Root oxygen use as a measure for ion uptake from slightly different nutrient solutions

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Root oxygen use as a measure for ion uptake from slightly different nutrient solutions

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Abstract

The current system of nutrient supply in horticulture may be improved by ion specific measuring in combination with real-time nutrient adjustments. Frequent measurements of the solution around the roots and related adjustments of the solutions supplied, will result in plant roots experiencing smaller fluctuations in nutrient concentrations and ratios than in the current system of analyzing and adjusting the nutrients once in 1-2 weeks. To find out how much effect on growth more frequent measurements and adjustments could have, we measured the oxygen uptake of roots growing in nutrient solutions of different ionic compositions. The assumptions underlying this approach are that nutrient uptake requires energy and the energy is generated by the oxidation of assimilates. Thus root oxygen use is a measure of the energy use for ion uptake and via assimilate use also related to above-ground production. To measure root oxygen use in optimal and slightly sub-optimal nutrient solutions, four air-tight vessels to grow plants in nutrient solutions were constructed. Each vessel was connected to an airbag and a peristaltic pump to circulate the air. Consequently tomato, cucumber and sweet pepper young plants were grown on optimal and slightly sub-optimal nutrient solutions. Oxygen sensors recorded the decrease in oxygen concentration. So far, a 5-18% higher oxygen consumption was found for a NO_3 solution as compared to a NO_3/Cl solution. In conclusion root oxygen use for a whole root system during growth can be accurately measured and as a first result the uptake of nitrate requires more energy than the uptake of chloride. Consequences for the application of ion specific measurements as well as for the definition of optimised target solutions for the root environment are discussed.

Keywords: root respiration, assimilates, ion specific meter, oxygen sensor

INTRODUCTION

The Dutch greenhouse industry has the goal to reach zero emissions of nutrients and pesticides by 2027 (Van Paassen and Welles, 2010). Nutrient unbalances and the accumulation of unwanted elements are the principal reasons for growers to discharge drainage water (Blok et al., 2012). Nowadays, the nutrient solution is corrected every 7-14 days based on drainage water samples analyzed by external laboratories. However, this method is inaccurate because the ions intake may vary in the course of a single day according to changes in climate conditions and within 14 days due to the development stage (Voogt and Van Os, 2012). Ion specific measurements have been suggested as a possible solution to increase accuracy (van den Boogaard et al., 2003). Such ion specific measurements can be operated at a nursery and could potentially give fast information on the nutrient dynamics in the solution and push toward a more accurate control of the fertigation (Bamsey et al., 2012). Several technologies can measure ion-specific concentrations. The Micro HPLC (high-performance liquid chromatography or capillary electrophoresis) is a promising one. In particular, the machine "Celine" by The Sensory Factory (TSF) is now at an advanced stage of development (Blok et al., 2012). Apart from the emission reduction, other potential benefits of the ion specific measurements are: the lower costs for water and nutrients due to longer recirculation and therefore, a higher water and nutrients efficiency; the business opportunities for service industries (manufactories, sensor technologies, consultancy); the higher yield due to higher plant energy efficiency (Blok et al., 2012). Acquiring a higher yield

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is crucial for the practical application of such technology in the horticultural industry.

However, it is still uncertain whether more accurate nutrient solutions translate into growth effects (Marcelis et al., 2003; Elings et al., 2004; Massa et al., 2011). Our present understanding is that when plants absorb nutrients, they burn assimilates with oxygen (Bouma and De Visser, 1993; Nakamura and Nakamura, 2016). When plants have to deal with nutrient unbalance, even more assimilates may be burnt to maintain the equilibrium uptake. Hence, fewer assimilates can be invested above ground, resulting in a production loss (Rewald et al., 2016). Following this line of thought, oxygen consumption by the roots may be used as proof of higher energy consumption at unbalanced nutrient ratios (Cannell and Thornley, 2000).

The object of the present study was to establish that root respiration allows to measure the oxygen need of the plant roots to absorb specific nutrient solutions. By doing so, the study aimed to indirectly prove the negative effect of nutrients unbalance on plant growth, and to show the applicability of the ion specific measurement as a technique to improve yield and the environmental sustainability of greenhouse horticulture. To do this, individual plants were grown inside hermetically closed vessels filled with a nutrient solution (no solid substrate). The oxygen consumption was measured as a drop in oxygen level in the vessel.

MATERIAL AND METHODS

To measure the oxygen use by plant roots when taking up nutrients, an airtight closed system was made to study roots without outside air interference (Blok and Gérard, 2013). The system consisted of a 4.0 L vessel with 2.0 L of nutrient solution, one plant, an 5-L air storage bag and a peristaltic pump that circulated the air through the air containing part of the vessel (Figure 1). Several openings in the lid were made. The plant stem went through the middle of the lid (±1 cm) and the remaining hole was closed by using a malleable sealing paste (terostat VII, Gerstaecker), preventing air exchange between the vessel and outside air while still allowing the stem to increase in diameter. The lid contained 3 smaller openings: one air inlet; one air outlet, both connected to the peristaltic pump an air storage bag within the closed system; and one opening for the oxygen sensor (Sendot Research, Houten, The Netherlands), which was placed in the nutrient solution to measure the real-time oxygen concentration. The oxygen consumption was calculated by using the slope of the regression line of oxygen decrease over time (Figure 2). A continuous supply of air bubbles in the nutrient solution was maintained with a stainless steel air stone connected to the air inlet, all part of the closed system (Figure 1). A stirrer at the bottom inside the vessel was used to create water flow. All tubes in the system had an inside diameter of 6 mm.

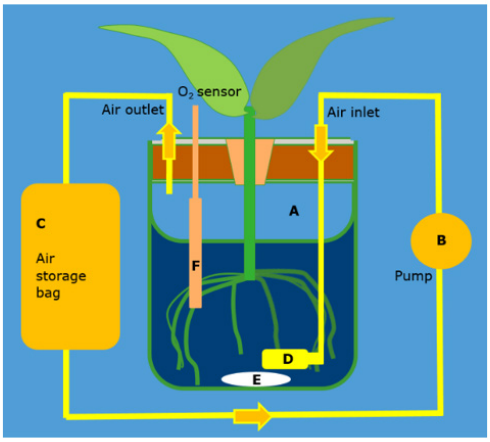

Figure 1. Closed system with an air-tight vessel containing nutrient solution, plant roots and air (A), a peristaltic pump (B) and an air storage bag (C) with inside the vessel an air stone (D), a stirrer (E) and an O_2 sensor (F).

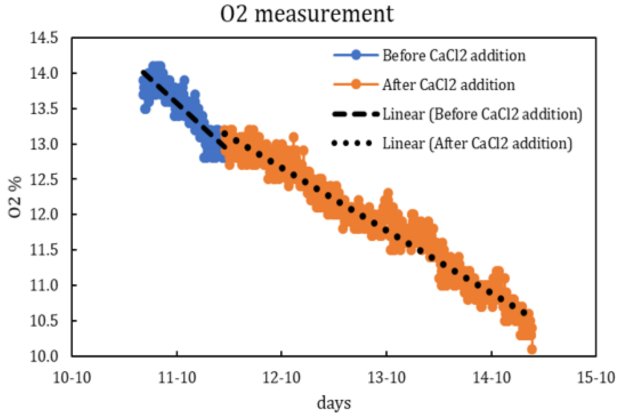

Figure 2. O_2 measurement in the root environment before and after $CaCl_2$ addition in a pepper plant.

First a smaller test was performed with a single bell pepper plantlet with two oxygen sensors in one vessel which measured the oxygen consumption in air and water in a standard nutrient solution (Figure 3). The standard nutrient solution is based on the average nutrient take-up of a certain crop per amount of water (De Kreij et al., 1999). The oxygen concentration started at saturation (21 mg L^{-1} in air, >8 ppm in water). The first test consisted of short runs measuring oxygen concentration to illustrate the effect on oxygen consumption of a growing plantlet over time. Thereafter, the effect of bubbling air in the nutrient solution on oxygen consumption in water and air was tested comparing T1 and T2 without bubbling air and T3 with bubbling air using an air stone, all in a closed system to get an impression how oxygen is distributed between air and water.

Figure 3. Vessel with pepper plant, used in the first tests.

The main experiment was performed with 4 closed systems with alternating two nutrient solutions (Figures 1 and 4).

NO_3/Cl – cucumber

Each system contained a vessel with a cucumber plant and an oxygen sensor in the nutrient solution. Two different nutrient solutions were made: a standard reference solution (Ref) and an alternative solution (Alt) containing more Cl^- and less NO_3^- (Table 1). At the start, 2 vessels were filled with Ref solution and 2 vessels with Alt solution. The oxygen concentration started at saturation (>8 ppm). Each system was closed and the oxygen concentration gradually decreased over time while the roots consumed oxygen. After 3-7 days, depending on plant height, the oxygen concentration reached the minimum set at <4ppm. Below this concentration, roots were believed to react to sub-optimal oxygen levels by decreasing the slope in oxygen use. On reaching 4 ppm oxygen, nutrient solutions in all vessels were changed and re-saturated with oxygen. The 2 vessels which received the Ref solution at the start now received the Alt solution and vice versa. The systems were closed again. The test was repeated with 4 new cucumber plants.

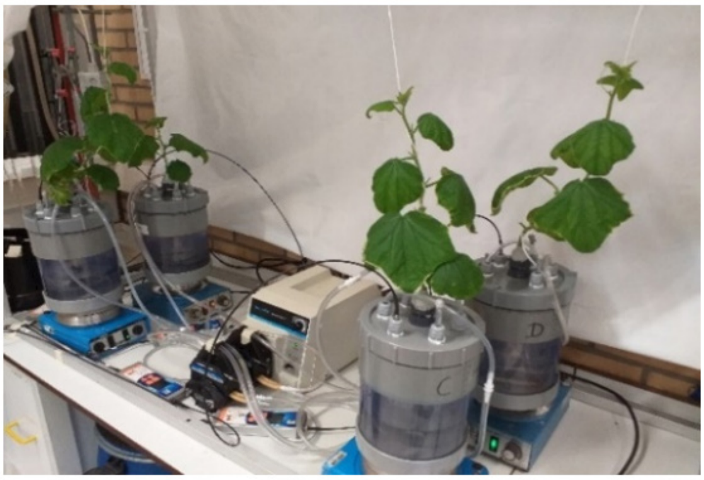

Figure 4. Four closed systems as Figure 1 with four cucumber plants. The O_2 sensor is inserted in the nutrient solution of each vessel.

Table 1. The concentrations of NO_3^-, Cl^- and SO_4^{2-} of the reference (Ref) and alternative (Alt) solutions.

Start solution	EC (mS cm^{-1})	NO_3^- (mmol L^{-1})	Cl^- (mmol L^{-1})	SO_4^{2-} (mmol L^{-1})
Ref NO_3/Cl	2.6	16.9	<0.1	3
Alt NO_3/Cl	2.6	7.3	10.4	2.4
Ref NO_3/SO_4	2.7	16.2	0.1	4.4
Alt NO_3/SO_4	2.7	8.5	0.1	9.9

NO_3/Cl – tomato

The same starting solutions, Ref NO_3/Cl and Alt NO_3/Cl (Table 1) were used in this next test with tomato plants under constant conditions (light: 16 h d^{-1} 600W Full spectrum LED light (LUMERI PRO-LINE 600), T: 20°C, RH: 70%). Instead of swapping once, we now alternated the nutrient solution 5 times, totaling 6 runs.

NO_3/SO_4 – tomato

New starting solutions were made and tested with tomato plants. The Alt solution contained less NO_3^- (8.5 mmol L^{-1}) and more SO_4^{2-} (9.9 mmol L^{-1}) compared to the Ref solution (16.2 mmol L^{-1} NO_3^-, 4.4 mmol L^{-1} SO_4^{2-}) (Table 5). Again the solutions were alternated between the vessels. Since it had been found that root damage occurred when using a magnetic stirrer, waterflow was now created by water jets from 2 vertical tubes parallel to the vessel wall driven by a peristaltic pump.

RESULTS AND DISCUSSION

Oxygen consumption of the growing plantlet's roots increased over time (Tables 2 and 3). In Table 3 the rate of oxygen decrease from the solution and the air was calculated separately. This showed that bubbling air through the solution at T3 lowered the decrease in oxygen from the solution, even though the total oxygen uptake was a lot higher (Table 3). This illustrates that active air supply to a solution is needed to prevent depletion of the solution. It also shows depletion of a solution may occur even though air with a sufficient amount of oxygen is circulating over the solution surface.

For the NO_3/Cl runs the oxygen consumption by the roots increased from the Alt to Ref solution and decreased from the Ref to Alt nutrient solution by 17% for cucumber and 9% for tomato runs (Table 4). The slope of the decrease in oxygen consumption is taken as an indication of oxygen consumption in mg O_2 h^{-1} g^{-1} FW roots (example in Figure 2). Finally, the change in slope is expressed as a %, of which the value of the denominator is always taken as the value of the standard nutrient solution. This means that the roots in the treatment with the nitrate-rich nutrient solution are 9-17% more energy-consuming than in the treatment where part of the nitrate is replaced by chloride.

Table 2. First results of measurements of O_2 consumption per hour of a bell pepper plant with approximately 10 g of fresh root mass at the start.

Date	O_2 consumption (mg h^{-1})	Particularity
11 Sep	2.0	1.0 L air 2.0 L water
12 Sep	2.7	1.0 L air 2.0 L water
13-14 Sep	2.8	4.0 L air 2.0 L water
18-22 Sep	3.3	4.0 L air 2.0 L water
25-30 Sep	4.4	4.0 L air 2.0 L water

Table 3. O_2 consumption from air, water and total for a root system of a bell pepper plant on T1 (16 OCT), T2 17 OCT), and T3 (18 -21 OCT). An air flow rate of 25 mL min^{-1} over the solution was used at T1 and T2 without a bubble stone. At T3, the air was passed through the solution with a bubble stone at an air speed of 25 mL min^{-1}.

O_2 consumption (mg h^{-1}) measured in:	T1	T2	T3
Air	3.8	4.0	6.2
Liquid solution	0.29	1.12	0.09
Total	4.10	5.07	6.29

Table 4. Difference in O_2 consumption by plantlet roots expressed in Δmg h^{-1} and % between alternative (Alt) and reference (Ref) nutrient solution of NO_3/Cl runs (using Table 5a starting concentrations). Cucumber runs: 2 NOV until 6 DEC 2019. Tomato runs: 10-25 March 2020.

O_2 consumption difference	From Alt to Ref		From Ref to Alt
Cucumber			
Δ mg h^{-1}	+0.87±0.15		-0.39±0.11
%	+22%±3%		-13%±3%
Relative %		17%	
Tomato			
Δ mg h^{-1}	+3.19±1.01		+0.84±0.67
%	+36%±9%		+18%±8%
Relative %		9%	

The NO_3/SO_4 runs were performed with tomato plantlets. In the NO_3/SO_4 runs, the Alt solution where part of the nitrate was replaced by sulfate showed a higher oxygen consumption by the roots compared to the Ref solution, expressed as -20% lower oxygen consumption by the Ref solution (Table 5), i.e. the opposite of the NO_3/Cl runs.

A possible explanation may be that NO_3^- is a luxurious ion (on average 30% more in nutrient solution present than needed for the plant, because more cations the plant needs to take up compared to anions). SO_4^{2-} is a bivalent ion, which may require more energy to take up compared to monovalent Cl^- (Marschner, 2012), resulting in higher oxygen consumption. The 7.3 mmol L^{-1} NO_3^- may result in shortage of time, which may result in less ion uptake and a decrease in oxygen consumption.

Table 5. Comparison between NO_3/Cl and NO_3/SO_4 runs of starting concentrations (mmol L^{-1}) of reference and alternative solution (EC 2.7, pH 5.8) and the difference in O_2 consumption between Ref and Alt solution by the roots of tomato plantlets.

Comparison	NO_3/Cl		NO_3/SO_4	
	NO_3^-	Cl^-	NO_3^-	SO_4^{2-}
Reference	16.9	<0.1	16.2	4.4
Alternative ratio	7.3	10.4	8.5	9.9
O_2 consumption (variation)		+12% (5-18%)		-20% (8-33%)

CONCLUSIONS

This article shows the first steps in measuring oxygen consumption by roots in different nutrient solutions. So far, we have seen an effect on oxygen consumption by partly replacing one nutrient by another nutrient (NO_3^- to Cl^- and NO_3^- to SO_4^{2-}). Partly substitution of NO_3^- by Cl^- resulted in 12% less oxygen consumption, while partly substitution of NO_3^- by SO_4^{2-} showed a 20% higher oxygen consumption. This relatively large effect is interpreted as proof of the relevance of ion-specific control of the root solution, even if the levels of ions present in the nutrient solution are sufficient to prevent nutrient deficiencies. The need for ion-specific control also justifies continuous attention for the use of ion-specific measuring equipment such as the “Celine” apparatus. Experiments with suboptimal but not growth limiting ratios of other elements will follow just as experiments with ion-specific measuring equipment.

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