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## *Sphagnum* moss is a promising growth substrate in arctic bramble container cultivation

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#### ABSTRACT

Development of container cultivation methods for arctic bramble (*Rubus arcticus* L.) is currently underway. The aim of this study was to evaluate *Sphagnum* moss and two substrate mixes containing peat and coir or perlite as alternatives for a pure peat substrate in arctic bramble container cultivation, with particular interest on *Sphagnum* moss. The experiment was conducted in plastic high tunnel in plant towers with three planting levels (Top, Middle, Bottom). The substrates used were unfertilised peat (UP), an UP and perlite mix (80/20 by dry loose volume) (UPP), an UP and commercial coir mix (50/50) (UPCoir) and unfertilised *Sphagnum* moss (SM). Plant vigour was higher in SM compared to UP and UPP, while total fruit yield, mean fruit weight and individual drupelet weight were higher in SM compared to UPP. Both plant vigour and fruit yield were substantially reduced on Middle and especially Bottom level, compared to the Top level of plant towers. Water retention measurement of pure substrate materials showed higher air volume content in SM compared to UP or pure coir material. We conclude that *Sphagnum* moss is a highly promising substitute for peat as a substrate in arctic bramble container cultivation.

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**KEYWORDS** Container grown plants; greenhouse soils; intensive cropping; protected cultivation; *Rubus arcticus* L

#### Introduction

Arctic bramble (*Rubus arcticus* L.) is a high value berry crop that is both harvested from the wild and cultivated for a niche market, mainly in Finland. It has an exquisite flavour that is highly sought after, and could potentially become a more important crop than it currently is. Marketing the arctic bramble as a luxury product requires a modernised cultivation system that can reliably produce high-quality fruit.

The arctic bramble is a perennial rhizomatous plant with annual flowering ramets typically reaching 10– 30 cm in height (Ryynänen 1972, 1973). The ramets originate as root or basal suckers, which undergo terminal flower initiation while still underground and remain dormant over winter (Ryynänen 1972, 1973). After first (terminal) flowering in the spring, there is often second phase of flowering associated with the growth of axillary shoots (Ryynänen 1973). Cross-pollination by insects is necessary for arctic bramble fruiting (Tammisola and Ryynänen 1970; Tammisola 1988). In traditional field cultivation, arctic bramble plants compete poorly against weeds (Ryynänen 1973; Hellqvist 2000; Kokko et al. 2012), and the emergence of fungal diseases has caused a decline in arctic bramble cultivation in Finland (Koponen et al. 2000). As with other berry crops, development of soilless, protected cultivation is under way for extended season, better quality fruit and more efficient harvest. Within protected cultivation systems, there is also interest in vertical farming, using towers of stacked containers or similar arrangements, which have been found to allow higher yields in limited space (Fernández et al. 2018).

Intensive crop production in containers requires a growth substrate with highly optimised cultivation properties. Substrate total pore space and pore size distribution determine the ability of the substrate to contain air and thus provide plant roots with oxygen, while also retaining water for the plant between irrigations. As the substrate dries, a progressively lower water potential (stronger matric suction) is required to pull water out of progressively smaller pores against the capillary force, filling the pores with air instead. The volume of pores emptying of water between 0 and -10 hPa is often considered the baseline for air volume (AV) immediately after watering (Gruda and Schnitzler 2004; Michel 2010), assuming the largest

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pores in the substrate are drained simply by gravity. The pore space emptying between -10 and -100 hPa is considered the measure of plant available water (Michel 2010), since in this water potential range the plant roots are able to effectively extract water. Gruda and Schnitzler (2004) divide the capacity of available water into easily available water (EAV) (released between -10 and -50 hPa) and water buffer capacity (WBC) (released between -50 and -100 hPa), where the former represents water optimally available for growth while the latter allows plants time to adapt physiologically as drought conditions begin to set in. Much of the water retained in the substrate at -100 hPa may be still marginally available for plants, but this level of drought should not occur in normal cultivation (Gruda and Schnitzler 2004; Michel 2010). For cultivation purposes, it is essential to maximise the content of pore space fractions that contain AV and plant available water, as opposed to the pore space containing less available water or the space taken by solid substrate material. Pore size distribution of a substrate is greatly affected by the degree of substrate compression, usually measured via an increase in bulk density (Raviv and Lieth 2008), as well as the inherent structure of the substrate material.

The selection of growth substrate for container cultivation involves consideration for availability, economical feasibility, meeting the specific requirements of the crop and the production system and, increasingly, ecological sustainability (Gruda 2019). Peat is traditionally the most common growth substrate used in soilless horticulture. It is compostable, locally available in many areas such as northern Europe, and has many favourable physical and chemical properties for container cultivation. Michel (2010) reviews the main physical properties of peat as a growth substrate, such as traits relating to water retention, wettability and physical stability. However, the ecological sustainability of peat use has been frequently questioned (Gruda 2012; Carlile and Coules 2013; Neumaier and Meinken 2015; Gruda 2019). This necessitates finding alternative substrates with equal or better cultivation properties compared to peat. Ideally, new substrate materials and mixes could improve or complement the suboptimal traits of peat, or add new properties such as biofertilisation or biostimulation (Gruda 2019).

Sphagnum mosses (Sphagnum spp.), common in northern hemisphere peat bogs, are the primary material of origin for high-quality horticultural peat. There is also a growing interest in the use of Sphagnum moss as such, to substitute peat in container horticulture (see for example Aubé et al. 2015; Müller and Glatzel 2021). While in Finland *Sphagnum* moss is harvested from natural bogs (Silvan et al. 2017), *Sphagnum* farming has also been investigated (Pouliot et al. 2015; Gaudig et al. 2017). The properties of *Sphagnum* moss as a growth substrate have been studied by Emmel (2008) and Kämäräinen et al. (2018, 2020). To our knowledge, the use of *Sphagnum* moss in arctic bramble container cultivation has not been previously investigated.

Coir, or fibrous material separated from coconut mesocarp, is a waste product of coconut production. It is widely used as a substrate in soilless cultivation and is available as processed commercial mixes, including more coarse material (cocofibre or coir proper) and finer parenchymatous material (Neumaier and Meinken 2015). These have relatively good air capacity, capillarity and structural stability, with little risk of nitrogen immobilisation via decomposition (Domeno et al. 2009; Neumaier and Meinken 2015). In a previous study (Tommila et al. 2022), we evaluated the suitability of coir as a substrate for arctic bramble compared to standard horticultural peat.

This study was focused on evaluating *Sphagnum* moss in particular as a possible alternative for peat in arctic bramble soilless cultivation. In addition to peat and *Sphagnum* moss, we included two other substrates where peat was partially substituted with either a commercial coir mix or perlite. Our first hypothesis was that, compared to pure peat, some of the three substrates would have improved water retention characteristics and increase arctic bramble plant vigour and fruit yield.

The plants were grown during summer in a high plastic tunnel, in containers stacked into plant towers with partly circulated fertigation flow. We observed the effect of tower level in this arrangement to see, whether the plants on lower levels would tolerate the partial shading by uppermost plants in a high tunnel environment. Our second hypothesis was that, compared to the uppermost level, containers on lower levels would have no substantially reduced arctic bramble plant vigour or fruit yield.

#### Materials and methods

#### **Experimental setup**

The cultivation experiment on arctic bramble cv. 'Alli' was conducted from June to October 2016 in a high polyethene tunnel under natural light (60°14'N 25°1'E). The experiment included four substrate treatments with six replicate plant towers for each, in total 24 towers arranged in three rows as randomised complete

block design where each row constituted a block with two replicate towers. Two additional towers were placed as buffers at each end of a row. Arctic bramble cv. 'Mesma', cv. 'Mespi' and the selection 154 were planted in the buffer towers to ensure pollination of the experimental plants.

The containers used were three-lobed 3.5 L containers (Jiangxi Bolai Plastic Industries, Yichun, Jiangxi, China) stacked six on top of each other. The base of the tower was elevated from floor level, setting the lowest planting level 85 cm and the highest 165 cm above floor level, allowing for the shoots to hang at convenient picking height. Spacing was 240 cm between the rows and 85 cm between the towers in a row. The containers were designed to drain excess water into lower planting levels, allowing irrigation flow from the top downwards. Irrigation of the towers was arranged via a shared drip system, with four drips placed in each of the two topmost containers and one in each of the four lower containers. The towers were fertigated with 1.4 mS cm<sup>-1</sup> Turve-Superex (Kekkilä, Finland, N-P-K 12-4.7-27). Fertigation was given for 2 min at a time, three times per day. In total 12 drips per tower were installed, one in the middle of each container and additional ones in each of the three lobes of the two uppermost containers.

Since the containers were stacked in alternating orientations, each draining two levels downward, the containers in each tower formed two parallel drainage flow systems with three levels each, as described by Tommila et al. (2022). These were labelled from top down as the Top, Middle and Bottom level, each consisting of two adjacent containers. Most data were collected and analysed using this set of two containers as an experimental unit. Thus, within blocks, the experiment was arranged as a split-plot design with the growth substrate as a main plot and the planting level as a split plot. Plant growth and yield data are presented per container.

The plant material was propagated by dividing the root system of container-grown, cold-stored  $(-2^{\circ}C)$  arctic bramble plants. The plants were divided into roughly equally sized units with approximately 20 suckers in each, and three units were planted in each container. Propagation and planting took place on 6 June, five days after dormant plant material was brought into the high tunnel to initiate growth. Pollination of flowers relied mainly on natural insects, assisted by a bumblebee hive (Minipol, Koppert Biological Systems, Romulus, MI, U.S.A.) during the first flowering in late June. Predatory mites *Neoseiulus cucumeris* and *Phytoseiulus perisimilis* (Biotus Oy, Forssa, Finland) were used to control thrips and spider mites in the tunnel. The harvest of fruit yield took place weekly from 29

July through 23 September (53–109 days from planting). The On 30 September (116 days from planting), the experiment was concluded with final growth and soil measurements.

#### Substrate treatments

As a control substrate treatment, we used 100% unfertilised peat (UP) (Luonnonturve, von Post 2-4, Kekkilä, Vantaa, Finland), thereafter termed UP. A mix of 80% UP and 20% perlite, thereafter termed UPP, was included to assess whether the perlite addition in a peat-based substrate would be useful for improving substrate aeration in the cultivation system used. Third, a commercial coir mix of 85% fine coir material and 15% cocofibre (art. 11. 1932, Legro, Helmond, Netherlands) was used as a 50% mix with UP, thereafter termed UPCoir. As a fourth treatment, Sphagnum moss was used as such, thereafter termed SM. The sphagnum moss (mainly S. fuscum, S. magellanicum, S. balticum and S. rubellum) was harvested as a 30 cm surface layer from the ombotrophic Neva-Lyly mire (Karvia, Finland) in 2014, air-dried, ground and sifted through a 40-mm sieve. According to the information from manufacturers, the pH of the peat was 4.2 and that of coir was 6.9, while the EC of peat was 4.0 mS  $cm^{-1}$  and that of the coir was 0.2 mS  $cm^{-1}$ .

Mixed substrates were prepared by measuring, by volume, dry, loose component materials into a through for a total 40 (UPCoir) or 50 (UPP) L of substrate at a time, and mixing by hand. The peat material was acquired in a lightly compressed state and loosened by hand before mixing, while other materials were acquired in a loose state. Just prior to mixing, 15 and 5 g L<sup>-1</sup> of fine-ground horticultural chalk (Aito Puutarha-kalkki, Nordkalk, Parainen, Finland) was added to peat and *Sphagnum* moss, respectively. After this preparation, the substrate was watered and mixed more to initiate water absorption in the pores. In planting, the containers were filled with wetted loose material, which was manually compressed very lightly around the plant and watered on the surface.

We aimed to adjust the pH of peat and *Sphagnum* moss close to that of coir, while assuming perlite would have no effect on the final substrate pH. The pH of the substrate materials was measured in 2-hour water infusion with 1 L of substrate in 1 L of tap water at room temperature (UltraBasic-10 Benchtop Meter, Denver Instrument, Bohemia, NY, U.S.A.), resulting in 3.4 for peat, 3.8 in *Sphagnum* moss and 6.2 in coir substrate. In order to estimate the necessary amounts of chalk, 5–20 g L<sup>-1</sup> was then added to samples of peat and *Sphagnum* moss and infused for 2 or 24 h before pH measurement.

#### Water retention of substrate materials

In planting, we did not aim for a specific substrate bulk density, but used a planting protocol that would result in minimal compression in all substrates. The same protocol, without the plants, was used in 3.5-litre reference containers to produce soil samples from which the water retention of three main substrate materials (UP, Sphagnum moss and 100% coir, thereafter referred to as UP, SM and Coir) was measured at -10, -30, -50 and -100 hPa matric suction in a sand box (08.01 Sandbox, Eijkelkamp Soil & Water, Giesbeek, Netherlands). The measurement was conducted as described by Dane and Topp (2002) and the soil sample preparation and the calculation of total pore volume were conducted as described by Tommila et al. (2022). Dry bulk densities used in the calculation (Table 1) were observed from the soil sample as done by Tommila et al. (2022) and the particle densities used were sourced from literature (Table 1). The pore space emptying between 0 and -100 hPa was divided into AV, EAV and WBC as per Gruda and Schnitzler (2004). The water retention data on coir was also used for substrate material comparisons in Tommila et al. (2022) and was first published there.

### Physical and chemical observations of the substrates

The pH and electric conductivity (EC) of drainage water from the plant towers were measured every four weeks from 6 July to 28 September. Water samples were taken from the total drainage volume collected from the Bottom unit of each tower after a single irrigation. The water samples were measured for the pH (UltraBasic-10 Benchtop Meter, Denver Instrument, Bohemia, NY, U.S.A.) and EC (Jenway 4020 Conductivity Meter, Cole-Parmer, Vernon, II, U.S.A.). Substrate water content was measured on 30 September from one plant tower per experimental block per substrate treatment (Water Content Meter, Grodan, Roermond, Netherlands), using the instrument setting for peat type substrates. The

**Table 1.** The measured bulk densities (g cm<sup>-3</sup>), presumed solid particle densities (g cm<sup>-3</sup>) and estimated solid particle and total pore volumes (% of substrate volume) of three primary substrate materials.

	Bulk density, g cm <sup>-3</sup>	Solid density, g cm <sup>-3</sup>	Solid volume %	Pore volume %
Peat	0.068	1.5 <sup>1)</sup>	4.5	95.5
Moss	0.031	1.4 <sup>1)</sup>	2.2	97.8
Coir	0.062	1.4 <sup>2)</sup>	4.4	95.6

<sup>1)</sup>Kämäräinen et al. (2018).

<sup>2)</sup>Gruda and Schnitzler (2004).

measurement was made within two hours of an irrigation cycle, in cool humid autumn conditions to assess the maximum water content retained in each substrate after irrigation. One measurement was made from each container lobe and the means of six measurements (the lobes of the two containers within an experimental unit) were used in statistical analysis.

#### Plant growth observations

All fully or partially developed fruits of generally marketable quality were included in fruit yield, and their number and total weight per experimental unit were recorded. The number of unmarketable (dried or mouldy) berries was recorded separately from marketable yield. The number of drupelets in berries was counted from two samples that included all berries harvested from 4 to 11 August and from 1 to 8 September, to calculate the average drupelet weight. Mean fruit weight was calculated separately for the whole yield and for the samples used for drupelet counts. On 30 September, all flowers that had not developed into fruits were collected and counted. Fruit set for the total number of flowers (harvested and spoiled fruits plus undeveloped flowers) per container was calculated. The remaining aboveground shoot mass, referred to as vegetative growth, was harvested on 30 September, dried for three days at 60°C and weighed.

#### Statistical analysis

Data were subjected to analysis of variance (ANOVA) using ANOVA procedure of SAS. Means were separated using Tukey's Studentized Range (HSD) test. Results are presented as the arithmetic mean, with positive and negative standard error. For ANOVA, data on fruit set and fruit spoilage were subjected to Arcsine transformation.

#### Results

#### Water retention of the substrate materials

Both volumetric water retention (Figure 1(A)) and mass ratio of retained water to substrate dry matter (Figure 1(B)) differed between substrate materials at all matric tension levels (p < 0.001). In SM, volumetric water content was consistently lower than in other materials (Figure 1(A)), while water mass ratio was substantially higher (Figure 1(B)). The differences between Coir and UP were small at all matric tension levels (Figure 1(A,B)).

The estimated volume of air-filled pores at -10 hPa was substantially higher in SM (36%) compared to coir



**Figure 1.** Water content of Coir, unfertilised peat and *Sphagnum* moss (SM) in sand box at 0, -10, -30, -50 and -100 hPa matric tension, as percentage of the total substrate volume (A) and as total water per gram of dry matter (B). Different lower case letters indicate statistically significant differences between the substrate materials at each matric tension level (p < 0.05) by Tukey's test. Values are means of three replicates. Vertical bars present  $\pm$  SE.

(29%) and UP (26%). The capacity of EAV, defined as the pores emptying between -10 and -50 hPa, was smaller in coir (25%) compared to SM (31%) and UP (32%). The WBC, defined as the pores emptying between -50 to -100 hPa, was relatively small in all substrates.

### Physical and chemical observations of the substrates

Water content in the containers in plant towers at the end of growth season was affected by substrate (p < 0.001) and tower level (p < 0.001), with no interaction between factors (Figure 2). Mean water content, including all levels, was higher in UP (66%) and lower in SM and UPP (56 and 55%, respectively) (Figure 2). Including all substrates, water content was highest on Top level (64%) and lower on Middle and Bottom level (58 and 56% respectively). In UPCoir and SM, water content was higher on Top level and reduced by 7–15 percent points on either Middle or Bottom level, respectively (Figure 2).

Mean drainage water EC was similar to the fertigation solution in SM (1.5 mS cm<sup>-1</sup>) and lower in other substrates (1.0–1.2 mS cm<sup>-1</sup>) at the first observation on 6 July, but increased in all substrates by the second observation date on 3 August, and remained on similar levels (1.6–2.3 mS cm<sup>-1</sup>) on 31 August and 28 September. The EC remained consistently lower in UPP than in SM. Mean drainage water pH on July 6 was higher than expected in all substrates (7.0–7.4), but decreased by 3 September (6.5–6.9) and 28 September (6.4–6.9). There were no consistent differences in pH between the substrates on observation dates.



**Figure 2.** Water content in different substrates and tower levels in arctic bramble plant towers on 30 September (16 weeks from planting), as percentage of the total substrate volume. Different upper case letters indicate statistically significant differences between the substrates (p < 0.05) by Tukey's test. Different lower case letters indicate statistically significant differences between the levels within each substrate (p < 0.05) by Tukey's test. Values are means of three replicates. Vertical bars present ±SE.

#### Vegetative growth

Vegetative growth, measured as the amount of aboveground vegetative dry matter, was affected by substrate (p < 0.001) and tower level (p < 0.001), with no interaction between factors (Figure 3). Between the substrates, vegetative growth per container ranged from 43.6 g in UPP to 67.0 g in SM (Figure 3). When comparing UP to other substrates, it is notable that SM produced 32% more dry matter, while UPP and UPCoir did not differ from the 50.9 g in UP (Figure 3). Between tower levels, there was a 55% reduction from 92.8 g on Top level to 42.1 g on Middle level and 67% reduction from Top level to 30.2 g on Bottom level (Figure 3).

#### Fruit yield and fruit set

Arctic bramble fruit yield was affected by substrate and tower level, with no interaction between factors. The yield per container was distinctly higher on SM than on UPP and on Top level compared to lower levels (Table 2). Comparing other substrates to UP, the yield was increased by 49% on SM and by 14% on UPCoir, and reduced by 20% on UPP (Table 2). Comparing lower tower levels to Top level, the yield was reduced by 31% on Middle level and by 52% on Bottom level (Table 2).

Number of fruits per container was likewise affected by substrate and tower level (Table 2), while mean fruit weight was only affected by substrate (Table 3).



**Figure 3.** Aboveground vegetative dry matter per container of arctic bramble plants grown on different substrates and levels in plant towers in a high tunnel. Different upper case letters indicate statistically significant differences between the substrates (p < 0.05) by Tukey's test. Different lower case letters indicate statistically significant differences between the levels within each substrate (p < 0.05) by Tukey's test. Values are means of six replicates. Vertical bars present ±SE.

**Table 2.** The mean fruit yield and number of flowers per container, fruit set as percentage of flowers, fruit spoilage as percentage of set fruits and number of harvested fruits per container of arctic bramble plants grown on different substrates and levels in plant towers in a high tunnel. Values are means of six replicates, followed by  $\pm$  standard error. Different lower case letters indicate statistically significant differences between the means within each factor by Tukey's test (p < 0.05). n.s = not significant.

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Factor	Level	Fruit yield, g per container	Nr of flowers per container	Fruit set %	Fruit spoilage %	Nr of harvested fruits per container
Substrate	UP	21.1 ± 3.6 ab	67 ± 8 ab	82±2 a	54±4 a	26.8 ± 3.9 ab
	UPP	17.9 ± 3.8 b	59 ± 10 b	85 ± 2 a	53 ± 2 a	24.4 ± 4.4 b
	UPCoir	26.8 ± 3.3. ab	63 ± 7 ab	88±1a	45 ± 2 a	29.5 ± 3.0 ab
	SM	32.8 ± 3.2 a	81±8 a	85 ± 2 a	45 ± 2 a	36.4 ± 3.0 a
Level	Тор	35.8 ± 3.0 a	108 ± 6 a	87±2a	56±1a	42.1 ± 2.8 a
	Middle	22.7 ± 2.9 b	54 ± 4.0 b	85 ± 1 ba	46 ± 2 b	26.1 ± 2.7 b
	Bottom	15.5 ± 2.1 b	41 ± 3 b	83 ± 2 b	45 ± 3 b	19.6 ± 2.2 b
р	Substrate (S)	0.007	0.025	n.s.	0.016	0.034
	Level (L)	<0.001	<0.001	0.045	0.004	<0.001
	$S \times L$	n.s.	n.s.	n.s.	n.s.	n.s.

Compared to UP, the high-yielding substrate SM had a substantially higher number of fruits (by 35%), while both SM and UPCoir had a substantially higher fruit weight (by 19 and 24% respectively) (Table 2). Number of flowers per container was affected by both substrate and tower level (Table 2). Compared to UP, SM had a substantially higher number of flowers (Table 2). Between tower levels, the number of flowers was much lower and fruit set was slightly lower on Middle and Bottom level than Top level, resulting in lower yield despite lower fruit spoilage rate (Table 2).

#### Harvest accumulation and fruit weight

Highest weekly harvests were picked on 29 July (53 days from planting) and 1–16 September (87–102 days from planting) (Figure 4A). Differences between substrates remained relatively small until late August and became more substantial in early September (Figure 4A). While there was no pause between the first and second harvest phase, weekly harvests increased considerably between 25 August and 8 September on UPCoir and SM, where the total yield ended up being the highest (Figure 4(A), Table 2). Thus, on higher-yielding substrates, early harvest contributed a smaller portion of the eventual yield. On 25 August, 64% of the total yield had accumulated on UPP and 56% on UP, but only 41% on both UPCoir and SM. On 29 July, 29% of the eventual yield had accumulated on UPP, but only 10% on SM.

Mean fruit weight in weekly harvest batches varied through the harvest season, being lowest from 4 to 11 August and highest from 1 to 9 September (Figure 4 (B)). Thus, we combined these four batches into two fruit samples to further examine the number of drupe-lets as an indicator of pollination success and as a factor affecting fruit weight. The mean fruit weight of all substrate treatments was 0.84 g for the total yield, 0.70 g for the early harvest sample (4–11 August) and 1.01 g for the late harvest sample (1–8 September).

#### Fruit structure

In the two fruit samples used for structural analysis, mean fruit weight increased from early to late harvest by 19–31%, depending on substrate, and the mean number of drupelets per fruit increased by 23–30%, while there was little or no change in mean drupelet weight (Table 3). In both samples, fruit weight was affected by substrate but not by tower level (Table 3).

**Table 3.** The mean fruit weight, number of drupelets per fruit and drupelet weight in fruit samples collected from 4 to 11 August (harvest phase 1) and from 1 to 8 September (harvest phase 2) from arctic bramble plants grown on different substrates and levels in plant towers in a high tunnel. Values are means of six replicates, followed by  $\pm$  standard error. Different lower case letters indicate statistically significant differences between the means within each factor by Tukey's test (p < 0.05). n.s = not significant.

Factor	Level	Fruit weight, g		Nr of drupelets per fruit		Drupelet weight, mg	
		Phase 1	Phase 2	Phase 1	Phase 2	Phase 1	Phase 2
Substrate	UP	0.66 ± 0.03 bc	0.86 ± 0.06 ab	20.4 ± 1.3 a	26.5 ± 1.4 b	34 ± 2 a	32 ± 1 b
	UPP	0.56 ± 0.02 c	0.78 ± 0.05 b	20.8 ± 1.0 a	29.1 ± 1.5 ab	28 ± 1 b	27 ± 1 c
	UPCoir	0.78 ± 0.04 a	0.97 ± 0.05 ab	22.9 ± 0.9 a	31.9 ± 1.4 a	34±1 a	30 ± 1 bc
	SM	0.71 ± 0.03 ab	1.02 ± 0.04 a	19.3 ± 0.9 a	27.7 ± 0.8 ab	37 ± 2 a	37±1a
Level	Тор	0.65 ± 0.03 a	0.96 ± 0.04 a	18.8 ± 0.7 b	29.4 ± 1.0 a	35 ± 1 a	33±1a
	Middle	0.73 ± 0.03 a	0.95 ± 0.04 a	21.8 ± 1.1 a	29.4 ± 0.9 a	34 ± 2 b	33±1a
	Bottom	0.66 ± 0.03 a	0.82 ± 0.06 a	21.9 ± 0.8 a	27.6 ± 1.6 a	30 ± 1 b	30±1a
Ρ	Substrate (S)	< 0.001	0.008	n.s.	0.040	<0.001	< 0.001
	Level (L)	n.s.	n.s.	0.018	n.s.	0.032	n.s.
	S×L	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.



**Figure 4.** Weekly fruit yield per container (A) and mean fruit weight (B) over the harvest season of arctic bramble plants grown on different substrates in plant towers in a high tunnel. Values are means of six replicates. Vertical bars present  $\pm$ SE.

On SM, the fruit weight was consistently higher than on UPP (Table 3). The substrate also affected drupelet weight, which was consistently lower on UPP than on UP or SM (Table 3). The number of drupelets was affected by the substrate during late harvest, but not in a way obviously connected to the fruit weight. Tower level had an effect during early harvest, when the number of drupelets was lowest on Top level and drupelet weight was lowest on Bottom level (Table 3), though neither of these effects translated to fruit weight.

#### Discussion

This study combined two highly topical issues; the use of *Sphagnum* moss as growth substrate to substitute peat and multiple planting levels to increase space use efficiency in protected cultivation. Our results show that a pure *Sphagnum* moss growth substrate performed at least as well for arctic bramble as peat-based substrates. With regard to our first hypothesis, plant vigour was higher in SM compared to UPP, while fruit yield was higher in SM compared to UPP. Aeration, defined as volume of air-filled pores at -10 hPa, was higher in *Sphagnum* moss than in peat

or coir, without compromising the capacity of EAV, defined as the reduction of water content between -10 and -50 hPa (Figure 1(A)). With regard to our second hypothesis, on the plant tower arrangement, both plant vigour and fruit yield were substantially reduced on Middle and especially Bottom level compared to Top level. Thus, we consider arctic bramble to be poorly suited for multi-level cultivation, at least in this particular system.

In recent years there has been rapid development of processed, peat and *Sphagnum* moss based growth substrate mixes for container cultivation. Michel (2010) notes that while all available substrate materials are less than ideal in some respects, peat and peat-based mixes remain essential in combining good water retention and good aeration. However, Michel (2010) also acknowledges that especially the aeration of peat substrates can be improved by mixing other materials. Rivière et al. (1990) distinguish four types of growth substrate, based on AV content (aeration) and water availability: (I) aerated with high water availability and high water buffering capacity (II) less aerated (III) highly aerated with low water availability (IV) aerated with high water availability but low buffering capacity. Without accounting for Sphagnum moss, Michel (2010) estimates that only white peats fit in type I while more decomposed peats and fine coir material represent type II. In this scheme, type III includes various inorganic and coarse organic materials, and only rockwool fits in type IV (Michel 2010). Sphagnum moss is the natural precursor of white peat, with a relatively complicated pore structure, and its physical properties (see Kämäräinen et al. 2018) suggest it would fit in type I or type II of Rivière et al. (1990). Overall, all substrate materials used in this study had relatively similar water retention profiles in terms of volumetric water content, at matric suction levels suitable for cultivation (Figure 1(A)). In terms of water mass ratio, Sphagnum moss retainer much more water compared to coir and peat (Figure 1 (B)) due to its lower bulk density.

Kämäräinen et al. (2018) demonstrated a higher capacity of plant available water in Sphagnum moss biomass compared to light peat at similar bulk densities. In our study, the bulk density of Sphagnum moss in reference containers was roughly half of that of other substrate materials (Table 1). Measuring the water retention of the materials in these bulk densities, we found the AV higher in Sphagnum moss compared to other materials, and the capacity of EAV similar to peat and slightly lower compared to coir (Figure 1(A)). Similarly, water content measured from the plant towers was lower in Sphagnum moss and peat-perlite mix compared to peat or peat-coir mix (Figure 2), suggesting higher air content assuming near-equal amounts of total pore space. Although natural substrate compression in this experiment may have resulted in higher bulk densities in plant towers than were observed in the reference containers, the water content measurements are compatible with better aeration in Sphagnum moss compared to other substrates (Figure 1(A)). This improved aeration could significantly improve growth conditions by increasing root oxygen supply, although the sensitivity for root oxygen stress varies between species.

The effects of substrate mixing on bulk density and water retention, as well as substrate performance on specific crops, are difficult to predict. Emmel (2008) found *Sphagnum* moss and its mixes with peat or peat and clay were all generally highly suitable for ornamental plants, although some *Sphagnum* types had a harmful effect on seedlings. In Oberpaur et al. (2010), *sphagnum* moss mixes with either varying contents of composted pine bark or 40% content of compost or humus had physical and chemical properties close to peat, but only the two latter mixes performed well as growth substrate. Kämäräinen et al. (2020) found peat-*Sphagnum* moss mixes to improve the growth of sweet

basil (Ocimum basilicum) and verbena (Verbena × hybrida) compared to both light peat and pure Sphagnum moss material, while acknowledging that the result cannot be extrapolated to all plant species due to different root aeration requirements. Further, Kämäräinen et al. (2020) propose that a superior combination of aeration and EAV in pure Sphagnum moss could be achieved if the moss material is cultivated at a lower bulk density than is usual with peat, adjusted optimally for the plant species, cultivation system and irrigation regime. Our results on cultivating arctic bramble on minimally compressed substrates supports this hypothesis, as the plant growth was more vigorous and the AV higher in Sphagnum moss compared to peat, which in our experiment had substantially higher bulk density. Pure Sphagnum moss has naturally a lower bulk density than peat, and its density could be easily adjusted further with controlled compression.

In this study, we aimed to provide the same irrigation regime for all substrate treatments and, if possible, for all tower levels. This goal was complicated by the unequal growth of the plants, resulting in higher transpiration particularly on Top level. Although there was no continuous monitoring of soil water content during the growth season, we visually estimated that the plants on Top level often suffered from some degree of drought stress between irrigation cycles, while the substrate on lower levels seemed to remain relatively wet. However, when the maximal water content retained in the plant tower units was measured in cool, humid autumn conditions, it was the same or higher on Top level compared to the lower levels (Figure 2).

The commercial plant tower system used in this study is designed to fertigate the lower tower levels in part by utilising drainage water and leached nutrients from the upper levels, which makes the distribution of water and nutrients unpredictable and difficult to manage. In this setting, growth conditions on different levels in a high tunnel can be expected to differ in light, water and nutrient availability, as well as air temperature and relative humidity. The major differences between tower levels in vegetative growth, flowering and fruit yield might have been caused by scarcity of light on the lower levels, or by nutrient sequestration on the upper levels. We have previously found sequestration of nutrients a problem in similar plant tower arrangements (unpublished).

While we attempted to control for the pH and fertiliser use in different substrates, this approach may be inherently in conflict with the goal of comparing the best possible application of each substrate material. In Tommila et al. (2022), the comparison between horticultural peat and coir substrate was deemed problematic due to the short cultivation period and the start fertiliser included in the peat. In the current study, since we couldn't acquire the coir and Sphagnum moss materials with start fertilisation comparable to common horticultural peat substrates, we chose an unfertilised, unlimited peat product. The application of lime in peat and Sphagnum moss just prior to planting, which was intended to adjust the pH to the level of coir, inevitably required differing amounts of lime, thus affecting the availability of calcium in the substrate. The pH 6.2 initially measured for the coir material would be only slightly higher that the average pH 5.9 of Finnish soils used in arctic bramble field cultivation (Kokko et al. 2012). Ultimately, due to the lack of a well-tested protocol, the liming resulted in higher than expected drainage water pH levels in all substrates especially during early growth. However, based on drainage water analyses, the pH levels of all four substrates were close to each other. While the optimal pH range for arctic bramble is not known, Ryynänen (1973) estimated the species to be widely tolerant of both acidic and neutral pH levels.

The fruit size of the aggregate fruits of *Rubus* spp. depends on the number and size of drupelets, each of which develops from a single pollinated carpel. In this study, fruit size in general was smaller than expected, especially during the first harvest phase. In our previous experiments on cv. 'Alli' (unpublished), we have observed that fruit weight of ~1.5 g is possible in protected cultivation, while in field cultivation (Kostamo et al. 2013), fruit weight of  $\sim 1$  g could be expected. However, in Tommila et al. (2022), fruit weight of cv. 'Alli' was similar to the current study, but with a smaller number of larger drupelets. In our experience, relatively high drupelet weight is a common trait in protected arctic bramble cultivation, compared to field cultivation, and may result in high fruit weight if successful pollination produces a sufficient number of drupelets.

In this study, the differences in drupelet weight between the substrates were reflected by the differences in fruit weight, notably between peat-perlite mix and Sphagnum moss, of which the latter had consistently higher drupelet weight and fruit weight (Table 3). Plants grown on Sphagnum moss also had a much higher amount of vegetative growth (Figure 3) and larger numbers of flowers and harvested fruits (Table 2). Therefore, we conclude that the drupelet weight and associated fruit weight may have been limited by the general plant vigour. However, in this case, a higher drupelet weight could have been expected on Top level compared to lower tower levels, which was not the case (Table 3). It is possible that the plants and developing fruits on Top level were exposed to water stress during summer days, due to higher leaf area and transpiration and insufficient water reserve, which could have reduced drupelet and fruit size.

During the first harvest phase, fruit weight was also reduced by the relatively low number of drupelets. While there was no difference in drupelet weight between the first and second harvest phase, the number of drupelets increased substantially by the second phase, especially on Top level where the number of drupelets was initially lower than on Middle or Bottom levels (Table 3). The factors affecting arctic bramble pollination and fruit set are poorly understood, but the lower number of drupelets per fruit during the first harvest phase suggests less successful pollination.

Overall, pollination success in arctic bramble is highly variable and low fruit set and incomplete fruit development are common problems (Vool et al. 2009; Kokko et al. 2012; Kostamo et al. 2018). In this study, both fruit set (Table 2) and the number of drupelets in 'Alli' (Table 3) were considerably higher than we observed previously in a greenhouse (Tommila et al. 2022), suggesting better pollination conditions. The bumblebee hive used in the tunnel appeared to have become inactive by the time of second flowering, but wild bumblebees were abundantly present. It has been proposed (Hiirsalmi 1975) that arctic bramble pollination may be favoured by moderately high air relative humidity, and affected negatively by the low relative humidity that typically occurs on warm sunny days early in the flowering season. Hiirsalmi (1975) found that the combination of misting and placing a beehive in a plastic high tunnel produced the highest yield on a relatively dry summer, but on a wet summer misting made no difference. In our previous greenhouse experiment on 'Alli', we found that fruit set was higher at 60% relative humidity compared to 40 or 80% (unpublished). In this study, pollination may have been negatively affected by high temperatures and low relative humidity during the first flowering, particularly on Top level. At the end of the second harvest phase, fruit weight decreased again considerably (Figure 4(B)), while the weekly harvest came close to zero (Figure 4(A)), as is usual in our experience, due to poor pollination success between the season's last few flowers.

Other than pollination, it is not known which factors might affect the fruit set or drupelet count in arctic bramble. Jean and Lapointe (2001) found that poor carbohydrate supply can cause fruit abortion and reduce the number of developing drupelets in cloudberry, which largely relies on rhizome carbohydrate reserves for fruiting, but to our knowledge this has not been studied on arctic bramble. Drying or other spoilage of developing fruits is a common problem in arctic bramble, caused by various fungal diseases and possibly other, poorly known factors (Kokko et al. 2012; Kostamo et al. 2015). While in our previous greenhouse experiment the number of spoiled fruits was small (Tommila et al. 2022), in this study the amount of yield loss by spoilage was remarkably high (Table 2). Overall, it is likely that the one-week interval between pickings was too long in the warm humid environment of the plastic tunnel, resulting in overripening and spoiling of fruits that otherwise would've been marketable.

In conclusion, we found that pure Sphagnum moss is a promising option for arctic bramble, potentially outperforming peat-based substrates. However, further research is necessary so that the irrigation and fertilisation regimes can be optimised for the unique properties of this material with arctic bramble, as with other specific crops. Likewise, in order to assess the cultivation properties of different substrate materials, the irrigation and fertilisation regime would have to be both better controlled and separately optimised for different materials. Different irrigation properties of substrate materials might also require adjustment in other aspects of container cultivation technology, such as shifting towards new standards in container architecture, to maximise lateral diffusion and dispersion of water in the substrate. Finally, we note that investment in climate control and frequent picking can be highly important in arctic bramble protected cultivation to prevent yield loss due to spoilage of developing and ripening fruits.

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