



Full Length Article

Characterisation of yacon tuberous roots and leaves by DART-TOF/MS



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ABSTRACT

Yacon [*Smallanthus sonchifolius* (Poepp. et Endl.) H. Robinson] is a plant grown worldwide originating in the Andes region. Yacon is grown for its sweet tuberous roots and leaves used for the preparation of herbal infusions. Twenty-six yacon landraces' leaves and roots (both peeled and unpeeled) have been analysed by DART-TOF/MS. The method has been optimised and the fingerprints of the mass spectra have been statistically processed by PCA and LDA statistical analysis. The DART method has succeeded in differentiating between the yacon landraces according to their genotype and geographical origin.

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1. Introduction

Yacon [*Smallanthus sonchifolius* (Poepp. et Endl.) H. Robinson] from the *Asteraceae* botanical family is a native plant of the Andes region, where it is cultivated for its sweet tuberous roots. The roots contain fructooligosaccharides (FOS) and inulin, which are best-known as sources of prebiotics with a positive effect on human health [1,2]. The yacon leaves and tuberous roots are rich in polyphenols [3]. The polyphenolic component of the yacon leaves have a strong antioxidant effect; yacon leaves can, therefore, be used in the human diet to prevent chronic diseases [4]. Yacon is eaten raw as a fruit and is prized for its sweetness, mainly by children. In the Andean region, yacon is cultivated from Colombia to Argentina, mostly in small areas and for the farmer's own consumption. Peru, is the country with the largest biodiversity of yacon. In Peru, more than 200 accessions have been found, while in Bolivia and Ecuador, 40 and 32 accessions have been observed, respectively [5]. Due to its attributes, yacon has expanded to other regions outside the Andes. Currently, it is grown in Brazil, the Czech Republic, China, Japan, New Zealand, the Philippines, Russia, South Korea,

the U.S.A and other countries [6–10]. The Czech Republic has 26 accessions; it is the largest collection outside of the Andean region. There is huge morphological, chemical and molecular variability in yacon [11]. The yacon tuberous root flesh colour can be white, cream, white with purple striations, purple, pink or yellow. Their skin can be brown, pink, purple, cream, or ivory white, and contains resin tubes filled with yellow crystals. These properties are typical for all genotypes [12,13]. The total saccharide content in the yacon tuberous roots (80.62–224.7 g/kg dm.) [1], the FOS content (2.1–70.8 g/100 g dm.) [14] and the content of the total phenols in the leaves and tuberous roots are influenced by the genotype [3,15].

DART (Direct Analysis in Real Time) is a novel ionisation technique of mass spectrometry. DART principles and advantages have been well described in literature [16–18]. This technique is used for the inspection of food and raw food materials. Possible applications of the DART-TOF/MS technique are summarised in literature [16,19]. The aim of this work was to characterise the tuberous roots and leaves of 26 yacon accessions with the DART-TOF/MS technique.

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Table 1
Description of the yacon accessions.

Codes	Qualitative characteristics			Year of introduction to the Czech Republic	Chromosome number (2n)
	Colour of root skin	Colour of root flesh	Colour of leaves		
PER01	greyish orange	yellow white	greenish yellow	2005	58
PER02	greyish orange	white	greenish yellow	2005	58
PER03	purplish red	orange yellow	dark green	2005	58
PER04	purplish grey	yellow white	greenish yellow	2005	58
PER05	purplish red	yellow white	dark green	2005	87
PER06	purplish grey	white	dark green	2005	58
PER07	greyish orange	yellow white	greenish yellow	2005	58
PER08	purplish grey	white	greenish yellow	2005	58
PER09	greyish orange	orange yellow	dark green	2005	58
PER10	white	orange yellow	dark green	2005	58
PER11	greyish orange	yellow white	greenish yellow	2005	87
PER12	purplish grey	white	dark green	2005	87
PER13	white	white	greenish yellow	2005	87
PER14	greyish orange	yellow white	dark green	2005	87
PER15	greyish orange	yellow white	greenish yellow	2008	^a
BOL20	purplish red	yellow white	dark green	1995	58
BOL21	greyish orange	yellow white	greenish yellow	2007	58
BOL22	purplish red	yellow white	greenish yellow	2007	58
BOL23	purplish grey	white with purple red strains	dark green	2007	58
BOL24	greyish orange	yellow	greenish yellow	2007	58
ECU40	purplish red	yellow white	dark green	1994	58
NZL51	white	orange yellow	greenish yellow	1993	58
NZL52	purplish grey	white	dark green	1993	58
NZL53	purplish grey	orange	dark green	2007	116
NZL54	purplish grey	orange	dark green	2007	116
GER30	purplish grey	yellow white	greenish yellow	1994	58

^a Not measured.

2. Materials and methods

2.1. Plant material

For this study, 26 yacon accessions cultivated in the climatic conditions of the Czech Republic were analysed. The colour of the leaves and tuberous roots-root skin and flesh (assessed a year after the introduction to the Czech Republic), along with a chromosome number for each of the accessions, are shown in Table 1. The yacon accessions came from different climatic regions: Bolivia (BOL 20, BOL 21, BOL 22, BOL 23, and BOL 24), Ecuador (ECU 40), Germany (GER 30), Peru (PER 01, PER 02, PER 03, PER 04, PER 05, PER 06, PER 07, PER 08, PER 09, PER 10, PER 11, PER 12, PER 13, PER 14, and PER 15), and New Zealand (NZL 51, NZL 52). Accessions NZL 53 and NZL 54 were obtained by *in vitro* induced polyploidisation from accession NZL 51 [20]. The yacon collection is kept by the Faculty of Tropical AgriScience, the Czech University of Life Sciences Prague. The plant material was cultivated in field conditions on experimental plots of the Faculty of Tropical AgriSciences, the Czech University of Life Sciences Prague. The plots lie at an average altitude of 286 m above sea level (50°04' north latitude and 14°26' east longitude), and have loamy soils. The yacon leaves and tuberous roots were harvested in October 2014 after 146 and 157 days of cultivation, respectively. The samples (leaves and tuberous roots) for the analyses were taken randomly from 2 to 4 plants of each clone.

The leaves were removed from the fourth top node of the main stem of each plant. The weight and size of the tuberous roots used in the analyses fluctuated between 100 and 300 g and 12–20 cm, respectively, depending on which landraces were used.

2.2. DART/TOF-MS method

The parameters of the DART ion source for the measurements were as follows: a positive ionisation mode, a grid voltage of 350 V, an autosampler velocity of 1 mm/s, and an ionisation gas temper-

ature of 300 °C. A DART SVP 100 ion source (IonSense, Saugus, MA, USA) was used. DART consists of a DART controller and a total ionisation source. For the measurements, helium gas (SIAD, Prague, Czech Republic; purity: 4.8) was used, whereas nitrogen gas was used in the standby mode. Nitrogen was generated by a nitrogen generator (Peak Scientific, NM32LA at an operating pressure of 6.0 bar) and purified by an RMSN-4 Agilent universal trap. The pressure of helium and nitrogen on the default input of the DART source was set at 5.5 bar using reduction valves. The DART source was coupled with a TOF-MS by a vacuum interface (IonSense). At the interface, an 83 mm long and a 3.18 mm diameter ceramic tube (for the DART-100 ion source) was used. Evacuation of He from the space of the interface (to prevent a low vacuum being indicated by the TOF-MS measurement accessories calibrated for nitrogen) was performed using an MZ 2NT Vacuum Pump (Vacuubrand, Wertheim, Germany). Samples were analysed using a 12 DIP-it[®] autosampler. For the measurements, DIP-it (IonSense, Saugus, MA, USA) sampling rods were used. The DART ion source can operate at temperatures between 50 and 500 °C. The velocity of the autosampler movement ranges from 0.2 to 10.0 mm/s. The default flow of the reaction gas (helium) was set at 3.5 l/min. The DART source was controlled by the DART – SVP 3.0.3b software (IonSense, Saugus, MA, USA). The mass spectrometer was operated in both positive and negative ion modes; the fragmentor voltage was 175 V and the skimmer voltage was 65 V. For data acquisition and processing, an Agilent MassHunter Workstation Acquisition B.04.00 (Agilent Technologies, Santa Clara, USA) and an Agilent MassHunter Workstation Software Qualitative B.04.00 were used. For mass spectral studies, the total ion current (TIC) chromatogram was registered in the range of *m/z* 100–1500. A TOF LC/MS 6224 mass detector (Agilent Technologies, Santa Clara, USA) was used. A rough vacuum in the TOF-MS was created by an Edwards E2M28 vacuum pump (Grawley, West Sussex). Tuning of TOF-MS was carried out before each set of samples using an API-TOF Reference Mass Solution Kit (Agilent Technologies, Santa Clara, USA).

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