

Soil oxygen and water dynamics underlying hypoxic conditions in the root-zone of avocado irrigated with treated wastewater in clay soil

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ABSTRACT

Alternative water sources for agriculture are in high demand in a world with diminishing fresh water (FW) availability. Treated wastewater (TWW) offers a reliable alternative, but increasing evidence is pointing to damage to TWW irrigated orchards planted in clay soils related to soil hypoxia. The mechanisms responsible for this hypoxia have not been extensively studied to date. The aim of this study was to elucidate meaningful insights into the mechanisms responsible for the hypoxia in TWW irrigated orchards planted in clay soils using a novel approach whereby parameters describing the soil oxygen and water temporal dynamics are analyzed. To that end, soil oxygen and soil water tension (SWT) measurements from a two year field experiment comparing TWW to FW irrigation in an avocado orchard planted in a clay soil (60 % clay) were used. The deterioration in oxygen levels occurred as the irrigation season progressed, and the oxygen availability decreased with depth (10–35 cm depth). During August–September, when the lowest oxygen concentrations were measured, the water content at which oxygen supply matched oxygen consumption at 35-cm depth did not differ between treatments (~50 mbar), but the TWW irrigated soil experienced ~47 % more time at wetter conditions. Lower oxygen decline rates were observed in the TWW irrigated plots which countered the previous concept that TWW leads to increased soil oxygen consumption. The findings point towards the rate of soil drying as the prime cause of differences – TWW irrigated plots dried in a rate which is nearly 4-times smaller than that in FW irrigated plots during the dark and light hours, reflecting slower drainage and water uptake respectively. It is suggested that soil hypoxia induced by the low soil drainage in TWW irrigated clay soils impairs tree water uptake, which further hinders the soil oxygen levels. Based on these results management tools are suggested to allow sustainable irrigation with TWW in the future. Furthermore, the work demonstrates how analysis of parameters describing the oxygen hourly changes can be utilized to gain mechanistic insights into processes affecting the oxygen regime in the soil.

1. Introduction

Fresh water (FW) scarcity is becoming a problem of increasing significance globally. Use of treated wastewater (TWW) as a main source of water for agriculture offers a solution for irrigation, turning this waste product into a valuable resource. TWW not only supplies a reliable source of water but also enables nutrient recycling, leading to reduced fertilizer requirements (Bar-Tal, 2011). However, in clay soils irrigated with TWW, damage has been reported in orchards of subtropical species such as citrus and avocado with drops in yield of up to 40 % in Hass avocado (Assouline et al., 2015). These crops are prime candidates for cultivation in water scarce countries for which TWW irrigation may be suitable (Rodríguez Pleguezuelo et al., 2018), and so investigating the

sustainability of TWW orchard-irrigation is a matter of priority.

In previous work of our group, we studied the effect of TWW on the oxygen conditions in the root zone of an avocado orchard planted in a clay soil (Yalin et al., 2017). In that work it was shown that the root zone of TWW irrigated trees reached lower minimum oxygen levels and experienced overall longer periods in low oxygen levels as compared with FW irrigated trees. We furthermore found a negative correlation between yield and the duration of oxygen levels below 10 % V/V (widely considered to be a threshold of oxygen damage to plants). Research in citrus comparing physiological parameters of the trees in different soils has further substantiated that the damage associated with TWW was more pronounced in a clay soil compared to a sandy loam soil (Paudel et al., 2016) and in later publications the damage was linked to lack of

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oxygen (Paudel et al., 2019, 2017).

The aim of the work described here was to identify the edaphic conditions which act to create hypoxic soil conditions in TWW irrigated orchards planted in clay soil by investigating the oxygen dynamics (i.e. the changes in oxygen by time) and its relations to the soil water regime (i.e. water input and water content).

The soil oxygen dynamics, i.e. the change in oxygen by time, can be described as a simple mass balance:

$$\frac{\Delta \text{Oxygen}}{\Delta \text{time}} = \frac{\text{Oxygen Supply} - \text{Oxygen demand}}{\Delta \text{time}} \quad (1)$$

Where Δ Oxygen is the change in O_2 amount in a given soil volume, oxygen supply is the amount of O_2 entering that soil volume and oxygen demand is the O_2 lost from that soil volume, in a given time (Δ time). Oxygen supply is driven predominantly by O_2 diffusion from the atmosphere into the ground, and since O_2 diffusion rate through water is ~ 4 orders of magnitude smaller than in air, is a function of the amount and size of air filled conductive pores. The oxygen demand is mainly a result of biological respiration and is thus a function of root and soil biota respiration rate (Ben-Noah and Friedman, 2018).

The elevated SAR levels in TWW are known to degrade soil structure and to thus cause narrowing of the soil pores (Gharaibeh et al., 2016), decreasing oxygen penetration to dry soil (Horn and Smucker, 2005). These changes related to SAR are also known to cause poor water infiltration into the soil and higher retention of water in low soil water tensions (Assouline et al., 2016; Assouline and Narkis, 2011; Schacht and Marschner, 2014), which may impede oxygen supply into the soil after irrigations. The addition of a constant supply of organic matter with TWW may increase microbial activity and consequently is expected to increase soil respiration rates as was previously found in a clay soil (Paudel et al., 2018). It was thus hypothesized that the poor water conductivity and the degradation of macro-pores in TWW irrigated soil would retard oxygen supply to the soil depth while the amelioration of microbial activity by the organic matter inputs would accelerate oxygen consumption and increase the oxygen demand component, jointly leading to longer periods of hypoxia with lower oxygen levels. While, standard methods have been previously utilized to establish the occurrence of the individual changes in soil properties mentioned above, the integrated effect of these changes on the oxygen regime has not been assessed quantitatively under field conditions.

In this work a new approach was utilized whereby parameters describing the hourly changes in soil oxygen and water levels based on *in-situ* measurements are given a mechanistic interpretation based on a conceptual model. By using field measurements, we aimed to reach representing results of the complex field interactions affecting soil oxygen. The insights gained by this work lay the basis for designing reliable mitigation methods. Furthermore, currently there are no agreed indices to describe the soil oxygen regime (Ben-Noah and Friedman, 2018). The new approach described here defines measurable indices of oxygen in drip irrigated agriculture, which may be widely used as a diagnostic tool of soil oxygen conditions in the field.

2. Methods

2.1. Research site

The experiment was conducted during the years 2012–2014 in an avocado orchard in the Western Galilee Experiment Station, Acre, Israel (Acre, 32°55'52"N 35°06'23"E) and included two treatments – irrigation with treated wastewater (TWW) or with freshwater (FW). The soil in the site is a grumusol containing 60 % clay mostly smectite, a cation exchange capacity of 48.7 meq/100 g, 5.0 % organic matter by combustion, and 2.8 % calcium carbonate. Major soil indices for the individual treatments are described in Table 1. The selected orchard included Hass avocado, grafted on VC-66 West-Indian rootstock; these were chosen as the focus of research because they were most affected by TWW in previous experiments. Irrigation with either TWW or FW began since planting in 1995. TWW was supplied by the Acre wastewater treatment plant, which receives municipal wastewater from the city of Acre. The TWW goes through secondary treatment and is then stored in the Shomrat-Hazorea reservoir prior to distribution. Major indices of the irrigation water quality are detailed in Table 1. Irrigation was performed using 2 above-ground driplines (1.6 L/h) for each tree line. Irrigation amounts (1–5 mm a day) and timing (1–3 times a week) were according to the local agronomic recommendations, and were equal for the two treatments. Fertilizers were given through fertigation, and the N, P, and K concentrations in the TWW were supplemented with fertilizer up to an equivalent amount to the fertilizer applied in the FW irrigated plots (annual amounts average: 253 kg N/ha, 35.6 kg P/ha and 267 kg K/ha). Measurements were performed in three plots of each treatment. Each plot consisted of nine trees, from which only the central three trees were used for measurements. More details of the study site and field measurements are given in previous work (Yalin et al., 2017).

2.2. In-situ soil measurements

Soil oxygen and soil water tension (SWT) were measured in proximity to the drippers located ~ 50 cm south from each measurement tree trunk at depths of 10, 20 and 35 cm. Oxygen was measured using a galvanic type Pb cell (KE-50, Figaro engineering Inc. Japan). The oxygen sensors were placed in perforated plastic chambers (Fig. 1) and set into the soil. Data from the oxygen sensors was logged using data-loggers (Campbell Sci., Logan, UT) once per three hours and the raw reading were calibrated according to pre-installation calibration. Soil water tension was measured using transducer equipped digital tensiometers (Mottes Tensiometers Israel) and the data was logged once per half an hour directly onto the Mottes internet-site through remote transmission.

2.3. Conceptual model

To define indices describing the oxygen dynamics and to give the measured values a mechanistic interpretation, a conceptual model was formed. The oxygen galvanic cell readings at a certain depth represent the partial pressure of oxygen in the sensor-chamber (Fig. 1). We relate the changes in oxygen partial pressure in this chamber as a simplified mass balance between the rate of oxygen supply to the chamber volume

Table 1

Major indices of the soil (0-30 cm depth) and irrigation water quality in the Acre site in TWW vs FW irrigated plots. Electric conductivity (EC), Na, Ca, Mg, and Cl concentrations, sodium adsorption ratio (SAR), and biological oxygen demand (BOD); average and standard deviation of soil samples performed in November 2013 (N = 3) and periodical water samples performed between March and November during the years 2012-2014 (N = 22).

| | Treatment | EC dS/m | Na meq/L | Ca meq/L | Mg meq/L | Cl meq/L | SAR (meq/L) ^{0.5} | BOD mg/L |
|-------|-----------|-------------|-------------|-------------|-------------|-------------|-------------------------------|-------------|
| Soil | TWW | 1.54 ± 0.46 | 7.56 ± 1.52 | 3.95 ± 2.08 | 2.31 ± 1.01 | – | 4.42 ± 0.32 | – |
| | FW | 0.69 ± 0.11 | 1.53 ± 0.21 | 3.03 ± 0.86 | 1.79 ± 0.49 | – | 1.01 ± 0.24 | – |
| Water | TWW | 1.62 ± 0.13 | 6.61 ± 1.14 | 5.11 ± 1.61 | 2.98 ± 1.05 | 5.68 ± 1.07 | 3.34 ± 0.72 | 18.1 ± 16.2 |
| | FW | 0.86 ± 0.08 | 0.96 ± 0.18 | 5.18 ± 0.83 | 3.19 ± 0.59 | 2.78 ± 5.29 | 0.47 ± 0.08 | – |

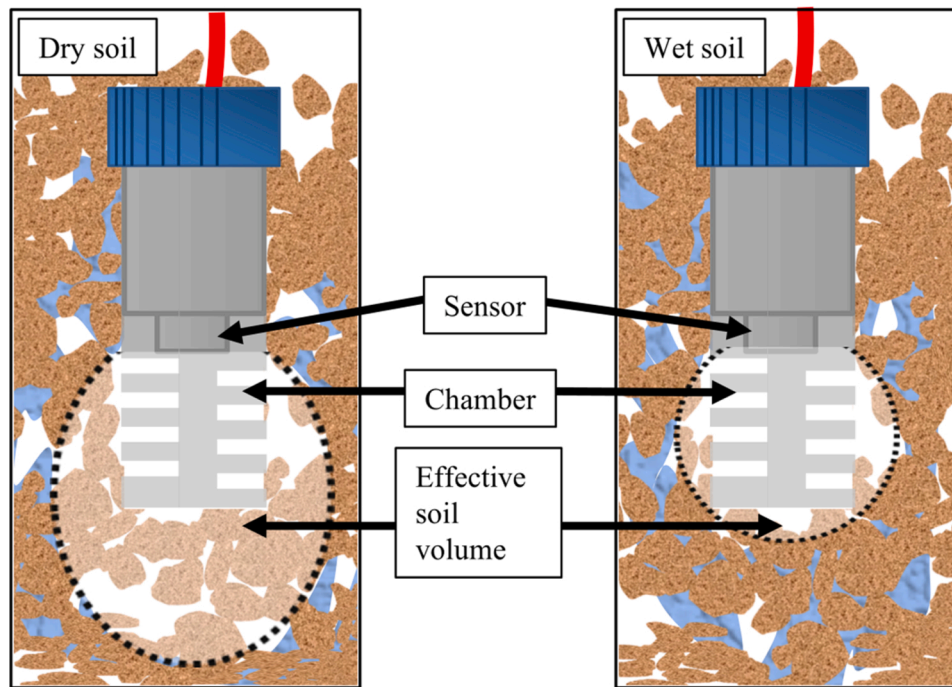


Fig. 1. Schematic representation of the effective soil volume (i.e. the soil volume connected by air filled pores to the measurement chamber), around the O_2 sensors under dry soil conditions (left) and wet soil conditions (right).

and the rate of oxygen consumed from the same chamber, as described in Eq. 1 in the introduction.

The model indices and the main effects considered are presented with an example of the values measured in 35 cm depth in a single plot (Fig. 2). According to our observations, the oxygen dynamics ($\Delta O_x/\Delta T$, the change in oxygen partial pressure by time) in this study, at the relevant depth on this site (35 cm), can be divided into four distinct stages, described using the time (T) passed from irrigation start (h). (1) Basal oxygen state ($T = -6$ to 0), for which $\Delta O_x/\Delta T \sim 0$. The oxygen concentration at this phase, the night prior to irrigation is an expression of the soil most aerated conditions, affected by soil structure, measurement depth but also by residual effects of previous irrigation events. (2) Oxygen linear decline phase ($T = 6-12$), for which $\Delta O_x/\Delta T < 0 \sim \text{constant}$. The oxygen decline during this phase (immediately after irrigation end) is attributed to soil saturation, i.e., no air-conducting pores exist, at least in the soil layer above the sensors (state A in Fig. 2), which is assumed to nullify the oxygen supply due to the order of magnitude lower oxygen diffusion through water as compared to air. Assuming oxygen supply rate = 0, and following Eq. 1, we derive that the value of $\Delta O_x/\Delta T$ in this phase equals to the oxygen consumption rate which expresses the soil respiration rate, and is thus a measure of the sum of microbial and root respiration. The decline rate is expected to diminish from its constant rate if oxygen levels begin to limit the biological activity, or due to arrival of oxygen supply. (3) Oxygen minimum ($T > 12$), for which $\Delta O_x/\Delta T = 0$, the time at which a shift between oxygen decline and oxygen incline occurs, marks the conditions at which oxygen supply matches oxygen consumption. To attain sufficient oxygen supply, air-filled pores conducting oxygen down to the measuring depth are required. Since the macro-pores (D in Fig. 2) are the first to drain when soil water content decreases, and since these pores are the most conductive pores, a sharp change from oxygen decline to oxygen increase will occur at this time. The time of oxygen minimum is hence referred to as the oxygen relief point. We refer to the SWT value at which this pore opening occurs as a critical SWT and relate this value to the size and abundance of macro-pores, and expect it to reflect the soil mechanical composition and structure (which may be affected by irrigation water quality). The time until oxygen relief is thus affected by

both the critical SWT and the water content, controlled by the water input, water drainage and water uptake by the plants. (4) After the oxygen relief point, oxygen gradually rises up to the basal values according to the oxygen supply rate (assumed to be negligible up to this stage), or until an additional irrigation occurs.

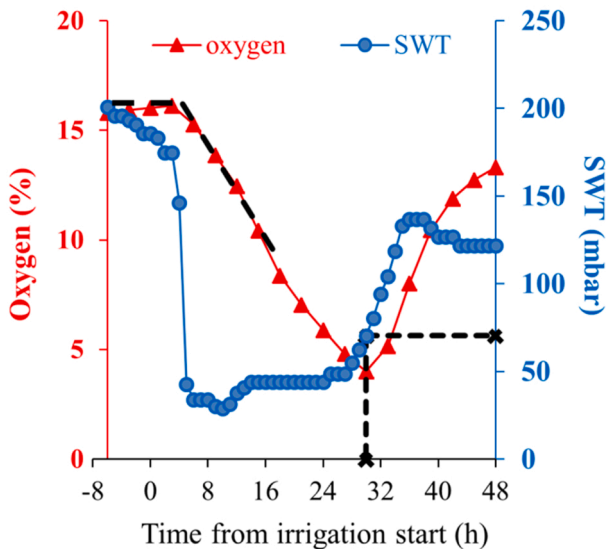
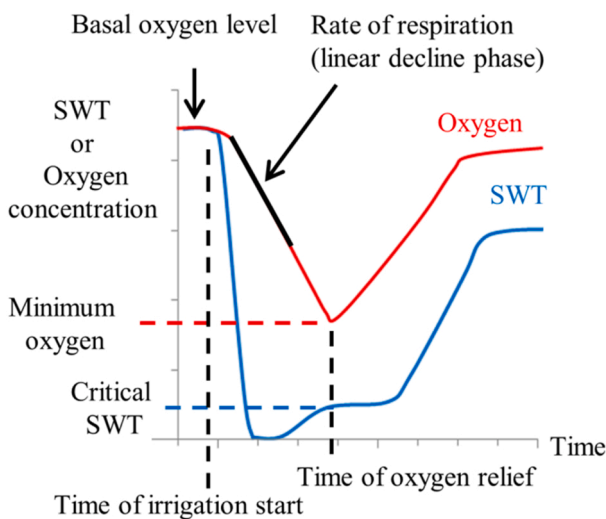
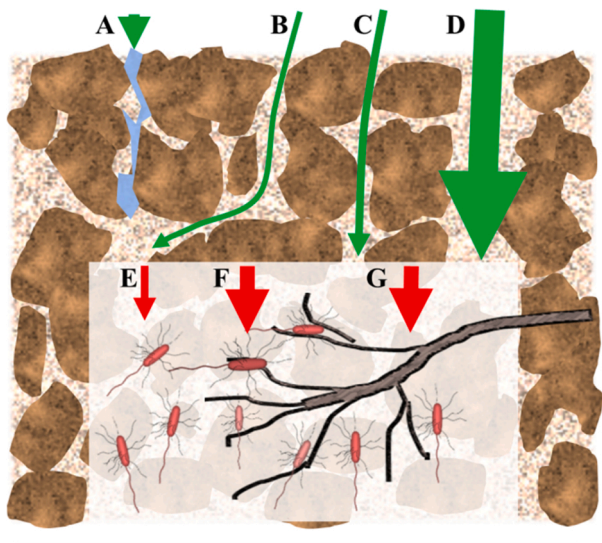
Changes in SWT values during the light hours (06:00–18:00) on days with no irrigation are attributed to plant water uptake, while the changes during the dark hours (18:00–06:00) are attributed to redistribution of water in the soil which we refer to as drainage.

2.4. Retention curve

In order to represent the soil column above the sensors, soils were sampled from a depth of 5–30 cm during September 2014 from a tree adjacent to the tree where the continuous measurements were performed. The samples were air dried, grinded, sieved (<2 mm) and kept in dry and cool conditions until measurements. Retention curves were performed using Hyprop 2 (Hydraulic Property Analyzer, METER Group, Inc., Pullman, WA, USA). To that end, soils were packed into a 250-mL cylinder to achieve a bulk density of 1 g/cm³ and were then allowed to saturate with free water suction for one week. The relatively low bulk density was chosen to prevent the heterogeneous compaction of the soil which may be created due to soil swelling during the wetting processes. To better mimic the field situation, we used synthetic soil solutions for this step. This solution contained Na, K, Ca and Mg chloride salts in concentrations equivalent to the different treatments' soil salinity and SAR according to 2:1 water extractions of the soils. The Hyprop measured curves were then analyzed using the Hyprop-fit software (METER Group AG.) yielding parameters according to the Van-Genuchten equation:

$$\theta_v = \theta_R + \frac{\theta_s - \theta_R}{1 + (\alpha \cdot \text{SWT}^n)^{1-1/n}} \quad (2)$$

Where θ_v is the volumetric water content at a given SWT, θ_s is the water content at saturation; θ_R is the residual water content, SWT is the water tension, α and n are parameters related to the air penetration value and pore size-distribution respectively.



(caption on next column)

Fig. 2. Conceptual model of oxygen dynamics in the soil. Top – Schematic representation of the major processes controlling the oxygen supply rate (green arrows) and the oxygen consumption rate in the root zone (red arrows). The rate of oxygen supply in water filled pores (A) is assumed zero; narrow pores with high tortuosity (B) supply oxygen at a lower rate than those with less tortuosity (C), both considered negligible as compared to the oxygen supply rate through macro-pores (D). Oxygen is consumed by free soil biota (E), root-associated biota (F), and the root (G) – each in its own rate (symbolized by arrow size). Middle – Dynamics of oxygen concentration in relation to the dynamics of soil water tension (SWT) at a given depth in the root zone of a drip irrigated orchard during an irrigation cycle. The parameters used to describe the model are presented. Bottom – Real data oxygen and SWT dynamics measured at 35 cm depth in a single plot during an irrigation cycle in August 2013. The parameters used to analyze the dynamics according to the model are overlain in black dash lines.

Additional retention curves were performed using a pressure plate, where soil samples of ~2-cm thickness were wetted by slow suction of free distilled water for ~48 h and then the desired pressure was attained using a pneumatic pump and retained for ~48 h. Soil water content was then measured gravimetrically for each point on the retention curve for three soil replicates.

2.5. Data analysis

The relative number of readings below 10 % V/V oxygen ($\tau_{10} = \frac{n_{\text{Oxygen} < 10\%}}{n_{\text{Oxygen}}}$) is used to describe the severity of oxygen stress in a specific time period, $n_{\text{Oxygen} < 10\%}$ is the number of readings under 10 % oxygen, and n_{Oxygen} is the total number of readings in this time period. 10 % V/V oxygen was chosen as a threshold because it is generally accepted as a value below which oxygen stress to plants is initiated (Glinski and Lipiek, 2018).

Data selection, aggregation, and transformation were performed to produce average SWT and oxygen dynamics during irrigation cycles with defined properties – such as similar irrigation levels, or similar dates. The data set from sensors with improbable readings or missing data was omitted and so the number of repetitions averaged differs between the different depths. Statistical analysis was performed with JMP® pro 14 statistical software (SAS institute inc., USA)

3. Results

3.1. Overview

3.1.1. Annual oxygen trends

An overview of the oxygen conditions during the two years of the experiment is presented in Fig. 3, where the relative number of readings below 10 % V/V oxygen (τ_{10}) is used to describe the severity of oxygen stress in each season. The crude division into three month periods shows a repeating trend, most noticeable for the TWW irrigated plots at 35 cm depth – during the winter months at which there was no irrigation, the soil oxygen levels improved, so that the lowest τ_{10} values occurred in the spring time. Then τ_{10} values increased as the irrigation season continued towards maximum values either in the summer or autumn when the irrigation levels were highest. Because the more hypoxic conditions occurred during the irrigation season, the focus below is on this period of the year only.

3.1.2. Weekly oxygen trends

During the irrigation season a weekly pattern occurred in all the depths in both treatments (Fig. 4). On irrigation days (Sundays, Tuesdays, and Thursdays) the drop in SWT that occurred after the irrigation commenced was followed by a decrease in oxygen levels, then as soil dried, a rise in oxygen values occurred, approaching their values prior to irrigation. The dynamics during this progress in each irrigation cycle is the focus of the following segments. The background colors in Fig. 4 are

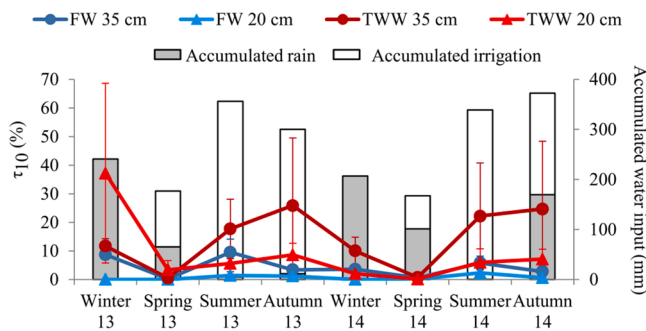


Fig. 3. The τ_{10} values describing the time ratio below 10 % oxygen per season, $\tau_{10} = \frac{n_{\text{oxygen} < 10\%}}{n_{\text{oxygen}}}$ where $n_{\text{oxygen} < 10\%}$ is the number of readings under 10 % oxygen, and n_{oxygen} is the total number of readings, in TWW- and FW- irrigated plots at 20 cm and 35 cm depth – average and standard error values from three plots; and accumulated water inputs as rain water or irrigation in each period. Winter – December to February; Spring – March to May; Summer – June to August; Autumn – September to November (2013 and 2014). Precipitation data from Acre weather station at Moag.gov.il.

aimed at emphasizing that the irrigation on set days three times a week creates a gap of two full days of drying between the Thursday and Sunday irrigations, contrary to the Sunday and Tuesday irrigations which are followed by only one full day of drying. The tight irrigation intervals between Sundays and Thursdays caused a worsening in soil oxygen and wetness which was alleviated during the Saturday dry day.

This worsening especially affected the TWW irrigated plots (SI Fig. S1) and so to avoid the accumulated weekly damage in the comparison between treatments and to investigate where the weekly deterioration stems from the segments below examine the dynamics during Sunday irrigation cycles.

3.2. Irrigation cycle dynamics

3.2.1. SWT dynamics

The irrigation cycle dynamics of SWT and soil oxygen are compared between the two treatments using average values from four consecutive Sunday irrigation events of 8.5–9.6 mm each, during August–September 2013 (Fig. 5 and Table 2). SWT values during the 6 h prior to the irrigation start (at 06:00) were nearly constant with slightly lower values at the lower soil depths (The 20 cm depth in FW irrigated soil skews from this trend which we suspect is unreliable due to only one functional sensor at that depth). These initial SWT values were lower in the TWW irrigated plots as compared to FW irrigated plots in all depths. Following irrigation, the soil reached similar minimum SWT values at the three depths (between 20–50 mbar). The rise in SWT values during drying was greater at the 10 cm depth as compared to the deeper soil layers and in the TWW irrigated plots, the rise during the 48 h after irrigation start followed a clear depth gradient with changes of 154 ± 19 , 88 ± 6 , and 57 ± 9 mbar (average and SE) in the 10, 20 and 35 cm depths respectively. These rises in SWT in the TWW irrigated plots were smaller as compared to the rises in FW irrigated plots, so that TWW irrigated plots were left at lower SWT values at the end of the irrigation cycle in all the depths

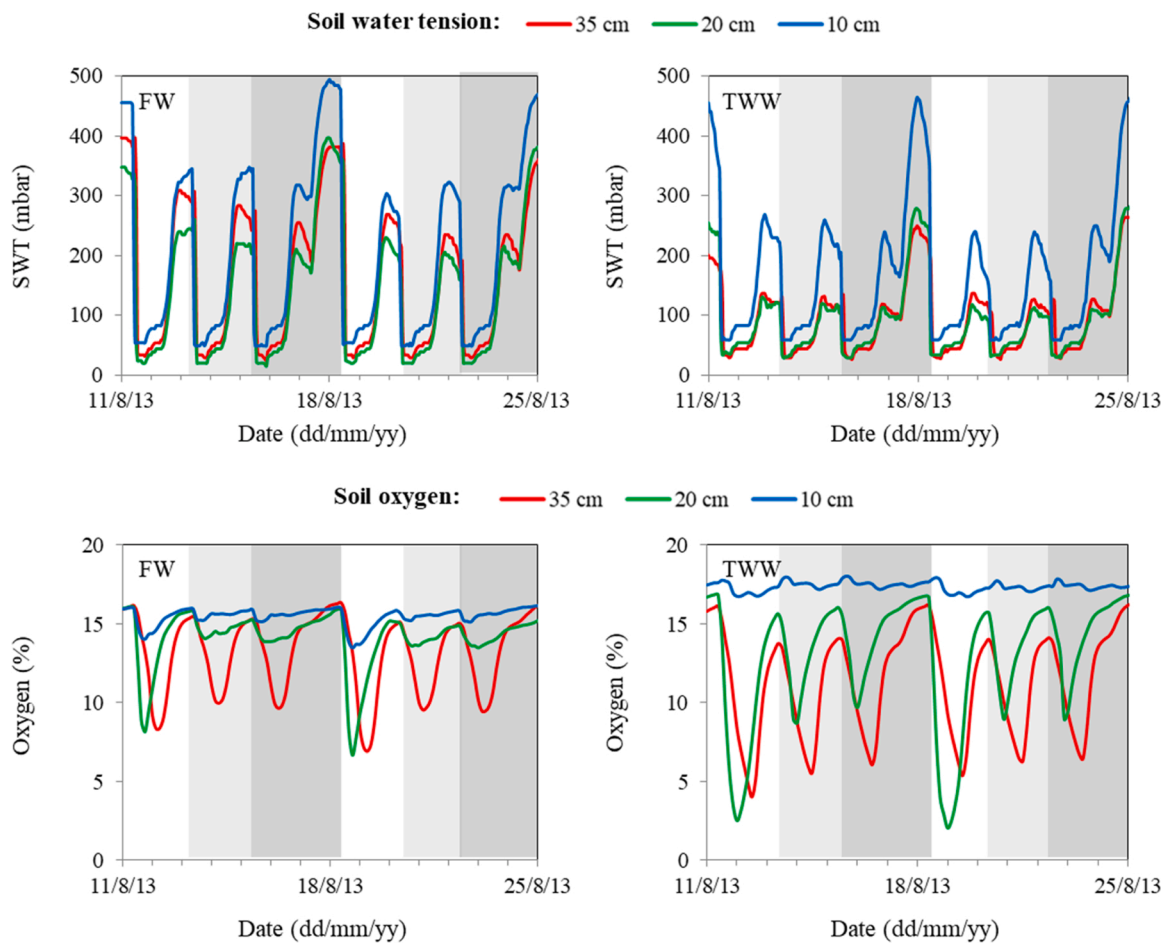


Fig. 4. Two-week (11/08/2013–25/08/2013) course of soil water tension (SWT, top) and soil oxygen levels (bottom) at depths of 10, 20 and 35 cm in a FW-irrigated plot (left) and a TWW-irrigated plot (right). Background colors denote the irrigation cycles: white - Sunday irrigation; light grey – Tuesday irrigation; dark grey – Thursday irrigation. Note that the third weekly cycle is of three days, while the other two are of two days each.

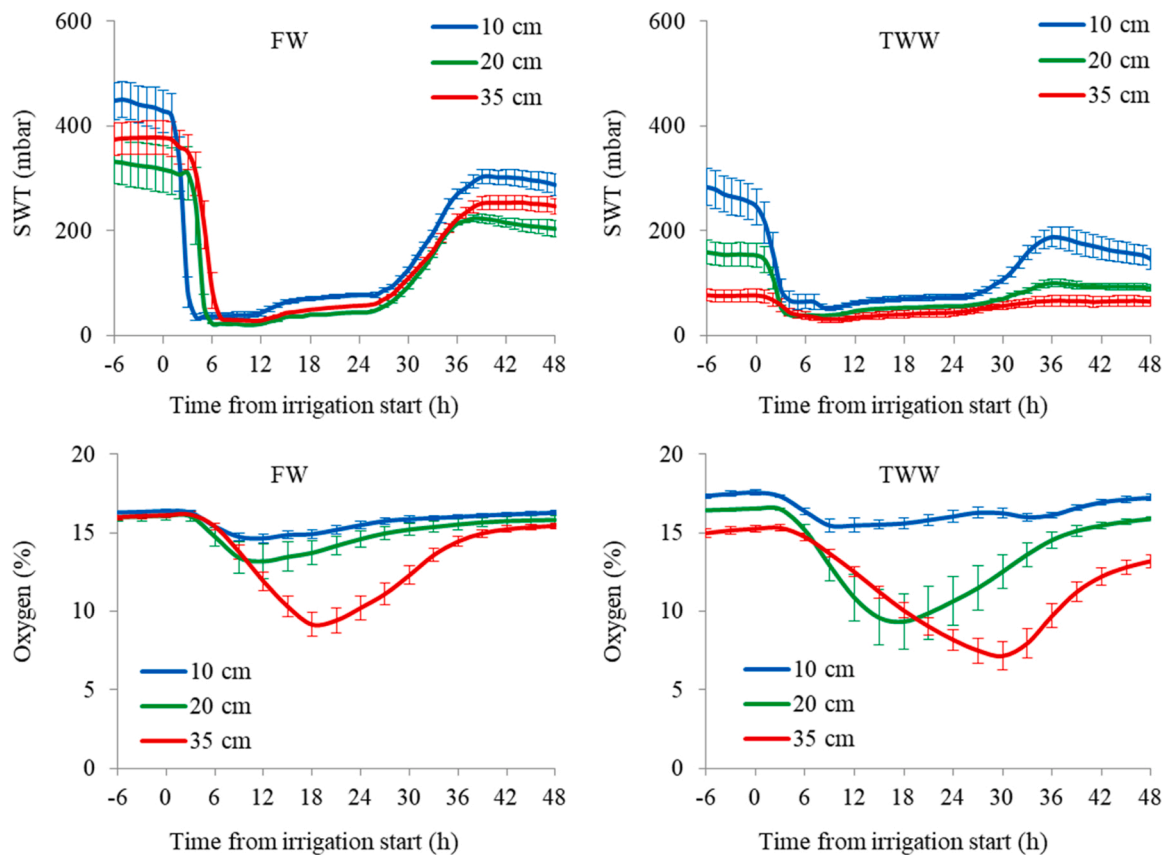


Fig. 5. Soil water tension (SWT, top) and soil oxygen dynamics (bottom) during Sunday irrigation cycles in FW-irrigated plots (left) vs. TWW-irrigated plots (right). Showing average and standard error values of four irrigation events with irrigation levels between 8.5 and 9.6 mm (August–September 2013). A summary of the statistical differences appears in SI Table S1.

Table 2

Indices of SWT and soil oxygen dynamics at the different depths in FW- vs. TWW- irrigated plots, average and standard error values of four consecutive Sunday irrigation events with irrigation levels between 8.5 and 9.6 mm (August–September 2013) and p-value for the differences between treatments in each depth. Numbers in brackets denote the number of samples.

| Depth | 10 cm | | | 20 cm | | | 35 cm | | | |
|---|-----------|----------------|----------------|---------|-----------------|----------------|---------|-----------------|-----------------|---------|
| | Treatment | FW | TWW | p-value | FW | TWW | p-value | FW | TWW | p-value |
| SWT value before irrigation (mbar) | | 441 ± 36 (8) | 266 ± 34 (12) | 0.003 | 325 ± 43 (4) | 155 ± 22 (12) | 0.020 | 377 ± 31 (12) | 76 ± 12 (8) | <0.001 |
| Minimum SWT (mbar) | | 35 ± 6 (8) | 50 ± 4 (12) | 0.065 | 20 ± 0 (4) | 37 ± 1 (12) | <0.001 | 27 ± 1 (12) | 29 ± 5 (8) | 0.77 |
| SWT value before next irrigation (mbar) | | 296 ± 16 (8) | 158 ± 19 (12) | <0.001 | 209 ± 12 (4) | 93 ± 6 (12) | <0.001 | 252 ± 14 (12) | 66 ± 10 (8) | <0.001 |
| Basal oxygen levels (% V/V) | | 16.3 ± 0.1 (8) | 17.1 ± 0.1 (4) | <0.001 | 16.0 ± 0.3 (12) | 16.5 ± 0.1 (8) | 0.146 | 16.1 ± 0.2 (12) | 15.1 ± 0.3 (12) | 0.009 |
| Minimum oxygen (% V/V) | | 14.6 ± 0.3 (8) | 14.6 ± 0.4 (8) | 0.945 | 12.9 ± 1.1 (12) | 9.0 ± 1.8 (8) | 0.093 | 8.9 ± 0.8 (12) | 7.1 ± 0.9 (12) | 0.124 |
| Time of oxygen relief (h from irrigation start) | | 10.5 ± 0.6 (8) | 14.6 ± 1.7 (8) | 0.053 | 16.5 ± 1.3 (12) | 18.0 ± 0.8 (8) | 0.327 | 19.0 ± 0.6 (12) | 28.0 ± 1.3 (12) | <0.001 |
| Basal oxygen after irrigation (% V/V) | | 16.2 ± 0.1 (8) | 17.1 ± 0.2 (8) | 0.003 | 15.8 ± 0.3 (12) | 15.7 ± 0.2 (8) | 0.766 | 15.3 ± 0.2 (12) | 12.7 ± 0.5 (12) | <0.001 |

compared to the FW irrigated plots.

3.2.2. Oxygen dynamics

Basal oxygen levels (from 6 h before irrigation and up to irrigation) remained nearly constant ranging between 15 % V/V and 17 % V/V (Fig. 5 and Table 2) and at 35 cm depth TWW irrigated plots were slightly (~1 % V/V) but significantly (p-value = 0.009) lower. The decline in oxygen following the irrigation started at similar times at all depths (~6 h from irrigation start). At 35 cm depth the oxygen decline was linear for at least 6 h from the decline start ($R^2 > 0.8$ for all repetitions for the period of 6–12 h from irrigation start). The rate of oxygen

decline at 35 cm depth based on linear approximation for this period was 0.57 ± 0.06 percent oxygen per hour in the FW irrigated plots, higher than that in the TWW irrigated plots 0.39 ± 0.03 percent oxygen per hour (p-value = 0.012 for comparison between treatments). According to the conceptual model the decline rate in this phase is equivalent to the soil respiration rate. Using the capsule volume (20 cm^3) and open surface area (34.1 cm^2) the oxygen decline rates translate to aerial respiration rates of $1.75 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$ and $1.20 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$ in the FW and TWW irrigated plots respectively (see SI for further details).

The time until oxygen minimum which we relate to as the oxygen relief point differed between treatments at 35-cm depth. In TWW

irrigated plots oxygen relief occurred ~ 9 h later than in the FW irrigated plots (p -value <0.001). The longer decline time lead the TWW irrigated plots to decline down to minimum oxygen values lower by ~ 1.8 % V/V compared to the FW irrigated plots, but the difference was not statistically significant (p -value 0.124). The rise rate from the minimum oxygen values at 35 cm depth was at a rate of 0.45 ± 0.08 percent oxygen per hour in the TWW irrigated plots as compared with 0.30 ± 0.04 percent oxygen per hour in the FW irrigated plots (p -value = 0.111). At 35 cm-depth the final oxygen levels 48 h from the Sunday irrigation start and just before the Tuesday irrigation start were lowered by 2.4 ± 0.4 % V/V in the TWW irrigated plots as compared to their values before irrigation, while at the FW irrigated plots oxygen levels were lowered by only 0.7 ± 0.1 % V/V from the basal values before the irrigation began (p -value = 0.003 for the comparison between treatments).

3.3. Water content changes

3.3.1. Water retention curves

Water retention curves performed in a pressure plate where the samples were not forced to a defined bulk density and expansion was promoted by saturation with distilled water showed that potentially the TWW irrigated soils retain greater amounts of water at low SWT values (p -value <0.001), with a water content of 53.5 ± 0.1 % by mass in the TWW irrigated soil as compared to 47.4 ± 0.6 % in the FW irrigated soils at 100 mbar tension. Retention curves performed using Hyprop devices (SI Fig. S2) where the soils were packed to a predefined bulk density, and saturated with simulated soil solutions, showed similar values but a smaller difference between the treatments (49.7 % V/V and 46.9 % V/V in the TWW and FW irrigated soils respectively at 100 ± 1 mbar).

3.3.2. Water dynamics in volumetric terms

The SWT dynamics during the August-September irrigation cycles 2013 (Fig. 5) were translated into volumetric terms to represent the differences in water volume uptake and drainage. The volumetric water content (θ_v), presented in Fig. 6, was calculated from the 35-cm SWT data-set using the Van-Genuchten parameters derived for each treatment, from the full retention curves (SI Fig. S2). Prior to irrigation the θ_v in TWW irrigated soils was ~ 13 % V/V higher than in FW irrigated plots (p -value <0.001). These values represent saturation levels (water filled volume/total pore volume) of 85 % V/V in the TWW irrigated plots as compared to a saturation level of 64 % V/V in the FW irrigated plots. Following irrigation, the TWW irrigated plots reached a maximum water content of $\theta_v = 56.8 \pm 0.8$ % and the FW irrigated plots $\theta_v = 54.5 \pm 0.1$ %. During the hour before sunset very little drying occurred, then during

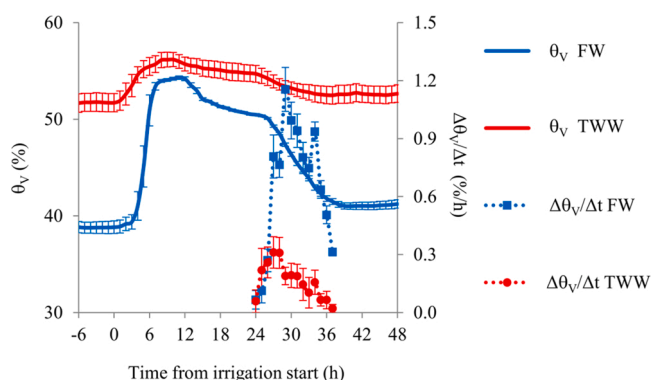


Fig. 6. The soil volumetric water content (θ_v) at 35-cm depth during a Sunday irrigation cycle and the hourly θ_v change ($\Delta\theta_v/\Delta t$) during the light hours (06:00-19:00) the day after irrigation. Based on SWT measurements from four irrigation events, with irrigation levels between 8.5-9.6 mm in three FW-irrigated plots vs. two TWW irrigated plots, transformed according to van-Genuchten parameters, calculated from the soils' water retention curves (2013).

the night (between 19:00 on the day of irrigation and 05:00 the following morning) the decrease in soil moisture was $\Delta\theta_v = 0.8 \pm 0.1$ % in the TWW irrigated plots and almost four times greater in the FW irrigated plots, $\Delta\theta_v = 3.0 \pm 0.3$ % (p -value <0.001). During the light hours (between 06:00 and 19:00), the day after irrigation the loss of water was again nearly four times greater in the FW irrigated plots (9.1 ± 0.4 % V/V) as compared to the TWW irrigated plots (2.3 ± 0.1 % V/V; p -value <0.001), these differences in the daytime water loss are emphasized in the hourly changes in θ_v also presented in Fig. 6.

3.4. Annual deterioration

3.4.1. Seasonal changes in oxygen minimum and maximum

The seasonal oxygen deterioration as described by the minimum and maximum oxygen values in each Sunday irrigation was more accentuated in the 2014 irrigation season despite seemingly similar irrigation management regimes and climatic conditions (Fig. 7). The maximum oxygen values declined in the 2014 season by ~ 4 % V/V in the TWW irrigated plots, from 18.1 ± 0.8 % V/V down to 14.1 ± 0.9 % V/V between May and September 2014, while in the FW irrigated plots a smaller decline of only ~ 1 % occurred from 17.0 ± 0.2 % V/V down to 16.0 ± 0.3 % V/V. The seasonal decrease trend was even more substantial in regards to the minimum oxygen values, again with greater effect on the TWW irrigated plots which declined ~ 11 % V/V from 16.4 ± 0.9 % V/V down to 5.2 ± 1.8 % V/V between May and September 2014 as compared to a decline of ~ 8 % V/V in the FW irrigated plots, from 15.6 ± 0.4 % V/V down to 7.2 ± 1.0 % V/V. The major shift in the oxygen range occurred in both years and both treatments during June when irrigation levels increased from a range of 2-7 mm per irrigation up to a range of 8-12 mm per irrigation.

To further understand the changes in the transition months between irrigation volumes, the oxygen and SWT measurements at 35 cm depth during four irrigation cycles with the typical irrigation volumes within each month of this transition in the 2014 irrigation season, were averaged (Fig. 8). In the May-June irrigations, significant rises occurred in the SWT values between 6-12 h from irrigation start, equivalent to a change of 6.1 ± 1.2 % V/V in the soil water content of FW irrigated and 3.3 ± 0.9 % V/V in the TWW plots as compared to the later months when very little drying occurred before sunset of the irrigation day, 0.2 ± 0.1 % V/V in the FW irrigated plots in August (p -value = 0.004 for comparison of the months) and 0.4 ± 0.2 % V/V in the TWW irrigated plots (p -value = 0.035 for comparison of the months). The TWW- irrigated plots in all months consistently incurred lower SWT values as compared to the FW irrigated plots, even in the May-June irrigations, the average water content during 48 h of the irrigation cycle was ~ 11 % V/V higher in the TWW irrigated plots as compared to the FW irrigated plots (p -value <0.001 for comparison of the treatments).

The lower minimum oxygen values in July and August corresponded with longer periods until oxygen relief occurred as compared to May-June. During the May-June irrigations oxygen relief occurred ~ 12.8 h from irrigation start in both treatments, while in August the FW irrigated plots oxygen decline persisted ~ 5 h more (17.8 ± 0.8 h from irrigation start; P -value = 0.008 for the comparison of the months) and in the TWW irrigated plots ~ 14 h more (26.3 ± 0.5 h from irrigation start; p -value <0.001 for comparison of the months). The gap in time till oxygen relief between treatments in the height of the irrigation season was ~ 9 h, similarly to that in the 2013 season (Fig. 5). In all three periods displayed, it can be seen that soil oxygen and SWT were hindered at the end of the irrigation cycle as compared to the beginning of the irrigation cycle, this as well worsened as the months progressed from a decrease of 0.7 ± 0.1 % V/V in the May-June in the TWW irrigated plots up to 2.8 ± 0.5 % V/V in August irrigations (p -value = 0.002 for comparison of the months), and a decrease of 0.4 ± 0.1 % V/V in each irrigation in the FW irrigated plots during May-June irrigation up to 1.2 ± 0.1 % V/V in the August irrigations (P -value <0.001 for comparison of the months).

Assembling the data from 45 Sunday irrigation cycles during the

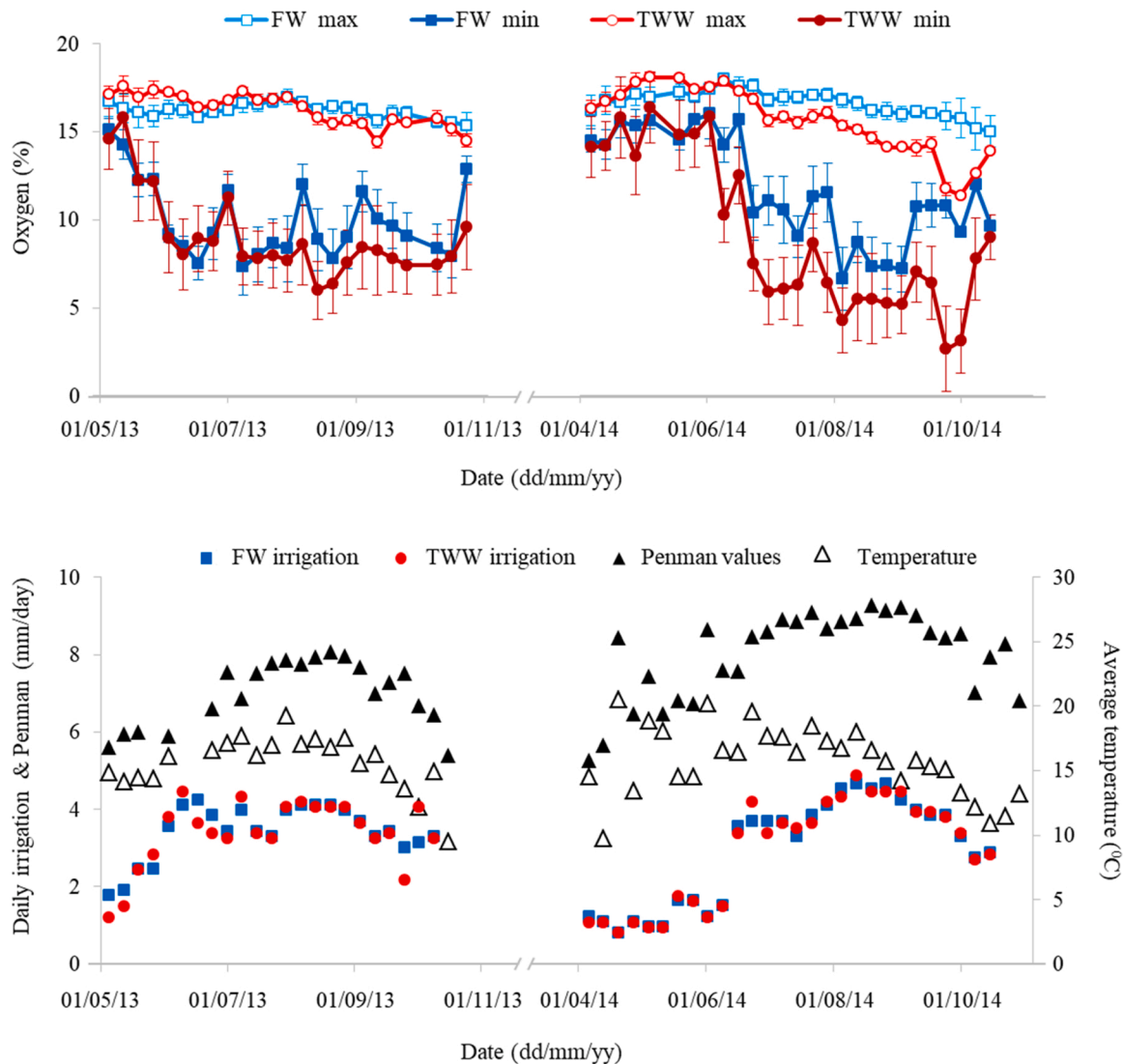


Fig. 7. Sunday irrigation-cycle maxima (max) and minima (min) oxygen values in TWW- vs. FW- irrigated plots at 35 cm depth, average and standard error from three sensors per treatment (top). And average daily irrigation volume applied for each treatment during that week of irrigation, and Penman daily evaporation values together with daily average temperature at 2-m height (bottom; weather data from Acre weather station at Moag.gov.il).

2013 and 2014 (Fig. 9) indicates a significant (p -value <0.001) decrease of the maximum oxygen levels in each irrigation cycle as the elapsed time from the start of the irrigation season increased (Fig. 9A, SI Fig. S3). This effect was smaller for the FW irrigated plots decreasing by only 0.006 percent oxygen per day and slightly higher for the TWW irrigated plots decreasing by 0.020 percent oxygen per day (p -value <0.001 for the treatment effect on the slope). As for the minimum oxygen values in each irrigation cycle, this relations to the time was smaller and a stronger fit was established to the irrigation amount given in each irrigation cycle (p -value <0.001 ; Fig. 9B, SI Fig. S4). Also here the effect was greater in the TWW irrigated plots, decreasing by ~ 1.3 percent oxygen per mm of irrigation given as compared to 0.9 percent oxygen per mm irrigation in the FW irrigated plots (p -value <0.001 for the treatment effect on the slope). The effect of the irrigation levels was also evident for the time span from irrigation start to the oxygen minimum in each irrigation cycle (p -value <0.001 ; Fig. 9C, SI Fig. S4). Increasing the time span to oxygen minimum by ~ 1.8 h per mm of irrigation given in the TWW irrigated plots as compared with ~ 1.0 h per mm of irrigation in the FW irrigated plots (p -value <0.001 for the treatment effect on the slope). The integration of the data from these two correlations revealed that ΔO_x (the size of oxygen decline in each irrigation cycle) correlated with Δt (the time span from irrigation start to oxygen minimum in each

irrigation cycle; SI Fig. S5). Calculating the average rate of oxygen decline based on ΔO_x and Δt for the 45 irrigation events assembled into these charts gave a higher decline rate in the FW irrigated plots (0.49 ± 0.02 percent oxygen per hour) as compared to the TWW irrigated plots (0.35 ± 0.01 , percent oxygen per hour; p -value <0.001).

4. Discussion

4.1. General

Soil oxygen decreased and the differences between treatments increased as soil water inputs increased. This link can be seen in the extent of hypoxia during the height of the irrigation season, at which TWW hindered soil aeration the most as compared to FW irrigation (Figs. 3 and 7). Accordingly the most aerated conditions occurred during the spring time at which the differences between treatments were marginal. A possible explanation for the recuperation in oxygen during the winter months is the long periods of drying between rain events which allow the soil to aerate (Assouline and Narkis, 2013; Yalin et al., 2017), or alternatively leaching by rain water which decreases the soil SAR (SI Fig. S6) which may lead to soil structure improvement. The more hypoxic conditions during the irrigation season may increase the

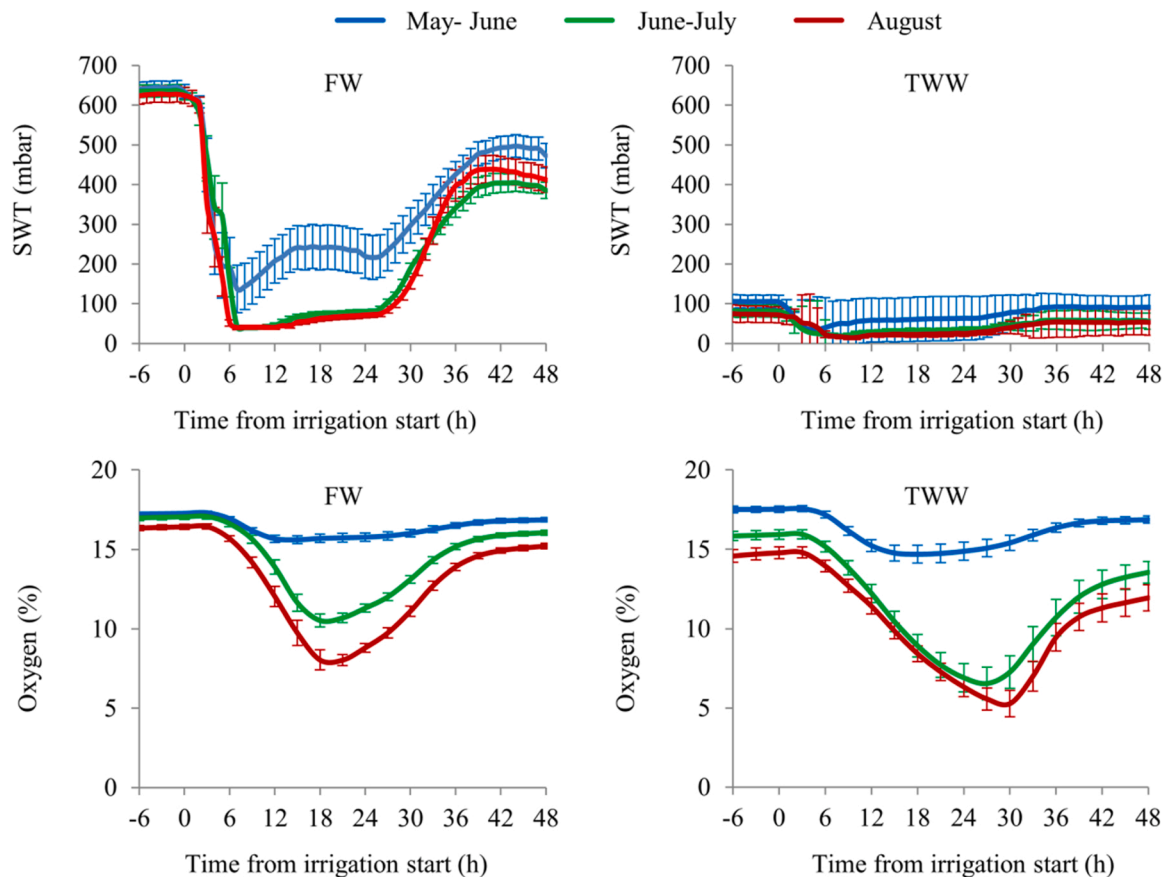


Fig. 8. Soil water tension (SWT, top) and soil oxygen dynamics (bottom) at a depth of 35 cm during Sunday irrigations in FW-irrigated plots (left) vs. TWW-irrigated plots (right). Showing average and standard error values of four irrigation cycles during each period, May-June, June-July and August (2014) from three oxygen sensors per treatment and two SWT sensors per treatment.

risk to plants such as avocado because the yield determining stages of this crop occur in this period in Israel (Salazar-García et al., 2013).

During the irrigation season the decline in oxygen levels was tightly linked to the irrigation events (Fig. 4). A worsening in soil conditions occurred through the irrigation week when there was only one day of drying between irrigations. This worsening was partially alleviated between the Thursday and Sunday irrigations when there was an additional day of drying. The TWW irrigated soils were more affected by this weekly worsening, and so to identify the source of the differences between the treatments it was chosen to focus on the dynamics occurring during the Sunday irrigations.

4.2. Soil oxygen dynamics

The high frequency measurements of soil oxygen in-situ produce a novel opportunity for the understanding of soil mechanisms which affect the oxygen regime. To the most part studies of soil oxygen have not performed high frequency measurement (see Friedman and Naftaliev, 2012 for example) in part due to the cost of setting permanent measuring devices. The few studies that have employed high frequency measurements of soil oxygen in field conditions, did not relate to the hourly changes in oxygen and how they are affected by edaphic conditions (Kallestad et al., 2008; Liptzin et al., 2011; Owens et al., 2017; Shahzad et al., 2019).

To define indices that describe the oxygen dynamics and to give them a mechanistic interpretation a conceptual model presented in the methods section was formed. Data series from irrigation cycles with defined properties – such as similar irrigation levels, or similar dates were than analyzed using this conceptual model.

During August-September (Fig. 5), when the most hypoxic

conditions were recorded, the basal oxygen levels before irrigation commenced were lower than the atmospheric oxygen concentrations, with the highest basal oxygen values appearing in the TWW irrigated soils at 10 cm depth averaging at $17.1 \pm 0.1\%$. These values are close to previous reports of oxygen at these depths (Kallestad et al., 2008; Meek et al., 1983; Owens et al., 2017), and are in line with the perception of the soil gaseous phase as being poor in oxygen as compared to the external atmosphere. At 35 cm depth oxygen levels were lower at the TWW irrigated plots suggesting that despite the improvement in soil aeration on Saturdays TWW irrigated plots began the irrigation week with unfavorable conditions. The oxygen decline began ~6 h from the irrigation start in both treatments in all the measurement depths. Oxygen decline time and decline amplitude (i.e. the size of oxygen decline) increased with depth as expected because oxygen supply is diffusion limited and because of the sequential drying of the layers above the measuring point (Ben-Noah and Friedman, 2018). In accordance, the lowest oxygen values and the longest time with low oxygen occurred at 35 cm depth. The differences between treatments were also amplified at this depth.

During the August-September irrigations (Fig. 5), the oxygen decline at 35 cm depth progressed linearly for at least 6 h from the decline start. The slope of oxygen ($\Delta O_x/\Delta t = \text{change in oxygen by time}$) which was calculated during this period from irrigation start was related to as a measure of the soil respiration rate according to the conceptual model. Greater respiration rates were established according to this methodology in the FW irrigated plots (0.57 ± 0.06 percent oxygen per h) as compared to the TWW irrigated plots (0.39 ± 0.03 percent oxygen per h) during the August to September 2013 irrigations presented in Fig. 5 at 35 cm depth. These trends and values were also found when the oxygen decline rate was calculated in a cruder method according to the ratio

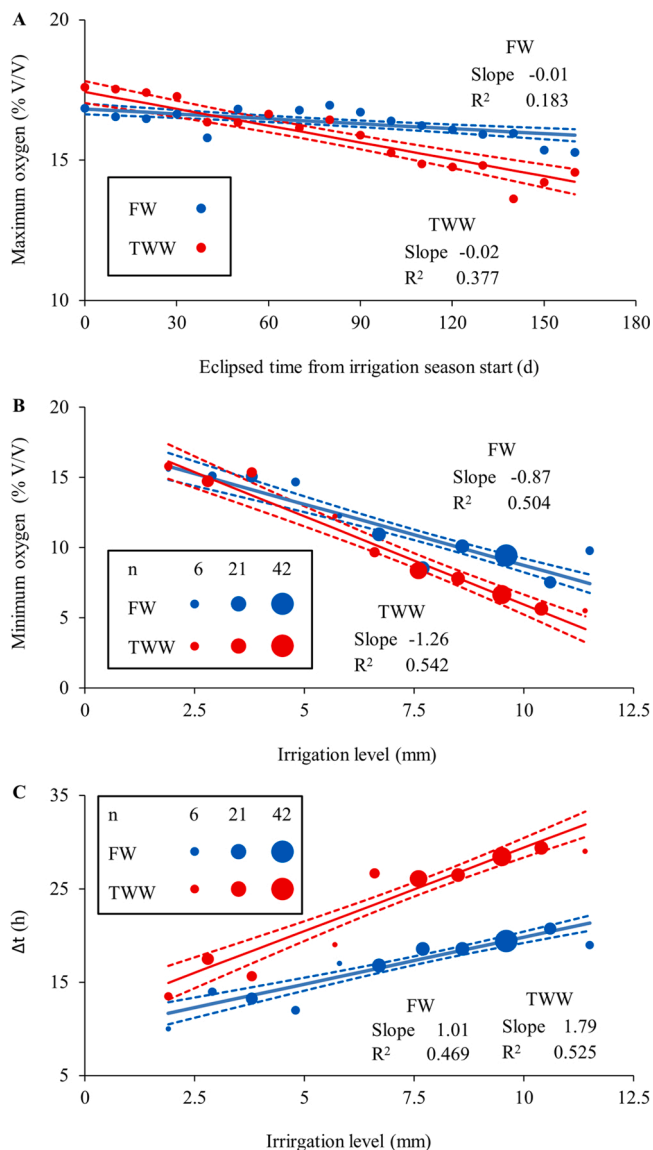


Fig. 9. Correlations between the maximum oxygen value in each irrigation cycle and the elapsed time from the start of the irrigation season (estimated as 1st of April) (A); the minimum oxygen value and the irrigation amount in each irrigation cycle (B); the time span from irrigation start to the time of minimum oxygen concentration (Δt) and the irrigation level in each irrigation cycle (C); in TWW vs. FW irrigated plots. Markers represent the average value for each irrigation amount. In B and C the marker size represents the number of measurements (n). And, linear regression lines (full lines) and their 0.05 confidence intervals (dashed lines). Assembly of soil measurements at 35-cm depth of 45 Sunday irrigations (2013 and 2014).

between the oxygen change during irrigation to the time span between irrigation start and oxygen minimum for 45 Sunday irrigation cycles from 2013 and 2014 (0.49 ± 0.02 and 0.35 ± 0.01 percent oxygen per hour in the FW and TWW irrigated plots respectively). Previous studies show contradicting trends regarding the effects of TWW on soil and root respiration, with variations even within the irrigation season (Elifantz et al., 2011; Meli et al., 2002; Morugán-Coronado et al., 2011; Paudel et al., 2016, 2018). We expect that the main cause for the smaller respiration in TWW irrigated soils at 35 cm depth in this study is the poor root growth we previously reported for the TWW irrigated trees (Yalin et al., 2017), as soil respiration in the bulk soil is known to be significantly lower than in the rhizosphere (Finzi et al., 2015). Additionally, it is possible that the amount of organic matter added with the

wastewater was negligible as compared to the extensive litter layer typical of avocado orchards. Notably, a bias might be suspected in relations to the rate of oxygen decline – in more saturated conditions the connectivity between air-filled pores is smaller so that less of the soil volume is connected to the measuring-chamber volume to consume the oxygen in it (Fig. 1). This bias is explained through Eq. 3 describing the dependence of the measured rate of oxygen decline ($\frac{\Delta O_x}{\Delta t}$) on the specific soil respiration (S, the oxygen consumption rate per volume of soil), the effective soil volume (V_{soil}), the chamber volume ($V_{chamber}$) and the air filled pore fraction (θ_{air} , approximated to change by negligible size within the effective volume)

$$\frac{\Delta O_x}{\Delta t} = \frac{S \times V_{soil}}{\theta_{air} \times V_{soil} + V_{chamber}} \quad (3)$$

This bias may be the underlying cause of greater decline rates ($\frac{\Delta O_x}{\Delta t}$) we observed during instances when the soil was drier and the effective soil volume was greater, for example seen in the 20 cm depth (Fig. 4) during Sundays which were drier as compared to other week days. To minimize this bias, the volume of the measuring chamber was minimized in our setup. Furthermore, it should be stressed that the differences in soil water content between TWW and FW irrigated soils during the period for which our comparison was performed (6–12 h from irrigation start, Fig. 4) were minimal so that this bias had little effect on the trend identified.

To confirm the plausibility of the measured values a comparison to the literature was performed. The standard methods used to measure respiration rates are based on aerial measurements of oxygen change at the soil surface, and respiration rates are standardly expressed as mass change per aerial surface. Using the interface between the oxygen capsule and the soil as the effective surface area measured, and relating to the measurement volume as being identical to the capsule volume we calculated respiration rates of $1.75 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$ and $1.20 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$ in the FW and TWW irrigated plots respectively. This respiration rate is nearly an order of magnitude smaller than the average values collated by Ben-Noah and Friedman (2018) for evergreen forests ($7.8 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$) and field crops ($12.2 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$); but in the range of measurements cited therein of $0.9\text{--}3.9 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$ in a tropical rainforest at 34°C . Furthermore, it should be noted that while standard measurements of soil respiration are performed at the soil surface so as to represent the whole soil profile and specifically the top-soil where most of the soil respiration occurs (Spohn et al., 2016), the measurement chamber used here represents a smaller and deeper part of the soil profile, which may quite likely be smaller than the whole soil profile, especially under wet soil conditions that limit the connectivity in the soil (Moldrup et al., 2000).

The observation that TWW irrigated plots reached similar or lower oxygen values at 35 cm, despite the slower oxygen decline rate (Fig. 5) was related to delayed oxygen relief in the TWW irrigated plots. The decline in TWW irrigated plots at 35 cm depth lasted 9 h more than the FW irrigated plots in August-September of both the 2013 (Fig. 5) and 2014 (Fig. 8) irrigation seasons. The extended period of oxygen decline not only implicated unto the minimum oxygen values but also meant that oxygen remained at low values during the light hours the day after irrigation; the period of exposure to hypoxia in itself has great physiological impact on plants (Armstrong and Drew, 2002), as we previously demonstrated in relation to the reduced yields in this orchard (Yalin et al., 2017).

The rise in oxygen values after the oxygen relief point was slightly but not significantly faster in TWW irrigated plots as compared to FW irrigated plots (Fig. 7), which may be ascribed to the lower respiration rates in the TWW irrigated plots. However, the short period remaining between oxygen relief and the start of the next irrigation, combined with the lower oxygen concentration in this point of time in the TWW irrigated plots, did not allow the oxygen values to recover to their basal levels prior to the next irrigation cycle. This decline in oxygen levels

during the Sunday irrigation events repeated itself during the Tuesday irrigation events leading to an overall weekly decline in oxygen.

The lowering of oxygen levels through the irrigation season (Fig. 7) which was more noticeable in the TWW irrigated plots was related to two factors. First, was the decline in the basal oxygen levels prior to irrigation (the maximum values, Fig. 9A) which we related to the insufficient time for oxygen levels to recuperate between the end of oxygen decline and the following irrigation event. This effect was greater in the TWW irrigated soils because they had less time to recuperate, and started their recuperation at lower values. The second effect was the effect of irrigation amount (Fig. 9B) affecting the minimum oxygen, while this effect was also seen in the FW irrigated plots; the effect on TWW irrigated plots was ~40 % greater. We attributed the oxygen minimum values to the time to oxygen relief (Figs. 5 and 8), this link was further established by the correlation between the time to oxygen minimum in each irrigation cycle and the irrigation levels and the inter-correlation between the two dependent parameters (Fig. 9C and SI Fig. S5). The dramatic differences occurring between the oxygen dynamics in the beginning of June and that in the end of June (Fig. 8) further support the idea that slow processes such as changes in soil chemistry, water quality or climate cannot be the explanatory factors behind the hindering of soil oxygen levels during the season but rather the fast shift in irrigation levels.

4.3. Soil water dynamics

During the height of the irrigation season (Figs. 5 and 8) the driest soil conditions were reached prior to the Sunday irrigations because there was an additional day of drying between the Thursday irrigation and Sunday as compared to the other weekly irrigations. Despite the additional day of drying, on Sunday the TWW irrigated soils started at significantly wetter conditions throughout the soil profile as compared with the FW irrigated plots. The changes in SWT dynamics with depth after irrigation followed similar depth trends in the TWW and FW treatments. The low hydraulic conductivity associated with TWW did not induce measurable differences in the arrival time of the wetting front to the deeper soil layers (~6 h in both treatments). The maximum water content reached after irrigation was close in both treatments representing the field saturation and around 80–85 % of the water holding capacity of this soil according to our lab water retention measurements. No significant drying occurred during the day of irrigation at the August-September Sunday irrigations. As expected the drying process was faster at the shallower soil depths, it is assumed that evaporation played a minor role in the drying of the top layers, because of the difference between treatments and also because of the thick leaf-litter layer, and so the faster drying at the top soil layer can be ascribed mainly to drainage and to the superficial root system typical of avocado (Chanderbali et al., 2013). The soil in the TWW irrigated plots showed much slower drying, which translated to a volume of water 4-fold smaller lost from the 35 cm depth in the TWW irrigated plots as compared to the FW irrigated plots during the light hours the day after irrigation (Fig. 6), during this time drainage is expected to be negligible pointing to lower transpiration of the TWW irrigated trees as the main driving factor behind this difference. In a numerical study in similar soil conditions, Russo et al. (2020) predicted that the osmotic stress due to the elevated Cl^- levels in TWW would cause a ~20 % decrease in transpiration rate of avocado as compared to the transpiration of FW irrigated trees. This prediction was affirmed by sap flow measurements in an avocado orchard where it was also suggested that salinity was the major factor for the deterioration of TWW irrigated trees water uptake (Nemera et al., 2020). We propose, that if the damage to the tree water uptake was entirely due to salinity, the damage would also be occurring in sandy soils irrigated with similar water quality, but several studies have found very limited damage to TWW irrigated crops in sandy soils (Paudel et al., 2016 for example). It is therefore proposed that the direct cause of decreased water uptake in TWW irrigated trees is oxygen stress on the root. A decrease in water

uptake is one of the first responses of plants and specifically avocado to hypoxia (Armstrong and Drew, 2002; Gil et al., 2009), thus it is quite likely that the exposure of TWW irrigated trees to longer periods with hypoxia and lower oxygen levels brings about significant changes in water uptake as our results imply. Notwithstanding, the long term deterioration of the TWW irrigated trees expressed in smaller canopy size and root growth (Yalin et al., 2017) most probably played a part in the diminished water uptake, as can be seen during the spring months (Fig. 8). This long term deterioration may also be related to hypoxia, and may also reflect interactions between hypoxia and elevated salt concentrations which has been described for many plant species (Barrett-Lennard, 2003; Barrett-Lennard and Shabala, 2013) and for which some evidence was found in the form of elevated Na levels within the TWW irrigated tree xylem (Yalin et al., 2017). The differences observed in the moisture changes during the night hours are ascribed to slower drainage below the root zone in TWW irrigated plots as compared to FW irrigated plots, this could be explained by the hindering of water conductance in the TWW irrigated plots as has been established in the literature (Assouline et al., 2016; Gharaibeh et al., 2016). As the changes in drying rate seem to play a cardinal part of the difference between treatments, it is proposed that this difference in drainage was the initial instigator of the differences between treatments, then leading to damage to the trees and eventually to the differences in water uptake as well.

The SWT values at which oxygen relief occurred which we refer to as critical SWT (Fig. 2) were nearly equal in both treatments at 35 cm during August-September 2013, 50 ± 1 mbar in FW irrigated plots and 52 ± 5 mbar in TWW irrigated plots (Fig. 5). In modeling work the range of 20–55 mbar was established as the critical range at which transpiration is negatively affected due to insufficient oxygen supply in a clay soil at 288 K at 25 cm depth (Bartholomeus et al., 2008), the proximity of this range to the value established here supports the validity of our model. The similar values in the two treatments were not in line with the hypothesis that the high SAR in TWW irrigated plots would lead to narrowing of the soil pores, so that they would only be drained at higher SWT values. A possible explanation may be the field salinity compensating the effect of sodicity, demonstrated by the fact that retention curves performed with simulated soil solutions showed negligible differences between treatments in the water contents at the low SWT range (SI Fig. S3), while retention curves performed with distilled water showed significant differences between the treatments. The close critical SWT values indicate that it is the rate of soil drying that was the factor differentiating the two soils.

From the seasonal changes in SWT we deduce that during May-June the irrigation levels were sufficiently small in relation to evapotranspiration to support a substantial rise in SWT in the hours between the end of irrigation and sunset of the irrigation day (Fig. 8), thus permitting oxygen penetration substantially sooner. This link is further established through the correlation between oxygen relief time and the irrigation levels throughout the two irrigation seasons (Fig. 9C).

5. Summary and conclusions

Out of the myriad of negative effects associated with TWW irrigation (Tal, 2016), lack of oxygen is singled out as the most probable cause of damage to orchards irrigated with TWW in clay soils (Paudel et al., 2019; Stemke, 2016; Yalin et al., 2017). This link to hypoxia is further substantiated because of the manifestation of damage in avocado, one of the most hypoxia sensitive species (Schaffer et al., 2013) rather than in more tolerant species such as pear (Tarchitzky et al., 2015), and the occurrence of this damage in clay rather than sandy soils (Paudel et al., 2016).

In this work a novel approach was used to investigate the factors leading to the more hypoxic conditions in TWW irrigated clay soils based on hourly changes in soil oxygen and soil water tension. A conceptual model was developed using field measurements to yield a mechanistic interpretation of the findings. The soil respiration rate

calculated according to the model was equivalent to $1-2 \text{ g O}_2 \text{ m}^{-2} \text{ d}^{-1}$ which corresponds with the range given by former studies employing standard methods. Also, the critical SWT (below which soil is wet enough to cause oxygen supply rate to be smaller than the oxygen consumption rate) was found to be consistent with estimates reached using other methodologies for clay soils ($\sim 50 \text{ mbar}$). The method also showed reproducible values when comparing the two measuring years. The reproducibility of the parameters, and the consistency with the literature, attest the validity of the approach used here. The use of *in-situ* measurements while challenging, gives the findings added value as they best represent the actual agricultural environment.

The critical SWT did not vary between the two water types, but soil drying rates were 4-times smaller in the TWW irrigated soil as compared to FW irrigated soil, leading to $\sim 47\%$ more time at water levels that did not permit oxygen relief. The difference in drying rate was attributed to both low drainage and low tree water uptake. The slow drying in TWW irrigated plots led to longer periods at which oxygen supply to the root zone did not match oxygen demand. The longer times of oxygen deprivation in TWW irrigated plots caused oxygen decline to proceed to lower values, exposing the plants to greater oxygen stress and preventing oxygen recovery before the following irrigation. As irrigation amounts increased through the season, minimum oxygen levels decreased with greater effect in the TWW irrigated plots. This trend matches with a rise in time until oxygen relief point, with increasing irrigation amounts, again with greater effect in the TWW irrigated plots. Additionally, in the TWW irrigated soils the insufficient time for oxygen to recuperate during the Sunday and Tuesday irrigation cycles lead to a weekly and seasonal decline of the upper range of the oxygen levels. It is deduced that the hindering of soil oxygen levels in the TWW irrigated soils originates from the impedance of water drainage caused by the elevated SAR in the TWW. We argue that the prolonged periods with low oxygen levels in TWW irrigated plots then act as a prime cause hindering tree water uptake, causing negative feedback to further deprive the tree roots from the necessary oxygen.

The lower rate of oxygen decline in TWW irrigated plots contradicts the previous perception that the increased respiration rates in TWW irrigated soils associated with the addition of labile organic matter is responsible for the hypoxia. Accordingly, it is suggested that upgrading the TWW to a tertiary quality with lower organic matter content will not act to mitigate the hypoxia. We state that while decreasing TWW sodicity and SAR by desalination or dilution with FW may eliminate soil structure deterioration and thus hypoxia formation in the root zone, this solution is usually difficult to achieve and in most cases where TWW is the only water source for irrigation is not a valid solution. In contrast, this study lays a basis for designing management practices that will allow long term sustainable use of TWW for irrigation in clay soils. It is hence suggested that the key to improving the soil oxygen regime in TWW irrigated orchards planted in clay soils is retaining the soil at drier conditions than the critical SWT (found to be $\sim 50 \text{ mbar}$ in the research site) for longer periods. This can be achieved by decreasing the frequency of irrigation (e.g., 2 times a week instead of 3 times a week) which will permit more time for soil oxygen to recuperate between irrigations. To maintain the plant water requirements, an undesired increase irrigation amounts is required, which in drip irrigated orchards can be resolved by dispersing the irrigation over a greater surface area around the tree (e.g., by adding additional dripper lines). This will promote faster soil drying due to a smaller irrigation volume at each point. Another method for the sustainable use of TWW, which is suggested following this study, is the formation of pumice-filled ditches under the irrigation line, so that trees establish their roots into this well aerated media, thus eliminating the negative effect of the TWW used for irrigation. Indeed, in a nearby avocado orchard, where these suggested management practices were tested (by measurement of sap flow rate, overall transpiration and canopy conductance), they were shown to have promising results in alleviating the negative effects of TWW (Nemera et al., 2020). Overall, this study and the conceptual model

developed, not only decipher the effect of TWW on the soil oxygen regime, but also lay a basis for designing agronomical solutions for sustainable cultivation of crops using TWW. Furthermore, the insights gained in this study regarding soil oxygen dynamics have many environmental implications on issues such as carbon turnover in soil, the fate of pollutants and greenhouse-gas evolution in soil.

Author's declaration

All authors have participated in (a) conception and design, or analysis and interpretation of the data; (b) drafting the article or revising it critically for important intellectual content; and (c) approval of the final version.

This manuscript has not been submitted to, nor is under review at, another journal or other publishing venue.

The authors have no affiliation with any organization with a direct or indirect financial interest in the subject matter discussed in the manuscript.

Declaration of Competing Interest

The authors report no declarations of interest.

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:<https://doi.org/10.1016/j.still.2021.105039>.

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