Maximising fructo-oligosacharide production in yacon

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Abstract
There is renewed interest around the world in fructo-oligosaccharide and herbal tea production from the storage roots and leaves and stems, respectively, of the South American vegetable, yacon (Smallanthus sonchifolius). Little agronomic information is available on how best to grow yacon for these products and a programme of research has begun in New Zealand to define its agronomic requirements. Results are presented from three field trials conducted at Pukekohe Research Station. Information was collected on the yield and carbohydrate and fructan components of the storage roots of four yacon lines compared to Jerusalem artichoke (Helianthus tuberosus). The yacon lines varied in storage-root yield from 71 to 96 t/ha with the fructan level varying from 240 to 320 mg/g DM, but with no significant differences in the total carbohydrate content (565 mg/g DM). Jerusalem artichoke produced 36 % less tubers and 21 % less fructans than yacon storage roots, but it had a similar total carbohydrate content. The yield and fructan content of Jerusalem artichoke was similar to the lowest yacon lines. Thin layer chromatography showed that the majority of the fructans in the Jerusalem artichoke had a degree of polymerisation (DP) above 10 while the yacon fructans were predominant in the range 3-9. Two trials examined the effects of time of harvesting and removal of the top growth in autumn on yacon root production. The highest yield of yacon roots (96 t/ha) was obtained by harvesting the crop once the top growth had stopped growing in June, with earlier harvests (March, April, May) giving reduced yields. Removal of top growth in the autumn months before the plant had stopped growing also reduced subsequent root yield. Results were sufficiently encouraging to warrant the examination of yacon production on a commercial scale in an ongoing research programme.

Introduction
Yacon (Smallanthus sonchifolius syn. Polymnia sonchifolius - Asteraceae) is a tall, perennial herb from South America, related to sunflowers. It develops tuber-like storage roots in the range 100 to 500 g, but sometimes heavier than 1 kg (Grau, 1993). Traditionally these roots have been eaten raw or cooked as a sweet, juicy vegetable. However, there is emerging interest in using the roots as a source of fructo-oligosaccharides (FOS) and the herbage as a medicinal, herbal tea (National Research Council, 1989; Rea, 1994; Grau and Rea, 1997). This interest has contributed to the rapid growth of an industry in functional foods — products that improve health when consumed. FOS are classified as a prebiotic food that is indigestible in the upper alimentary canal but stimulates the growth and activity of beneficial bacteria in the lower-gut, improving health (Gibson and Roberfroid, 1995). In this paper FOS are defined as a fructan based carbohydrate with a degree of polymerisation (DP) in the range of 3 to 9. The digestibility of FOS decreases as chain length increases; shorter chains are more available as an energy source for the desirable lower-gut bifidobacteria than longer chain inulins (DP >10) (Farnworth, 1993; Gibson and Roberfroid, 1995; Kolida et al., 2002). FOS are manufactured by the enzymatic treatment of sucrose or obtained from crops that contain high levels of fructans, such as chicory (Cichorium intybus), Jerusalem artichoke (Helianthus tuberosus), burdock (Arctium lappa) or yacon (Suzuki, 1993). Both chicory and Jerusalem artichoke have been cultivated in Europe for the production of fructans with FOS being obtained by hydrolysis from the longer chain oligosaccharides (Kosaric et al., 1984; Baert and Bockstaele, 1993; Frese, 1993). In comparison, yacon is an undeveloped crop, but it has the advantage over chicory or Jerusalem artichoke that the sugars are naturally oligofructoses within the low DP range that give health benefits and can be
extracted without the need for hydrolysis (Grau and Rea, 1997). Dried yacon leaves are traditionally used in South America to prepare an antidiabetic tea. Recent laboratory animal experiments support this use (Grau and Rea, 1997; Aybar et al., 2001). Greater international interest is now being shown in the production of FOS and herbal tea from yacon, but production outside South America is still in its infancy and little published agronomic information is available.

Yacon is a native of the subtropical and warm-temperate environment of the Andean mountains, between the equator and the tropic of Capricorn (23°S) and within the altitudinal range of 600 to 3500 m (Grau and Rea, 1997). The herbage is frost sensitive with leaves damaged at -1°C, but the crop can be grown successfully in colder environments provided there is a 6-7 month frost-free growing period to allow the crop to mature. Yacon grows best on fertile, free draining soils but is a crop with large leaves and a high transpiration capacity. Consequently, it requires adequate water for good production. The crop is usually established by planting crown-pieces containing dormant buds (turions) because the storage roots have no buds so cannot produce shoots (Grau, 1993). In South America, crops are planted from September to November in furrows with row widths varying from 70 to 100 cm and plant spacing varying from 60 to 140 cm with fertiliser and irrigation, as appropriate for the soil conditions (Grau and Rea, 1997).

Yacon was introduced into New Zealand from Ecuador in the early 1980s (Endt, 1983) as a new exotic vegetable, and although some initial commercial production took place (Grau and Halloy, 1994) it did not become an established crop. Two further clones of yacon from north-western Argentina were introduced into New Zealand in 1994 (Martin et al., 1997). The early commercial production of yacon grown as a vegetable on Great Barrier Island (latitude 36°S) and experimental research in the South Island (latitude 45°S) showed that yacon can successfully grow to produce tuber-like roots over a wide range of latitudes in New Zealand (Grau, 1993; Grau and Halloy, 1994; Martin et al., 1997). Nevertheless, agronomic information is lacking on how best to grow yacon to optimise crop production and quality for the production of FOS and herbage, or both. The trials reported here are part of a wider programme being undertaken to develop an understanding of the agronomic requirements of yacon in New Zealand. Jerusalem artichoke was included in one trial to provide a comparative assessment with other crops used for FOS production.

**Materials and Methods**

Two yacon accessions (Y42, Y43) were collected from north-western Argentina in 1993 by Dr Alfredo Grau as part of the MAF Technology new crops programme (Martin et al., 1997). Two further lines originated from the introductions from Equador by Landsendt Nursery, Oratia, Auckland in the early 1980s (Endt, 1983), and were obtained from the Landsendt Nursery (accession name - Landsendt) and a private garden in the Waikato (accession name - Ohaupo).

Three field trials were conducted at Pukekohe Research Station on a Patumahoe clay loam where the plants were grown on ridges formed by potato-ridging equipment. Prior to planting, the trial areas were fertilised with 1 t/ha 30 % potassic superphosphate.

**Trial 1: preliminary trial comparison with Jerusalem artichoke**

This trial compared the root yield and carbohydrate content of the four different yacon accessions with Jerusalem artichoke. The field trial was a randomised block layout with four replicates. The Jerusalem artichoke planting stock was obtained from a private garden in the Waikato. Twenty plants of the accessions Y42 and Y43 were propagated in winter 1998 from crown pieces held at Crop & Food Research, Lincoln, and transferred to Pukekohe in August 1998 and then grown in a greenhouse. In early September 1998, mature crowns of the Landsendt line and Jerusalem artichoke were broken up into pieces of about 50 g, dipped in a mixture of 5 g Benlate and 15 g Thiram in 10 L of water and grown in pots in the greenhouse. Mature crowns of the Ohaupo line were sourced and broken up into 50 g propagules, dipped in fungicide and planted in pots in October 1998. All lines were taken out of the greenhouse in late October/early November to harden the plants prior to field planting on 11 November 1998. Individual plots consisted of a single row of five plants spaced 30 cm apart and 1.5 m between rows with the three inner plants recorded for yield. The plants were irrigated by overhead sprinklers every three days for the first two weeks after planting. They were irrigated during dry periods. On 23 February 1999 the plants were side dressed with NPK fertiliser (12:10:10) at a rate of 50 kg/ha. Weeds were controlled by hand weeding and
directed spraying with Roundup®. Looper caterpillars were controlled by spraying with Orthene® on 14 January, 5 February and 17 March 1999 and with Decis® on 27 February at the recommended label rates using a tractor mounted boom sprayer.

The trial was harvested on 14 June 1999. The tops of the plants were harvested close to ground level by hand, and the three recorded plants in each plot were hand-dug. Storage roots were separated from the crown and washed, counted and weighed. Samples were dried at 80°C for dry matter (DM), and carbohydrate analyses.

**Carbohydrate analysis**

For the carbohydrate analyses dried samples (100 mg) were weighed into 50 ml screw capped tubes. Distilled water (25 ml) was added and the tubes heated to 80°C with agitation to disperse the material. The solutions were cooled to room temperature and diluted to 50 ml with distilled water with 1 ml aliquots of these solutions used for analysis. The total water-soluble carbohydrate content of the extracts was determined by the phenol-sulphuric assay (Dubois et al., 1956) using partially hydrolysed chicory inulin (0-80 μg; FrutafitHD, Swift NZ Ltd) as a standard. The fructan content of the extracts was determined using the Megazyme (Megazyme International, Ireland Ltd) fructan assay kit. Samples were also analysed qualitatively by thin-layer chromatography. Aliquots of extracts containing approximately 50 μg of carbohydrate were applied to TLC plates (20 x 20 cm, Kieselgel 60 F254, Merck, Germany) and developed once in propan-1-ol/ethyl acetate/water (5/3/2 v/v/v) at room temperature. Fructooligosaccharides were visualised using the ketose-specific, urea-phosphoric acid stain method (Wise et al., 1955).

**Trial 2: time of harvest of roots**

The trial was planted on 10 November 1999 as a randomised block design with four harvest treatments and three replicates. The trial was established using crown pieces of the Ohaupo line weighing about 80 g each. Individual plots consisted of three rows 1.5 m apart, each containing 7 plants at 30 cm spacings. The five central plants of the middle row were recorded. The trial received a side dressing of 50 kg N/ha as calcium ammonium nitrate on 31 December 1999, six irrigations with 40 mm of water between 14 January and 29 March 2000 and six applications of Monitor® (1.5 l/ha) between 26 January and 13 April 2000 to control aphids and looper caterpillars. Weeds were controlled by hand weeding until canopy closure. The harvest dates were 10 March, 11 April, 15 May and 7 June 2000. Top growth and root production were recorded from the harvested plants and samples were taken and oven dried at 80°C for dry matter analyses.

**Trial 3: time of harvest of top growth and effect on root production**

The trial was planted on 10 November 1999 as a randomised block design with four replicates of four cutting treatments using the Ohaupo line, as in trial 2. Each plot consisted of three rows 1.5 m apart with 10 plants spaced 30 cm apart in each row. The six plants in the centre row were recorded for yield and the rest acted as guard plants. The trial management was the same as in trial 2, except that the side dressing of 50 kg/ha N as calcium ammonium nitrate was applied on 19 January 2000.

The four cutting treatments were: a stubble height of 100-150 mm in March (8-10), in April (11-13), in May (9-11), and in March with a repeat cut in May 2000. The top growth was separated into leaf and stem components and samples were taken for dry matter analyses at each harvest. The root harvest for all treatments was undertaken during 11-16 August 2000; roots were washed, weighed and samples were taken for DM analysis. All DM samples were oven-dried at 80°C.

**Analysis of results**

The trial data were statistically analysed by analysis of variance using GENSTAT package (GenStat, C., 2000). Data were transferred to the log scale where required to stabilise the variance and retransformed to display the results.

**Results**

**Trial 1**

The yacon accessions produced an average of 3.8 kg roots/plant with the production of the individual lines varying from 3.2 to 4.3 kg/plant and an average individual storage root weight of 213 g (Table 1). At the plant population used this equated to a crop yield of 71-96 t/ha. These roots had a DM content of 11.1 % with a total carbohydrate content of 565 mg/kg DW, of which the fructan content was 292 mg/kg DW. There was no superior yacon line in storage root yield. The Ohaupo line produced the largest individual roots (mean 275 g), but its fructan content was lower than Y43 and Landsendt lines.
The tuber yield of Jerusalem artichoke was 35% less than that of the yacon lines, with Jerusalem artichoke tubers an average weight of 26 g, which was 8 times lighter than the yacon roots. The total carbohydrate content of Jerusalem artichoke tubers was lower than, but not statistically different to, the yacon roots. The fructan content of the Jerusalem artichoke was 38% lower than yacon lines Y43 and Landsendt, but similar to the Ohaupo line. The TLC chromatogram showed intense staining at the origin of the Jerusalem artichoke extract, indicating carbohydrate material above DP10, in contrast to the yacon extracts in which all carbohydrate had moved off the origin, indicating that they were all less than DP9 and predominantly in the DP3-6 range (Fig. 1). The 13.2% DM of the Jerusalem artichoke tubers was significantly higher than the highest yacon storage root DM of 11.8%.

Table 1. The yield, mean weight, DM % and total carbohydrate and fructan content of the storage roots of four yacon lines and Jerusalem artichoke tubers.

<table>
<thead>
<tr>
<th>Yacon lines</th>
<th>Root yield (kg/plant)</th>
<th>Root yield (t/ha)</th>
<th>Mean root weight (g)</th>
<th>DM % roots (%)</th>
<th>Total carbohydrate (mg/g DW)</th>
<th>Fructan (mg/g DW)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Y42</td>
<td>4.3</td>
<td>96</td>
<td>184</td>
<td>11.8</td>
<td>520</td>
<td>290</td>
</tr>
<tr>
<td>Y43</td>
<td>3.8</td>
<td>85</td>
<td>182</td>
<td>11.5</td>
<td>580</td>
<td>320</td>
</tr>
<tr>
<td>Landsendt</td>
<td>3.2</td>
<td>71</td>
<td>212</td>
<td>10.3</td>
<td>590</td>
<td>320</td>
</tr>
<tr>
<td>Ohaupo</td>
<td>4.1</td>
<td>92</td>
<td>275</td>
<td>10.8</td>
<td>570</td>
<td>240</td>
</tr>
<tr>
<td>Artichoke</td>
<td>2.5</td>
<td>55</td>
<td>26</td>
<td>13.2</td>
<td>490</td>
<td>230</td>
</tr>
</tbody>
</table>

LSD (0.05) 1.48 23.5 66.2 1.1 ns 56
P value 0.014 0.014 <0.001 <0.001 0.46 0.009

Trial 2
The highest yacon root yield of 4.33 kg/plant, equivalent to 96 t/ha, was achieved by the latest harvest in June (Table 2). Earlier harvesting in March, April or May was progressively detrimental to crop yield with root yields respectively 12, 30, or 59% of the final yield. The plants harvested in March had only 41% of the number of roots of the June harvest, but by the April harvest 74% of the roots were present and 85% by the May harvest (Table 2). The April harvested roots had a slightly lower dry matter percentage than the March harvested roots (9.94 v. 10.82%), but these earlier harvests were 2% lower than later harvests in May and June. The herbage yields were similar for the April, May and June harvests with the herbage yield almost doubling between March and April harvests (Table 2).

Table 2. The effect of time of harvest on the yield, number and DM % of yacon storage roots and the fresh herbage yield.

<table>
<thead>
<tr>
<th>Time of harvest</th>
<th>Fresh herbage</th>
<th>Root yield</th>
<th>Root number</th>
<th>DM % Roots (%)</th>
<th>Root yield (t/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>weight (kg/plant)</td>
<td>(kg/plant)</td>
<td>(no./plant)</td>
<td>(%)</td>
<td>(t/ha)</td>
</tr>
<tr>
<td>March</td>
<td>1.2</td>
<td>0.52</td>
<td>11</td>
<td>10.82</td>
<td>11</td>
</tr>
<tr>
<td>April</td>
<td>2.3</td>
<td>1.32</td>
<td>20</td>
<td>9.92</td>
<td>29</td>
</tr>
<tr>
<td>May</td>
<td>2.4</td>
<td>2.58</td>
<td>23</td>
<td>12.04</td>
<td>57</td>
</tr>
<tr>
<td>June</td>
<td>2.3</td>
<td>4.33</td>
<td>27</td>
<td>12.67</td>
<td>96</td>
</tr>
</tbody>
</table>

LSD (0.05) 0.38 1.07 2.95 0.79 26
P value <0.001 <0.001 <0.001 <0.001 <0.001

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Table 3. The effect of different times of herbage removal in autumn on the leaf and stem production of yacon and on the subsequent storage root yield and DM %.

<table>
<thead>
<tr>
<th>Cut time</th>
<th>Leaf production (t/ha DM)</th>
<th>Stem production (t/ha DM)</th>
<th>Total herbage production (t/ha DM)</th>
<th>Fresh root yield (t/ha)</th>
<th>DM % roots (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>March</td>
<td>2.1</td>
<td>1.5</td>
<td>3.6</td>
<td>39.6</td>
<td>9.98</td>
</tr>
<tr>
<td>April</td>
<td>3.0</td>
<td>3.1</td>
<td>6.1</td>
<td>41.3</td>
<td>7.84</td>
</tr>
<tr>
<td>May</td>
<td>2.7</td>
<td>2.8</td>
<td>5.6</td>
<td>59.4</td>
<td>8.68</td>
</tr>
<tr>
<td>March &amp; May</td>
<td>3.9</td>
<td>2.3</td>
<td>6.2</td>
<td>33.6</td>
<td>8.17</td>
</tr>
<tr>
<td>LSD (0.05)</td>
<td>0.57</td>
<td>0.55</td>
<td>1.2</td>
<td>9.48</td>
<td>1.30</td>
</tr>
<tr>
<td>P value</td>
<td>0.003</td>
<td>&lt;0.001</td>
<td>0.003</td>
<td>&lt;0.001</td>
<td>0.02</td>
</tr>
</tbody>
</table>

Trial 3

Yacon harvested in April or May, or harvested in March and again in May gave a mean total herbage production of 6 t/ha DM, but the single harvest in March gave only 59% of the herbage yield of the later harvests (Table 3). The repeat harvesting in March and May produced the highest leaf percentage (63%) in the total herbage and this was 14% higher than the single harvests of April and May. The repeat harvesting had the greatest detrimental effect on the subsequent root yield and gave 57% of the root yield of the treatment defoliated in May. Single defoliations in March or April gave 67-69% of the May defoliation root yields (Table 3). The highest root dry matter of 10.0% was recorded from the March-harvested treatment with the April and May harvests and the March and May repeat harvests, having a lower dry matter percentage, within the range 7.8-8.7% (Table 3).

Figure 1. Thin layer chromatogram of the root extract of Jerusalem artichoke (Ht) and four yacon lines, Ohaupo (Oh), Landsendt (La), Accession 42, and Accession 43, showing the intense carbohydrate staining for Jerusalem artichoke remaining on the chromatogram origin while the yacon carbohydrate gives intense staining within the polymerisation range DP3-6 with low levels of sucrose (S) and monosaccharides (M).

Discussion

Yacon is a traditional food crop in South America (National Research Council, 1989), but interest around the world is focused on root FOS for use as a functional food as well as the dried herbage as a herbal tea (Grau and Rea, 1997). These trials show that yacon can produce high root yields in northern New Zealand with the maximum,
extrapolated, plot yields of 96 t/ha being similar to reported crop yields in South America (Grau and Rea, 1997). The root yield/plant of 4.3 kg at Pukekohe is higher than that recorded at Lincoln of 2.9 kg/plant (Martin et al., 1997), or at Mosgiel of 1.7 kg/plant (Grau, 1993), and is likely to be related to the warmer conditions for growth at Pukekohe. Yacon requires 6-7 months of good growing conditions to reach maturity, temperatures of 18-25°C and little or no frost (National Research Council, 1989; Grau and Rea, 1997). This regime climate is predominantly found in northern New Zealand.

The potential of yacon for the production of FOS was highlighted in the first trial where the yacon lines gave 28 to 72 % higher root production and 4 to 39 % higher fructan content than Jerusalem artichoke, depending on the accession. The production of low DP fructans by yacon agrees with published results (Ohyama et al., 1990) and indicates that FOS could be extracted from yacon without the need to enzymatically hydrolyse inulins, as is required for chicory or Jerusalem artichoke. The fructan component (52-59 %) of the carbohydrates was similar in range to yacon grown in Equador, but total carbohydrate levels were much less (Hermann et al., 1998). There was a variation of 90 mg/g dry weight in the fructan component of the four yacon lines in the first trial, indicating the possibility of selecting high fructan-producing lines. Large germplasm collections of yacon exist in Peru and Equador (Rea, 1994), but research on compositional diversity suggests that greater variation in carbohydrates is more likely due to the effects of growing and storage conditions than genetic differences (Hermann et al., 1998). Research on factors that influence total sugar content and the FOS component of yacon is the subject of ongoing research at the University of Waikato (N. Wong, pers. comm.).

The DM % of the yacon roots harvested from mature plants ranged from 10.3 to 12.7 % and was similar to the results of Hermann et al., (1998) on South American material, but much less than the 15 to 30 % DM reported in other results (Rea, 1994; Grau and Rea, 1997). Harvesting yacon roots before the plants have completed their growth cycle reduced the DM content of roots. All treatments in the herbage removal trial gave low DM %, but whether this effect is related to the herbage removal remains uncertain.

The time of harvesting trial showed that it was important for yacon to be left until top growth had stopped in winter before harvesting the roots otherwise the root yield was reduced. Very large gains in production were made in the last two months of growth and consequently early harvesting of a crop is likely to severely limit yield potential. This suggests that to maximise crop yields from yacon there is a need to grow this crop in an environment where growth conditions are without frosts in April and May. Planting yacon earlier in the spring may result in the crop maturing earlier in the autumn, but this possibility has not been tested.

Harvesting the yacon herbage in the autumn while the crop is actively growing also reduces subsequent root harvest. Consequently, any development of yacon as a duel purpose crop for both root and herbage production involves compromising between the desirability of early harvests of the foliage to achieve good quality, and the likely reduction in root production if harvesting takes place before plant senescence. Production of yacon herbage may be better undertaken independently of root production with semi-permanent beds established and harvested with multi-harvest systems. This has not been evaluated.

Considerable research is required before yacon can be confidently grown as a commercial crop for FOS production or herbal tea manufacture. Research has already been undertaken to identify appropriate herbicides for weed control (Scheffer et al., 2002) and the plant populations to optimise root yields, but further research is needed to define the fertiliser requirements and pre and postharvest factors that influence crop quality. In addition to optimising the agronomic requirements of yacon, there is a need to develop mechanised methods for handling the crop. Yacon storage roots are attached to a woody crown, and following lifting with a potato digger the separation of the crown for replanting from the storage roots must be mechanised to minimise the labour input required.

These trials results are only the beginning of a programme to develop yacon as a new crop for New Zealand. The production and quality of yacon has been sufficiently promising to encourage the evaluation of yacon on a commercial scale for the production of FOS in a continuing research programme.

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References


