# Rice irrigation management effects on water productivity, grain quality and food safety

by

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Submitted in fulfilment of the requirements for the degree of

Master of Science (Research)

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Submitted in fulfilment of the requirements for the degree of Master of Science (Research)

Deakin University (Australia), December 2019



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#### Abstract

The rice sector is facing great challenges in the coming years of not only achieving high yields to meet global food demand but also to use less water, energy, and other inputs per unit of production. This also needs to be achieved without compromising the environment and maintaining food safety.

Rice farming systems in Uruguay are at the leading edge of productivity and fields are fully irrigated and continuously flooded. Water is becoming increasingly scarce due to environmental concerns, climate change reducing water availability and competition from other sectors. New irrigation techniques need to be developed to use less water. These techniques will also need to minimize off-site impacts while preserving grain yield, quality and food safety. Increases in water productivity would allow rice production to expand and/or allow the allocation of water to irrigate other crops and/or other users such as urban and industrial. In addition, increases in water productivity will reduce pumping costs, improving the economic results and sustainability of the rice industry.

The focus of this study was to determine irrigation techniques that increase water productivity (WP), allowing a reduction in water input without negatively affecting grain yield in Uruguay. Between 2009 to 2015, a total of ten experiments were conducted in the northern, central and eastern rice growing regions of Uruguay. Treatments included: early continuous flooding (C), alternate wetting and drying (AWD), intermittent flooding until panicle initiation (IP) and intermittent flooding during all crop growth periods (I). The irrigation treatments were investigated in a delayed flood, drill-seeded rice production system. All treatments were planted on dry soil. In treatment C which represents the traditional irrigation management regime (i.e., control), flooding started 15-20 days after emergence and a water layer of 10 cm above the soil surface was maintained throughout all the crop cycle. In treatments IP and I, the water level alternated between 10 cm and 0 cm and was reestablished when the soil was still saturated. The AWD treatment allowed the soil to dry periodically (water depletion of 50% of soil available water) until panicle initiation. After this period, the field was continuously flooded as the control

treatment. IP and I led to significant savings in irrigation water inputs in the North and Central regions (averaged 35% or - 3986 m<sup>3</sup> ha<sup>-1</sup>) in relation to C. In the East region, AWD allowed for a 29% (-2067 m<sup>3</sup> ha<sup>-1</sup>) water saving in relation to the control but resulted in a significant yield loss of 1339 kg rice ha<sup>-1</sup> (15% reduction) in relation to C. WP was increased by 0.25kg m<sup>-3</sup> (23%) in IP and 0.68 kg m<sup>-3</sup> (62%) in I, in relation to the control C. The whole grain percentage was significantly reduced with I in the North region only. Techniques that maintained the soil at saturated water conditions like intermittent flooding, allowed a reduction of water input with no significant effects on grain yield, which led to a significant increase in WP in relation to the control C treatment.

A second objective of this study was to determine the inorganic arsenic (iAs) accumulation in rice grain in two contrasting soils commonly used for rice production in Uruguay. This research project also aimed to identify alternative irrigation management techniques to traditional flooding that could be used to limit or reduce the inorganic arsenic accumulation in the grain and to determine differences in the iAs levels within the most commonly planted rice varieties in Uruguay. To this end, five experiments were conducted with a split plot design with four blocks over three rice growing seasons from 2014 until 2017. The experimental sites included two irrigation treatments: continuous flooded (C) and alternate wetting and drying (AWD). The split plots included different varieties: *Indicas* and *Japonicas*. Average iAs accumulated in rice grain was 0.07 mg kg<sup>-1</sup>, well below international limits, even under the C irrigation technique. It was found that iAs accumulation in rice grain can be further reduced by the implementation of AWD in certain soils. *Japonica varieties* had a lower accumulation of iAs in rice grain, in comparison with *Indicas* at both sites.

In summary, this study identified irrigation techniques that used significantly less irrigation water while maintaining rice grain yield and therefore increasing water productivity, across a range of typical irrigated rice growing environments in Uruguay. Intermittent irrigation until panicle initiation was found to be the lowest risk technology that allowed a reduction in irrigation water used without negatively affecting rice yield, leading to a significant increase in water productivity. Grain yield was not reduced with irrigation techniques that maintained soil moisture above or near saturated conditions. When the soil moisture dropped below saturation even during the vegetative period, yield was found to be affected negatively. Alternate Wetting and Drying techniques allowed soil moisture to drop below saturation and yield was affected negatively.

Inorganic Arsenic levels (iAs) in two experimental rice growing sites evaluated in Uruguay were found to be well below the limit proposed by the international standards CODEX of 0.20 mg kg<sup>-1</sup> (FAO and WHO, 2019). Alternative irrigation management techniques such as AWD, resulted in lower levels of iAs accumulated in rice grain in relation to continuous flooded treatment at one of the evaluated experimental sites in Uruguay. Rice variety was found to significantly affect iAs uptake and accumulation in rice grain. *Japonica* varieties were found to accumulate lower amounts of iAs in grain relative to *Indicas*.

Based on the results obtained using @risk, an average income loss of implementing IP in relation to C of -53.7 US\$ ha<sup>-1</sup> was expected, considering the average rice price of 217 US\$ ha<sup>-1</sup> and a water price of 0.017 US\$ m<sup>-3</sup> with 90% of probability. It was found in most cases that a loss in profitability occurred by implementing alternative irrigation technologies such as IP, I, AWD in relation to the control treatment continuous flooding (C) using @risk modelling. Higher economic loss was registered in the East followed by the Central site. However, in the North, a lower net economic loss of implementing alternative irrigation management was found and the economic difference of implementing alternative irrigation managements could be negative or positive depending on water and rice prices variations. Traditional continuous flooding irrigation technique the most adopted practice in Uruguay in order to achieve the highest yield potential. As water payment in Uruguay is currently based on a fixed cost per irrigated hectare not by volume of water used, changes beyond flood management practices would likely be necessary in order for producers to be incentivized to implement alternative irrigation techniques that increase water productivity while improving the economic results. New irrigation technologies, geolevelling, automation of rice irrigation systems and rice breeding to develop cultivars that tolerate non-flooded conditions, could also play an important role for the successful implementation of alternative irrigation techniques on rice fields in the future.

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#### **CHAPTER 1**

#### **1. General Introduction**

# 1.1 Background - Problem Context (water scarcity and availability)1.1.1 Worldwide

Rice is the major staple food crop globally with more than 50 kg consumed per person annually and is the largest irrigated crop in the world with a higher water demand in relation to other cereal crops (Pimentel et al., 2004; FAO, 2018). Additionally, a growing population and rising global food requirements will contribute to an increase in water use demand, increasing the competition for this resource within agricultural, industrial and urban users (Bouman et al., 2007a).

Water is a resource that is becoming increasingly scarce in some parts of the world (Tuong and Bouman, 2003; Rijsberman, 2006; Mekonnen and Hoekstra, 2016). This can be attributed to the increased occurrence of drought periods, aquifer over extraction and loss of water quality by sewage, chemical pollution and salinization (Meybeck et al., 1996; Bouman et al., 2007a; Reba et al., 2013; Famiglietti, 2014). The Mississippi aquifer (MRVA) in the USA is an example which is declining at 0.15m per year (Yazoo Mississippi Delta Joint Water Management District (YMD), 2013, cited by Massey et al., 2014) due to increased extraction of groundwater for crop production. It is very likely that environmental, social and political demands will increase in the future due to a reduction in stream flows according to climate change projections (Christensen et al., 2007; Gaydon et al., 2010; Pittock, 2003). Water scarcity is imposed on farmers not only by drought, but also by environmental flow legislations and decision makers in some countries. As an example, water available for agriculture has been reduced and irrigators have experienced a reduction in their water allocations since 1997 in Australia (Gaydon et al., 2010). Rice cultivation on soils with high percolation losses are restricted in some countries by a water use limit policy and electromagnetic soil surveys which has led to a reduction in water used, improving water productivity (Beecher et al., 2002; Humphreys and Robinson 2003).

Climate change predictions by many models are indicating increases in temperature, more weather variability (Stocker et al., 2013) and higher frequency, duration and severity of water shortages (Spinoni et al., 2014) which would limit water available for irrigation and rice production in the future (Lobell, 2007; Lyman et al., 2013; Peng et al., 2004; Wassmann et al., 2009a, 2009b). According to Climate change projections for Australia, average Murray-Darling stream flows will be reduced from 16 to 48% by 2100, affecting water allocation for irrigation (Christensen et al., 2007; CSIRO, 2008; Gaydon et al. 2010; Pittock, 2003).

#### 1.1.2. Uruguay

The rice sector in Uruguay is divided in three regions: East (118391 ha), North (33 448 ha) and Central (12 618 ha) representing 72%, 20% and 8% of total annual rice planted area with an average annual rice yield of 8.5 ton ha<sup>-1</sup> (DIEA MGAP, 2018). The Uruguayan rice sector has been one of the most integrated in the country, which has contributed to rapid adoption of technologies and increased yields at one of the highest rates worldwide (Carracelas et al., 2017b, 2019a). Uruguay ranks seventh amongst rice exporters globally with an average (5 seasons) national total rice production of 1.4 million tons of paddy rice per year (DIEA MGAP, 2018) of which more than 90% is exported worldwide (FAO, 2018). The current economic scenario of high production costs and low grain prices resulted in a reduction in the rice cultivated area to 145000 ha in the 2018-2019 season, total grain production declined to 1.2 million tons as annual rice yield was 8.3ton ha<sup>-1</sup> (DIEA MGAP, 2019).

Uruguay has a subtropical to temperate climate with a great deal of secure water resources and an average annual rainfall ranging from 1200 mm (Southwest) to 1600 mm (Northeast) (Castaño et al. 2011). Average rainfall during the rice-growing season from October to March over a 17-year period (1988-2015) was 624 mm ranging from 301 - 934 mm per year (Carracelas, 2017a, b, GYGA, 2018). There is an opportunity to optimize rainfall capture and reduce irrigation inputs by implementing alternative irrigation management practices. Most of the water used to irrigate rice is pumped (56%) in Uruguay (DIEA MGAP, 2017). For these reasons, lowering the irrigation cost (water and pumping-energy cost) to increase profit are the main drivers for the

implementation of water saving techniques by farmers. The highest proportion of water to irrigate rice in this country is sourced from dams (54%) built for irrigation purposes (DIEA MGAP, 2017).

Rice is planted on dry field conditions and early flooded from tillering until 20 days before harvest which is also done on dry soils. There is an opportunity to optimize rainfall captured by implementing alternative irrigation techniques in which the rice crop is not continuously flooded, and significant water savings can be obtained. This would reduce irrigation pumping costs as the cost of energy is an increasingly pressing issue for farmers in this country. Water savings obtained by implementing such irrigation techniques would also leave more water available to irrigate other cereal crops and pastures and would create an opportunity for land-owners to make more profit and reduce risk by diversification of their products. Additionally, in dry years water stored in the reservoirs may not be enough to irrigate 100% of rice fields flooded during the entire growing season, affecting rice yields

The importance of testing and developing irrigation techniques that use less water while preserving crop yields in the case of Uruguay, is mainly associated with a mix of drivers such as assisting farmers to cope with water scarcity in dry years, reducing irrigation pumping costs, promoting expansion of rice crop area, minimizing environmental impacts, i.e. water footprint, greenhouse gas emissions and reducing heavy metals accumulation in grain.

Intermittent irrigation and safe alternate wetting and drying have been shown to be promising alternative irrigation techniques for reducing water use and increasing water productivity (Tabbal et al., 2002; Belder et al., 2004; Lampayan et al., 2005). Additional benefits such as improved food safety, through reduced arsenic accumulation in rice grain (Yang et al., 2017; Carrijo et al., 2018; Li et al., 2019) and reduced environmental impacts have also been shown (Linquist et al., 2015; Tarlera et al., 2016). In adopting these practices in Uruguay, it is important to research and understand the main factors affecting the success of these alternative irrigation techniques over a range of environmental and management conditions specific to Uruguay.

#### 1.2 Rice and Water

#### **1.2.1** Water Stress effects on rice plants

Rice is a crop very sensitive to water stress (Tuong et al., 2005; Bouman et al., 2007a), which is associated with its shallow root system (Parent et al., 2010), as well as other factors like diseases, weeds or nutrients. Rice plants are most sensitive to drought during flowering which is the rice critical period (Bouman and Tuong, 2001). Rice plants under water deficit, respond with a variety of mechanisms that can affect light interception, photosynthesis, phenology and grain yield components such as the number of panicles, grains per panicle and grain weight. The timing, duration, frequency and severity of the water stress imposed will determine the drought impact on grain yield. When the water stress is imposed during the vegetative period, the plant mechanisms are associated with a reduction in light interception and photosynthesis (decline in leaf area, closure of stomata, leaf rolling, leaf senescence and roots growth is prioritized), change in phenology, delay in flowering date and also a reduction in the number of tillers and, thus, panicles per hectare. On the other hand, when the water stress occurs during the reproductive period, yield components will be affected by a reduced number of spikelets, lower grain weight and increasing grain sterility which decreases the number of grains per panicle (Bouman and Tuong, 2001).

#### 1.2.2 Rice water use and requirements

Rice farming systems in Uruguay are one of its largest water consumers as traditional continuous flooding is the main irrigation technique implemented by farmers to secure the highest yields to maximize profit.

Irrigation water use ranges from 11000 to 14000 m<sup>3</sup> ha<sup>-1</sup> with an average of 12500 m<sup>3</sup> ha<sup>-1</sup> (Battello et al., 2009). There are high variations depending on soil characteristics and landscape topography/slope. Several authors reported rice water requirements for growth within the range from 4000 to 7000 m<sup>3</sup> ha<sup>-1</sup> (Pringle, 1994; Tabbal et al., 2002; Bouman et al., 2007a; Massey et al., 2014). Evapotranspiration recorded in rice crops in Uruguay ranged from 5500 to 6800 m<sup>3</sup> ha<sup>-1</sup> (Böcking et al., 2008). Conversely, the reported evapotranspiration rates during rice growing seasons in Australia were much

higher than those reported values ranging between  $11000 - 12000 \text{ m}^3 \text{ ha}^{-1}$  (Thompson, 2002). Net water input (irrigation water supply + rain – surface drainage) ranged from 15000 - 15600 m<sup>3</sup> ha<sup>-1</sup> in Australia for continuous flooded irrigation (Dunn and Gaydon, 2011).

The main benefits of flooding rice crops are related to more effective weed control (Baldwin and Slaton, 2001; Marchesi and Chauhan., 2019), an increase in nutrient availability (Dunn and Gaydon, 2010), lower disease incidence (Cartwright and Lee, 2001) and thermal insulation/protection from cold during micros-sporogenesis (William and Angus, 1994). However, in Uruguay with higher air temperatures, Roel, (2005) found no differences in temperature within the canopy by the application of a deep-water layer during this critical period.

Some disadvantages of the traditional continuous flooding technique presents against alternative irrigation techniques like the alternate wetting and drying (AWD) are associated with higher arsenic (As) accumulation in rice grain (Linquist et al., 2015; Yang et al., 2017; Carrijo et al., 2017; Carrijo et al., 2018; Seyfferth et al., 2018) and higher greenhouse gas (GHG) emissions (Linquist et al., 2015; Tarlera et al., 2016; Seyfferth et al., 2018).

Several water-saving irrigation techniques have been developed to reduce water use, associated irrigation costs or save water for other purposes (Bouman et al., 2007a) but they may have a negative impact on grain yield as rice is very sensitive to water stress (Tuong et al., 2005). Rice yields can be reduced under non-saturated soil conditions (Bouman and Tuong, 2001) and this could be associated with the shallow rice root system (Parent et al., 2010) as well as other factors like diseases, weeds or nutrients.

#### **1.2.3 Water Productivity**

Water Productivity (WP) is defined as kilograms of rice grain produced per unit of input water (kg m<sup>-3</sup>) (Bouman et al., 2007a). There are different definitions for WP according to the type of water used in its calculation:

- Irrigation water productivity (WPi) is defined as the kilograms of grain produced per m<sup>3</sup> of irrigation water input.
- Total water productivity (WPir) is defined as the rice yield over volume of total water inputs including irrigation and rainfall.
- Evapotranspiration water productivity (WP<sub>ET</sub>), is defined as rice yield over m<sup>3</sup> of evapotranspirated water.
- Transpiration water productivity (WP<sub>T</sub>), is rice yield over volume of transpirated water.

WPi and WPir information is valuable for irrigation engineers, managers and farmers that are interested in optimizing the productivity of irrigation water and total water resources including rainfall. These indices are also of interest to regional water resource planners interested in the amount of grain that be produced with available water resources (Bouman, et al., 2007a). Conversely, rice breeders are interested in the productivity of transpired water (WPt) or evapo-transpired water (WP<sub>ET</sub>) when selecting for more water-efficient cultivars. Reported WP<sub>ET</sub> average values ranged from 1.0 to 1.5 kg m<sup>-3</sup> for different irrigation techniques (Bouman et al., 2007b, Sudhir-Yadav et al., 2012) which were aligned with WP<sub>ET</sub> values reported in Uruguay in previous studies (Böcking et al., 2008).

The parameter WP can be improved by optimizing field layout and implementing irrigation management techniques that allow a reduction in water use while maintaining or increasing grain yields.

Irrigation techniques that maximize WP have many potential benefits. These include: allowing an increase in annual rice planted area as dams are the main source of water and a limiting factor for the expansion of rice crops, allocation of fresh water to other uses (urban or industrial), improved economic results through reduced pumping costs, increased total grain production to meet the growing demand for food worldwide, minimizing risks of water scarcity and improved sustainability of the rice sector.

#### **1.3** Alternative irrigation techniques for rice

It is important to continue developing irrigation techniques that use less water while preserving crop yields to cope with water scarcity while increasing total amount of grain to meet the increasing food demand worldwide. The most common irrigation practice for rice production is continuously flooded (C). The alternative irrigation techniques for rice are saturated soil culture (SSC) or intermittent irrigation (I) and alternate wetting and drying (AWD) (Tuong et al., 2005). Other less common irrigation management techniques used in rice crops are sprinkler irrigation (Muirhead, 1989) and aerobic rice techniques (Kato et al., 2009). Aerobic rice culture, furrow and sprinkler irrigation were beyond the scope of this study.

This thesis focused on the evaluation of irrigation management practices such as SSC, I and AWD because they have the potential to reduce water inputs without reducing grain yield when implemented properly. Furthermore, these technologies do not require extensive capital investment to be implemented.

#### **1.3.1 Intermittent Irrigation (I) or Saturated Soil Culture (SSC)**

This irrigation management technique consists in keeping the soil always saturated. Several studies conducted around the world, have reported that this irrigation management practice can reduce water inputs without reducing grain yield, thereby increasing WP. (Heenan and Thompson, 1984; Bouman and Tuong, 2001; Bouman et al., 2007a; Dong et al., 2001; Li, 2001; Tabbal et al., 2002; Marco and Marella, 2006; Thompson and Griffin, 2006; Böcking et al., 2008; Roel et al., 2011; Lavecchia et al., 2011; de Avila et al., 2015; Massey et al., 2014; Massey et al., 2018).

Bouman and Tuong 2001, found in several experiments an average water saving of 23 % without significantly reducing rice grain yield with saturated soil culture (SSC) management treatment in relation to continuously flooded irrigation (C). Further studies in direct seeded rice in non-puddled soil, determined a water saving of 49%, yield reduction of -4% and WP increased by 89% in rice grown under SSC compared to C (Tabbal et al., 2002). The main drivers of reducing water inputs by implementing

alternative irrigation techniques were associated with a reduction in percolation and floodwater runoff losses in relation to C (Bouman and Tuong, 2001; Dong et al., 2001; Li, 2001). Additionally, many authors reported that this reduction in irrigation water inputs was associated with an increase in rainfall capture by implementing intermittent irrigation technique (Li, 2001; Massey et al., 2014; de Avila et al., 2015; Massey et al., 2018). This technique allowed for a water input saving that ranged from 22% up to 76% by optimizing rainfall capture, without reducing grain yield compared with the C control (de Avila et al., 2015). Experiments conducted in the North of Uruguay across two seasons with intermittent irrigation, resulted in an average water saving of 25 % (ranging from 12% to 35%), WPi increased by 24% from 0.78 to 0.97 kg m<sup>-3</sup> in relation to C techniques and rice yield was not reduced in irrigated field areas where the soil was always kept saturated (Böcking et al., 2008). However, a significant yield reduction as soils dry out on the top of levees of -8% was found, which was equivalent to - 807 kg ha<sup>-1</sup> on intermittent compared to continuous flooding. Water saving through the implementation of I in relation to C across other studies were 12 % (Marco and Marella, 2006) and 35% (Henderson et al., 2008). Experiments conducted in the central region of Uruguay determined a water input saving of 18% with no significant differences in rice yield and an increase in WP from 0.88 to 1.04 kg m<sup>-3</sup> for I and C, respectively (Lavecchia et al., 2011). In Australia, the I irrigation until panicle initiation achieved a water input saving of 23 %, with no differences in grain yield in relation to the conventional method C (Heenan and Thompson, 1984). Further studies determined similar grain yields and increases in water productivity ranging from 0,06 to 0,23 kg m<sup>-3</sup> obtained with I irrigation in relation to C techniques (Thompson and Griffin, 2006).

However, when the intermittent irrigation technique was not implemented properly and soil was allowed to dry out, rice yield and grain quality could be penalized. Some authors recorded a significant rice yield loss when intermittent irrigation techniques were implemented (Borrell et al., 1997; Thompson, 1999; Henderson et al., 2008), when soils were not kept in a saturated condition (Lavecchia et al., 2011). Thompson, (1999), found in Australia that this technique reduced both water input and yield by more than 10%. Borrell et al., 1997, determined water savings of 34 % but yield losses of 16-34% with furrow irrigation and raised beds to facilitate the implementation of this alternative intermittent irrigation technique. Higher infiltration capacity soils and higher slopes impose a great challenge to correctly implement intermittent irrigation techniques and to keep the soil in a saturated condition.

Different water management practices have been tested in order to be able to implement and adapt safe alternative irrigation techniques. These focus on allowing quick re-flooding and uniformly levelled rice fields, allowing the soil to always remain saturated. Massey et al. (2014), found that intermittent irrigation combined with a multiple inlet rice irrigation (MIRI) system can be successfully implemented on commercial farm fields on clay soils, leading to a reduction in water used (4990 m<sup>-3</sup> ha<sup>-1</sup>) and a positive effect on grain yield in relation to continuous flooding. It was found that a reliable irrigation system with ample irrigation delivery well capacity allowed a quick reflooding of the field, ensuring water stress was not limiting production. Expertise in using the MIRI systems plus an integrated weed and disease management program was found as the key success elements for adapting intermittent irrigation to commercial farms (Massey et al., 2014).

In the USA, the Multiple Inlet Rice Irrigation (MIRI) allowed water to be distributed to all paddies simultaneously, with polypipe tubes installed perpendicular to levees (Vories et al., 2005 cited by Massey et al., 2014). Straight levees determined water savings of 17% (9650 m<sup>-3</sup> ha<sup>-1</sup>) while MIRI used with straight levees was found to reduce water inputs by 30% (7830 m<sup>-3</sup> ha<sup>-1</sup>). Zero grade leveling was determined as the most efficient water savings technique, with a reduction in water use of 55% (5008 m<sup>-3</sup> ha<sup>-1</sup>) in relation to contour-levee fields (11170 m<sup>-3</sup> ha<sup>-1</sup>) (Smith et al., 2007). It was found that I irrigation combined with the MIRI system could be successfully implemented on commercial farm fields on clay soils, leading to an important reduction in water used while increasing grain yields for most of the evaluated varieties in relation to continuous flooding (Yazoo Mississipi Joint Water Management District (YMD), 2013 cited by Massey et al., 2014).

#### **1.3.2 Alternate Wetting and Drying (AWD)**

This irrigation management technique consists of alternating saturated and unsaturated soil conditions by modifying the irrigation intervals, allowing soil water to drop down until the soil type reaches an aerobic state before the field is re-flooded. AWD is a water-saving technology where irrigation water is applied a few days after the disappearance of the ponded water. Hence, the field gets alternately flooded and non-flooded.

AWD is a promising management practice in reducing water inputs, improving WP, reducing arsenic (As) accumulation in rice grain and decreasing greenhouse gas emissions (GHG) (Linquist et al., 2015; Tarlera et al., 2016; Yang et al., 2017; Carrijo et al., 2018). However, there is a high degree of variation in rice yield response depending on timing, duration and severity during the drying event of this technique. It has been widely researched in different parts of the world such as China, northwest India, Philippines (Lampayan et al., 2005), USA (Linquist et al., 2015; Carrijo et al., 2017; Carrijo et al., 2018) and Australia (Humphreys et al., 2005; Humphreys et al., 2006; Dunn and Gaydon, 2011).

Most experiments reported a significant yield reduction in the AWD treatments compared with the control continuous flooded, with large variability in the results according to different soils type and differences in water stress level. Bouman and Tuong (2001), reported in most experiments a yield reduction up to 70% in the AWD treatments, compared with the control continuous flooded technique. However, WP was improved due to reduced water inputs. The authors attributed the large variability in those results to differences in irrigation frequency, different type of soils and hydrological conditions. Research in loamy and sandy soils with deeper groundwater tables in the Philippines showed a significant reduction in water inputs but yield was penalized compared with the flooded control treatments (Tabbal et al., 2002).

However, some experiments conducted on heavy soils in China and the Philippines reported a reduction in water used by implementing AWD of 15-30%, without significantly reducing grain yield in relation to continuous flooding (Tabbal et al., 2002; Belder et al., 2004; Lampayan et al., 2005). The authors hypothesized that in these fields shallow ground water depths (0.10-0.40 m) allowed roots to still have access to water even during the drying periods in AWD.

According to IRRI, the "safe" alternate wetting and drying technique allows a reduction in water used without penalizing rice grain yield when water depth dropped to no more than 15 cm below soil surface and the field is re-flooded to a water layer of 5 cm (IRRI, 2019). This is aligned with "Safe AWD" recommendations reported by different authors where ponded water depths never dropped below the root zone (Lampayan et al., 2009). It was also found that there was no yield penalty when soil water potential was higher than -20 kPa (Carrijo et al., 2017). Studies by Yang et al., (2017), found that moderate AWD allowed water savings, increased rice yields and rice quality improved (water table was maintained at 0.10 to 0.15 m and soil water potential was between -10 to -15 kPa). Additionally, it was found no yield penalty occurred even under severe AWD (soil water potential was lower than -20 kPa) when roots can still have access to water (Carrijo et al., 2018). The total amount of water used in rice crops in Australia was found to be reduced by 15% on average, when implementing delayed continuous flooding and implementing AWD techniques during the vegetative growth period. It was found that this alternative irrigation technique increased the crop growth duration period, increasing the risk of cold damage during the reproductive phase (Dunn and Gaydon, 2011).

#### **1.4 Rice and arsenic**

Increasing or maintaining grain yields while reducing heavy metal content in food is a great challenge the rice sector will increasingly be facing in the future. Arsenic levels in food are continuously monitored as they are frequently associated with high risk factors in food nutritional safety (Al-Saleh and Abduljabbar, 2017; Mitra et al., 2017).

Rice has naturally higher levels of As (Williams et al., 2007) as rice plants have a greater ability to absorb and accumulate it in the grain in relation to other staple food crops (Das et al., 2004). Additionally, the traditional continuous flooded irrigation technique favors the availability and absorption of this heavy metal by rice plants (Williams et al., 2007).

Arsenic is actually the heavy metal of most concern for the rice industry. Arsenic levels in food are strongly regulated by international standards and its compliance influences access to international markets which is crucial for exporting countries. The recommended inorganic arsenic (iAs) levels for polished and brown rice in the CODEX are 0.2 and 0.35 mg kg<sup>-1</sup>, respectively (FAO and WHO, 2019).

The levels of As and their forms in rice grain have previously been found to be affected by irrigation, varieties, fertilization and natural presence in air, soils and waters (Meharg and Zhao, 2012; Linquist et al., 2015; Mitra et al., 2017).

Continuous-flood irrigation\_techniques, and the anaerobic conditions that they foster can increase the availability and absorption of As by plants (Williams et al., 2007, Fendorf and Kocar, 2009). Several research studies worldwide have reported that AWD is an alternative technique that has led to reduce As accumulation in rice grain, contributing to improved food safety (Linquist et al., 2015; Das et al., 2016; Lahue et al., 2016; Yang et al., 2017; Carrijo et al., 2018; Li et al., 2019).

Differences within varieties have been reported in the As levels accumulated in root tillers and grain (Zhu et al., 2008). Accumulation of As in grain was found to be higher in *Indica* rice varieties compared to *Japonicas* (Jiang et al., 2011).

A key factor that influences As accumulation in paddy rice is soil type (Meharg and Zhao, 2012) which depends on the sediments that it is originated from. Reported natural concentrations of As in soils ranged from 5 to 10 mg kg<sup>-1</sup> worldwide (Han et al., 2003; Hossain et al., 2008) which are well below the Canadian limit for agricultural soils of 12 mg kg<sup>-1</sup> (CCME, Canadian Environmental Quality Guidelines). Soils with higher As content resulted in greater As accumulation in rice grain (Quintero et al., 2014). Arsenic concentration in the soil solution would reflect the bioavailability of arsenic because rice roots absorb As mostly from the soil solution (Xu et al., 2008). Arsenic bioavailability has been found to increase under reduced soil conditions (Kumarathilaka et al., 2018).

Another natural source of arsenic into the rice cropping systems is the As transported through irrigation water (Meharg and Zhao, 2012). There are also other possible

sources of arsenic such as industrial, urban pollution, contamination of irrigation water, use of fertilizers and pesticides contaminated with arsenic (Meharg and Zhao, 2012)

In the effort to mitigate rice accumulation of As there are other options like fertilization with minerals (Fe, S, P and Si) that competes with As uptake (Mitra et al., 2017). As an example, silicon management is a promising sustainable soil amendment that could increase plant-available Si to compete with As for root uptake (Seyfferth et al., 2018). Other strategies to reduce arsenic accumulation in rice grain are related to gene editing technologies (Mitra et al., 2017).

The implementation of the mentioned mitigation managements options and changes in cultivation practices are likely to be adopted in environments in which arsenic concentrations are an issue. This would contribute to reduce the levels of As to promote food safety, consumer health and sustainability of the rice sector globally.

Alternative irrigation management techniques tested in our study could not only potentially improve water productivity but additionally, could also reduce the accumulation of arsenic in rice grain.

#### 1.5 Aims and Objectives

#### 1.5.1 Research Gaps

Based on the review of literature conducted in this study, there were two broad research gaps identified which needed further research to develop management techniques to optimize rice water use productivity and rice quality in Uruguay. These were:

• Optimal irrigation management techniques that can be adapted for different soils and environments within the rice sector in Uruguay, allowing an increase in water productivity without reducing grain yields and quality.

• Alternative irrigation techniques to the traditional continuous flooding strategy that reduce heavy metals accumulation like inorganic arsenic in rice grain while reducing water used without penalizing yield and quality.

#### **1.5.2 Project justification**

It is important to continue developing irrigation technologies that use less water while preserving crop yields for the sustainability of the rice crops in Uruguay. Irrigated rice crops are the highest fresh water consumers and it is very likely that environmental, social and political demands will increase in the future, reducing the water availability within the sector.

This is the first integrated analysis study of different irrigation management practices in experiments conducted with different soil types and slope situations in all three rice-growing regions of Uruguay. Similar irrigation studies have been conducted around the world but on different soil types, slopes, irrigation management and field layout techniques under hydrological conditions different to those in Uruguay. These experiments were very important to be able to determine and adapt irrigation management practices to local conditions in Uruguay.

Increasing water scarcity has been an issue for some countries around the world (Mekonnen and Hoekstra, 2016). In contrast Uruguay has a great deal of secure water resources (streams, rivers, lagoon) and a subtropical to temperate climate with a high average annual rainfall. There is an opportunity to optimize rainfall capture and reduce irrigation inputs by implementing alternative irrigation management practices.

In summary, the importance of investigating irrigation technologies that use less water while preserving crop yields in the case of Uruguay is mainly driven by:

\*coping with water scarcity in dry years; climate change predictions by many models are indicating increases in temperature, more weather variability (Stocker et al., 2013) and higher frequency, duration and severity of water shortages (Spinoni et al., 2014) which would limit water available for irrigation and rice production in the future (Lobell, 2007; Lyman et al., 2013; Peng et al., 2004; Wassmann et al., 2009a, 2009b). In drought years, water stored in the reservoirs in Uruguay may not be enough to irrigate 100% of rice fields flooded during the entire growing season.

\* promoting the expansion of rice crop area as water is a limiting factor for the expansion of rice crops. Dams built for irrigation purposes are the main water source (54%) in Uruguay (DIEA MGAP, 2017).

\* increasing water availability for other irrigated cereal crops, pastures or other uses. Increasing water use efficiency would contribute to increase the irrigated agriculture area, creates an opportunity for land and water owners to make more profit and reduce risk by diversification of their products.

\* reducing irrigation pumping costs as most of the water used to irrigate rice in Uruguay is pumped (56%) (DIEA MGAP, 2017) while the cost of energy is an increasingly pressing issue for farmers in this country. Lowering the irrigation cost to increase profit and having enough water to adequately irrigate the crop to secure crop yield potential are the main drivers for the implementation of water saving techniques by farmers in Uruguay.

\* minimizing environmental impact; greenhouse gases emissions (Linquist et al., 2015; Tarlera et al., 2016).

\* food safety issues associated with the accumulation of heavy metals like arsenic in grain. Disadvantages of the traditional continuous flooding technique are higher arsenic (As) accumulation in rice grain (Linquist et al., 2015, Yang et al., 2017; Carrijo et al., 2018; Seyfferth et al., 2018).

#### 1.5.3 Research Aim

This study aimed to investigate options for improving water use productivity and understanding the drivers of arsenic accumulation in rice grain in Uruguay. Specific questions for these two areas were: 1. What irrigation management techniques will allow a reduction in water used without negatively affecting rice grain yield and quality? How much water can be saved using these alternative irrigation techniques? What is the increase in water productivity (WP) that alternative irrigation techniques could offer compared with continuous flooded irrigation systems?

2. What is the accumulation of inorganic arsenic in rice grain in different soil types in Uruguay? What is the impact on food safety and reduction on inorganic arsenic accumulation in rice by implementing mitigation management practice such as AWD irrigation techniques? Is there any difference in the accumulation of inorganic arsenic within the commonly planted varieties in Uruguay?

#### **1.6 Publications arising from this project**

#### 1.6.1 Description of all publications published and how are they linked

This research study aimed to address key questions in two published papers related to: irrigation management techniques effects on water productivity, grain quality and arsenic accumulation in rice grain.

In the first paper (Chapter 2), we aimed to identify irrigation management techniques that allow a reduction in water used without negatively affecting rice grain yield and quality. Additionally, in this study we aimed to quantify how much water can be saved using these techniques and what was the increase in water productivity compared with the traditional continuous flooded irrigation system. The hypothesis was that during the crop vegetative phase it would be possible to adjust the traditional management, reducing irrigation water used with no effects on grain yield, quality and increasing water productivity. The objective of this paper was to determine irrigation management practices and techniques that increase water productivity WP (kg m<sup>-3</sup>) allowing a reduction in water input without negatively affecting grain yield and quality.

The evaluated alternative irrigation techniques in our first study not only would contribute to improve water productivity but also would help to identify mitigation practices to reduce the accumulation of heavy metals in rice grain. For this reason, in the second paper (Chapter 3) we aimed to determine what is the accumulation of inorganic arsenic in rice grain in different soil types in Uruguay with some of the irrigation techniques (C and AWD) investigated in the first paper. The objective was to determine what was the impact on food safety and quantify the reduction of inorganic arsenic accumulation in rice by implementing different mitigation management practices in rice grain in two contrasting soil sites, commonly used for rice production in Uruguay. Additionally, we aimed to investigate if there were different responses with different varieties.

In summary, alternative irrigation techniques that would contribute to improve water productivity were addressed in paper 1 Chapter 2, while food safety issues specially the accumulation of heavy metals like arsenic in grain under different irrigation management for different varieties were published in paper 2 Chapter 3.

#### **1.6.2 List of Publications**

• Chapter 2 was published as follows:

Carracelas, G., Hornbuckle, J., Rosas, J., Roel, A., 2019. Irrigation management strategies to increase water productivity in Oryza sativa (rice) in Uruguay. Agric. Water Manag. 222, 161–172. <u>https://doi.org/10.1016/j.agwat.2019.05.049</u>

• Chapter 3 was published as:

Carracelas, G., Hornbuckle, J., Verger, M; Huertas, R., Riccetto, S., Campos, F and Roel, A. 2019. Irrigation management and variety effects on rice grain Arsenic levels in Uruguay. Journal of Agriculture and Food research. <u>https://doi.org/10.1016/j.jafr.</u> 2019.100008

#### **CHAPTER 2**

# 2. Published paper 1. Irrigation management strategies to increase water productivity in Oryza sativa (rice) in Uruguay

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**Keywords:** Alternate Wetting and Drying; Intermittent, Yield, Quality, Water productivity

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#### **2.1 Highlights**

This study identified irrigation techniques that used significantly less irrigation water while maintaining rice grain yield and therefore increasing water productivity, across a range of typical irrigated rice growing environments in Uruguay.

Intermittent irrigation techniques that maintained soil moisture above or near a saturated condition, allowed a reduction in irrigation water input without negatively affecting rice yield, leading to a significant increase in water productivity.

Alternate Wetting and Drying techniques allowed soil moisture to drop below saturation and yield was found to be affected negatively.

#### **2.2 Abstract**

Traditional rice irrigation systems in Uruguay are fully irrigated and early continuously flooded irrigation accounts for a high volume of water used. The purpose of this study was to determine irrigation techniques that increase water productivity (WP) allowing a reduction in water input without negatively affecting grain yield in Uruguay. Ten experiments were conducted over a six-year period from 2009 to 2015, in three experimental units located among the major rice growing regions. Treatments included: early continuous flooding (C), alternate wetting and drying (AWD), intermittent flooding until panicle initiation (IP) and intermittent flooding during all crop growth period (I). All treatments were planted on dry soil. In treatment C flooding started 15-20 days after emergence and a water layer of 10 cm above the soil surface was maintained throughout all the crop cycle. In treatments IP and I, the water level alternated between 10cm and 0cm and was re-established when the soil was still saturated. The AWD treatment allowed the soil to dry periodically (water depletion of 50% of soil available water) until panicle initiation. IP and I over three seasons led to significant savings in irrigation water inputs in the North and Central regions (averaged 35% or - 3986 m<sup>3</sup> ha<sup>-1</sup>) in relation to C. In the East region, AWD allowed for a 29%(-2067 m<sup>3</sup> ha<sup>-1</sup>) water saving in relation to the control over four seasons but determined a significant yield loss of 1339 kg rice ha<sup>-1</sup> (15% reduction) in relation to C. WP was increased by 0.25kg m<sup>-3</sup> (23%) in IP and 0.68 kg m<sup>-3</sup> (62%) in I, in relation to the control C. Whole grain percentage was significantly reduced with I in the North region only. Techniques that maintained the soil water at saturated conditions like intermittent flooding, allowed a reduction of water input with no significant effects on grain yield, which led to a significant increase in WP.

#### **2.3 Introduction**

Continuously flooded rice is the largest irrigated crop in the world with a higher water demand in relation to other cereal crops (Pimentel et al., 2004) and the major staple food crop with 54kg consumed per person annually (FAO, 2018). Increasing grain yields and maintaining grain quality while reducing water use, is a great challenge for the rice sector globally. Rising global food demand will increase water use requirements and competition for this resource that is becoming increasingly scarce in some parts of the world (Tuong and Bouman, 2003; Rijsberman, 2006; Mekonnen and Hoekstra, 2016). This can be attributed to competition from other sectors, environmental concerns, and climate change predictions, like increased occurrence of drought periods, aquifer over-extraction, loss of water quality by sewage, chemical pollution, and salinization (Meybeck et al., 1996; Bouman et al., 2007a, Siebert et al., 2010, Reba et al., 2013, Famiglietti, 2014). Climate change predictions by many models are indicating increases in temperature, more weather variability (Stocker et al., 2013) and higher frequency, duration and severity of water shortages (Spinoni et al., 2014) which would limit water availability for irrigation and rice production in the future (Peng et al., 2004; Lobell, 2007; Wassmann et al., 2009a, 2009b; Gaydon et al., 2010; Lyman et al., 2013).

Rice is also the largest irrigated and water consumer crop in Uruguay. Early continuous flooding is the main irrigation technique implemented by farmers to secure the highest yields to maximize profit. Rice is planted on dry soil conditions, flooded from 15-25 days after emergence when rice plants have 3-5 leaves (V3 -V5 according to Counce et al., 2000), and maintained with a water layer of 5-10 cm until 20 days before harvest. Rice grown in Uy requires from 8000 to 15000 m<sup>3</sup> ha<sup>-1</sup> of water (Battello et al., 2009; Böcking et al., 2008; Roel et al., 2011; Lavecchia et al., 2011; Riccetto et al., 2017). Several authors reported rice water requirements for growth within the range from 3550 to 7000 m<sup>3</sup> ha<sup>-1</sup> (Pringle, 1994; Tabbal et al., 2002; Bouman et al., 2007a; Massey et al., 2014). This information is in concordance with the data reported by Blanco et al., 1984, where only 45% or 6000 m<sup>3</sup> ha<sup>-1</sup> were evapotranspirated from the 13300 m<sup>3</sup> ha<sup>-1</sup> irrigation water input for continuous flooded rice in Uruguay. Those results are also aligned with the ones reported by Böcking et al. (2008), where evapotranspiration ranged from 5500 to 6780 m<sup>3</sup> ha<sup>-1</sup> in
three studies conducted in the North of Uruguay. The main benefits of flooding the rice crops are related to more effective weed control, an increase in nutrient availability, lower disease incidence, and thermal insulation/protection from cold during microsporogenesis (Williams and Angus, 1994; Dunn and Gaydon, 2011). In some countries like Australia, a deep layer of water (0.20-0.25m) is used during flowering to protect pollen from low temperature (Humphreys et al., 2006). Conversely, the application of deep-water layer during this critical period allowed no differences in temperature within the canopy in Uruguay (Roel, 2005). Some potential disadvantages of the traditional continuously flooded (C) technique are associated with higher arsenic (As) accumulation in rice grain (Linquist et al., 2015; Carrijo et al., 2017; Yang et al., 2017; Carrijo et al., 2018; Seyfferth et al., 2018), and higher Green House Gas (GHG) emissions (Linquist et al., 2015; Tarlera et al., 2016; Seyfferth et al., 2018) in relation to alternative irrigation techniques like alternate wetting and drying (AWD).

Uruguay has a subtropical to temperate climate with a great deal of secure water resources (river, streams, lagoon) and an average annual rainfall ranging from 1200 mm (Southwest) to 1600 mm (Northeast) (Castaño et al., 2011). Average rainfall during the rice growing season from October to March over a 17-year period (1988-2015) was 624 mm ranging from 301 - 934 mm per year (Carracelas et al., 2017b, GYGA website). Rainfall is not evenly distributed during the crop season and for this reason rice cannot be grown without the addition of irrigation water in this country, as grain yields are highly penalized resulting in non-harvestable yields. All rice cultivated in Uruguay is irrigated during most of the crop cycle. There is an opportunity to optimize rainfall captured by implementing alternative techniques like intermittent irrigation (Massey et al., 2014; de Avila et al., 2015). The importance of studying and continuing to develop irrigation techniques that use less water while preserving crop yields in Uruguay are also driven by the desire to reduce irrigation pumping costs and promote expansion of rice crop area. Most of the water used to irrigate rice is pumped (56%) in Uruguay (DIEA MGAP, 2017) and the cost of energy is a pressing issue for farmers. Lowering the irrigation cost to increase profit and maintaining enough water to irrigate adequately to secure crop yield potential is one of the main drivers for the implementation of water saving techniques by farmers in Uruguay.

Water is a limiting factor for the expansion of rice and other crops. Dams built for irrigation purposes are the main water source (54%) (DIEA MGAP, 2017). Increasing water use efficiency and building new dams would contribute to an increase in irrigated area. Additionally, if more water is available to irrigate other cereal crops and pastures, this would create an opportunity for land-owners to make more profit and reduce risk by diversification of their products. In drought years water stored in the reservoirs in Uruguay may not be enough to irrigate 100% of rice fields flooded during the entire growing season. New water management techniques have the potential to help farmers cope with water scarcity in dry years.

Worldwide, several water saving irrigation techniques have been implemented to reduce water input, reduce associated irrigation costs, or save water for other purposes (Bouman et al., 2007a) but they may have a negative impact on grain yield as rice is very susceptible to water stress (Tuong et al., 2005). Much of the research outputs have conflicting result in the impacts of alternative irrigation systems on grain yields. Rice yields can be reduced under non-saturated soil conditions (Bouman and Tuong, 2001, Tuong et al., 2005; Parent et al., 2010; Sudhir-Yadav et al., 2012), and this could be associated with the shallow rice root system (Parent et al., 2010) as well as other factors like diseases, weeds or nutrients. However, other studies reported a significant reduction in water input without affecting rice grain yield and therefore improving water productivity (Tabbal et al., 2002; Belder et al., 2004; Lampayan et al., 2005). It is imperative to research and find out the main factors affecting the success of these alternative irrigation techniques over a range of environmental, soil and management conditions specific to each country. Alternative irrigation techniques need to be locally adapted and developed to use less water and minimize off-site impacts while preserving grain yield and quality. Intermittent irrigation and safe alternate wetting and drying are a promising alternative irrigation technique, not only for reducing water input and to increase water productivity, but also to minimize water footprint, environmental impact, greenhouse gas emissions and food safety issues, especially the accumulation of heavy metals like Arsenic in grain (Linquist et al., 2015; Tarlera et al., 2016; Yang et al., 2017; Carrijo et al., 2017; Carrijo et al., 2018; Seyfferth et al., 2018).

This paper is an integrated analysis of different irrigation management practices in experiments conducted at different sites with different soil and slope situations representative of the three rice growing regions of Uruguay. The main objective of this research was to determine irrigation management practices and techniques that increase WP without negatively affecting grain yield. In addition, we were looking to identify an optimal irrigation management that could be implemented across all environments or if different techniques need to be developed for each region. This study tested the hypothesis that during the crop vegetative phase it is possible to adjust the traditional early irrigation flooding management, without affecting grain yield, reducing irrigation water input and consequently increasing water productivity (WPi and WPir). WPi can be defined as the kilograms of grain produced per m<sup>3</sup> of irrigation water inputs and WPir is rice yield over volume of water inputs by irrigation and rain (WPir) (kg m<sup>-3</sup>). Evapotranspirated water, was also reported in this work (Bouman et al., 2007a).

## 2.4 Methods

### 2.4.1 Study site description

The Uruguayan rice sector is divided in three regions: East (118391 ha), North (33 448 ha) and Central (12 618 ha) representing 72%, 20% and 8% of total annually rice planted area (DIEA MGAP, 2018) (Figure 2.1). There was one experimental unit per region: In the North (Lat:-30.50S, Long:-57.12W) experiments were conducted during the seasons: 2011/12 - 2013/14 - 2014/15; in the Central region (Lat:-32.18S, - 55.17W), experiments were conducted during the seasons: 2011/12 - 2013/14 - 2014/15; in the Seasons: 2011/12 - 2012/13 - 2013/14; in the East region (Lat:-33.27S, Long:-54.17W), the experiments were conducted throughout seasons 2009/10, 2010/11, 2011/12 and 2012/13 .

The 10 experiments were conducted in typical soil types of each region. Soil properties determined in a laboratory for the different field sites are presented in Table 2.1.



**Figure 2.1.** Location of the National Institute of Agricultural Research(INIA) rice field experimental sites, reference weather stations (INIA) and rice areas of Northern, Central and Eastern Uruguay (DIEA MGAP, 2018).

**Table 2.1.** Soil property descriptions for each experimental unit North, Central and East. Soil fertility and parameters information were determined in private and INIA soil laboratories. Soil texture information for the first horizon 0-30cms.

| Soil Parameter                     | Region             |                 |            |  |
|------------------------------------|--------------------|-----------------|------------|--|
|                                    | North              | Central         | East       |  |
| pH (water)                         | 6.7                | 5.4             | 5.9        |  |
| Organic Matter %                   | 3.4                | 1.1             | 2.1        |  |
| P - Bray 1                         | 1.3                | 5.3             | 3.8        |  |
| P Citric Acid (ppm)                | 12.3               |                 | 6.9        |  |
| K (meq/100g)                       | 0.29               | 0.13            | 0.18       |  |
| Texture                            |                    |                 |            |  |
| Sand %                             | 6                  | 17              | 30         |  |
| Silt %                             | 25                 | 60              | 43         |  |
| Clay %                             | 69                 | 23              | 28         |  |
|                                    |                    | *               |            |  |
| Soil                               | Vertisol           | Planosol        | Brunosol   |  |
| * Soil texture information for the | first horizon A (0 | - 30cms). SIGRA | S webpage. |  |

## 2.4.2 Field Management

Typical rotation in the experimental sites consisted in one year of rice followed by two to three years of perennial pastures (mixes of grasses and legumes). Minimum tillage was done in the previous summer, 6-9 months before the planting date. Land preparation consisted in one- or two-discs plowings to control weeds and incorporate previous crop (pasture) residue Additionally, 1 landplane was done and contour levees of 20-30 cms height were constructed. Tillage operations, sowing, pre, postemergence weed controls and first Nitrogen application was done on dry soils before permanent flooding.

The planting date was mainly in October in all sites (from late September up to early November) as dictated by local weather conditions. In the Northern region the crop was planted on 3/11, 25/9, 25/9, in the Central region planting date was on 19/10,

16/10, 1/10 and in Eastern region crop was planted on 1/10, 8/10, 22/10 and 19/10 for the different consecutive seasons evaluated. All treatments were dry seeded with a commercial direct drill machine of 13 line (Semeato) at all sites. Soil moisture content ranged from 35–46 mm / 10 cm; normally the crop is planted with soil moisture content around field capacity.

Indica type cultivars were planted at all sites. In the North and Central region, the cultivar planted was INIA Olimar. Seeding rate of this variety was 160 kg seed ha<sup>-1</sup>. In the East region INIA Olimar was planted in the first season (160 kg ha<sup>-1</sup>) and El Paso144 in the following seasons at 143 kg ha<sup>-1</sup>, as this variety was the main one planted in this region.

Fertilization management of the crop consisted of basal application of Nitrogen (16 -  $30 \text{ kg N ha}^{-1}$ ), Phosphorus ( $30 - 46 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$ ) and Potassium ( $18 - 99 \text{ kg K}_2\text{O ha}^{-1}$ ) plus two urea fertilization in coverage at tillering prior to the flood and panicle initiation ( $12.4 - 55 \text{ kg N ha}^{-1}$  each) based on soils fertility analyses results. In the central region 30 kg ZnSO4 ha<sup>-1</sup> was also applied in season 2018-2019. Herbicide applications to control weeds varied across seasons and regions according to their degree of incidence.

## 2.4.3 Field Crop and Water measurements

The main information collected at the experimental sites included the following variables:

<u>-Rice yields</u> (kg ha<sup>-1</sup>) at 14% moisture. The area harvested in the middle of plot was 6.1 m<sup>2</sup> in the East. In the North and Central region three samples of 5.1m<sup>2</sup> each (10 rows X 3metres) were harvested per plot and averaged. The rice samples were mechanically threshed. Grain yields were normalized to 14% moisture. Harvest was done manually when grain moisture was lower than 21% and average green percentage was lower than 8%, according to rice industry recommendations. Grain percentage was visually separated and weighted from a 50-gr sample and moisture

contents was determined using an electronic moisture tester (Steinlite) from a 100g sample in the laboratory.

<u>-Industrial Grain Quality</u>. Whole grain percentage was determined with the cylinder of "Trieurs" specific to each variety. This parameter is defined as the unbroken grains of rice and large broken grains whose length is equal or greater than <sup>3</sup>/<sub>4</sub> of the average length of whole grains. Total white percentage is an estimate of the amount of whole and broken grains that are produced in the milling of cargo rice to a degree of whiteness that ranges from 37 to 40 degrees. It was determined with a grinder and a white grade meter. Chalking percentage is estimated visually and includes the whole and broken rice grains that present an opaque aspect like chalk, in 50% or more of the grain. All parameters were determined in INIA and ACA (Rice Growers Association) Laboratories.

<u>-Water input</u> (WI) volume (m<sup>3</sup> ha<sup>-1</sup>) were measured in all regions with helicoidal flowmeters (ARAD, WMR in the East and DOROT / KAPA brand of 110 mm size in the North and Central). Flowmeters were installed at the entrance of each plot to allow independent management of each irrigation treatment. In the North and Central region irrigation was by gravity from a dam while in the east water input was pumped from the river.

Total water (WT) includes irrigation water input plus rainfall measured during the crop cycle.

<u>-Water Productivity (WP)</u> (kg m<sup>-3</sup>) is defined as kilograms of rice grain produced per unit of input water (Bouman et al., 2007a).

<u>Irrigation Water Productivity (WPi)</u> it was determined by the relationship between the rice yield at 14% of moisture (kg) and Irrigation Water Input (WI).

<u>Total Water Productivity (WPir)</u> was calculated considering rainfall + Irrigation Water Input (WT).

<u>Evapotranspiration Water productivity (WP<sub>ET)</sub></u>, was estimated as rice yield (14%) registered by irrigation treatment in each region, over cumulative weight of crop

evapotranspirated water (ETc). Crop Evapotranspiration (ETc) was calculated based on the equation:  $ETc = ETo \times Kc$ , using a crop coefficient average factor Kc=1.04, weighed by crop period (Kc initial: 1.05 for 0-55 days after emergence - DAE, Kc mid: 1.20 (55-95 DAE) and Kc end: 0.75 for late season growth stage (from 95 DAE) (Allen, 1998). An average crop cycle from emergence to harvest of 141 days was considered. Average number of days from emergence to flowering (50%) was 96 and harvest was done 45 days after flowering (Table 2.6). Potential Evapotranspiration (ETo) was obtained from locally modified Penman equation - FAO (Allen, 1998) adjusted for the conditions of Uruguay, available at: http://www.inia.org.uy/disciplinas/agroclima/penman.htm (Table 2.2, 2.3).

<u>-Moisture content in the soil</u> was determined in the AWD treatment in the East region. The methods used were gravimetric, with weekly measurements at a depth of 0-15 and 15-30 cm, and by capacitance probes FDR (Decagon Devices, EC-5) with continuous measurements, installed at a depth of 0-10 cm. The available water storage capacity for the East region soil was determined by the difference between the volumetric moisture at field capacity and the volumetric moisture at permanent wilting point. Both parameters were obtained from the tension-humidity curve obtained using the Richards method (Richards, 1948).

<u>-Flowering date</u> percentage was determined by visually counting the emerged panicles every second day in a monitored area of 1-meter length with three replications per plot. When 50% of total panicles were flowering, this date was recorded as flowering and used to estimate the number of days from rice emergence.

<u>-Weather parameters</u> were retrieved from INIA (National Institute for Agricultural Research) meteorological stations in the North (Salto Grande), East (Treinta y Tres) and Central (Tacuarembó) (GRAS available at: <u>www.inia.uy/gras/Clima/Banco-datos-agroclimatico</u>).

Daily weather parameters (average from 2009-2015) included: Solar Radiation (kJ m<sup>-</sup> <sup>2</sup> d<sup>-1</sup>), Minimum and Maximum Temperature (Tmin. °C, Tmax. °C), Vapour pressure (%), Rainfall (mm), Wind speed (ms<sup>-1</sup>) (Table 2.2). Quality control and filling/correction of weather data were performed based on NASA-POWER

(http://power.larc.nasa.gov/ as described in Grassini et al., 2015; Carracelas et al., 2017a, b). Information is also available at: GYGA - www.yieldgap.org. A rainfall gauge to determine rainfall was additionally installed in each experimental site. Effective precipitation - EP (mm) was calculated considering surface runoff water according to the precipitation index method and is available at <a href="http://www.inia.uy/gras/Monitoreo-Ambiental/Balance-H%C3%ADdrico/Calculo-Precipitacion-Efectiva">http://www.inia.uy/gras/Monitoreo-Ambiental/Balance-H%C3%ADdrico/Calculo-Precipitacion-Efectiva</a>.

Parameters like Evap. "Tank A", EP, ET0 and ETc were calculated for the seasons where the experiments were conducted for each region. (East: 2009/10, 2010/11, 2011/12; Central: 2011/12, 2012/13, 2013/14 and North: 2011/12, 2012/13 and 2014/15) (Table 2.2, 2.3).

**Table 2.2.** Description of the climate parameters across the regions registered in the nearest INIA meteorological stations from the experimental sites for the East Central and North region. Average of 6 seasons from Oct to March (2009 -2015).

| Parameters  | Region |            |                |  |
|---|--------|------------|----------------|--|
| i arameters   | North  | Central    | East           |  |
| Solar Radiation (kJ m <sup>-2</sup> d <sup>-1</sup> ) | 21968  | 21583      | 20190          |  |
| Minimum temperature (degrees Celsius)                 | 17     | 16         | 15             |  |
| Maximum temperature (degrees Celsius)                 | 29     | 27         | 27             |  |
| Vapour pressure (kPa)                                 | 2.2    | 2.1        | 2              |  |
| Wind speed (m s <sup>-1</sup> )                       | 1.9    | 2.1        | 2.4            |  |
| Total, Precipitation (mm)                             | 915    | 929        | 736            |  |
| Effective Precipitation EP (mm)                       | 661    | 706        | 540            |  |
| Evaporation "Tank A" (mm)                             | 939    | 817        | 889            |  |
| ET <sub>0</sub> Penman (mm)                           | 685    | 641        | 614            |  |
| Etc (mm)  | 712    | 665        | 639            |  |
| Weather station location INIA                         | Salto  | Tacuarembo | Treinta y Tres |  |
| Latitude (S)  | -31.3  | -31.7      | -33.2          |  |
| Longitude (W)   | -57.9  | -55.8      | -54.3          |  |

**Table 2.3.** Weather parameters registered by season in each reference weather station in the East, Central and North region. Effective precipitation (EP mm), Evaporation "Tank A" (E."Tank A"), Potential (ET<sub>0</sub>) and crop (ETc) evapotranspiration. Average number of days from emergence until harvest considered was 141.

| Region  |         | Parameters (mm) |                     |                           |     |
|---|---------|-----------------|---------------------|---------------------------|-----|
| Weather Station   | Season  | EP              | Evap.<br>''Tank A'' | ET <sub>0</sub><br>Penman | Etc |
|   | 2009-10 | 819             | 747                 | 562                       | 585 |
| E   | 2010/11 | 371             | 1048                | 631                       | 656 |
| <b>East</b> - INIA Treinta y Tres $(33.28, 54.3W)$      | 2011/12 | 446             | 894                 | 655                       | 681 |
| (-33.2 <b>3</b> , -34.3 W)                              | 2012/13 | 525             | 868                 | 610                       | 634 |
|   | Average | 540             | 889                 | 614                       | 639 |
|   | 2011/12 | 553             | 928                 | 657                       | 683 |
| <b>Central</b> –<br>INIA Tacuarembo<br>(-31.7S, -55.8W) | 2012/13 | 679             | 774                 | 639                       | 664 |
|   | 2013/14 | 887             | 748                 | 626                       | 651 |
|   | Average | 706             | 817                 | 641                       | 665 |
| <b>North -</b> INIA Salto<br>(-31.3S, -57.9W)           | 2011/12 | 580             | 1004                | 722                       | 751 |
|   | 2013/14 | 673             | 946                 | 661                       | 687 |
|   | 2014/15 | 731             | 866                 | 672                       | 699 |
|   | Average | 661             | 939                 | 685                       | 712 |

# 2.4.4 Treatments and experimental design

Three irrigation management practices were evaluated in each region (North Central and East). Continuous traditional flooding (C) that represents the most common rice flood management (control), Intermittent irrigation until panicle initiation (IP) and a third treatment that varied across the region to be able to impose higher water stress in plants: Intermittent during all cycle (I) in North/ Central regions and alternate wetting and drying (AWD) in the East (Figure 2.2).

In treatment C, flooding started 15-20 days after emergence and a water layer of 10 cm above the soil surface was maintained after flooding throughout all the crop cycle.

Irrigation water input filled out the bays within levees and replenish evapotranspirated water. In treatment IP and I the water layer alternated between 10cm and 0cm above the soil surface and was re-established when the soil was still saturated. The AWD treatment permitted the soil to dry periodically (allowing a water depletion of 50% of soil available water) until panicle initiation. The common treatments along the three regions were C and IP and the third treatment was I in North / Central and AWD in East region.

The experimental design in the East region was a complete randomized block design with four blocks. In the North and Central region, the experimental design was a split plot with 2 blocks. Main plots were the field layout (FL) while irrigation treatment was the split plot. Average size of the plots was: 695 m<sup>2</sup> (main plot) and 232 m<sup>2</sup> (split plot). Total experimental area was in average 6200 m<sup>2</sup>.



**Figure 2.2.** Summary of irrigation treatments tested in different regions of Uruguay. Traditional continuous flooding (C) and intermittent flooding until panicle initiation (IP) as common treatments across all regions, intermittent flopding during all crop cycle (I) in North and Central region and alternate wetting and drying (AWD) flood management tested only in East region.

### 2.4.5 Data analysis

A linear mixed effects model was used to fit each one of the response variables (Irrigation Water input, Total Water input, Rice Yield, Water Productivity (WPi, WPir, WP<sub>ET</sub>), and Grain Quality parameters) for all the experiments, with Irrigation, Region, Block and Irrigation\*Region interaction as fixed effects, and Year and Irrigation\*Year interaction as random effects. An Analysis of variance was then performed followed by means separation using the Tukey test. The analyses were performed using the packages lme4 (Bates et al., 2015) and emmeans (Lenth, 2018) in R software (R Core Team, 2018). Following the significance of statistical analyses outputs, the irrigation information was not presented by Season. The tested interaction irrigation\*season was non-significant for most parameters evaluated: Grain Yield, Irrigation water input, Total water input, Water Productivity (WPi, WPir, WP<sub>ET</sub>) and Chalkiness. The same criteria were applied for the irrigation by region interaction.

## **2.5 Results**

## 2.5.1 Irrigation Water used and Total Water input

Traditional continuous flooding irrigation resulted in the highest water input in all regions. A significant interaction between region and irrigation treatments was detected (P<0.05). In the North region, WI savings, relative to control treatment of 28% (4133 m3 ha-1) and 42% (6217 m<sup>3</sup> ha<sup>-1</sup>) was determined for IP and I, respectively. In the Central Region, intermittent irrigation treatments allowed a significant WI saving in average of 34% (2798m<sup>3</sup> ha<sup>-1</sup>) in relation to C. In the East region, AWD determined a significant WI reduction, of 29% (2067 m<sup>3</sup> ha<sup>-1</sup>) in relation to C. A non-significant water use reduction WI of 14% (1016 m<sup>3</sup> ha<sup>-1</sup>) was measured in the IP treatment in relation to C for this region (Table 2.4, Figure 2.3).

|   | Water Input (m <sup>3</sup> ha <sup>-1</sup> ) |                               |  |  |  |
|---|--|-------------------------------|--|--|--|
| Treatments  | Irrigation<br>(WI)                             | Irrigation +<br>Rainfall (WT) |  |  |  |
| Irrigation *Region  |  |                               |  |  |  |
| East  |  |                               |  |  |  |
| 1. Continuous (C)   | 7101 a   | 12594 a                       |  |  |  |
| 2. Intermittent until panicle initiation (IP)   | 6085 ab  | 11870 ab                      |  |  |  |
| 4. Alternate Wetting and Drying (AWD)   | 5034 b   | 10840 b                       |  |  |  |
| Central   |  |                               |  |  |  |
| 1.Continuous (C)  | 8187 a   | 16087 a                       |  |  |  |
| 2. Intermittent until panicle initiation (IP)   | 5847 b   | 13747 b                       |  |  |  |
| 3. Intermittent during all crop cycle (I)   | 4932 b   | 12832 b                       |  |  |  |
| North   |  |                               |  |  |  |
| 1.Continuous (C)  | 14711 a  | 21428 a                       |  |  |  |
| 2. Intermittent until panicle initiation (IP)   | 10578 b  | 17295 b                       |  |  |  |
| 3. Intermittent during all crop cycle (I)   | 8494 c   | 15210 c                       |  |  |  |
| Average   | 7886   | 14656                         |  |  |  |
| CV%   | 13.42  | 4.96                          |  |  |  |
| P<0.05  | ***  | ***                           |  |  |  |
| Irrigation * Season - P<0.05  | NS   | NS                            |  |  |  |
| Means followed by different letters are significantly different with a probability less than 5% (P < 0.05). Signif. codes: '***' 0.001 '**' 0.01 '*' 0.05; NS: non-significant differences. CV: |  |                               |  |  |  |

**Table 2.4.** Average Irrigation Water Input (WI) and Total average Water Input (WT= Irrigation plus Rainfall)  $m^3 ha^{-1}$  for different irrigation systems and rice regions in Uruguay.

0.05). Signif. codes: \*\*\*\* 0.001 \*\*\* 0.01 \*\* 0.05; NS: non-significant differences. CV: coefficient of variation.

Total average water savings, WT (irrigation + rainfall) for IP and I treatments, relative to the control treatment of 24% and 17% were recorded for the North and Central region, respectively. In the East region, WT savings of 14% were measure in AWD in relation to C. A non-significant interaction was registered between Irrigation and Season for water input (P<0.05) (Table 2.4),



**Figure 2.3.** Irrigation water input (WI: m<sup>3</sup> ha<sup>-1</sup>) for different treatments and rice regions in Uruguay. Black dot represents least-square means, grey bars are indicating standard errors. Different letters are significantly different with a probability less than 5%.

# 2.5.2 Rice Yield and Water Productivity

There was no significant interaction between irrigation and region for yield and WP (P<0.05). Rice Yield in the East was 21 % higher (1716 kg ha <sup>-1</sup>) than the average yield recorded for the North and Central Region (Table 2.5).

There were no significant differences in rice grain yield between continuous flooded and intermittent (I, IP) irrigation treatments. The AWD treatment resulted in a significant yield reduction of 1339 kg rice ha<sup>-1</sup> (14.6% reduction) in relation to C (Table 2.5, Figure 2.4, 2.5).

**Table 2.5.** Rice grain yield (kg ha<sup>-1</sup>, 14% moisture) and Water Productivity, kg rice grain per m<sup>3</sup> of water (kg m<sup>-3</sup>) considering only irrigation water input (WPi) and total water (WPir) irrigation + rainfall) during the crop cycle, by irrigation treatments and regions.

| The state of the  | Rice Yield             | Water Productivity (WP)<br>kg m <sup>-3</sup> |                                   |  |
|---|------------------------|---|-----------------------------------|--|
| I reatments   | (kg ha <sup>-1</sup> ) | WPi-<br>Irrigation                            | WPir-<br>Irrigation +<br>Rainfall |  |
| Irrigation  |                        |   |                                   |  |
| 1.Continuous (C)  | 9194 a                 | 1.09 c  | 0.59 b                            |  |
| 2. Intermittent until panicle initiation (IP)   | 8755 a                 | 1.34 b  | 0.64 ab                           |  |
| 3. Intermittent during all crop cycle (I)   | 8710 ab                | 1.77 a  | 0.71 a                            |  |
| 4. Alternate Wetting and Drying (AWD)   | 7855 b                 | 1.37 abc                                      | 0.62 ab                           |  |
| Average   | 8628                   | 1.39  | 0.64                              |  |
| CV%   | 3.75                   | 14.49   | 5.75                              |  |
| P<0.05  | ***                    | ***   | ***                               |  |
| Region  |                        |   |                                   |  |
| I. Central - Ce   | 7628 a                 | 1.49 a  | 0.55 b                            |  |
| II. North - N   | 8485 a                 | 0.88 b  | 0.48 b                            |  |
| III.East - E  | 9772 b                 | 1.81 a  | 0.89 a                            |  |
| Average   | 8628                   | 1.35  | 0.62                              |  |
| CV%   | 4.3                    | 15.71   | 6.34                              |  |
| P<0.05  | ***                    | ***   | ***                               |  |
| Irrigation*Region P<0.05  | NS                     | NS  | NS                                |  |
| Irrigation*Season - P<0.05  | NS                     | NS  | NS                                |  |
| Means followed by different letters are significantly different with a probability less than 5% ( $P < 0.05$ ). Signif. codes: '***' 0.001 '**' 0.01 '*' 0.05; NS: non-significant differences. CV: coefficient of variation. |                        |   |                                   |  |



**Figure 2.4.** Rice grain yields, (kg ha<sup>-1</sup>, 14 % moisture) produced by different irrigation treatments (A) and Uy rice growing regions (B). Black dot represents least-square means, grey bars are standard errors, red arrow lines indicates confidence interval by Tukey. Different letters are significantly different with a probability less than 5%.



**Figure 2.5.** Rice grain yields, (kg ha<sup>-1</sup>, 14 % moisture) by irrigation treatments for each region: Central, East and North. Black dot represents least-square means, grey bars are indicating standard errors. Different letters are significantly different with a probability less than 5%.

Average water productivity (WPi) levels ranged from 1.09 recorded in the traditional control C to 1.77 kg m<sup>-3</sup> in the intermittent irrigation treatment I. Total water productivity (WPir) was on average 0.64 kg m<sup>-3</sup> (rainfall + irrigation) with no differences within IP, AWD and C treatments. The I treatments resulted in a significantly higher WPi and WPir in relation to the traditional control C treatment (Table 2.5, Figure 2.6).



**Figure 2.6.** Water productivity (WPi =kg m<sup>-3</sup>) considering only irrigation water used by irrigation treatment (A) and by region (B). Black dot represents least-square means, grey bars are indicating standard errors, red arrow lines indicates confidence intervals by Tukey. Different letters are significantly different with a probability less than 5%.

The highest WPi (kg m<sup>-3</sup>) was obtained with intermittent irrigation during all the crop cycle (I) in all regions 1.77 kg m<sup>-3</sup> (Figure 2.6, 2.7). Intermittent irrigation determined a significant increase in WPi in relation to the control continuous flooded treatment of 62% and 23% for I and IP, respectively. AWD determined a non-significant increase of 25% in WPi in relation to C.



**Figure 2.7.** Water productivity (kg m<sup>-3</sup>) considering only irrigation water input (WPi) by irrigation treatments for each region: Central, East and North. Black dot represents least-square means, grey bars are indicating standard errors. Different letters are significantly different with a probability less than 5%.

Average water productivity WPi and WPir in the East region was 1.81 and 0.89 kg m<sup>-3</sup> respectively. The lowest values of those parameters were observed in the North 0.88 and 0.48 kg m<sup>-3</sup> for WPi and WPir in that order. The Central region registered values of WPi 1.49 and WPir 0.55 kg m<sup>-3</sup>, 69 % and 15 % higher respectively compared to the North region.

Evapotranspiration water productivity (WP<sub>ET</sub>) was 1.37 kg m<sup>-3</sup> for C and 1.31 kg m<sup>-3</sup> for IP and I with no significant differences within treatments but it was significantly reduced to 1.15 kg m<sup>-3</sup> when AWD technique was implemented (Figure 2.8). Significant differences were also registered of WP<sub>ET</sub> by region. The highest WP<sub>ET</sub> was estimated for the East region (1.55 kg m<sup>-3</sup>) and no differences were registered between the North and Central regions with an average value of 1.16 kg m<sup>-3</sup>. The analyzed interactions (irrigation\*region and irrigation\*season) were not significantly different also for WP<sub>ET</sub>, like the results obtained for WPi and WPir (P<0.05).



**Figure 2.8.** Water productivity (WP<sub>ET</sub>) as a function of evapotranspirated water by irrigation treatment (A) and by region (B). Black dot represents least-square means, grey bars are indicating standard errors, red arrow lines indicates confidence intervals by Tukey. Different letters are significantly different with a probability less than 5%.

# 2.5.3 Grain Quality

Implementing alternative water-saving irrigation techniques did not influence grain quality parameters such as white grain and chalkiness percentages, for all regions. In addition, whole grain percentage was not affected negatively in the East and Central regions (Table 2.6). **Table 2.6.** Industrial quality parameters percentage and number of days from emergence to flowering (50%) for different irrigation techniques and rice regions in Uruguay.

|   | Industrial Quality % |                |            | Flowering                   |
|---|----------------------|----------------|------------|-----------------------------|
| Treatments                                    | White<br>Grain       | Whole<br>Grain | Chalkiness | from<br>emergence<br>(days) |
| Irrigation *Region                            |                      |                |            |                             |
| East  |                      |                |            |                             |
| 1. Continuous (C)                             | 70.4 a               | 58.3 a         | 4.2 a      | 87 a                        |
| 2. Intermittent until panicle initiation (IP) | 70.7 a               | 62.1 a         | 4.5 a      | 94 b                        |
| 4. Alternate Wetting and Drying (AWD)         | 70.8 a               | 59.6 a         | 3.9 a      | 96 b                        |
| Central                                       |                      |                |            |                             |
| 1.Continuous (C)                              | 69.1 a               | 67.9 a         | 5.9 a      | 97 a                        |
| 2. Intermittent until panicle initiation (IP) | 68.8 a               | 66.0 a         | 5.1 a      | 97 a                        |
| 3. Intermittent during all crop cycle (I)     | 68.8 a               | 66.3 a         | 6.0 a      | 97 a                        |
| North   |                      |                |            |                             |
| 1.Continuous (C)                              | 68.3 a               | 62.2 a         | 1.2 a      | 98 a                        |
| 2. Intermittent until panicle initiation (IP) | 68.2 a               | 59.5 ab        | 1.1 a      | 100 a                       |
| 3. Intermittent during all crop cycle (I)     | 68.0 a               | 56.5 b         | 2.7 a      | 100 a                       |
| Average                                       | 69                   | 62             | 3.9        | 96                          |
| CV%   | 1                    | 4              | 45         | 3                           |
| P<0.05  | NS                   | ***            | NS         | ***                         |
| Irrigation*Season -P <0 .05                   | **                   | *              | NS         | ***                         |

Means followed by different letters are significantly different with a probability less than 5% (P < 0.05). Signif. codes: '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05; NS: non-significant differences. CV: coefficient of variation.

Both IP and AWD treatments, delayed flowering by around 1 week in the East region. There was no significant effect on number of days to flowering after emergence by implementing alternative irrigation techniques in the North and Central region (Table 2.6).

Intermittent irrigation (I), led to a significant reduction in whole grain percentage of 5.7% in relation to C only in the North region (Figure 2.9).



**Figure 2.9.** Whole grain percentage for different irrigation treatments and rice regions in Uruguay. Black dot represents least-square means, grey bars indicate standard errors. Different letters are significantly different with a probability less than 5%.

## 2.6 Discussion

#### 2.6.1 Irrigation management effects on Water Input

Traditional continuous flooding has been the main irrigation technique for rice implemented by farmers in Uruguay. Irrigation water input measured under continuous flooded conditions averaged 10000 m<sup>3</sup> ha<sup>-1</sup> (ranging from 7000 to 15000 m<sup>3</sup> ha<sup>-1</sup>). Total water input averaged 16700 m<sup>3</sup> ha<sup>-1</sup> when rainfall was included, ranging from 12600 to 21400. The big differences measured between regions are associated with the soil characteristics (texture, organic matter) (Table 2.1) and land gradients. Rice in the East and Central region is cultivated on lower percolation and lower infiltration rate soils (planosols) compared to the North areas (vertisols). Slopes in the North region are also higher and field layout techniques in this region are different with lower height and closer contour levees in comparison to the Central and East region. In this region higher runoff water losses normally occur to maintain the crop being continuously flooded. This information is aligned with the irrigation water use reported in continuous traditional irrigation by other authors not only in Uruguay but also around the world: net water input (irrigation water plus rainfall minus surface drainage) of 15000-15600 m<sup>3</sup> ha<sup>-1</sup> in Australia (Dunn and Gaydon, 2011), irrigation water applied from 13140 to 24050 m<sup>3</sup> ha<sup>-1</sup> (Linquist et al., 2015), field measured applied irrigation averaged 8720 ranging from 2440 to 18800 m<sup>3</sup> ha<sup>-1</sup> in USA (Massey et al., 2018), total water input (rain plus irrigation) in field experiments and farmer fields ranged from 6500 to 15250 m<sup>3</sup> ha<sup>-1</sup> in China and from 5770 to 35004 m<sup>3</sup> ha<sup>-1</sup> in Philippines (Bouman et al., 2007a), total water input including rainfall measured in experiments ranged from  $11710 - 14300 \text{ m}^3 \text{ ha}^{-1}$  in Brazil (de Avila et al., 2015). Rice receives more irrigation water than is needed according to crop evapotranspiration requirements using traditional irrigation methods. The estimated average crop evapotranspiration from emergence until crop harvest (ETc) in this study was 6720 m<sup>3</sup> ha<sup>-1</sup> with some differences within regions. The highest value was registered in the North (7120 m<sup>3</sup> ha<sup>-1</sup>) followed by the Central (6650 m<sup>3</sup> ha<sup>-1</sup>) and East region (6390 m<sup>3</sup> ha<sup>-1</sup>) (Table 2.3). It was found in validation experiments adapting alternative irrigation techniques on commercial farms on clay soils that 6000 m<sup>3</sup> ha<sup>-1</sup> of irrigation water input for rice is an achievable target with no yield or quality penalties (Massey et al., 2014).

Alternative irrigation techniques tested in this paper determined water use savings, in all regions evaluated. In the North and Central, the intermittent irrigation IP and I determined a significant input water saving of 28% (3237 m<sup>3</sup> ha<sup>-1</sup>) and 41% (4736 m<sup>3</sup> ha<sup>-1</sup>) on average in relation to the control continuous flooded respectively. There is a chance to optimize rainfall capture and reduce irrigation inputs by implementing alternative irrigation management practices. In the East water input saved was lower in relation to the other regions by implementing intermittent irrigation IP (14%, 1016  $m^{3}ha^{-1}$ ) and even under the more stressed AWD treatment (29%, 2067  $m^{3}ha^{-1}$ ). The lower rainfall received during the crop cycle in the East in relation to the other regions (Table 2.2), determined a lower opportunity to optimize rainfall capture by the implementation of alternative irrigation techniques. Average rainfall in our studies from Oct to March was 574 mm, 670 mm and 795 mm for East, North and Central regions, respectively. Average rainfall of the three regions (680 mm) was 9 % higher than the historical average over a 17-year period (624 mm) (GYGA website, Carracelas, et al., 2017b). It has been reported by many authors in several studies an increase in rainfall capture by implementing intermittent irrigation techniques and a reduction in irrigation water inputs (Massey et al., 2014; de Avila et al., 2015; Massey et al., 2018). The main reasons of reduced water inputs identified by other authors

were also associated with a reduction in percolation (Sudhir-Yadav et al., 2012) and lower floodwater runoff losses (Bouman et al., 2007b). Reported average water savings of 12 % and 18% during two consecutive years, were recorded by Dunn and Gaydon, (2011). Results from this paper agree with international work and show that intermittent irrigation management has a significant potential to increase WP across Uruguay.

# 2.6.2 Irrigation management effects on Grain Yield, Quality and Water productivity

The implementation of intermittent irrigation until panicle initiation = IP technique (common experimental treatment along the three regions) confirmed that it is possible to reduce water use during the vegetative non-critical period without reducing significantly the rice grain yield and not affecting grain quality, therefore, increasing WP. An average irrigation water saving of 25% (approximately 2500 m<sup>3</sup> ha<sup>-1</sup>) and a WP increase of 23% from 1.09 to 1.34 kg m<sup>-3</sup> (0.25 kg of grain increase per m<sup>3</sup> of water) were achieved by implementing the IP irrigation technique in comparison to the traditional continuous flooded practice. A non-significant yield loss of 4.8% were registered in the IP in comparison with C. This result is aligned with information obtained around the world where it was found in several experiments a water saving of 23% on average (5 - 50% range), without significantly reducing grain yield by comparing intermittent saturated soil conditions treatments with continuously flooded (C) (Heenan and Thompson; 1984, Borrell et al., 1997; Bouman and Tuong, 2001; Tabbal et al., 2002).

Results reported in our experiments with the AWD treatment tested (allowed a 50% depletion of available water) indicated a yield loss of 15% in relation to C as soil was allowed to dry down. In this paper we confirmed that rice yield can be reduced when soil moisture was below saturation as it was found and reported by other authors (Bouman and Tuong, 2001; Tuong et al., 2005; Parent et al., 2010; Sudhir-Yadav et al., 2012). Carrijo et al., 2017 also found that yield was reduced by 23 % in AWD treatments compared to C when soil water potential was lower than -20 kpa. However, there is a high degree of variation in rice yield response to AWD depending on timing,

duration and severity during the drying event of this technique. Some studies reported a reduction in water input by 15–30% without a significant impact on yield (Tabbal et al., 2002; Belder et al., 2004; Lampayan et al., 2005), which were associated with a lower level of stress imposed to rice plants and local climatic conditions, soil properties (pH, OM, texture) and field slopes In some situations of shallow ground water depths (0.10-0.40m) roots can still have access to water even during drying periods in AWD, like what happens in intermittent irrigation where the soil is always kept saturated. In safe AWD recommendations, soil water depth reaches no more than 0.15m below the surface and the field is reflooded with the aim to minimize yield penalties to a standing 0.05m water depth (Lampayan et al., 2009; Lampayan et al., 2015). Sudhir-Yadav, et al. (2011a, b), reported an optimum irrigation soil tension of -20kPa at 0.20m for AWD to reduce irrigation water input without affecting grain yields and therefore improving WPi and WPir. Other authors also found no yield penalty when soil water potential was higher than -20kPa (Carrijo et al., 2017; Yang et al., 2017) or roots were able to provide total transpiration water demand from deeper soil layers (Carrijo et al., 2018).

Industrial grain quality parameters like whole grain, total white, chalkiness and green (%) are important in Uruguay as poorer grain quality results in reduced paddy price. When irrigating intermittently during all the irrigation cycle (I), whole grain was affected negatively in the North region only. Land surface gradients and infiltration rate in these soils are higher, which makes the successful implementation of this alternative irrigation technique more difficult to maintain soil moisture levels always under saturated conditions uniformly. Additionally, temperature and solar radiation are higher in the North compared to the Central and East region (Table 2.2), which could increase the stress level and the risk of negatively affecting whole grain percentage. This could be attributed to a higher sensitivity of this parameter to higher levels of water stress imposed to plants during the grain filling period. This parameter fell below the limit of 58% threshold set by the milling industry and would induce a payment penalty.

Input irrigation water productivity (WPi) was on average 1.39 kg m<sup>-3</sup> and total WPir was 0.64 kg m<sup>-3</sup> averaged across all regions. Intermittent irrigation implemented during the entire crop cycle (I) resulted in the highest values of those parameters 1.77

kg m<sup>-3</sup> and 0.71 kg m<sup>-3</sup> for WPi and WPir, respectively, compared to 1.09 and 0.59 kg m<sup>-3</sup> in the control. Water productivity values reported in this study are very good compared with ranges reported internationally: 0.2 -0.4 kg m<sup>-3</sup> in India with continuous flooded, 0.3-1.1 kg m<sup>-3</sup> in Philippines, (Bouman and Tuong, 2001, Sudhir-Yadav et al., 2012). WP considering total water input equals 0.4 kg m<sup>-3</sup> (ranging from 0.2 to 1.2) (Bouman et al., 2007a). There are several definitions of WP as it was pointed out by Bouman et al., 2007a, which denotes the amount kg rice grain (yield) over volume of water used. Water productivity can be defined as the values reported in this paper that consider the rice yield over volume of water inputs by irrigation (WPi) and rice yield over volume of water inputs by irrigation and rainfall (WPir). This information is valuable for irrigation engineers, managers and farmers that are interested in optimizing the productivity of irrigation water and total water resources rainfall and irrigation water, and also for regional water resource planners that could be interested in the amount of grain/food that can be produced with total water resources (Bouman, et al., 2007a). On the other hand, rice breeders are interested in the productivity of the amount of transpired water (WPt) or evapotranspiration (WPET), for selecting more water efficient cultivars. Bouman et al., 2007b and Sudhir-Yadav et al., 2012 reported WP<sub>ET</sub> average values that ranged from 1.0 to 1.5 kg  $m^{\text{-3}}$ using the simulation crop model Oryza with no significant differences within several irrigation water tension threshold. This information is aligned with estimated average WP<sub>ET</sub> values determined in this work, 1.15, 1.16 and 1.55 kg m-<sup>3</sup> for the North, Central and East regions, respectively. Additionally, no significant differences were registered for C IP and I irrigation treatments with an average WP<sub>ET</sub> of 1.33 kg m-<sup>3</sup>. However, AWD determined a significant WPET reduction of 13% which was mainly explained by the significant reduction of grain yield when this irrigation technique was implemented.

This study helped to identify irrigation techniques that use significantly less water while maintaining rice grain yield and therefore increasing WP, across a range of typical irrigated rice growing regions in Uruguay.

More research is needed in AWD and validation studies before promoting wide-scale adoption of this alternative technique. Further research is also required to evaluate ranges of "safe" alternate wetting and drying management strategies that maintain soil water depletion in a range that does not reduce rice grain yields. Intermittent irrigation until panicle initiation is the most promising irrigation technique to save water without penalizing grain yields and quality across Uruguay. If the 25% water saved by implementing the IP technique is used to promote the expansion of rice crops in Uruguay, an additional 32000 ha of rice could be annually cultivated, equivalent to 0.26 Mt of total rice production over the already 1.4 Mt available for trade would be possible. Widespread adoption of intermittent irrigation techniques could have the potential to expand rice crop area and significantly increase total rice production in Uruguay. However, results were obtained on experimental plots where irrigation is easy to manage. Under commercial conditions the implementation of intermittent irrigation would be more challenging associated with scalability and agronomic concerns such as weeds and nutrients. There is a risk of losing yield, quality and total income by implementing alternative irrigation techniques on larger scales. Therefore, the implementation of this technology will be limited unless an economic incentive is applied for farmers to use water more efficiently as has been reported in other studies worldwide (Bouman et al., 2007a; Linquist et al., 2015). In the current scenario of increasing production costs, low grain prices and lacking economic incentives to adopt water saving techniques, continuous flooding from 15-20 days after emergence is likely to remain the standard adopted and recommended practice in Uruguay, unless policy incentives are put in place. IP is a potential successful viable irrigation alternative to be validated across Uruguay while AWD would need more research before wide-scale adoption.

## **2.7 Conclusions**

Alternative irrigation techniques like intermittent irrigation in North, Central and alternate wetting and drying (AWD) in the East region allowed a significant water use saving of 5175 (35%), 2798 (34%), and 2067 m<sup>3</sup> ha<sup>-1</sup> (29%), respectively, compared to the early continuous flooded systems. Average irrigation water input was 7900 m<sup>3</sup> ha<sup>-1</sup> and total irrigation water input plus rainfall was 14700 m<sup>3</sup> ha<sup>-1</sup> in the continuous flooded treatment.

Rice yield was not negatively affected when intermittent irrigation techniques were implemented, and soils were maintained above saturation. Alternating wetting and drying conditions with 50% of soil available water depletion determined a yield loss of 1339 kgs (15%) in relation to the traditional continuous flooded treatment.

Average water productivity for all treatments considering only irrigation water (WPi) and total with rainfall (WPir) was 1.39 kg m<sup>-3</sup> and 0.64 kg m<sup>-3</sup>, respectively. Water productivity was significantly increased with the implementation of intermittent irrigation techniques by 0.25 kg m<sup>-3</sup> (from 1.09 to 1.34) with IP until panicle initiation and by 0.68 kg m<sup>-3</sup> (from 1.09 to 1.77) with I during all irrigation period in relation to the continuous flooded treatment. -Evapotranspiration WP<sub>ET</sub> was not affected by the implementation of intermittent irrigation (IP, I), in relation to the continuous flooded control C (average WP<sub>ET</sub> = 1.33 kg m<sup>-3</sup>). AWD determined a significant reduction of 0.20 kg m<sup>-3</sup> in WP<sub>ET</sub> in relation to C.

Industrial quality (white grain % and chalkiness %), was not affected negatively by implementing alternative irrigation technics in all regions. However, intermittent irrigation during the entire crop cycle, reduced whole-grain percentage in the North.

Intermittent irrigation until panicle initiation (IP) was shown in this study to be a technology that allowed a significant increase in water productivity without negatively affecting rice grain yield, with no effect on grain industrial quality and a significant reduction in irrigation water used in experimental conditions across all regions.

Further research should look to validate and adapt these technologies on commercial fields.

# **CHAPTER 3**

# **3.** Published paper **2.** Irrigation management and variety effects on rice grain Arsenic levels in Uruguay.

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# **3.1 Highlights**

Inorganic Arsenic levels (iAs) in Uruguay (average 0.07 mg kg<sup>-1</sup>) were found to be well below the limit proposed by the international standards of 0.20 mg.kg<sup>-1</sup>.

Alternate Wetting and Drying (AWD) irrigation technique resulted in lower levels of iAs accumulated in rice grain at one of the evaluated experimental sites in Uruguay.

Rice variety was found to significantly affect iAs uptake and accumulation in rice grain.

*Japonica* varieties were found to accumulate lower amounts of iAs in grain in relation to *Indicas*.

### **3.2 Abstract**

Rice is the most important staple component of the human diet worldwide. The higher amounts of arsenic accumulation in its grain in relation to other crops, determines a potential toxicity risk to humans. This research project aimed to determine the inorganic arsenic accumulation in rice grain (iAs) in two contrasting soil sites, Paso Farias-Artigas (PF) and Paso de la Laguna-Treinta y Tres (PdL), with two different mitigation practices, in Uruguay. These being firstly irrigation management techniques and secondly the use of different varieties. Five experiments were conducted with a split plot design with four blocks over three rice growing seasons from 2014 until 2017. The experimental sites included two irrigation treatments: continuous flooded (C) and alternate wetting and drying (AWD). The split plots included different varieties: *Indicas* and *Japonicas*. Average iAs accumulated in rice grain were 0.07 mg kg<sup>-1</sup>, well below international limits, even under the C irrigation technique. It was found that iAs accumulation in rice grain can be further reduced by the implementation of AWD in certain soil types. *Japonica* varieties had a lower accumulation of iAs in rice grain, in comparison with *Indicas* at both sites.

# **3.3 Introduction**

Growing demand for food around the world is expected to expand rice production by 1.1 percent to almost 510 million tons in 2018/19 (FAO, 2018). Rice is the most important staple component of the human diet worldwide with an average consumption of 54 kg of grain per person per year (FAO, 2018). Arsenic content in rice presents a risk to human health; it has been classified as a carcinogen class 1 and its toxicity depends on its chemical form. Different species of As are grouped into organic and inorganic and both constitutes the "total arsenic" content. The inorganic forms arsenite As<sup>III</sup> and arsenate As<sup>V</sup>, being more toxic for human health than the organic forms, such as monomethylarsonate (MMA) and dimethylarsinate (DMA) (Smith et al., 2002; Befani et al. 2017). The major component species of total arsenic in rice grain is inorganic arsenic ((As<sup>III</sup> and As<sup>V</sup>)), which are associated with negative health impacts like cancers (Meharg et al., 2009), hypertension, diabetes, and premature births (WHO, 2011). Arsenic levels in food are concerning as they are

frequently associated with high risk factors in food nutritional safety (Al-Saleh and Abduljabbar, 2017; Mitra et al., 2017).

Rice has naturally higher levels of As (Williams et al., 2007) as plants have a greater ability to absorb and accumulate it in the grain in relation to other staple food crops (Das et al., 2004). Arsenic (As) absorption by rice plants occurs through different transporters depending on arsenic speciation. As<sup>V</sup> uptake occurs mainly through phosphate transporters, while As<sup>III</sup> and methylated forms of As uptake occurs through non-specific aquaporins, mainly responsible of silicic acid uptake (Awasthi et al., 2017). Soil characteristics are very important to determine As content and its availability for plants, but As availability also depends on: pH, redox potential, organic matter content, cation exchange capacity, and concentration of iron oxides (Romero-Freire et al., 2014). When redox potential reaches high levels (200 -500mV), the predominant arsenic specie is AsV which has lower water solubility and, thus, generally reduced bioavailability. Solubility rises when an alkaline pH or high reductive conditions promotes the reduction of As<sup>V</sup> into As<sup>III</sup>. In an intermediate condition when redox potential is between 0 - 100 mV, Arsenic solubility depends on dissolution of iron oxides. At high redox potential,  $Fe^{+2}$  is oxidized to  $Fe^{+3}$ , precipitating as iron oxides or hydroxides, forming an iron plaque. (Masscheleyn et al., 1991). The iron plaque acts to adsorb As and reduces the absorption of As by plants (Tripathi et al., 2014). Organic matter also can reduce the mobilization of As in soils. In India, composted municipal waste successfully reduced native soil As mobilization in the rhizosphere by acting as a binding mediator (Bhattacharyya et al., 2003).

Arsenic is a natural component in primary minerals, therefore it is also found naturally in soils. The As concentration in uncontaminated soils of the world varies from 5 - 10mg kg<sup>-1</sup> (Han et al., 2003; Hossain et al., 2008). When this chemical element is partitioned into the aqueous soil phase rather than the solid phase, it has the potential to be uptaken by plants and can be a problem from a health perspective (Fendorf and Kocar, 2009). The levels of As and their forms in rice grain have previously been found to be affected by irrigation, varieties, fertilization and natural presence in air, soils and waters (Meharg and Zhao, 2012; Linquist et al., 2015; Mitra et al., 2017). Traditional (i.e., continuous) rice flood management can increase the bioavailability and absorption of As by plants. Under anaerobic soil conditions, arsenate is reduced to arsenite which is much more mobile in solution and more easily absorbed by rice roots (Williams et al., 2007). Additionally, many bacteria are induced to use Mn or Fe oxides as electron acceptors leading to their dissolution, increasing As displacement in the aqueous phase (Fendorf and Kocar, 2009).

Several studies have shown that continuous flooded irrigation results in the highest absorption of As by rice crops. AWD (alternate wetting and drying) is an irrigation technique that allows soil water to subside until the soil reaches an aerobic state in unsaturated soil conditions. According to IRRI (http://www.knowledgebank.irri.org/) AWD is a water-saving technology where irrigation water is applied a few days after the disappearance of the ponded water. Hence, the field gets alternately flooded and non-flooded. This technique allows a reduction in water used without penalizing rice grain yield when water depth dropped to no more than 15 cm below soil surface (safe AWD) and field is re-flooded to a water layer of 5 cm. An increase in oxygen concentration in the rhizosphere may increase redox potential, limiting As mobilization (Seyfferth et al., 2018). Several studies have reported that AWD could lead to a reduction in the accumulation of As in grain (Yang et al., 2017; Carrijo et al., 2018; Li et al., 2019), thereby contributing positively to food safety while lowering the environmental impact of rice crops and reducing greenhouse gas emissions (Linquist et al., 2015; Tarlera et al., 2016). In relation to rice yield response to AWD, there is a high degree of variation depending on timing, duration and severity during the drying event of this technique. Previous experiments conducted in Uruguay, reported a yield loss of 15% with the AWD treatment tested that allowed a 50% depletion of available water, relative to continuously flooded management (Carracelas et al., 2019b). It was also reported by other authors that rice yield can be reduced when soil moisture was below saturation (Bouman and Tuong, 2001; Tuong et al., 2005; Parent et al., 2010; Sudhir-Yadav et al., 2012; Carrijo et al., 2017). However, some studies reported no significant impact on rice grain yield with safer AWD techniques (Tabbal et al., 2002; Belder et al., 2004; Sudhir-Yadav et al., 2011a, b; Lampayan et al., 2015; Carrijo et al., 2017; Yang et al., 2017; Carrijo et al., 2018).

Differences within varieties have been reported in the As levels accumulated in root tillers and grain (Zhu et al., 2008). Accumulation of As in grain was found to be

higher in *Indica* rice varieties compared to *Japonicas* (Jiang et al., 2011). More than 95% of the As absorbed remains in the roots and only 1% is accumulated in the grain (Rahman et al., 2007).

Arsenic levels in food are strongly regulated and international standards are being continuously debated and revised. Recommended inorganic arsenic (iAs) levels for polished and brown rice in the CODEX are 0.2 and 0.35 mg kg<sup>-1</sup>, respectively (FAO and WHO, 2019). Compliance with these standards influences access to international markets which is crucial for exporting countries like Uruguay. Regional Mercosur technical regulation on maximum limits of As in foods are 0.30 mg kg<sup>-1</sup> (MERCOSUR, 2011). The 0.30 mg kg<sup>-1</sup> is the maximum total As permitted content to the edible part of the food product. This Technical Regulation does not apply to foods for infants and young children. The iAs concentration for infant rice products limit is below 0.10 mg kg<sup>-1</sup> in the USA (FDA, 2016).

Given the permanent review of international standards in terms of safety, it is important to have local information on cultivated rice varieties and management mitigation alternatives for reducing the levels of As to promote food safety, consumer health, sustainability and competitiveness of the rice sector in Uruguay.

Rice is the largest irrigated crop in Uruguay with 164500 ha cultivated annually (DIEA MGAP, 2018). National total rice production is 1.4 million tons of paddy rice per year, of which more than 90% is exported worldwide. As such, Uruguay ranks seventh in terms of global rice exports and is one of the main exporters in South America (FAO, 2018). Continuous flooding is the main irrigation technique implemented by farmers and the most planted varieties are *Indicas*, to secure the highest yields, which were shown by several authors to maximize As uptake in rice grain yields. The Uruguayan rice sector is divided in three regions: East (118391 ha), North (33 448 ha) and Central (12 618 ha) representing 72%, 20% and 8% of total annually rice planted area (DIEA MGAP, 2018). Dams built for irrigation purposes that capture rainfall water are the main water source (54%) especially in the North and Central region while in the East the main water sources are rivers, lagoons and dams on a smaller proportion (DIEA MGAP, 2017). These water sources have reported low arsenic values (Verger., 2015; Falchi et al., 2018; Mañay et al., 2019) which were

below the limits of 0.050 mg L<sup>-1</sup> (Class2a) and 0.0050 mg L<sup>-1</sup> (Class 3) for irrigation surface water (Decreto N° 253/79, 1979). High levels of As in groundwater have been reported in some wells located in the south-west region of Uruguay (Mañay et al., 2018), 52% wells were above the limit recommended for drinking water of 0.010 mg L<sup>-1</sup> (WHO, 2017 ) while 27% were higher than the national limit in Uruguay of 0.020 mg L<sup>-1</sup> (UNIT 833, 2008). Falchi et al., 2018, reported in the main rice region of Uruguay (East) lower groundwater As levels in average of 0.0063 mg L<sup>-1</sup> (0.0022 – 0.0095 mg L<sup>-1</sup>) which were below local and the international limits (UNIT 833, 2008, WHO, 2017). This is unlikely to be an issue as rice is not cultivated in the south-west region and currently no underground water from aquifers is pumped for irrigation purposes in the rice sector in Uruguay.

The general objective of this paper was to determine the iAs accumulation in rice grain in two contrasting soils sites, Paso Farias - Artigas (PF) located in the North region and Paso de la Laguna - Treinta y Tres (PdL) in the East region, commonly used for rice production in Uruguay. This research project also aimed to identify alternative irrigation management techniques to traditional flooding that could be used to limit or reduce the iAs accumulation in grain and to determine differences in iAs levels within the two most commonly planted rice varieties in Uruguay.

The specific aims of this research were to: 1. determine if continuous flooded conditions can increase the bio-availability of As in soils, resulting in a higher accumulation of As in grain in relation to the alternative irrigation technique AWD, 2. determine if *Indica* varieties promote higher levels of absorption and accumulation of As in the grain in relation to *Japonicas* and 3. investigate if soil types have an influence on the levels of As accumulation in rice grain.

#### **3.4 Methods**

### **3.4.1 Study site description**

Experiments were conducted in two experimental units located in Paso Farias, Artigas department in the Northern region (PF: Lat: -30.50S, Long: -57.12W) and in Paso de la Laguna, Treinta y Tres department, in the Eastern region (PdL: Lat: -33.27S, Long:

-54.17W) of Uruguay (Figure 3.1). This study was conducted throughout the rice growing seasons of 2014/15 - 2015/16 - 2016/17 in PdL and during season 2014/15 - 2016/17 in PF. These study sites have soils which are typical of the rice growing regions in Uruguay.



**Figure 3.1.** Locations of rice field experimental sites in Paso Farias (PF) in northern Uy and Paso de la Laguna (PdL) in eastern Uy.

### **3.4.2 Field management**

For all years, the planting date ranged from 29 September to 03 October and 08 October to 14 October for the PF and PdL locations, respectively.

Land preparation consisted of a minimum tillage performed in the summer, approximately six to nine months prior to rice planting. Disc plowing was used to control weeds and incorporate previous pasture residues. Additionally, one landplane operation was done and contour levees of 20–30 cms height were constructed. Tillage operations, sowing, pre, post-emergence weed controls and first Nitrogen application were all done on dry soils before permanent flooding. Typical rotation in the experimental sites consisted of one year of rice followed by two to three years of perennial pastures (mixes of grasses and legumes).

Soil property information for each field site was determined at the INIA soil laboratory (Table 3.1).

| Soil Parameter      | Experimen                       | Experimental Site        |  |  |
|---------------------|---------------------------------|--------------------------|--|--|
|                     | Paso Farias, PF                 | Paso de la Laguna, PdL   |  |  |
| pH (water)          | 7.1                             | 5.9                      |  |  |
| Organic Matter %    | 4.7                             | 2.1                      |  |  |
| P Citric Acid (ppm) | 4.5                             | 6.9                      |  |  |
| K (meq/100g)        | 0.24                            | 0.18                     |  |  |
| Texture             |                                 |                          |  |  |
| Sand %              | 10                              | 30                       |  |  |
| Silt %              | 38                              | 43                       |  |  |
| Clay %              | 52                              | 27                       |  |  |
| Soil                | Vertisol (Itapebí Tres Arboles) | Brunosol (La Charqueada) |  |  |

**Table 3.1.** Soil parameters information determined in INIA soil laboratory. Soil

 texture information for the first horizon (0-30cms) Source: SIGRAS, webpage.

*Indica* and *Japonica* type cultivars were planted at both sites (Figure 3.1). Direct sowing of rice was performed using a six-row (PF location) or nine-row (PdL location) Semeato brand grain drill (<u>https://www.semeato.com.br/</u>). Row spacings were 17 and 20 cm for the PF and PdL sites, respectively. Sowing density ranged from 145 kg ha<sup>-1</sup> to 165 kg ha<sup>-1</sup> depending on the variety as the sowing rate was adjusted by germination percentage and weight of seeds to get the target of around 500 viable seeds m<sup>-2</sup>.

Fertilization of the crop was based on soil fertility analyses for each site. In PF it consisted of a basal application of Nitrogen (18 kg N ha<sup>-1</sup>), Phosphorus (46 kg  $P_2O_5$  ha<sup>-1</sup>) and Potassium (36 kg  $K_2O$  ha<sup>-1</sup>) plus two urea applications at tillering, prior to the flooding and panicle initiation (35 kg N ha<sup>-1</sup> each). In PdL the basal fertilization was at 12 kg N ha<sup>-1</sup>, 66 kg  $P_2O_5$  ha<sup>-1</sup> and 45 kg  $K_2O$  ha<sup>-1</sup> while urea fertilization at tillering and panicle initiation was 23 kg N ha<sup>-1</sup> each at each application.

Weed controls varied accordingly to the type of weeds and their degree of incidence across sites and seasons, as per INIA's recommendations. In PdL the chemical products used to control weeds were: glyphosate, propanil, quinclorac, clomazone, exocet, cibelcol and ciperof. In PF: glyphosate, clomazone pyrazosulfuron, metsulfuron and penoxsulam at the standard recommended doses and rates was used. Applications of fungicides to control diseases were not necessary.

### 3.4.3 Treatments and experimental design

The experimental design was a split plot with 4 blocks in both the PF and PdL sites. Main plots consisted of Irrigation treatments while Variety formed the split plot. Four varieties (3 *Indicas* and 1 *Japonica*) were tested in PF: INIA Olimar, ElPaso144, INIA Merin (*Indicas*) and INIA Parao (*Japonica*). Also 4 varieties were evaluated in PdL: INIA Olimar, ElPaso144 (*Indicas*) and INIA Parao, INIA Tacuarí (*Japonica*) (Figure 3.2).

Two irrigation management practices were compared at each site. Continuous traditional flooding (C) that represents the most common rice flood management (control) and the alternative irrigation method: alternate wetting and drying (AWD). In treatment C, flooding started 20-30 days after emergence and a water layer of 10 cm above the soil surface was maintained after flooding throughout all the crop cycle. For the AWD treatment, the soil was permitted to dry periodically, allowing a water depletion of 50% of soil available water in the first 20 cm of the soil, which was equivalent to 22-25 mm for the soils at PF and PdL. A water balance was conducted for each site to manage the irrigation in the AWD treatment considering the effective precipitation, crop evapotranspiration and soil water storage capacity. Effective precipitation - EP (mm) was calculated considering the rainfall and surface runoff water according to the precipitation index method and is available at http://www.inia.uy/gras/Monitoreo-Ambiental/. The evapotranspiration was retrieved from INIA weather stations ( http://www.inia.uy/). The available water storage capacity for the soils was determined by the difference between the volumetric moisture at field capacity and the volumetric moisture at permanent wilting point. Both parameters were obtained from the tension-humidity curve obtained using the
Richards method (Richards, 1948). Additionally, moisture content in the soil was determined in the AWD treatment in the PdL site. The methods used were gravimetric, with weekly measurements at a depth of 0-15 and 15-30 cm, and by capacitance probes FDR (Decagon Devices, EC-5) with continuous measurements, installed at a depth of 0-10 cm.

This management technique (AWD) resulted in oxic and anoxic conditions in the soil (saturated and unsaturated), until panicle initiation. After this stage the crop was continuously flooded and managed as per the control treatment C (Figure 3.2).



**Figure 3.2.** Irrigation treatments (traditional continuously-flooded (C) and alternate wetting and drying (AWD)) and varieties (*Indicas and Japonicas*) tested in the two experimental sites, Paso Farias (PF) and Paso de la Laguna (PdL).

The source of irrigation water was different between sites. In PF, irrigation water source was from a reservoir (gravity irrigation) while in PdL, the irrigation water was pumped from the local river (Olimar).

#### 3.4.4 Chemicals and crop parameters measured

The parameters measured were: 1. In the Soil: Total arsenic (tAs) and Bioavailable arsenic (bioAs) at sowing, sampled at two depths: 0-15cm and 15-30cm . Bioavailable As at the end of the crop cycle (harvest) was also measured at the same two soil depths. 2. In the Water: Arsenic (As), at 5 and 6 periods during the flooding for AWD and C treatments, respectively. 3. In the crop: Inorganic arsenic in polished grain. All samples were analyzed in the Technological Laboratory of Uruguay - LATU. Additionally, pH and redox potential were measured in irrigation water at each sampling moment (0, 5, 10, 30, 45, 60 days from the start of flooding in C and 0, 5, 10, 30, 45 days in AWD). Also, the crop was harvested to determine rice yield for each treatment.

#### 3.4.4.1 Arsenic in Soil

Bioavailable Arsenic (bioAs) and total Arsenic (tAs) were determined at two soil depths at two stages throughout the growing season: sowing (Initial) and harvesting (End). bioAs represents specifically-sorbed As in soils that may be potentially mobilized due to changes in pH or P addition (Wenzel et al., 2001). Soil samples were made to pass through a 2 mm sieve, dried until constant weight and homogenized in a porcelain mortar (ASTM, 2015). For tAs analysis 1 g of dried soil was digested with 10 mL of nitric acid in a microwave oven (Millestone, Ethos One, Italy) and the digests were diluted up to 30 mL with deionized water (United States Environmental Protection Agency, 2007). Inductively coupled plasma- Optical emission Spectrometry was used to determine total arsenic in soil samples (ISO, 2007). bioAs was extracted using 0,05 M NH4H2PO4 (Stroud et al., 2011; Wenzel et al., 2001). Five grams of soil was mixed with 25 mL of 0.05 M NH4H2PO4 and shaken at room temperature for 16 h in an orbital shaker (GLF 3016, Deutschland). The samples were centrifuged at 3000g for 15 min and the supernatants were filtered through a 0.45-mm membrane filter. Graphite furnace atomic absorption spectrometry was used to determine bioavailable As in soil samples. 10  $\mu$ g of palladium nitrate (Pd(NO3)2) and 6  $\mu$ g of magnesium nitrate (Mg(NO3)2) were used as regular modifier in a transversally heated graphite furnace with Zeeman correction (Perkin Elmer, AA 800, USA).

#### 3.4.4.2 Arsenic in Water

Graphite furnace atomic absorption spectrometry was used to determine As in water samples (ISO, 2003). As was measured at 193.7 nm in an Atomic Spectrometer lengthwise heated with deuterium background correction (Perkin Elmer, AA 200, USA). 15  $\mu$ g of palladium nitrate (Pd (NO<sub>3</sub>)<sub>2</sub>) and 10  $\mu$ g of magnesium nitrate (Mg (NO<sub>3</sub>)<sub>2</sub>) were used as a regular matrix modifier. Sampling was done at 0, 5, 10, 30, 45 and 60 days after flooding in the continuous irrigation treatment while in AWD treatment sampling was done up to 45 days after flooding only as flooding started after panicle initiation and the duration of this period was shorter in this treatment.

Redox potential and pH were measured in the field using a portable device Horiba model D-52-meter manual platinum electrode (Kurosawa et al., 2013; Tarlera et al., 2016). This device allowed the recording of instantaneous measurements at each sampling event (0,5,10,30,45,60) days after flooding in C and 0,5,10,30 and 45 days in AWD. Five replicates measurements were taken between rows at 10 cm soil depth, in each of the four blocks in the flooded soil.

**Table 3.2.** Analytical detection and quantification limits of the methodologies used to determine inorganic arsenic in grain (iAs), soil (tAs and bioAs) and water (As) by the Technological Laboratory of Uruguay.

|                              | <b>Rice Grain</b>                          | S                                      | Water  |                                     |
|------------------------------|--|--|--|-------------------------------------|
| methodology limits           | Inorganic As<br>(iAs mg kg <sup>-1</sup> ) | Total As<br>(tAs mg kg <sup>-1</sup> ) | Bioavailable As<br>(bioAs µg L <sup>-1</sup> ) | Arsenic<br>(As mg L <sup>-1</sup> ) |
| Detection Limit (DL)         | 0.03                                       | 0.6                                    | 10   | 0.001                               |
| Quantification limit<br>(QL) | 0.06                                       | 3                                      | 20   | 0.003                               |

The limits of detection and quantification to determine arsenic in water, soil and grain by the Technological Laboratory of Uruguay (LATU) are presented in Table 3.2. In order to perform statistical analyses, when a sample was below the analytical detection limit (DL) it was considered as half of the value of DL and when a sample data was higher than the limit of detection but lower than the limit of analytical quantification (QL), the mean value between both analytical limits was used.

#### 3.4.4.3 Inorganic Arsenic (iAs) in polished grain

Polished rice grain samples were frozen until grinding and were grinded with a blade mill to pass a 1 mm sieve. 1 gr of milled rice was digested with 10 mL of 0.28M Nitric Acid (Merck, 65% for analysis) in 50 mL plastic tubes, 90 minutes at 95°C in a preheated water bath (GLF 1083, Deutschland). The extracts were diluted with 6.7 mL of deionized water, centrifuged at 3000 rpm for ten minutes and filtered with a 0.45 µm nylon syringe filter. The filtrate pH was adjusted to 6-8.5. High performance liquid chromatography (Flexar, Perkin Elmer, USA) coupled to inductively coupled plasma mass spectrometry (Nex Ion 350 D, Perkin Elmer, USA) was used to determine inorganic arsenic as the sum of two inorganic forms of arsenic, arsenite and arsenate (Kubachka et al., 2012). Hamilton PRP-X100 anion exchange column (5µ, 4,6 x150 mm) was used, and 10 mM ammonium phosphate dibasic (99,5 % pure, Crystals, Mallinckrodt) at pH of 8.25 ( $\pm$  0,05) was used as mobile phase. As was monitored at m/z of 75 with standard cell mode. Calibration curves were prepared with arsenite (998 mg  $L^{-1}$ ), arsenate (1000 mg  $L^{-1}$ ) stock standards from Spex Certiprep (USA), Monosodim acid methane arsonate sesquihydrate MMA (≥99.5%) from ChemService (USA) and Cacodylic Acid- DMA (>99.0%) from Sigma Aldrich (USA). Every 20 samples, one blank, two fortified samples, and one certified reference material (1568b Rice Flour, National Institute of Standards and Technology, USA; and 7532a, Brown Rice Flour National Metrology Institute of Japan) were included as quality control samples. Certified reference materials (1568b and 7532a) were used to assess the accuracy of total As concentration and As speciation for rice flour.

## 3.4.4.4 Rice yields (kg ha<sup>-1</sup>)

Harvest was done manually in the middle of experimental treatments plots when grain moisture was lower than 21%. Harvested area was  $5.95m^2$  (7rows X 5 meters) in PdL and  $5.1m^2$  each (10 rows X 3 meters) in the PF site. The rice samples were mechanically threshed, and grain yields were normalized to 14% moisture.

#### **3.4.5 Data analysis**

Statistical analyses were all performed in R software (R Core Team, 2018) using the emmeans (Lenth, 2018) and nlme packages (Pinheiro et al., 2019). A linear mixed effect model was used to fit each of the response variables. Analyses of variance was followed by means separation using the Tukey test. For iAs in grain and rice yield, the fixed effects considered were: Site, Irrigation, Varieties and their interactions. Block, Irrigation and Season were considered as random effects according to a split-plot experimental design. All other soil and water measured parameters were also analyzed using the linear mixed effect model.

# **3.5 Results**

#### 3.5.1 Total Arsenic (tAs) and Bioavailable Arsenic (bioAs) in soils

Average initial tAs in the soil at sowing was 2.14 mg kg<sup>-1</sup> in PF site, while in the soils at PdL site tAs was 69% significantly higher with an average value of 3.62 mg kg<sup>-1</sup>. Additionally, bioAs was 15.1  $\mu$ g L<sup>-1</sup> (99%) higher in PdL compared to PF (Table 3.3).

There were no significant differences in tAs and bioAs within soil samples in different soil layers (depths: 0-15 cm and 15-30 cm). Also, the interaction site\*soil depth was not significant for these soil parameters (P<0.05).

At both sites, average bioavailable As concentrations increased during the rice growing season. There were no significant differences in the levels of bioAs registered at harvest within sites. Additionally, no significant differences were measured in the bioAs levels, within the two irrigation treatments evaluated, C and AWD (Table 3.4).

**Table 3.3.** Total Arsenic (tAs) and Bioavailable Arsenic (bioAs) registered in soil samples taken initially (sowing of rice) at different soil depths (0-15 cm and 15-30 cm) in two experimental sites: Paso de la Laguna (PdL) and Paso Farias (PF).

|                         | Arsenic in Soils at sowing (Initial)        |                      |  |  |
|-------------------------|---|----------------------|--|--|
| Classification criteria | Total Arsenic<br>(tAs mg kg <sup>-1</sup> ) | Bioavailable Arsenic |  |  |
| Site                    |   | (010115 µg L )       |  |  |
| PdL                     | 3.62 a                                      | 30.30 a              |  |  |
| PF                      | 2.14 b                                      | 15.21 b              |  |  |
| Average                 | 2.88  | 22.76                |  |  |
| CV%                     | 27.56                                       | 15.40                |  |  |
| P<0.05                  | ***   | ***                  |  |  |
| Depth (P<0.05)          | NS  | NS                   |  |  |
| Site* Depth (P<0.05)    | NS  | NS                   |  |  |

Means followed by different letters are significantly different with a probability less than 5% (P < 0.05). Signif. codes: '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05; NS: non-significant differences. CV: coefficient of variation.

**Table 3.4.** Bioavailable arsenic in soils (bioAs  $\mu$ g.L<sup>-1</sup>) at harvest time by irrigation treatments and the interaction with experimental sites, Paso de la Laguna (PdL) in the East and Paso Farias (PF) in the North.

| Classification criteria               | Bioavailable<br>Arsenic in Soils<br>(bioAs µg. L <sup>-1</sup> )<br>Final-Harvest |
|---------------------------------------|---|
| Irrigation                            | NS  |
| Site                                  | NS  |
| Irrigation*Site                       |   |
| Site -PdL                             |   |
| 1.Continuous (C)                      | 27.00 a   |
| 2. Alternate Wetting and Drying (AWD) | 34.67 a   |
| Site-PF                               |   |
| 1.Continuous (C)                      | 40.88 a   |
| 2. Alternate Wetting and Drying (AWD) | 29.31 a   |
| Average                               | 32.96   |
| CV%                                   | 11.71   |
| P<0.05                                | *   |

Means followed by different letters are significantly different with a probability less than 5% (P < 0.05). Signif. codes: '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05; NS: non-significant differences. CV: coefficient of variation.

## 3.5.2 Arsenic, pH and Redox potential (Eh) in Water

Average Arsenic levels registered in the irrigation water were 0.00224 mg L<sup>-1</sup>. Arsenic levels in the irrigation water were 55% higher in PdL in relation to the PF site. The AWD treatment resulted in a significant As reduction in irrigation water of 24% in relation to C for both sites. (Table 3.5).

**Table 3.5.** Average irrigation water Arsenic levels, pH and Redox Potential, measured

 during the flooding period by Site and Irrigation management.

| Classification<br>criteria | Arsenic in water<br>(As mg L <sup>-1</sup> ) | рН     | Redox<br>Potential<br>(Eh mV) |  |
|----------------------------|--|--------|-------------------------------|--|
| Site                       |  |        |                               |  |
| PdL                        | 0.00272 a                                    | 5.50 a | 79.68 a                       |  |
| PF                         | 0.00176 b                                    | 6.09 b | 83.72 a                       |  |
| Average                    | 0.00224                                      | 5.80   | 81.7                          |  |
| CV%                        | 22.72  | 2.17   | 54.57                         |  |
| P<0.05                     | ***  | ***    | *                             |  |
| Irrigation                 |  | •      |                               |  |
| С 0.00255 а                |  | 5.75 a | 104.73 a                      |  |
| AWD                        | 0.00193 b                                    | 5.84 a | 58.67 b                       |  |
| Average                    | 0.00224                                      | 5.80   | 81.7                          |  |
| <i>CV</i> %                | 22.68  | 2.17   | 54.59                         |  |
| P<0.05                     | **   | NS     | ***                           |  |
| Site* Irrigation           | * Irrigation NS                              |        | ***                           |  |

Means followed by different letters are significantly different with a probability less than 5% (P < 0.05). Signif. codes: '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05; NS: non-significant differences. CV: coefficient of variation.

Water pH was significantly higher (11 %) at the PF site in the North (6.1) relative to the PdL site in the East (5.5). There were no significant differences in the pH registered in irrigation water between C and AWD at either site. The interaction between irrigation and site was not significant for the pH parameter (Table 3.5).

Average Redox Potential of water was 81.7 with non-significant differences between both experimental sites (PF and PdL). Average pH and Eh evolution trend (two seasons) for each irrigation treatment and experimental site are presented in Figure 3.3. In C, values were measured right after the establishment of permanent flood (2030 days after emergence) while in the AWD this measurement started during the flooding period from panicle initiation until 20 days before harvest.



**Figure 3.3.** (A,B) Redox Potential Eh (mV) and (C,D) pH evolution (average) at different days from the start of flooding, for each irrigation treatment, continuous (C) and alternate wetting and drying (AWD) and for each experimental site, Paso de la Laguna (PdL) and Paso Farias (PF).

The initial values of redox potential in C were higher in PdL in comparison with PF, following a reduction of these values in both treatments during the flooding period (Figure 3.3A). At the final sampling event (60 days after flooding) PdL reached lower negative values while in PF, Eh values were almost zero. In AWD treatments, both sites had very similar Eh trends, with PF having higher values at the first sampling event, while PdL also reached lower negative values at the final sampling date (Figure 3.3B).

The pH values were initially lower (acid) in PdL in relation to PF and increased during the flooding period, tending to a value near neutrality (pH 6.0) at both treatments and sites (Figure 3.3C and 3.3D). At PF, pH values were always higher than levels registered at PdL.

## 3.5.3 Arsenic in Grain

Average Inorganic arsenic values registered in polished grain across both sites were 0.07 mg kg<sup>-1</sup>. This parameter was significantly lower in PF (0.043 mg kg<sup>-1</sup>) than in PdL (0.091 mg kg<sup>-1</sup>) (Table 3.6).

Significant differences within varieties were registered while the interaction between irrigation:variety for iAs was not significant. (Table 3.6). *Japonica* cultivars INIA Parao and INIA Tacuarí resulted in the lowest iAs values in relation to *Indica* type cultivars EP144 and INIA Olimar. Average values were 0.03 mg kg<sup>-1</sup> lower in *Japonica* cultivars compared to *Indica* varieties (0.05 vs 0.08 mg kg<sup>-1</sup>, respectively). However, no significant differences were registered between INIA Parao and INIA Merín (Figure 3.4).





**Table 3.6.** Inorganic Arsenic (iAs mg kg<sup>-1</sup>) levels accumulated in polished rice grain by sites, irrigation treatments and main varieties cultivated in Uruguay.

| Classification criteria                  | Inorganic Arsenic in Grain (iAs mg kg <sup>-1</sup> ) |  |  |  |
|--|---|--|--|--|
| Site                                     |   |  |  |  |
| PdL (Paso de la Laguna – Treinta y Tres) | 0.091 a   |  |  |  |
| PF (Paso Farias – Artigas)               | 0.043 b   |  |  |  |
| Average                                  | 0.067   |  |  |  |
| CV%                                      | 4.524   |  |  |  |
| <i>P</i> <0.05                           | ***   |  |  |  |
| Irrigation                               |   |  |  |  |
| 1.Continuous (C)                         | 0.069 a   |  |  |  |
| 2. Alternate Wetting and Drying (AWD)    | 0.064 a   |  |  |  |
| Average                                  | 0.067   |  |  |  |
| CV%                                      | 4.540   |  |  |  |
| <i>P</i> <0.05                           | *   |  |  |  |
| Variety                                  |   |  |  |  |
| Tacuarí (Japónica)                       | 0.046 c   |  |  |  |
| Parao (Japónica)                         | 0.057 bc  |  |  |  |
| Merín (Indica)                           | 0.076 ab  |  |  |  |
| EP144 (Indica)                           | 0.077 a   |  |  |  |
| Olimar (Indica)                          | 0.079 a   |  |  |  |
| Average                                  | 0.067   |  |  |  |
| CV%                                      | 6.651   |  |  |  |
| <i>P</i> <0.05                           | ***   |  |  |  |
| Irrigation*Site                          |   |  |  |  |
| PdL                                      |   |  |  |  |
| 1.Continuous (C)                         | 0.086 a   |  |  |  |
| 4. Alternate Wetting and Drying (AWD)    | 0.097 a   |  |  |  |
| PF                                       |   |  |  |  |
| 1.Continuous (C)                         | 0.053 a   |  |  |  |
| 4. Alternate Wetting and Drying (AWD)    | 0.032 b   |  |  |  |
| Average                                  | 0.067   |  |  |  |
| <i>CV</i> %                              | 6.134   |  |  |  |
| P<0.05                                   | ***   |  |  |  |
| Irrigation * Variety                     |   |  |  |  |
| P<0.05                                   | NS  |  |  |  |

Means followed by different letters are significantly different with a probability less than 5% (P < 0.05). Signif. codes: \*\*\*\*' 0.001 \*\*\* 0.01 \*\* 0.05; NS: non-significant differences. CV: coefficient of variation. (.) = non-estimated A significant interaction between Irrigation and Site was detected for iAs (Table 3.6). There were no differences within irrigation treatments in the PdL site (average 0.091 mg kg<sup>-1</sup>), while AWD determined a significant iAs reduction in grain of 0.02 mg kg<sup>-1</sup> (39.5%) in relation to the traditional continuous flooding in the PF site (Figure 3.5).



**Figure 3.5.** Inorganic arsenic (mg kg<sup>-1</sup>) accumulation in polished rice white grain by irrigation management: C: continuous and AWD alternate wetting and drying recorded in different regions. Black dot represents least-square means, grey bars are indicating standard errors, red arrow lines indicates confidence intervals by Tukey. Different letters are significantly different with a probability less than 5%.

# 3.5.4 Rice Yield

Average harvested rice yield in this study was 8567 kg ha <sup>-1</sup>. Values of this parameter reported in the PdL site were 21 % higher (1577 kg ha <sup>-1</sup>) than the mean yield recorded for the PF site. Significant differences in rice yield were recorded for region, irrigation management and varieties evaluated (Table 3.7).

**Table 3.7.** Rice grain yield (kg ha<sup>-1</sup>, 14% moisture) registered in two sites of Uruguay, by irrigation treatments and main varieties planted in Uruguay.

| Classification criteria                  | Rice Yield (kg ha <sup>-1</sup> ) |  |  |  |
|--|-----------------------------------|--|--|--|
| Sites                                    |                                   |  |  |  |
| PdL (Paso de la Laguna – Treinta y Tres) | 9500 a                            |  |  |  |
| PF (Paso Farias – Artigas)               | 7635 b                            |  |  |  |
| Average                                  | 8567                              |  |  |  |
| CV%                                      | 2.14                              |  |  |  |
| P<0.05                                   | ***                               |  |  |  |
| Irrigation                               |                                   |  |  |  |
| 1.Continuous (C)                         | 9230 a                            |  |  |  |
| 2. Alternate Wetting and Drying (AWD)    | 7904 b                            |  |  |  |
| Average                                  | 8567                              |  |  |  |
| CV%                                      | 2.42                              |  |  |  |
| P<0.05                                   | ***                               |  |  |  |
| Variety                                  |                                   |  |  |  |
| Tacuarí                                  | 6279 d                            |  |  |  |
| Parao                                    | 8095 c                            |  |  |  |
| Merín                                    | 10203 a                           |  |  |  |
| EP144                                    | 9289 ab                           |  |  |  |
| Olimar                                   | 8971 b                            |  |  |  |
| Average                                  | 8567                              |  |  |  |
| CV%                                      | 2.86                              |  |  |  |
| P<0.05                                   | ***                               |  |  |  |
| Irrigation*Site                          | L                                 |  |  |  |
| P<0.05                                   | NS                                |  |  |  |
| Irrigation * Variety                     | L                                 |  |  |  |
| P<0.05                                   | NS                                |  |  |  |

Means followed by different letters are significantly different with a probability less than 5% (P < 0.05). Signif. codes: '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05; NS: non-significant differences. CV: coefficient of variation.

The AWD irrigation treatment resulted in a significant yield reduction of 14% (-1326 kg ha<sup>-1</sup>) in comparison to the traditional continuous flooding irrigation technique.

The highest rice yields were registered with INIA Merín (10203 kg ha<sup>-1</sup>) and EP144 (9289 kg ha<sup>-1</sup>) both *Indica* varieties, followed by INIA Olimar with no significant difference with EP144. The *Japonica* cultivars INIA Parao had a significantly lower rice grain yield in relation to *Indica* cultivars (15% reduction) and INIA Tacuarí reported the lowest rice grain yield (Table 3.7, Figure 3.6).



**Figure 3.6.** Rice grain yields (kg ha<sup>-1</sup>, 14% moisture) for different varieties by irrigation techniques. Black dot represents least-square means, grey bars are indicating standard errors, red arrow lines indicates confidence intervals by Tukey. Different letters are significantly different with a probability less than 5%.

#### **3.6 Discussion**

#### **3.6.1** Arsenic concentration in soils

In the Brunosol soils at the PdL site, average initial tAs at sowing was significantly higher (69%) in comparison to the Vertisols soils at the PF site. One of the natural sources of Arsenic into paddy rice crops can be derived from the soil type (Meharg and Zhao, 2012) which depends on the sediments that it originated from. The levels of As found in the soils at sowing in the two experimental sites located in the PdL and PF sites (3.62 and 2.14 mg kg<sup>-1</sup>, respectively) were below the reported natural world

concentration of As in soils of 5 mg kg<sup>-1</sup> (Hossain et al., 2008), of 5-10 mg kg<sup>-1</sup> (Han et al., 2003) and well below the Canadian limit for agricultural soils of 12 mg kg<sup>-1</sup> (CCME, Canadian Environmental Quality Guidelines). The differences within sites could be associated with lower organic matter (%) and clay percentage recorded in the soils at PdL in relation to PF, as reported in previous studies by Verger, 2015. Studies performed by Quintero et al in 2010, in Entre Rios-Argentina found an average soil tAs value of 2.9 mg kg<sup>-1</sup>, ranging from 1.6 mg kg<sup>-1</sup> in fluvial sediments soils, 3.9 mg kg<sup>-1</sup> in Vertisols of central-south and 4.1 mg kg<sup>-1</sup> in wetlands soils of the north.

Arsenic concentration in the soil solution would reflect the bioavailability of As because rice roots absorb As mostly from the soil solution (Xu et al., 2008). No differences in the levels of bioAs were registered at different soil depths during both crop stages when measurements were taken (sowing and harvest) and no differences in bioAs was detected within irrigation treatments. The bioAs at sowing in the soils of our study was 15.1  $\mu$ g.L<sup>-1</sup> (99%) higher in the PdL site compared to the PF site. However, no significant differences were recorded in bioAs levels within regions during the final sampling at harvest. Average bioAs levels increased during the cropping cycle from sowing to harvesting (Table 3.3 and 3.4). Arsenic bioavailability has been found to increase under reduced soil conditions, as Fe oxyhydroxides to which As is adsorbed are dissolved and become available to the rice roots (Kumarathilaka et al., 2018). Other authors have reported that arsenic transported through water during irrigation could be another natural source of As into the rice cropping systems (Meharg and Zhao, 2012). However, the average Arsenic levels registered in the irrigation water in this study, were very low in relation to the limited restriction values for irrigation water (Decreto N° 253/79, 1979). For this reason, the increase in the bioAs during the crop growth period is likely not related to arsenic transported through irrigation water and was associated with the reduced soil conditions. Additionally, tAs in the soil across sites was also very low and below the reported natural values around the world (Hossain et al., 2008, Han et al., 2003) and well below the limit for agricultural soils according to the Canadian Environmental Quality Guidelines (CCME). While there are other possible sources of As (anthropogenic) such as industrial/urban pollution for paddies downstream of large population centers, contamination of irrigation water, use of fertilizers and pesticides contaminated with arsenic (Meharg and Zhao, 2012), these are generally not relevant

in Uruguay rice growing situations. The amount of phosphate fertilizers used in Uruguay and particularly in this study was very low and currently no organic manure is used in Uruguay rice systems, hence this is unlikely to be a source of Arsenic contamination. Therefore, the measured increases in bioAs in the soils over the two experimental sites during the cropping period are likely due to soil/water interactions causing reduced soil conditions and greater bioavailability of As. However, it is important to highlight that bioAs levels were found to be low across both sites and unlikely to be an issue at current levels.

### 3.6.2 Arsenic concentration in water, pH and Redox Potential

The average As levels measured in the irrigation water during this study was 0.00224 mg L<sup>-1</sup>, which is aligned with values reported in two sites of Ecuador of 0.00142 and 0.00307 mg L<sup>-1</sup> (Otero et al., 2016) and were below the limited restriction values for irrigation surface water of 0.05 mg L<sup>-1</sup> (Class 2a) and 0.005 mg L<sup>-1</sup> (Class3) (Decreto N° 253/79, 1979) and well below the limited restriction values for human water consumption of 0.02 mg L<sup>-1</sup> (UNIT 833, 2008) and 0.01 mg L<sup>-1</sup> (WHO, 2017). This information is aligned with the reported average arsenic values of <0.0015 mg L<sup>-1</sup> in the irrigation surface water collected in lagoons, irrigation channels and rice fields in the East region of Uruguay (from <0.0005 mg L<sup>-1</sup> to 0.0036 mg L<sup>-1</sup>) (Verger., 2015, Falchi et al., 2018). Mañay et al. 2019, reported a mean arsenic level of 0.0009 mg L<sup>-1</sup> in irrigation channels and 0.00087 mg L<sup>-1</sup> in a Lagoon, which is one of the most important water resources for rice irrigation in the East region in Uruguay. Significant differences where registered between the two sites, with As levels in the water 55% higher at the PdL site (0.00274 mg L<sup>-1</sup>) compared to the PF site (0.00157 mg L<sup>-1</sup>). However, As levels in water measured across both sites in this study were very low.

Redox potential (Eh) declined and reached a lower and negative minimum value at the final sampling event, which reflected a more reductive soil condition at PdL in comparison with PF. Meharg and Zhao, 2012, determined that As liberation into the soil occurs when Eh is below +250mV at pH=7. An increase of As availability when Eh decreases was also reported by Marin et al., 1993. Eh values and trend reported at the PdL site were aligned with information reported by Tarlera et al., 2016. According

to Masschelyn et al., 1991, an increase in solubility of arsenic can occur due to the reduction of iron oxy-hydroxides within the reported range of Eh. When Eh drops below 150 mV at pH = 7, arsenic solubility may increase due to the reduction of Fe<sup>+3</sup> to Fe<sup>+2</sup> (Marschner et al., 2012). Arsenic mobilization in paddy soils can be strongly impacted by soil redox potential. However, this effect can be difficult to quantify using measurements at a single point in time, as fluctuations of soil Eh can be high during the rice growing season (Meharg and Zhao, 2012). Takahashi et al., 2004, found that the increase in As concentration in the soil solution occurs simultaneously with the rise in Fe and Mn concentration. This author affirms that the solubility of As is strongly regulated by Fe reduction in aqueous systems. The slightly more reductive soils conditions registered at the PdL site are likely associated with the higher arsenic water levels found in this site and with the higher inorganic arsenic contents measured in the grain at this site compared to PF.

According to Honma et al., 2016, the recorded trend of redox potential reduction and pH increase found in this study, could potentially correspond to situations where the availability of As can be reduced (Figure 3.3). This condition may have further contributed to the very low levels of iAs accumulated in grain found in this study.

## **3.6.3** Arsenic concentration in grain

Inorganic arsenic values in white polished grain averaged 0.07 mg kg <sup>-1.</sup> across the study sites. Reported values of iAs in this study were below international and regional legislation limits established for human health and food safety: 0.20 mg kg<sup>-1</sup> for iAs in polished rice grain by the CODEX ALIMENTARIUS (FAO and WHO, 2019), and 0.30 mg kg<sup>-1</sup> for tAs (MERCOSUR, 2011). Globally, reported values of tAs ranged from 0.05 mg kg<sup>-1</sup> to 0.38 mg kg<sup>-1</sup> while reported values of iAs ranged from 0.03 mg kg<sup>-1</sup> up to 0.25 mg kg<sup>-1</sup> (Table 3.8 and Figure 3.7).

| Countries with<br>Reported values   | Inorganic Arsenic<br>iAs (mg kg <sup>-1</sup> )Total Arsenic<br>tAs (mg kg <sup>-1</sup> ) |                       | Rice<br>Type | References            |  |  |  |
|---|--|-----------------------|--------------|-----------------------|--|--|--|
| Taiwan  | 0.25   | 0.38                  | W            | Williams et al., 2005 |  |  |  |
| Argentina   | -  | 0.34                  | М            | Quintero et al., 2010 |  |  |  |
| Argentina   | 0.06   | 0.33                  | М            | Quintero et al., 2014 |  |  |  |
| Argentina   | 0.08   | 0.30                  | W            | Oteiza et al., 2019   |  |  |  |
| Argentina   | 0.10   | 0.30                  | *            | Farías et al., 2015   |  |  |  |
| France  | -  | 0.28                  | W            | Meharg et al., 2009   |  |  |  |
| Australia   | 0.16   | 0.28                  | W            | Rahman et al., 2014   |  |  |  |
| USA   | 0.11   | 0.28                  | W            | Meharg et al., 2008   |  |  |  |
| USA   | 0.08   | 0.26                  | W            | Williams et al., 2005 |  |  |  |
| USA   | -  | 0.26                  | W            | Linquist et al., 2015 |  |  |  |
| Australia   | -  | 0.26                  | W            | Phuong et al., 1999   |  |  |  |
| USA   | 0.10   | 0.25                  | W            | Meharg et al., 2009   |  |  |  |
| USA   | 0.10   | 0.27                  | W            | Zavala et al., 2008   |  |  |  |
| China   | 0.15   | 0.23                  | W            | Zhu et al., 2008      |  |  |  |
| Brazil  | 0.11   | 0.22                  | W            | Batista et al., 2011  |  |  |  |
| Italy   | 0.13   | 0.22                  | R            | Williams et al., 2005 |  |  |  |
| Vietnam   | -  | 0.21                  | W            | Phuong et al., 1999   |  |  |  |
| Spain   | -  | 0.20                  | W            | Meharg et al., 2009   |  |  |  |
| Japan   | -  | 0.19                  | W            | Meharg et al., 2009   |  |  |  |
| Spain   | 0.08   | 0.17                  | Р            | Williams et al., 2005 |  |  |  |
| Thailand  | 0.17   | 0.17 0.17             |              | Rahman et al., 2014   |  |  |  |
| Europe  | 0.08   | 0.16                  | *            | Williams et al., 2005 |  |  |  |
| Italy   | 0.11   | 0.15                  | W            | Meharg et al., 2009   |  |  |  |
| China   | 0.16   | 0.14                  | W            | Meharg et al., 2009   |  |  |  |
| Thailand  | -  | 0.14                  | W            | Meharg et al., 2009   |  |  |  |
| Bangladesh  | 0.08 0.13  |                       | W            | Meharg et al., 2009   |  |  |  |
| Bangladesh  | 0.08   | 0.13                  | W            | Williams et al., 2005 |  |  |  |
| USA   | -  | < 0.12                | W            | Carrijo et al., 2018  |  |  |  |
| Thailand  | 0.08   | 0.11                  | J            | Williams et al., 2005 |  |  |  |
| India   | 0.09   | 0.10                  | В            | Rahman et al., 2014   |  |  |  |
| Pakistan  | 0.08   | 0.09                  | В            | Rahman et al., 2014   |  |  |  |
| India   | 0.03   | 0.07                  | W            | Meharg et al., 2009   |  |  |  |
| Canada  | 0.05   | 0.07                  | Wi           | Williams et al., 2005 |  |  |  |
| Egypt   | -  | 0.05                  | W            | Meharg et al., 2009   |  |  |  |
| India   | 0.03   | 0.05                  | WB           | Williams et al., 2005 |  |  |  |
| Ecuador   | 0.12   | _                     | W            | Otero et al., 2016    |  |  |  |
| Uruguay   | 0.07 -   |                       | W            | Current study         |  |  |  |
| References: W: white, M: mix  | ted; *: not specified; B: b  | asmati; R: risotto; J | : jasmine; W | i: wild; P: paella    |  |  |  |
| International Limits  | iAs = $0.2$ (polished)- $0.35$ (husked). CODEX (FAO and WHO, 2019)                         |                       |              |                       |  |  |  |
| *** If the tAs concentration is below or equal to the limit established for iAs, no further testing is required, and the sample is determined to be compliant with the legislation. If the tAs concentration is above the limit for iAs, follow-up testing shall be conducted to determine the iAs (FAO and WHO, 2019). |  |                       |              |                       |  |  |  |

**Table 3.8.** Inorganic Arsenic (iAs) and total Arsenic (tAs) in rice grain reported in

 different studies worldwide and international limit established for food safety.

The measured iAs values in both experimental sites in this study, were generally lower in comparison with reported values by other authors (Figure 3.7). Accumulation of iAs in rice grain varies across studies could be explained mainly by the wide range of environments, different varieties, soil types, water sources and differences in cropping systems-management. Amongst these limited studies, results from this study in Uruguay were found to be in the lower range of recorded iAs results in rice grain. Most of the rice producing countries reported mean iAs levels below the international limit established by the CODEX (FAO and WHO, 2019).



**Figure 3.7.** Inorganic arsenic accumulation in rice grain reported by country. Red line indicates the international limit for iAs in polished rice grain established for human health and food safety in the CODEX ALIMENTARIUS (FAO and WHO, 2019).

In this study, the highest accumulation of iAs in rice grain was found at the PdL site. Similarly, the highest As values were registered in the soils (tAs and bioAs at sowing) and in the water at this site. This information is aligned with Quintero et al., 2014, that reported highest As accumulation in grain, in cultivated soils with higher As content such as in wetlands in Northern Argentina.

Although overall iAs accumulated in rice grain were low, it was found that levels can be further reduced by the implementation of alternative irrigation management techniques. The AWD techniques used in this study had a significant reduction in iAs accumulation in grain of 40% at the PF site. This information agrees with information reported by Linquist et al., 2015, with a 58% reduction of tAs in polished rice by implementing AWD (from 0.37 to 0.16 mg kg <sup>-1</sup>) in relation to the continuously flooded rice system. Carrijo et al. (2018), also determined that grain total tAs concentration decreased by 56 to 68%, in AWD that allowed the soil to dry out from 45 days after sowing until flowering (50% heading) until it reached 25 – 35 % of soil volumetric water content. However, in less severe treatments such as "safe" AWD where the field was reflooded when the water table reached 15 cm below soil surface, the authors, didn't find a reduction in total As accumulation in relation to the continuously flooded treatment (Carrijo et al., 2018). Other studies with a higher severity of water stress imposed with AWD have also reported a significantly higher reduction in the accumulation of arsenic in rice grain (Das et al., 2016; Lahue et al., 2016).

The lower reduction in iAs measured in this study could be explained by the lower severity of the AWD treatment as it was only implemented until panicle initiation and allowed a water depletion of 50% of the available water. Additionally, at one of the sites (PdL), no differences in inorganic As accumulated in rice grain within irrigation treatments (C and AWD) were found. The soil type and field characteristics of lower slope at the PdL site (table 3.1), could favor the anoxic saturated conditions for longer periods in relation to the PF site. Most likely this didn't allow the development of aerobic conditions in the soil for long enough periods to decrease the soil bioavailability of As at the PdL site.

## 3.6.4 Grain Yield

Grain yield was found to be affected significantly by irrigation method. Despite AWD being shown as an alternative irrigation technique that can reduce As accumulation in rice grain under certain conditions, it was found in this study that yield was reduced by 14% in the AWD treatments (7904 kg ha<sup>-1</sup>) in comparison to continuous flooded treatments C (9230 kg ha<sup>-1</sup>). This information is in agreement with previous studies reported worldwide by other authors (Bouman and Tuong, 2001; Tuong et al., 2005;

Parent et al., 2010; Sudhir-Yadav et al., 2012; Carrijo et al., 2017) and also in Uruguay by Carracelas et al., 2019b, where AWD resulted in a significant yield loss of 1339 kgs ha<sup>-1</sup> in comparison to the traditional continuous flooded treatment. The yield losses associated with the AWD treatment would likely limit the implementation of this technique in commercial farms. In other studies rice yield was not negatively affected when soils were maintained above saturation or rice plants had access to water using "safe" AWD irrigation techniques (Tabbal et al., 2002; Belder et al., 2004; Sudhir-Yadav et al., 2011a, b; Lampayan et al., 2015; Carrijo et al., 2017; Yang et al., 2017; Carrijo et al., 2018). However, under saturated soil conditions and even "safe" AWD practice, no reduction in iAs accumulated in rice grain was reported (Carrijo et al., 2018). The implementation of a mitigation management option, such as AWD, that reduces the crop yield is likely to only be adopted in environments in which arsenic concentrations are an issue.

#### 3.6.5 Rice variety

Another option to reduce arsenic accumulation in rice grain is by selecting cultivars that accumulate low arsenic levels. *Japonica* cultivars included in this study (Tacuarí and Parao) were found to have on average 35% less accumulation of iAs in rice grain, in comparison to *Indica* cultivars (Olimar and El Paso 144) when grown under the same conditions. However, *Indica* varieties in this study reported significantly higher yields in relation to *Japonicas*, 9488 vs 7187 kg ha<sup>-1</sup>, respectively. Despite yield being reduced on average by 2301 kg ha<sup>-1</sup> (24%) in *Japonicas* in relation to the *Indica* cultivars, some varieties such as INIA Tacuarí do obtain a price premium related to higher quality that compensate for the lower yields.

In summary, the inorganic arsenic accumulated in rice grain in Uruguay, was found to be very low and below international limits on the two experimental sites monitored in this study. Therefore, the implementation of the mitigation management practices developed in this study are unlikely to be needed for mitigating arsenic uptake in rice, unless arsenic concentrations in areas outside the study sites were significantly different.

# **3.7 Conclusions**

Inorganic Arsenic accumulated in polished rice grain grown in the Paso Farias (PF) and Paso de la Laguna (PdL) sites were found to be below the regional (MERCOSUR, 2011) and international limits established by CODEX (FAO and WHO, 2019). Total Arsenic levels in Irrigation water and soils were found to be very low at both sites, which resulted in low levels of iAs accumulated in rice grain at these sites across the monitoring period. The relative higher levels of iAs registered at the PdL site in relation to the PF site can be associated with the higher level of tAs and bioAs in the soil at sowing and with the higher As level in the water, measured at the PdL site.

This study showed that irrigation management and varieties have the potential to affect iAs accumulation in rice grain in Uruguayan growing environments. Even though the levels of iAs accumulated in rice grain were low, this study showed that it was possible to further reduce those levels with irrigation management practices such as AWD on certain soil types and growing conditions. It was also confirmed that *Japonica* varieties accumulate lower amounts of iAs in rice grain in relation to *Indicas* across both experimental sites.

This research was conducted in two specific sites in the rice growing regions in Uruguay, and while these sites are typical for the rice growing regions of Uruguay a more extensive broader study would help provide a comprehensive picture of any likely arsenic issues. Future studies should look to perform regional scale sampling on a wide-scale across a large number of rice fields in order to further understand grain iAs levels spatially across the whole rice sector in Uruguay.

# **CHAPTER 4**

## 4. General discussion

# 4.1 Impact of implementing alternative irrigation techniques on rice

Traditional continuous flooding has been the main irrigation technique used in rice production by farmers in Uruguay. This has allowed the rice industry to achieve high ranking yields on a worldwide-scale (Carracelas et al., 2017a, 2019a; GYGA, 2019). It has also allowed the rice sector to become one of the most successful and most integrated agricultural industries in Uruguay. Rice yield in Uruguay has increased at a rate of 126 kg ha<sup>-1</sup> year<sup>-1</sup> over the past 17 years, reaching 8.3 ton ha<sup>-1</sup> in 2018-19 (Figure 4.1A).



**Figure 4.1. A.** Trend in average rice yields in Uy (2002-2019). B. Trends in cultivated rice area (ha) and total grain production in Uy (2002-2019) (DIEA MGAP, 2019).

Rice cultivated area in Uruguay reached an historical maximum of 195000 ha in 2010-11 and has been slowly declining to one of the lowest cultivated area of 145000 ha in 2018-19 (Figure 4.1B). Total rice production in the country has also declined from 1.6 million ton in 2010-11 to 1.2 million ton in 2018-19 (Figure 4.1B). This reduction in cultivated area and total rice production can be associated with the lack of profit obtained, due to increasing production costs and a reduction in rice grain price (Figure 4.2).



Figure 4.2. Trend in rice prices (USD per ton) in Uy (2002-2019). (ACA, 2019).

In order to maintain the sustainability and profitability of rice crops in Uruguay, it is important to maintain and increase rice grain yield while continuing to develop irrigation technologies that not only use less water but reduce costs. Additionally, these irrigation technologies need to maintain crop yields and quality but also maintain low levels of inorganic arsenic accumulated in rice grain. This will allow export and access to the most lucrative international markets.

The two manuscripts published during this study were the first integrated analyses of different irrigation management practices conducted in different rice growing regions of Uruguay. These studies reported the impacts of irrigation management on water productivity, grain yield, quality and accumulation of iAs in rice grain for the most common varieties cultivated.

The amount of irrigation that was applied using conventional flood irrigation practices was 10,000 m<sup>3</sup> ha<sup>-1</sup> and ranged from 7000 to 15,000 m<sup>3</sup> ha<sup>-1</sup> across the different study sites. The alternative tested irrigation technique, intermittent until panicle initiation (IP) allowed a water input saving of 25% compared to the continuous flooded systems (C). A non-significant rice yield loss of 5% was found when the soils were maintained above saturation with the IP technique. However, when soil moisture dropped below saturation, yield was found to be affected significantly, by as much as 15% with the alternate wetting and drying (AWD) technique in comparison to C.

Irrigation water productivity (WPi) was significantly increased with the implementation of intermittent irrigation techniques by 23% with IP. However, WP was not significantly improved with the AWD technique. All water productivity values reported in this study were high compared with ranges reported internationally. Average water productivity for all treatments considering only irrigation water (WPi), total with rainfall (WPir) and evapotranspiration (WPET) were 1.39 kg m<sup>-3</sup>, 0.64 kg m<sup>-3</sup> and 1.33 kg m<sup>-3</sup> respectively.

Industrial quality (white grain % and chalkiness %) was found not to be affected negatively by implementing alternative irrigation technics in all regions. However, intermittent irrigation during the entire crop cycle (I), was found to reduce significantly whole grain percentage in the North.

The inorganic arsenic accumulated in polished rice grain grown at both sites was found to be below the international limits for polished rice of 0.20 mg kg<sup>-1</sup> for iAs established by CODEX (FAO and WHO, 2019). Total Arsenic levels in irrigation water and soils were found to be very low at both sites, which explained the low levels of iAs accumulated in rice grain at these sites. The relative higher levels of iAs registered at the PdL East site in relation to the PF North site can be associated with the higher level of tAs and bioAs in the soil at sowing and with the higher As level in the water, measured at the PdL site. In summary, iAs registered levels were lower than the international and regional limits established for food safety and were also very low in comparison to other rice producing countries. This study reported the first results of arsenic level in soils, water and inorganic arsenic (iAs) accumulated in rice polished grain. Additionally, this research has shown that irrigation management and varieties have the potential to affect iAs accumulation in rice grain in Uruguay. Alternative irrigation management techniques such as alternate wetting and drying (AWD) resulted in lower levels of iAs accumulated in rice grain only at one of the evaluated sites PF in the North. Additionally, rice *Japonica* varieties were found to accumulate lower amounts of iAs in grain in relation to *Indicas*. Even though the levels of iAs accumulated in rice grain were low, it was possible to further reduce those levels with irrigation management practices such as AWD on certain soil types and by cultivating *Japonica* varieties across both experimental sites. It would be important to explore opportunities of any possible marketing advantage associated to the low levels of iAs registered in Uruguay.

The regional differences observed for irrigation use, water productivity, grain quality and iAs accumulation in grain were primarily due to differences in soil characteristics (e.g., texture, OM) and field slope. Rice in the Eastern and Central regions is cultivated on planosol and brunosol soils with low permeabilities and low OM. In contrast, soils in the Northern region are vertisols having high clay contents and steeper slopes. The latter makes it difficult to implement the alternative irrigation techniques that were investigated, in part because the shorter levee heights limit the freeboard available to capture rainfall.

This research identified potential irrigation techniques for improving WP without reducing grain yields and quality, which most likely would have a positive impact for the rice sector in Uruguay. The intermittent irrigation until panicle initiation was found to be the most promising irrigation technique to save water without penalizing grain yields and quality. In fact, some farmers in Uruguay are already implementing this technique, especially in situations where the amount of water in the reservoirs (dams) is not enough to irrigate properly the crop during the entire season and in situations to reduce pumping costs and capitalise on rainfall water.

Developing, adapting and implementing alternative irrigation technologies that use less water while preserving crop yields in Uruguay would promote the expansion of rice crop area and reduce irrigation and associated pumping costs.

# **4.1.1 Impact of implementing alternative irrigation on rice cultivated area and total rice grain production in Uruguay.**

Results from this research study may be of benefit to increase the total cultivated rice area in Uruguay. If the results of this research were widely adopted by producers, rice production in Uruguay could be expanded. For example, the 25% irrigation savings attained using IP could expand rice area by approximately 36,250 ha. This would represent an increase of approximately 0.29 Mt above and beyond the 1.20 Mt that is traded each year (Figure 4.3). Total rice area cultivated in Uruguay in 2019 was 145.000 ha with an average rice yield of 8.3 tons ha<sup>-1</sup> (Figure 4.1) (DIEA MGAP, 2019).



**Figure 4.3.** Potential increase in rice cultivated area (ha) and total production (ton) in Uy by the implementation of alternative irrigation technique (IP) considering information of season 2018-2019 and the yield potential of Uruguay. Assumptions: 25% of water input saving in IP, 5% of yield reduction with IP in relation to C, Actual yield (18/19) = 8.3 ton ha<sup>-1</sup>, Potential yield (achievable) = 11.2 ton ha<sup>-1</sup> (Carracelas et al., 2017a, b), potential cultivated area with continuous flooded = 195 000 ha (based on maximum historical value registered in Uruguay in 2010-11).

The estimated potential total grain production in the country with traditional C irrigation is 2.2 Mt considering the achievable yield potential on commercial farms of 11.2 t ha<sup>-1</sup> annually (80% of yield potential = 14 t ha<sup>-1</sup>) (Carracelas et al., 2017a, 2019a) and the highest historical cultivated area in the country of 195.000 ha, irrigated with traditional continuous flooding technique (Figure 4.1B) (DIEA MGAP, 2011). In this scenario and assuming that the economic situation turns more positive and attractive to cultivate rice in Uruguay, if IP is implemented instead of C, the total potential area to be planted with rice would increase up to a maximum of 244 000 ha increasing total rice production to 2.6 Mt ha<sup>-1</sup>. This would have a significant impact in the revenue from this sector and contribution to the economy of Uruguay.

## 4.1.2 Economic impact of implementing alternative irrigation management

Water cost in Uruguay has been based on a fixed cost per irrigated hectare with an average water cost of 214 US\$ ha<sup>-1</sup> across the last 17 year period (Figure 4.4).



**Figure 4.4.** Trend in water prices (USD) and rice grain (kg per ha), from 2002 until 2018/19 (DIEA MGAP, 2019).

From an economic point of view, in a situation where water is payed by irrigated hectare, net losses of implementing IP would be of 95 US\$ ha<sup>-1</sup> considering the non-significant 5% yield losses (average = -439 kg ha<sup>-1</sup>) recorded in this study in relation to C (average rice price = 217 US\$ ha<sup>-1</sup>, ACA, 2019). In this situation where water is charged on a per hectare basis and irrigation systems are by gravity with no pumping costs involved, 0% or no yield loss is allowed in order to not have a net income loss. In cases where it would be possible to reduce the associated irrigation costs, the reduction benefit should be higher than the net income loss of implementing IP of 95 US\$ ha<sup>-1</sup>, for a farmer to have an economic benefit of implementing this technology.

In contrast if water payment would be by volume of water used at 0.017 US\$/m<sup>3</sup>, the net economic loss by implementing IP is reduced in average across sites to 53 US\$ ha<sup>-1</sup> (Table 4.1). This estimation was calculated considering an average water input of 12500 m<sup>3</sup> ha<sup>-1</sup> (Battello et al., 2009) at 217 US\$ ton<sup>-1</sup> from 2002 until 2019 (ACA, 2019) with an average water cost of 214 US\$ per hectare (DIEA MGAP, 2019). Other factors or variable costs such as pumping cost, nitrogen fertilization and weeds control were not included in this analysis as they were beyond the scope of this research.

**Table 4.1.** Economic loss difference considering only water value cost by implementing the alternative irrigation techniques at rice price of 217 US\$ per ton. and a base water value of 0.017 US\$ per  $m^3$  for different regions.

| Region   | Irrigation | Water Input<br>m <sup>3</sup> ha <sup>-1</sup> (Wi) | Rice Yield kg<br>ha <sup>-1</sup> (Y) | Total<br>irrigation<br>water cost<br>US\$ ha <sup>-1</sup><br>(WC) | Total income<br>US\$ ha <sup>-1</sup> (I) | Economic<br>difference<br>US\$ ha <sup>-1</sup><br>(Ed) | Net Economic loss<br>difference of implementing<br>alternative irrigation<br>(Ned) |                |
|--|------------|---|---------------------------------------|--|---|---|--|----------------|
| -  | С          | 7101  | 10338                                 | 121  | 2243                                      | 2123  | US\$   | Compariso<br>n |
| East   | IP         | 6085  | 9899                                  | 103  | 2148                                      | 2045  | -78  | IP -C          |
|  | AWD        | 5034  | 8999                                  | 86   | 1953                                      | 1867  | -177   | AWD - C        |
|  | С          | 8187  | 8193                                  | 139  | 1778                                      | 1639  |  |                |
| Central  | IP         | 5847  | 7754                                  | 99   | 1683                                      | 1583  | -55  | IP -C          |
|  | Ι          | 4932  | 7709                                  | 84   | 1673                                      | 1589  | -50  | I - C          |
|  | С          | 14711   | 9050                                  | 250  | 1964                                      | 1714  |  |                |
| North  | IP         | 10578   | 8611                                  | 180  | 1869                                      | 1689  | -25  | IP -C          |
|  | Ι          | 8494  | 8566                                  | 144  | 1859                                      | 1714  | 1  | I - C          |
| AVERAGE  | С          | 10000   | 9194                                  | 170  | 1995                                      | 1825  |  |                |
| common treatments  | IP         | 7504  | 8755                                  | 128  | 1900                                      | 1772  | -53  | IP-C           |
| Formulas: WC=Wi*water.price (0.017 US\$/m <sup>3</sup> ); I=Y*grain.price (217 US\$/ton; Ed=I-WC; Ned=Ed.IP-Ed.C |            |   |                                       |  |   |   |  |                |

The net economic loss by implementing IP is highly variable across sites, ranging from -25 to -78 US dollars (Figure 4.5A). In sites like in the North, with higher irrigation water volumes used, the lowest economic loss difference was found with IP and almost no economic difference was found by implementing intermittent irrigation during the entire crop cycle.

The associated irrigation pumping, and costs reduction benefit should be higher than the average net income loss of implementing IP of -53 US\$ for a farmer to have an economic benefit of implementing IP (Figure 4.5A). Differences in this parameter were registered within regions, with higher value in the East (-78 US\$ ha<sup>-1</sup>) followed by the Central (-55 US\$ ha<sup>-1</sup>) while the lowest value was estimated for the North (-25 US\$ ha<sup>-1</sup>) (Table 4.1).



**Figure 4.5.** A. Net economic difference (US\$) of implementing IP in comparison with traditional technique C, with a base price of \$217/ton (USD) and a water value of \$0.017 m<sup>-3</sup> (USD). B. Percentage of maximum allowed yield loss with IP in order to breakeven by implementing an alternative irrigation technology.

The maximum yield loss allowed in order to not have a net income loss, with this input and output price situation, would be on average 2.4% within regions, 0.8%, 2.2% and 4.2% for East, Central and North region, respectively (Figure 4.5B).

The net income loss of implementing alternative irrigation (IP, I or AWD) in relation to the control C decreases with the increase in the water value price for all regions (Table 4.2).

**Table 4.2.** Economic difference by implementing the alternative irrigation technique

 IP in comparison with the traditional C at different water values. The economic

 difference is estimated as the difference of gross margins (Yield\*Rice price – Water

 used \*Water value) within irrigation techniques.

| Sensitivity analyses with variation in water values per m3   |                          |   |       |       |       |       |       |       |
|--|--------------------------|---|-------|-------|-------|-------|-------|-------|
| Price Va   | ariation %               | (-) 60% (-) 40% (-) 20% <b>Base</b> (+) 20% (+) 40% (+) 60% |       |       |       |       |       |       |
| Water va   | lue US\$ / m3            | 0.007   | 0.010 | 0.014 | 0.017 | 0.020 | 0.024 | 0.027 |
| Rice pri   | ce US\$ /ton             | 217   |       |       |       |       |       |       |
| Region   | Treatments<br>comparison | Economic difference by alternative irrigation technique     |       |       |       |       |       |       |
| Fast   | IP - C                   | -88   | -85   | -81   | -78   | -75   | -71   | -68   |
| East   | AWD - C                  | -188  | -185  | -181  | -177  | -174  | -170  | -167  |
| Control  | IP - C                   | -79   | -72   | -62   | -55   | -48   | -39   | -32   |
| Central  | I - C                    | -82   | -73   | -59   | -50   | -40   | -27   | -17   |
| North  | IP - C                   | -66   | -54   | -37   | -25   | -13   | 4     | 16    |
| Norui  | I - C                    | -62   | -43   | -18   | 1     | 19    | 44    | 63    |
| Average  | <b>IP - C</b>            | -78   | -70   | -60   | -53   | -45   | -35   | -28   |
| Base water price = $0.017 \text{ US}/\text{m}^3$ = 19 rice bags per ha at 10.85 US\$/bag (average 2002-2019, ACA, 2019; DIEA MGAP, 2019) and 12500 m3/ha water used with C traditional technique (Battello et al., 2009) |                          |   |       |       |       |       |       |       |

In a situation with higher volumes of water used, such as in the North, no income loss occurs from the base price for the intermittent (I) treatment while no further income loss would occur if the price of water increases up to 0.024 US\$ m<sup>-3</sup> by implementing intermittent irrigation until panicle initiation (IP). However, the extreme (low and high) prices of water tested in this analysis would have a low probability of occurrence in Uruguay (Figure 4.6 and 4.7) (Table 4.2). For this reason, a more complex approach to this analysis was considered necessary including variation within water and rice prices and the different probabilities of occurrence.

#### 4.1.3 Risk analysis

Based on the results obtained in this study, an economic analysis of the implementation for each of the alternative irrigation management strategies was conducted using an add-in to Microsoft Excel that allows analysis of possible outcomes and the probability of occurrence (@RISK, 2019). This tool allowed comparisons to be run for each alternative irrigation strategy in relation to the control continuous flooded, based on different combinations (10000 iterations) of historical

data prices of water and rice grain variations over a period of 17 years from 2002 until 2019 (Figure 4.2, 4.4). The measured irrigation water used and rice grain yield values for each irrigation treatment in this study were also considered for this analysis (Table 4.1).

Historical rice grain prices were adjusted with a normal distribution, with an average of 217 US\$ ha<sup>-1</sup> and a standard deviation of 45.1 (Figure 4.6). In order to avoid unreal extreme prices, they were truncated to a maximum and minimum price of 330 US\$ ton<sup>-1</sup> and 100 US\$ ton<sup>-1</sup> respectively based on historical information.



**Figure 4.6.** Distribution of rice grain prices (Normal) based on historical information over 17 years period from 2002 to 2019 (ACA, 2019).

According to Figure 4.6, it is expected with a 90% of probability that rice grain prices would range within 141 and 292 US\$ ton<sup>-1</sup> with an average of 217 US\$ ton<sup>-1</sup>.

In relation to water prices, a Pert distribution was adjusted accordingly to the maximum, minimum and average values registered within the 2002 to 2019 period (DIEA MGAP. 2019) (Figure 4.7).



**Figure 4.7.** Distribution of water prices per volume of water based on historical information from 2002 until 2019 (Pert distribution) (DIEA MGAP, 2019) considering an average total water use of 12500 m3 ha<sup>-1</sup> (Battello et al., 2009).

The most expected water price value is 0.017 US\$  $m^{-3}$  ranging from 0.011 up to 0.023 US\$  $m^{-3}$  (90% of probability).

The simulation results (10000 iterations considering rice and water price) for the common treatments evaluated across the three regions (IP – C) determined an average income loss of implementing IP in relation to C of -53.7 US\$ ha<sup>-1</sup>. Within 90% of probability this parameter would range from -89.5 to -17.5 US\$ ha<sup>-1</sup> (Figure 4.8).



**Figure 4.8.** Economic difference (US\$) of implementing alternative irrigation IP in relation to the traditional continuous flooding C with different price variations (water and rice grain), average of all regions (North Central and East) (result of 10000 iterations, SD: Standard Deviation).

The economic difference of implementing alternative irrigation technologies (IP, I, AWD) in relation to the control treatment continuous flooding (C), determined in most cases a loss in profitability with some variability across sites and irrigation techniques (Figures 4.9, 4.10, 4.11).

In the East region, the average net economic loss of implementing IP vs C was -78.3 (ranging from -112.1 to -44.4) (Figure 4.9A). The AWD management in this region, determined the highest net economic loss of -178 (-246 to -109), as yield was significantly penalized with this irrigation treatment (Figure 4.9B).



**Figure 4.9.** Economic difference (US\$) of implementing alternative irrigation techniques (A: IP, B:AWD) in relation to the traditional continuous flooding C with different price variations (water and rice grain), in the East region (result of 10000 iterations, SD: Standard Deviation).

In the Central region the net economic loss of implementing alternative irrigation management was lower than in the East. The average net economic loss of implementing IP vs C was -56.2 (ranging from -91.8 to -19.8) (Figure 4.10A). A

similar average net economic loss of implementing I vs C was registered in this region -50.8 (ranging from -91.6 to -8.9) (Figure 4.10B).



**Figure 4.10.** Economic difference (US\$) of implementing alternative irrigation techniques (A: IP, B: AWD) in relation to the traditional continuous flooding C with different price variations (water and rice grain), in the Central region (result of 10000 iterations, SD: Standard Deviation).
Conversely in the North region, average net economic loss of implementing IP vs C was considerably lower than the reported in the other regions with an average of -26.4, ranging from -67.5 to positive economic difference of 15.5 (Figure 4.11A). Almost no economic difference of implementing I vs C was registered in this region with an average of -1.4 (ranging from -53.5 to 51.6) (Figure 4.11B).



**Figure 4.11.** Economic difference (US\$) of implementing alternative irrigation techniques (A: IP, B: AWD) in relation to the traditional continuous flooding C with different price variations (water and rice grain), in the North region (result of 10000 iterations, SD: Standard Deviation).

In summary, in the Central and East region most likely scenarios (90% of probability) of implementing alternative irrigation management strategies are associated with a loss of profit. Conversely in the North region, the economic difference of implementing alternative irrigation management strategies could be negative or positive depending on water and rice prices variations. These simulation results would be applicable in situations where water cost is considered per volume of water used and no other irrigation costs are considered (labour, energy, pumping costs). This could happen in commercial situations where the water source is stored in reservoir, irrigation is by gravity and water competes with other alternative uses like irrigation of other crops or pastures within a farming system. Some land and water owners that cultivate their own crops could consider the results of this analysis to decide the most appropriate irrigation strategy to implement in their rice crops.

However, it is important to consider that water cost in Uruguay is based on a fixed cost per irrigated hectare not by volume of water used and currently there are no economic incentives for farmers to adopt water saving techniques in many situations. Additionally, most of Uruguayan farmers (70%) lease both the land and water so it is unlikely that water saved will be used to increase the cultivated rice land area. In this situation, the implementation of alternative irrigation techniques can threaten total rice grain production on a large-scale farm (Bouman and Tuong, 2001) as they are normally associated with yield loss. Additionally, the presence of a water layer operates like an insurance, which is important to minimize risks and potential yield reductions if there is an operational irrigation problem in the field. The implementation of alternative irrigation techniques in larger areas in commercial farms will be more challenging and higher yield losses than the average 5% measured in this study can be found when these are not implemented properly, and soils are allowed to dry down.

In the current scenario of increasing production costs, low grain prices and lacking economic incentives to adopt water saving techniques, continuous flooding from 15-20 days after emergence is likely to remain the standard adopted practice in Uruguay, unless policy incentives are put in place, such as tradable water rights or altering the way farmers are charged for irrigation water used. These would allow to reduce irrigation cost and water cost while improving the economic result for farmers that use water more efficiently. Additionally, total rice cultivated area could be increased in Uruguay. Under the current cost and price structures, the outcomes of this study are not favourable for the adoption of alternative irrigation techniques. However, these outcomes might be different if other irrigation costs such as energy cost for pumping are considered and also if economic conditions change in the future.

## **CHAPTER 5**

## 5. General conclusions and future research needs

This study identified irrigation management options that improved water productivity which was the first aim of this project. Alternative irrigation techniques to the traditional continuous flooding, like intermittent irrigation, maintained the soils under saturated conditions and allowed a reduction in irrigation water inputs and increased water productivity without penalising grain yields and quality.

Alternative irrigation techniques like intermittent irrigation in northern - central and alternate wetting and drying (AWD) in the eastern region, allowed a significant water use saving of 35%, 34% and 29%, respectively, compared to the early continuous flooded systems. Water productivity was significantly increased with the implementation of intermittent irrigation techniques by 0.25 kg m<sup>-3</sup> with IP until panicle initiation and by 0.68 kg m<sup>-3</sup> with I during all irrigation period in relation to the continuous flooded treatment. However, in alternate wetting and drying management, yield was found to be affected negatively as the soil dropped below saturation, even during the vegetative period. A yield loss of 1339 kgs (15%) was registered in AWD in relation to the traditional continuous flooded treatment.

The intermittent technique was identified in this study as the optimal irrigation management strategy that could be adapted and implemented in different soils and environments within the rice sector in Uruguay. Additionally, the chances of success of this management was higher when it was implemented only until panicle initiation. This technique is a potential viable irrigation alternative to be validated across Uruguay while alternate wetting and drying AWD would need more research in terms of timing, duration and severity of the dry period, before wide-scale adoption.

The key elements to be able to implement this technology on large commercial scale farms will be the ability to quickly flood and re-establish the layer of water during the intermittent irrigation period to achieve a uniform irrigation across the whole rice field. It is also important to consider that implementation of this management technique involves a higher risk of potentially reducing productivity during an operational irrigation problem in the field. This technique would also allow farmers to capitalise on rainfall water reducing the blue water footprint. Additionally, this alternative irrigation technique could be a valuable management option to be implemented in dry years when water stored in the reservoirs may not be enough to irrigate rice fields continuously flooded during the entire growing season.

In order to address the second aim of this study, the inorganic arsenic in rice grain was measured in two sites, under different irrigation management with two different varieties types (*Indicas* and *Japonicas*). Alternate Wetting and Drying (AWD) irrigation technique resulted in lower levels of iAs accumulated in rice grain at only one of the evaluated experimental sites. Rice variety was found to significantly affect iAs uptake and accumulation in rice grain. *Japonica* varieties accumulated lower amounts of iAs in grain in relation to *Indicas*.

The inorganic arsenic levels in the two experimental rice growing sites evaluated in Uruguay, were very low (0.07 mg.kg<sup>-1</sup>) and below the limit proposed by the international standards of 0.20 mg.kg<sup>-1</sup>. For this reason, the implementation of another alternative irrigation management such as AWD is unlikely to be needed for mitigating arsenic uptake in rice in Uruguay. Additionally, it was also found that the AWD management technique implemented during the vegetative period was not effective in significantly reducing the low levels of iAs accumulated in rice grain on certain soil types and growing conditions (PdL). Furthermore, AWD technique allowed soil moisture to drop below saturation and rice grain yield was found to be affected negatively.

Further research should validate on larger scale commercial fields, the most promising technologies identified in this study to improve water productivity. Additionally, available technologies need to be developed and adapted to Uruguay rice growing environments such as remote sensing and in-field sensing and water flow measurement devices in order to facilitate the implementation and monitoring of irrigation management in rice crops. This would contribute to the successful implementation of alternative irrigation technologies on large commercial farms, improve irrigation uniformity, maximize yields, increase water productivity and

reduce the associated irrigation cost, contributing to the sustainability of rice cultivation in Uruguay.

Remote sensing, geo-levelling and automation of rice irrigation systems will be key elements in increasing rice productivity. These technologies would facilitate the successful implementation of alternative irrigation management strategies such as those investigated in this study, would reduce issues associated with the increasing lack of labour within the rice sector in Uruguay and help reduce risk of implementing these alternate irrigation strategies.

Additional research is required to evaluate ranges of "safe" alternate wetting and drying management strategies that maintain soil water depletion in a range that does not reduce rice yields, allow an increase in water productivity and a significant reduction in iAs accumulated in grain for different soils and environments within the rice sector in Uruguay. It would be important to consider if there is any possible marketing advantage associated with the low levels of iAs in rice grain detected in Uruguay, which are well below the international limits.

Subsequent studies should focus on improving the understanding on how different levels of water stress and aerobic periods (duration, timing and severity) at different phenological crop periods impact yields and profitability. Development of monitoring techniques, such as low cost soil/crop sensors, that allow farmers to safely use these techniques is a key requirement for its successful implementation over large areas.

Rice breeding will be important to identify and develop new cultivars that tolerate non-flooded conditions without penalising rice grain yields and industrial quality while allowing to accumulate lower amounts of iAs in grain and other heavy metals.

Future studies should look to perform regional scale sampling across many rice fields in order to further understand grain As levels spatially across the whole rice sector in Uruguay.

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# **APPENDIX 1. AUTHORSHIP STATEMENT Chapter 2** 1. Details of publication and executive author

| 1. Details of publication and executive author  |   |   |  |  |  |
|---|---|---|--|--|--|
| Title of Publication  |   |   | Publication details  |  |  |
| Irrigation management strategies to increase water productivity<br>in Oryza satiya (rice) in Uruguay  |   | Agricultural Water Management 222<br>(2019) 161–172 |  |  |  |
| in Oryza sativa (nee) in Oruguay  |   | (2019) 101-172.                                     |  |  |  |
|   |   | https:/   | //doi.org/10.1016/j.agwat.2019.05.049                                  |  |  |
| Name of executive author  | School/Institute/Division if E<br>based at Deakin; Organisation<br>and address if non-Deakin  |   | Email or phone   |  |  |
| Julio Gonzalo Carracelas<br>Garrido   | School of Life and<br>Environmental Sciences  | 5   | gcarracelas@deakin.edu.au /<br>gcarracelas@inia.org.uy                 |  |  |
| 2. Inclusion of publication i   | n a thesis  |   |  |  |  |
| Is it intended to include this publication in a higher<br>degree by research (HDR) thesis? Ye   |   | Yes<br>Yes  | No If Yes, please complete Section<br>If No, go straight to Section 4. |  |  |
| 3. HDR thesis author's decl   | aration   |   |  |  |  |
| Name of HDR thesis author if<br>different from above. (If the<br>same, write "as above")  | School/Institute/Divis<br>at Deakin   | School/Institute/Division if t<br>at Deakin         |  | Thesis title   |  |
| As above  | School of Life and En<br>Sciences   | School of Life and Environmer<br>Sciences           |  | al Rice irrigation strategies effects on<br>water-productivity, grain-quality<br>and food-safety |  |
| intellectual content, etc.)<br>Conception of the project, design of methodology and experimental protocol, running experiments, sample collection, data collection and analysis, writing the paper, drafting the manuscript, revising and editing the manuscript for submission |   |   |  |  |  |
| <i>I declare that the above is an accurate description</i><br><i>of my contribution to this paper, and the</i><br><i>contributions of other guthers are as described</i>  |   |   | ature<br>date  | October 14 <sup>th,</sup> 2019   |  |
| below.  |   |   |  |  |  |
|   |   |   | Signature Redacted by Library  |  |  |
|   |   |   |  |  |  |
| 4. Description of all author contributions  |   |   |  |  |  |
| Name and affiliation of author  | Contribution(s) (for example, conception of the project, design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.) |   |  |  |  |
| John Hornbuckle   | Professor supervisor. Revising the manuscript critically for drafting and final submission.   |   |  |  |  |
| Juan Rosas  | Statistical analysis advising, revising the statistical analyses description in the manuscript  |   |  |  |  |
| Alvaro Roel   | Conception of the project, design of methodology and experimental protocol, revising the manuscript   |   |  |  |  |

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I agree to be named as one of the authors of this work, and confirm:

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that the description in Section 4 of my contribution(s) to this publication is accurate, that the data on which these findings are based are stored as set out in Section 7 below. If this work is to form part of an HDR thesis as described in Sections 2 and 3, I further consent to the incorporation of the publication into the candidate's HDR thesis submitted to Deakin University and, if the higher degree is awarded, the subsequent publication of the thesis by the university (subject to relevant Copyright provisions).

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| John Hornbuckle |                 | 15-10-2019 |
|                 |                 |            |
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| Juan Rosas      | red             | 14-10-2019 |
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## **APPENDIX 2. AUTHORSHIP STATEMENT. Chapter 3.**

| 1. Details of publication and   | executive author  |   |   |   |  |
|---|---|---|---|---|--|
| Title of Publication  |   |   | Publication details                                   |   |  |
| Irrigation management and variety effects on rice grain Arsenic levels in Uruguay.  |   | Journal of Agriculture and Food Research.<br>https://doi.org/10.1016/j.jafr.2019.100008 |   |   |  |
| Name of executive author  | School/Institute/Division if<br>based at Deakin; Organisation<br>and address if non-Deakin  |   | Email or phone  |   |  |
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|   |   |   |   |   |  |
| 2. Inclusion of publication in  | 1 a thesis  | Vac   | /No   | If Vac along complete Castion 2   |  |
| by research (HDR) thesis?   | ication in a higher degree  | Yes   | / 100   | If No, go straight to Section 4.  |  |
| 3. HDR thesis author's decla  | aration   |   |   |   |  |
| Name of HDR thesis author if<br>different from above. (If the same<br>write "as above")   | nesis author if<br>School/Institute/Division if<br>at Deakin  |   | ased Thesis title                                     |   |  |
| As above  | School of Life and En<br>Sciences   | School of Life and Environmental<br>Sciences  |   | Rice irrigation strategies effects on<br>water-productivity,<br>grain-quality and food-safety |  |
| If there are multiple authors, give a full description of HDR thesis author's contribution to the publication (for example, how much did you contribute to the conception of the project, the design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.)<br>Conception of the project, design of methodology and experimental protocol, running experiments, sample collection data collection and analysis, writing the paper drafting the manuscript revising and editing the |   |   |   |   |  |
| manuscript for submission<br><i>I declare that the above is an accurate description of</i> Signature October 14 <sup>th</sup> , 2019  |   |   |   |   |  |
| <i>my contribution to this paper, and the contributions of other authors are as described below.</i>  |   | and   | date  |   |  |
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| 4. Description of all author contributions  |   |   |   |   |  |
| Name and affiliation of author  | Contribution(s) (for example, conception of the project, design of methodology or experimental protocol, data collection, analysis, drafting the manuscript, revising it critically for important intellectual content, etc.) |   |   |   |  |
| John Hornbuckle   | Professor supervisor. Revising the manuscript critically for drafting and final submission.   |   |   |   |  |
| Melissa Verger  | Melissa Verger Laboratory analysis. lab. methods description and revising the manuscri  |   |   | escription and revising the manuscript  |  |
| Raquel Huertas  | Laboratory analysis and lab. methods description  |   |   | s description   |  |
| Sara Riccetto   | Sample collection and   | Sample collection and revising the manuscript   |   |   |  |
| Federico Campos   | Revising the manuscript and added content, references to the discussion   |   |   |   |  |
| Alvaro Roel         Conception of the project, design of methodology and experimental protocol, revising the manuscript   |   |   |   |   |  |

## 5. Author Declarations

I agree to be named as one of the authors of this work, and confirm: that I have met the authorship criteria set out in the Deakin University Research Conduct Policy,

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## **APPENDIX 3.** Other Published articles (Summary of paper 1)

## MANEJO DEL RIEGO Y PRODUCTIVIDAD DEL AGUA EN EL CULTIVO DE ARROZ EN URUGUAY

## <u>G. Carracelas</u><sup>1</sup>, J. Hornbuckle <sup>2</sup> J. Rosas<sup>4</sup> y A. Roel<sup>3</sup>

#### PALABRAS CLAVE: Intermitente, AWD, Rendimiento

#### 1. INTRODUCCIÓN

La mayor parte del agua utilizada para regar el arroz se bombea (56%) en Uruguay y las represas construidas con fines de riego son la principal fuente de agua (54%) (DIEA MGAP, 2017). Manejos de riego alternativos podrían determinar una mejora en el resultado económico de la actividad por un ahorro en los costos asociados al riego (mano de obra, costos de bombeo, entre otros). En años de seguía, el agua almacenada en las represas puede no ser suficiente para regar el cultivo de forma continua durante todo su ciclo. Más agua disponible, permitiría expandir el área de arroz o permitiría regar otros cultivos de cereales y pasturas, reduciendo el riesgo mediante la diversificación de productos. Aumentar los rendimientos y mantener la calidad industrial del grano de arroz, mientras se reduce el gasto de agua, es un gran desafío para el sector arrocero. Los altos costos del cultivo sumado a los bajos precios de comercialización del grano dificultan la implementación de nuevas alternativas de riego que estén asociadas a un mayor riesgo por pérdidas de rendimiento o menor calidad de grano. El objetivo principal de este trabajo es determinar técnicas de manejo del riego que aumenten la eficiencia y la productividad del agua, sin afectar negativamente el rendimiento y calidad del grano. La productividad del agua de riego se define como los kilogramos de arroz por m<sup>3</sup> de agua de riego (WPi) (Bouman et al., 2007).

## 2. MATERIALES Y MÉTODOS

Este documento es un resumen de un análisis integrado de 10 experimentos de riego realizados durante un periodo comprendido entre 2009 y 2015, en suelos típicos de cada región arrocera del Uruguay (Carracelas et al., 2019). Los tratamientos de riego evaluados en todas las regiones fueron: Inundación continua tradicional (C) e Intermitente hasta primordio (IP). Intermitente durante todo el ciclo de cultivo (I) solo en la región Norte-Centro y AWD en el Este. (Figura 1).

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Figura 1. Tratamientos de riego evaluados en diferentes regiones arroceras del Uruguay.

En el tratamiento C, la inundación se mantuvo con una lámina de agua de 10 cm durante todo el ciclo de cultivo.

En el tratamiento IP e I, la lámina de agua se dejaba resumir alternando entre 0 cm y10 cm y se restablecía cuando el suelo aún estaba saturado (barro liquido).

El tratamiento AWD permitía que el suelo se secara con un agotamiento del agua del 50% del agua disponible, a partir del cual se volvía a saturar, alternando suelo seco y saturado durante el periodo vegetativo, hasta primordio.

## 5. RESULTADOS DE LA INVESTIGACIÓN

El gasto de agua de riego promedio fue de 7886 m<sup>3</sup> ha<sup>-1</sup> y el gasto total de agua de riego incluyendo la lluvia fue de 14656 m<sup>3</sup> ha<sup>-1</sup> en el tratamiento de inundación continua. Las diferentes técnicas alternativas de riego evaluadas permitieron un ahorro significativo en el gasto de agua en relación con el riego continuo (Figura 2A). En la región Norte, los manejos intermitentes determinaron ahorros de agua de riego, del 28% en IP y del 42% en I, en relación con C. En la Región Central, dichos manejos (IP, I) permitieron un ahorro significativo de agua de riego en promedio del 34% en relación con el control C. En la región Este, el manejo AWD determinó una reducción significativa del gasto de agua del 29% en relación con C.





Figura 2. A. Gasto de agua de riego y total (riego + lluvias) y B. Productividad del agua de riego (kg m<sup>-3</sup>) para los manejos de riego y regiones.

La productividad promedio del agua para todos los tratamientos considerando solo agua de riego fue de 1.39 kg m<sup>-3</sup> (Figura 2B). Los manejos de riego alternativos determinaron aumentos significativos de este parámetro en todas las regiones. El registro más alto de productividad de agua se obtuvo con el tratamiento I, con un valor de 1.8 kg m<sup>-3</sup> en el Centro.





Figura 3. A. Rendimiento en grano de arroz (kg ha<sup>-1</sup>, 14% de humedad), B. Calidad Industrial del grano (Entero %) para cada tratamiento de riego y regiones arroceras.

En relación con el rendimiento, no se registraron diferencias significativas entre los tratamientos C, IP e I. El tratamiento AWD, resultó en una reducción significativa del rendimiento de 1339 kg de arroz ha<sup>-1</sup> (15%) en relación con C (Figura 3 A). La calidad industrial, porcentaje de grano entero no se vio afectado negativamente en las regiones Este y Centro. Sin embargo, en la región Norte, el riego intermitente I, determinó una reducción significativa en este parámetro cercana al 6% en relación con C (Figura 3 B).

Se destaca la técnica de riego intermitente hasta el inicio de primordio que permitió un ahorro importante del gasto de agua (25%) y un aumento significativo en la productividad del agua de riego (23%), sin afectar el rendimiento de arroz y la calidad industrial del grano. Los resultados se obtuvieron en parcelas experimentales donde el riego es fácil de manejar. El éxito en la implementación exitosa de manejos alternativos de riego, con amplias capacidades de flujo de entrada de agua que permita una inundación rápida y uniforme de la chacra. A su vez se requiere de un programa integrado de manejo agronómico, control de malezas, fertilización y enfermedades acorde al nuevo manejo (Massey et al., 2014). La implementación será limitada a menos que exista un incentivo económico como reducción de los costos de bombeo del riego, costo de la energía, y también de una reducción del costo total del agua. En el escenario actual de altos costos de producción, bajos precios de los granos

y sin incentivos económicos, el riego continuo con inundaciones tempranas seguirá siendo la práctica de mayor adopción en Uruguay para la concreción del alto potencial de rendimiento del cultivo.

## **3. CONCLUSIONES**

Este estudio identificó técnicas de riego (IP), que utilizaron significativamente menos agua de riego al tiempo que mantuvieron el rendimiento de arroz sin afectar la calidad industrial y, por lo tanto, aumentaron la productividad del agua de riego en una amplia gama de ambientes de cultivo de arroz irrigado en Uruguay.

Las técnicas de riego que mantuvieron el suelo siempre saturado (IP, I) permitieron una reducción del gasto de agua sin afectar negativamente el rendimiento del arroz, lo que determinó un aumento significativo en la productividad del agua. Sin embargo, el riego intermitente durante todo el ciclo del cultivo redujo significativamente el porcentaje de granos Enteros en el Norte. Cuando el suelo se seca al implementar la técnica AWD, el rendimiento fue afectado negativamente.

Investigaciones futuras deberán validar y adaptar estas tecnologías para ser implementadas con éxito en chacras comerciales. A su vez, es necesario evaluar diferentes estrategias de secado en AWD que mantengan el agotamiento del agua del suelo en niveles que no afecten el rendimiento y calidad de arroz.

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**APPENDIX 4.** Graphical Abstract of paper 2



**APPENDIX 5.** Literature review diagram