



Switchgrass biomass yield and composition and soil quality as affected by treated wastewater irrigation in an arid environment

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ARTICLE INFO

Keywords:

Drought
Sustainability
Cropping system diversification
Bioenergy feedstock
Soil salinization

ABSTRACT

Freshwater (FW) scarcity as a result of prolonged drought has reduced FW availability to agriculture in the arid west Texas region in order to meet demands from other sectors. Alternatively, there is enormous potential to utilize treated urban wastewater (TWW) for agricultural irrigation. However, the soil salinization potential of TWW is a concern as it can be detrimental to crops and soil quality. Alternative crops that are both less water-intensive and salt-tolerant are therefore needed to sustain this region's agriculture. Switchgrass is a perennial grass that is well adapted to grow on marginal lands and is a novel crop for lignocellulosic bioenergy feedstock. However, its performance when irrigated with TWW on arid soils of far west Texas is largely unknown. This field study evaluated the yield potential and composition of switchgrass biomass as affected by TWW along with soil quality changes, using a split-plot experimental design. Results indicate that biomass yields were not affected by TWW irrigation and there were no significant differences between TWW and FW across years. With respect to biomass composition, cellulose and lignin contents were lower, while ash content was significantly higher in TWW treatment. Theoretical ethanol production was not affected. Soil salinity and sodicity increased overtime but this increase was more prominent under TWW irrigation. However, application of gypsum and sulfur significantly reduced soil sodicity. These results indicate that switchgrass can tolerate soil salinity induced by TWW application and therefore can be successfully grown on these marginal arid soils as a bioenergy feedstock.

1. Introduction

Climate change has caused changes in weather patterns that resulted in frequent high temperature events and erratic rainfall patterns creating extended periods of drought, especially in semi-arid and arid regions such as Southwest United States [1,2] including far west Texas. Recent model projections have also showed that the Southwest US is going to experience more frequent and hotter droughts in future [3–5]. Same is the case for the state of Texas with higher drought risk expected in the latter half of 21st century [6]. This exacerbates the long-term freshwater (FW) shortages in these areas and puts FW supply systems at an increased risk of water shortage [1] to meet agricultural and municipal water demands. Furthermore, increased FW demand from municipal and industrial sector, due to growing population and industrialization, has resulted in diversion and reduced allocation of FW to agriculture sector. In 2020, water needs in Texas were projected as 51

and 28%, respectively for agriculture and municipal sectors. However, by 2070, water needs by municipal sector is projected to increase to 39% and that of agriculture will reduce to 36% (TWDB, 2017). This is a substantial reduction in irrigation water availability to agriculture and puts agricultural productivity at risk [7]. It is therefore important that better water management strategies must be adopted to efficiently utilize available water resources and keep agriculture sustainable in this region.

United States Department of Agriculture (USDA) recommends diversifying water resource profile in order to meet agricultural water demands [8]. Using non-traditional water resources such as treated wastewater from urban water treatment plants is one approach that could augment FW supplies and help maintain agricultural productivity in this region [7,9]. El Paso county located in the far west Texas region has an extremely arid climate and is part of the middle Rio Grande basin. Agriculture in this region largely relies on FW supplies from the Rio

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<https://doi.org/10.1016/j.biombioe.2021.106160>

Received 7 December 2020; Received in revised form 10 May 2021; Accepted 6 June 2021

Available online 19 June 2021

0961-9534/© 2021 Published by Elsevier Ltd.

Grande river [10], which receives its water from snowpack on mountains of southern Colorado. Cropping pattern in this region is dominated by water-intensive crops including cotton (*Gossypium hirsutum* L.), pecan (*Carya illinoensis* L.) and alfalfa (*Medicago sativa* L.) [11]. However, recent droughts and reduced snow water equivalent has severely impacted the FW availability in the river. This has reduced FW allocation to the irrigation districts and farmers are forced to abandon fields dedicated to annual crops such as cotton and divert the little available water to salvage perennial cash crops such as pecans [12]. The city of El Paso produces about 7900 ha-meter of treated wastewater (TWW), of which only 13% is reused for industrial and commercial landscape irrigation. Therefore, there is great potential for diverting this water to agricultural irrigation and improve agricultural sustainability.

Treated wastewater, on the other hand, contains higher dissolved salt concentrations and could negatively affect soil quality by causing soil salinization and reduce crop productivity [13,14]. While cotton can tolerate soil salinity, both pecan and alfalfa are salt-sensitive crops and are not suitable for TWW irrigation. Therefore, it is important to diversify cropping pattern and find alternative crops that are both salt-tolerant and less water intensive than traditional crops. At this juncture, growing bioenergy crops such as switchgrass (*Panicum virgatum* L.) could be a potential alternative due to its tolerance to environmental stresses and increased demand for lignocellulosic biomass feedstocks in bioenergy production [8]. Growing such bioenergy crops on abandoned marginal lands, a characteristic of this region, can expand their acreage into non-traditional growing areas, enhance ecosystem services of marginal lands [15] and help generate farm income to farmers [16], in addition to meeting the mandated Renewable Fuels Standards (RFS2) goal to use 36 billion gallons of renewable bio-based fuels by 2022 [17].

There is a growing interest in non-traditional fuels such as bioethanol produced from bioenergy feedstocks. In U.S. corn-based bioethanol production is at forefront in bioenergy production and is a first-generation biofuel production technology [18]. However, due to the ease of availability of lignocellulosic biomass feedstocks, lignocellulosic biofuel production has gained importance as a second-generation bioenergy production [8,19]. Warm season perennial C_4 grasses such as switchgrass and miscanthus (*Miscanthus giganteus* L.) have gained increased attention as potential sources of lignocellulosic feedstock for biofuel production [20]. Switchgrass, is considered as a strong and model candidate for bioenergy feedstock due to its desirable traits including; perenniality, high water and nutrient use efficiencies, adaptability to marginal lands with low input requirement, tolerance to environmental stresses, high biomass yield potential and improved carbon sequestration [21–27]. Switchgrass was even shown to produce more renewable energy than the non-renewable energy consumed [28]. Due to these desirable properties, switchgrass can be a suitable candidate for bioenergy crop production in arid west Texas.

Moreover, there are two types of switchgrass based on their habitats i.e., upland and lowland [23]. Lowland ecotypes were reported to produce more biomass than upland ecotypes, have delayed flowering and late maturity with thick stems and dense bunching, enabling them to better adapt to the southern latitudes such as the current study site [29,30]. Among the lowland ecotypes, “Alamo” cultivar was shown to be promising with wider adaptability spanning across different USDA hardness zones including Texas [10,31,32]. Previous studies have evaluated switchgrass biomass production on various marginal lands but its performance under TWW irrigation on degraded arid soils of west Texas has not been tested yet.

Due to TWW's tendency to increase soil salinization, the precise effects of TWW irrigation on agronomic performance of switchgrass, its biomass yield and composition are largely unknown and needs investigation. Therefore, the specific objectives of this study were: 1) to evaluate biomass yield potential, biomass compositional changes and theoretical ethanol production in switchgrass grown with TWW irrigation; 2) to quantify changes in rootzone soil salinity and sodicity

overtime with TWW irrigation. We hypothesize that TWW irrigation deteriorates soil quality as a result of soil salinization, which negatively effects switchgrass plant growth and reduced biomass yields and biomass quality.

2. Materials and methods

2.1. Study site and experimental design

A three-year field study was started in April of 2017 at the Texas A&M AgriLife Research and Extension Center El Paso, TX, USA (31° 39' 27.31" N, 106° 16' 8.32" W) (Fig. 1). The study site is characterized as having an arid climate with an annual average precipitation of ~0.17 m and a potential evapotranspiration rate of 1.94 m [8]. Mean annual temperature ranges from −3.6 °C in winter to 35.8 °C during summer. The dominant soil map unit at the study site was Saneli Silty Clay loam (clayey over sandy or sandy-skeletal, montmorillonitic calcareous, thermic Vertic Torrifluvents). A split-plot randomized complete block experimental design was used in this study with irrigation water type as the main plot factor and soil amendment application as the subplot factor. The two water types included fresh (FW) and treated urban wastewater (TWW). Amendment application consisted of either a combined application of gypsum and elemental sulfur (GS) or a no-amendment control (NA). All treatment combinations were replicated three times. There were a total 12 individual experimental plots with each plot measuring 5.6 m long and 2.6 m wide.

2.2. Irrigation water source and analyses

Regular tap water was used as the source of freshwater, which is filtered and chlorine-disinfected Rio Grande river water. Treated wastewater was sourced from a local municipal wastewater treatment plant (Roberto Bustamante). The treatment process for TWW consisted of screening, de-gritting, pre-aeration, primary settling, aeration, secondary settling and chlorine disinfection. Both FW and TWW samples were collected at their source during each growing season using pre-cleaned 0.5 L polyethylene bottles. Collected samples were stored at 4 °C until further chemical analysis is completed. Sub samples of both waters were filtered through a 0.45 μ m syringe filter and analyzed for chemical properties including pH, electrical conductivity (EC), cations (calcium- Ca^{2+} , magnesium- Mg^{2+} , sodium- Na^+ , potassium- K^+ , ammonium- NH_4^+), and anions (chloride- Cl^- , nitrate- NO_3^- , sulfate- SO_4^{2-} , phosphate- PO_4^{3-}) using methods described in the Standard Methods for the Examination of Water and Wastewater by American Public Health Association (APHA). Electrical conductivity and pH of waters were measured on aliquot samples using a Fisher brand accumet XL600 multichannel benchtop meter (Fisher Scientific Com, NH). The cations and anions were determined using ion-exchange chromatography on a Dionex ICS-1100. (Dionex Corporation, Sunnyvale, CA). Relevant chemical properties of both FW and TWW used in this study are given in Table 1.

2.3. Soil amendment application

Elemental sulfur (S) and Gypsum ($CaSO_4 \cdot 2H_2O$) were added to respective amended plots at the rates equivalent to 10 and 1 $Mg\ ha^{-1}$ and were applied only once at the beginning of the experiment (2017) before planting and incorporated into the soil to a depth of 0.15 m. Gypsum was added to the soil with the main aim of countering soil sodicity that may increase as a result of irrigation water application, especially TWW. Also, it is a common practice for growers to add elemental S to solubilize native $CaCO_3$ (up to 10% by weight in the upper 0.75 m) to aid in in-situ gypsum formation. The goal behind this amendment application is to test if such application would affect the switchgrass biomass production other than the irrigation water, as these amendments are normally included in a farmer's soil management

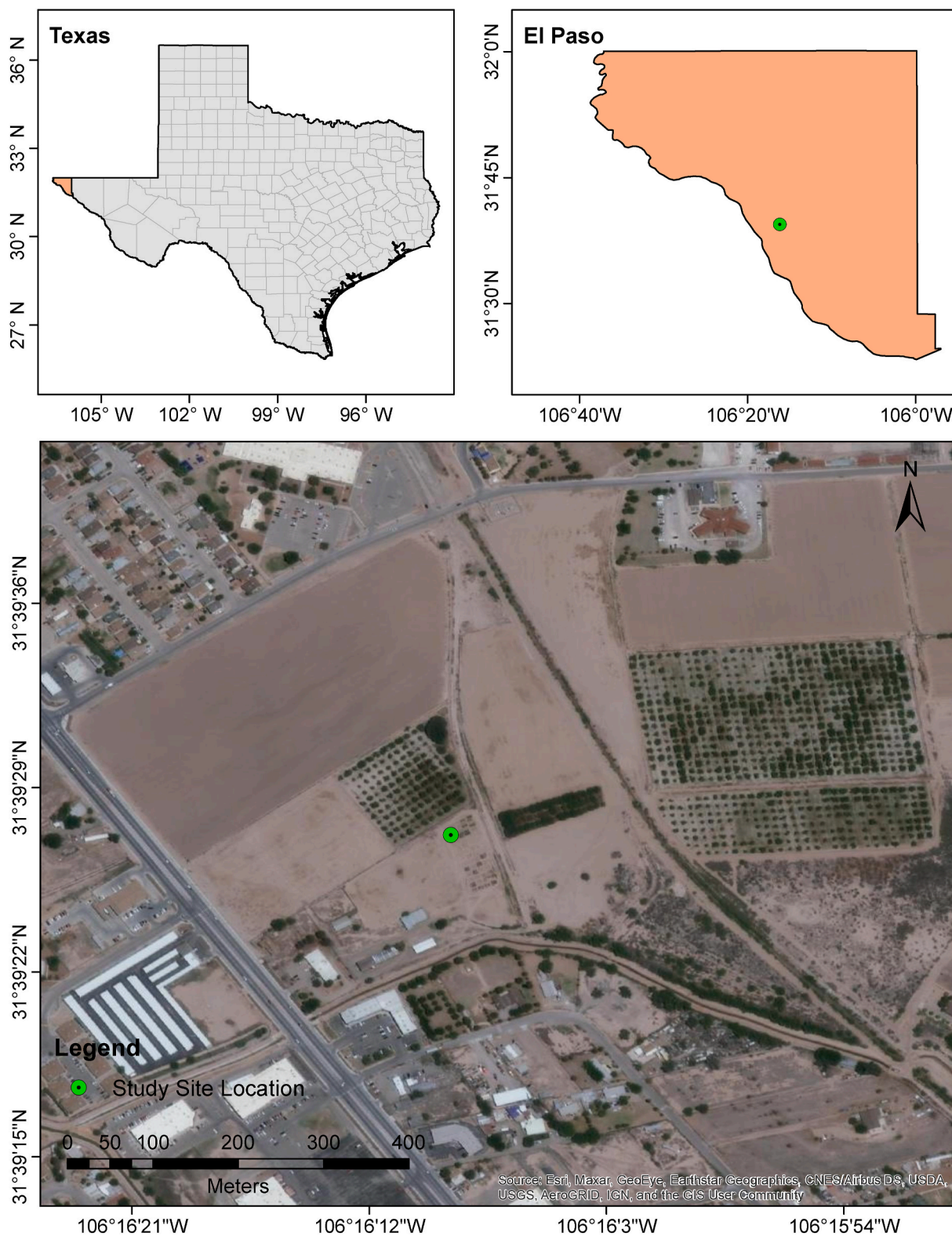


Fig. 1. Graphical illustration of study site in El Paso, TX.

routine in this region and are also capable of transiently increasing soil salinity, specifically gypsum.

2.4. Plot management

Initial soil preparation at the experiment site included tilling using a

disking harrow and then leveling using a field cultivator to prepare a seedbed. Individual plots were made by making raised earthen berms (~0.20 m in height). Each individual plot was separated from each other by a 0.60 m wide buffer strip to avoid any edge effects of treatments and lateral percolation of irrigation water into adjacent plots. An inter-row spacing of 0.90 m and intra-row spacing of 0.05 m was followed as

Table 1

Chemical composition of fresh and treated wastewaters used in this study (mean \pm SE, $n = 4$).

Property	Fresh Water	Treated wastewater
pH	6.82 \pm 0.06	6.89 \pm 0.04
EC _{iw} (dS m ⁻¹)	0.75 \pm 0.07	1.80 \pm 0.30
SAR	2.96 \pm 0.39	5.42 \pm 0.60
SAR Adj	2.85 \pm 0.28	5.90 \pm 0.57
Na ⁺ (mg L ⁻¹)	107 \pm 6.40	268 \pm 37.0
NH ₄ ⁺ (mg L ⁻¹)	3.49 \pm 1.55	6.44 \pm 2.44
K ⁺ (mg L ⁻¹)	13.5 \pm 2.13	23.5 \pm 2.71
Mg ²⁺ (mg L ⁻¹)	13.9 \pm 2.73	22.8 \pm 3.97
Ca ²⁺ (mg L ⁻¹)	105 \pm 8.27	146 \pm 12.1
F ⁻ (mg L ⁻¹)	0.33 \pm 0.06	1.30 \pm 0.97
Cl ⁻ (mg L ⁻¹)	75.9 \pm 9.45	241 \pm 46.7
NO ₃ ⁻ (mg L ⁻¹)	4.58 \pm 0.43	60.2 \pm 10.9
PO ₄ ³⁻ (mg L ⁻¹)	3.84 \pm 2.86	6.42 \pm 0.58
SO ₄ ²⁻ (mg L ⁻¹)	121 \pm 14.4	254 \pm 42.9
CO ₃ ²⁻ + HCO ₃ ⁻ (mg L ⁻¹)	116 \pm 10.5	93.0 \pm 6.17

EC_w: Electrical conductivity of water; SAR: Sodium Adsorption Ratio of water.

per the literature review. There were three rows of switchgrass in each plot and lowland cultivar “Alamo” was transplanted after initial germination in small pots in the greenhouse. Transplantation was completed in late May of 2017. Before transplanting, starter fertilizer was applied to all plots in bands at a rate of 120, 120, and 120 kg ha⁻¹ of nitrogen (N), phosphorus (P₂O₅) and potassium (K₂O), as urea, mono-ammonium phosphate and sulfate of potash, respectively. All fertilizer was applied only at the beginning of the experiment. Weeds in the plots were controlled by a combination of manual weeding and application of a post-emergence herbicide (Atrazine). All plots were irrigated by flood irrigation method as it is the most common practice in this region. Irrigation was scheduled every 3–4-week intervals based on the evapotranspiration rates of each month. A total of 8 irrigations were scheduled with each irrigation event receiving 0.0762 m of water. At the end of a cropping season, each experimental plot received a total of 0.61 m of irrigation water.

2.5. Soil sampling and analyses

Before the onset of the experiment (March 2017), soil samples were collected to characterize the general soil properties of the study site. Soils were collected randomly from four different regions of the site from both 0–0.15 and 0.15–0.30 m depths. These soils were analyzed for their

Table 2

Initial physical and chemical characteristics of study site soils at 0–15 and 15–30 cm depths (mean \pm SE, $n = 4$).

Soil characteristic	0–15 cm	15–30 cm
Sand (%)	41.9 \pm 0.81	48.9 \pm 6.04
Silt (%)	33.5 \pm 0.94	28.1 \pm 5.92
Clay (%)	24.5 \pm 0.93	23.0 \pm 0.50
Texture class	Loam	Loam
Organic Matter (%)	0.60 \pm 0.01	0.64 \pm 0.02
CEC (cmol _c kg ⁻¹)	12.5 \pm 0.32	12.9 \pm 0.18
Bulk Density (g cm ⁻³)	1.31 \pm 0.03	1.44 \pm 0.24
Saturated Paste Extract		
pH	7.37 \pm 0.02	7.25 \pm 0.04
EC _e (dS m ⁻¹)	0.86 \pm 0.10	1.32 \pm 0.31
SAR (mmol l ⁻¹) ^{0.5}	1.96 \pm 0.21	2.81 \pm 0.57
Soluble Na ⁺ (mmol _c l ⁻¹)	4.83 \pm 0.79	7.11 \pm 0.92
Soluble K ⁺ (mmol _c l ⁻¹)	0.66 \pm 0.02	0.57 \pm 0.05
Soluble Ca ²⁺ (mmol _c l ⁻¹)	11.6 \pm 1.09	12.3 \pm 1.71
Soluble Mg ²⁺ (mmol _c l ⁻¹)	0.75 \pm 0.11	0.98 \pm 0.12
Soluble Cl ⁻ (mmol _c l ⁻¹)	13.7 \pm 1.59	7.06 \pm 0.65
Soluble SO ₄ ²⁻ (mmol _c l ⁻¹)	10.2 \pm 0.35	11.5 \pm 1.02

CEC: Cation Exchange Capacity of soil; EC_e: Electrical conductivity of soil saturated paste extract; SAR: Sodium Adsorption Ratio of soil saturated paste extract.

physical and chemical characteristics as presented in Table 2. Soil particle size was measured using the hydrometer method as given in Gavlak et al. (2003). The cation exchange capacity of the soil was estimated following the Bower method of Na⁺ saturation using 1 M sodium acetate solution with pH adjusted to 8.2, followed by ethanol rinsing and replacing adsorbed Na⁺ by NH₄⁺ using a 1 M ammonium acetate solution with pH adjusted to 7.0 (Richards, 1954). The exchange capacity of the soil was calculated by measuring the Na⁺ concentration in the 1 M ammonium acetate extract as given in Richards, (1954). The electrical conductivity of soil (EC_e) and pH were measured on soil saturated paste extracts (SPE) following the methods given in Richards (1954) using Fisher brand accumet XL600 multichannel benchtop meter (Fisher Scientific Com, NH). Water-soluble cations (Na⁺, K⁺, Ca²⁺, and Mg²⁺) were analyzed on saturated paste extracts by inductively coupled plasma-optical emission spectrophotometry using a PerkinElmer Avio 200 spectrophotometer (PerkinElmer, Inc., MA). Sodium adsorption ratio (SAR) of soils was estimated using the equation given in Chaganti et al. [8].

The soil organic matter content was determined using the loss on ignition method (Gavlak et al., 2003).

Pre-study (May 2017) and end of the study (December 2019)) soil samples were collected from each experimental plot at 0–0.15 m and 0.15–0.30 m soil depths using a 0.05 m diameter soil auger. Three random soil samples were collected from each experimental plot at each depth and were composited. Composite soil samples were air-dried, mixed and ground to pass through a 2-mm sieve to achieve uniformity. Pre-study and end of the study soil samples from all experimental plots were analyzed for specific soil quality indices including EC_e, and SAR using methods described above.

2.6. Switchgrass biomass harvesting and analysis

Switchgrass plants were harvested in November after ~180 days (May–Oct) of growth every year from all three rows in each test plot using hand implements. Plants were cut at the base, approximately 0.15 m above the soil surface. Total biomass from each plot was bagged and weighed for fresh weight. Bagged biomass was dried in a forced air oven at 70 °C until a constant weight was reached. Total dry weight of above-ground biomass was recorded for each individual test plot. Sub-samples of dried switchgrass biomass from each treatment were ground to pass through a 1-mm sieve using a commercial grinder and ~0.015 kg of sample was sent to external forage laboratory (Dairy one laboratory, Ithaca, NY, USA) for determining the composition of biomass including water-soluble carbohydrates (WSC), neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin and ash content using near-infrared reflectance spectroscopy [33]. Cellulose content was calculated as the difference between ADF and lignin, while hemicellulose content was calculated as the difference between NDF and ADF. Theoretical ethanol production (TEP, L ha⁻¹) was calculated using the equations [22,34] given below:

$$\text{TEP}_{\text{ss}} = \text{WSC (g kg}^{-1}\text{)} \times F_1 \times F_2 \times 1.267 \text{ (ml g}^{-1}\text{)} \times \text{dry biomass weight (kg ha}^{-1}\text{)} \quad (1)$$

$$\text{TEP}_{\text{C+H}} = \text{Cellulose + hemicellulose content (g kg}^{-1}\text{)} \times F_1 \times F_2 \times F_3 \times F_4 \times 1.267 \text{ (ml g}^{-1}\text{)} \times \text{dry biomass weight (kg ha}^{-1}\text{)} \quad (2)$$

where, F_1 is the coefficient of conversion of sugar to ethanol (0.51); F_2 is the conversion efficiency of sugar to ethanol (0.85); F_3 is the coefficient of conversion cellulose and hemicellulose to sugar (1.11); F_4 is the conversion efficiency of cellulose and hemicellulose to sugar (0.85) [22] and 1.267 is the specific volume of ethanol [33]. Total theoretical ethanol production was calculated as a sum of TEP_{ss} and TEP_{C+H}.

2.7. Statistical analysis

All data were checked for normality and equality of variances tests

and were subjected to analysis of variance using the General Linear Model (GLM) repeated measures analysis in SPSS (v.27) to determine the significance (at 5% level) of main (water type) and sub-plot (amendment application) factors and their respective interactions. Water type and amendment effects were considered as fixed effects and year was considered as a repeated measure in the model. When soil quality measurements were considered (EC_e and SAR), depth of the soil was also included in the model as a fixed factor in addition to water type and amendment application. When statistically significant (at $P \leq 0.05$ or unless otherwise stated), treatment means were separated using the Tukey's HSD test. Correlation analysis was used to assess the relationship between TEP and switchgrass biomass yields. Similarly, to assess the relationship between switchgrass biomass yields and soil EC_e and SAR; a simple linear model was fit using the *lm()* function in R Studio. All graphics were generated using Sigma Plot v.14.5.

3. Results and discussion

3.1. Biomass yields

During the year of 2017, there were no reportable biomass yields, therefore, yields for 2018 and 2019 years are only reported (Table 3). The switchgrass biomass yields ranged from a low of 9.1 Mg ha^{-1} to a high of 12.6 Mg ha^{-1} across all treatments and two years (Table 3). Year, water type and amendment application and their respective interactions did not have any significant effect on switchgrass biomass yields (Table 3). In general, mean biomass yields were higher in the year 2018 compared to 2019. Average biomass yields for FW and TWW across both the years were 11.5 and 10.6 Mg ha^{-1} , respectively. Application of gypsum and sulfur also did not produce any yield advantage relative to unamended treatments. Mean yields were 10.7 and 11.5 Mg ha^{-1} for unamended and GS treatments, respectively.

Our study results concur with those reported by Ganjegunte et al. [10], who also reported that switchgrass biomass yields for the "Alamo" did not differ significantly in the first three years between water types and soil amendment applications under greenhouse experimental conditions. Also, these authors reported no effect of amendment application on switchgrass yields, a result consistent with the current study. It can be assumed that the amendment application would counter soil salinity and sodicity and would facilitate better biomass production. However, Ganjegunte et al. [10] attributed this affect to the high native calcium sources (calcite 10% and gypsum 2.5% by weight, respectively) found in the upper 0.75 m of soils in the study area [10] that could supply in situ Ca^{2+} for up to 3 years. Therefore, application of external gypsum did not produce any added benefit. Similarly, in a related study conducted in the same study site, Chaganti et al. [8] also reported no yield response of

bioenergy sorghum to gypsum and sulfur application.

On the other hand, irrigating with TWW did not negatively affect biomass production of switchgrass. This result is contrasting our original hypothesis where we assumed that TWW irrigation would increase soil salinity and sodicity overtime and decrease biomass production. Our soil quality results show that in fact, both soil salinity and sodicity increased after three years (Figs. 3 and 4) but there was no biomass reduction observed. A simple regression analysis also revealed that there was no significant relationship between switchgrass biomass yields and either soil salinity or sodicity (Fig. 5). Contrastingly, Ganjegunte et al. [10] reported decreased switchgrass biomass yields after 6 years of continuous irrigation with TWW. In a greenhouse study, Pica et al. [35] also reported that the switchgrass biomass yields were affected by low quality produced water irrigation. However, Zanetti et al. [36] showed that lowland switchgrass cultivar "Alamo" could tolerate salinity up to 14 dS m^{-1} without any effect on biomass production. It should be noted that soil salinity in the Ganjegunte et al. [10] and Pica et al. [35] studies increased beyond 14 dS m^{-1} and therefore likely resulted in reduced switchgrass biomass yields. In the current study, the soil salinity did not exceed 14 dS m^{-1} after three years of irrigation with TWW and GS application and thus the biomass production was not negatively affected.

Regardless, the yields reported in this study under irrigated conditions are comparable or well above those reported in literature under various water regimes (irrigated vs. non-irrigated) and soil conditions (marginal vs. non-marginal) using the same "Alamo" cultivar [31,37–42, 47]. For example, Emery et al. [47] reported switchgrass yields ranging between 9 and 12 Mg ha^{-1} , when two cultivars were evaluated for two years under simulated drought conditions (conditions like what found in the current study area) in Southwest Michigan, USA. Similarly, Fu et al. [37] found switchgrass biomass yields averaged at 12.6 Mg ha^{-1} over two years when evaluated under marginal soil conditions in Northern China. Ameen et al. [38], on the other hand reported lower biomass yields ($\sim 8.1 \text{ Mg ha}^{-1}$) when switchgrass was grown on a semi-arid land for two-years. Nevertheless, our results support the findings from literature that switchgrass is indeed a drought-tolerant crop in addition to being salt-tolerant and therefore could be a viable alternative to water-intensive crops grown in this region.

3.2. Biomass quality

3.2.1. Nitrogen

The nitrogen concentration of switchgrass biomass ranged between 2.9 g kg^{-1} to 5.9 g kg^{-1} (Table 4) and was significantly affected by water type and amendment application. Irrigating with TWW significantly increased biomass N concentrations. Biomass nitrogen concentration in TWW treatment was almost twice the concentration of N found in FW irrigated biomass. Mean biomass N for TWW and FW treatments stood at 3.2 and 5.7 g kg^{-1} , respectively. This increase in biomass N concentration can be mostly attributed to the addition of readily available N through TWW (in the form of nitrate, NO_3^-), which facilitated greater uptake of N by switchgrass plants. On the other hand, biomass N concentrations significantly decreased ($P < 0.1$) in GS soils relative to non-amended soils (Table 4). This is most likely due to increase in soil salinity after GS application, which negatively impacted the N uptake from soil [43]. Wastewater application should have had the same effect as TWW also increased soil salinity. However, we believe that the extra nitrogen supply in a readily available form negated the salinity affect in the TWW irrigated soils and in fact contributed for higher N uptake by switchgrass plants. This ultimately shows the alternative potential of TWW to supply N to bioenergy feedstocks grown on marginal lands. However, higher N content in bioenergy feedstock is not desirable as having high N can affect the efficiency of biomass conversion to biofuel during pyrolysis [44,45]. Nevertheless, the biomass N levels found in this study are closer to those reported by Aurangzaib et al. [23] in the same 'Alamo' switchgrass cultivar and by Wilson et al. [44] in an upland

Table 3

Switchgrass biomass yields (mean \pm SE) for two water and soil amendment treatments during the 2018 and 2019 cropping seasons.

Cropping year		2018	2019
Water type	Amendment	Biomass Yield (Mg ha^{-1})	
FW	NA	11.4 ± 2.59^a	10.5 ± 0.82
FW	GS	12.6 ± 2.01	11.8 ± 1.77
TWW	NA	10.7 ± 0.97	10.2 ± 1.23
TWW	GS	12.4 ± 2.46	9.1 ± 1.05
Source		$P > F$	
Year		0.28	
Water type		0.47	
Amendment		0.55	
Year \times water type		0.68	
Year \times Amendment		0.58	
Water type \times Amendment		0.72	
Year \times Water type \times Amendment		0.56	

^a Columns or rows with same or no letters indicate that there were no significant differences at $P < 0.05$, Tukey's test. FW: Freshwater; WW: Treated Wastewater; NA: No Amendment; GS: Gypsum + Sulfur.

Table 4Composition of switchgrass biomass for two water and soil amendment treatments at the end of the third year (2019 data) (mean \pm SE).

Biomass parameter	Nitrogen	Hemicellulose	Cellulose	Lignin	WSC	Ash	TEY
Water type (W)	—g kg ⁻¹ —	g ⁻¹					—L ha ⁻¹ —
FW	3.20 \pm 0.96 b ^a	30.9 \pm 1.16	39.1 \pm 1.13 a	7.32 \pm 0.26 a	1.72 \pm 0.15	4.76 \pm 0.54 a	4119 \pm 271
WW	5.68 \pm 0.92 a	28.5 \pm 2.31	35.2 \pm 0.80 b	6.28 \pm 0.44 b	2.67 \pm 0.59	6.48 \pm 0.77 b	3334 \pm 300
Amendment (A)							
NA	4.70 \pm 1.03 A	30.8 \pm 1.26 A	36.8 \pm 1.11	7.15 \pm 0.25	1.97 \pm 0.48	5.14 \pm 0.38	3740 \pm 279
GS	4.18 \pm 0.85 B	28.6 \pm 2.21 B	37.5 \pm 0.82	6.45 \pm 0.45	2.42 \pm 0.26	6.11 \pm 0.92	3712 \pm 384
Source			P > F				
W	<0.05	0.28	<0.05	<0.05	0.19	<0.05	0.25
A	<0.1	<0.1	0.43	0.11	0.41	0.14	0.95
W x A	0.83	0.89	0.6	0.72	0.87	0.82	0.47

^a Columns with same or no letters indicate that there were no significant differences between water type (lowercase) and amendment (uppercase) treatments at $P < 0.05$, Tukey's test. FW: Freshwater; WW: Treated Wastewater; NA: No Amendment; GS: Gypsum + Sulfur.

switchgrass cultivar.

3.2.2. Water soluble carbohydrates

Water soluble carbohydrates (simple soluble sugars) concentrations ranged from 1.5 to 2.9 g 100 g⁻¹ across two years, water types, soil amendment treatments (Table 4). Water type and amendment application did not cause any significant differences in WSC concentrations after three years of study. However, it should be noted that there was a trend of higher WSC concentrations in the switchgrass plants irrigated with TWW. This is contrasting to the premise that increase in salinity could reduce K uptake and negatively affect tissue rehydration processes that are crucial for sugar transport and metabolism [8]. Similarly, Chaganti et al. [8] also showed that the WSC concentrations in biomass sorghum did not reduce, even though soil salinity increased after wastewater application. Previously, Ganjegunte et al. [10] also reported that WSC concentrations in switchgrass biomass did not differ between water types and soil amendment treatments under greenhouse conditions, a result similar to the current study. No response of WSC to changes in salinity can be attributed to the higher salinity tolerance of switchgrass than the levels of soil salinity observed here, which ultimately did not affect the sugar metabolism. Nonetheless, the levels of WSC observed in switchgrass biomass in the Ganjegunte et al. [10] study are much higher (7–16 g 100 g⁻¹) than those observed in this study. In addition, other studies have also reported rather higher soluble sugar concentrations in switchgrass biomass than what was found in the current study [22,23,25,34]. Lower soluble sugar concentrations observed in this study can be attributed to the marginal nature of our soils and more importantly due to a delayed harvesting in late fall. The time of harvesting and plant maturity was shown to significantly influence non-structural carbohydrate dynamics in switchgrass plants with soluble sugar content decreasing as plants mature with time [23].

3.2.3. Hemicellulose and cellulose

Hemicellulose concentrations ranged between 29.7 and 32.0 g 100 g⁻¹ (Table 4) and did not significantly differ between the two water types. However, GS application significantly ($p < 0.1$) reduced hemicellulose concentration in switchgrass biomass. Mean hemicellulose concentrations of amended (GS) and non-amended treatments were 28.6 and 30.8 g 100 g⁻¹, respectively. On the other hand, cellulose concentrations fell between 34.6 and 39.3 g 100 g⁻¹ and significantly ($p < 0.05$) differed between the two water treatments with no effect of soil amendment application. Switchgrass irrigated with TWW had significantly lower cellulose concentrations relative to FW irrigated plants. Mean cellulose concentrations were 39.2 and 35.2 g 100 g⁻¹ for FW and TWW treatments, respectively. Increased salt stress associated with TWW and GS application likely resulted in lower holocellulose (hemicellulose + cellulose) concentrations in plant cell wall due to possible alterations in cellulose synthesis pathways [46]. Even though TWW or GS application reduced the levels of either cellulose or hemicellulose in switchgrass biomass, the levels observed in this study are well within the

range reported in several recent studies [10,22,23,25,47]. Higher cellulose and hemicellulose concentrations are more desirable in lignocellulosic biomass feedstocks during the biochemical conversion process for liquid bioethanol synthesis [48,49].

3.2.4. Lignin

Switchgrass biomass lignin concentration ranged between 5.9 and 7.6 g 100 g⁻¹ across two water types and soil amendment treatments (Table 4). Irrigating with TWW irrigation significantly ($P < 0.1$) decreased biomass lignin concentration in switchgrass biomass. Mean lignin concentrations were 7.3 and 6.3 g 100 g⁻¹ for FW and TWW treatments, respectively. On the other hand, soil amendment application did not cause any significant changes in biomass lignin levels. The lignin content observed in this study is consistent with those reported by some previous studies [10,23,50,51]. For example, in a study conducted by Aurangzaib et al. [23] in Iowa, lignin concentrations in lowland 'Alamo' cultivar increased in switchgrass biomass with delayed harvesting time but never exceeded 6 g 100 g⁻¹. Similarly, Lemus et al. [50] reported a mean biomass lignin concentration of 6.1 g 100 g⁻¹ when 20 switchgrass cultivars were evaluated for their biomass yield and quality in southern Iowa. In the same study, the lignin concentration in 'Alamo' cultivar was found to be 5.7 g 100 g⁻¹ [50]. Our results however, match very close to the Ganjegunte et al. [10] study, where biomass lignin concentrations fell in the approx. range of 4–7 g 100 g⁻¹ when the same 'Alamo' cultivar was grown under greenhouse experimental conditions. On the other hand, Yan et al. [51] and Alexander et al. [52] reported rather very high lignin concentrations in 'Alamo' switchgrass cultivar with concentrations exceeding 20 g 100 g⁻¹. Lignin is a complex cross-linked polymer and is highly recalcitrant that adds stability to the cell wall. However, its insolubility in water and as a "glue" between cellulose and hemicellulose, presence of higher lignin contents is a major barrier to bioethanol production [48,53] and therefore feedstock with low lignin content is preferred.

3.2.5. Ash

Switchgrass biomass ash contents ranged from 4.2 to 6.9 g 100 g⁻¹ across all treatments (Table 4). Irrigating with TWW significantly increased biomass ash content relative to FW treatment. Wastewater treatments had 36% higher ash content than their FW counterparts. Though soil amendment application did not have a statistically significant effect on biomass ash content, there was a definite trend observed where GS treatments had higher ash contents than non-amended treatments. Mean ash contents were 6.1 and 5.1 g 100 g⁻¹ for GS and non-amended treatments, respectively. Higher ash contents in TWW and GS treatments can be attributed to higher soil salinity observed in these soils, which resulted in higher mineral uptake by plants. Higher ash contents in switchgrass biomass under elevated salinity conditions were also reported by the Ganjegunte et al. [10] study. These authors also found higher ash contents under TWW irrigation than FW irrigation. Ash contents in this study ranged between 6.5 and 12 g 100 g⁻¹ in switchgrass

biomass [10]. However, it is important to note that the ash contents found in this study are consistent with those reported under non-saline conditions elsewhere [23,25,44,50]. For example, Tang et al. [25] reported ash contents ranging between 5.0 and 8.2 g 100 g⁻¹ in switchgrass biomass feedstock when grown on semi-arid lands. Similarly, Aurangzaib et al. [23] and Wilson et al. [44] also found ash contents decreasing to levels closer to 7.0 g 100 g⁻¹ as switchgrass plants matured. On the other hand, Yan et al. [51] and Ameen et al. [22] reported much lower concentrations of ash (~1.5–2 g 100 g⁻¹) in switchgrass biomass. Generally, lower ash concentrations are favored as higher ash contents reduce the efficiency of biochemical conversion process by reducing hydrocarbon yields while also increasing maintenance costs [23,54].

3.2.6. Theoretical ethanol production (TEP)

Theoretical ethanol production varied between 3334 and 4119 L ha⁻¹ (Table 4) among different treatments. Though TEP was lower in TWW treatment than FW, the differences were not statistically significant. Soil amendment application also did not cause any significant differences in ethanol production. Theoretical ethanol production is a function of biomass yield and WSC, cellulose and hemicellulose contents [22]. In this study, biomass yields were not significantly affected by either water type or amendment application though there were some effects observed on biomass composition. A strong positive correlation ($R = 0.94$) was also seen with biomass yields (Fig. 2), which indicates that TEP is highly dependent on biomass yields in addition to feedstock composition. A similar result was reported by Ameen et al. [22], where there was a strong correlation between TEP and biomass yields. However, the average TEP reported in the Ameen et al. study (2211 L ha⁻¹) was lower than the TEP (3726 L ha⁻¹) seen in this study, largely due to lower biomass yields [22] than what were seen in the current study. However, TEP levels observed in our study are closer to other studies, such as 3691, 3701, 3753, and 4273 L ha⁻¹ reported by Schmer et al. [34], Adler et al. [55], Scaglione-Mellor et al. [56], and Liu et al. [57], respectively.

3.3. Soil quality

3.3.1. Soil salinity

Temporal changes in soil salinity (EC_e) after application of two water types and soil amendments are presented in Fig. 3. Statistically significant effects of year ($P < 0.001$), watertype ($P < 0.001$) amendment ($P < 0.1$), and depth ($P < 0.001$) were observed on soil salinity. Also, there were some significant interactions between year \times water ($P < 0.001$), year \times amendment ($P < 0.05$), year \times depth ($P < 0.001$), watertype \times depth ($P < 0.01$) and year \times watertype \times depth ($P < 0.01$). After three years, regardless of irrigation water and soil amendments, soil salinity significantly increased across both the depths. On average soil salinity increased by 68% from 0.88 to 1.47 dS m⁻¹ in FW irrigated soils across both the depths. This is an expected result due to the arid climate of this region that is characterized by high temperatures and low precipitation and irrigation water application not exceeding evapotranspiration. This results in net salt accumulation in the soil profile due to inadequate salt leaching. Irrigation with TWW however, worsened this effect with higher increases in soil salinity observed relative to both FW and pre-study baseline soils (Fig. 3). Soil salinity across both the depths in TWW irrigated plots was twice as much as in FW irrigated plots and almost quadrupled after three years relative to baseline soils. Higher soil salinity associated with TWW irrigation can be attributed to its high dissolved salt concentrations (Table 1) [8,10,58–60] and therefore continuous application likely resulted in higher salt accumulation in the root zone.

On the other hand, gypsum and sulfur application significantly increased (Fig. 3) soil salinity across both the depths regardless of irrigation water type. This is not surprising as gypsum is a neutral “salt” and releases Ca²⁺ and SO₄²⁻ ions upon dissolution into soil solution and thus can potentially increase soil electrical conductivity transiently [61]. Highest soil EC_e values were observed consistently in the WW-GS treatment at both 0–15 (Figs. 3A) and 15–30 cm (Fig. 3B) depths with salinities reaching beyond the threshold of 4 dS m⁻¹. However, this cannot be considered as a negative result as gypsum application is largely considered beneficial due to Ca²⁺ release, which helps in maintaining soil structure and improve soil water infiltration, especially in soils affected by sodium [58,62–64]. Nevertheless, higher salinities in WW-GS treatment can be attributed to the cumulative effect of salt

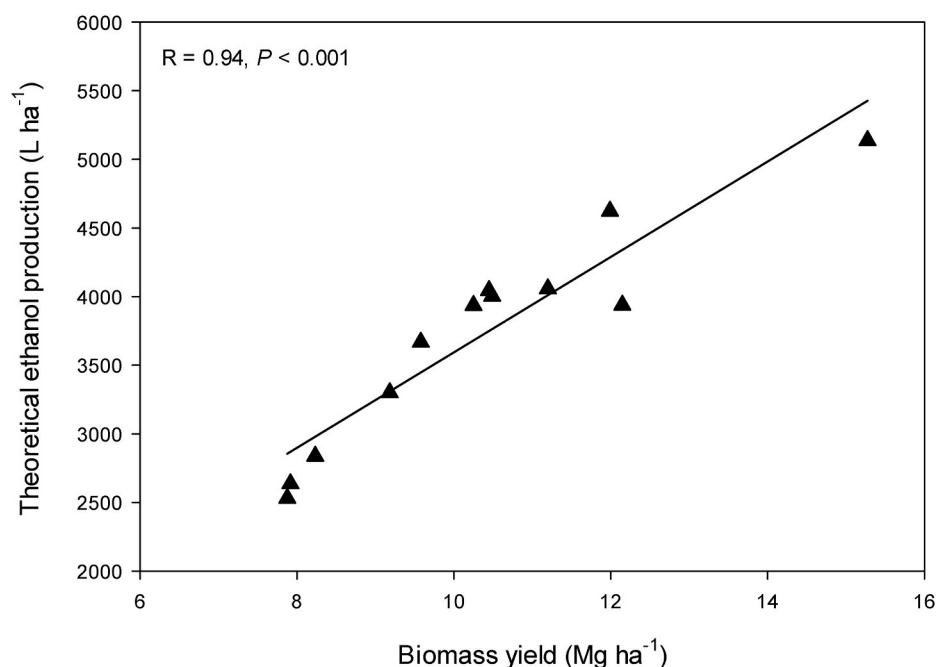


Fig. 2. Correlation between biomass yield and theoretical ethanol production (2019 data).

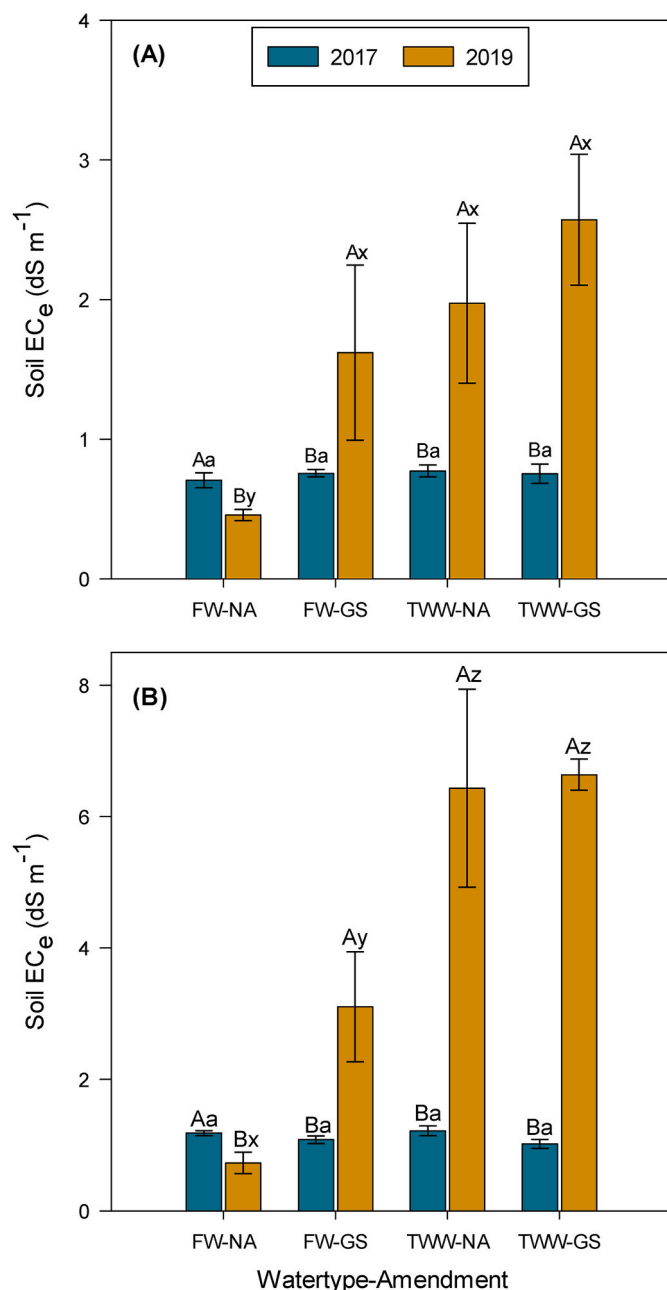


Fig. 3. Changes in soil salinity (EC_e) (mean \pm SE) at (A) 0–15 cm and (B) 0–30 cm soil depths as affected by water type and amendment combinations before (2017) and after the end (2019) of the experiment. Lower case letters indicate significant differences between treatment combinations within each year at $P < 0.05$, Tukey's test. Upper case letters indicate significant differences between years within each treatment combination at $P < 0.05$, Tukey's test. FW; Freshwater; TWW; Treated wastewater; NA: No amendment; GS: Gypsum + sulfur amendment.

accumulation through TWW and gypsum + sulfur application. Soil depth on the other hand, also had significant effect on soil salinity. In general, after three years, soil EC_e was significantly higher in the 15–30 cm depth compared to the upper 0–15 cm layer across the two water types and soil amendment treatments (Fig. 3A and B). This was likely due to downward leaching of salts by irrigation water [8]. More importantly, it should be noted that increase in soil salinity either by TWW application or gypsum + sulfur application did not cause significant yield reductions in switchgrass biomass. Linear regression also revealed no relationship between biomass yields and soil salinity (Fig. 5)

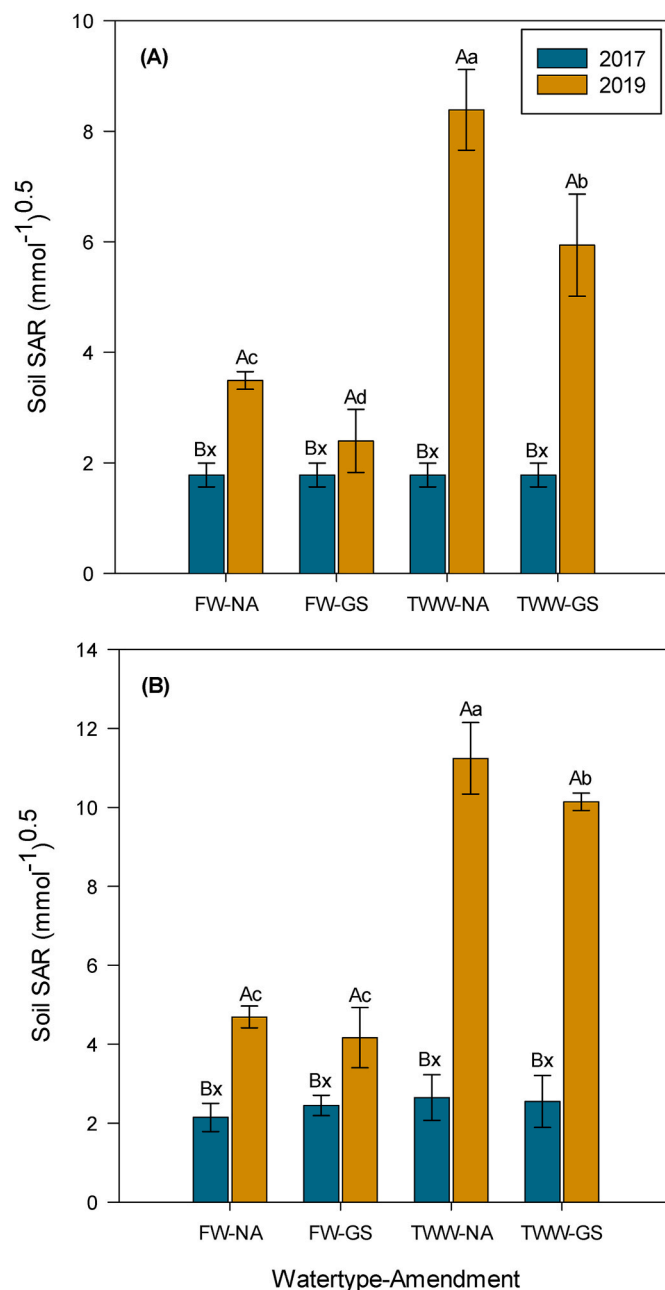


Fig. 4. Changes in soil sodicity (SAR) (mean \pm SE) at (A) 0–15 cm and (B) 0–30 cm soil depths as affected by water type and amendment combinations before (2017) and after the end (2019) of the experiment. Lower case letters indicate significant differences between treatment combinations within each year at $P < 0.05$, Tukey's test. Upper case letters indicate significant differences between years within each treatment combination at $P < 0.05$, Tukey's test. FW; Freshwater; TWW; Treated wastewater; NA: No amendment; GS: Gypsum + sulfur amendment.

proving that yields were not affected by changes in soil salinity. This is likely due to higher salinity threshold (~ 14 dS m⁻¹) of switchgrass [36] than the soil salinity levels observed in this study. This highlights that switchgrass as a bioenergy feedstock, can be successfully grown on marginal lands with TWW under elevated soil salinity.

3.3.2. Soil sodicity

Changes in soil sodicity (SAR) overtime at both depths are given in Fig. 4A and B. Individual effects of year ($P < 0.001$), water type ($P < 0.001$), amendment ($P < 0.05$) and depth ($P < 0.001$) were all

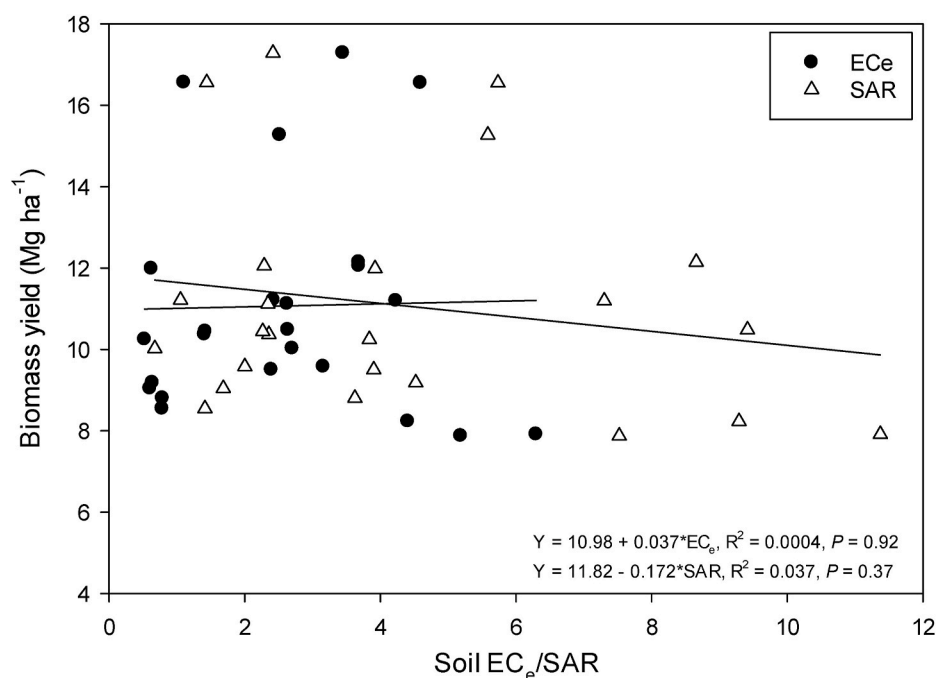


Fig. 5. Relationship between soil EC_e and SAR and switchgrass biomass yields. Soil EC_e and SAR data is across all water and soil amendment treatments averaged across both the depths (all data from 2018 to 2019 years).

statistically significant. There were also significant interactions observed between year \times watertype ($P < 0.001$), year \times depth ($P < 0.05$). Results show that soil sodicity generally increased after three years regardless of water type and amendment application. Soil SAR increased almost three times from 2.1 to 6.3 at the end of third year. Increase in SAR is generally due to the natural accumulation of Na^+ that is being added through irrigation waters that increases Na^+ ion concentration in soil solution relative to Ca^{2+} and Mg^{2+} ions. Moreover, “valence dilution” [58] could also increase monovalent cation concentration (specifically, Na^+), relative to divalent cations such as Ca^{2+} and Mg^{2+} that likely contributed for increase in soil SAR. After three years, the overall soil sodicity increased by approximately 3 times at both 0–15 and 15–30 cm depths (Fig. 4A and B) compared to pre-study baseline soils. At the same time, the relative increase in soil sodicity was much higher in the TWW irrigated soils compared to FW irrigated soils. This was an expected outcome as TWW contains high Na^+ concentration compared to FW (Table 1). Increase in soil SAR due to TWW application is a common result reported in the literature [8,10,65,66].

Application of gypsum and sulfur on the other hand, significantly reduced soil SAR in both FW and TWW irrigated plots after three years when compared to non-amended plots (Fig. 4). Gypsum is a well-known traditional soil amendment applied to correct soil sodicity as it readily supplies Ca^{2+} to counter Na^+ in the soil solution or on exchange complex and aids in maintaining soil structure and water permeability [64]. Therefore, lower soil SAR values observed here are due to gypsum solubilization and enrichment of soil solution with Ca^{2+} that facilitated Na^+ removal and its leaching into deeper layers. This is further substantiated by higher soil SAR values generally seen in the lower 15–30 cm depth than at 0–15 cm (Fig. 4A and B). Regardless, soil SAR reduced by 20% and 18% in the FW-GS and WW-GS treatments across both the depths, respectively, relative to their non-amended treatments. These results are in conjunction with those reported by Chaganti et al. [8], who also reported significant reductions in soil SAR with gypsum application in bioenergy sorghum plots. Similarly, in a greenhouse study, Ganjgunte et al. [10] also reported higher SAR values in untreated TWW irrigated soils and at lower soil depths due to Na^+ leaching. Many other studies have also reported significant reductions in soil SAR after gypsum application [58,67–70]. Overall, these results highlight that

long-term application of TWW can increase soil sodification, but application of gypsum can help keep soil SAR values well below the threshold of 13, above which the effects of sodium on soil structure become more apparent. More importantly, increased soil sodicity did not affect switchgrass biomass yields as regression analysis did not show any significant trend between soil SAR and biomass yields (Fig. 5).

4. Conclusions

Our results show that biomass yields and consequently, theoretical ethanol production, was not affected by TWW irrigation, although there were some changes in biomass quality. Results also indicate that TWW can exacerbate soil salinity and sodicity. Sodicinity from TWW can negatively affect soil quality over long-term. However, gypsum application negated soil sodicity suggesting that an appropriate soil management practice should be in place to mitigate sodic hazard of TWW. Nevertheless, increase in soil salinity and sodicity did not negatively affect switchgrass biomass production, which likely is due to higher salinity tolerance of switchgrass than the levels observed in this study. In conclusion, our results are novel in that they will have important implications in diversifying cropping pattern in arid west Texas while also extending acreage of bioenergy crops into marginal lands of this region, without competing for arable lands used in food production. We believe that these results are also pertinent and could help diversify crop regimes in arid and semi-arid regions elsewhere in the world, where water availability and salinity issues are prevalent. Above all, use of TWW for irrigating such bioenergy crops, helps extend the fresh water supplies to more demanding sectors and results in efficient water reuse for producing clean energy.

Data availability statement

Data is available from the corresponding author upon reasonable request.

Author contributions

Vijayasatya N. Chaganti: Data collection, Formal analysis, Writing –

writing, review & editing. Girisha Ganjegunte: Conceptualization, experimental design, study implementation, Writing – review & editing. Manyowa N. Meki: Conceptualization, Writing – review & editing. James R. Kiniry: Conceptualization, Writing – review & editing. Genhua Niu: Conceptualization, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This project was supported by the South-Central Sun Grant Initiative administered through Oklahoma State University and USDA Project No. 2017-68007-26318 led by TWRI, through the National Institute for Food and Agricultural's Agriculture and Food Research Initiative. Part of Dr. Ganjegunte's salary was supported by USDA-NIFA Hatch project (Accession No. 1001806 and Project number: TEX0-1-9162). The authors thank John Clark, Carlos Castro, Sahara Jordan, Luis Ramirez, Priscilla Reyes, Andres Gomez and Mariana Munoz for assistance in field and laboratory work. The authors would also like to specially thank the anonymous reviewers for their valuable comments that helped improve this manuscript.

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