Identification of Phenotypic Traits Associated with Tuber Yield Performance in Non-Staking Cultivation of Water Yam (Dioscorea alata L.)

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Abstract: For sustainable food production, labor-saving cultivation systems are required. Yam, a major food crop, is cultivated mostly with supports such as stakes (staking), which increases tuber yield (TY) but involves high material and labor costs. We, therefore, focused on non-staking water yam cultivation, in which no stakes are used. The effects of different cultivation methods (staking vs. non-staking) on TY, yield components, shoot traits, and tuber shape of six cultivars were investigated in a two-year field experiment, and phenotypic traits related to yielding ability in non-staking cultivation were analyzed. Averaged across years and cultivars, TY was significantly lower (by 19%) in non-staking than in staking cultivation because of smaller single-tuber weight. TY was significantly affected by the cultivation × cultivar interaction. We found no difference among cultivars in staking cultivation. In non-staking cultivation, Yamatomakousha and Shirokoushaman 1 cultivars had higher TY than the other cultivars, which was similar to their TY in staking cultivation. Shoot dry weight and vine number were closely associated with TY in both cultivations, whereas lower tuber length-to-width ratio was strongly related to higher TY only in non-staking. Tubers of Yamatomakousha and Shirokoushaman 1 were more rounded than those of other cultivars. In non-staking cultivation, these two cultivars showed a higher vine number and, thus, maintained higher TY owing to higher above-ground growth. Therefore, rounded tubers and high vine number are target traits for non-staking cultivation of water yam.

Keywords: labor-saving cultivation; yam; temperate region; tuber yield; cultivar difference; vine development; tuber shape; apical dominance

1. Introduction

Water yam (Dioscorea alata L.) is one of the most important edible yam species [1]. This species, which is also commonly known as greater yam, winged yam, ube, or purple yam, is widely grown in tropical and subtropical countries for food and income [2]. Among yam species, water yam has superior yielding ability and environmental adaptability because of its greater single-tuber weight, tolerance to a low-water environment, and growth adaptability to low-fertility and alkaline soils compared with other Dioscorea species [3–5]. Therefore, water yam may be vital for food security and income generation under future climate change.

In Japan, Chinese yam (Dioscorea opposita Thunb.), water yam, and Japanese yam (Dioscorea japonica Thunb.) are mainly cultivated. Chinese yam accounts for 88% of total yam production in Japan [6], but detailed data on water yam and Japanese yam production are not publicly available. Water yam production in Japan is limited to the southwestern islands of Kyushu and Okinawa (regions of transition from temperate to subtropical climate) in Kagoshima and Okinawa Prefectures [5]. Its total production is low, and it is not widespread in the domestic food market, possibly because water yam is native to the tropics and is, therefore, more susceptible to low temperatures than the other yam
species [7]. Less information is available on the yield potential and cultivation of water yam in temperate regions than in tropical and subtropical regions, and cultivation systems adapted to temperate regions are not established.

Low-cost and labor-saving cultivation is important for achieving sustainable crop production. Regarding yams, research on reducing production costs is progressing, such as studies on optimum seed sett size [8,9] and seed tuber production methods using vine cuttings [10,11]. Meanwhile, yam cultivation in many regions follows traditional farming methods such as slash-and-burn agriculture [12], and innovation in cultivation technology has not progressed much. Cultivation methods such as the effects of fertilizer application [13,14], planting dates [15], and density [16] on water yam have been studied, but little is known about labor-saving cultivation methods.

Yams, which are vine plants, are generally cultivated with supports such as stakes [17,18]. Tuber yield is higher in yams supported with stakes than in non-staked ones owing to the promotion of vine elongation and vigorous above-ground growth [10,19–22], but the use of stakes for support involves the costs of the stakes, labor for their installation, and training [23], and staked plants are susceptible to damage by strong winds such as typhoons. Hence, non-staking cultivation might be an effective, low-cost, and labor-saving approach. However, only a few studies have focused on improving tuber yield in non-staked cultivation [24–27], and factors involved in cultivar suitability for non-staking cultivation remain unknown.

The aim of the present study was to obtain basic knowledge for cost and labor-saving non-staking cultivation of water yam. We performed a two-year field experiment in a temperate region in southwestern Japan to investigate (1) cultivar differences in tuber yield and yield-related traits in staking and non-staking cultivation and (2) target traits involved in yield performance in non-staking cultivation.

2. Materials and Methods

2.1. Plant Materials

Six water yam (Dioscorea alata L.) cultivars, Ishigakizairai (Ishi, JP175478), Oogiiimo (Oogi, JP173294), Nagaimo (marukei) (Naga, JP173293), Nicchakousha (Niccha, JP175490), Yamatomakousha (Yama, JP175488), and Shirokoushaman 1 (Shiro, JP173295), obtained from the National Institute of Agrobiological Sciences Genebank, were used in this study. All cultivars are classified as Japanese landraces.

2.2. Location, Cultivation and Growth Conditions

A field experiment was conducted during the yam cropping seasons in 2020 and 2021 at the Ito Plant Experiment Fields and Facilities (33°59′ N, 130°2′ E), Faculty of Agriculture, Kyushu University, Fukuoka Prefecture, Japan. In each year, different upland fields with alluvial soil were used; soil characteristics are listed in Table S1. Fertilizers (20 g m⁻² N, P, K as compound fertilizer and 10 g m⁻² magnesium lime as basal dressing) were uniformly incorporated to a depth of 20 cm by a rotary tiller. Ridges of approximately 25 cm in height were made with white/black plastic film mulch.

Seed tubers were selected from tubers harvested in a previous year and stored at 15 °C. Whole tubers were cut into pieces of approximately 40 g per sett, treated with fungicide (0.5% benomyl; Benlate wettable powder, Sumitomo Chemical Co., Tokyo, Japan), and incubated in a plastic container with vermiculite at 25 °C. Sprouted setts were transplanted to the field at one sett per 1 m⁻² (1.0 m × 1.0 m) and 5 cm depth on 11 June 2020 and 23 June 2021.

In plots prepared by the staking method, vines were staked on 1.8 m plastic sticks, whereas no sticks were used in plots prepared by the non-staking method (Figure S1). Rainfall was the only source of water. No herbicide was applied, and hand weeding was conducted when necessary. At approximately 2 months after transplanting, 10 g m⁻² N, P, K as compound fertilizer was applied 15 cm away from each plant as top dressing.
In each year, the experimental design used a split-plot arrangement; the main plot was the cultivation method (staking vs. non-staking), and the subplot was a cultivar with three replications. A subplot consisted of six rows 6.0 m long (36 m²) in 2020 and six rows 4.0 m long (24 m²) in 2021. The total area was 384 m² in 2020 and 230 m² in 2021.

Meteorological conditions during the growth season are presented in Table S2. The daily means of precipitation, temperature, and solar radiation were recorded at the weather station of the Ito Plant Experiment Fields and Facilities, located approx. 100 m away from the field. Total precipitation, average temperature, and average solar radiation for the growth period were 1455 mm, 21.7 °C, and 14.6 MJ m⁻² day⁻¹ in 2020 and 1279 mm, 21.8 °C, and 14.8 MJ m⁻² day⁻¹ in 2021.

2.3. Measurements

At physiological maturity, 2 or 3 plants per 6 m² in 2020 and 2 plants per 4 m² in 2021 were harvested manually from the center of a row of each plot on 7 December. We measured the number of primary vines, tuber number, individual tuber weight, and maximum tuber length and width. After tuber weighing, a moderate-size tuber from each plot was dried (80 °C, 72 h) and used to calculate moisture content. Tuber yield was calculated based on 80% moisture content. Small tubers (<100 g) were considered unmarketable and excluded from the yield survey.

In order to evaluate shoot dry weight, the aboveground parts (leaves and vines/stems) from two plants were harvested at the time of tuber harvesting from the center of a row of each plot, completely air-dried, and weighed.

2.4. Statistical Analyses

Statistical analyses were performed by using SPSS v. 28.0 software (IBM, Tokyo, Japan). The statistical model used a split-plot treatment structure. Analysis of variance (ANOVA) was used to test the effects of cultivation method and cultivar on tuber yield and yield components (number of tubers, single tuber weight). The cultivation method and cultivar were considered fixed effects, and the year and replication (block) were considered random effects for the tuber yield and yield components. When the F-test result of the ANOVA was significant \( p < 0.05 \), Fisher’s protected least significant difference (LSD) test was used to detect significant differences between means. Simple linear regression analyses were used to evaluate the relationships between tuber yield and yield components, shoot and tuber traits, and the relationships among relative tuber yield, shoot dry weight, and vine number by calculating the ratio of values in non-staking cultivation to values in staking cultivation and the tuber length-to-width ratio.

3. Results

3.1. Tuber Yield, Yield Components, Shoot Traits, and Tuber Shape

The effects of cultivation method and cultivar on tuber yield, yield components, and shoot traits were analyzed by ANOVA (Table 1). When averaging across cultivars, tuber yield was lower by 19% \( p < 0.01 \) and tuber weight by 29% \( p < 0.001 \) in non-staking cultivation than in staking cultivation, but tuber number did not differ, indicating that tuber yield was lower in non-staking cultivation because of lower tuber weight (Table 1). A significant cultivation × cultivar interaction \( p < 0.05 \) was found for tuber yield. We, therefore, analyzed the cultivar difference of tuber yield for each cultivation method and the effect of cultivation method on tuber yield for each cultivar (Figure 1a). In staking cultivation, tuber yield was not significantly different among all six cultivars (av. 1777 g plant⁻¹, Figure 1a). In non-staking cultivation, tuber yield was significantly higher in Yama (1939 g plant⁻¹) and Shiro (2413 g plant⁻¹) than in the other four cultivars (av. 1063 g plant⁻¹), and the mean yield was 109% of that in staking cultivation in Yama and 120% in Shiro, but it was 52–75% in the other four cultivars, especially Ishi and Niccha, which had significantly lower yields in non-staking cultivation (Figure 1a). Although the effects of cultivar on tuber number \( p < 0.001 \) and tuber weight \( p < 0.01 \) were significant, these
variables were not affected by the cultivation × cultivar interaction (Table 1). Our data demonstrate that yielding ability in non-staking cultivation differs among water yam cultivars.

**Table 1.** Tuber yield, number of tubers, tuber weight, and shoot dry weight of six water yam cultivars in staking and non-staking cultivation in 2020 and 2021.

<table>
<thead>
<tr>
<th>Cultivar (A)</th>
<th>Cultivation (B)</th>
<th>Tuber Yield (g Plant⁻¹)</th>
<th>Number of Tubers (Plant⁻¹)</th>
<th>Tuber Weight (g Tuber⁻¹)</th>
<th>Shoot Dry Weight (g Plant⁻¹)</th>
<th>Number of Vines (Plant⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nagaimo (marukei)</td>
<td>Staking</td>
<td>1777</td>
<td>2.9</td>
<td>673</td>
<td>324</td>
<td>9.1</td>
</tr>
<tr>
<td></td>
<td>Non-staking</td>
<td>1434</td>
<td>3.0</td>
<td>477</td>
<td>251</td>
<td>9.3</td>
</tr>
<tr>
<td>Shirokoushaman 1</td>
<td>Staking</td>
<td>1569 ab</td>
<td>3.5 bc</td>
<td>466 a</td>
<td>264 a</td>
<td>11.0 c</td>
</tr>
<tr>
<td></td>
<td>Non-staking</td>
<td>1425 ab</td>
<td>1.9 a</td>
<td>761 c</td>
<td>263 a</td>
<td>5.8 a</td>
</tr>
<tr>
<td>Yamatomakousha</td>
<td>Staking</td>
<td>1317 a</td>
<td>2.3 a</td>
<td>581 ab</td>
<td>259 a</td>
<td>6.6 a</td>
</tr>
<tr>
<td></td>
<td>Non-staking</td>
<td>1253 a</td>
<td>2.7 ab</td>
<td>492 ab</td>
<td>343 b</td>
<td>11.3 c</td>
</tr>
<tr>
<td>Nicchakousha</td>
<td>Staking</td>
<td>1859 bc</td>
<td>3.8 c</td>
<td>510 ab</td>
<td>343 b</td>
<td>11.3 c</td>
</tr>
<tr>
<td></td>
<td>Non-staking</td>
<td>2210 c</td>
<td>3.6 c</td>
<td>641 bc</td>
<td>364 b</td>
<td>11.5 c</td>
</tr>
</tbody>
</table>

ANOVA:
- A × B
- A
- B
- A × B
- ns, not significant.

Values of a parameter followed by the same letter did not differ significantly between cultivars (ANOVA followed by Fisher’s LSD test, *p* < 0.05). ANOVA results: *** *p* < 0.001; ** *p* < 0.01; * *p* < 0.05; ns, not significant.

**Figure 1.** Effect of cultivation method × cultivar interaction on (a) tuber yield and (b) vine number. The data were averaged across the 2020 and 2021 growing seasons. Values are means ± S.E. of 6 replications derived from different plots. The same letters above bars indicate no significant difference in the LSD test at the 5% level. Values in parentheses indicate ‘non-staking’ values (%) relative to ‘staking’ values. ** *p* < 0.01; * *p* < 0.05; † *p* < 0.10; ns, not significant.

The cultivation method significantly affected shoot dry weight (*p* < 0.001) but not the number of vines (Table 1). Shoot dry weight averaged for all cultivars was significantly (by 23%) lower in non-staking than in staking cultivation, whereas that averaged across cultivation methods was higher in Yama and Shiro. The number of vines was higher in Yama, Shiro, and Ishi, and vine number was significantly affected by cultivation × cultivar interaction (*p* < 0.05). Among all six cultivars, Ishi had the highest vine number (12.9 plant⁻¹) in staking cultivation (Figure 1b), whereas Yama (12.6 plant⁻¹) and Shiro (13.3 plant⁻¹) had the highest vine number in non-staking cultivation and those values
were higher by 26% \((p < 0.10)\) in Yama and by 38\% \((p < 0.05)\) in Shiro than in staking cultivation.

We also analyzed the effects of cultivation method and cultivar on morphological traits of tubers by ANOVA (Table 2). Cultivation method significantly affected tuber length and width \((p < 0.01)\), but not tuber length-to-width ratio. Tuber length and tuber width averaged across cultivars were significantly lower (by 9\% and 12\%, respectively) in non-staking than in staking cultivation. Tuber length and width and their ratio differed significantly among the cultivars \((p < 0.001)\). Tuber length averaged across cultivation methods was shorter in Niccha, Yama, and Shiro, and tuber width tended to be larger in Yama and Shiro than in the other cultivars. The tuber length-to-width ratio in cultivars was as follows: Shiro, Yama < Niccha < Naga, Oogi < Ishi, indicating that the cultivars can be classified into four groups according to tuber roundness. Tuber length and width and their ratio were not significantly affected by cultivation \(\times\) cultivar interaction.

### Table 2. Tuber length, tuber width, and tuber length-to-width ratio of six water yam cultivars in staking and non-staking cultivation in 2020 and 2021.

<table>
<thead>
<tr>
<th>Cultivation ((A))</th>
<th>Tuber Length ((cm))</th>
<th>Tuber Width ((cm))</th>
<th>Tuber Length-to-Width Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staking</td>
<td>21.8</td>
<td>8.6</td>
<td>2.9</td>
</tr>
<tr>
<td>Non-staking</td>
<td>19.8</td>
<td>7.6</td>
<td>3.0</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Cultivar ((B))</th>
<th>Tuber Length ((cm))</th>
<th>Tuber Width ((cm))</th>
<th>Tuber Length-to-Width Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ishigakizairai</td>
<td>26.6 b</td>
<td>5.8 a</td>
<td>4.8 d</td>
</tr>
<tr>
<td>Oogiimo</td>
<td>28.3 b</td>
<td>7.7 b</td>
<td>4.0 c</td>
</tr>
<tr>
<td>Nagaimo (\text{marukei})</td>
<td>27.6 b</td>
<td>7.5 b</td>
<td>3.9 c</td>
</tr>
<tr>
<td>Nicchakousha</td>
<td>14.9 a</td>
<td>8.2 bc</td>
<td>2.0 b</td>
</tr>
<tr>
<td>Yamatomakousha</td>
<td>14.0 a</td>
<td>9.2 cd</td>
<td>1.6 a</td>
</tr>
<tr>
<td>Shirokoushaman 1</td>
<td>13.4 a</td>
<td>10.3 d</td>
<td>1.3 a</td>
</tr>
</tbody>
</table>

ANOVA: **NS**

Values of a parameter followed by the same letter did not differ significantly between cultivars (ANOVA followed by Fisher’s LSD test, \(p < 0.05)\). ANOVA results: *** \(p < 0.001)\; ** \(p < 0.01)\; ns, not significant.

3.2. Relationship between Tuber Yield and Yield Components, Shoot Traits, and Tuber Shape

A simple linear regression analysis was conducted to identify traits related to tuber yield in both cultivation methods (Table 3). In staking cultivation, tuber yield was highly positively correlated with shoot dry weight \((p < 0.001)\) and the number of tubers \((p < 0.01)\) and was positively correlated with vine number \((p < 0.05)\), but it was not significantly correlated with tuber weight or shape traits. In non-staking cultivation, tuber yield was highly positively correlated \((p < 0.001)\) with the number of tubers, tuber weight, shoot dry weight, vine number, and tuber width but was negatively correlated with tuber length \((p < 0.05)\) and tuber length-to-width ratio \((p = 0.001)\). These results suggest that tuber number, shoot dry weight, and vine number are related to tuber yield in both cultivation methods.

3.3. Key Traits of Non-Staking Adaptability on Tuber Yield

Next, we focused on target traits for non-staking adaptability and used the value of non-staking to the value of the staking ratio as its indicator. When all plot data were pooled, relative tuber yield was significantly positively correlated with relative shoot dry weight \((p < 0.001, \text{Figure 2a})\); relative shoot dry weight was also significantly positively correlated with relative vine number \((p < 0.001, \text{Figure 2b})\); and relative vine number was significantly negatively correlated with tuber length-to-width ratio \((p < 0.001, \text{Figure 2c})\). These results clarified that high tuber yield in non-staking cultivation was due mainly to high shoot biomass, which in turn resulted from the increase in vine number closely associated with high tuber roundness.
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Table 3. Correlation coefficients between tuber yield and the number of tubers and vines, tuber weight, shoot dry weight, and tuber length, width, and their ratio across the 2020 and 2021 growing seasons (n = 36).

<table>
<thead>
<tr>
<th>Cultivation</th>
<th>Variable</th>
<th>Tuber Yield (g Plant⁻¹)</th>
<th>r</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staking</td>
<td>Number of tubers (plant⁻¹)</td>
<td>0.504</td>
<td>0.002</td>
<td>**</td>
</tr>
<tr>
<td></td>
<td>Tuber weight (g tuber⁻¹)</td>
<td>0.274</td>
<td>0.105</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Shoot dry weight (g plant⁻¹)</td>
<td>0.778</td>
<td>&lt;0.001</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Number of vines (plant⁻¹)</td>
<td>0.366</td>
<td>0.028</td>
<td>*</td>
</tr>
<tr>
<td></td>
<td>Tuber length (cm)</td>
<td>−0.319</td>
<td>0.526</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Tuber width (cm)</td>
<td>0.259</td>
<td>0.127</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>Tuber length-to-width ratio</td>
<td>−0.121</td>
<td>0.483</td>
<td>ns</td>
</tr>
<tr>
<td>Non-staking</td>
<td>Number of tubers (plant⁻¹)</td>
<td>0.771</td>
<td>&lt;0.001</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Tuber weight (g tuber⁻¹)</td>
<td>0.603</td>
<td>&lt;0.001</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Shoot dry weight (g plant⁻¹)</td>
<td>0.883</td>
<td>&lt;0.001</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Number of vines (plant⁻¹)</td>
<td>0.629</td>
<td>&lt;0.001</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Tuber length (cm)</td>
<td>−0.380</td>
<td>0.022</td>
<td>*</td>
</tr>
<tr>
<td></td>
<td>Tuber width (cm)</td>
<td>0.783</td>
<td>&lt;0.001</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Tuber length-to-width ratio</td>
<td>−0.519</td>
<td>0.001</td>
<td>***</td>
</tr>
</tbody>
</table>

*** p < 0.001; ** p < 0.01; * p < 0.05; ns, not significant.

Figure 2. Relationships between (a) relative tuber yield and relative shoot dry weight, (b) relative shoot dry weight and relative vine number, and (c) relative vine number and tuber length-to-width ratio for six cultivars under staking or non-staking cultivation in two years. The values shown are the ratios of values in non-staking cultivation to values in staking cultivation. Error bars indicate the S.E. of 6 replicates. Pearson’s correlation coefficient (r) was calculated with the data combined across the two years (n = 36).

4. Discussion

Water yam tuber yields in staking (17.8 t ha⁻¹; 1777 g plant⁻¹) and non-staking (14.3 t ha⁻¹; 1434 g plant⁻¹) cultivation were comparable to those obtained in field trials in Côte d’Ivoire (15.5–24.6 t ha⁻¹) [13]. Non-staking cultivation resulted in 19% lower yields than staking cultivation did (Table 1), consistent with the reported lower yields under non-staked conditions [10,19]. Tuber yield across both cultivation methods differed significantly among cultivars and ranged from 1253 to 2210 g plant⁻¹ (Table 1). We found that tuber yield was significantly affected by cultivation method × cultivar interaction (Table 1). No differences in yield were found among cultivars in staking cultivation; in non-staking cultivation, the yield of four cultivars decreased, but that of Yama and Shiro did not (Figure 1a), which was a remarkable characteristic rarely reported in water yam [25,26]. Single tuber weight, length, and width were significantly lower in non-staking cultivation, but no difference was found in the number of tubers (Table 1). Therefore, the reduction of tuber yield in non-staking cultivation may be caused by a decreased tuber weight, mainly
because of a smaller tuber size. In a yield trial of *Dioscorea* spp., single tuber weight was reduced in non-staked cultivation [10,20].

Above-ground biomass at harvest is closely related to tuber yield in *D. rotundata* [28–30] and *D. cayenensis* [31]. In this study, shoot dry weight at harvest was significantly lower in the non-staking plants than in the staked ones (Table 1), and significant positive correlations were found between tuber yield and shoot dry weight and vine number in both cultivation methods (Table 3). These results suggest that non-staking cultivation suppressed above-ground growth, thereby reducing the amount of carbon assimilates translocated from above-ground organs to tubers. Srivastava and Gaiser [32] reported that fertilizer application increased tuber yield in *D. rotundata* by increasing above-ground biomass, but it did not affect dry matter partitioning to tubers. These authors, therefore, inferred that translocation efficiency (translocation amount per above-ground biomass) in yam is almost constant regardless of nutrient status. To improve tuber yield in water yam, it seems necessary to enhance shoot biomass production to accumulate more carbon assimilates for translocation.

The number of tubers and vines per plant was approximately three and nine, respectively, with no significant effect on the cultivation method (Table 1), indicating that three vines, on average, were formed on a single tuber. *Dioscorea rotundata* reportedly forms one vine per tuber [33], which differs from our result. Therefore, we suggest that in water yam, new adventitious buds are induced from a single tuber formed on a vine. Thus, the source–sink unit during tuber enlargement consists of leaves on three vines and a single tuber.

The number of vines was positively correlated with tuber yield (Table 3) and was significantly affected by the cultivation method × cultivar interaction (Table 1). Cultivars with higher yield in non-staking (Yama and Shiro) had higher vine numbers (Table 1, Figure 1b). It can be inferred that a sufficient number of vines, especially in non-staking cultivation, contributes to higher yields via vigorous shoot growth.

Interestingly, tuber yield was strongly negatively correlated with tuber length-to-width ratio in non-staking ($r = -0.519^{***}$), indicating that tuber yield was higher at higher tuber roundness (Table 3). Using the value of non-staking to the value of staking ratio, we found that high tuber yield in non-staking was due mainly to high shoot dry weight, which in turn resulted from high vine number per plant closely associated with high tuber roundness. In non-staking cultivation, cultivars with more rounded tubers had a smaller reduction in vine number, thereby maintaining above-ground growth and ensuring sufficient sourcing capacity and thus maintaining higher tuber yields. This could be a mechanism for the non-staking adaptability in the tuber yield of water yam.

Tuber morphology in water yam varies greatly among cultivars and lines, and tuber shape has been classified as round, oval (based on length), cylindrical or long cylindrical (based on width), and irregular or deformed [34,35]. Tuber shape segregates in F$_1$ populations derived from parental lines of *D. alata* with different tuber shapes [36,37], suggesting that tuber morphological traits are controlled by genetic factors. In our study, single tuber weight decreased in non-staking cultivation, but tuber length-to-width ratio was unaffected by cultivation (Table 2), indicating that tuber shape is determined by genotype rather than the environment. Recently, Bredeson et al. [3] identified a QTL for tuber shape on chromosome 7 by genome-wide association study (GWAS) of water yam lines. Ehounou et al. [38] developed two mapping populations of water yam by hand pollination and detected several major QTLs for tuber size (length-to-width ratio) on linkage group 16. The future use of those QTLs for line selection and breeding should enable modification of tuber shape.

In general, budding in yam tubers is apically dominant, and adventitious bud differentiation occurs early in the head and tail parts and late in the middle part [39]. It is likely that vigorous growth of initially elongated vines in a staked plant prevents the induction of other adventitious buds on the seed tuber. In non-staking cultivation, biotic or abiotic stresses are more likely to inhibit the growth of the shoot apex, which in turn
induces adventitious bud differentiation and, thus, new vine growth. In the present study, we observed a significant cultivation method × cultivar interaction for vine number per plant and found significant cultivar differences in both cultivations (Table 1, Figure 1b). A significant correlation between vine number and tuber length-to-width ratio was observed only in non-staking cultivation (Figure S2). Thus, cultivars with high tuber roundness are more likely to induce adventitious buds and primary vines without support; that is, their apical bud dominance is less effective, leading to the development of multiple additional vines. Since the phytohormones auxin and cytokinin are the main regulators of apical dominance [40] and auxin is involved in tuber morphogenesis in root and tuber crops [41], these hormones may be responsible for cultivar differences in vine formation and tuber roundness in non-staking. This is an interesting point, and we plan to investigate the phytohormone-mediated regulation of tuber morphology and its interaction with adventitious bud and vine induction.

Our field experiment demonstrated that cultivars with higher tuber roundness had higher vine number, and thus maintained higher tuber yield owing to higher above-ground growth when grown in non-staking. It is possible that these traits were not found in the cultivation of water yam, which is normally grown with stakes. This finding is a simple and clear result, and we believe that it provides important information for the low-cost and labor-saving cultivation of water yam. However, because the number of tested cultivars was rather small and they were grown at a single site, and some areas are seriously affected by yam anthracnose disease, which can be a major risk for non-staking cultivation, it will be necessary to conduct yield trials with more cultivars in tropical and subtropical regions in order to assess the non-staking adaptability of tuber yields.

In future research, the relationship between tuber morphology and vine development will be analyzed from a physiological aspect to clarify why roundness varieties have higher yields in non-staking cultivation. We will also focus on light-intercepting traits of the cultivars at the canopy level based on shoot characteristics (vine growth, leaf area and its thickness, and photosynthetic ability).

5. Conclusions

To identify the key traits involved in tuber yield performance in water yam, we conducted a two-year field experiment under staking and non-staking cultivation. The results showed that yielding ability differs among cultivars only in non-staked condition. We found two cultivars, Yama and Shiro, out of six cultivars, show higher tuber yield than the other cultivars in non-staking, which performed equally well in both cultivations. Tubers of Yama and Shiro were more rounded than those of other cultivars. Shoot dry weight and vine number were closely associated with tuber yield in both cultivations, whereas lower tuber length-to-width ratio (tuber roundness) was strongly related to higher tuber yield only in non-staking. These findings suggest that water yam cultivars with higher tuber roundness have higher vine number and thus maintain higher tuber yield owing to higher shoot growth when grown in non-staking. Thus, rounded tubers and high vine number are target traits for labor-saving, non-staked cultivation.

Supplementary Materials: The following materials are available online at: https://www.mdpi.com/article/10.3390/agronomy12102323/s1, Table S1: Soil characteristics of the experimental fields at Faculty of Agriculture, Kyushu University, Fukuoka, Japan, in 2020 and 2021; Table S2: Monthly precipitation, mean air temperature, and solar radiation in Ito Plant Experiment Fields and Facilities during the 2020 and 2021 growing seasons; Figure S1: Water yam cultivation in the experimental field in 2021. (a) Overall view of the field. (b) Staking and non-staking cultivation in the plot; Figure S2: Relationships between tuber length-to-width ratio and vine number of six cultivars under staking and non-staking cultivation in two years. Bars indicate S.E. of 6 replicates. Pearson’s correlation coefficient (\(r\)) was calculated with the data combined across two years (\(n = 36\)).
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References
7. Olorunda, S.O.; Macklon, E.S. Effects of storage and chilling temperature on low absorption salt retention capacity and respiratory pattern in yam tubers. J. Sci. Agric. 1976, 27, 405–412. [CrossRef]


