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Characterisation of cell wall polysaccharide profiles of apricots (*Prunus armeniaca* L.), peaches (*Prunus persica* L.), and pumpkins (*Cucurbita* sp.) for the evaluation of fruit product authenticity

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Abstract

Cell wall polysaccharides were investigated for their suitability as markers for quality and authenticity control of fruit products. For this purpose, the alcohol-insoluble residue (AIR) from several cultivars of apricots and peaches of different harvest seasons, provenances, and stages of ripeness was extracted and subsequently fractionated into acid- and EDTA/alkali-soluble pectins, hemicellulose, and cellulose. Each fraction was analysed for its neutral sugar composition by gas chromatography. In addition, analyses were also carried out on several cultivars of pumpkins because of their potential for use in fraudulent admixtures. Within the respective fruit species, characteristic neutral sugar profiles of the AIR and its fractions were observed, which were found to be independent of the cultivar, harvest season, and provenance. The fruit specific saccharide composition may be used for the differentiation of fruit products devoid of carbohydrate-based hydrocolloids. Furthermore, the isolated hemicellulose may also allow the detection of admixtures of non-specified fruit in complex fruit products, such as jams, spreads, and fruit preparations.
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1. Introduction

Apricots (*Prunus armeniaca* L.) and peaches (*Prunus persica* L.) are fruits of high economic relevance. Adulterations of apricot-based products with cheaper fruits or vegetables like peaches and pumpkins, resulting in deterioration of product quality, are difficult to detect. In order to protect the consumer as well to as avoid unfair competition it is absolutely essential to check the composition of a food at all levels of the production process, from the raw material to the finished product in terms of authenticity and quality. The various strategies for the detection of fraudulent admixtures to fruit products have recently been reviewed (Fügel, Carle, & Schieber, 2005; Reid, O'Donnell, & Downey, 2006). Numerous former studies focussed on the presence of characteristic phenolic compounds for the determination and differentiation of plant species (Alonso-Salces et al., 2004; Zimmermann & Galensa, 2007).

However, taking the phenolic composition as a parameter of food authenticity is not without problems and limitations. For example, the admixture of apricots to peach jam can easily be detected via the phenolic profile, whereas the detection of the more likely adulteration of the usually more expensive apricot jam by addition of peaches is impossible (García-Viguera, Ferreres, Tomás-Barberán, Gil, & Tomás-Lorente, 1992; Tomás-Lorente, García-Viguera, Ferreres, & Tomás-Barberán, 1992). Recent advances in food analytical chemistry, in particular the application of hyphenated techniques such as LC–MS and LC-NMR, have also demonstrated that some phenolic

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compounds like phloridzin and isorhamnetin glycosides previously considered typical markers of apples and pears, respectively, may also be present in other genera (Hilt et al., 2003; Schieber, Keller, Streker, Klaiber, & Carle, 2002). Furthermore, several studies have shown that the phenolic composition undergoes qualitative and quantitative variations with different cultivars, stage of maturity, geographical origins and storage conditions (Lee, Kagan, Jaworski, & Brown, 1990; Lee, Park, & Choi, 2001; Senter, Robertson, & Meredith, 1989; Veberic & Stampar, 2005). In addition, owing to their susceptibility towards heat and oxygen, some polyphenolics may undergo degradation during industrial processing. Indisputable authentication of food products implies the need of highly sophisticated and cost-intensive analytical methods like NMR spectroscopy and IRMS (Pfammatter, Maury, & Théthaz, 2004; Schmidt, Roßmann, Stöckigt, & Christoph, 2005). DNA-based technologies using PCR technique are highly sensitive but inapplicable to heated, acidic products, since the DNA has been shown to degrade under these conditions (Adam & Zimm, 1977; Bauer, Weller, Hammes, & Hertel, 2003; Hupfer, Hotzel, Sachse, & Engel, 1998; Meyer, Rosa, Hischenhuber, & Meyer, 2001).

Studies on plant cell wall polysaccharides carried out so far aimed at their physiological function as a dietary fibre and their structure and organisation in fruits and vegetables. In terms of food quality, the main attention has been paid to cell wall changes during ripening in order to optimise textural attributes and cell wall-dependent quality characteristics (Waldron, Parker, & Smith, 2003). However, studies on the characterisation of the neutral sugar composition of the major cell wall constituents, pectin, hemicellulose, and cellulose, particularly with respect to authenticity control are scarce. Recently, an innovative approach has been reported for strawberries and cherries (Fügel, Carle, & Schieber, 2004). Therefore, the objective of the present study was to evaluate the possibility of differentiation of fruit species belonging to the same genus on the basis of the isolated cell wall components and their neutral sugar composition. Due to their economical significance, the main focus was on the investigation of apricots and peaches, both representing the genus *Prunus*. Pumpkins, which are likely candidates to be used for fraudulent purposes because of their carotene-based colour, were also included in this study.

2. Materials and methods

2.1. Plant material

Fresh apricots (*Prunus armeniaca* L.) and peaches (*Prunus persica* L.) of different cultivars and provenances and yellow-fleshed pumpkins (*Cucurbita* sp.) were obtained from the local market (Stuttgart, Germany) (Table 1). IQF fruits (individually quick frozen after lye peeling (peaches) and blanching) were provided by Wild (Eppelheim/Heidelberg, Germany). Fresh apricots and peaches were (lye) peeled and manually cored. An aliquot of selected fruit varieties remained unpeeled to determine the influence of (lye) peeling on cell wall polysaccharide composition. The pumpkins were manually peeled, cored, blanched at 85 °C for 10 min, and subsequently mashed through a sieve (mesh size: 1.5 mm). Fruits were harvested in 2003, 2004, 2005, and 2006. The fruits of 2003 and 2004 were stored, lyophilised in a Steris Lyovac® GT 4 Lyophilizer (Steris, Hürth, Germany) and frozen at –20 °C, respectively, to avoid enzymatic degradation of the cell wall material.

2.2. Alcohol-insoluble residue (AIR)

Apricots, peaches, and pumpkin purées were freeze-dried and ground with liquid nitrogen to a fine powder in a cutter (Stephan und Söhne & Co., Hameln, Germany). The lyophilisate (25 g) was homogenised in boiling aqueous ethanol (300 mL, 80%, v/v) using an Ultra-Turrax blender. After boiling for 1 h, the insoluble solids were collected on a Büchner funnel. Ethanol extraction was repeated until a clear extract was obtained. The AIR was stirred overnight in pure acetone, passed through a G1 glass sinter filter and air-dried at 50 °C for 24 h. The dried AIR was ball-milled (Retsch, Haan, Germany), sealed in lever lid glass bottles and kept in a desiccator until further analysis.

2.3. Sequential extraction of the AIR

The sequential extraction was based on a method described previously (Fügel, Förch, Carle, & Schieber, 2005) and optimised for apricots and peaches by an additional extraction step with hydrochloric acid. The AIR (800 mg) was suspended in 50 mL of diluted hydrochloric acid (0.05 M) and stirred at 60 °C for 1 h. After centrifugation at 15,000*g* for 20 min, the pellet was washed twice with 50 mL of distilled water. The supernatants were pooled, dialysed exhaustively against distilled water for 48 h using dialytic membranes (type 36/32, pore size 25–50 Å, Roth, Karlsruhe, Germany). Subsequently, the HCl-soluble pectin (HSP) fraction was freeze-dried. For further extraction of the residue, alkaline EDTA solution (0.05 NaOH; 0.5 mM EDTA) was used at 30 °C for 1.5 h. The suspension was centrifuged for 20 min at 15,000*g* and the remaining pellet washed twice with distilled water. The supernatants were pooled, adjusted to pH 6.5–7.0 with HCl, dialysed for 48 h against distilled water and freeze-dried to obtain the NaOH/EDTA-soluble pectin (OHEP) fraction. The final extraction was carried out using 50 mL of aqueous sodium hydroxide solution (16%, w/w) for 5 h at 30 °C. After centrifugation at 15,000*g* for 20 min, the pellet was rinsed twice with distilled water. The supernatants were pooled and adjusted to pH 6.5–7.0 with HCl, followed by the procedure described for the previous fraction to yield the hemicellulose (HC) fraction. The remaining pellet consisting of insoluble solids such as lignin and cellulose (C fraction) was suspended in 50 mL of distilled water, dialysed and lyophilised.

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