Title: Impacts of soil incorporation of pre-incubated silica-rich rice residue on soil biogeochemistry and greenhouse gas fluxes under flooding and drying

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Abstract

Incorporation of silica-rich rice husk residue into flooded paddy soil decreases arsenic uptake by rice. However, the impact of this practice on soil greenhouse gas (GHG) emissions and elemental cycling is unresolved particularly as amended soils experience recurrent flooding and drying cycles. We evaluated the impact of pre-incubated silica-rich rice residue incorporation to soils on pore water chemistry and soil GHG fluxes (i.e., CO₂, CH₄, N₂O) over a flooding and drying cycle typical of flooded rice cultivation. Soils pre-incubated with rice husk had 4-fold higher pore water Si than control and 2-fold higher than soils pre-incubated with rice straw, whereas the pore water As and Fe concentrations in soils amended with pre-incubated straw and husk were unexpectedly similar (maximum ~0.85 μM and ~450 μM levels, respectively). Pre-incubation of residues did not affect Si but did affect the pore water levels of As and Fe compared to previous studies using fresh residues where straw amended soils had higher As and Fe in pore water. The global warming potential (GWP) of soil GHG emissions decreased in the order straw (612±76 g CO₂-eq m⁻²) > husk (367±42 g CO₂-eq m⁻²) > ashed husk = ashed straw (251±26 and 278±28 g CO₂-eq m⁻²) > control (186±23 g CO₂-eq m⁻²). The GWP increase due to pre-incubated straw amendment was due to: a) larger N₂O fluxes during re-flooding; b) smaller contributions from larger CH₄ fluxes during flooded periods; and c) higher CH₄ and CO₂ fluxes at the onset of drainage. In contrast, the GWP of the husk amendment was dominated by CO₂ and CH₄ emissions during flooded and drainage periods, while ashed amendments increased CO₂ emissions particularly during drainage. This experiment shows that ashed residues and husk addition minimizes GWP of flooded soils and enhances pore water Si compared to straw addition even after pre-incubation.

47 **Keywords**

48 arsenic; carbon dioxide; methane; nitrous oxide; silicon; soil flooding; soil management

49

1. Introduction

The management of rice paddy soils influences global greenhouse gas (GHG) emissions (Delwiche and Cicerone, 1993; Neue, 1997), nutrient dynamics (Kaewpradit et al., 2008; Penido et al., 2016; Seyfferth et al., 2013), and arsenic (As) availability to rice (Arao et al., 2009; Li et al., 2009; Linquist et al., 2015; Seyfferth and Fendorf, 2012; Seyfferth et al., 2016). As a staple food for over 50% of the global population, rice yield is directly linked to global food security. The highest rice yield is typically obtained under flooded paddy cultivation in which soils are flooded and remain so until grain filling. The timing of the drainage impacts both As uptake by rice and soil GHG emissions. (Adviento-Borbe et al., 2015; Kim et al., 2012; Linquist et al., 2012; Linquist et al., 2015). While rice paddy flooding is beneficial for yield, it exacerbates both CH₄ emissions and As mobilization in soils due to anaerobic metabolic reactions under reduced soil conditions (Linquist et al., 2012; Linquist et al., 2015). The mobilized As, in turn, may be taken up by rice (Ma et al., 2008) and transferred to grain (Carey et al., 2010), where it can compromise yield (Duxbury et al., 2003; Panaullah et al., 2009) and impact human health (Banerjee et al., 2013; Williams et al., 2005) upon consumption. Decreasing As uptake is a primary local-to-global food security goal, but sustainable solutions must consider its impacts on yield and its biogeochemical implications such as short- and long-term GHG emissions.

Soil incorporation of silica is an emerging method to decrease toxic As uptake by rice and storage in grain (Seyfferth et al., 2016). Silica affects rice As concentrations because dissolved silica (silicic acid) and reduced arsenic (arsenous acid) are chemically similar and share root transporters (i.e., Lsi1 and Lsi2) in rice (Ma et al., 2008). Due to weathering and leaching, many rice soils are depleted in plant-available silica (Savant et al., 1997a), which leads

to upregulation of Si transporters in rice roots and inadvertently increases the potential for As uptake. Addition of silica to soil may decrease As uptake by downregulation of Si transporters and competition between Si and As for uptake (Ma et al., 2008). However, Si addition may also cause As desorption from soil solids (Luxton et al., 2006), stabilization of poorly crystalline Fe (oxyhydr)oxides in soil and on root-bound Fe (oxyhydr)oxide plaque (Schwertmann and Thalmann, 1976), and co-precipitation with As-adsorbing Fe (oxyhydr)oxides in bulk soil and in plaque (Swedlund and Webster, 1999). The overall impact of Si on As in rice reflects an interplay of the aforementioned plant physiological and soil chemical processes.

Soil silica amendments can affect grain As concentrations and GHG emissions, but the impacts depend on the type of silica added (Ma et al., 2014; Penido et al., 2016; Seyfferth and Fendorf, 2012; Seyfferth et al., 2016). Under typical flooded paddy cultivation, soil incorporation of silica gel decreases grain As concentrations without affecting soil CH₄ production or rice yield (Li et al., 2009; Seyfferth and Fendorf, 2012), whereas silica-rich diatomaceous earth increases grain As concentrations (Seyfferth and Fendorf, 2012). Soil incorporation of silica-rich rice straw increases grain As (Ma et al., 2014) and exacerbates soil CH₄ production (Penido et al., 2016) and emissions (Liu et al., 2014; Vibol and Towprayoon, 2010; Wang et al., 2012; Wassmann et al., 2000). In contrast, soil incorporation of silica-rich rice husk, a holistic yet seldom used soil amendment, decreases inorganic grain As levels by 25-50% without affecting yield or CH₄ production (Seyfferth et al., 2016). Far fewer studies have focused on multiple GHG emissions and As cycling due to rice husk incorporation than due to rice straw incorporation. To evaluate the impact of husk residue incorporation on GHG emissions and compare it to other silica-rich amendments, research efforts need to consider the

95 impact of husk amendment on soil biogeochemistry for multiple flooding and drying events and
96 CH₄, CO₂ and N₂O emissions simultaneously.

97 Factors that promote CH₄ emissions include strongly reduced soil conditions and labile
98 carbon availability, whereas N₂O emissions are promoted under suboxic soil conditions and in
99 response to NO₃⁻ fertilization (Zou et al., 2005). Organic residue incorporation such as rice straw
100 is known to increase CH₄ production and efflux (Conrad et al., 2012; Weber et al., 2001).
101 Flooded rice fields, particularly those amended with organic residues, are thus major sources of
102 CH₄ emissions and comprise between 9 and 13% of global anthropogenic emissions (Pedersen et
103 al., 2006). While CH₄ emissions tend to dominate in flooded rice production (Wassmann et al.,
104 2000), periods of suboxic soil conditions such as those that occur during drainage may increase
105 N₂O emissions (Adviento-Borbe et al., 2015). Moreover, the management history (e.g.,
106 incorporation of organic matter such as plant residues) of the soil also affects the microbial
107 community structure, which in turn affects GHG production and fluxes (Lagomarsino et al.,
108 2016).

109 We previously reported that rice husk incorporation to flooded soil led to higher
110 dissolved silica, lower dissolved As, and lower dissolved CH₄ concentrations than rice straw
111 incorporation in a six-week flooded incubation study without plants (Penido et al., 2016). That
112 study utilized a 1% residue:soil ratio, which was designed to provide silica benefits for multiple
113 growing seasons and thus recurrent flooding and drying cycles. The findings from that work
114 open the following question: *To what extent does pre-incubation under flooded conditions affect*
115 *biogeochemical cycling and GHG emissions when amended soils are re-flooded and again*
116 *dried?* This question is relevant to evaluate how biogeochemical processes of amended soils
117 respond to multiple flooding-drying cycles, and to inform long-term soil management practices.

To address that question, we experimentally tested the influence of soil incorporation of pre-incubated silica-rich rice residues of straw, husk, ashed husk and ashed straw over dynamic soil moisture conditions. We simulated a flooded rice cultivation cycle that consists of a wetting up-flood-drainage cycle of the approximate length of a growing season. We conducted the experiment under laboratory conditions (i.e., controlled temperature) without plants to isolate the amended soil impacts and avoid confounding effects. We monitored biogeochemical dynamics in pore water and CO₂, CH₄, and N₂O fluxes, and calculated the 100-year global warming potential (GWP) of these GHG emissions. We hypothesized that soil incorporation of pre-incubated 1) rice husk would lead to the highest pore water Si concentrations and lower dissolved As and GWP than rice straw; 2) rice straw would lead to the highest pore water As concentrations and GWP, mainly as CH₄, due to strongly reduced soil conditions; and 3) ashed residues would lead to the lowest GWP because these residues have more recalcitrant carbon.

2. Materials and methods

2.1 Soil sampling and experimental set-up

Soils and treatments were identical to those described in Penido et al. (2016). Briefly, air-dried soil collected from 2 – 30 cm depth (to avoid collection of overlying grass vegetation) at the University of Delaware's Newark farm, Newark DE, USA was used for the experiments. This soil was chosen because of its similar weathering extent, plant-available silica and arsenic concentration to rice paddy soils in Cambodia (Seyfferth et al., 2014). Soil characteristics were reported in Penido et al. (2016) and are briefly summarized in Table 1. During collection, care was taken to remove vegetation and roots while keeping the soil structure

as intact as possible. Approximately 3.5 kg soil was added to each of 15 acid-washed, 4 L high-density polyethylene pots.

For treatments, 1% (w/w) of rice residues were incorporated by hand into the soil; these consisted of rice husk, rice straw, ashed husk, or ashed straw. Both husk and straw were ground to a powder using a Wiley Mill prior to soil incorporation. The total As, Si, Fe and P was < 0.02, 4028, 6.7, and 10.8 mmol kg⁻¹ for husk and 0.11, 2128, 15.1, and 33.9 mmol kg⁻¹ for straw (Penido et al., 2016). The C content of both husk and straw is ~36% and the N content is ~0.45% for husk and ~0.6% for straw (Kajiura et al., 2015). Ashed husk residues were obtained from a mill in Battambang, Cambodia where fresh husk is used as a fuel source and incompletely combusted at ~450° C, as is done in some rice-growing countries (Haeefe et al., 2009; Savant et al., 1997b). Ashed straw was created in the laboratory by combustion of the fresh straw at 450° C in a muffle furnace. The creation of the ashes concentrated As, Si, Fe, and P by at least a factor of two, but it should be noted that these are not true ashes as they represented incompletely combusted materials that were a mixture of ash and charred material. Each treatment and non-amended control was replicated three times for a total of 15 pots. Prior to the start of the experiment soils were amended, flooded for 6 weeks (Penido et al., 2016), and then allowed to air-dry at room temperature (22°C) for over two months. This allowed us to test our main question on amended soils that have been exposed to flooding and drainage for more than one cycle (i.e., pre-incubation) as expected in actively managed rice paddies.

To monitor pore water chemistry, Rhizon pore water sampling devices (Eijkelkamp Soil & Water, The Netherlands) were inserted into a pre-drilled hole approximately 1 cm from the top rim of each pot. The 10-cm long Rhizon samplers, with 0.2 µm pore size, penetrated the soil at a 45 degree angle to allow for weekly pore water sampling (Penido et al., 2016; Seyfferth and

Fendorf, 2012; Seyfferth et al., 2016). To ensure a tight seal for GHG measurements, each port was sealed with silicone adhesive and allowed to dry prior to manipulation of soil moisture.

2.2 Soil moisture manipulation

Soil moisture was manipulated to approximate the dynamic conditions experienced during traditional flooded rice cultivation, which lasts approximately 90-120 days from seed to harvest. The soil moisture regime consisted of three distinct phases: wetting up (Phase I, initial dry to 100% soil moisture over 2 d), flood (Phase II, soil moisture 100% for 60 d) and dry down (Phase III, soil moisture < 100% and continually decreasing over 25 d). In Phase I, the pots were flooded using double-deionized 18 M Ω -cm (DDI) water delivered with an irrigation system to ensure simultaneous wetting of each pot. During this time, each pot received 542 mL of water during each wetting every 12 hours for the first two days of the experiment, and a total of 2.147 liters, the level of which was approximately 5 cm below the rim of the pot and at least 5 cm above the soil surface.

Pots remained flooded for six weeks, and were then allowed to dry down over another six-week period. To maintain water levels during the six-week flooded period accounting for evaporation, DDI water was added weekly (< 100 mL) after GHG flux measurements, moisture measurements, and pore water samples were taken. After six weeks of flooding, no additional water was added during the six-week dry down period. The experiment was carried out at room temperature (i.e., 22°C) to avoid confounding effects with changes in air temperature.

2.3 Pore water sampling and analysis

To measure impacts of amendment and soil moisture on pore water chemistry and controls on gas fluxes, pore water was collected 1-2 times per week through Rhizon samplers into acid-washed vials via a needle stop-cock assembly (Penido et al., 2016; Seyfferth and Fendorf, 2012; Seyfferth et al., 2016). Briefly, empty and pre-cleaned vials were capped with septa and crimp sealed in an oxygen-free atmosphere and evacuated with a hand-held vacuum pump just prior to sampling. The septa of evacuated vials were pierced with needles that were attached to the Rhizon samplers with luer fittings. Once the stop-cock was opened, pore water flowed into the vial under an inert N₂ atmosphere. Pore water samples were taken after each gas flux and soil moisture measurement and before the water additions. Each pore water sample was immediately analyzed for Eh using a platinum electrode (Thermo/Orion) with a built-in silver/silver chloride reference and Eh values are reported relative to the standard hydrogen electrode. The sample was then analyzed for pH using a calibrated probe (Thermo/Orion), total Fe, Si, and P using ICP-OES (Thermo) and As using ICP-MS (Agilent 7500) against known standards after acidification to 2% acid with trace metal grade HNO₃. For metal and nutrient analysis, quality assurance and quality control protocols included duplicate measurements, blanks, and standard checks every 20 samples.

2.4 Soil gas flux, temperature and moisture measurements

Soil gas fluxes of CO₂, CH₄, and N₂O were measured using a closed dynamic flux chamber (Pumpanen et al., 2004) coupled with a Picarro G2508 analyzer (Picarro Inc., Sunnyvale, CA, USA) (Petrakis et al., 2017). The Picarro G2508 analyzer measures gas concentrations for CO₂ (0.02-2 μmol mol⁻¹ ± 0.05%), CH₄ (0.5-15 μmol mol⁻¹ ± 0.05%), and N₂O (0-400 μmol mol⁻¹ ± 0.05%). The instrument was calibrated using factory standards that included

high precision gases and tests against other similar units (Picarro Inc., Sunnyvale, CA, USA). Prior to each gas flux measurement, surface soil/water temperature was obtained using an infrared handheld thermometer (8500H, NuBee, USA). During each gas flux measurement, each pot was sealed to create a closed dynamic loop system to measure changes in gas concentration for a total of 5 min, which included a dead band of the first 2 min and a 3 min observation time for calculations (see the following section). After each measurement, the instrument was allowed to re-equilibrate with laboratory atmospheric concentrations between 2-5 min (i.e., CO₂ <450 ppm; CH₄ <2 ppm; and N₂O <1 ppm,) prior to measuring the next pot. Following each gas flux measurement, soil moisture was measured using a Delta-T SM150 sensor (Delta-T Devices, Cambridge, UK); however, when soils were completely flooded, 100% soil moisture was assumed.

Gas flux measurements were taken throughout Phases I, II, and, III, with more frequent measurements during Phases I and III when rapid changes in soil moisture occurred due to soil wetting up and drying down (Kim et al., 2012). During Phase I, an initial measurement was taken on dry soil prior to any water additions, and then every 2-3 hours during days 1-2 in the rewetting period. During Phase II, measurements were taken three times per day on day 3 (once flooding was achieved), once per day on days 4 through 6 (first days of completely flooded conditions), and weekly thereafter (constant flooding). During the latter part of Phase II and during Phase III, gas fluxes were measured twice per week for the last 4 weeks of the drying phase.

2.5 Soil gas flux calculations

Measured gas concentrations were used to calculate gas fluxes of CO₂, CH₄, and N₂O (in μmol m² s⁻¹), using the following equation (Steduto et al., 2002):

$$\text{Soil GHG Flux} = \frac{\delta c}{\delta t} \frac{V}{S} \frac{Pa}{RT} \quad \text{Equation 1}$$

where c is the mole fraction of CO₂, CH₄, or N₂O in μmol mol⁻¹, t is the observation time of 180 s (3 minutes after removing a dead band of 2 minutes), V is the total volume of the closed system (0.00163 m³), S is the surface area of the soil in the pot (0.03 m²), P_a is the atmospheric pressure inside the chamber (100.96 kPa), R is the universal gas constant (8.3×10⁻³ m³ kPa mol⁻¹ K⁻¹), T is the surface water or surface soil temperature (°K), and we applied a linear fit to calculate $\delta c/\delta t$ (Pearson et al., 2016). For QC, only slopes from the linear regression that were significantly different from zero ($P < 0.05$) were used to compute fluxes (Petrakis et al., 2017).

2.6 Data analysis

We report instantaneous GHG emissions and GHG emissions for the entire length of the experiment (i.e., 90 days) to describe instantaneous and then cumulative treatment effects. We calculated the GWP for each treatment and control over the entire experiment using the average of individual (CO₂, CH₄, and N₂O) GHGs and the conservative 100-yr GWPs of 265 for N₂O and 28 for CH₄ relative to CO₂ that do not include climate-carbon feedbacks (Myhre et al., 2013) to express emissions as g CO₂ equivalent units per m² (g CO₂-eq m⁻²). Consequently, the GWP are reported as the sum of CO₂, CH₄, and N₂O emissions expressed in g CO₂-eq m⁻² for the length of the experiment (i.e., 90 days).

We analyzed GHG fluxes and the GWP per phase (i.e., wetting up, flood, and drying down) to provide information on temporal dynamics and influence of wetting-drying conditions. These analyses only included data obtained and duration of time for individual phases, where

Phase I included data from $t = 0 - 0.14$ wk, Phase II included data from $t = 0.154 - 8.32$ wk, and Phase III included data from $t = 9.32 - 12.74$ wk. The GWP for each treatment and control was computed as described above but for each individual phase.

We verified that all data from each treatment and control met quality control protocols, were normally distributed, and variance was homogeneous. One-way ANOVAs and post-hoc Tukey tests were performed to find differences in the overall GWP among treatments. Two-way repeated measurements ANOVAs were conducted to evaluate treatment and temporal (i.e., Phase) effects and their interaction on response variables i.e., GHG fluxes, GWP, pore water Eh and pH, and pore water concentrations of Si, As, Fe, and P. All statistical tests were performed with SPSS v. 23 software.

3. Results

3.1 Soil moisture, redox and pH dynamics

Soil moisture, Eh, and pH were similar among treatments and control and changed through time due to dynamic water regime across Phases I, II, and III (Fig. 1). Soil moisture sharply increased from 0% to 100% within the first two days during Phase I and remained at 100% through Phase II and the first part of Phase III until it decreased to 15-20% (Fig. 1A). Eh showed an opposite pattern to soil moisture and decreased from an initial 120-160 mV during Phase I to a minimum of 40 – 100 mV, during Phase II before increasing to levels approaching 200 mV during Phase III (Fig. 1B); however, no significant differences were observed among treatments. pH values did not significantly differ among treatments and control but differed with Phase, ranging from 6.8 to 7.2 for Phases I and II before dropping to significantly lower values ($F = 26.6$; $p < 0.001$) as low as 6.3 as the soils dried (Fig. 1C).

3.2 Pore water Fe, As, Si and P dynamics

Statistically significant differences in pore water chemistry due to treatment were evident with Fe, As, Si, and P concentration dynamics in pore water (Fig. 2). While the pattern of an initial increase during Phase I, leveling off during Phase II, and decrease during Phase III was consistent for all four solutes, the magnitude of change differed among the treatments for each solute.

The maximum pore water Fe in husk and straw was as high as $\sim 450 \mu\text{M}$ and was 2-fold higher than the other treatments and control (Fig. 2A). These results are supported by a two-way repeated measurements ANOVA that showed significant differences by treatment ($F = 9.7$, $P < 0.0001$), phase ($F = 4.5$, $P = 0.035$), and their interaction ($F = 5.4$, $P < 0.0001$). Post hoc tests revealed that both straw and husk treatments had significantly higher ($p < 0.05$) average dissolved Fe in pore water compared to ashed husk, ashed straw and non-amended control. However, the husk treatment lagged behind the straw treatment during both the increase in Phase I and the decrease to nondetectable levels in Phase III (Fig. 2A).

Pore water As concentrations were initially $\sim 0.2 \mu\text{M}$ for amended soils and non-amended controls, but thereafter deviated from each other (Fig. 2B). Pore water As in non-amended soil increased to $0.4 \mu\text{M}$ during Phase II before decreasing to $< 0.05 \mu\text{M}$ during Phase III. Both ashed straw and ashed husk had similar patterns of pore water As, which showed an increase to the highest levels of $\sim 0.65 \mu\text{M}$ by week 2 before slowly decreasing throughout the experiment. Straw amendment also showed a rapid increase to $\sim 0.65 \mu\text{M}$ during the first two weeks, but remained at this level to week 7 and then increased to the highest value of $\sim 0.85 \mu\text{M}$ by week 9 before decreasing rapidly to $< 0.05 \mu\text{M}$ during Phase III. In contrast, pore water As in husk

amended soils exhibited a gradual increase to $\sim 0.85 \mu\text{M}$ from weeks 1-8 and after which lagged behind straw as it decreased to $< 0.05 \mu\text{M}$ in Phase III (Fig. 2B). These results are supported by a two-way repeated measurements ANOVA that showed significant differences by treatment ($F = 5.5$, $P < 0.0001$), phase ($F = 45.1$, $P < 0.0001$), and their interaction ($F = 2.7$, $P = 0.032$). Post hoc tests revealed that the average pore water As was significantly higher ($p < 0.05$) in husk and straw treatments compared to control.

Pore water Si concentrations varied considerably with treatment (Fig. 2C). Non-amended control soils had the lowest Si levels of $\sim 200 \mu\text{M}$ throughout the experiment. Amended soils had similar patterns of an initial increase in pore water Si during Phase I and a relatively steady level during Phase II before dropping off during the latter part of Phase III, but varied in magnitude among treatments. Pore water Si was the highest in husk amended soils, ranging $\sim 600 - 900 \mu\text{M}$, followed by ashed husk, ranging $320 - 500 \mu\text{M}$, while straw and ashed straw had similar low levels of between 250 and $350 \mu\text{M}$ (Fig. 2C). These results are supported by a two-way repeated measurements ANOVA that showed significant differences by treatment ($F = 258.4$, $P < 0.0001$) and phase ($F = 485.9$, $P < 0.0001$) but not their interaction ($F = 1.6$, $P = 0.163$). Post hoc tests revealed that the average pore water Si was decreased in the following order husk > ashed husk > straw = ashed straw > non-amended control.

Pore water P initially increased during Phase I, reached a relatively steady level during Phase II and a sharply decreased during Phase III in all treatments. Similar to Fe and As trends, pore water P in husk amended soil showed a lag compared to straw amended soil in which levels were lower than in straw amended soils until week 4 and then increased to higher levels in weeks 8-10 before returning to low levels at the end of Phase III (Fig. 2D). However, in contrast to Fe, As and Si levels, pore water P concentrations were higher in ashed amendments than in fresh

amendments (Fig. 2D). Ashed husk and ashed straw had similar and relatively high initial concentrations of 40 μM that increased to nearly 60 μM by week 4 before they slowly decreased to ~45 μM by week 10 and sharply decreased to ~25 μM at the end of Phase III. Non-amended control and straw amended soils had similar pore water P levels that were initially 25 μM and increased to ~40 μM by week 4 and remained steady until they sharply decreased at the end of Phase III. These results are supported by a two-way repeated measures ANOVA that showed significant differences by treatment ($F = 19.6$, $P < 0.0001$) and phase ($F = 25.2$, $P < 0.0001$), but not their interaction ($F = 2.2$, $P = 0.075$). Post hoc tests revealed that the average pore water P was significantly higher ($p < 0.05$) in ashed straw and ashed husk treatments.

3.3 Soil GHG fluxes

Moisture phases and silica-rich rice residue treatments impacted soil GHG fluxes. Soil CO_2 fluxes were the most variable and were relatively high during Phase I (maximum 4 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ for straw -amended soils) before leveling off to a near zero flux during Phase II and then increasing during Phase III (maximum 2 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ in ashed husk- and straw-amended soils) as the soils dried (Fig. 3A). These results are supported by a two-way repeated measures ANOVA that showed significant differences by treatment ($F = 5.5$, $P < 0.0001$) and phase ($F = 137.6$, $P < 0.0001$), and their interaction ($F = 2.0$, $P = 0.042$). Post hoc tests revealed significant differences ($p < 0.05$) showing that the average CO_2 efflux was highest from straw- and husk-amended soils, and was highest in Phase I and lowest in Phase II.

In contrast to CO_2 , CH_4 fluxes were near zero during Phase I and the latter part of Phase III for all treatments, but deviated by treatment during Phase II and the start of Phase III (e.g., >50 $\text{nmol CH}_4 \text{ m}^{-2} \text{ s}^{-1}$ from straw-amended soils) (Fig. 3B). During this time, straw amendment

led to occasionally high soil CH₄ fluxes up to ca. 200 nmol m⁻² s⁻¹ that were highly variable and were likely due to ebullition. In contrast, husk amendment led to low fluxes similar to ashed husk and ashed straw amendments and control until week 6 when a steady increase in soil CH₄ flux up to 50 nmol CH₄ m⁻² s⁻¹ was observed that lasted until the week 10, the beginning of Phase III (Fig. 3B). These results are supported by a two-way repeated measures ANOVA that showed significant differences by treatment (F = 2.4, P = 0.49) and the interaction between treatment and phase (F = 2.2, P = 0.028), but not with phase (F = 1.382, P = 0.252). Post hoc tests revealed that the average soil CH₄ flux was significantly higher (p<0.05) in straw treatments than in ashed straw treatments, but did not significantly differ with phase.

Soil N₂O fluxes were variable and relatively high for straw amendments up to 22 nmol m⁻² s⁻¹ during Phase I and the beginning of Phase II, but for all amendments were low during the remainder of Phase II and increased to ca. 10 nmol m⁻² s⁻¹ during Phase III (Fig 3C). A two-way repeated measures ANOVA showed significant differences by treatment (F = 4.8, P < 0.0001) and phase (F = 5.1, P = 0.006), but not their interaction (F = 1.4, P = 0.183). Post hoc tests revealed significant differences (p<0.05) and show that the average soil N₂O efflux was highest in straw treatments compared to all other treatments and control, and was highest in Phase III.

3.4 Treatments and control GWP

Considering the sum of all three measured GHGs together over the entire experiment on a CO₂-equivalent basis, straw amendment led to the highest GWP observed in this study of 612 ± 76 g CO₂-eq m⁻², which was 228% higher than control (Fig. 4). Husk amendment led to GWP of 367 ± 42 g CO₂-eq m⁻², which was 40% less than straw but was 96% higher than control. Ashed

husk and ashed straw amendments led to ca. 40% higher GWP than control but lower than fresh residues and were 251 ± 26 and 278 ± 28 g CO₂-eq m⁻², respectively (Fig. 4).

To capture the dynamics of GWP on a temporal basis, we report results per Phase. Using a two-way repeated measures ANOVA, significant differences were observed by treatment ($F = 9.7$, $P < 0.0001$), Phase ($F = 37.7$, $P < 0.0001$), and their interaction ($F = 2.2$, $P = 0.028$). Post hoc tests showed that GWP was highest in Phases II and III and was significantly higher in straw treatments than all other treatments and control. The increase in GWP relative to control due to straw was mostly as enhanced N₂O and CH₄ emissions during Phase II, but CO₂ emissions during Phases II and III also contributed (Table 2). In contrast, the increase in GWP due to husk was a result of enhanced CO₂ and CH₄ emissions during Phases II and III (Table 2). Contributions of N₂O and CH₄ for GWP was about an order of magnitude less from husk-amended soils compared to straw amendments during Phase II (Table 2). The increases in GWP for ashed husk and ashed straw amendments compared to control were mostly as increased CO₂ during Phases II and III and these amendments did not exhibit the increase in CH₄ emissions observed with the fresh residues.

4. Discussion

We sought to understand how soil incorporation of rice husk and rice straw residues would affect pore water chemistry and GHGs when amended soils that had been previously incubated under flooded conditions (Penido et al., 2016) are re-flooded and again dried. Incorporation of residues at levels used in these experiments (1% w/w) was designed to provide soluble silica that could benefit plants for multiple growing seasons; thus, it is imperative to

evaluate the impacts of that practice on pore water chemistry and GHG emissions over multiple wet-dry cycles.

Our results support our first hypothesis as rice husk incorporation led to the highest average pore water Si, which was 4-fold higher than control and 2-fold higher than straw incorporation (Fig. 2C). These pore water Si results are consistent with the findings reported in the initial incubation study (Penido et al., 2016). Taken together, these results indicate that soils amended with rice husk will have significantly higher concentrations of pore water Si available for plant uptake after a series of wet-dry cycles, more so than straw- or ash-amended soils. Soil incorporation of fresh husk into flooded rice soils has also resulted in a 25 – 50% decrease in inorganic As in rice grain without affecting dissolved CH₄ levels in 3 rice cultivars (Seyfferth et al., 2016).

For all treatments and control, pore water Fe and As increased as soils were flooded due to reductive dissolution of Fe (oxyhydr)oxides and As release, and decreased as soils dried due to Fe oxidative precipitation and As adsorption (Fig. 2A-B) (Pedersen et al., 2006; Smedley and Kinniburgh, 2002). While it is possible that some Fe(III)-OM complexes contribute to the Fe signal (Steinmann and Shotyk, 1997), Fe(II) is the dominant form and thus high pore water Fe are an indication of reductive Fe dissolution (Ponnamperuma, 1972). In contrast to our hypotheses, average pore water As and Fe was unexpectedly similar when fresh husk- and rice straw-amended soils were reflooded (Fig. 2A and B). In our previous work, Penido et al. (2016) observed 200 – 1600 µM Fe and 1 – 1.6 µM As in pore water upon straw incorporation and < 100 µM Fe and 0.25 to 0.45 µM As upon husk incorporation. After the re-flooding and drying reported in the present study, pore water Fe and As reached a maximum of ~400 µM and ~0.8 µM, respectively, for both husk and straw amendments (Fig. 2A-B). These values are generally

lower for straw amendment and higher for husk amendment than during the first flooding cycle (Penido et al., 2016). Similar to results from husk, incorporation of ashed husk and ashed straw also led to higher pore water Fe and As during re-flooding (average $\sim 100 \mu\text{M}$ Fe and $\sim 0.4 \mu\text{M}$ As, Fig. 2A-B) than the initial incubation (average $\sim <50 \mu\text{M}$ Fe and $\sim 0.2 \mu\text{M}$ As, Penido et al., 2016). These data suggest that during the first incubation, the strong reducing conditions due to fresh straw amendment (Penido et al., 2016) drove Fe reduction, As release and reduction, and formation of volatile As compounds that likely left the system via outgassing (Huang et al., 2012). This resulted in lower pore water As concentrations than expected for straw-amended soils during re-flooding (Fig. 2B). However, the modest reducing conditions observed in the first incubation with husk or ash amended soils (Penido et al., 2016) did not favor As volatilization (Huang et al., 2012) and upon drying favored retention of As (adsorption/co-precipitation) onto neoformed Fe (oxyhydr)oxides that were readily released upon the re-flooding (Neubauer et al., 2008) in the present study (Fig. 2A-B). The lag in maximum pore water Fe and As in husk amended soils relative to straw amended soils may indicate differences in neoformed Fe minerals and/or mechanisms of As retention upon drying arising from differences in silica and organic matter (Jones et al., 2009) between husk and straw after the first incubation (Penido et al., 2016), but further research is needed in order to confirm that claim.

Our results partially support our second hypothesis as incorporation of rice straw led to the highest GHG emissions and consequently the largest GWP (Fig. 4). However, our observations do not support that the rice straw GWP increase would be mainly an effect of enhanced CH_4 emissions due to strongly reducing conditions (Fig. 1, Table 2). The reducing conditions due to pre-incubated straw amendment reported here were not as strong as those observed in Penido *et al.* (2016). In fact, no significant differences in average Eh between

treatments were observed in the present study (Fig. 1). This lack of strongly reducing conditions favored less CH₄ production in straw-amended soils than was hypothesized. While CH₄ emissions were important, N₂O emissions contributed more to GWP for straw-amended soils in the present study (Table 2). Rice straw is known to have a high labile C content and upon soil incorporation and flooding results in a drop in soil redox potential (Wang et al., 1992). These strongly reducing conditions due to straw amendment typically increase CH₄ emissions under continuous flooding (Bronson et al., 1997; Kimura et al., 1992; Kludze and Delaune, 1995; Wang et al., 1992; Wassmann et al., 2002; Wassmann et al., 2000; Yagi and Minami, 1990). However, our data (Fig. 1B and 4; Table 2) in conjunction with that of Penido et al. (2016) indicates that much of the labile C in fresh straw that typically results in strongly reducing conditions after soil incorporation and flooding is oxidized to CO₂ and/or outgassed, particularly upon drying/drainage. This results in C loss from the soil. Noteworthy, rice farmers in Northern California, USA who incorporate fresh straw and flood fields during the winter fallow season observe rapid decomposition of the material when flooded soils are drained for field preparation (Fitzgerald et al., 2000). When these dried and pre-incubated soils are re-flooded, soil microorganisms have access to less labile pools of organic carbon, similar to those encountered when soils are amended with husk. This labile carbon limitation could also explain the similar extent of reductive dissolution of Fe oxyhydr(oxides) and As release observed in pre-incubated straw and husk amended soils in the present study.

Regardless of the similar average Eh observed (Fig 1B), we observed the largest cumulative GWP from straw-amended soils, which were 40% higher than those from husk-amended soils, 60% higher than those from ash-amended soils, and 70% higher than those from non-amended control soils (Fig. 4). The increase in overall GWP due to straw amendment was

mainly due to larger N₂O fluxes during re-flooding (early Phase II), as well as smaller components from larger CH₄ fluxes during flooded periods and CH₄ and CO₂ fluxes at the onset of drainage (Table 2; Figure 3). Recent work has highlighted the importance of measuring GHG emissions during end-of-season drainage, which shows that enhanced N₂O emissions occur when the percentage of water-filled pore spaces (WFPS) decrease (Adviento-Borbe et al., 2015). Because of a similar decrease in WFPS, mid-season drainage events also result in higher N₂O emissions (Towprayoon et al., 2005). Our data show that when pre-incubated soils are re-flooded, N₂O emissions are enhanced particularly within the first 1-2 weeks for soils amended with straw. Rice straw also has a higher N content and increases soluble organic N within the first 5 days of incubation, which stimulates microbial activity, N mineralization, and N₂O flux within the first 10 days of incubation (Lou et al., 2007). Some prior work has shown enhanced N₂O upon N fertilizer application, which was not as great as with straw amendment (Bronson et al., 1997; Sander et al., 2014). However, the frequency of sampling (i.e., once every one to two weeks) and the late initiation of sampling (i.e., typically after rice transplantation) may have missed spikes in N₂O emissions due to straw amendment as soils are reflooded in prior work (Towprayoon et al., 2005). Similar to our findings, Linquist et al. (2015) observed elevated N₂O emissions before fields were flooded in Arkansas, USA rice soils into which the preceding crop residues had been incorporated.

Compared to straw, far fewer studies have been conducted on rice husk residue incorporation into paddy soil. Our previous work showed that soil incorporation of husk did not result in as strong of reducing conditions as straw incorporation in flooded soils, with consequently higher Eh and lower dissolved CH₄ levels (Penido et al., 2016). However, when those soils are re-flooded (in the present study), no difference in total CH₄ fluxes was observed

between husk- and straw-amended soils (Fig. 4). Instead, higher N_2O and to a lesser extent CO_2 were responsible for the 2-fold higher GWP from straw amended soils compared to husk amended soils (Fig. 4). As discussed above, the partial decomposition of straw during the initial incubation and subsequent drying (during which labile C was likely converted to CO_2 and outgassed) rendered straw-amended soils more labile C-limited in the present study and on par with husk-amended soils. As a result, reducing conditions were similar between husk and straw amended soils in the present study (Fig. 1B). Thus, a similar extent methanogenesis was observed in husk- and straw-amended soils (Table 2, Figure 3B).

Our results provide support for our third hypothesis that ashed residues would lead to the lowest GWP of amended soils (Fig. 4). Burning of rice straw and rice husk residues for an energy source is practiced commonly in Asia (Savant et al., 1997b). The burned residues that remain that we have termed “ashes” are not true ashes, but incompletely combusted material that is concentrated in Si, As, and P, depleted in labile C, and do not affect soil pH upon incorporation (Fig. 1C). Burning of residues undoubtedly leads to increased emissions of C as CO_2 during incomplete combustion and leaves behind recalcitrant C (Gustafsson et al., 2009). These C emissions due to burning are not accounted for in the GWP estimates presented in this study. However, we can estimate C losses upon burning by using our measurements that burning 15 kg of fresh husk to make ash results in 9.35 kg of mass loss. Then, by applying the %OM = $1.72 \times \text{\%TOC}$ conversion factor used for soils, approximately 36% of the original 15 kg was lost as C. Thus, including the C lost during husk burning would yield C emissions that are roughly 36% higher than we measured here. Moreover, it is estimated that burning straw to make ashed straw results in 10% higher C emissions than burning husk to make ashed husk (Kajiura et al., 2015). Due to the loss of labile C during burning, it is no surprise that ashed amendments led to

the lowest GWP of amended soils that we measured (Fig. 4). If these burning C losses were included, we might expect GWP from ashed residues to be comparable to fresh residues, but we would also need to account for C losses during the first incubation (Penido et al., 2016) and other C costs associated with transportation of the materials, and the impact that plants would have (e.g., root C exudation, C fixation) on the C cycle in these systems for a full life cycle assessment. Noteworthy, incorporation of these burned materials increased CO₂ emissions from soils compared to control (Fig. 4). We also observed higher dissolved P concentrations in ashed materials (Fig. 2D), which may have provided an advantage for CO₂-respiring aerobic and anaerobic microorganisms in ash-amended soils compared to control, and could also be plant-available (Ehlers et al., 2010). Finally, to fully address the long-term implications for GWP we must consider multiple flooding-drying cycles where the impact of initial C emissions by combustion of ashes are likely to be compensated by lower long-term GHG emissions as result of the chosen management practice.

While As was also concentrated in ashed residues, the rate of release was fastest during re-flooding to a maximum of ~0.6 to 0.7 μ M As and then pore water As slowly declined throughout the flooded period, and was decoupled from pore water Fe (Fig. 2A and B). These observations indicate that a retention mechanism other than adsorption onto reducible Fe (oxyhydr)oxides was responsible for the retention of As in ash-amended soils upon drying in the initial study (Penido et al., 2016). This As was readily released upon initial re-flooding, but was slowly retained in the soil over time. These results will require further investigation across multiple rewetting and drying cycles to evaluate their persistence and the underlying mechanisms. Furthermore, burning of silica-rich rice residues, particularly at higher temperatures, enables cristobalite formation (Savant et al., 1997b), which is less soluble than the

poorly-crystalline opaline Si that forms in fresh tissues. This practice thus leads to a lower concentration of pore water Si compared to fresh residues upon incorporation (Fig. 2C and Seyfferth et al., 2016). Taken together, despite its ability to concentrate and release higher pore water P, the low pore water Si concentrations and decrease in air quality (Gustafsson et al., 2009) from rice residue burning render ashed residues not ideal to sustainably enhance Si for rice plants; however, it may be an effective P fertilizer. Furthermore, the ability of ashed residues to decrease pore water As over time (Fig. 2B) may be due to adsorption onto the burned material and should be further explored.

5. Conclusions

While the results reported here are focused on soils without plants to avoid confounding effects, these data show that major biogeochemical differences exist among rice residues in terms of their potential for GHG emissions and cycling of arsenic and nutrients over recurrent flooding and drying cycles. Pre-incubation of straw appears to minimize As release and CH₄ emissions upon re-flooding likely because of As and C outgassing during the initial incubation, yet pre-incubation of husk does affect Si release. Husk amendment could thus be beneficial for plants even after recurrent flooding and drying cycles. Plants will undoubtedly affect GHG fluxes due to plant-mediated gas exchange through aerenchyma and C exudation into the rhizosphere, and will directly influence cycling of As and nutrients (i.e., Si) via plant-uptake. It is likely that increased plant nutrition due to increased Si from husk amendment will increase plant growth and thus the rate of plant-mediated gas exchange and C exudation from roots and plant uptake. Future research on rice husk incorporation should focus on plant-based studies at the field scale under natural weather variability with measurements of multiple GHGs. In particular, studies

should focus on the wetting-up and flooded periods prior to transplanting, which we show is important for N₂O fluxes even when plants are absent and especially where pre-incubated straw has been incorporated to test for long-term effects of this management practice.

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568 **References**

- 569 Adviento-Borbe MA, Padilla GN, Pittelkow CM, Simmonds M, van Kessel C, Linquist B.
570 Methane and Nitrous Oxide Emissions from Flooded Rice Systems following the End-of-
571 Season Drain. *Journal of Environmental Quality* 2015; 44: 1071-1079.
- 572 Arao T, Kawasaki A, Baba K, Mori S, Matsumoto S. Effects of water management on cadmium
573 and arsenic accumulation and dimethylarsinic acid concentrations in Japanese rice.
574 *Environmental Science & Technology* 2009; 43: 9361-9367.
- 575 Banerjee M, Banerjee N, Bhattacharjee P, Mondal D, Lythgoe PR, Martinez M, et al. High
576 arsenic in rice is associated with elevated genotoxic effects in humans. *Scientific Reports*
577 2013; 3:2195.
- 578 Bronson KF, Neue HU, Singh U, Abao EB. Automated chamber measurements of methane and
579 nitrous oxide flux in a flooded rice soil .1. Residue, nitrogen, and water management.
580 *Soil Science Society of America Journal* 1997; 61: 981-987.
- 581 Carey AM, Scheckel KG, Lombi E, Newville M, Choi Y, Norton GJ, et al. Grain Unloading of
582 Arsenic Species in Rice. *Plant Physiology* 2010; 152: 309-319.
- 583 Conrad R, Klose M, Yuan Q, Lu YH, Chidthaisong A. Stable carbon isotope fractionation,
584 carbon flux partitioning and priming effects in anoxic soils during methanogenic
585 degradation of straw and soil organic matter. *Soil Biology & Biochemistry* 2012; 49:
586 193-199.
- 587 Delwiche CC, Cicerone RJ. Factors Affecting Methane Production under Rice. *Global*
588 *Biogeochemical Cycles* 1993; 7: 143-155.
- 589 Duxbury JM, Mayer AB, Lauren JG, Hassan N. Food chain aspects of arsenic contamination in
590 Bangladesh: Effects on quality and productivity of rice. *Journal of Environmental*
591 *Science and Health Part a-Toxic/Hazardous Substances & Environmental Engineering*
592 2003; 38: 61-69.
- 593 Ehlers K, Bakken LR, Frostegard A, Frossard E, Bunemann EK. Phosphorus limitation in a
594 Ferralsol: Impact on microbial activity and cell internal P pools. *Soil Biology &*
595 *Biochemistry* 2010; 42: 558-566.
- 596 Fitzgerald GJ, Scow KM, Hill JE. Fallow season straw and water management effects on
597 methane emissions in California rice. *Global Biogeochemical Cycles* 2000; 14: 767-776.
- 598 Gustafsson O, Krusa M, Zencak Z, Sheesley RJ, Granat L, Engstrom E, et al. Brown Clouds over
599 South Asia: Biomass or Fossil Fuel Combustion? *Science* 2009; 323: 495-498.
- 600 Haefele S, Knoblauch C, Gummert M, Konboon Y, Koyama S. Black carbon in rice-based
601 systems: characteristics and opportunities. In: Woods WI, Teixeira W, Lehmann J,
602 Steiner C, Prins AW, editors. *Terra Preta Nova: a Tribute to Wim Sombroek*. Springer,
603 New York, 2009, pp. 445-463.

604 Huang H, Jia Y, Sun GX, Zhu YG. Arsenic Speciation and Volatilization from Flooded Paddy
605 Soils Amended with Different Organic Matters. *Environmental Science & Technology*
606 2012; 46: 2163-2168.

607 Jones AM, Collins RN, Rose J, Waite TD. The effect of silica and natural organic matter on the
608 Fe(II)-catalysed transformation and reactivity of Fe(III) minerals. *Geochimica Et*
609 *Cosmochimica Acta* 2009; 73: 4409-4422.

610 Kaewpradit W, Toomsan B, Vityakon P, Limpinuntana V, Saenjan P, Jogloy S, et al. Regulating
611 mineral N release and greenhouse gas emissions by mixing groundnut residues and rice
612 straw under field conditions. *European Journal of Soil Science* 2008; 59: 640-652.

613 Kajiura M, Wagai R, Hayashi K. Optimal Thermolysis Conditions for Soil Carbon Storage on
614 Plant Residue Burning: Modeling the Trade-Off between Thermal Decomposition and
615 Subsequent Biodegradation. *Journal of Environmental Quality* 2015; 44: 228-235.

616 Kim DG, Vargas R, Bond-Lamberty B, Turetsky MR. Effects of soil rewetting and thawing on
617 soil gas fluxes: a review of current literature and suggestions for future research.
618 *Biogeosciences* 2012; 9: 2459-2483.

619 Kimura M, Miura Y, Watanabe A, Murase J, Kuwatsuka S. Methane Production and Its Fate in
620 Paddy Fields .1. Effects of Rice Straw Application and Percolation Rate on the Leaching
621 of Methane and Other Soil Components into the Subsoil. *Soil Science and Plant Nutrition*
622 1992; 38: 665-672.

623 Kludze HK, Delaune RD. Straw Application Effects on Methane and Oxygen-Exchange and
624 Growth in Rice. *Soil Science Society of America Journal* 1995; 59: 824-830.

625 Lagomarsino A, Agnelli AE, Pastorelli R, Pallara G, Rasse DP, Silvennoinen H. Past water
626 management affected GHG production and microbial community pattern in Italian rice
627 paddy soils. *Soil Biology & Biochemistry* 2016; 93: 17-27.

628 Li RY, Stroud JL, Ma JF, McGrath SP, Zhao FJ. Mitigation of arsenic accumulation in rice with
629 water management and silicon fertilization. *Environmental Science & Technology* 2009;
630 43: 3778-3783.

631 Linquist BA, Adviento-Borbe MA, Pittelkow CM, van Kessel C, van Groenigen KJ. Fertilizer
632 management practices and greenhouse gas emissions from rice systems: A quantitative
633 review and analysis. *Field Crops Research* 2012; 135: 10-21.

634 Linquist BA, Anders MM, Adviento-Borbe MAA, Chaney RL, Nalley LL, Da Rosa EFF, et al.
635 Reducing greenhouse gas emissions, water use, and grain arsenic levels in rice systems.
636 *Global Change Biology* 2015; 21: 407-417.

637 Liu C, Lu M, Cui J, Li B, Fang CM. Effects of straw carbon input on carbon dynamics in
638 agricultural soils: a meta-analysis. *Global Change Biology* 2014; 20: 1366-1381.

- 639 Lou YS, Ren LX, Li ZP, Zhang TL, Inubushi K. Effect of rice residues on carbon dioxide and
640 nitrous oxide emissions from a paddy soil of subtropical China. *Water Air and Soil*
641 *Pollution* 2007; 178: 157-168.
- 642 Luxton TP, Tadanier CJ, Eick MJ. Mobilization of arsenite by competitive interaction with
643 silicic acid. *Soil Science Society of America Journal* 2006; 70: 204-214.
- 644 Ma JF, Yamaji N, Mitani N, Xu X-Y, Su Y-H, McGrath SP, et al. Transporters of arsenite in rice
645 and their role in arsenic accumulation in rice grain. *Proceedings of the National Academy*
646 *of Sciences* 2008; 105: 9931-9935.
- 647 Ma R, Shen JL, Wu JS, Tang Z, Shen QR, Zhao FJ. Impact of agronomic practices on arsenic
648 accumulation and speciation in rice grain. *Environmental Pollution* 2014; 194: 217-223.
- 649 Myhre G, Shindell D, Breon F-M, Collins W, Fuglestad J, Huang J, et al. Anthropogenic and
650 Natural Radiative Forcing. In: Stocker TF, Qin D, Plattner G-K, Tignor M, Allen SK,
651 Boschung J, et al., editors. *Climate Change 2013: The Physical Science Basis.*
652 *Contribution of Working Group I to the Fifth Assessment Report of the*
653 *Intergovernmental Panel on Climate Change.*, Cambridge, United Kingdom and New
654 York, NY, USA., 2013.
- 655 Neubauer SC, Emerson D, Megonigal JP. Microbial oxidation and reduction of iron in the root
656 zone and influences on metal mobility. In: Violante A, Huang P, Gadd GM, editors.
657 *Biophysico-Chemical Processes of Heavy Metals and Metalloids in Soil Environments,*
658 *Hoboken, NJ, 2008, pp. 339-371.*
- 659 Neue HU. Fluxes of methane from rice fields and potential for mitigation. *Soil Use and*
660 *Management* 1997; 13: 258-267.
- 661 Panaullah GM, Alam T, Hossain MB, Loeppert RH, Lauren JG, Meisner CA, et al. Arsenic
662 toxicity to rice (*Oryza sativa* L.) in Bangladesh. *Plant and Soil* 2009; 317: 31-39.
- 663 Pearson AJ, Pizzuto JE, Vargas R. Influence of run of river dams on floodplain sediments and
664 carbon dynamics. *Geoderma* 2016; 272: 51-63.
- 665 Pedersen HD, Postma D, Jakobsen R. Release of arsenic associated with the reduction and
666 transformation of iron oxides. *Geochimica Et Cosmochimica Acta* 2006; 70: 4116-4129.
- 667 Penido ES, Bennet AJ, Hanson TE, Seyfferth AL. Biogeochemical impacts of silica-rich rice
668 residue incorporation into flooded soils: Implications for rice nutrition and cycling of
669 arsenic. *Plant and Soil* 2016; 399: 75-87.
- 670 Petrakis S, Seyfferth AL, J. K, Inamdar S, Vargas R. Influence of experimental extreme water
671 pulses on greenhouse gas emissions from soils. *Biogeochemistry* 2017; In Press.
- 672 Ponnamperna FN. The chemistry of submerged soils. *Advances in Agronomy* 1972; 24: 29-96.

673 Pumpanen J, Kolari P, Ilvesniemi H, Minkkinen K, Vesala T, Niinisto S, et al. Comparison of
674 different chamber techniques for measuring soil CO₂ efflux. *Agricultural and Forest*
675 *Meteorology* 2004; 123: 159-176.

676 Sander BO, Samson M, Buresh RJ. Methane and nitrous oxide emissions from flooded rice fields
677 as affected by water and straw management between rice crops. *Geoderma* 2014; 235:
678 355-362.

679 Savant NK, Datnoff LE, Snyder GH. Depletion of plant-available silicon in soils: A possible
680 cause of declining rice yields. *Communications in Soil Science and Plant Analysis* 1997a;
681 28: 1245-1252.

682 Savant NK, Snyder GH, Datnoff LE. Silicon management and sustainable rice production.
683 *Advances in Agronomy*, Vol 58. 58, 1997b, pp. 151-199.

684 Schwertmann U, Thalmann H. Influence of Fe(II) , Si , and pH on formation of lepidocrocite and
685 ferrihydrite during oxidation of aqueous FeCl₂ solutions. *Clay Minerals* 1976; 11: 189-
686 200.

687 Seyfferth AL, Fendorf S. Silicate mineral impacts on the uptake and storage of arsenic and plant
688 nutrients in rice (*Oryza sativa* L.). *Environmental Science & Technology* 2012; 46:
689 13176-13183.

690 Seyfferth AL, Kocar BD, Lee JA, Fendorf S. Seasonal dynamics of dissolved silicon in a rice
691 cropping system after straw incorporation. *Geochimica Et Cosmochimica Acta* 2013;
692 123: 120-133.

693 Seyfferth AL, McCurdy S, Schaefer MV, Fendorf S. Arsenic concentrations in paddy soil and
694 rice and health implications for major rice-growing regions of Cambodia *Environmental*
695 *Science & Technology* 2014; 48: 4699-4706.

696 Seyfferth AL, Morris AH, Gill R, Kearns KA, Mann JN, Paukett M, et al. Soil incorporation of
697 silica-rich rice husk decreases inorganic arsenic in rice grain. *Journal of Agricultural and*
698 *Food Chemistry* 2016; 64: 3760-3766.

699 Smedley PL, Kinniburgh DG. A review of the source, behaviour and distribution of arsenic in
700 natural waters. *Applied Geochemistry* 2002; 17: 517-568.

701 Steinmann P, Shotyk W. Chemical composition, pH, and redox state of sulfur and iron in
702 complete vertical porewater profiles from two Sphagnum peat bogs, Jura Mountains,
703 Switzerland. *Geochimica Et Cosmochimica Acta* 1997; 61: 1143-1163.

704 Swedlund PJ, Webster JG. Adsorption and polymerization of silicic acid on ferrihydrite, and its
705 effect on arsenic adsorption. *Water Resources Research* 1999; 33: 3413-3422.

706 Towprayoon S, Smakgahn K, Poonkaew S. Mitigation of methane and nitrous oxide emissions
707 from drained irrigated rice fields. *Chemosphere* 2005; 59: 1547-1556.

708 Vibol S, Towprayoon S. Estimation of methane and nitrous oxide emissions from rice field with
709 rice straw management in Cambodia. *Environmental Monitoring and Assessment* 2010;
710 161: 301-313.

711 Wang JY, Zhang XL, Xiong ZQ, Khalil MAK, Zhao X, Xie YX, et al. Methane emissions from a
712 rice agroecosystem in South China: Effects of water regime, straw incorporation and
713 nitrogen fertilizer. *Nutrient Cycling in Agroecosystems* 2012; 93: 103-112.

714 Wang ZP, Delaune RD, Lindau CW, Patrick WH. Methane Production from Anaerobic Soil
715 Amended with Rice Straw and Nitrogen Fertilizers. *Fertilizer Research* 1992; 33: 115-
716 121.

717 Wassmann R, Aulakh MS, Lantin RS, Rennenberg H, Aduna JB. Methane emission patterns
718 from rice fields planted to several rice cultivars for nine seasons. *Nutrient Cycling in*
719 *Agroecosystems* 2002; 64: 111-124.

720 Wassmann R, Buendia LV, Lantin RS, Bueno CS, Lubigan LA, Umali A, et al. Mechanisms of
721 crop management impact on methane emissions from rice fields in Los Banos,
722 Philippines. *Nutrient Cycling in Agroecosystems* 2000; 58: 107-119.

723 Weber S, Lueders T, Friedrich MW, Conrad R. Methanogenic populations involved in the
724 degradation of rice straw in anoxic paddy soil. *Fems Microbiology Ecology* 2001; 38: 11-
725 20.

726 Williams PN, Price AH, Raab A, Hossain SA, Feldmann J, Meharg AA. Variation in arsenic
727 speciation and concentration in paddy rice related to dietary exposure. *Environmental*
728 *Science & Technology* 2005; 39: 5531-5540.

729 Yagi K, Minami K. Effect of organic-matter application on methane emission from some
730 Japanese paddy fields. *Soil Science and Plant Nutrition* 1990; 36: 599-610.

731 Zou JW, Huang Y, Jiang JY, Zheng XH, Sass RL. A 3-year field measurement of methane and
732 nitrous oxide emissions from rice paddies in China: Effects of water regime, crop residue,
733 and fertilizer application. *Global Biogeochemical Cycles* 2005; 19.
734
735