysis of total phenolics and antioxidant capacity were basically performed as previous reported (Volden, Bengtsson, & Wicklund, 2009). Freeze-dried samples (200 mg) were extracted with 15 mL cold methanol and placed in an orbital shaker overnight at 4 °C. The suspension was centrifuged at 31,000 × *g* for 10 min at 4 °C, and the residue was extracted again using the same method. The supernatants were combined for detection of total phenolics and antioxidant activity. Total phenolics were determined using Folin–Ciocalteu's reagent (Sigma–Aldrich, Shanghai, China) by reading the absorbance at 760 nm. Gallic acid