



Intermittence in irrigation management and nitrogen optimize yield and water use efficiency in baby lettuce

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Abstract

Aim of the study: To evaluate the influence of irrigation management strategies and nitrogen (N) on baby lettuce growth, yield, and water use efficiency in a protected environment.

Area of study: Botucatu, SP, Brazil.

Material and methods: The experiments were conducted in two consecutive cycles, and the treatments corresponded to the combination of irrigation management strategies with N doses. Irrigation strategies comprised 3 possibilities: continuous irrigation (Cont); intermittent irrigation with three irrigation pulses at one-hour intervals (Int1); and intermittent irrigation with irrigation depth split and applied at 7:00, 11:00, and 15:00 h (Int2). Two N doses were evaluated, 100 and 130 kg/ha of N, applied in daily fertigation according to the irrigation management strategies.

Main results: Intermittent irrigation strategies (Int1 and Int2) promoted the highest vegetative growth, increased leaf water content, and increased total yield (49.55 and 55.30; 46.83 and 49.50 t/ha) and marketable yield (46.77 and 52.44; 45.11 and 47.17 t/ha) in the first and second cycles, respectively, in addition to optimizing water use efficiency. The N dose of 130 kg/ha increased the total yield (51.61 t/ha), marketable yield (48.98 t/ha), and water use efficiencies of total yield (53.60 kg/m³) and marketable yield (50.88 kg/m³) only in the first crop cycle.

Research highlights: The use of intermittence in irrigation management proved to be more efficient in obtaining high yields and maximized the efficiency of use of water as a factor of production.

Additional key words: pulse irrigation; fertigation; drip irrigation; vegetative growth; *Lactuca sativa* L.

Abbreviations used: C1 (first cycle); C2 (second cycle); Cont (a single irrigation event); DAIS (days after the start of the application of irrigation strategies); DAT (days after transplantation); ET_c (crop evapotranspiration); ET_o (reference evapotranspiration); HC (head circumference); HI (harvest index); Int1 (intermittent irrigation with three irrigation pulses at one-hour intervals); Int2 (intermittent irrigation with the application of the irrigation depth at 7:00, 11:00 and 15:00 h); LA (leaf area); MFM (marketable leaf fresh mass); MY (marketable yield); NMFM (nonmarketable leaf fresh mass); NML (number of marketable leaves); NMY (nonmarketable yield); NNML (number of nonmarketable leaves); PH (plant height); RCI (relative chlorophyll index); RWC (leaf relative water content); SD (stem diameter); SL (stem length); TFM (total leaf fresh mass); TNL (total number of leaves); TY (total yield); V (base saturation); WUE (water use efficiency); WUE_{MY} (water use efficiency of marketable yield); WUE_{TY} (water use efficiency of total yield).

Citation: Pereira, LS; Saad, JCC; Cardoso, AII; Silva, MA; Lima, CJGS; Silva, EM (2023). Intermittence in irrigation management and nitrogen optimize yield and water use efficiency in baby lettuce. Spanish Journal of Agricultural Research, Volume 21, Issue 4, e1202. <https://doi.org/10.5424/sjar/2023214-20482>

Received: 25 May 2023. **Accepted:** 02 Nov 2023.

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| Funding agencies/institutions | Project / Grant |
|--|---------------------------------|
| National Council for Scientific and Technological Development (CNPq) | 140682/2017-1 and 307457/2022-2 |

Competing interests: The authors declare no conflicts of interest.

Introduction

Lettuce (*Lactuca sativa* L.), belonging to the Asteraceae family, is one of the most commercialized and consumed leafy vegetables in Brazil and worldwide. It is a source of vitamins and minerals and has various cultivars of the head and leaf types, with or without head formation, with purple or green leaves, according to each cultivar, and can be cultivated in the field or protected environment. It is very sensitive to water stress and, therefore, requires good irrigation management to meet its water needs (Pereira et al., 2022).

Irrigation management consists of applying the irrigation depth required by crops at the right time and in the ideal amount throughout the cultivation cycle to avoid water deficit or excess. Proper irrigation management can promote a significant increase in productivity and water use efficiency (WUE) by crops, as well as improve the quality of harvested products (Abdelraouf et al., 2019; Pereira et al., 2019, 2022).

An efficient irrigation management strategy that can maximize lettuce yield is pulse irrigation. This irrigation strategy involves applying the irrigation depth split in short intervals, followed by an off period and another irrigation period. This cycle is repeated until the irrigation depth required by the crop is completely applied. Studies indicate that this irrigation management strategy can maximize the yield of crops such as green bean (El-Mogy et al., 2012), soybean (Eid et al., 2013), coriander (Zamora et al., 2019), onion (Madane et al., 2018a, b), orange (Abdelraouf et al., 2019), sugar beet (Mehanna et al., 2020) and peanuts (Silva et al., 2022).

In addition to increasing crop yield, the pulse irrigation management strategy also significantly reduces the applied irrigation depth by approximately 20%, with an increase in yield (Madane et al., 2018a), contributes to improving the quality of harvested products (Elnesr et al., 2015; Madane et al., 2018a), and maximizes plant growth rates, nutrient accumulation, biomass production, and WUE (Madane et al., 2018a, b; Abdelraouf et al., 2019; Zamora et al., 2019; Menezes et al., 2020a, b; Mehanna et al., 2020).

Nitrogen (N) is one of the nutrients most required by lettuce crops and actively participates in the synthesis of amino acids, proteins, and chlorophyll, in addition to favoring vegetative growth (Perchlik & Tagedere, 2018; Consentino et al., 2022). However, the response of lettuce yield to N fertilization depends on the cultivar, soil, and local climatic conditions (Sylvestre et al., 2019) and on the form of fertilizer application. In general, the recommendation of conventionally applied N fertilization for lettuce is approximately 100 to 130 kg/ha, split and applied three times throughout the cycle (Van Raij et al., 1997).

The determination of adequate doses of nutrients to be applied and the best fertilization management strategy for lettuce cultivation still needs to be researched, since the

productivity of agricultural crops is sustained by adequate fertilization management practices (Souza et al., 2017; Sobucki et al., 2019). Fertigation is the simultaneous application of water and fertilizers by an irrigation system. This technique can maximize the yield of crops such as lettuce and optimize the efficiency of the use of N fertilizers as a factor of production (Souza et al., 2017; Pereira et al., 2022). However, inadequate fertigation management, especially in a protected environment, associated with the application of high doses of nutrients such as N, increases salt concentrations on the soil surface and compromises water absorption.

The hypothesis of this study is that intermittent irrigation with fertigation increases lettuce yield, reduces the N dose applied, and optimizes the efficiency of use of water as a factor of production. This study aimed to evaluate the influence of irrigation management strategies and N on baby lettuce growth, yield, and WUE in a protected environment.

Material and methods

Experimental site

The experiments were conducted in two consecutive cycles under conditions of protected cultivation in an area located at the School of Agricultural Sciences (FCA), Botucatu, SP, Brazil (22°51'03" S latitude, 48°25'37" W longitude, and 786 m altitude). The first cycle (C1) was conducted from October 31 to December 5, 2019, and the second cycle (C2) was conducted from December 13, 2019, to January 12, 2020.

The protected environment was made of a metal structure with a single arch, 31 m long × 7 m wide, covered with a transparent plastic film of low-density polyethylene (150 µm thick), ceiling height of 1.85 m and a height of 4.50 m at the highest point. The sides, front, and back were covered with 50% black shade nets, measuring 1.85 m in height.

Experimental design and treatments

A randomized block design in a 3 × 2 factorial scheme with four replicates was used, with treatments consisting of the combination of three daily irrigation management strategies: continuous irrigation (Cont - a single irrigation event); intermittent irrigation with three irrigation pulses at one-hour intervals (Int1); and intermittent irrigation with the application of the irrigation depth at 7:00, 11:00 and 15:00 h (Int2); associated with two N doses (100 and 130 kg/ha of N), which correspond to the minimum and maximum doses recommended by Van Raij et al. (1997). The experimental design is shown in Fig. 1a.

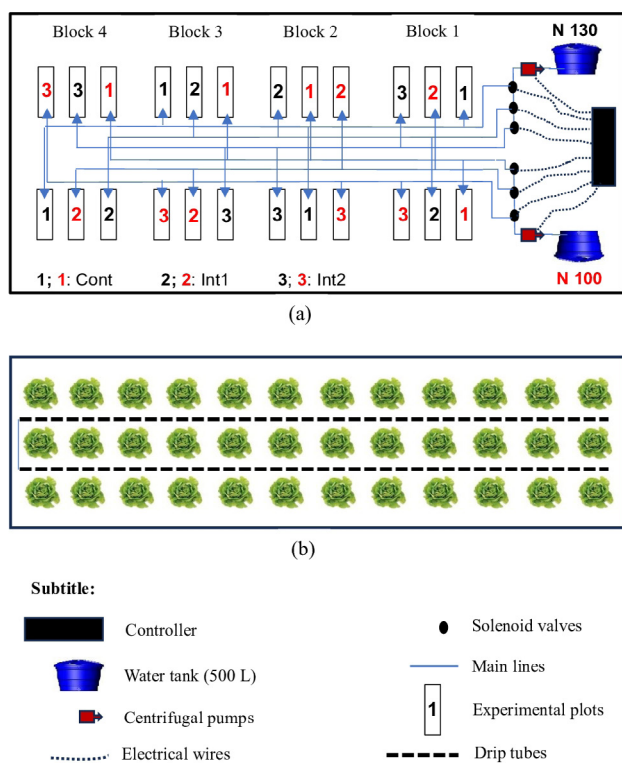


Figure 1. Layout of the experimental design (a) and details of the experimental plots (b).

The daily irrigation depths applied in Int1 and Int2 were divided into equal times and amounts in each irrigation event. Top-dressing N was applied through daily fertigation following irrigation management strategies. The irrigation events started at 7:00 h and continued throughout the day, according to the treatments. Daily irrigation depths varied throughout the crop cycle but were the same among treatments.

The experimental plots consisted of 2.0 m² beds with three rows of twelve plants, totaling 36 plants per plot. Only the ten plants in the central row were used for evaluation (Fig. 1b).

Experiment description

The soil inside the greenhouse is classified as *Nitossolo Vermelho distroférico* (Ultisol) with a sandy loam texture in the 0-0.20 m layer (Santos et al., 2013). The soil was prepared with a rotary hoe, and based on chemical analysis, liming was performed to raise base saturation (V) to 80% and correct acidity (Van Raij et al., 1997). Limestone was applied broadcast at a dose of 1.60 Mg/ha and incorporated to a depth of 0.20 m.

After 30 days of correction, the soil showed the following chemical and physical-hydraulic attributes: pH (CaCl₂) = 6.0; P (resin) = 50 mg/kg; V = 75%; organic matter = 19 g/dm³; CEC = 76 mmol/L; Ca²⁺ = 46 mmol/L; Mg²⁺ = 10 mmol/L; K = 1.8 mmol/L; electrical conductivity = 362

μS/cm; Al³⁺ = 0 mmol/L; H+Al = 19 mmol/L; S = 15 mg/kg; B = 0.31 mg/kg; Cu = 5.5 mg/kg; Fe = 19 mg/kg; Mn = 11.5 mg/kg; Zn = 7.0 mg/kg; and the following physical attributes: sand = 634 g/kg; silt = 99 g/kg; clay = 267 g/kg; bulk density = 1.39 kg/dm³; particle density = 1.98 g/cm³; total porosity = 47%; field capacity = 0.1974 m³/m³; and permanent wilting point = 0.1546 m³/m³.

Basal fertilization with N, P and K was performed manually in the beds based on the chemical analysis of the soil and the fertilization recommendations proposed by Van Raij et al. (1997). In total, 40, 300, and 100 kg/ha of N, P₂O₅, and K₂O, through the sources urea (45% of N), single superphosphate (18% P₂O₅), and potassium chloride (60% K₂O), respectively, were applied in each of the two crop cycles at 7 days before transplanting the seedlings. The basal N fertilization corresponded to the application of 40 kg/ha of the recommended N. Hand-held hoes were used to raise the beds, with dimensions of 0.80 × 2.50 × 0.30 m in width, length, and height, respectively, spaced 0.50 m apart.

Pelletized seeds of the baby green romaine lettuce Astorga Rijk Zwaan were used. The variety was chosen due to its increasing acceptance in the market because it is new, smaller, and cultivated year-round. The seeds were sown in thin plastic polystyrene trays of 200 cells in the commercial substrate Plantmax Hortaliças™. The seedlings were kept in a protected environment and transplanted to the beds 30 days after sowing, with 3 to 5 true leaves, at a spacing of 0.20 m × 0.20 m between rows and between plants, respectively.

Following the irrigation management strategies, the top-dressing N doses were applied in the form of urea through daily fertigation. The doses were split into equal amounts and applied throughout the crop cycles, totaling 16 fertigation events per cycle. Fertigation began at 10 days and ended at 26 days after transplantation in both crop cycles, with direct injection of the solutions by the pumps.

Irrigation was applied with a drip system consisting of two Eletroplas® centrifugal pumps (Model ICS-50AB) with a power of 1/2 HP and a flow rate of 1.8 m³/h. For each pump, the following items were installed: a water tank (500 L) to dilute N doses, a disc filter (120 mesh), a manometer for pressure control, and three main lines with a nominal diameter (ND) of 25 mm, with NaanDanJain solenoid valves (Model S390-2 W-4 24VAC) positioned at the beginning of each main line to allow differentiation of irrigation management strategies (Fig. 1a).

The lateral lines of the irrigation system were NaanDanJain polyethylene drip tubes (AmnonDrip PC CNL) with an ND of 17 mm, measuring 2.5 m in length, with built-in pressure-compensated anti-drain emitters, equidistantly spaced every 0.20 m, with a nominal flow rate of 2.0 L/h operating at a pressure of 1 bar, with a water application efficiency of 92.50%. Two drip tubes installed between the three planting rows in each bed were used (Fig. 1b).

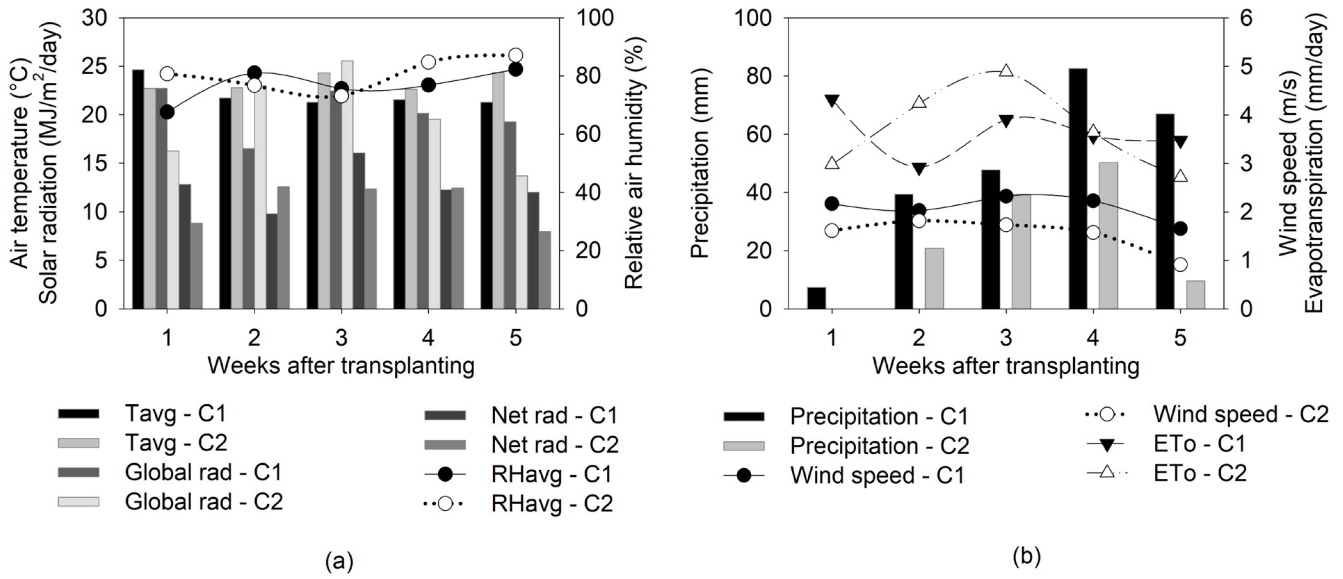


Figure 2. Weekly means of the meteorological variables average temperature (Tavg), global solar radiation (Global Rad), net radiation (Net Rad), average relative air humidity (RHavg) (a) and precipitation, wind speed and reference evapotranspiration (ETo) (b) outside the greenhouse during the experimental periods of the firsts (C1) and second cycle (C2).

Irrigation times were set in a prototype controller developed with a Microchip PIC 16F628a microcontroller, with an eight-digit numeric display to establish the exact operating times of each solenoid valve, connected to a keyboard that established the operator’s interface with the microcontroller. Five relay drivers were installed in the controller to establish the microcontroller connections with the two pumps (one relay/pump) and solenoid valves (one relay/two valves), actuating one solenoid valve per pump synchronously (Fig. 1a).

The water used in the irrigation came from an artesian well and was classified as C1S1, indicating no risks of soil salinization or sodification. Daily irrigation management was performed by the climatic method based on reference evapotranspiration (ETo) outside the greenhouse, obtained by the Penman–Monteith method adapted by FAO (Allen et al., 2006). Crop evapotranspiration (ETc) or daily irrigation depth was determined by the relationship: $ETc = ETo \times Kc$, where Kc is the crop coefficient (Allen et al., 2006). The Kc used were those obtained by Bastos et al. (1996), equal to 0.52 for phase I (1-10 days after transplantation - DAT), 0.80 for phase II (11-25 DAT), and 0.92 for phase III (26 DAT - until the harvest of the cycle).

External climatic data of air temperature and humidity, solar radiation, and wind speed were obtained from an automatic weather station belonging to the School of Agricultural Sciences (FCA) in Botucatu-SP, located at a distance of 100 m from the greenhouse, and were used to calculate daily ETo (Fig. 2). The Solar Radiometry Laboratory of the Department of Bioprocesses and Biotechnology of the FCA of Botucatu-SP provided net radiation data.

The micrometeorological variables air temperature, relative humidity, and wind speed inside the greenhouse were monitored by a weather station that performed readings continuously every second and stored the averages every 15 min in a Campbell Scientific CR10X datalogger.

The daily means of the micrometeorological variables temperature, relative humidity, and wind speed during the period in which lettuce crop remained in the greenhouse after transplantation were 21.57 °C, 78.47%, and 0.0031 m/s, ranging from 17.50 to 26.50 °C, 67.69 to 94.04% and 0.0015 to 0.0050 m/s in C1, respectively. In C2, the means of these variables were 22.73 °C, 81.26%, and 0.0032 m/s, ranging from 19.62 to 24.81 °C, 80.93 to 95.64%, and 0.0 to 0.0251 m/s, respectively (Fig. 3b,c).

Irrigation management from the seedling transplant period until 7 DAT consisted of a daily irrigation event starting at 8:00 a.m. The irrigation depths applied in all treatments were 96.28 and 86.08 mm in C1 and C2, respectively.

Harvest was performed at 35 and 30 DAT in C1 and C2, respectively, identifying the commercial standard as the harvest point, with possible formation of heads and maximum vegetative development, with no signs of flowering.

Characteristics analyzed

Vegetative growth and yield components

The relative chlorophyll index (RCI) was obtained in three plants per plot, with measurements taken on two sides of the upper part of the leaves at 7, 14, and 21

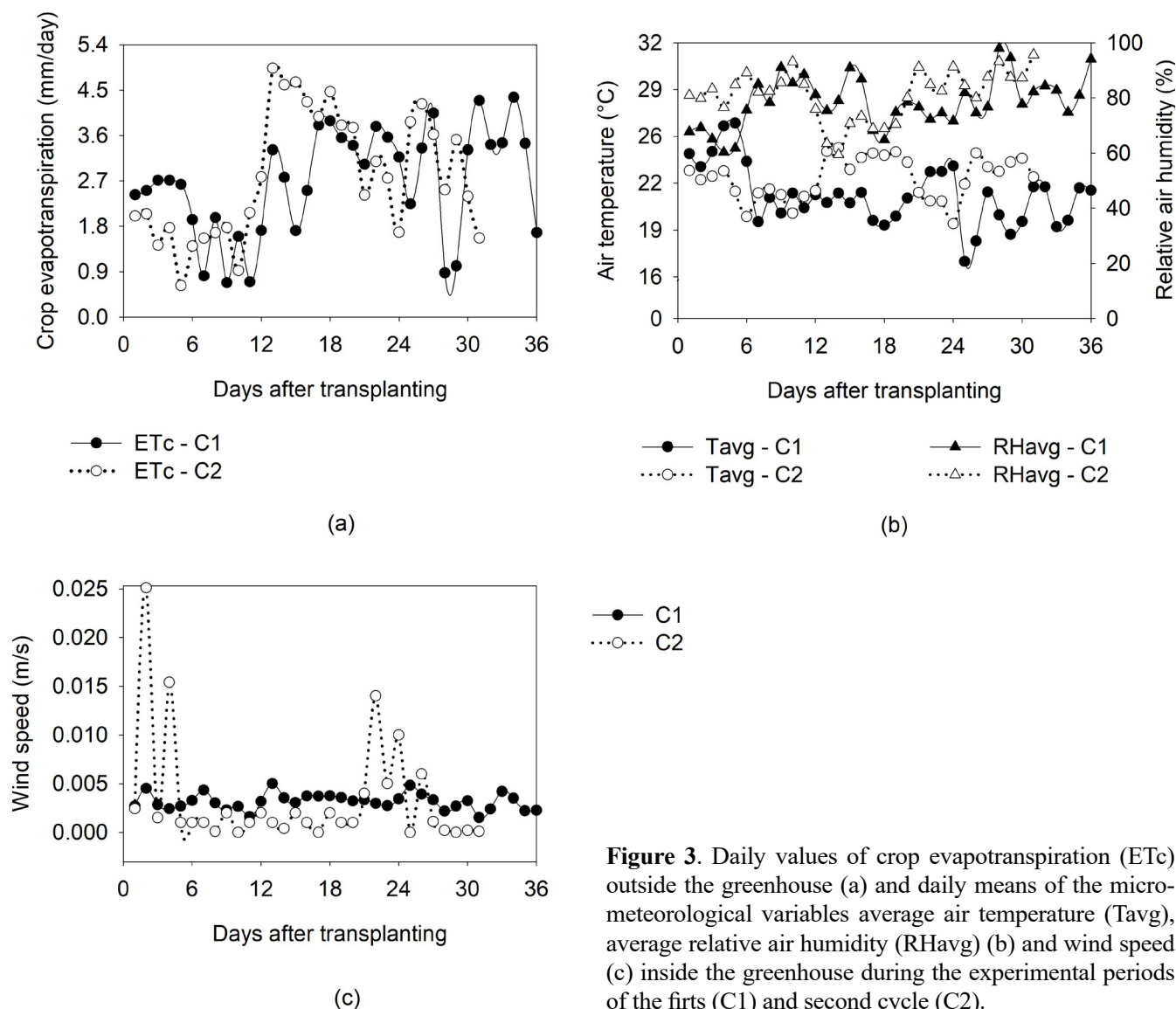


Figure 3. Daily values of crop evapotranspiration (ETc) outside the greenhouse (a) and daily means of the micro-meteorological variables average air temperature (Tavg), average relative air humidity (RHavg) (b) and wind speed (c) inside the greenhouse during the experimental periods of the first (C1) and second cycle (C2).

days after the beginning of the application of irrigation management strategies in both cycles, with a portable chlorophyll meter (SPAD-502Plus, Minolta). Leaf relative water content (RWC, %) was determined 30 days after transplanting, according to the methodology described by Badr & Brüggemann (2020).

At the end of each cycle, the following parameters were evaluated in three plants per plot: stem diameter (SD, mm), measured below the insertion of the first leaf with a 150-mm steel digital caliper (ZAAS-1.0004); stem length (SL, cm), also measured with a digital caliper after total defoliation of the plants; plant height (PH, cm), measured between the collar and the end of the highest leaves with a graduated ruler; and head circumference (HC, cm), measured between the opposite margins of the leaf disc with a graduated ruler.

The total number of leaves (TNL), the number of marketable leaves (NML), and the number of nonmarketable leaves (NNML) per plant were determined

by manually counting the leaves and leaf area (LA, m²) determined using a benchtop leaf area meter (LI-COR-3100). Total leaf fresh mass (TFM), marketable leaf fresh mass (MFM), and nonmarketable leaf fresh mass (NMFM) (g/plant) were obtained by weighing the leaves on a digital precision scale 0.01 g, considering as marketable the leaves that were in good phytosanitary condition, without external damage, suitable for commercialization and consumption (Rezende et al., 2017). The evaluations of these variables were carried in three plants per experimental plot.

Yield, water use efficiency, and harvest index

Total (TY), marketable (MY), and nonmarketable (NMY) yields in tons per hectare were determined by weighing the aerial part of the plants (3 plants/plot) on a precision digital scale and extrapolating the values to one hectare. Water use efficiencies of total yield (WUE_{TY}) and

Table 1. Physiological and growth variables of lettuce as a function of irrigation management strategies and nitrogen in two crop cycles: Means and standard deviation of relative chlorophyll index (RCI), relative water content (RWC), stem diameter (SD), stem length (SL), plant height (PH) and head circumference (HC).

| | RCI | | | RWC (%) | SD (mm) | SL (cm) | PH (cm) | HC (cm) |
|--------------------------|-------------|-------------|-------------|-------------|-------------|------------|-------------|-------------|
| | 7 days | 14 days | 21 days | | | | | |
| First cycle (C1) | | | | | | | | |
| Strategy | | | | | | | | |
| Cont | 32.03±1.19 | 39.54±1.75 | 37.78±0.66b | 80.80±1.00b | 12.14±0.76 | 11.38±1.88 | 15.47±0.58b | 21.91±1.50b |
| Int1 | 32.08±1.29 | 39.71±2.16 | 39.89±1.06a | 83.26±2.37a | 12.78±1.03 | 12.77±0.99 | 17.81±0.42a | 25.00±1.09a |
| Int2 | 33.88±2.76 | 40.60±2.50 | 41.24±2.33a | 87.59±1.56a | 13.45±1.78 | 12.52±1.12 | 18.14±1.00a | 25.73±0.90a |
| N (kg/ha) | - | - | - | - | - | - | - | - |
| 100 | 32.60±1.66 | 38.78±1.96b | 39.02±1.48 | 83.79±3.52 | 12.81±1.64 | 12.06±1.49 | 17.05±1.44 | 23.83±2.24 |
| 130 | 32.73±2.38 | 41.12±1.59a | 40.26±2.41 | 84.03±3.33 | 12.77±0.99 | 12.39±1.48 | 17.23±1.39 | 24.60±1.82 |
| ANOVA | | | | | | | | |
| STR | ns | ns | ** | ** | ns | ns | ** | ** |
| N | ns | ** | ns | ns | ns | ns | ns | ns |
| STR × N | ns | ns | ns | ns | ns | ns | ns | ns |
| Second cycle (C2) | | | | | | | | |
| Strategy | | | | | | | | |
| Cont | 34.86±2.00b | 37.47±6.29 | 35.97±0.41b | 80.13±1.03b | 10.13±0.96b | 9.48±1.89 | 15.34±1.11b | 21.00±0.80b |
| Int1 | 39.27±1.96a | 40.82±7.94 | 38.76±3.18a | 84.38±1.40a | 11.17±0.98a | 11.09±1.33 | 17.68±0.82a | 23.71±0.41a |
| Int2 | 43.54±5.27a | 42.49±4.25 | 40.16±5.69a | 87.79±3.83a | 11.21±0.62a | 11.38±1.44 | 17.43±0.88a | 24.12±0.91a |
| N (kg/ha) | - | - | - | - | - | - | - | - |
| 100 | 39.94±4.57 | 41.70±6.54 | 39.22±5.07 | 84.61±2.95 | 10.91±1.13 | 11.08±1.95 | 16.77±1.32 | 22.72±1.49 |
| 130 | 38.50±5.46 | 38.82±6.24 | 37.37±2.94 | 83.59±2.00 | 10.76±0.83 | 10.22±1.43 | 16.87±1.53 | 23.16±1.71 |
| ANOVA | | | | | | | | |
| STR | ** | ns | * | * | * | ns | ** | ** |
| N | ns | ns | ns | ns | ns | ns | ns | ns |
| STR × N | ns | ns | ns | ns | ns | ns | ns | ns |

STR: irrigation management strategies. Means followed by different letters in the columns differ from each other by Tukey test at 0.05 significance level. * and ** indicate significance at 0.05 and 0.01, respectively. ns: non-significant interaction between the factors.

marketable yield (WUE_{MY}) were calculated by the ratio between yield (kg/ha) and the applied irrigation depth (m^3) (Zamora et al., 2019). The harvest index (HI) was obtained by the ratio between the marketable fresh mass of shoots (g/plant) and the total fresh mass of shoots (g/plant) (Santos et al., 2011).

Statistical analysis

The data were subjected to analysis of variance at a 0.05 significance level. In the case of a significant effect, a qualitative analysis of the comparison of means by the Tukey test at the 0.05 significance level was performed. The statistical analyses were performed in the computer program SAS v. 9.4 (SAS Institute, Cary, NC, USA). The figures were created in SigmaPlot v. 11.0 (Systat Software Inc., San Jose, CA, USA).

Results

Vegetative growth and yield components

Table 1 shows that N did not influence the RCI 7 and 21 days after implementing irrigation strategies (DAIS) in C1. However, at 14 DAIS in this cycle, the 130 kg/ha N dose significantly increased the RCI (41.12). Irrigation strategies influenced the RCI at 21 DAIS in C1 and at 7 and 21 DAIS in C2. The highest RCI values at 21 DAIS in C1 (39.89 and 41.24) and at 7 (39.27 and 43.54) and 21 (38.76 and 40.16) DAIS in C2 were observed with intermittent irrigation strategies (Int1 and Int2), respectively.

The RWC increased with the use of irrigation strategies Int1 and Int2, which promoted increments in RWC of 2.95 and 7.75% in C1 and 5.03 and 8.72% in C2 compared to the Cont strategy, respectively (Table 1). The means of

Table 2. Yield and growth components of lettuce as a function of irrigation management strategies and nitrogen in two crop cycles: Means and standard deviation of total number of leaves (TNL), number of marketable leaves (NML), number of non-marketable leaves (NNML), leaf area (LA), total leaf fresh mass (TFM), marketable leaf fresh mass (MFM) and nonmarketable leaf fresh mass (NMFM).

| | TNL | NML | NNML | LA | TFM | MFM | NMFM |
|--------------------------|----------------|--------------|------------|-------------------|---------------|----------------|------------|
| | (leaves/plant) | | | (m ²) | (g/plant) | | |
| First cycle (C1) | | | | | | | |
| Strategy | | | | | | | |
| Cont | 28.16±2.05b | 23.41±2.12b | 4.75±1.44 | 0.159±0.02 | 147.55±16.62b | 138.76±18.73b | 8.79±2.97 |
| Int1 | 29.83±2.62ab | 25.12±2.99ab | 4.70±1.37 | 0.169±0.01 | 170.14±17.63a | 159.02±16.16ab | 11.12±7.49 |
| Int2 | 32.00±1.60a | 27.62±2.07a | 4.37±0.93 | 0.175±0.01 | 189.74±18.77a | 178.28±17.34a | 11.45±4.05 |
| N (kg/ha) | - | - | - | - | - | - | - |
| 100 | 29.97±2.57 | 24.83±3.05 | 5.13±1.27a | 0.158±0.01b | 161.24±27.84b | 150.82±26.35b | 10.41±4.27 |
| 130 | 30.02±2.72 | 25.94±2.79 | 4.08±0.95b | 0.179±0.01a | 177.05±18.29a | 166.55±17.88a | 10.49±6.05 |
| ANOVA | | | | | | | |
| STR | ** | ** | ns | ns | ** | ** | ns |
| N | ns | ns | * | ** | * | * | ns |
| STR × N | ns | ns | ns | ns | ns | ns | ns |
| Second cycle (C2) | | | | | | | |
| Strategy | | | | | | | |
| Cont | 29.73±2.28b | 26.75±1.60b | 3.00±0.90 | 0.149±0.01b | 130.64±29.24b | 123.85±27.29b | 6.79±2.52 |
| Int1 | 33.03±2.20a | 29.91±1.90a | 3.11±1.53 | 0.169±0.02ab | 160.26±20.25a | 153.38±18.34a | 6.87±3.58 |
| Int2 | 34.46±2.21a | 30.50±2.90a | 3.95±1.07 | 0.183±0.01a | 169.71±16.42a | 160.29±16.47a | 9.41±3.16 |
| N (kg/ha) | - | - | - | - | - | - | - |
| 100 | 32.13±2.59 | 28.75±2.47 | 3.37±1.09 | 0.173±0.02 | 157.81±29.66 | 149.62±27.74 | 8.19±2.94 |
| 130 | 32.69±3.34 | 29.35±3.02 | 3.33±1.39 | 0.161±0.02 | 149.26±25.71 | 142.06±24.60 | 7.19±3.54 |
| ANOVA | | | | | | | |
| STR | ** | * | ns | ** | ** | ** | ns |
| N | ns | ns | ns | ns | ns | ns | ns |
| STR × N | ns | ns | ns | ns | ns | ns | ns |

STR: irrigation management strategies. Means followed by different letters in the columns differ from each other by Tukey test at 0.05 significance level. * and ** indicate significance at 0.05 and 0.01, respectively. ns: non-significant interaction between the factors.

SD, 11.17 and 11.21 mm, were also significantly higher in irrigation strategies Int1 and Int2 in C2, respectively.

The N factor did not influence RWC, SD, PH, or HC in any of the two crop cycles. SL was not influenced by any of the treatments (strategy and N), but the values of this variable ranged from 9 to 13 cm in the two crop cycles (Table 1).

The irrigation strategies Int1 and Int2 promoted significant increases in PH and HC in both crop cycles. Gains of 13.13 and 14.71% in PH were recorded in C1, and gains of 13.23 and 11.99% were recorded in C2, respectively, compared to the Cont treatment. For the HC variable, the increments were 12.36 and 14.84% in C1 and 11.42 and 12.93% in C2, respectively, when compared to the Cont irrigation strategy (Table 1).

Table 2 shows that irrigation strategies influenced the TNL and NML of lettuce. In C1, the highest means of TNL (32.00 leaves/plant) and NML (27.62 leaves/plant) were

obtained with irrigation strategy Int2. In C2, the Int1 and Int2 strategies increased the TNL (33.03 and 34.46 leaves/plant) and NML (29.91 and 30.50 leaves/plant), respectively.

The N factor did not influence the TNL and NML variables in the two crop cycles. However, this factor promoted significant changes in the NNML in C1, and the N dose of 130 kg/ha caused a 20.46% reduction in the production of nonmarketable leaves (Table 2).

LA was significantly influenced by the N factor in C1 when its highest value (0.179 m²) was obtained at the N dose of 130 kg/ha. In C2, only the irrigation strategy factor caused significant differences in this variable, and the highest LA values (0.169 and 0.183 m²) were observed with irrigation strategies Int1 and Int2, respectively. However, the greatest differences occurred between the Int2 and Cont strategies (Table 2).

Irrigation strategies influenced TFM and MFM. The highest means of TFM (170.14 and 189.74 g/plant) and

Table 3. Yield (t/ha), water use efficiency (kg/m³), and harvest index of lettuce as a function of irrigation management strategies and nitrogen in two crop cycles: Means and standard deviation of total yield (TY), marketable yield (MY), non-marketable yield (NMY), water use efficiency of total yield (WUE_{TY}), water use efficiency of marketable yield (WUE_{MY}), and harvest index (HI).

| | TY | MY | NMY | WUE _{TY} | WUE _{MY} | HI |
|--------------------------|-------------|--------------|-----------|-------------------|-------------------|-----------|
| First cycle (C1) | | | | | | |
| Strategy | | | | | | |
| Cont | 43.01±4.97b | 40.81±5.51b | 2.19±0.74 | 44.67±5.17b | 42.39±5.72b | 0.94±0.02 |
| Int1 | 49.55±5.06a | 46.77±4.75ab | 2.78±1.87 | 51.46±5.25a | 48.57±4.93ab | 0.95±0.02 |
| Int2 | 55.30±5.44a | 52.44±5.10a | 2.86±1.01 | 57.43±5.65a | 54.46±5.29a | 0.94±0.01 |
| N (kg/ha) | - | - | - | - | - | - |
| 100 | 46.36±8.11b | 44.36±7.75b | 2.60±1.06 | 48.77±8.42b | 46.07±8.05b | 0.94±0.02 |
| 130 | 51.61±5.32a | 48.98±5.25a | 2.62±1.51 | 53.60±5.53a | 50.88±5.46a | 0.95±0.01 |
| ANOVA | | | | | | |
| STR | ** | ** | ns | ** | ** | ns |
| N | * | * | ns | * | * | ns |
| STR × N | ns | ns | ns | ns | ns | ns |
| Second cycle (C2) | | | | | | |
| Strategy | | | | | | |
| Cont | 38.12±8.51b | 36.42±8.02b | 1.69±0.63 | 44.29±9.89b | 42.31±9.32b | 0.95±0.01 |
| Int1 | 46.83±5.86a | 45.11±5.39a | 1.72±0.89 | 54.40±6.81a | 52.41±6.26a | 0.96±0.01 |
| Int2 | 49.50±4.82a | 47.17±4.84a | 2.35±0.79 | 57.50±5.60a | 54.77±5.62a | 0.95±0.01 |
| N (kg/ha) | - | - | - | - | - | - |
| 100 | 46.05±8.63 | 44.00±8.15 | 2.05±0.73 | 53.50±10.03 | 51.12±9.47 | 0.95±0.01 |
| 130 | 43.58±7.50 | 41.78±7.23 | 1.75±0.88 | 50.63±8.72 | 48.54±8.40 | 0.95±0.01 |
| ANOVA | | | | | | |
| STR | ** | ** | ns | ** | ** | ns |
| N | ns | ns | ns | ns | ns | ns |
| STR × N | ns | ns | ns | ns | ns | ns |

STR: irrigation management strategies. Means followed by different letters in the columns differ from each other by Tukey test at 0.05 significance level. * and ** indicate significance at 0.05 and 0.01, respectively. ns: non-significant interaction between factors.

MFM (159.02 and 178.28 g/plant) in C1 were observed in irrigation strategies Int1 and Int2, respectively, but the greatest differences in MFM were observed between Int2 and Cont. In C2, intermittent irrigation strategies (Int1 and Int2) again increased TFM (160.26 and 169.71 g/plant) and MFM (153.38 and 160.29 g/plant), respectively (Table 2).

The N factor also significantly influenced TFM and MFM in C1. The highest means of TFM (177.05 g/plant) and MFM (166.55 g/plant) were obtained with the application of 130 kg/ha of N (Table 2).

Yield, water use efficiency, and harvest index

TY and WUE_{TY} increased significantly using irrigation strategies Int1 and Int2 in C1. The highest values of TY

(49.55 and 55.30 t/ha) and WUE_{TY} (51.46 and 57.43 kg/m³) were found in the before-mentioned irrigation strategies, respectively. The MY (52.44 t/ha) and WUE_{MY} (54.46 kg/m³) were highest in Int2 (Table 3). In C2, irrigation strategies Int1 and Int2 maximized TY (46.83 and 49.50 t/ha), MY (45.11 and 47.17 t/ha), WUE_{TY} (54.40 and 57.50 kg/m³), and WUE_{MY} (52.41 and 54.77 kg/m³), respectively (Table 3).

The N doses influenced TY, MY, WUE_{TY}, and WUE_{MY} only in C1. The highest means of these variables, 51.61 t/ha, 48.98 t/ha, 53.60 kg/m³ and 50.88 kg/m³, respectively, were observed at the N dose of 130 kg/ha (Table 3).

The variables NMY and HI were not influenced by the treatments applied. However, in all treatments, the means of HI for the baby romaine lettuce cultivar evaluated in this study were higher than 0.94 (Table 3).

Discussion

Crop evapotranspiration (ET_c) and micrometeorological variables

The ET_c varied between the minimum and maximum limits of 0.68 to 4.35 mm/day in C1 and from 0.62 to 4.92 mm/day in C2 (Fig. 3a). The irrigation depths applied for all treatments in C1 and C2 were 96.28 and 86.08 mm per cycle, respectively. The irrigation depths applied in all treatments were lower than those water requirements of lettuce recommended by Andrade Júnior & Klar (1997) of 120 mm per cycle. These differences in irrigation depths can be explained by the climatic variations or variations in sowing periods and also those related to soil, cultural management and cycle of cultivars.

The internal micrometeorological conditions prevailing during C1 and C2, 35 and 30 days after transplantation (Fig. 3b, c), respectively, were favorable to the growth and development of the baby green romaine lettuce Astorga, considering that the average air temperature inside the greenhouse, in both cycles, was between the lower, 17 °C, and the upper, 28 °C, basal temperatures recommended by Abu-Hamdeh & Almitani (2016) for lettuce crops.

Air temperature is a determining factor for plant development stages and regulates transpiration rates and water status in the plant as a function of stomatal control. In lettuce cultivation, air temperatures well above the ideal range (17 to 28 °C) tend to reduce commercial quality and head formation, increase the production of leaves unfit for marketing, advance the flowering stage, and promote greater stem elongation, in addition to stimulating the production of latex and bitter flavor in the leaves.

The means of internal relative air humidity ranged from 67.69 to 94.04% and from 80.93 to 95.64% in C1 and C2, respectively (Fig. 3b). The relative humidity is a climate component that greatly influences plant transpiration rates and evapotranspiration, with ideal values for plant growth within the range from 60 to 90% (Allen et al., 2006; Santosh et al., 2017). However, high values of this component (80 to 100%) may favor the incidence of fungal diseases, limiting lettuce yield (Clarkson et al., 2014). Despite the high relative humidity observed inside the greenhouse (Fig. 3b), there was no incidence of severe fungal diseases that could compromise lettuce growth and yield.

The internal wind speed ranged from 0.0015 to 0.0050 m/s and from 0.0 to 0.0251 m/s in C1 and C2, respectively (Fig. 3c) and was always close to zero during the experimental periods, even on days with the approach of cold fronts with high external wind speeds. The daily values of wind speed observed in this study were below the range of 0.5 to 0.7 m/s, considered ideal for the growth and leaf gas exchange of plants grown in greenhouses (Santosh et al., 2017). In addition, this low air movement made it impossible to use the algorithm proposed by Allen

et al. (2006) to obtain the reference evapotranspiration of Penman–Monteith inside the greenhouse.

Vegetative growth and yield components

RCI is an indirect nondestructive tool used in N fertilization programmes since it reflects the intensity of green color in the leaves due to the chlorophyll content and the level of N present in leaf tissues. The increase in RCI with the application of 130 kg/ha of N at 14 DAIS in C1 indicates a higher efficiency of N assimilation because this nutrient favors protein and chlorophyll synthesis. Thus, a lower chlorophyll content reduces photosynthesis and the physiological activity of plants (Perchlik & Tegeder, 2018; Ali et al., 2019).

Despite the lower RCI values observed with the application of 100 kg/ha of N in C1 (Table 1), lettuce plants grown under this condition showed no symptoms of nutritional deficiency, such as yellowing and leaf drop, and completed the entire cycle normally. The RCI value of 41.12 obtained in this study is higher than that reported by Sobucki et al. (2019) of 20.00 for the lettuce cv. Stella with the application of 180 kg/ha of N. This divergence in RCI may be related to the cultivars used, the efficiency of N assimilation, and the evaluated periods.

The increments observed in RCI with the use of intermittent irrigation strategies (Int1 and Int2) are attributed to the decrease in the soil moisture front speed, which increases the permanence time and stability of soil water in the root region of crops as a consequence of the greater horizontal movement of soil water, optimizing the absorption of water and nutrients (Abdelraouf et al., 2019; Menezes et al., 2020a,b; Pereira et al., 2022). El-Mogy et al. (2012) and Eid et al. (2013) observed the highest RCI in green bean and soybean plants using intermittent irrigation of four pulses per day.

RWC is a physiological variable that has been used to indicate water deficit in plants since it reflects water status in the leaves (Badr & Brüggemann, 2020). The prolonged intervals between irrigations (1 h and 4 h), in addition to ensuring higher RCI values, as a probable response to the highest levels of chlorophyll and leaf N, also provided greater hydration of the protoplasm.

The greater water availability throughout the day and the higher water content in plant tissues optimize the stomatal opening process and transpiration, consequently reducing leaf temperature and maximizing photosynthetic efficiency. In this context, it is likely that irrigation management strategies Int1 and Int2 have mitigated the effects of water and thermal stresses that occur naturally throughout the day, even in a protected environment. Increasing the frequency of daily fertigation reduces soil water tension in the surface layer throughout the day, increases transpiration rates, and favors the influx of nutrients into lettuce roots (Xu et al., 2004).

It should also be added that the decrease in RWC is characteristic of plants grown under water deficit conditions (Badr & Brüggemann, 2020). The results obtained in this study corroborate those reported by Elnesr et al. (2015) and Zamora et al. (2019), who also found higher RWC in tomato leaves and coriander plants with the use of intermittent irrigation strategies, respectively.

Intermittent irrigation strategies also promoted significant increases in lettuce SD, PH, and HC. These results are due to the higher RCI and the higher water content in plant tissues observed under these strategies (Table 1), favoring vegetative growth and physiological activity. According to Zamora et al. (2019), intermittent irrigation is advantageous because it maximizes the vegetative growth of plants.

The higher HC values observed in Int1 and Int2 demonstrate the greater adaptability of the baby green romaine lettuce cv. Astorga to the microclimatic conditions inside the plastic greenhouse and to the management conditions, increasing the efficiency of water relations and photosynthetic rates. Madane et al. (2018a) mention that short intervals between irrigation events favor the optimal growth of crops and ensure balance in the water supply and nutrients throughout the growth period. Increases in plant height and fruit circumference using the intermittent irrigation strategy have also been reported in other plant species, such as green beans (El-Mogy et al., 2012), tomatoes (Elnesr et al., 2015), onion (Madane et al., 2018a, b) and sugar beet (Mehanna et al., 2020).

Despite the absence of significance of the treatments, the SL of the baby romaine lettuce cultivar used in this study ranged from 9 to 13 cm (Table 1). In crisphead lettuce, stem elongation is a characteristic that indicates flowering, whereas a shorter stem favors head formation, improves the quality of the product and is desirable in processing; the ideal and acceptable values are within the range from 6 to 9 cm (Yuri et al., 2017). However, the lettuce cultivar studied here showed no signs of flowering, changes in flavor, or a tendency toward elongation due to possible high temperatures occurring inside the greenhouse (Fig. 3b) but formed slightly compact heads even in the treatment that had lower SL values.

The number and size of leaves determine the yield of leafy vegetables such as lettuce. These characteristics depend on the availability of water and N in the soil and directly influence the interception of solar radiation and, consequently, photosynthetic production. In this context, the increments in TNL and NML observed in irrigation strategies Int1 and Int2 are attributed to the better maintenance of soil moisture in the surface layer throughout the day since the greater number of daily irrigations optimizes the efficiency of water application in the root zone of drip-irrigated plants and reduces water stress in the roots (Abdelraouf et al., 2019).

It is also likely that, with the continuous application of irrigation depth, lettuce plants have optimized the

mechanism of adaptation to daily stresses (water and thermal) and limited the number and size of leaves to avoid water loss by transpiration. Xu et al. (2004) found a significant decrease in water absorption by lettuce plants between 10:00 and 14:00 h with continuous application of daily fertigation. The results found in this study corroborate those obtained by El-Mogy et al. (2012) and Madane et al. (2018b), who also observed lower production of leaves in green bean and onion plants, respectively, with the strategy of continuous application of the irrigation depth.

The application of 130 kg/ha of N significantly reduced the production of nonmarketable leaves of lettuce in C1 (Table 2). This result is explained by the greater availability of N with the application of this treatment since the application of high levels of this nutrient is beneficial for vegetative growth, increases production per plant, and improves the quality of harvested lettuce (Sylvestre et al., 2019; Consentino et al., 2022).

The increase in N availability in the soil in both N fertilization treatments in C2 justifies the absence of a significant effect and the reductions of 34.30 and 18.38% in NNML with the applications of 100 and 130 kg/ha of N, respectively, when compared to the values of this variable in C1 (Table 2). Souza et al. (2017) found that the application of 171 kg/ha of N via fertigation increased the number of marketable leaves of the curly lettuce cv. Vera. It is important to highlight that the increase in temperature and relative humidity inside the greenhouse observed in C2 did not favor the increase in the NNML.

The increments in LA observed in treatments with 130 kg/ha of N in C1 and in the irrigation strategies Int1 and Int2 in C2 (Table 2) reflect the higher TNL also observed in these treatments, whose plants produced larger leaves and at greater intensity, maximizing the interception of solar radiation and the capacity to produce photoassimilates necessary for vegetative growth. Increased LA has been common in crops irrigated with an intermittent irrigation strategy (El-Mogy et al., 2012; Eid et al., 2013; Mehanna et al., 2020).

Nitrogen increases the rates of net CO₂ assimilation, RuBisCO activity, and leaf chlorophyll levels in plants, in addition to increasing leaf area and maximizing vegetative growth, yield, and, consequently, biomass production (Perchlik & Tegedere, 2018). The LA value of 0.179 m² observed in this study with the use of 130 kg/ha of N is close to those obtained by Stagnari et al. (2015) and Sobucki et al. (2019), who found higher LA, 0.156 and 0.170 m², in the lettuce cvs. Bionda Degli Ortolani and Stella with applications of 150 and 180 kg/ha of N, respectively.

The TFM and MFM values increased with both intermittent irrigation strategies in the two crop cycles and under the application of 130 kg/ha of N in C1 (Table 2). These results are due to the higher water content present in leaf tissues and the increase in the number and size of leaves in general observed in these treatments, directly leading to gains in leaf area and, consequently, in leaf fresh mass.

The gains in TFM and MFM of the baby green romaine lettuce Astorga with the application of 130 kg/ha of N in C1 are also justified by the reduction of NNML caused by this treatment (Table 2). The results found in this study are similar to those reported by Consentino et al. (2022), who obtained a higher fresh mass of the lettuce cv. Canastra with the application of 60 and 120 kg/ha of N in treatments not inoculated by microorganisms. However, these results differ from those reported by Souza et al. (2017), who observed higher TFM and MFM of the cv. Vera with the application of 171 kg/ha of N.

Yield, water use efficiency, and harvest index

Yield is a variable totally dependent on the physiological behavior and vegetative growth of crops and on the management strategies employed, and it is characterized as an important economic indicator of cultivated areas. In this study, intermittence in daily irrigation management promoted significant increases in RCI, RWC, PH, HC, TNL, NML, TFM, and MFM due to the greater water availability in the soil throughout the day. These results justify the increase in the TY and MY of lettuce when irrigated using irrigation management strategies Int1 and Int2. An increase in yield with the use of intermittence in irrigation management has also been reported for other vegetable crops, such as coriander (Zamora et al., 2019), onion (Madane et al., 2018a, b), and sugar beet (Mehanna et al., 2020).

The N dose of 130 kg/ha promoted considerable gains of 10.17 and 9.43% in TY and MY, respectively, compared to the N dose of 100 kg/ha in C1 (Table 3). N is a fundamental primary macronutrient in the synthesis of amino acids, proteins, and chlorophyll and acts directly on cell formation and division, in addition to increasing the vegetative growth and yield of crops such as lettuce (Souza et al., 2017; Perchlik & Tegedere, 2018; Sylvestre et al., 2019). The higher the RCI and RWC were, the greater the expression of vegetative growth, and the gain in yield observed at the N dose of 130 kg/ha in C1 indicated an adequate N fertilization level.

The highest means of TY and MY of the baby green romaine lettuce cv. Astorga evaluated in the present study, 51.61 and 48.98 t/ha, are higher than those reported by Sylvestre et al. (2019), of 20.30 and 44.60 t/ha, obtained in the summer and winter seasons for the crisphead lettuce Vanda with the conventional application of 60 and 120 kg/ha of N, respectively. Thus, it can be inferred that these variations in yield due to N fertilization depend, among other factors, on the cultivar used, form and time of fertilizer application, and local edaphoclimatic conditions.

WUE represents the amount produced for each m³ of water applied to the crop. The results of this study indicated that the WUE_{TY} and WUE_{MY} were significantly higher under irrigation strategies Int1 and Int2 (Table 3). Generally, higher WUE in plants is followed by lower yield because, under stress conditions, such as water deficit, plants maximize the

efficiency of using water as a factor of production because the gain in yield is not proportional to the amount of water added (Pereira et al., 2019).

As observed in this study, the irrigation strategies Int1 and Int2 promoted significant gains in TY and MY (Table 3) and consequently optimized WUE as a production factor. The results of this study corroborate those reported by El-Mogy et al. (2012), Eid et al. (2013), Abdelraouf et al. (2019), Mehanna et al. (2020), and Silva et al. (2022), who also found increases in WUE with the use of intermittent irrigation strategies in green bean, soybean, orange, sugar beet, and peanut crops, respectively.

For the N factor, it was possible to observe that the N dose of 130 kg/ha increased WUE_{TY} and WUE_{MY} by 9.01 and 9.45% compared to the N dose of 100 kg/ha in C1, respectively (Table 3). These results are related to the gains in TY and MY also observed in this treatment since the application of adequate levels of N fertilizers during plant growth promotes higher yield and better quality of harvested products, in addition to maximizing the economic benefits (Du et al., 2018).

HI was not influenced by the treatments applied, and its values ranged from 0.94 to 0.96 (Table 3). HI is an index that expresses the commercial quality of lettuce, and the ideal values are above 0.70 (Rezende et al., 2017). This indicates that the treatments applied in this study were efficient in obtaining a high harvest index, keeping the crop free of injuries and pathogens, and that the microclimatic conditions inside the greenhouse were adequate for the baby lettuce cultivar used in this study.

This study allowed us to conclude that intermittent irrigation management at 1-h and 4-h intervals favored vegetative growth, increased yield and optimized the water use efficiency of lettuce grown in a protected environment. Intermittent fertigation did not favor the reduction of the N dose to be applied, and the application of 130 kg/ha of N only in the first cycle of baby lettuce is recommended. Further studies with the use of higher intermittence, varied time intervals between fertigation events, and a higher number of N levels to be applied are needed.

Authors' contributions

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