Review

Overview of a Lab-scale Pilot Plant for Studying Baby Leaf Vegetables Grown in Soilless Culture

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Investigating several environmental factors affecting plant growth implies having sound experimental facilities equipped to test individual factors in lab-scale although applicable later at the industrial scale. Sometimes, detailed information is hardly given in a manuscript that allows for replications by other authors, maybe due to the shortening of pages requested by journal publishers and editors. A system and methodology was developed for qualitative and quantitative analyses of baby leaf vegetables (BLV) raised in floating growing systems (FGS). Lab-scale pilot plants (LSPP) were developed in 2 greenhouses differing in structure and equipment, suitable for different growing seasons in a continental climate. The equipment and technology allowed multiple treatments and replicates for sound statistical design and data analyses. Environmental conditions and cultural techniques were studied in major and minor species (white mustard, Brassica alba L. Boiss; black mustard, Brassica nigra L. Koch; garden cress, Lepidium sativum L.; water cress, Nasturtium officinale R. Br.; rocket salad, Eruca sativa Mill.; perennial wild rocket, Diplotaxis tenuifolia L. DC.; corn salad, Valerianella olitoria L.; baby spinach, Spinacia oleracea L.) to determine best cultivation techniques in a standard soilless culture system (SCS) for BLV, based on FGS. Considering that SCS can improve raw material quality at harvest, and enhance the postharvest shelf-life of many vegetables and herbs, a standardized growing system is required to obtain premium quality raw material in terms of commercial stage, low nitrate content and long shelf-life. Among the SCS used, the FGS are suitable systems to grow leafy vegetables because the plants can grow at high densities, thereby producing high yields, and in a short time. FGS are based on sub-irrigation technology, avoiding over-head irrigation and contact between nutrient solution and edible parts, and result in greater qualitative and quantitative yields than the traditional cultivation techniques, reducing pollution, crop and substrate residues, leading to clean raw material with potential low microbiological load. The FGS is a modern technology that could be exploited more to enhance yield, quality and safety of fresh and fresh-cut BLV. The LSPP installed are providing the basis for expanding the research to other species and agronomic factors.

Key Words: deep flow systems, floating growing systems, growing efficiency, LSPP, raw material quality.

Introduction

Innovative soilless growing systems

Innovative growing systems and technologies have been developed to enhance raw material production, guarantee safety, diversify the fresh produce available to consumers and prolong shelf-life (Fontana and Nicola, 2008, 2009). Protected cultivation is increasingly shifting from traditional culture systems in soil to soilless culture systems (SCS) (Nicola and Fontana, 2007). Among different SCS, the floating growing system (FGS) is a sub-irrigation system that consists of trays floating on a water bed or hydroponic nutrient solution (HNS) (Fontana and Nicola, 2008; Pimpini and Enzo, 1997; Thomas, 1993). FGS can be considered an efficient system for producing leafy vegetables with high added value, processed as fresh-cut produce, satisfying the requirements of the production chain (Nicola...
et al., 2009). FGS is suitable for producing uniform raw material, with short production cycle, and high plant density (Nicola and Fontana, 2014; Selma et al., 2012). FGS increases the precision of fertilizer application to plants by reducing water leaching during irrigation. For managing mineral plant nutrition, tissue ion accumulation can be controlled by varying the macroelement-ratio in the HNS or its composition (Fontana and Nicola, 2008; Santamaria and Elia, 1997). FGS reduces microbial contamination, and eliminates soil and chemical residue spoilage allowing softer washing procedures, such as eliminating chlorine from the water sanitation process, resulting in less stress for the leaf tissue (Fontana and Nicola, 2008; Nicola and Fontana, 2014; Scuderi et al., 2011). FGS can be implemented either with a continuous flotation system (FL) or with an ebb-and-flow system (EF). EF is scheduled with drying (ebb) periods to avoid hypoxia, the situations in which the oxygen concentration is a limiting factor for the plant growth (Morard and Silvestre, 1996; Nicola et al., 2007). Previous studies comparing FL and EF irrigations to grow baby leaf vegetables (BLV) in individual tray per water bed indicated that FL gave greater leaf fresh mass production in corn salad (Valerianella olitoria L.) and in rocket salad (Eruca sativa Mill.), although leaf nitrate accumulation was higher (Fontana and Nicola, 2008; Nicola et al., 2003). In both cases the HNS was applied once a week. Some BLV grown in FL may rapidly deplete oxygen dissolved in the HNS as a result of root respiration (Marfà et al., 2005). To avoid the negative effect of hypoxia on plants, growers schedule drying periods with EF, or aerate the HNS to enrich it with oxygen to ensure the growth and the functionality of the roots (Lara et al., 2011; Tesi et al., 2003). In general, HNS aeration increases some vegetative growth parameters of the shoots, e.g., number of leaves, leaf area and fresh or/and dry matter, with respect to non-aerated conditions (Nicola et al., 2015; Niñirola et al., 2011, 2012; Tesi et al., 2003).

Information on experimental plants for SCS

Leafy vegetables represent an important sector of horticultural production, and cover around the world a multitude of species. Iceberg lettuce is one of the main ingredients of salad mixes, but at present other types of lettuce and BLV are increasingly consumed as fresh-cut product, sole or as part of salad mixes (Martínez-Sánchez et al., 2012). Major species used are represented by many lettuce types and cultivars (Lactuca sativa L.), chicory species and cultivars (Cichorium intybus L.; Cichorium endivia L.), corn salad, cresses (water cress, Nasturtium officinale R. Br.; garden cress, Lepidium sativum L.), rocket (rocket salad; perennial wild rocket, Diplotaxis tenuifolia L. DC.), baby spinach (Spinacia oleracea L.), chards (Beta vulgaris ssp. vulgaris; Beta vulgaris ssp. cicla), baby leaf mustard, Brassica alba L. Boiss; black mustard, Brassica nigra L. Koch; red mustard, Brassica juncea L. Czern; mizuna, Brassica rapa L. ssp. nipposinica). In Italy, 100000 metric tons of BLV are produced annually, of which 60000 metric tons are exported (Della Casa and Dall’Olio, 2013, http://adm-distribuzione.it/wp-content/uploads/2014/10/Marca-2013-prof-Della-Casa.pdf, September 7, 2015). Annually, 91000 metric tons of fresh-cut salads are produced in the UK, 69000 metric tons in France, and 43000 metric tons in Germany (Zucconi, 2014, http://www.freshpointmagazine.it/EN/quarta-gamma-italia-prima-per-incidenza-sul-totale-ortofrutta, September 7, 2015). Although SCS are the most intensive production systems in today’s horticulture industry (Gruda, 2009), little is known about the economics of the different types of growing systems and in every location, that would allow understand the feasibility of their expansion at the commercial level, given the increasing demand for BLV. Detailed investigations are required on each single species to obtain Good Agricultural Practices for all of them, and possibly a standardized growing system for several species. Investigating several environmental factors affecting plant growth implies having sound experimental facilities equipped to test individual factors in lab-scale although applicable later at the industrial scale. Sometimes, detailed information is hardly given in a manuscript that allows for replications by other authors, maybe due to the shortening of pages requested by journal publishers and editors. In a few articles detailed indications are given on the technical preparation and management of an experiment in SCS (Karam and Al-Daoood, 2005; Marfà et al., 2005; Rouphael et al., 2005), but little information is available about the description of the experimental sizing (Frezza et al., 2005; Huber et al., 2005; Sato, 2005; Thompson et al., 1998). Only a few authors such as López-Galarza et al. (2005) report indication about the suppliers, and some articles have details of the system also with the graphic scheme (Bar-Yosef et al., 2005; Karam and Al-Daoood, 2005; Marfà et al., 2005).

To study the effect of a growing system and test several treatments on BLV, it is important to have a standardized, homogeneous and replicable system in which most factors and inputs can be monitored. Consequently, we decided to build a lab-scale pilot plant (LSPP) based on FGS in order to have a representative and reliable experimental system. The experimental pilot system was built in scale with respect to a commercial greenhouse growing system. Sizing was studied to cope with the necessity of both obtaining a reliable amount of data and keeping the scientific precision of each trial. The LSPP was adjusted and enhanced according to the results and experiences from the trials, and it has been designed for further experimental implementations.
Description of the LSPP for FGS

Greenhouse setting

The pilot plant design was established in the Experimental Centre of the Department DISAFA (44°53’11.67”N; 7°41’7.00”E – 231 m a.s.l.) in Tetti Frati, Carmagnola (TO), Italy. In the location, commercial plastic and glass greenhouses are present, equipped with an automatically controlled heating system with homogeneous diffusion of the hot air into the environment. Greenhouses are equipped with an automatically controlled opening system to provide aeration. During the warm season, the greenhouses are covered with black shading systems, with a 50%-shade cloth. Greenhouses are equipped with water pipelines for easy and local access for HNS preparation. A plastic house is used for the nursery stage, and is equipped with an automatically controlled over-head irrigation system, while 2 glasshouses were dedicated to set 2 LSPP to accommodate several experiments according to the season.

The nursery plastic house has an internal area of ca. 210 m², and is equipped with 4 benches reaching a total bench area of ca. 65 m². Each glasshouse used for setting the LSPP has an internal area of ca. 125 m², with 3 benches each reaching a total bench area per glasshouse of ca. 42 m².

Water beds and FGS

Each bench of the LSPP has been split into 4 separated sealed water beds (2.50 m × 1.40 m; 0.15 m depth) to allow 4 treatments per bench, varying according to each experiment (e.g. HNS concentration, HNS aeration, species). Each bench is considered as a block, differing according to sun exposition (South, center, North). The design of the LSPP unit is in Figure 1, with the representation of one single treatment.

The creation of the sealed water beds on the benches has been obtained using a double layer of a white/black (upper/lower site) plastic film (Fig. 2). The white upper site serves the double purpose of light reflection and reduction of the heating effect (Siwek et al., 1994). Each water bed can hold up to 16 styrofoam cell trays (0.5 m × 0.3 m), each one to represent the experimental unit per each block. The space between the trays is covered with polystyrene strips in order to prevent the algae formation when beds are filled with the HNS and the experiments are running (Fig. 3). Algae growth, affected by light availability, is a common problem in FGS, decreasing yield and clogging the supply lines, as in other SCS (Coosemans, 1995). Moreover, the totally covered surface reduces as much as possible the HNS evaporation. According to the experimental design of each trial, reducing the number of trays floating on the water beds is possible by replacing the trays themselves with polystyrene tables.

Each water bed has been equipped with a fixed and independent HNS inlet/outlet system. HNS of different compositions is prepared at the start of each experiment in separated tanks (Fig. 1, blue path) that feed the beds by connecting tubes. Several pumps allow for filling with the HNS more than one bed at the same time according to treatments and random locations of each replication. During plant growth, HNS can be refilled by new HNS stocked in reservoirs connected to the pipeline (Figs. 1 and 4). Each reservoir is internally

Fig. 1. Scheme of the lab-scale pilot plant (LSPP). Blue and red paths are represented for a single treatment; the specific path has been randomly chosen for representation. Blue circuit refers to hydroponic nutrient solution (HNS) flow; red path refers to aeration flow.

Fig. 2. View of the benches and the water beds equipped for using a floating growing system (FGS) to grow baby leaf vegetables (BLV) in one glasshouse.
equipped with an air pump that at scheduled frequency and intensity stirs the internal HNS, to avoid the salt deposition, and to increase its aeration.

In each water bed 2 outlet devices have been installed enabling flow control of the HNS depending on the set of the experiment. A common sink drain stopper has been installed to be used for discharging the HNS only at the end of the experiment (Fig. 5A). Under this setting, a FL can be used for growing the BLV either under steady HNS or aerated HNS (see paragraph on aeration). A second outlet device has been placed with an overflow system that allows to control the level of the HNS and enable a flow-and-flow (FF) mechanism (Fig. 5B). Indeed, this control occurs by the natural fall of the HNS through the overflow column via the regulation of the column height. This system allows for implementing a recirculating flow collecting the HNS from each bed in the drainage collection tanks from where pumps control the HNS flow to re-enter into the water beds through faucets (Fig. 1). The FF system can be temporized and the HNS can be heated or cooled with the addition of temperature-controlling devices, according to the experimental setting.

The HNS composition and concentration has been defined according to results obtained in previous experiments conducted by the research group in Vegetable Crops & Medicinal and Aromatic Plants – VEGMAP (Fontana and Nicola, 2008; Nicola et al., 2004, 2007; Pignata et al., 2015). The HNS tested for all species had 6 N, 2 P, 6 K, 2 Mg, and 2.5 Ca (all in mmol·L⁻¹), with a 40/60 N-NO₃⁻/N-NH₄⁺ ratio. For water cress, different levels of N-P-K were introduced in some studies (Table 1 and Pignata et al., 2015). HNS was prepared by dissolving in tap water nutrients with a purity > 98% plus micronutrients.

The pH of the HNS was steadily monitored and kept close to 5.5 using acid or basic solutions in order to neutralize salts, avoiding the immobilization or the precipitation of salts itself, increasing the availability for plants (Amiri and Sattary, 2004; De Rijck and Schrevens, 1998).

Aeration treatment

The implementation of the aeration treatment took place after literature review, making technical improvements and tailored to our system. In our previous experiments related to EF (Nicola et al., 2004, 2010), this system was firstly obtained manually, lifting the styrofoam trays, and suspending them on planks above the HNS. This manual system, despite the positive results (data not shown), has some limits related to: the necessity of the continued presence of manpower to move the styrofoam trays according to the schedule; the time needed for the operations; the care required to avoid damages to the styrofoam trays and the plants; and the possibility to increase the microbial contamination due to the manipulation.

Starting from the publication in which EF and FL were simultaneously tested (Nicola et al., 2007), different aeration systems combined with FL, alternative to EF, have been studied. Before being implemented in LSPP, each system studied and its technical evolutions have been previously tested in a small scale prototype. The best solution has been found by creating a wide-
## Table 1. Summary of major treatments, growing cycle and season in relation to 2 methods of computing yield of baby leaf vegetables (BLV) grown in floating growing systems (FGS) using a lab-scale pilot plant (LSPP).  

<table>
<thead>
<tr>
<th>Species</th>
<th>FGS</th>
<th>HNS (^{+}) (mmol·L(^{-1}))</th>
<th>Crop Cycle</th>
<th>Crop season</th>
<th>Plant density (Plants·m(^{-2}))</th>
<th>Plant weight (g)</th>
<th>Total yield (g·m(^{-2}))</th>
<th>30-Plant sampling (g·m(^{-2}))</th>
<th>Discrepancies (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Brassica alba</em> L. Boiss(^w)</td>
<td>FL – no aeration</td>
<td>6-2-6</td>
<td>25</td>
<td>31</td>
<td>Winter</td>
<td>1569</td>
<td>1.36</td>
<td>1334</td>
<td>2132</td>
</tr>
<tr>
<td><em>Brassica alba</em> L. Boiss</td>
<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>13</td>
<td>12</td>
<td>Spring</td>
<td>1569</td>
<td>1.06</td>
<td>1329</td>
<td>1657</td>
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<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>8</td>
<td>9</td>
<td>Late spring</td>
<td>1569</td>
<td>1.01</td>
<td>1056</td>
<td>1580</td>
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<td><em>Brassica nigra</em> L. Koch(^w)</td>
<td>FL – no aeration</td>
<td>6-2-6</td>
<td>25</td>
<td>33</td>
<td>Winter</td>
<td>1569</td>
<td>1.51</td>
<td>1173</td>
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<tr>
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<td>6-2-6</td>
<td>8</td>
<td>13</td>
<td>Spring</td>
<td>1569</td>
<td>1.46</td>
<td>1596</td>
<td>2283</td>
</tr>
<tr>
<td><em>Lepidium sativum</em> L. ‘Cresso’(^w)</td>
<td>FL – no aeration</td>
<td>6-2-6</td>
<td>17</td>
<td>39</td>
<td>Winter</td>
<td>1961</td>
<td>0.68</td>
<td>1068</td>
<td>1341</td>
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<td><em>Lepidium sativum</em> L. ‘Cresso’</td>
<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>12</td>
<td>15</td>
<td>Spring</td>
<td>1961</td>
<td>1.29</td>
<td>2039</td>
<td>2536</td>
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<tr>
<td><em>Lepidium sativum</em> L. ‘Cresso’</td>
<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>4</td>
<td>13</td>
<td>Late spring</td>
<td>1961</td>
<td>0.83</td>
<td>1146</td>
<td>1413</td>
</tr>
<tr>
<td><em>Nasturtium officinale</em> R. Br(^w)</td>
<td>FL – no aeration</td>
<td>6-2-6</td>
<td>23</td>
<td>40</td>
<td>Winter</td>
<td>1961</td>
<td>0.72</td>
<td>1146</td>
<td>1413</td>
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<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>15</td>
<td>17</td>
<td>Spring</td>
<td>1961</td>
<td>1.05</td>
<td>1603</td>
<td>2060</td>
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<tr>
<td><em>Nasturtium officinale</em> R. Br</td>
<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>9</td>
<td>16</td>
<td>Late spring</td>
<td>1961</td>
<td>0.95</td>
<td>1517</td>
<td>1857</td>
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<td><em>Nasturtium officinale</em> R. Br</td>
<td>FL + 15' on + 45' off</td>
<td>12-2-6</td>
<td>4</td>
<td>20</td>
<td>Summer</td>
<td>1961</td>
<td>1.22</td>
<td>1532</td>
<td>2396</td>
</tr>
<tr>
<td><em>Nasturtium officinale</em> R. Br</td>
<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>11</td>
<td>24</td>
<td>Autumn</td>
<td>1961</td>
<td>1.24</td>
<td>2330</td>
<td>2431</td>
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<tr>
<td><em>Nasturtium officinale</em> R. Br</td>
<td>FL + 15' on + 15' off</td>
<td>6-2-6</td>
<td>12</td>
<td>24</td>
<td>Autumn</td>
<td>1961</td>
<td>2.12</td>
<td>3341</td>
<td>4153</td>
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<td><em>Eruca sativa</em> Mill(^w)</td>
<td>EF</td>
<td>6-2-6</td>
<td>11</td>
<td>16</td>
<td>Early spring</td>
<td>1961</td>
<td>0.66</td>
<td>812</td>
<td>1301</td>
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<td>11</td>
<td>16</td>
<td>Early spring</td>
<td>1961</td>
<td>0.73</td>
<td>1592</td>
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<td>12</td>
<td>16</td>
<td>Spring</td>
<td>1961</td>
<td>0.96</td>
<td>1971</td>
<td>1880</td>
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<td>7</td>
<td>11</td>
<td>Late spring</td>
<td>1961</td>
<td>0.67</td>
<td>1236</td>
<td>1314</td>
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<tr>
<td><em>Diplotaxis tenuifolia</em> L. DC. ‘Grazia’(^w)</td>
<td>EF</td>
<td>6-2-6</td>
<td>11</td>
<td>37</td>
<td>Early spring</td>
<td>1961</td>
<td>0.66</td>
<td>821</td>
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<td><em>Diplotaxis tenuifolia</em> L. DC. ‘Grazia’</td>
<td>FL + 120’ on + 120’ off</td>
<td>6-2-6</td>
<td>11</td>
<td>37</td>
<td>Early spring</td>
<td>1961</td>
<td>1.19</td>
<td>1454</td>
<td>2343</td>
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<td>20</td>
<td>Spring</td>
<td>1961</td>
<td>0.78</td>
<td>867</td>
<td>1526</td>
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<td><em>Valerianella olitoria</em> L.</td>
<td>FL + 15’ on + 15’ off</td>
<td>6-2-6</td>
<td>8</td>
<td>17</td>
<td>Late spring</td>
<td>1176</td>
<td>0.91</td>
<td>1346</td>
<td>1776</td>
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<tr>
<td><em>Valerianella olitoria</em> L.</td>
<td>FL + 15’ on + 15’ off</td>
<td>6-2-6</td>
<td>15</td>
<td>19</td>
<td>Spring</td>
<td>1176</td>
<td>0.77</td>
<td>859</td>
<td>911</td>
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<td><em>Spinacia oleracea</em> L. ‘Ballet’(^w)</td>
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<td>9</td>
<td>18</td>
<td>Late spring</td>
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<td>453</td>
<td>556</td>
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<td>6-2-6</td>
<td>15</td>
<td>14</td>
<td>Spring</td>
<td>1176</td>
<td>1.82</td>
<td>986</td>
<td>2141</td>
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</table>

\(^{+}\) FGS = Floating growing system; FL = Continuous flotation system; EF = Ebb-and-flow system; No aeration, on, off\(^w\) = No aeration, minutes on and minutes off of aeration during growth, respectively.  
\(^{\#}\) HNS = Hydroponic nutrient solution; 6-2-6: 6N-2P-6K; 12-2-6: 12N-2P-6K; 6-4-12: 6N-4P-12K; 12-4-12: 12N-4P-12K.  
\(^{x}\) Yield = Total yield: Yield (calculated from the total fresh mass); 30-Plant sampling: Yield (calculated from 30-plant sampling); Discrepancies: Yield computation discrepancies.  
\(^{\ast}\) Seeds provided by: Martin Bauer.  
\(^{\ast\ast}\) Seeds provided by: Jelitto.  
\(^{\ast\ast\ast}\) Seeds provided by: Enza Zaden.  
\(^{\ast\ast\ast\ast}\) Seeds provided by: Clause.  
\(^{\ast\ast\ast\ast\ast}\) Seeds provided by: Seminis.
spread tube system on the bottom of the beds to homogeneously insufflate air in the HNS (Fig. 1, red path; Fig. 5C). In each bed, 10.80 m of dripline tube (0.016 m outer diameter and 0.008 m thickness) were allocated with ca. 5 holes per m to diffuse air into the HNS (2.1 L·h⁻¹). Dripline tubes were anchored to the bottom of the beds by an inner stainless steel grid. A logic module (Zelio compact PLC, SR2B201FU 20i/o Telemecanique; RS Components S.p.A., Cinisello Balsamo (MI), Italy) was used to regulate airflow scheduling. The airflow is generated by a compressor (Mercure, made in P.R.C., provided by Nu Air Compressors and Tools S.p.A., Robassomero (TO), Italy).

Growing cycle

According to the experimental design, a defined number of 60-cell styrofoam trays were filled with ca. 50 cm³ per cell of a specific peat-based horticultural medium (Neuhaus Huminsubstrat N17; Klasmann-Deilmann® GmbH, Geeste, Niedersachsen, Germany) by an automatic filling machine in the farm Azienda Agricola Vivaistica Ricca Sebastiano (Carignano (TO), Italy).

BLV species under investigation were manually sown. Subsequently, a partially automated sowing system has been designed and developed due to: incomplete precision of manual sown particularly for species with seeds of reduced size (e.g., water cress); different worker skill in performing sowing; high number of styrofoam trays (often over 200) to be sown in the shortest possible time. This sowing system, called GiuWaPa, has been developed using a series of sieves and holes on the basis of the volume of the seeds of each species (Fig. 6). The sown styrofoam trays were placed into the nursery plastichouse, and daily overhead irrigated for 1 minute at 8 am and 2 pm.

Seedling thinning was performed after cotyledon expansion to reach a specific plant density, according to species used, canopy development expected, shade disturbance by plant canopies, and consequent potential effect on phytochemical profile of the raw material at harvest. In particular, the plant density was ca. (all in plants·m⁻²): 1569 for white mustard, and black mustard; 1961 for garden cress, water cress, rocket salad, and perennial wild rocket; 1176 for corn salad, and baby spinach (Table 1). After thinning, styrofoam trays were moved into the water beds.

Based on series of trials on HNS efficiency for BLV, 200 L of HNS were used to fill each water bed of the LSPP at the start of the growing period of the plants under flotation, that is a depth of ca. 0.06 m of solution available for trays to float on it. This amount of HNS has been proven to be sufficient for the entire growing cycle of 3–5 plants of BLV per cell of the 16 styrofoam trays used per water bed. Ultimately, the HNS requirement has been in average between 0.04 L/plant and 0.07 L/plant.

Before the start of the flotation and during the growing cycle several measurements were taken in order to check the growing conditions and recorded for further analyses and system considerations. Air temperature and humidity were constantly measured in the greenhouses with FAO standard thermohygrometers. The photosynthetic photon flux (PPF) was measured at canopy level 3 times a week with a Quantum sensor (model LI-1000 Data Logger; LI-COR, Inc., Lincoln, NE, USA). In the HNS, pH, conductivity and temperature were measured 3 times a week with Waterproof CyberScan PC 650 (Eutech Instruments Pte Ltd., Singapore) equipped with a submersible pH electrode (ECFC7252203B) and a conductivity/temperature probe (CONSEN9203J). Oxygen content dissolved in the HNS was measured 3 times a week with an osimeter (model YSI 550A; YSI, Inc., Yellow Springs, OH, USA).

Sampling size and statistical analysis

The statistical experimental designs used were randomized complete block design (RCBD). Regardless of the treatments, 3 blocks were always used during growing period and, eventually, during the following postharvest experiment. In similar cases generally 4 blocks are used while we decided to use 3 blocks after that some considerations were taken into account, in addition to the gradient created by the sunlight exposition, stated previously. According to F distribution used for ANOVA, using 3 blocks, thus 2 degrees of freedom, guarantees a greater statistical probability significance than using 2 blocks; while using 4 blocks, thus 3 degrees of freedom, guarantees a marginal greater statistical probability significance than using 3 blocks. On the other hand, using 4 blocks instead of 3 in the experimental design implies a great increase in time of operations in all the steps of the growing cycle (e.g., seedling, thinning, measurements in the greenhouse and in the HNS, harvesting operations, postharvest opera-
tions), increasing the effect of the sampling timing. Moreover, setting a fourth block in the experiments increases the need for resources both at level of materials, equipment, and instruments, and at level of manpower. Consequently, the lower statistical power of an experiment with 3 blocks than 4 blocks is compensated by the concentration of the time required to do all the activities and measurements in a reasonable time interval and the optimization of the resources.

Technical applications

Harvesting took place when plants were reaching commercial maturity according to species adapted for the fresh-cut industry. Plant densities studied throughout the trials were such that harvest was taking place when canopy was covering all tray surface (Fig. 7). Harvesting and raw material handling for measurements and analyses followed standard procedures also set throughout trials for replicable and comparable experiments, and for timing efficiency of sampling procedures. Harvesting was starting early in the morning also to avoid the hottest hours of the day. Tools used for harvesting were sanitized prior to use; the crew was taking particular attention to hand sanitation and personal dress and hygiene; gloves were always used to handle the raw material to avoid contamination. Raw material was immediately used for biometric determination. According to the experimental design set at any trial, the rest of the raw material was transferred to the post-harvest laboratory to be processed as fresh-cut product.

All fresh mass harvested was used to compute the yield per square meter. In this way, there is a better estimation of the agronomic yield compared with sampling a limited number of plants per treatment and calculated multiplying the plant density and the weight of the single plant obtained from a small sampling. Table 1 is presenting some results obtained in several trials with different treatments; the column “Yield discrepancies” gives the % of discrepancies between the 30-plant sampling and the real yield per square meter calculated considered the all fresh mass production: except for two cases, in all instances there is an overestimation if a 30-plant sampling is used, even reaching more than 100%. In average the overestimation can be ca. 35%.

Concluding remarks

A system and methodology was developed for qualitative and quantitative analyses of BLV grown in FGS. LSPP were developed in two greenhouses differing for structure, suitable for different growing seasons in a continental climate. The equipment and technology allowed multiple treatments and replicates for sound statistical design and data analyses. Environmental conditions and cultural techniques were studied in major and minor species (white mustard, black mustard, garden cress, water cress, rocket salad, perennial wild rocket, corn salad, baby spinach) to determine best cultivation techniques in a standard SCS for BLV, based on FGS. The LSPP installed are providing the basis for expanding the research to other species and agronomic factors.

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