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Investigation of bitterness in carrots (*Daucus carota* L.) based on quantitative chemical and sensory analyses

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Abstract

Bitterness is considered as an undesirable taste of carrots. Quantitative chemical analysis of potential bitter compounds of different carrot genotypes was combined with sensory analysis in order to identify key compounds likely to be responsible for the bitterness of carrots. Eight carrot genotypes ('Bolero', 'Mello Yello', 'Nairobi', 'Tornado', 'Purple Haze', 'Line 1', 'Line 2', and 'Line 3') representing extremes in sensory-perceived odour, flavour, and taste. Potential bitter compounds like polyacetylenes, isocoumarins and phenolic acids were quantified in the peel and the corresponding peeled carrot, and their contribution to bitterness in raw carrots was analysed by sensory profiling using multivariate data analysis. Falcarindiol and a di-caffeic acid derivative were highly related to bitterness in contrast to falcarinol and other potential bitter compounds. Falcarindiol and the di-caffeic acid derivative were primarily present in the peel whereas falcarinol was almost evenly distributed in the root. Investigation of bitterness revealed that high sugar content to some extent could mask the bitter perception of carrots. As falcarinol is the most bioactive of the carrot polyacetylenes the results of the present study indicate that there is a basis for improving the health effects of raw carrots without affecting sensory quality.

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Keywords: Daucus carota L.; Bitterness; Polyacetylenes; 6-Methoxymellein; Phenolic acids; Sensory profiling

1. Introduction

Bitter taste in vegetables often causes consumer rejection and is one of the main reasons for low preference scores in sensory evaluations of carrots (Kuusi & Virtanen, 1979; Alasalvar, Grigor, Zhang, Quantick, & Shahidi, 2001). Bitter and turpentine-like tastes in carrots are well documented (Sondheimer, 1957; Simon & Peterson, 1979; Simon, Peterson, & Lindsay, 1980a, 1982; Sejlåsen, Hoftun, & Bengtsson, 2001; Czepa & Hofmann, 2003). Carrots are able to produce sporadic bitterness when exposed to stress during growing, harvesting, transportation, storage, and processing (Lafuente, López-Galvez, Cantwell, & Yang, 1996; Sejlåsen et al., 2001; Talcott, Howard, & Brenes, 2001). During handling and distribution of carrots from field to consumer other less desirable odour and taste properties occur e.g. "harsh flavour". This sensory characteristic was first introduced by Simon et al. (1980a) to describe a strong burning turpentine-like flavour most clearly perceived at the back of the throat during and after chewing. "Harsh flavour" seems to be due to high contents of mono- and sesquiterpenes and low contents of sugars (Simon, Peterson, & Lindsay, 1980b).

6-Methoxymellein (3-methyl-6-methoxy-8-hydroxy-3,4dihydro-isocoumarin; 6-MM, Fig. 1) was the first bitter compound, identified in carrots (Sondheimer, 1957). Since then additional studies have suggested that bitterness in carrots is due to a multiplicity of several bitter compounds including phenolic acids, 2,4,5-trimethoxy benzaldehyde and polyacetylenes (Starkovsky, 1962; Phan, 1974; Hermann, 1990; Sejlåsen et al., 2001; Alasalvar et al., 2001; Czepa & Hofmann, 2003, 2004). The main phenolic acid, chlorogenic acid (5-caffeoylquinic acid, 5-CQA), can constitute up to 60% of the total phenolic acid content in carrots and has previously been described as having a mild bitter taste (Alasalvar et al., 2001). However, the contribution of 5-CQA and other phenolic acids to bitter taste in raw carrots is still unclear. Falcarinol [(Z)-heptadeca-1,9dien-4,6-divn-3-ol; FaOH], falcarindiol [(Z)-heptadeca-1,9-

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Falcarindiol (FaDOH), R=OH
Falcarindiol 3-acetate (FaDOAc), R =OAc

6-Methoxymellein (6-MM)

Fig. 1. Potential bitter and health-promoting compounds in carrots belonging to a group of polyacetylenes (FaOH, FaDOH and FaDOAc), isocoumarins (6-MM) and phenolic acids (*p*-coumaric acid, caffeic acid, ferulic acid and quinic acid).

dien-4,6-diyn-3,8-diol; FaDOH] and falcarindiol 3-acetate [(Z)-3-acetoxy-heptadeca-1,9-dien-4,6-diyn-8-ol; FaDOAc] (Fig. 1) are the main polyacetylenes in carrots (Czepa & Hofmann, 2003, 2004; Kidmose et al., 2004). Sensoryguided chromatographic fractionation has shown that these polyacetylenes have a major impact on bitter taste in carrots, and that the content of FaDOH might offer a new analytical measure for carrot quality (Czepa & Hofmann, 2003, 2004). However, a direct relationship between sensory-perceived bitterness of raw carrots and polyacetylenes has not been elucidated.

The bitter compounds may explain some of the health promoting effects of carrots, especially their anti-cancer effect (Hansen, Purup, & Christensen, 2003; Brandt et al., 2004; Kobæk-Larsen, Christensen, Vach, Ritskes-Hoitinga, & Brandt, 2005; Christensen & Brandt, 2006). In this context the polyacetylenes appear to be very promising with FaOH being the most bioactive of the carrot polyacetylenes (Hansen et al., 2003; Kobæk-Larsen et al., 2005; Zidorn et al., 2005; Christensen & Brandt, 2006). FaOH has shown pronounced cytotoxic activity against

human tumor cell lines *in vitro*, in concentrations down to 0.03 μg/ml and pre-clinical trials have demonstrated that FaOH also has *in vivo* anti-cancer activity (Matsunaga et al., 1990; Brandt et al., 2004; Kobæk-Larsen et al., 2005). Therefore polyacetylenes, and in particular falcarinol, seems to be a potential contributor to the anti-cancer effect of carrots, and perhaps also other health promoting effects (Brandt et al., 2004; Kobæk-Larsen et al., 2005; Christensen & Brandt, 2006). Consequently, there might be a correlation between the health promoting effects and bitterness of carrots.

Carrots are eaten raw or processed into a wide variety of products, including, cooked, canned, frozen and dehydrated products. The concentration of bitter compounds in carrots depends on genotype, location, growing and storage conditions and preparation methods (Hansen et al., 2003; Kidmose et al., 2004). Further, it has been shown that peeling may reduce the content of FaDOH and 6-MM but not the total content of FaOH and FaDOAc (Czepa & Hofmann, 2004).

In order to improve the health promoting effects and sensory quality of raw carrots, more knowledge needs to be provided. Consequently, it is important to determine the contribution and/or relationship of potential bitter compounds to bitterness in carrots and to investigate the possibility of masking bitterness with sugar content. The objectives of the present study were first to determine the relationship between potential bitter compounds (polyacetylenes, isocoumarins and phenolic acids) and sensory-perceived bitterness in raw carrot roots defined by sensory profiling using multivariate data analysis. A second aim was to determine the distribution of potential bitter compounds in the peel and the peeled roots of raw carrot as peeling of carrots may offer a possibility to change the bitter compound profile in carrot root products.

2. Materials and methods

2.1. Chemicals and standards

Trifluoroacetic acid (TFA) (99%) was obtained from Sigma-Aldrich (Steinheim, Germany). Acetonitrile (MeCN), methanol (MeOH), ethyl acetate (EtOAc), and formic acid were of Ratborn high-performance liquid chromatography (HPLC) grade (99.9% HPLC grade) obtained from Sigma-Aldrich. Sodium hydroxide (NaOH) was obtained from JT Baker (Deventer, The Netherlands) and sodium sulphate (Na₂SO₄) was obtained from Merck (Darmstadt, Germany). The water used for HPLC analysis was ultra pure generated by Elgastat Maxima Analytica Water Purification System (Elga Ltd., UK). All eluents for HPLC analysis were filtered through a 0.45 µm Minisart SRP 25 filter (Bie & Berntsen, Rødovre, Denmark) and degassed with ultrasound for 20 min before use.

Authentic standards of the polyacetylenes falcarinol (FaOH), falcarindiol (FaDOH) and falcarindiol 3-acetate (FaDOAc) and the isocoumarin 6-methoxymellein (6-MM)

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