D3.4 – Evaluation of a nitrate sensor for Dutch greenhouses at semi-practical scale

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List of abbreviations

AGRINUPES	Integrated monitoring and control of water, nutrients and plant protection products towards a sustainable agricultural sector.
DOC	Dissolved organic matter
ICP	Inductively Coupled Plasma
ISE	Ion selective electrodes dip probes
PLS	Partial Least Squares
UV-VIS	Ultraviolet-Visible

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Summary

According to the EU Water Framework Directive the emission of nutrients and plant protection products to water bodies should be reduced dramatically. In practice, threshold values for water bodies are exceeded due to leakages, discharges and unexpected problems from greenhouse facilities. Nitrate eutrophicates surface water. In soilless cultivation in The Netherlands, recirculation is mandatory, and discharge is regulated to yearly crop specific nitrogen concentrations. However, water authorities still measure exceedances in surface water, leading to an interest in having real-time and reliable measurements of nitrogen or nitrate for a faster intervention.

Adjustments in nutrient recipes in Dutch greenhouses are based on a 7-14 days analysis of drain water by sending it to a laboratory. This practice is suboptimal since the loss in nutrients is higher than necessary and it might even reduce crop development. Besides it is time consuming and often expensive. In areas where the results of the laboratory are not quickly available, a nitrate sensor may even play an important role in creating the right solution for the plants. With a real-time measuring nitrate sensor, it is possible to fertigate more accurately which increases the harvest/quality of the crop with a lower environmental footprint. Besides, such a nitrate sensor has also a large scientific value since the dynamic nitrate uptake by the crop can be studied at a scale of minutes instead of days.

A start has been made to analyse available methods for measuring nitrate. As a result, we will present an innovative optical fibre-based nitrate sensor using an algorithm which has been tested in a greenhouse setting at field conditions. Several tests are conducted which show that the nitrate measurement is not influenced by EC or ionic composition. Dissolved organic carbon and turbidity have a small influence on the nitrate measurement, but all the results are within the desired 10% accuracy. However, more extensive tests should be performed to get a better understanding of the effect of dissolved organic carbon and turbidity on the nitrate measurements. This sensor will enhance the efficiency and safety of greenhouse systems with respect to water reuse and recycling.

The work reported in this report will be presented at the 3rd International Symposium on Soilless Culture and Hydroponics, in Limassol (Cyprus) on 21-24 March 2021. Therefore, only after April 1st, 2021 the content of this deliverable will be available as a public document.

1. Introduction

Worldwide, in many regions with protected horticultural activities, threshold values for nitrate in water bodies are exceeded due to leakages, discharges and unexpected problems from greenhouse facilities. As an example, in the South-East of Spain aquifers are contaminated with nitrate leached from greenhouses which predominantly contributes to the eutrophication of surface water (Thompson et al., 2020).

In the Netherlands recirculation is mandatory for soilless cultivation systems and discharge is regulated according to crop specific nitrogen concentrations. Despite these regulations water authorities still often measure exceedances of nitrate in surface water in the vicinity of greenhouses (van der Salm et al., 2020).

Nitrate levels in drainage and discharges from greenhouses can be minimized by monitoring the nitrate concentration in the greenhouse extensively. Adjustments in nutrient recipes in greenhouses are based on weekly/bi-weekly laboratory analysis of the nutrient solution around the roots. This practice is suboptimal since nitrate levels at the roots deviate from the desired values in such a way that nitrate losses are higher than necessary which might even reduce crop development (Gieling et al., 2012). In addition, laboratory analysis can be time consuming, expensive or not even available in some areas. With a real-time measuring sensor for nitrate it is possible to optimize fertigation which increases the harvest or quality of the crop with a lower environmental footprint.

Commonly used in situ sensing technologies for real time analysis of nitrate in aqueous solutions are Ion Selective Electrodes dip probes (ISE) and optical sensors based on Ultraviolet-Visible (UV-VIS). Spectroscopy. UV-VIS spectroscopy has shown to be more effective over ISE sensors since the measurements are hardly influenced by pH, temperature or other anions (Edwards et al., 2001; Yeshno et al., 2019).

UV-VIS spectroscopy measures the amount of UV-light absorbed by an aqueous sample in a cuvette. The absorbance of UV-light by the sample depends on the concentrations of the absorbing components in the solution. Several methods exist to determine the nitrate concentration from the measured absorbance. Most are based on simple direct linear models and use the strong absorbance band of nitrate around 200-220 nm and 300-310 nm which are, respectively, more sensitive for low nitrate concentrations and nitrate concentrations higher than 0.2 g/L (Thomas and Burgess, 2017; Yeshno et al., 2019).

Dissolved organic matter (DOC) strongly interferes with nitrate analysis by UV-VIS spectroscopy since it also absorbs UV-light. Therefore, direct linear models cannot be applied in this case. A two-wavelength approach can account for DOC by correcting the measured absorbance at 200/220 nm with an absorbance measurement at 275/300 nm (Armstrong, 1963; Edwards et al., 2001). In addition, several multilinear regression models (PLS, PCA) in combination with pre-treatment methods have been developed to exploit the entire UV spectrum in order to deal with interferences of DOC on nitrate detection (Karlsson et al., 1995; Thomas et al., 2014).

So far UV-VIS spectroscopy has shown to be an effective and simple method for the detection of nitrate and accounting for DOC in natural water bodies, waste water and soil pore moisture without the use of any reagents (Avagyan et al., 2014; Causse et al., 2017; Rennie et al., 1979; Sarraguça et al., 2009; Thomas and Burgess, 2017; Yeshno et al., 2019). The existence of a method to measure nitrate in different hydroponic solutions and accounting for interferences (e.g. DOC) has not been described yet. Therefore, we present a novel comprehensive approach to measure nitrate in hydroponic solutions with UV-VIS spectroscopy which has the potential to enhance the efficiency and safety of greenhouse systems with respect to water reuse and recycling.

2. Materials and Methods

Experimental Setup

UV-VIS spectroscopy is used to measure nitrate concentrations in greenhouse hydroponic solutions. The solution is pumped through a flow cell containing a 10mm cuvette. The measuring system contains a deuterium light source (Ocean Optics model DH-2000-BAL), a spectrometer (Ocean Optics model HR4000) with a slit reflection probe and optical fibres. It measures the light intensity between 187 and 667 nm.

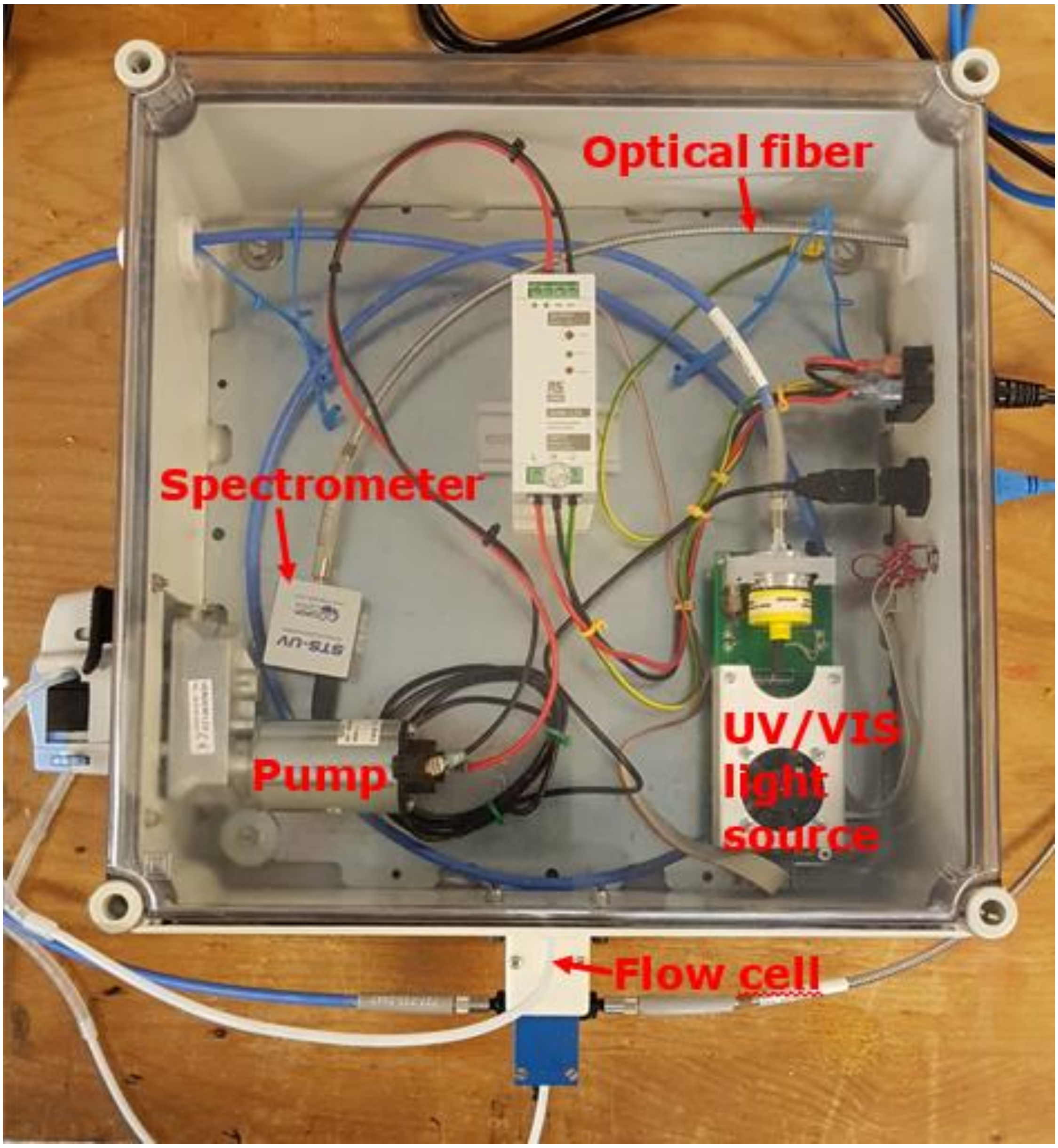

Figure 1. The UV-VIS spectroscopy system containing a pump, optical fibres, a spectrometer, a and a light source.

Since hydroponic solutions contain various ions which might cause interferences with the nitrate measurement, a Hoagland solution (Hoagland et al., 1938), which is one of the most standard hydroponic solution, was used as a reference solution (Table 1).

Table 1. The composition of the Hoagland solution.

Element	Concentration	Unit
EC	1.0	mS/cm
pH	5	-
NH4	<0.1	mmol/l
K	1.1	mmol/l
Na	<0.1	mmol/l
Ca	2.5	mmol/l
Mg	2.2	mmol/l
NO3	5.2	mmol/l
Cl	0.2	mmol/l
SO4	2.1	mmol/l
HCO3	<0.1	mmol/l
P	1	mmol/l
Fe	26	µmol/l
Mn	7.4	µmol/l
Zn	0.9	µmol/l
B	44	µmol/l
Cu	0.4	µmol/l
Mo	0.37	µmol/l

First 20 samples were measured to examine whether the measured absorbance values at all wavelengths were reproducible. Thereafter a total of 74 samples were collected for a period of 7 months consisting of supply and drain solutions from growing systems with various crops (64 samples) and consisting of surface water from greenhouse areas. At the day of measuring a duplicate sample was sent to an accredited laboratory which measured the nitrate concentration by Inductively Coupled Plasma (ICP). With the known nitrate concentrations, the measured light intensities of the samples and the reference solution, the system was calibrated, which process is described in the next section.

Theory

From the measured light intensities of the samples and the reference solution the absorbance was calculated via the Lambert-Beer equation:

$$A = -\log_{10}\frac{I}{I_0},$$

where I represents the light intensity after passing through the water sample, and I_0 the measured light intensity after passing through the reference solution. Lambert-Beer's law describes a linear relation between the absorbance and the concentration of a pure component. Since there are many components present in hydroponic solutions and the absorption of each individual component is unknown, a direct linear model cannot be used.

Partial Least Squares (PLS) is a multivariate statistical method to determine the concentration of a single component in complex solutions (Brereton, 2003; Geladi and Kowalski, 1986; Haaland and Thomas, 1988; Langergraber et al., 2003; Lepot et al., 2016; Mark and Workman, 2007; Sarraguça et al., 2009). Since greenhouse water contains several ions and organic components, like organic material from the substrate (coir and peat), plant protection products, root exudates, degrading parts from roots, leaves and fruits, PLS is used to set up the calibration.

To use PLS, the measured absorbance should be normalized to reduce overlapping peaks to allow for a better quantification of the individual components (Karlsson et al., 1995; Thomas and

Burgess, 2017). A common way for normalizing spectral data is to calculate the 2nd order derivative of the absorbance values according to:

$$\frac{d^2A_\lambda}{d\lambda^2} = \frac{A_{\lambda-h}+A_{\lambda+h}-2\cdot A_\lambda}{h^2}$$

with h as differentiation step (10 nm), A_λ the absorbance value at wavelength λ and $A\lambda\text{-}h$ the absorbance value at wavelength $\lambda\text{-}\ h$ (Thomas and Burgess, 2017).

With the 2nd order derivative of the absorbance values and the known concentrations of the 64 samples, a PLS1 regression was made which was used as calibration for the sensor system. The entire procedure of data progressing and executing the PLS regression was done with Matlab 2018b.

Validation

Since overfitting is a reported problem with PLS regressions (Crumpton et al., 1992), an experimental plan was set up containing 7 different tests to determine the accuracy of the calibration model, see Table 2.

Table 2. The description of the validation experiment.

Test	Description
A	a KNO_3 solution with different NO_3^- concentrations: 2, 5, 10, 15 and 25 mmol/l at the same EC of 3.0 mS/cm. KCl has been used to reach the desired EC.
B	a KNO_3 solution with a constant NO_3^- concentration of 15 mmol/l, but with different ECs: 2.0, 2.5, 3.0 and 3.5, 4.0. KCl has been used to reach the desired EC.
C	a standard tomato, cucumber and strawberry nutrient solution with the same EC and nitrate concentration which variates from Hoagland solution (table 1).
D	a standard tomato nutrient solution containing illite (6 mg/l), but with different concentrations of humic acid: 5, 10 and 15 mg/l.
E	a standard tomato nutrient solution containing humic acid (10 mg/l), but with different concentrations of illite: 3, 6 and 9 mg/l.
F	solutions of test D filtered.
G	solutions of test E filtered.

In each test the nitrate concentration was measured with the calibrated UV-VIS sensor and the measurement was compared with the result of the laboratory to determine the accuracy of the UV-VIS sensor. The samples in the above test were not included in the calibration. Test A and B were set up to test the interaction between the EC and the measurement of the sensor. The purpose of test C was to examine the influence of a different matrix of ions on the nitrate measurement. Samples of test D and E contain illite and humic acid to mimic respectively turbidity and organic material in greenhouse water (Van Ruijven et al., 2014). The results of test F and G was compared with test D and E to see the effect of suspended particles on the measurement. Based on communication with several growers, the desired accuracy of the sensor should be 10% in order to use the sensor for adapting the nutrient solution around the roots.

3. Results and Discussion

Calibration

After the repetitive measurements it become clear that the measured absorbance values below 260 nm and above 540 nm were not reproducible, so those wavelengths were excluded from the calibration. In Figure 2 the predicted concentration of nitrate in the 74 samples is plotted versus the reference concentration measured by the laboratory.

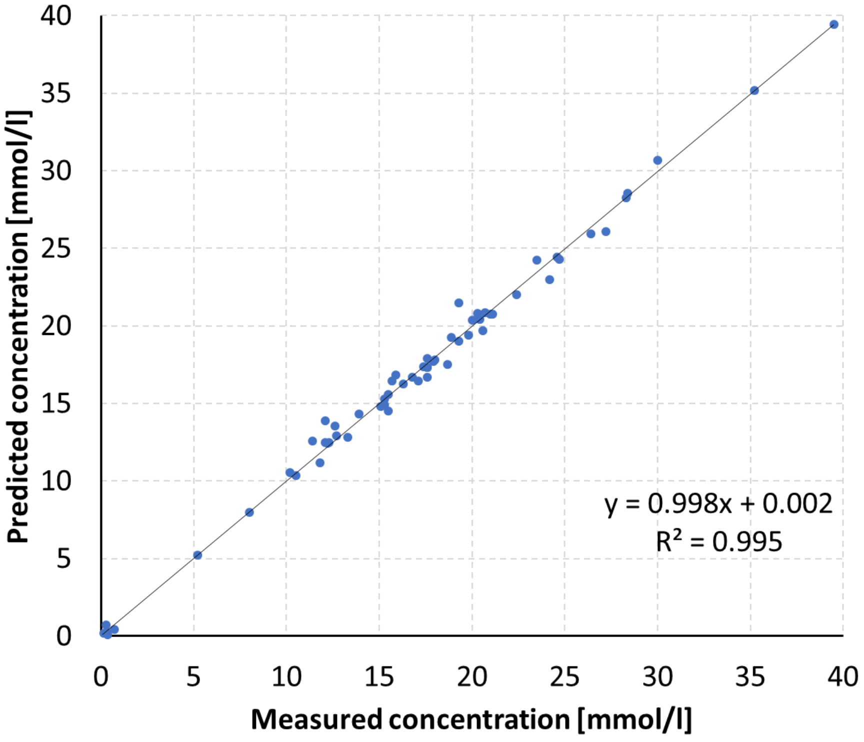

Figure 2. Correlation between measured nitrate concentration and predicted nitrate concentration.

The above figure clearly shows a very high correlation between measured and predicted concentrations of nitrate. To examine the performance of the calibration on unknown samples and to test the influence of EC, ionic composition, turbidity and DOC, the test in Table 2 was carried out.

Validation test

The results for test A are plotted below:

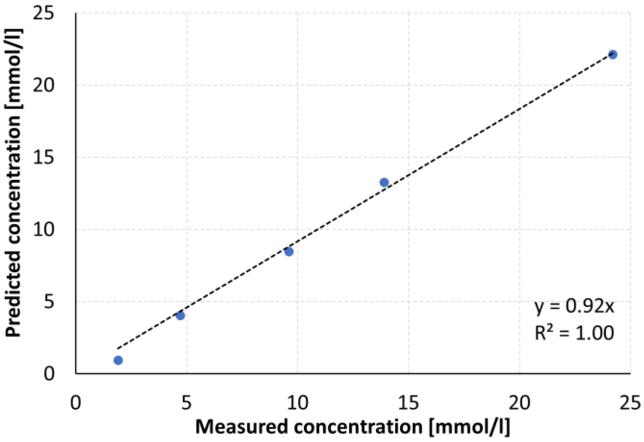

Figure 3. The predicted and measured nitrate concentration plotted for samples A1 until A5.

The above graph clearly shows that there is a very good correlation between the predicted nitrate concentration of the model and the measured nitrate concentration by the lab. The slope shows that the model underpredicts the nitrate concentration with 8% on average, but the accuracy is lower for sample A1 which has a low nitrate concentration. The reason is that the model makes a correction for other supposed UV-light absorbing constituents, but since nitrate is the only constituent which absorbs UV-light in the samples results in a correction, which is not necessary.

In the next graph the results of test B are shown.

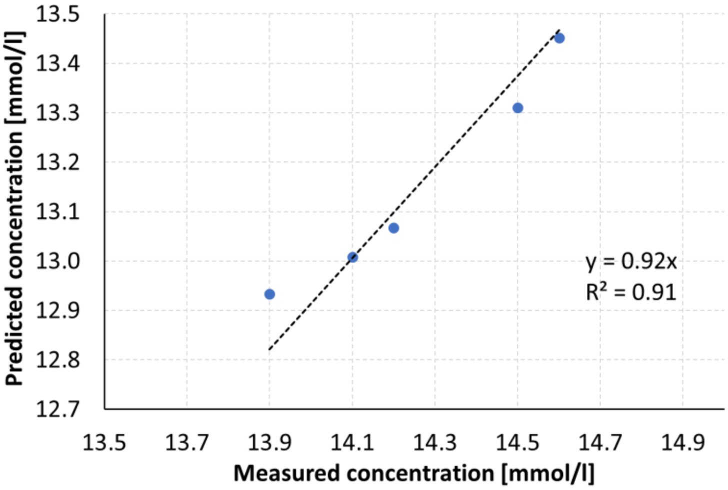

Figure 4. The predicted and measured nitrate concentration plotted for samples B1 until B5.

The above results also clearly show a strong correlation between the predicted and the measured concentration by the lab. Again, the model underpredicts the nitrate concentration with 8%. Since this underprediction was constant for all samples it is evident that the computation of the nitrate concentration by the model is not affected by the EC.

The figure below shows the results for test C:

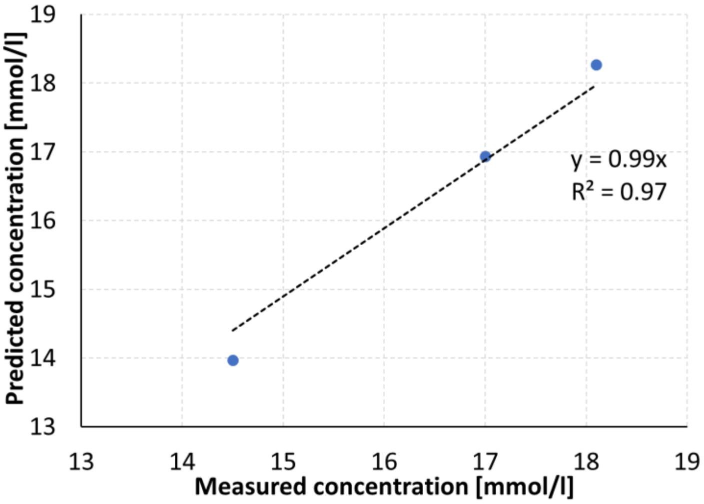

Figure 5. The predicted and measured nitrate concentration plotted for samples C1 until C3.

The above graph clearly shows that the model is able to predict the nitrate concentration in the three hydroponic solutions which contain no organic matter. The model can clearly distinguish 18, 17 and 14 mmol/l. Moreover, the ionic composition of the samples was slightly different, but the model is still able to predict the nitrate content in the samples.

As mentioned before hydroponic solutions can be turbid and can contain dissolved organic matter. To see what the influence of organic matter is test D was carried out and the results are shown the graph below:

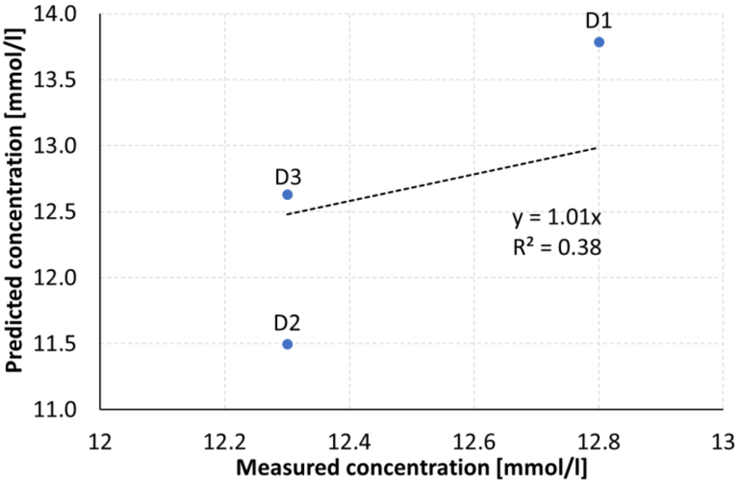

Figure 6. The predicted and measured nitrate concentration plotted for samples D1 until D3.

The above graph shows that there is no correlation between the predicted and measured concentration. Even though D2 and D3 have the same nitrate concentration the model predicts a higher nitrate concentration in sample D3 which has the highest humic acid (DOC) concentration. So, the presence of DOC has a significant influence on the prediction of the model. The dataset used for the calibration contains relatively more samples of supply water than of drain water, so it is to be expected that the results from test C are better than the results of test D.

Even though the low correlation between the measured and predicted concentration, the accuracy of the above samples is still within the desired 10% so the above results are very promising. The accuracy of the model can be increased in the future if more drain samples containing dissolved organic matter will be used for the calibration.

In the graph below the results of test E (varying turbidity levels) are shown:

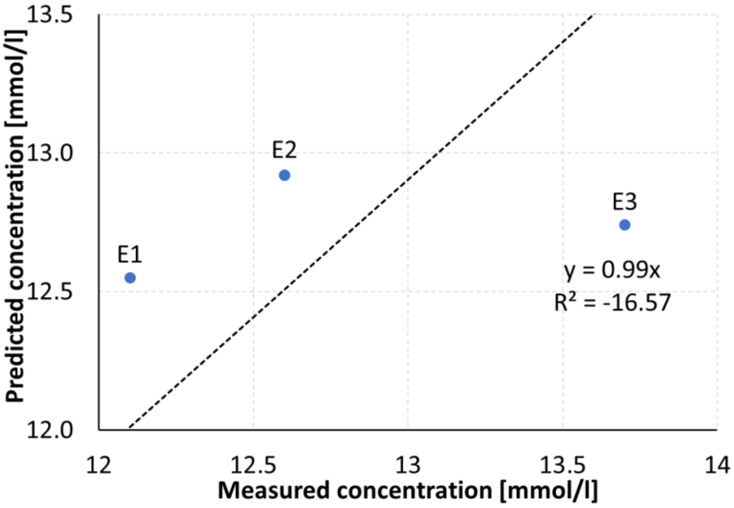

Figure 7. The predicted and measured nitrate concentration plotted for samples E1 until E3.

The above graph shows no correlation between the predicated and measured concentration. Even though the turbidity and nitrate concentration increase from E1 to E3, the predicted concentration by the model remains constant (±12.8). This could indicate that turbidity masks the nitration prediction. So, despite of an accuracy below the desired 10% it cannot be confirmed that the model can cope with varying turbidity levels perfectly. More tests should be performed to examine the influence of turbidity and to improve the model.

To see the effect of filtering the sample test F and G are performed:

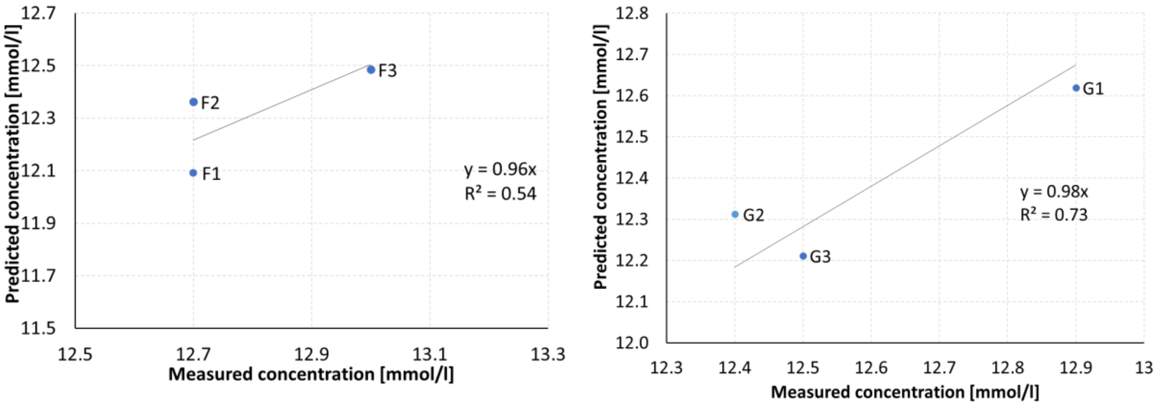

Figure 8. The predicted and measured nitrate concentration plotted for samples F until G.

The results of test F and G are significantly better compared to test D and E since the accuracy lies between the 5 and 1% so filtering the samples clearly improves the results. Test E and D were carried out three weeks later than test F and G which makes it more difficult to examine the effect of filtering the samples, since the nitrate concentration also changed over time.

Especially the measured concentration by the laboratory between E3 and G3 is significantly different. From experience we also know that the results of the analytical labs are not always 100% accurate, so this should also be kept in mind.

In general, turbidity and dissolved organic matter content in the hydroponic samples D, E, F and G were mimicked with respectively illite and humic acid. Here it is assumed that the absorption spectrum of these constituents is the same as for dissolved organic matter found in hydroponic solutions but this should be confirmed with more extensive tests.

The results of all the tests are summarized in Table 3.

Table 3. Summarized test results of the of the seven types of solutions (see Table 2) when comparing the laboratory analysis with the predicted nitrate values. (*) Average values per Solution.

Solution	Model (mmol/l)	Lab (mmol/l)	Difference (%)
A1	0.9	1.9	-50.2
A2	4.0	4.7	-14.3
A3	8.5	9.6	-11.8
A4	13.3	13.9	-4.5
A5	22.1	24.2	-8.5
A			**-17.9** *
B1	13.3	14.5	-8.2
B2	12.9	13.9	-7.0
B3	13.1	14.2	-8.0
B4	13.0	14.1	-7.7
B5	13.5	14.6	-7.9
B			**-7.8** *
C1	18.3	18.1	0.9
C2	14.0	14.5	-3.6
C3	16.9	17	-0.4
C			**-1.0** *
D1	13.8	12.8	7.7
D2	11.5	12.3	-6.5
D3	12.6	12.3	2.7
D			**1.3** *
E1	12.6	12.1	3.7
E2	12.9	12.6	2.5
E3	12.7	13.7	-7.0
E			**-0.3** *
F1	12.1	12.7	-4.8
F2	12.4	12.7	-2.7
F3	12.5	13	-4.0
F			**-3.8** *
G1	12.6	12.9	-2.2
G2	12.3	12.4	-0.7
G3	12.2	12.5	-2.3
G			**-1.7** *

The method explained in this paper can also be applied to measure nitrate in surface water. However, the absorbance band of nitrate around 200-220 nm is more sensitive for low nitrate concentrations, but the used UV-VIS system in this work cannot measure absorption in that range properly. So, in order to measure nitrate in surface water quantitatively some adaptations to the current system should be made. Nevertheless, the results of test A1 shows that it could be possible with the current model to measure at concentrations of 1 mmol/l (63 mg/l), which is close to the WHO standard for nitrate (50 mg/l). Therefore, the current model can also potentially be used as a qualitative method for nitrate measurements in surface water if more samples in the range of 0.5 -1 mmol/l NO_3^- are included in the calibration.

4. Conclusion

An advanced model has been developed which can be used to measure nitrate in hydroponic solutions with UV-VIS spectroscopy. To examine whether this calibration model can account for the most common interferences in hydroponic solutions, several tests were performed. The results show that the nitrate measurement is not influenced by EC or ionic composition. Dissolved organic carbon and turbidity have a small influence on the nitrate measurement, but all the results are within the desired 10% accuracy. However, more extensive tests should be performed to get a better understanding of the effect of dissolved organic carbon and turbidity on the nitrate measurements.

The results also show that the sensor is less accurate to measure nitrate in non-hydroponic solutions. This might indicate that a different approach is required to create a universal approach suitable to measure nitrate in hydroponic with very different compositions.

The UV-VIS spectroscopy system in combination with the developed model can be used for online monitoring of nitrate concentrations in hydroponic solutions, without the use of any reagents or need for recalibration, to enhance the efficiency and safety of greenhouse systems with respect to water reuse and recycling.

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