

Article

Effects of Elevated Atmospheric CO₂ Concentration and Water Regime on Rice Yield, Water Use Efficiency, and Arsenic and Cadmium Accumulation in Grain

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Abstract: (1) Background: Elevated atmospheric CO₂ concentration affects the growth and development of the rice crop. In Southern Brazil, rice is traditionally produced with continuous irrigation, implying a significant amount of water used. Besides, continuous flooding favors the uptake of toxic elements such as arsenic (As) and cadmium (Cd). In this work, one Brazilian rice cultivar (IRGA 424) was tested for the effects of elevated CO₂ concentration and different water regimes on rice yield, and As and Cd accumulation in grain. (2) Methods: Rice was grown in two CO₂ concentrations (400 and 700 μmol mol^{−1}) and two irrigation regimes (continuous and intermittent). It was evaluated the number of tillers, plant height, aboveground dry weight (ADW), water use efficiency (WUE), rice yield components, and As and Cd concentration in rice grain. (3) Results: Rice plants were taller and had a higher WUE when cultivated at e[CO₂]. The ADW and the rice yield component were not affected by CO₂ levels nor water regimes. Intermittent flood regimes had a lower average As concentration. The Cd concentration in the samples in both growing seasons and all treatments was below the limit of quantitation (8.76 μg kg^{−1}). (4) Conclusions: Enhanced CO₂ concentration did not affect rice yield, increased the WUE, and reduced As concentration in grains. Regarding water management, the intermittent regime enhanced WUE and promoted a reduction in As concentration in grains.

Keywords: ADW; climate change; free-air CO₂ enrichment; *Oryza sativa* L.; water management

1. Introduction

Rice (*Oryza sativa* L.) is a vitally important crop whose role in global food security is remarkable [1]. Along with wheat (*Triticum aestivum* L.) and maize (*Zea mays* L.), rice provides more than 42% of total calories consumed daily by humans [2]. Brazil and the U.S. are world rice exporters, with more than 60% of the total rice produced in the Americas [3]. This important crop is also threatened by climate change and by the declines in freshwater available. Agriculture will experience some of the most significant impacts of climate change due to its dependence on stable temperatures and reliable rainfall, being impacted by the increasing atmospheric concentration of CO₂ ([CO₂]) [4,5]. Climatic projections indicate increases in [CO₂] and corresponding increases in global average temperatures



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throughout the 21st century [6]. As a result, extensive efforts are underway to measure and determine ways to mitigate the impact of climate change on agricultural production [7,8].

Rice is grown using a continuous flood whereby a relatively constant flood depth is maintained throughout the growing season [9,10]. A continuous flood is primarily used because of its ability to suppress weeds [11], improve herbicide efficacy [12], stabilize labile nitrogen [13], and may moderate extreme temperatures during the reproductive stage when rice is susceptible [14]. However, this practice also leads to a higher amount of water used for irrigation [15]. Intermittent rice flooding, also known as alternate wetting and drying (AWD) irrigation, was developed by researchers at the International Rice Research Institute in Los Baños, Philippines [16,17] to assist farmers living in water-scare regions of Asia. Safe AWD [2] reduces irrigation applications by up to 50%, relative to continuous flooding, with little to no reductions in yield [15,18,19]. As such, it offers significant reductions in irrigation water use as water-intensive conventional flooding comes under increasing scrutiny as competition for freshwater grows [20].

Elevated atmospheric CO₂ concentration has a wide range of impacts on plant growth and development. As a C₃ species, rice has lower respiration rates and higher photosynthetic and metabolic efficiencies at high [CO₂] levels [21]. For example, a doubling of [CO₂] stimulated biomass accumulation of C₃ plants by up to 40% [22]. Root growth and development are also impacted [23], potentially resulting in higher uptake of nutrients and other soil substances. Nevertheless, the reductions in stomatal conductance and stomatal aperture associated with increased [CO₂] reduce transpiration losses [24], meaning that less water is transported through the xylem, and thus, less water and associated solutes are absorbed by roots. Thus, elevated CO₂ might increase root biomass and reduced the transpiration stream, impacting the uptake of nutrients and other materials.

Arsenic (As) and cadmium (Cd) are more available for plant uptake in anaerobic soils in continuously flooded rice fields [25,26]. Recent studies have raised concerns regarding As and Cd concentrations in rice grain [27,28]. Under anaerobic soil conditions, As is primarily present in its reduced form As (III), which is less bound to soil and, thus, more bioavailable to plants [29]. Increase [CO₂] and its effect on rice plants' growth and development could cause improvement in water use efficiency by regulating stomatal closure while also leading to higher As and Cd uptake from soil in rice grains.

Both the irrigation regime and CO₂ level impact the phenology [30,31] and physiology [32,33] of rice. However, studies investigating interaction(s) between these factors are limited. In particular, few reports regarding the combined impacts of an intermittent irrigation regime and increased [CO₂] on rice grain yield and other yield components or how these factors might influence As and Cd's accumulation in the grain. Thus, this study's objectives were to evaluate the effects of increased CO₂ and two irrigation water regimes on rice development, water use efficiency (WUE), and arsenic and cadmium accumulation in rice grain.

2. Materials and Methods

2.1. Experimental Conditions

The experiments were conducted during the 2017/18 growing season and repeated during the 2018/19 growing season. Both experiments were carried out in a wood-framed open-top chamber (OTC), measuring 1.90 × 1.90 × 2.00 m (width/depth/height) using a method previously described [34]. The sides of the OTC were covered by a 150 µm transparent plastic film (low-density polyethylene—LDPE) attached to the wooden frames. The CO₂ concentration was maintained by an automated CO₂ control system, which measured and adjusted the CO₂ concentration at 30 s intervals from rice planting to harvest [35].

The rice plants were grown in separate plastic bins (0.36 × 0.63 × 0.33 m) filled with rice paddy soil (Albaqualf) collected from the 0–20 cm soil profile from a nearby rice field. Plastic bins were filled with 23 cm of soil, leaving a 10 cm space for flooding. Rice seeds (variety IRGA 424 RI) were hand-planted in rows spaced 17 cm, with a seeding

rate equivalent to 110 kg of rice seeds ha^{-1} . At planting, it was applied the equivalent of 300 kg ha^{-1} of NPK (5-20-20) and during the irrigation cycle, urea (45% N) was applied at a rate of 150 kg ha^{-1} , in two different rice stages, the first immediately before rice flooding, and the second was performed at panicle initiation [36].

The experiments were carried out in a completely randomized design with a factorial arrangement with four replications per treatment. Factor A consisted of two levels of $[\text{CO}_2]$: ambient ($a[\text{CO}_2]$) at $400 \pm 50 \mu\text{mol mol}^{-1}$ and elevated ($e[\text{CO}_2]$) at $700 \pm 50 \mu\text{mol mol}^{-1}$, with rice being maintained for its entire cycle under these conditions. Factor B consisted of an irrigation management regime: continuous and intermittent flood. For each regime, the flood was initiated at V_4 [37]. A maximum flood depth of 10 cm was used for both irrigation treatments, but the continuous flood was maintained at 10 cm, irrigated daily, whereas the intermittent flood was allowed to subside until the soil surface was exposed to air when the flood was reestablished to a 10 cm depth. Both treatments were irrigated until grain maturity.

2.2. Variables Analyzed

For each bin, the following parameters were determined. To determine WUE, water use was monitored daily through graduated rulers installed in each bin and irrigated appropriately for each treatment. The total number of tillers at V_7 and plant height at R_3 were measured. When rice grains moisture content reached 21% (w/w), all plants were harvested, and the total number of panicles and grain yield (g) per bin was determined. A subset of 10 panicles from each bin was randomly selected, and the number of grains per panicle, spikelet sterility, and weight per thousand grains was determined. Spikelet sterility, expressed as a percentage of the total number of spikelets, was determined by counting the number of sterile spikelets separated from the filled grain sample. The total aboveground dry weight (ADW) was obtained after the straw samples were dried to a constant weight at 60 °C.

Determination of Arsenic and Cadmium in Rice Grain

Milled rice grain samples were analyzed for total arsenic (As) and cadmium (Cd) at the Biomaterials Contaminant Control Laboratory, Federal University of Pelotas, Pelotas, Brazil. Rice grain (100 g) was milled using a laboratory mill (Perten 3100, Perten Instruments, Sweden) equipped with a 35-mesh sieve. Next, the milled samples (0.50 g) were weighed using an analytical balance (AY220, Shimadzu, Philippines) and separately added to 100 mL chemically modified polytetrafluoroethylene (PTFE-TFM) vials to which 6 mL nitric acid (65%, Merck, Germany) was added. Before use, the nitric acid had been purified via distillation (Duopur, Milestone, Italy). Next, the samples were digested using a microwave oven (Multiwave 3000, Anton Paar, Austria), holding eight vials simultaneously. The digestion temperature and pressure were limited to 260 °C and 60 bar, respectively, as per the manufacturer's recommendations (Software Version v2.50, Anton Paar). The vials were capped, fixed to the rotor, and subjected to the following digestion program: (i) ramp from 0 to 1000 W for 5 min; (ii) maintenance at 1000 W for 5 min; (iii) cooling to 0 W for 20 min. The resulting solutions were transferred to volumetric flasks and volumes adjusted to 20 mL using ultrapure water ($18 \text{ M}\Omega \text{ cm}^{-1}$) produced by a Simplicity™ UV (Millipore, Germany) purification system. Analytical blank controls containing no rice grain were prepared as previously described to quantify background As and Cd concentrations.

Total As and Cd concentrations in digestion solutions were measured by inductively coupled plasma mass spectrometry (ICP-MS; NexION 300X, PerkinElmer, Waltham, MA, USA) using a concentric nebulizer (Meinhard, Golden, CO, USA), cyclonic-type nebulizer chamber (PerkinElmer), and quartz torch and quartz injection tube (2 mm internal diameter). The limits of detection (LOD) and quantification (LOQ) were calculated as 3- and 10-times background, respectively, as determined for As and Cd using non-fortified rice flour blanks. The sample preparation method used was optimized from the conditions described by Cerveira et al. [38].

2.3. Statistical Analysis

Statistical analyses were performed using R software [39]. Before the analysis of variance, the data were tested for normality. For the ANOVA, the growing-season effect was considered a random variable. When there was no difference between growing seasons, the data were combined. When ANOVA identified significant differences, Tukey's test was applied using 95% confidence intervals.

3. Results and Discussion

Rice tillering was affected by the growing season (Figure 1). The numbers of rice tillers measured during the 2017/18 growing season were not affected by atmospheric $[\text{CO}_2]$ or irrigation regimes (Figure 1A). In contrast, for the 2018/19 growing season, the average tiller number for the intermittent flooding treatment was higher under $e[\text{CO}_2]$ than $a[\text{CO}_2]$. No such differences were observed between $[\text{CO}_2]$ when the rice was grown using a continuous flood (Figure 1B). One explanation for the observed difference in tillering between growing season is that light incidence and temperature differs between growing season (Figure 2). Light and temperature were not controlled, as is typical for open-top chamber experiments conducted outdoors [34]. The minimum critical temperature for rice tillering is between 9 and 16 °C [36]. Figure 2 shows that minimum temperatures measured during the vegetative phase were lower than this threshold during the 2018/19 growing season; consequently, tiller production was reduced in 2018/19 compared to 2017/18. Low temperatures also alter the basal metabolism of plants, affecting all stages of crop development [40].

Rice plants in intermittent flooding produced more tillers at $e[\text{CO}_2]$ than at $a[\text{CO}_2]$. The increase in tillers and aboveground biomass was observed in another study with rice grown under $e[\text{CO}_2]$ [41]. A comparison between rice cultivars using FACE (free-air CO_2 enrichment) showed a general trend towards higher tillers production with enhanced CO_2 concentration [42]. The researchers noted differences in responses to elevated $[\text{CO}_2]$ among rice cultivars and growing season but did not find differences in tiller production when rice was grown using a continuous flood [42]. Increased tiller numbers with intermittent flooding were attributed to the periodic wetting–drying cycles increasing the plants' exposure to more sunlight and CO_2 . A rice plant's ability to produce tillers depends on several factors, including genetic aspects, seedling density, temperature, sunlight, nitrogen availability, and floodwater depth [37]. Sunlight enhances the tillering in rice because it enhances photosynthesis as the seeds' energy reserves are quickly exhausted [43]. Regarding CO_2 atmospheric concentration, in addition to the increase in aboveground biomass resulting from increased photosynthesis [44], higher CO_2 levels also increase root growth [22,23,45]. More roots lead to increased soil nutrients uptake [46], further enhancing tiller production.

Plant height was measured at flowering and harvest; in both stages, the plants were taller in the 2018/19 growing season than in 2017/18 (Figure 3). The shorter plants in the 2017/18 growing season were likely due to increased energy expenditure associated with tillering. In both growing seasons and independently of water management, plants growing under $e[\text{CO}_2]$ were taller than those grown at $a[\text{CO}_2]$. Similar results were obtained by another study with rice grown at $e[\text{CO}_2]$ [47]. Studies indicate that increases in plant height may involve the partition of nutrients when grown under $e[\text{CO}_2]$ conditions, directed to aboveground plant growth [48]. The transition from the vegetative to reproductive phase represents a critical period in crop development since, during this transition, numerous factors affecting growth often have significant impacts on final grain yield.

Regarding ADW, there were differences between growing seasons, but there was no consistent effect of CO_2 level or irrigation regime on ADW (Figure 4). For example, ADW was greatest under intermittent irrigation and $e[\text{CO}_2]$ in 2017/18, but the continuous flooding had higher ADW in the 2018/19 growing season. These results are different from other studies where rice grew taller at $e[\text{CO}_2]$ but had no corresponding ADW increases.

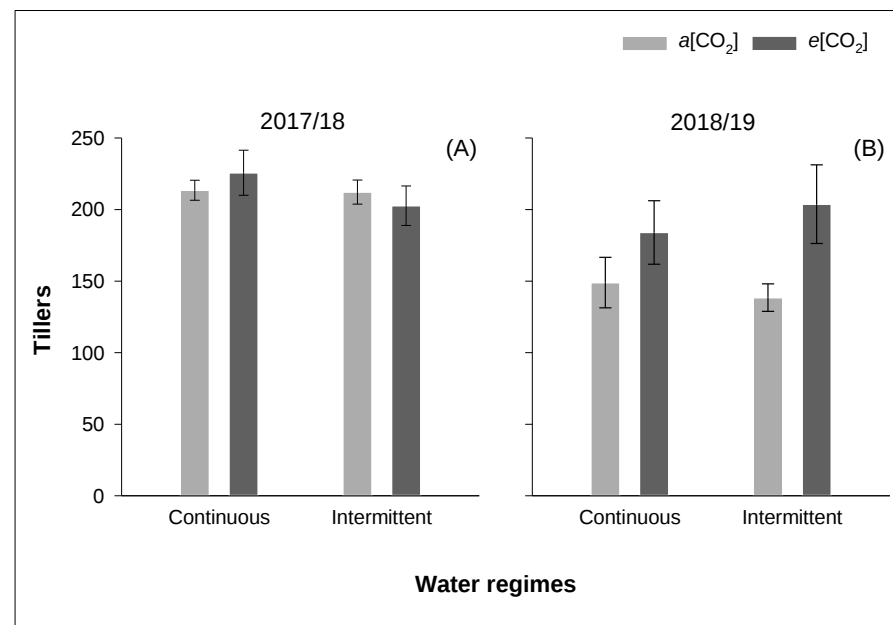


Figure 1. Effects of CO_2 concentration ($a[\text{CO}_2] = 400 \mu\text{mol mol}^{-1}$ and $e[\text{CO}_2] = 700 \mu\text{mol mol}^{-1}$) and irrigation regime (continuous vs. intermittent flood) on rice tillers per bin in 2017/18 (A) and 2018/19 (B). Error bars correspond to 95% confidence intervals about means.

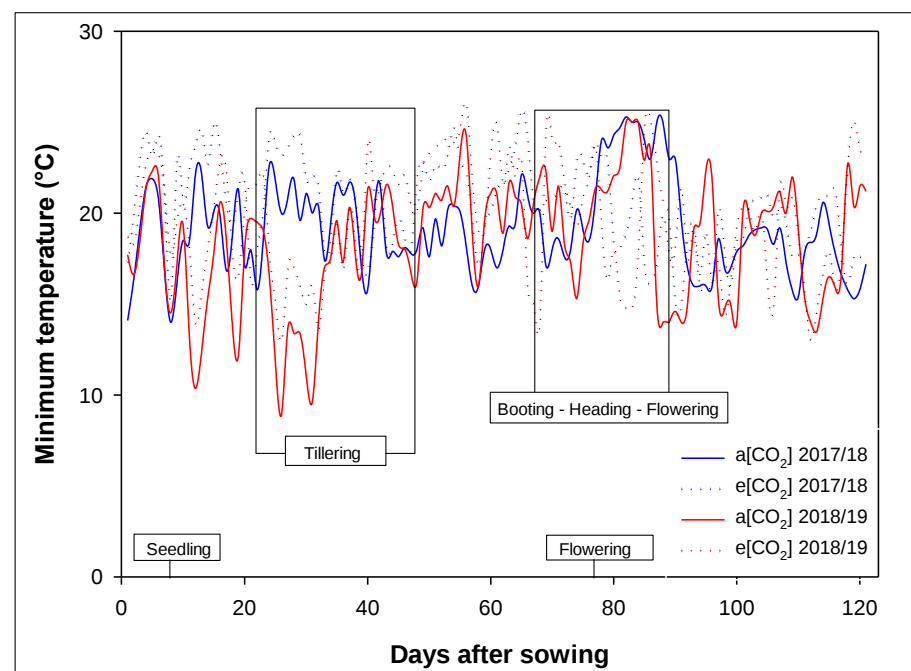


Figure 2. Average daily minimum temperatures measured inside the open-top chambers during 2017/18 and 2018/19 growing seasons maintained at two CO_2 concentrations ($a[\text{CO}_2] = 400 \mu\text{mol mol}^{-1}$ and $e[\text{CO}_2] = 700 \mu\text{mol mol}^{-1}$). The study was conducted in Pelotas, Rio Grande do Sul State, Brazil.

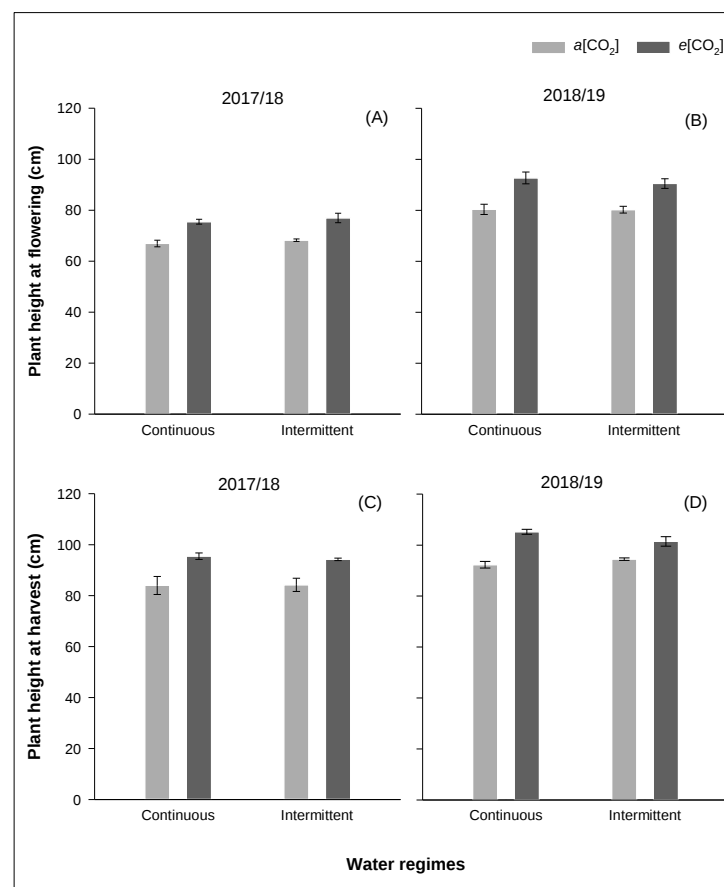


Figure 3. Effect of atmospheric CO_2 concentrations ($a[\text{CO}_2] = 400 \mu\text{mol mol}^{-1}$ and $e[\text{CO}_2] = 700 \mu\text{mol mol}^{-1}$) and water regimes (continuous vs. intermittent flood) on plant height at flowering (A,B) and plant height at harvest (C,D) in the two seasons growing (2017/18 and 2018/19). Error bars correspond to 95% confidence intervals.

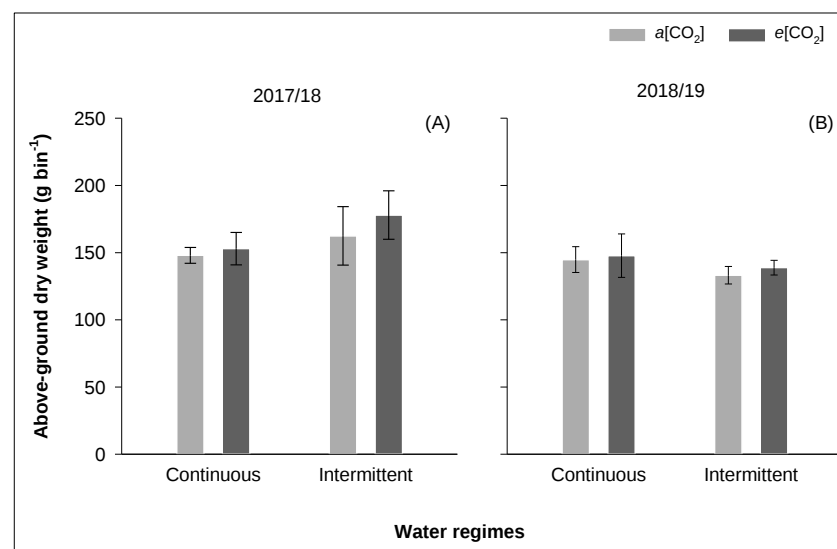


Figure 4. Effect of atmospheric CO_2 concentrations ($a[\text{CO}_2] = 400 \mu\text{mol mol}^{-1}$ and $e[\text{CO}_2] = 700 \mu\text{mol mol}^{-1}$) and irrigation water regimes (continuous vs. intermittent flood) on the aboveground dry weight of rice grown in open-top chambers in 2017/18 (A) and 2018/19 (B) growing seasons. Error bars correspond to 95% confidence intervals.

Rice yield components varied between growing seasons (Table 1). In 2017/18, there was no CO₂ and irrigation regime effect on panicle number, the number of grains per panicle, weight per thousand grains, grain yield, or spikelet sterility. In the 2018/19 growing season, CO₂ concentration and water management also had no clear impacts on most of these parameters, but panicle numbers and grains per panicle decreased, and spikelet sterility increased compared to 2017/18. The reduction in these important yield components in the 2018/19 growing season likely reflects less-than-optimal growing conditions (i.e., sunlight, cooler temperatures at tillage). Grain yields were generally higher for rice grown in $e[CO_2]$, but no statistical differences were owing to relatively high variability between the samples (Figure 5).

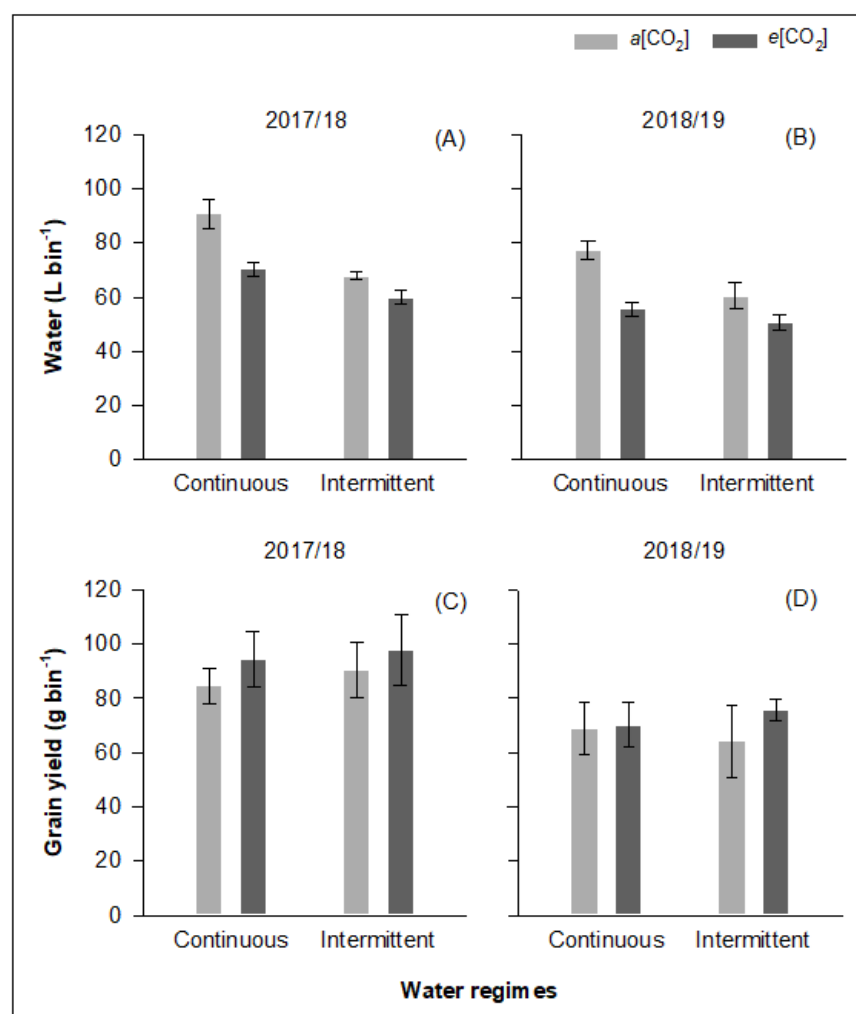


Figure 5. Effect of CO₂ concentrations ($a[CO_2] = 400 \mu\text{mol mol}^{-1}$ and $e[CO_2] = 700 \mu\text{mol mol}^{-1}$) and water regimes (continuous vs. intermittent flood) on the water used for irrigation (A,B) and grain yield (C,D) of rice plants. Error bars correspond to 95% confidence intervals.

In 2018/19, higher spikelet sterility and lower numbers of grains were observed. In the 2018/19 experiment, minimum air temperatures reached as low as 14 °C at the booting stage (Figure 2). Temperatures of 15 to 19 °C during reproduction impair microspore development, increasing spikelet sterility and, thus, reducing grain yield [49]. Other low-temperature studies [14,50] reported increased spikelet sterility when rice plants were exposed to low temperature at the early microspore stage, typically 10–12 days before heading. Therefore, higher spikelet sterility and reduced grain yield observed in the 2018/19 experiment were likely due to low temperatures during this critical time.

Table 1. Effects of CO₂ concentrations (400 $\mu\text{mol mol}^{-1}$ vs. 700 $\mu\text{mol mol}^{-1}$) and water regimes (continuous vs. intermittent) on rice panicle number m², number of grains per 10 panicles, weight per thousand grains, and spikelet sterility for two growing seasons.

Panicles (m ²)		Grains per 10 Panicles		1000-Grain Weight (g)		Spikelet Sterility (%)		
2017/18 Season								
[CO ₂]			Irrigation water regime					
	CON ^a	INT ^b	CON	INT	CON	INT	CON	INT
<i>a</i> [CO ₂] ^c	523 ± 50 ^{e ns}	520 ± 80	636 ± 66 ^{ns}	732 ± 29	18 ± 1 ^{ns}	18 ± 1	11 ± 1 ^{ns}	11 ± 1
<i>e</i> [CO ₂] ^d	520 ± 82	522 ± 68	711 ± 36	731 ± 53	18 ± 1	18 ± 1	10 ± 3	10 ± 3
CV(%) ^f	21.34		39.86		13.54		19.71	
2018/19 Season								
<i>a</i> [CO ₂]	580 ± 31 ^{ns}	552 ± 26	581 ± 65 ^{ns}	480 ± 143	17 ± 1 ^{ns}	17 ± 1	32 ± 4 ^{ns}	28 ± 2
<i>e</i> [CO ₂]	602 ± 37	574 ± 43	550 ± 136	649 ± 152	17 ± 1	18 ± 1	34 ± 9	31 ± 4
CV(%)	23.18		56.13		15.89		17.72	

^a Continuous flood (CON); ^b intermittent flood (INT); ^c *a*[CO₂] = 400 $\mu\text{mol mol}^{-1}$; ^d *e*[CO₂] = 700 $\mu\text{mol mol}^{-1}$; ^e mean ± standard deviation (n = 4); ^f coefficient of variation (CV); ^{ns} indicates no significant difference by Tukey's test.

As previously discussed, increased plant heights observed in 2018/19 may indicate that the rice plants invested in the growth of vegetative organs, such as stems and leaves, in higher quantities than in reproductive organs. Increased tillering and higher above-ground growth without a corresponding increase in grain yield were observed in another study [51]. When considering the allocation of photosynthetic carbohydrates in grain (sink) and leaf area (source), it is possible that spikelet sterility, caused by the low temperatures, may have reduced carbohydrate demand by the reproductive organs increasing translocation/retention of photosynthates to vegetative tissues [52].

The amount of water used to irrigate the plants was greater in 2017/18 than in the 2018/19 growing season, owing to warmer air temperatures (Figure 5). Comparing treatments, in both growing seasons, there was a greater amount of water used in the continuous compared to the intermittent flooding, results similar to those reported by others [15,18,19]. Regarding CO₂ treatments, less water was used under $e[CO_2]$ compared to the $a[CO_2]$ in both growing seasons. Many studies have shown that CO₂ enrichment limits stomatal opening, thus reducing transpiration losses [53,54].

The WUE was affected by irrigation management and atmospheric CO₂ concentration (Figure 6). Greater WUE was observed under intermittent irrigation compared to the continuous irrigation in both growing seasons, in a similar way that has been observed by other researchers [15,18,19]; regarding CO₂ effect, there was a greater WUE at $e[CO_2]$ compared to the $a[CO_2]$ in both growing seasons.

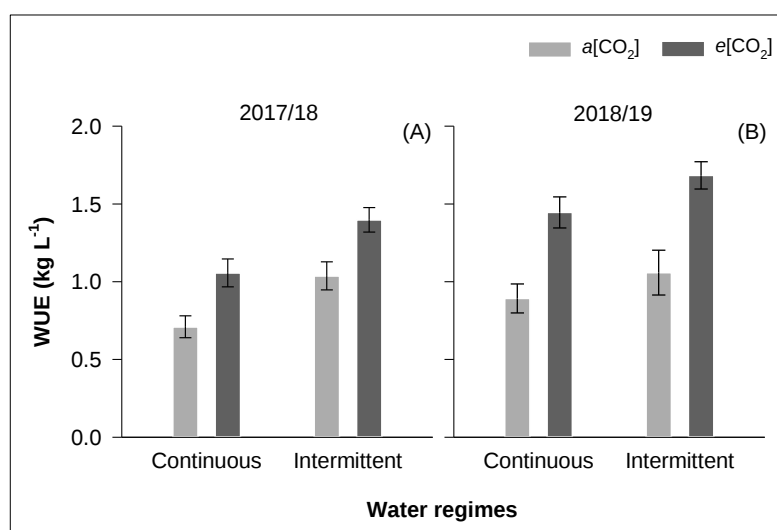


Figure 6. Effect of CO₂ concentrations [$a[CO_2]$ = 400 $\mu\text{mol mol}^{-1}$ and $e[CO_2]$ = 700 $\mu\text{mol mol}^{-1}$] and water regimes (continuous vs. intermittent flood) on the water use efficiency of rice plants in 2017/18 (A) and 2018/19 (B) growing season. Error bars correspond to 95% confidence intervals.

Total arsenic (As) concentration in milled rice grain was affected by the growing season and, thus, were analyzed separately (Figure 7). For As, the LOD was 1.37 $\mu\text{g kg}^{-1}$, and LOQ was 2.84 $\mu\text{g kg}^{-1}$. For Cd, the LOD was 4.64 $\mu\text{g kg}^{-1}$ and LOQ 8.76 $\mu\text{g kg}^{-1}$ for Cd. The trueness of the method was evaluated by analysis of a certified reference material of rice flour (CRM NBS 1568A—As: $290 \pm 30 \mu\text{g kg}^{-1}$ and Cd: $22 \pm 2 \mu\text{g kg}^{-1}$), agreement values of 107 and 109% were obtained for As and Cd, respectively.

Cd concentration in rice grain samples in both growing seasons and all treatments was below the limit of quantitation (8.76 $\mu\text{g kg}^{-1}$). Thus, this shows that either the soil had a lower concentration of available Cd or it was not absorbed. For As, the 2017/18 rice grain samples had higher concentration than those collected in 2018/19. The lower concentration of As in the 2017/18 growing season may be due to dilution, as there was a higher grain yield in 2017/18 compared to 2018/19. In the 2017/18 growing season, in the continuous flooding regime, there were no As differences between the two CO₂ treatments (Figure 7).

However, the intermittent flood regime had a lower average As concentration. A similar difference was observed in 2018/19, but higher overall As concentrations were obtained.

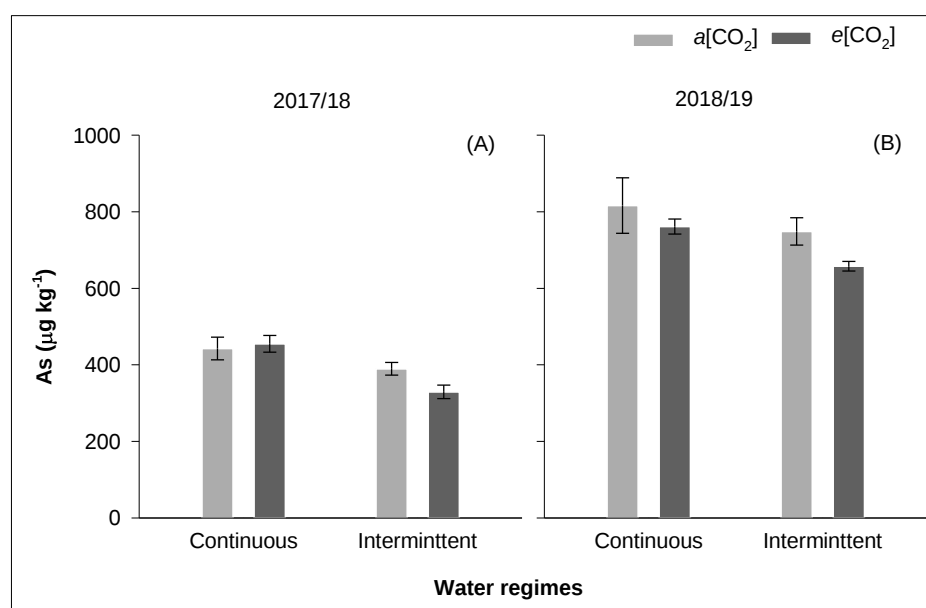


Figure 7. Effect of CO₂ concentrations ($a[\text{CO}_2] = 400 \mu\text{mol mol}^{-1}$ and $e[\text{CO}_2] = 700 \mu\text{mol mol}^{-1}$) and water regime (continuous vs. intermittent flood) on total arsenic ($\mu\text{g kg}^{-1}$) in husked rice grain plants in 2017/18 (A) and 2018/19 (B) growing season. Error bars correspond to 95% confidence intervals.

Researchers have investigated As uptake and concentration in rice grain along two central lines: toxicity in the plant [55,56] and accumulation in grain [26,57]. Studies have demonstrated that high [CO₂] reduces the concentration of several micronutrients, including iron, zinc, copper, calcium, and manganese, in rice grains and other species [58,59]. As previously mentioned, uptake reduction can be related to reductions in plant transpiration under $e[\text{CO}_2]$ conditions that change the nutrients uptake from the soil, translocated via water [48]. The present study corroborates findings where As concentrations in rice grain did not increase under $e[\text{CO}_2]$. Additional research may show that differences in As accumulation exists between rice varieties [60].

In a study comparing intermittent and continuous flooding, up to 24% reductions in As concentration were obtained in rice grains using an intermittent flood regime [61]. A study developed with different soil-water content to assess rice grains' toxic elements availability indicated lower concentrations of As in less saturated conditions [62]. An intermittent regime helps decrease As uptake, since oxygen can interfere with the bioavailability of As through the active root-associated microbiota involved in arsenic cycles [63]; this factor may be associated with our results. Our results also showed higher As concentration in rice grain, above the value $300 \mu\text{g kg}^{-1}$, which is the maximum allowed concentration in brown rice in Brazil [64]. It is essential to state that the absolute As concentration may not be taken into account, as our experiment was conducted in boxes in OTC with limited soil profile, and we can speculate that in field conditions, the magnitude of toxic elements uptake can be very different from what was observed in these conditions. For this reason, we use this information only for comparison between treatments.

However, in a monitoring study of total As concentration in rice grains from different Brazilian regions, concentrations ranged between $<2.6 \mu\text{g kg}^{-1}$ and $630 \mu\text{g kg}^{-1}$, indicating that rice grains were contaminated with values above the limit in the Rio Grande do Sul state [62]. The speciation study in the monitoring mentioned earlier indicated averages between 68 to $174 \mu\text{g kg}^{-1}$ of inorganic As in rice from the flooded system. Therefore, the total concentration study is not the best parameter to assess the risk of consuming this rice, and the grains may contain a high concentration of total As and, in this total,

low concentrations of most toxic species (inorganic As) [55]. Besides that, in the condition of $e[\text{CO}_2]$, several factors can be altered, influencing the As uptakes, such as higher root growth and changes in the composition of the rhizosphere. It is also essential to carry out FACE studies with different rice cultivars, as they may have different As uptake, translocation, and accumulation capacities in vegetable tissue and grains in field conditions.

4. Conclusions

This study showed that the rice cultivar IRGA 424 RI had higher growth under $e[\text{CO}_2]$, but there was no increase in grain yield. Moreover, rice grown at $e[\text{CO}_2]$ exhibited an increase in water use efficiency. While additional data are needed, the current study suggests that As concentration decreases in plants grown at $e[\text{CO}_2]$.

The intermittent water regime is more efficient using less water than the continuous regime. The total As concentration in rice grains decreased for plants grown in the intermittent regime. Although Cd was not detected in the samples, future studies need to determine the influence of $e[\text{CO}_2]$ and the water regime on the uptake and accumulation of Cd in rice grains.

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