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# Seasonal trends of starch and soluble carbohydrates in fruits and leaves of 'Abbé Fétel' pear trees and their relationship to fruit quality parameters

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#### ABSTRACT

Plants mainly store carbohydrates, a product of photosynthesis, as starch and the amount of distribution of starch differs between organs and species. The objective of this research was to study changes in carbohydrate reserves, mainly starch, in the fruits and leaves of 'Abbé Fétel' pear trees at different physiological stages during the 2012–2013 growing seasons. Trees were trained as a spindle system and grafted on two rootstocks, Sydo<sup>®</sup> and Quince C. The evolution of starch degradation in fruit was correlated with the fruit quality parameters, soluble sugars and organic acids. Starch in fruit started accumulating early, 50 days after full bloom (DAFB), and reached a maximum concentration during the middle phase of fruit development, 110 DAFB, several weeks before harvest. As starch degradation began, a constant increase of soluble carbohydrates occurred. The two rootstocks did not induce significant differences in starch concentrations, but were different in their amounts of soluble carbohydrates. Further, the starch degradation in fruits was highly related to fruit weight, soluble solids content, flesh firmness and Index of Absorbance Difference. Among soluble sugars and organic acids, glucose, fructose, sucrose and quinic acid were related to starch hydrolysis during fruit maturation. Understanding the changes in carbohydrate creation and degradation among vegetative and reproductive organs could lead to optimization of yield efficiency and production.

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# 1. Introduction

Early stages of reproductive development are highly dependent on current photosynthetic carbohydrates availability in fruitbearing evergreen trees, such as citrus and avocado (Finazzo et al., 1994; Scholefield et al., 1985). A different behavior characterizes deciduous tree species where initial growth and development of vegetative and reproductive organs is fueled by carbohydrates from stored reserves (Flore and Layne, 1999; Loescher et al., 1990). This contribution of stored carbohydrates to flowering and fruit development is partially dependent on the timing of season (Loescher et al., 1990). Therefore, the knowledge of phenological phases (vegetative and reproductive) development is crucial to

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http://dx.doi.org/10.1016/j.scienta.2016.08.008 0304-4238/© 2016 Elsevier B.V. All rights reserved. understand dynamics of carbohydrate reserves on fruit trees. In species that initiate shoot growth before anthesis, such as grapevines and kiwi, reserve mobilization is important for both shoot growth and flowering; until leaves produce enough assimilates to meet sink demand (Flore and Layne, 1999). A mechanism similar to the grape model has been postulated for apple because apple leaves are almost fully expanded before anthesis and main fruit development occurs after leaves fully develop (Loescher et al., 1990).

The life phases of the tree are mirrored by changes in sourcesink relationships like sink organs competing for a common pool of carbohydrates (Roitsch, 1999). All tree organs at some stage of their development act as sinks, i.e. consumers of assimilate. In terms of assimilate transport, the ability of a sink organ to import assimilates is known as sink strength (Ho, 1988). Sink strength of individual organs varies with the seasonal development pattern of the tree and age of the plant (Flore and Layne, 1999). The sink strength is a function of the ratio among endogenous hormones (Hansen, 1971).







Tree organs have an order of priority based on the strength of different sinks: seeds are the strongest followed by fruit, shoot apex, and leaves. The intermediate strength sinks are represented by cambium and root system and the weakest strength is the reserves accumulation (Cannell, 1985).

Actively growing pome fruit are supplied by the C-assimilates sorbitol and sucrose that are produced by leaf photosynthesis (Berüter et al., 1997). These C-sources are inputs for fruit metabolism and accumulate as fructose, sucrose, malic acid, and starch (Berüter et al., 1997; Berüter, 2004; Dugalic et al., 2014; Hudina and Štampar, 1999). Starch is synthesized in apple fruit up to 100–110 DAFB then its degradation begins. Starch hydrolysis produces glucose and fructose, important player in fruit ripening as substrate of respiration (Osorio and Fernie, 2014; Doerflinger et al., 2015). The process of hydrolysis is a determinant for the final internal fruit quality for measures like soluble sugars content (Berüter, 2004; Wegrzyn and McRae, 1995).

As the growing season proceeds, there are changes in metabolites in both source and sink organs that influence fruit quality (Berüter et al., 1997). Malic and citric acids are major products of the metabolism during fruit growth in apples and pears, which decrease at the maturity stage (Berüter, 2004; Hudina and Štampar, 1999). In pear fruit, the composition of soluble sugars and organic acids determines main characteristics like sweetness and sourness (Eccher Zerbini, 2002). Therefore, levels of soluble sugars and organic acids are important factors in determining the sensory quality of ripe fruit (Ackermann et al., 1992; Dugalic et al., 2014; Hudina and Štampar, 1999).

Physiological ripeness of fruit is assessed with maturity indices (Brookfield et al., 1997). Maturity at harvest is crucial for subsequent storage management, final consumers' acceptance (Kader, 2002), and it is determined by the exterior (shape, size, and appearance) and interior (taste and texture) characteristics of the fruit (Dewulf et al., 1999). Several fruit quality parameters such as soluble solids content (SSC), titratable acidity (TA), ratio of soluble solids to titratable acidity (SSC/TA), skin color, and flesh firmness (FF) are also utilized to choose the optimal harvest time. In apples, starch conversion into sugars is one of the most important indicators to predict optimal harvest dates (Brookfield et al., 1997; Peirs et al., 2002). For this purpose, the Starch Pattern Index (SPI) uses an iodine solution that provides a visual pattern for estimation, which uses a standard starch conversion chart, of the total starch content and degradation. The main disadvantage of this technique is the lack of a quantitative values for starch (Peirs et al., 2002). Conversely, in pear fruit the starch index is less employed even if some studies have reported the use of this procedure (Agar et al., 1999; Le Lezec and Belouín, 1994; Stow, 1988). However, Stow (1988) described this technique as an unreliable method to determine the optimum harvesting period of pear fruits.

Seasonal carbohydrates dynamics in different tissues and organs are well understood in many species such as apple (Hansen, 1971; Berüter et al., 1997; Brookfield et al., 1997), avocado (Scholefield et al., 1985; Whiley et al., 1996a,b), kiwi (Boldingh et al., 2000; Miller et al., 1998; Moscatello et al., 2011; Richardson et al., 1997), pistachio (Elloumi et al., 2014), and sweet cherry (Keller and Loescher, 1989) because many cultural practices that are common techniques for tree fruit production alter carbohydrates allocation dynamics. Nevertheless, information about European pear cultivation is scarce and demands of new markets, low yields and the ageing of existent orchards has triggered the need of orchard modernization for pear growers. Recently, innovations in the pear industry have shifted from low planting density orchards with vigorous rootstocks to high density planting  $(10-13,000 \text{ trees ha}^{-1})$ with quince rootstocks. This allows control of tree size, promote earlier bearing, and improve fruit quality (Musacchi, 2008). In Italy,

more than 90% of orchards are grafted on quince (*Cydonia oblonga*) rootstocks (Musacchi, 2011).

To better understand the effect of these innovations on pear quality, the present work determines the dynamics of accumulation and degradation of starch and soluble carbohydrates concentrations during the growing season, in fruits and leaves of 'Abbé Fétel' pear trees, trained as spindle systems and grafted on Sydo<sup>®</sup> and Quince C rootstocks. Additionally, the starch degradation evolution in fruit was evaluated to correlate it with fruit quality parameters, soluble sugars and organic acid content.

## 2. Materials and methods

#### 2.1. Plant material and sampling

The trial was carried out throughout two growing seasons (2012–2013) on seventeen-year-old pear trees (*Pyrus communis* L.) cv. 'Abbé Fétel', trained as a Spindle and grafted on two root-stocks: Sydo<sup>®</sup> (medium vigour) and Quince C (dwarfing). Planting distance was  $3.6 \text{ m} \times 1.4 \text{ m}$  for Sydo<sup>®</sup> and  $3.6 \text{ m} \times 0.7 \text{ m}$  for Quince C. The orchard (North-South oriented) was located at the Experimental Station of the Bologna Agriculture Faculty, in Cadriano, Italy (44°54′88.53″s; 11°38′59.30′′W). The orchard was managed following standard cultural practices (i.e. fertigation, disease and pest control). Homogeneous trees in size and vigour were selected within the orchard for the experimental trial on the basis of the Trunk Section-Cross Area (TSCA, cm<sup>2</sup>).

# 2.2. Fruit and leaf sampling

Timing of sample collections during the growing season were defined by days after full bloom (DAFB); a date registered each year. The full bloom, when 100% of the flowers were open, took place on March 30, 2012 and April 17, 2013. At each sampling date, 24 fruits held by 3-year-old branches in the middle layer of the tree (between 1.0–1.5 m from the ground) and from the external part of the canopy were collected randomly per treatment (2 rootstocks). Half of the fruit collected were used to determine the soluble carbohydrates and starch concentration, soluble sugars and organic acids, whereas the remaining 12 fruits were measured for fruit quality parameters. In addition, the leaves close to sampled fruit were collected at each sampling date.

Each sampling was done early in the morning, and the vegetal material was rapidly transferred inside a cooler and taken to the laboratory for analyses. Samples were collected at 97, 115, 131, 152, 171 and 195 DAFB in 2012, and at 51, 63, 79, 93, 107, 127 and 142 DAFB in 2013. The commercial harvest took place at 152 and 142 DAFB, respectively.

Fresh fruit weight was determined for 12 fruits per treatment from each sampling date. Average fresh weight (FW) was calculated for all leaves collected. Fruit and leaf dry weights (DW) were determined on subsamples placed in a freeze dryer for 7 and 3 days, respectively (HETO dry winner, DW3, Denmark).

## 2.3. Starch and soluble carbohydrates determinations

#### 2.3.1. Preparation of material

From each of the 12 fruits, one 2-mm-thick equatorial slice was excised and collected (3 biological replications with 4 pooled fruits per replication). Three samples of 50 leaves each were assembled from the leaves collected per sampling date. Fruit flesh and leaf samples were immediately frozen in liquid nitrogen and stored at -80 °C. Frozen samples were dried in a freeze dryer, reweighed and ground by a mill until a fine powder, and finally stored in airtight containers at room temperature. Three replications of 50 mg (DW)

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