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Influence of ventilation and media on potato (*Solanum tuberosum* L.) tuberization and its growth characteristics

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Considering the most suitable relationship between ventilation and media in this study, two mixtures of perlite and coir (3:1, light and 1:3, heavy v/v) were applied to determine the growth characteristics *var.* Sante of potato plantlets. As potato has high potential in minituber production, different levels of supplemental aeration (0, 12.5, 25 and 75%, v/v/min) were compared for providing root oxygen demand and increasing potato minituber yield. Applying sufficient aeration led to significant increase in dry matter, number, size, quality of minituber and most of the growth parameters such as leaf area index (LAI), leaf area index duration (LAID), harvest index (HI) and root : shoot ratio. The results showed that by applying aeration at 25% (v/v/min), tuber yield was increased significantly up to 162% and increase in tuber number was predominant at the rate of 3678 tuber/m^2. Desirable performance of aerated plant can be resulted from root oxygen demand of potato which is higher than normal ventilation in media. Heavy media was more responsive to aeration than light one. Thus, aeration at 25 and 12.5% (v/v/min) in heavy and light media respectively was considered as favorable level in rizo-sphere and had positive effect on tuberization.

Key words: Aeration, air-filled porosity, container system, media, oxygen deficiency, seed potato production.

INTRODUCTION

Reaching a commercial scale in soilless culture is motivated by a potential for increasing the crop productivity and efficiency. Widespread adoption of soilless container production in global food and plant production is the technical solution for problems including root diseases, root zone oxygen deficiency, fertility control which occurs in other systems.

The advantages of container production systems over ground production systems are simultaneously, greater water and nutrient efficiencies, less water consumption with more food production, applying recirculated systems, better cropping with higher salinity levels than soil grown plants. The reason is the connection between ample oxygen supply to the roots and their ability to exclude toxic ions and tolerate high osmotic pressure (Raviv et al., 2008). In container systems, gravitropism and hydrotropism frequently leads to accumulation of root mass at the bottom, which is the major part of total root biomass and may be exposed to oxygen deficiency due to the respiration of dense root mass and existence of a water layer at the bottom of the container (Bhattarai et al., 2006; Acuña et al., 2008).

Aeration is one of the critical factors influencing root and plant growth. Plant cells need oxygen for division and function. If rooting medium suffers from oxygen deficiency, plant will be injured severely or dead (Cherif et al., 1997). Especially in potato plant, due to large volume of biomass in root, oxygen deficiency plays more remark-

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able role. Respiration which produces energy for root growth and ion absorption requires oxygen. Metabolic processes like cell division, water movement into roots and mineral uptake can be prohibited by root oxygen deficiency and creates some changes in root system morphology, nutritional deficiencies and increased water stress. Roots will die after disruption of absorbing water and ions resulting from lack of adequate oxygen (Morard and Silvestre, 1996; Caron and Nkongolo, 2004). Selected physical aspects related to plant growth requirements, such as bulk density, total pore volume, structural stability, root resistance, water retention, rewetting, hydro-phobicity, water transport and oxygen transport are used to discuss the suitability of growing media for plant growth (Blok and Wever, 2008). The main role of physical characteristics of media is having suitable air-filled porosity for efficient oxygen diffusion and maintaining favorable water content for supplying water and nutrients and respiration of root. Measurements of bulk density, moisture and organic matter content can accurately predict air-filled porosities. Reductions in air-filled porosity at increasing moisture content are accompanied by an increase in permeability, apparently due to aggregations of fines (Richard et al., 2004). A growing media with higher water content has poor ventilation. Therefore increasing water holding capacity adversely affect on oxygen diffusion rate in media and plant growth and development (Cho et al., 2006). Aeration problems in growing media were examined by Caron et al. (2004, 2005) and Nkonogolo and Caron (1999). They clarified that gas diffusion is critical for diagnosing aeration problem in different media. It is important to emphasize that the need for measuring gas diffusion at different water content. The chosen water content should correspond as closely as possible to the container situation if the gas diffusivity is used as an index to assess plant performance. Wever et al. (2001) found out that with the air-filled porosity (AFP) less than 40% (v/v) the oxygen diffusion rate would have complete relationship with AFP and if it reduced to less than 30% (v/v), even in containers with small dimension 20 cm height and width, oxygen concentration would drop to less than 20%. This fact highlights the role of supplemental aeration for preventing oxygen drop-off.

Plantlet culture in hydroponics system is recently used as an efficient method to produce and propagate minituber, which are healthy seeds without any contamination to pathogens. Eradication of soil born plant diseases, prevention from inoculation of pathogens by sterilized root medium, increasing in growth rate, propagation and vigor of minitubers, multi harvesting, omission of terminal dominance, uniformity in size and higher number of minitubers are results of applying this method (Lommen and Struik, 1992; Lommen, 2007). The science of minituber production in soilless systems shows an improvement in seed potato production program. In order to reach the ideal efficiency in this technique some practical information should be enhanced. The purpose of this study was to investigate the role of media porosity and aeration of media on the main issues of dissolved oxygen deficiency in the potato root zone and mode of oxygen action on tuberization.

MATERIALS AND METHODS

This study was conducted during February - July 2007 at greenhouse of Mashhad Ferdowsi University, Iran. The treatments were arranged in a factorial experiment based on randomized complete block design with 4 replications, in order to determine the interaction of media composition and aeration on potato (*var.* Sante) minituber production under soilless conditions. The involving factors in this experiment included 4 levels of aeration in root zone and 2 compositions of media. The conditions for running the experiment were average temperature 21.6°C, mean partial humidity 79.4%, with 14 h photo period under natural light intensity.

The close container hydroponic systems were used to run this study. For applying supplementary ventilation of rooting medium at the bottom of each 10 l container (25 cm height and 22.5 cm diameter), a round air stone was installed that injected fresh air by an air pump to the media via separate flow meters. The flow meters were arranged to allow flowing of 1, 2 and 6 l air per min in terms of 12.5, 25 and 75% of the 8 l of media. The air pump was equipped with a timer which worked from sunlight to sunset (5:30 to 19:30). Aeration with fresh air was done at 4 levels, 0 (control), 12.5% (Low aeration), 25% (Moderate aeration) and 75% (High aeration) volume of media per min. Different levels of aeration were estimated based on dissolved oxygen (DO) changes after aeration in the hot time of day (that is 2 pm).

Among different commercial substrates which are used commonly, perlite with high air-filled porosity and coconut coir with high water holding capacity (WHC) were selected. These 2 substrates were prepared at 100:0, 75:25 (3:1), 50:50 (1:1), 25:75 (1:3) and 0:100 of perlite and coconut coir volume ratio respectively, used in preliminary experiment in order to specify the growth and development pattern of potato plantlets in these different compositions. From the results of preliminary experiment and also AFP and water holding characteristics of different compositions, two composition 3:1 and 1:3 of perlite and coir, named Light and Heavy media respectively, were selected for further study in the main experiment. The 3 week old *in vitro* plantlets were transplanted after 1 week acclimatization period with density 200 plants/m^2 (Lommen and Struik, 1992). Obtaining adequate number of *in vitro* plantlet was conducted using virus free potato plantlet. Virus free potato plantlet were made using tissue culturing of single nodal cuttings in modified Murashige and Skoog medium (Zamora et al., 1994).

Nutrient solution was continuously re-circulated 4 times per day and replaced every 3 weeks by applying some changes in the amount of N, P and Ca to 100, 141, 180 ppm after stolon initiation and to 60, 180, 160 ppm after tuber initiation, respectively. Also the pH and electrical conductivity (EC) of the solution were maintained at 5.8 and 2.2 to 2.5 mS/cm, respectively. The growth characteristics including stem number, plant height, leaf area, stem diameter and dissolved oxygen inside the media were measured at 14, 28, 42, 56, 70, 84, 91 and 105 days after transplanting (DATP) via non-destructive monitoring. The estimated leaf area index (LAI) was calculated by correlation between length, width and number of leaves with surface counted from checkered sheet (Hui et al., 2001). Oxygen concentration of media was monitored using a dissolved oxygen meter (Hana, YSI 55 -12 USA). Interval harvesting was done at 42, 70, 91 and 105 DATP (Lommen and Struik,

Table 1. Physical properties of the media composition tested.

Media	Max Water holding capacity (v/v)	Air-filled porosity in Max water holding (v/v)	Air-filled porosity one day after watering (v/v)	Total pore space (v/v)
Perlite	5.56e	10.66a	37.72a	69.92e
Perlite : Coir (3:1)	9.28d	10.77a	27.45b	80.18d
Perlite : Coir (1:1)	13.36c	10.66a	21.77c	84.15c
Perlite : Coir (1:3)	27.67b	11.26a	16.55e	89.36b
Coir	96.57a	11.34a	18.82d	96.28a

Mean separation within columns by Tukey's multiple range test at $p \leq 0.05$.

1992). Root length, number of root and stolon and dry matter of tubers were measured.

Harvested minitubers were classified based on their diameter into 4 groups according to C_1: <5, C_2: 5 – 10, C_3:10 – 15, C_4: > 15 mm diameter. Fresh weight and dry matter (oven dried for 72 h at 75C) of classified minitubers were determined. Time of tuber initiation was dated when 70% of plants of each plot had at least one tuber (>5 mm diameter). Physiological maturity was assigned when 70% of leaves turned yellow (Kawakami et al., 2004).

Field germination test was done by drying 2 minitubers per experimental unit on each class for 2 days, cured for 14 days and stored at 4°C in darkness and 80% RH for 3 weeks. Then, they were put in the pre-sprouting condition (15°C, 8 W/m^2 light and 80% RH) for 10 days, after treating with 2 ppm gibberellic acid for 10 min in order to break its dormancy. Field planting took place on the loamy soil with drip irrigation. Germination of 3 visible leaves was recorded every day. Analysis of variance for data and orthogonal mean comparison were performed by the general liner model (GLM) procedure of SAS (SAS institute inc., Cary, NC, USA, version 8.2.0). For mean separation significance, the Tukey's multiple range test at $p \leq 0.05$ was used.

RESULTS

Physical characteristics of media

Comparison between physical characteristics of different perlite and coir compositions indicated obvious difference between pure form of these compositions regarding maximum water holding and AFP which by increasing the % of coir in composition, water holding capacity (WHC) increased and AFP decreased (Table 1). Different compositions of perlite and coir showed same level of total pore space although this characteristic in perlite was less than coir. All different compositions had the same AFP in maximum water holding condition, but after 24 h significant difference became obvious. Composition 1:3 of perlite and coir showed 6.48% increases in AFP whereas composition 3:1 showed 17.06% increase in AFP. These 2 compositions (1:3 and 3:1) were selected, based on maximum WHC and AFP after 24 h, for further study in order to investigate the effect of supplemental aeration on these compositions. Composition 3:1 of perlite and coir was named as Light media regarding to its lower maximum water holding, and composition 1:3, due to higher maximum water holding, was named as Heavy media.

Plant growth in responses to aeration and media

Applying aeration in different levels (Low, Moderate and High) led to significant increase in dissolved oxygen level in both media (Table 2). Increasing in DO level resulted in improvement in plant growth characteristics, such as plant height, number of stem in each plant, stem diameter, length of largest root and number of stolon. This increasing trend was the same for both media (1:3 and 3:1 compositions) and showed significant difference compared to control. Also increasing the level of aeration led to remarkable increase in growing period and delay in physiologic maturity of plant. This delay was 10 days in Light media and 11 days in Heavy one. Furthermore, applying aeration led to two days acceleration in tuberization in Heavy media when compared to control.

Comparison the main effects of aeration and media in vegetative and reproductive growth rate of potato plantlets showed increasing in LAI, which was associated with increasing in aeration level. Leaf area index duration (LAID), also was increased in accordance to increasing in aeration level which was affected by increasing in LAI and longer growing period that happened in aerated levels comparing control group. The LAI and LAID were higher in Heavy media compared to Light one (Table 3). Root shoot ratio and harvest index (HI) was less in control group rather than aerated levels, although HI did not show any difference between 2 media treatments.

Minituber production

The interaction effects of aeration and media on the number of potato tuber produced showed significant effect in the number of minituber ($p < 0.0001$) in different aerated level and both kinds of media when compared to control group (Figure 1). Moderate and high level of aeration, when compared to Low level, had no effect on the number of minituber in Light media, whereas in Heavy

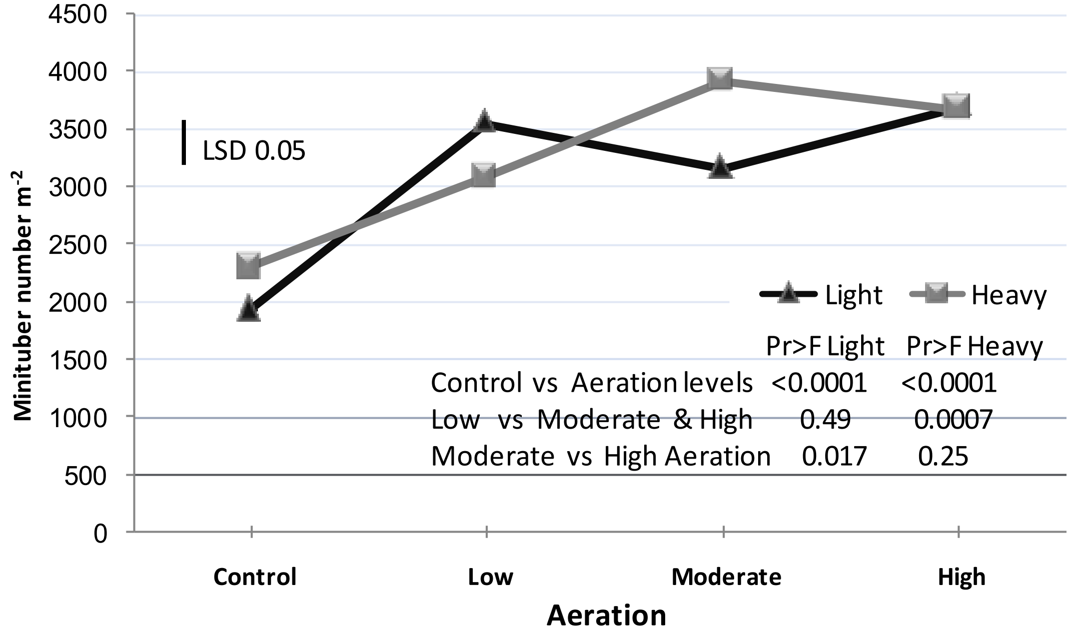

Figure 1. Changes of minituber number as affected by different type of media and aeration levels.

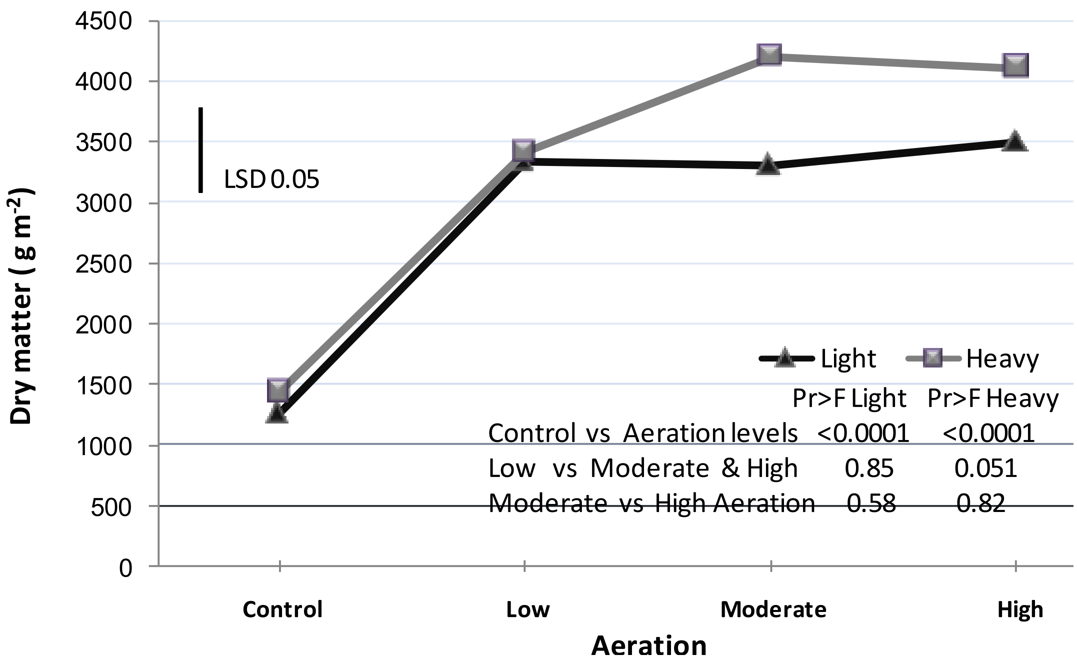

Figure 2. Changes of minituber dry matter as affected by Light and Heavy media in different aeration levels.

media the number of minitubers was increased by higher level of aeration significantly (p = 0.007) which was 3912/m^2 in Moderate level. In Heavy media, there was no difference in minituber numbers between Moderate and High level of aeration. But in Light media, the minituber numbers were significantly increased in High level of aeration (p = 0.017) which was 3687/m^2.

Orthogonal contrast of minituber dry weight in different

Table 2. Interaction effects of different aeration levels and media on potato plantlet growth characteristics.

Aeration levels	Dissolved oxygen (mg/l)		Plant height (cm)		Stem diameter (mm)		Stem number (per plant)		Largest root length (cm)		Stolon number (per plant)		DATP to tuberization		DATP to ripeness	
	Light	Heavy	Light	Heavy	Light	Heavy	Light	Heavy	Light	Heavy	Light	Heavy	Light	Heavy	Light	Heavy
Control	5.01d	4.84d	84.3b	82 b	4.25c	4.23c	1.04b	0.94b	19.75b	7.63b	3.31b	2.97b	39 a	38.25a	91.13c	90.75b
Low	7.09c	6.97c	113.3a	108 a	5.49b	5.74b	1.07ab	1.09a	40.04a	37.82a	8.56a	7.15a	36 c	37.5ab	96.25b	98.25a
Moderate	7.92b	7.65b	116.4a	122.1a	5.86ab	6.59ab	1.10ab	1.29a	35.80a	39.71a	8.24a	7.77a	38.3ab	36.5b	100.4ab	99.88a
High	8.35a	8.24a	112.6a	119.8a	6.56a	7.18a	1.24a	1.27a	42.49a	36.85a	8.35a	7.25a	36.9bc	36.1b	101.4a	101.2a

Aeration levels: Control = 0, Low = 12.5, Moderate = 25, High = 75 (% air V/media V/ min)
Light and Heavy media were 3:1, 1:3 of perlite and coir volume ratio, respectively.
Mean separation within columns by Tukey's multiple range test at $p \leq 0.05$.

Table 3. Main effect of different aeration levels and media on plant and tuber parameters.

Factor		LAI	LAID (m^2/m^2 /day)	Root / shoot	HI	Days to germination	% Minituber germination	Starch number
Aeration	Control	1.3 d	121.27d	3.1 b	63.71b	5.94a	88.28b	6.11c
	Low	2.7 c	266.19c	5.83ab	74.01a	5.31b	94.53a	9.55b
	Moderate	3.65b	369.92b	4.71ab	73.16a	4.56c	97.66a	9.73b
	High	4.01a	411.04a	6.64a	77.1 a	4.13c	96.88a	10.72a
Media	Light	2.78b	278.10b	5.20a	71.78a	5.09a	94.53a	8.43b
	Heavy	3.06a	306.11a	4.94a	72.21a	4.88a	94.14a	9.62a

Aeration levels: Control = 0, Low = 12.5, Moderate = 25, High = 75 (% air v/ media v/min).
Light and Heavy media were 3:1, 1:3 of perlite and coir volume ratio, respectively.
Mean separation within columns by Tukey's multiple range test at p ≤0.05.

level of aeration for both types of media showed significant increase in dry matter in aerated levels compared to control group ($p < 0.0001$), which was 1277, 1446 g/m^2 in control and 3489, 4099 g/m^2 in High level aeration in the Light and Heavy media, respectively (Figure 2). Moderate and High levels of aeration showed remarkable increase in dry matter comparing Low level in Heavy media ($p = 0.051$). However, Light media did not show any significant difference in this regards. These results indicated that Heavy and Light media showed the same reaction in potato production without applying aeration or using only low level of aeration. But, applying higher level of aeration (Moderate and High) in Heavy media due to higher WHC provided more favorable environment for plant growth. Without applying aeration there was no difference between two media because Light media had higher AFP which assisted in crop growth improvement.

The grading of minitubers in to classes (C1, C2, C3 and C4) showed the differences in minituber yield between two media more clearly. Minituber yield was raised up from 69.9 to 80.2, 79.7 and 78.4% in class C4 from control by applying Low, Moderate and High level of aeration respectively. It can be seen in Figure 3 that class

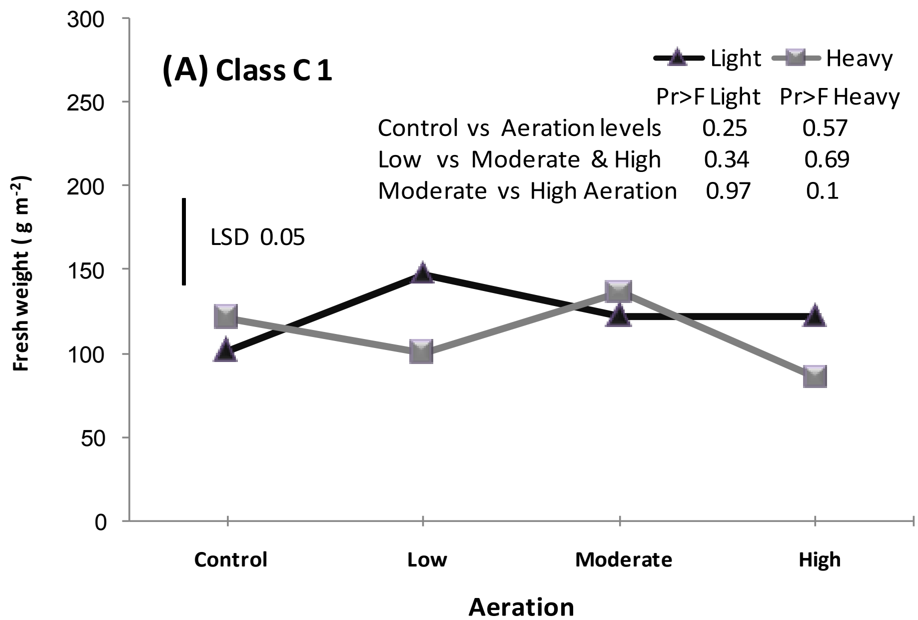

Figure 3A. Classification of minituber yield of potato plantlet under different levels of aeration and different media in class C1. Aeration levels: Control = 0, Low = 12.5%, Moderate = 25%, High = 75% (air v/ media v/ min).

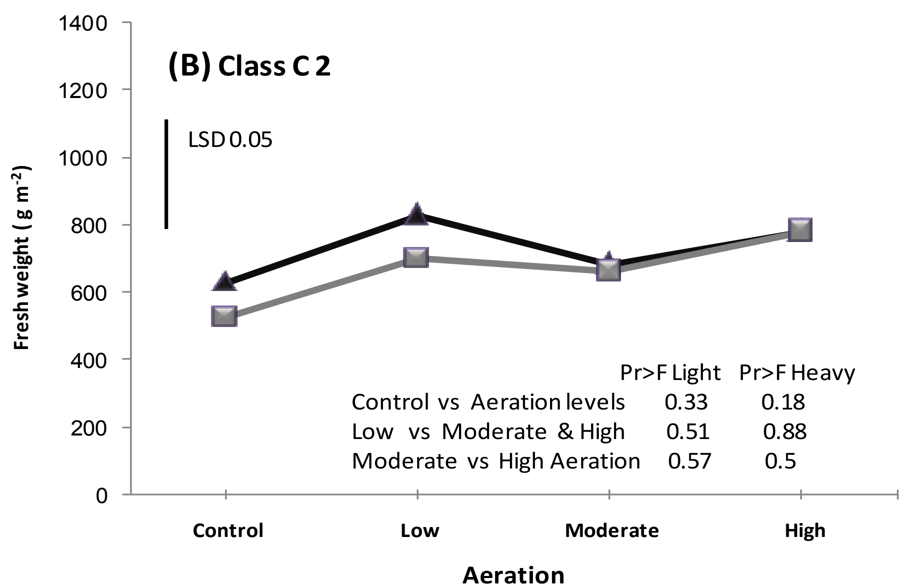

Figure 3B. Classification of minituber yield of potato plantlet under different levels of aeration and different media in class C2. Aeration levels: Control = 0, Low = 12.5%, Moderate = 25%, High = 75% (air v/ media v/ min)

C1 and C2 did not show any significant difference in tuber fresh weight (Figure 3A, 3B). The differences were shown mainly in tubers with larger size than 15 mm (C4) and C3. Class C3 of minitubers showed significant increase in fresh weight with aerated levels comparing to control group in both types of media. In Light media, moderate and High level of aeration rather than low level showed remarkable increase in tuber yield (p = 0.015)

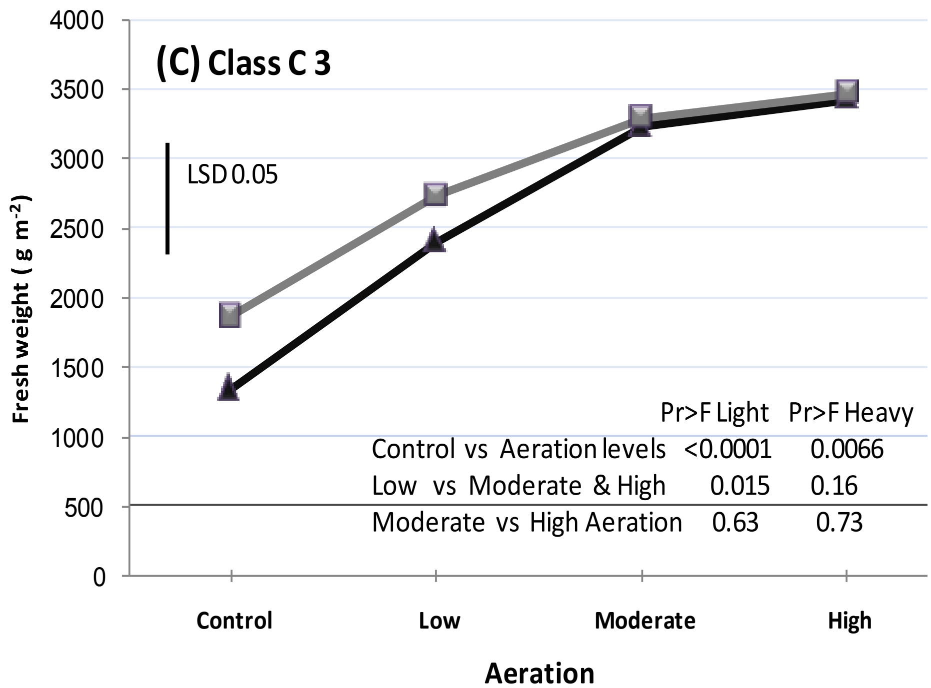

Figure 3C. Classification of minituber yield of potato plantlet under different levels of aeration and different media in class C3. Aeration levels: Control = 0, Low = 12.5%, Moderate = 25%, High = 75% (air v/ media v/ min).

(Figure 3C). Class 4 of minitubers also showed the lowest tuber yield in control group ($p < 0.0001$). In this class only, Heavy media showed significant difference in yield between Moderate and High level of aeration versus Low level (Figure 3D).

The main factor affecting the total yield was the yield of C4 minitubers class. Regarding total yield, control group showed the lowest minituber yield in both types of media ($P < 0.0001$) which was 7120 and 8132 g/m^2 for light and Heavy media, respectively. Increasing the level of aeration from low to Moderate and High led to significant increase in total yield in Heavy media ($p = 0.019$), which was 21598 g/m^2. Light media, due to its higher AFP, did not show any positive reaction to higher level of aeration (18497 g/m^2). Because the total minituber yield of Moderate and High aeration levels was similar in Heavy media (Figure 3E), it can be concluded that Low level of aeration 12.5% (v/v/min) is sufficient for minituber production in Light media while Moderate level of aeration probably provide the need of plant in Heavy media in container systems.

Minituber germination test

Germination ability of minitubers which were produced by applying different levels of aeration showed a very good (88 - 97%) germination rate after 4 to 6 days. There were no differences between media in germination, albeit aeration levels, due to the larger tubers and higher vigor, showed significantly higher and faster germination than the control (Table 3).

DISCUSSION

The growth characteristics of three levels of aerated plants in both light and heavy media demonstrated significantly greater number of stem, stolon, stem diameter, plant height, largest root length and longer plant growth period in both media than control (Tables 2). The greatest total minituber fresh and dry weights were obtained in the heavy media in Moderate and High aeration levels. Also, in the Heavy media, the highest minituber number appeared in Moderate level of aeration. It was clarified that applying supplemental aeration was needed to increase the growth parameters such as LAI, LAID, HI, root: shoot ratio, tuber yield, dry matter (up to 178%) and minituber number (up to 74%) significantly. Whereas there is no significant difference between Low versus Moderate and High level of aeration in Light media, Heavy media was more responsive to aeration system than Light one.

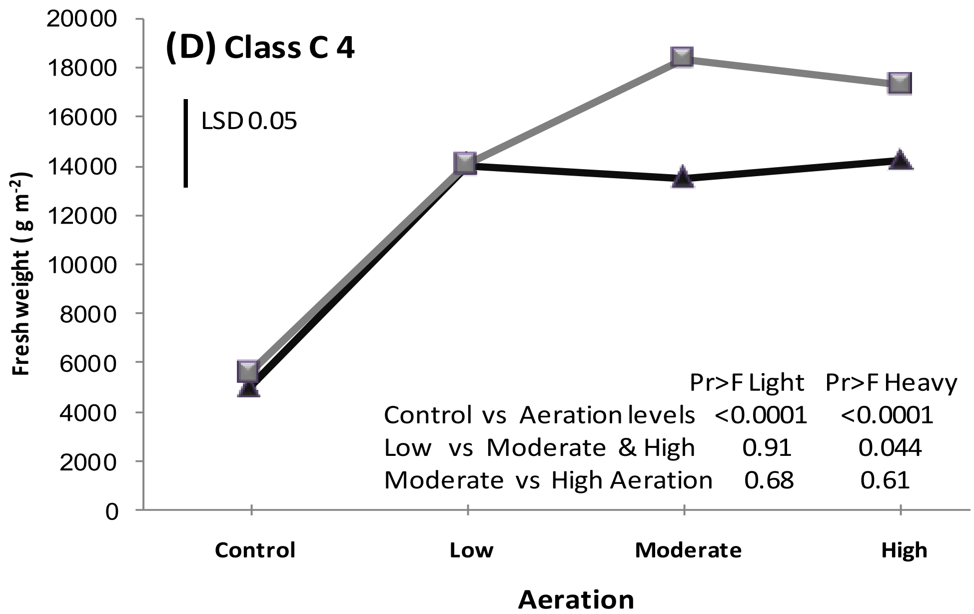

Figure 3D. Classification of minituber yield of potato plantlet under different levels of aeration and different media in class C4. Aeration levels: Control = 0, Low = 12.5%, Moderate = 25%, High = 75% (air v/ media v/ min).

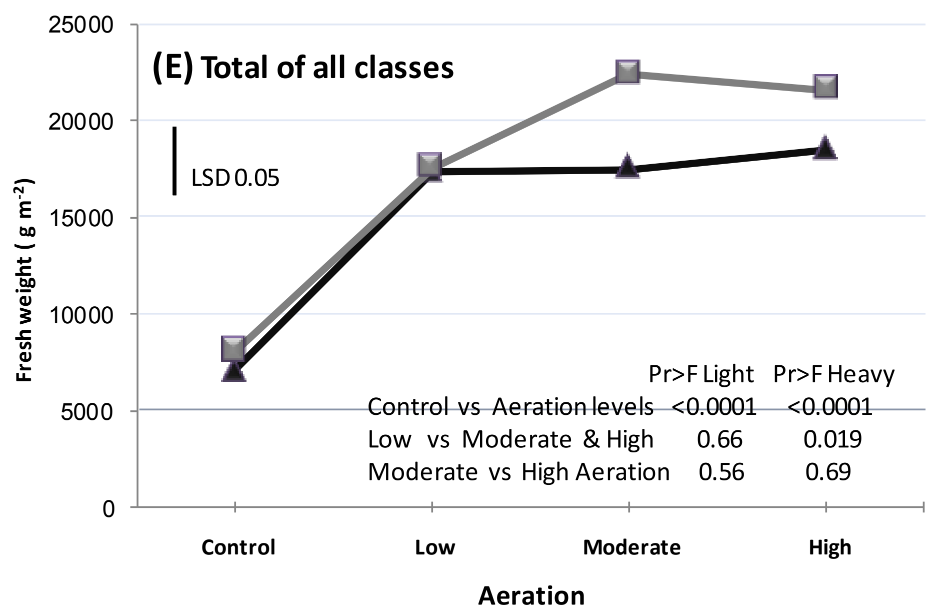

Figure 3E. Classification of minituber yield of potato plantlet under different levels of aeration and different media in class (sum of total classes). Aeration levels: Control = 0, Low = 12.5%, Moderate = 25%, High = 75% (air v/ media v/ min).

In some kinds of media high level of total porosity and a rather very high level of air-filled porosity results in low level of easily available water (Clemmensen, 2004). This was consistent with our results that by decreasing water holding capacity, air-filled porosity would increase in tested composition.

The AFP did not show any significant differences among different media in maximum water holding capacity. These results were similar to Cho et al. (2006), who clarifies that coir has smaller pores comparing perlite, whereas in available water condition coir, perlite and their compositions would be different regarding their AFP (Table 1). Finding an optimum growing medium for potato plantlets growth which is a tuber crop with high growth potential and requires high level of oxygen in underground parts, would be resulted from sufficient air supply and also having excessive water content for providing high plant nutrients and water demand. These findings were consistent with Morgan (2004) that mentions that many variables influence how much oxygen is available to the root system, including field factors such as compaction, soil type and structure, amount of humus, moisture content, the microbe populations present and, importantly, the amount of dissolved oxygen present in irrigation water and air-filled porosity.

Schroeder and Knaack (2007) showed the cucumber yield and nutrient content is not significantly affected by different types of media. All tested media have sufficient oxygen content to provide optimal cucumber plant growth, while different substrates have slightly different level of oxygen. They conclude that the cucumbers are able to take up enough amounts of nutrients and oxygen if the nutrient solution is enriched above the critical minimum oxygen, but mention that further investigations should be done with different O_2 levels at the same substrate. These contradictory results can be due to the differences in plant species, levels of O_2 or treatment duration. While in the current experiment comparison between two kinds of Light and Heavy media with sufficient AFP and WHC showed that they could not provide optimum growing medium for potato plantlet growth and producing of highest tuber yield. These differences can be related to the high biomass of potato root which is a tuber crop compared cucumber with scattered root. Morgan (2004) stated a more problematic issue in hydroponic systems which intermittent or mild oxygen deficiencies in the root zone of a crop often provide no visual signs, but yields, plant health and produce quality can be affected without the growers' knowledge that there is a problem with oxygen levels. Media based plants are just as prone to oxygen starvation in hydroponic systems as those grown in solution culture and no system of production is immune to oxygen depletion conditions. In a recent study, young cucumber plants were cultivated at different levels of oxygen in the supply of nutrient solution by Holtman et al. (2005). They found out that the oxygen levels inside the substrates are 0.5, 3.5 and 6 mg/l dissolved oxygen, which are defined as anoxia, critical oxygen level and sufficient oxygen for root systems respectively. Their results show that plants are grown using nutrient solution, which is oxygen saturated, have a higher leaf area comparing border plants, which are not subjected to continuous refreshment of nutrient solution. We conclude that the gas oxygen inside media is an important factor for plant development and roots may often suffer from hypoxia especially in the lower parts of slab unless water is refreshed regularly. So, oxygen supply for the root system inside substrates may be improved by optimalization of water management.

The increasing trend in tuber yield of different tuber sizes by applying aeration clarified that the tuber yield in aerated treatments had significant rise up to 162% with control group in class 3 and mainly class 4 of tubers (Figure 3). So, this theory can be stated that larger tubers have higher oxygen demand rather than normal substrate ventilation due to their more biomass and higher level of metabolism as well as less surface to volume ratio which is effective in oxygen diffusion to their internal mass. If oxygen in root zone is in a favorable condition, larger tubers will be produced; so potato plant will have more efficient and longer roots with higher number of stolon and can produce more and larger tubers, if root ventilation is reasonable with Moderate (25%, v/v media) aeration. This fact can be confirmed by comparing the successfully potato planting and producing of minitubers in aeroponics and bio-reactor systems (Akita and Takayama, 1994; Ritter et al., 2001; Factor et al., 2007).

Conclusion

Increasing in the O_2 concentration led to remarkable increase in size, number and quality of minitubers. Applying supplemental aeration was needed to increase tuber yield, dry matter and most of the growth parameters such as LAI, LAID, HI and root : shoot ratio significantly. Aeration provided favorable ventilation in root zone. Heavy media was more responsive to aeration technique than Light one. Thus, aeration at 25 and 12.5% (v/v/min) in Heavy and Light media respectively was considered as favorable level of aeration in rizo-sphere and had positive effect on tuberization. Therefore, the Heavy media is preferred to Light media when aeration facilities exist. This can result in promotion of potato seed industry.

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