Agricultural fertiliser from brewery effluent – the recovery of nutrients from the biomass of activated sludge and high rate algal pond treatment systems

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ABSTRACT

The disposal of waste biomass generated from biological wastewater treatment plants is a costly process and poses environmental threats to the receiving environment. This study aimed to determine the suitability of algae and waste activated sludge (WAS) produced from a brewery effluent treatment system as a fertiliser in agriculture. The change in soil characteristics and the growth of a crop fertilised with algae or WAS was compared with a conventional inorganic fertiliser. Swiss chard plants (Beta vulgaris) fertilised with anaerobically digested (AD) algae or WAS had a significantly higher mean biweekly yield (5.08 ± 0.73 kg/m²) when compared with the inorganic fertiliser control (3.45 ± 0.89 kg/m²; p < 0.0001). No difference was observed in the soil’s physical fertility when algae or WAS were applied to the soil (p > 0.05). The nitrogen applied to the soil from algae and WAS biomass appeared to leach out of the soil less than the nitrogen supplied by inorganic fertilisers. The application of WAS or algae on soil increased the soil’s sodium concentration and sodium absorption ratio from 774.80 ± 13.66 mg/kg to 952.17 ± 34.89 mg/kg and 2.91 ± 0.04 to 3.53 ± 0.13, respectively. Regulations on the application of algae or WAS on agricultural soils should be altered to consider the limit values for sodium.

Key words | agriculture, anaerobic digestion, biosolids, brewery effluent, high rate algal ponds, waste activated sludge (WAS)

HIGHLIGHTS

• Swiss chard production was similar when fertilised with fertiliser, waste activated sludge or algae.
• Anaerobically digested waste activated sludge and algae can replace inorganic fertiliser.
• Waste activated sludge and algae increased the soil’s sodium absorption ratio.
• Soil application rates of sludge should consider sodium limit values.

INTRODUCTION

Organic and inorganic pollutants are converted into biomass during the treatment of wastewater in biological systems such as high rate algal ponds (HRAP) and activated sludge (AS; Tchobanoglous et al. 2003). This biomass (biosolids) is separated from the treated effluent and disposed of. The disposal of waste biomass is a costly process and poses environmental threats if it is not carried out correctly (Hospido et al. 2010). Tchobanoglous et al. (2005) state that the ‘management of the solids and concentrated contaminants removed by effluent treatment has been, and continues to be, one of the most difficult and expensive problems in the field of wastewater engineering’. Practices and technologies need to be
developed that allow the sustainable disposal of biomass, especially where the recovery or reuse of resources trapped in the biomass are considered (Singh & Agrawal 2008). Wastewater biosolids are nutrient-rich organic solids which have the potential to be used for fertiliser, soil amendment or energy production (Dobhal & Singhal 2017; Elalami et al. 2020).

Biosolids can supplement or replace commercial fertilisers as they contain nitrogen, phosphorus and various micro-nutrients such as copper, zinc, molybdenum, boron, calcium, iron, magnesium, and manganese (Singh & Agrawal 2008; Engida et al. 2020). The organic matter content can be used to increase the cation exchange capacity (CEC) of the soil and can aid in amending soils that are carbon-depleted (Engida et al. 2020), and thus can potentially improve crop production. For example, the yield of maize, amaranthus, cowpea and crossandra fertilised with sewage sludge was the same as, or higher than, that of plants fertilised with a commercial inorganic fertiliser (Chideshwari et al. 2002; Engida et al. 2020). The yield of barley fertilised with waste activated sludge (WAS) applied to soils at 15 t/ha dry weight was significantly higher than barley fertilised with a commercial inorganic fertiliser (Antolín et al. 2005). After four years of annual application of WAS from a sewage treatment facility, the soil’s CEC, total organic carbon and available nitrogen increased significantly; however, so did the heavy metal concentration in the harvested grain (Antolín et al. 2005).

The majority of studies have reported an improvement in physical properties when WAS is applied to soils (Epstein 1975; Tsadilas et al. 1995; Lu et al. 2012). The application of WAS to soils increases its organic matter content, which aids in stabilising the soil structure by increasing interparticulate cohesion within aggregates and enhancing their hydrophobicity (Diacono & Montemurro 2010). Municipal solid waste applied to the soil, every two years, increased soil aggregate stability by 29%, thus increasing its resistance to erosion (Annabi et al. 2007). The application of sewage sludge to the soil has been shown to improve its water holding capacity, porosity and the bulk density of the soil (Epstein 1975; Ojeda et al. 2005).

Anaerobic digestion is a recommended stabilisation step prior to the land disposal of waste sludge as it allows the recovery of carbon into an energy source (biogas) and can increase the availability of nutrients to plants (Tchobanoglous et al. 2003). This process results in the conversion of protein-bound nitrogen to ammonia which can be utilised by plants and can increase the nutrient availability of biosolids. Anaerobically digested WAS had higher phosphorus availability than heat-dried WAS (Lyberatos et al. 2004). To date, there is only one publication which compares the use of AD to increase the nutrient availability of WAS to plants (Warman & Termeer 2005). Sludge pre-treated using AD resulted in a higher yield of Zea mays when compared with sludge that was composted (Warman & Termeer 2005). No other current literature has compared the use of AD to possibly increase the fertiliser value of effluent-grown algae and to document the subsequent effect on soil fertility.

High rate algal ponds and AS are effective brewery effluent treatment technologies which both produce a biomass than can be used in agriculture. The suitability of these biomasses as a fertiliser needs to be assessed and can aid in deciding which technology is favourable in a particular situation. Algae have resilient cell walls which can decrease their decomposition rate and thus reduce the availability of nutrients to plants when compared with WAS (Markou et al. 2012). Algal biomass cultured in piggery wastewater significantly increased the growth of wheat when compared with unfertilised treatments (Jenkins et al. 2017), however this was a pot experiment and only carried out for four weeks. This is the first study which compares the use of algae and WAS as a fertiliser replacement, where both the algae and the WAS are produced from effluent treatment systems that have been used to treat the same effluent.

Aims and objectives

The aim of this study was to determine the suitability of algae and WAS produced from a brewery effluent treatment system as a fertiliser in agriculture. This was done by comparing the change in soil characteristics and the growth of a crop fertilised with algae or WAS with a conventional inorganic fertiliser. This study will also determine what effect an AD pre-treatment step may have on the fertiliser quality and the fertility of the receiving soil.

The objectives of this study were to:

- compare plant growth and soil characteristics of plots fertilised with WAS, algae or inorganic fertiliser;
• compare plant growth and soil characteristics between soils fertilised with anaerobically digested and non-anaerobically digested algae or WAS; and
• determine the suitability of anaerobically digested and non-anaerobically digested WAS or algal biomass as an inorganic fertiliser replacement.

MATERIALS AND METHODS

Untreated brewery effluent was screened through a 500 μm drum filter (Autrex Industrial Screening, serial no. A 140/02, model no. R 015) to remove solids and then anaerobically digested in an up-flow anaerobic sludge blanket reactor. After AD the effluent was polished in a commercial AS system or a pilot HRAP system.

The majority of the post-AD effluent (800 m³/d) was treated in an AS system consisting of a rectangular aeration basin and clarifier operated by an independent commercial company (Proxa Pty Ltd, South Africa). The aeration basin was operated at a hydraulic retention time (HRT) between 0.4 and 0.5 days, while the dissolved oxygen concentration was maintained around 0.8 mg/l. Effluent was decanted from the aeration basin into a clarifier operated at an HRT between 0.3 and 0.4 days, which separated the suspended solids from the treated effluent. Settled sludge from the clarifier was also disposed of into the municipal sewage works,

TREATMENTS

Five treatments were prepared by adding the following soil amendments/fertilisers to a sandy loam top-soil (10% silt, 20% clay, 70% sand; Macvicar et al. 1977): (1) un-digested algae (algae); (2) anaerobically digested algae (AD-algae); (3) un-digested WAS (WAS); (4) anaerobically digested WAS (AD-WAS); and (5) a commercial inorganic fertiliser (inorganic-fertiliser) that served as a reference control (Hygrotech Pty Ltd, South Africa; registration number K5709; Act 36 of 1947). These fertilisers were applied to the soil at a nitrogen application rate equivalent to 80 kg/ha (Laboski & Peters 2012), two days before the soil was planted, and they were mixed into the top five centimetres of the soil.

Two semi-continuously-fed anaerobic digesters were used to digest the settled WAS and algal biomass. Both were made of plastic drums with a total volume of 220 l, a head space of 60 l and an operating volume of 160 l. They were stirred with a submersible mixer (Sobo, WP 400M, South Africa) for five minutes every half an hour, and were situated in a temperature-controlled room (37 °C). The digesters were seeded with sludge obtained from a biogas-producing up-flow anaerobic sludge blanket reactor at Ibhayi Brewery. They were fed once a day following the removal of the equivalent volume of digestate. The settled algae and WAS fed to the digester had a total solids content of 25 g/l. Each digester was fed 9.5 l of either algae or WAS per day resulting in a feeding rate of 1.5–2.0 g of TS/1 reactor/d
and an HRT of 16 d. These two digesters were operated for 60 days, before the digestate was collected and applied to the soil as a fertiliser.

**EXPERIMENTAL SPECIES, SYSTEM AND IRRIGATION**

Swiss chard (*Beta vulgaris* cv. Fordhoek giant) seedlings were purchased from a commercial nursery (Moorland Seedlings Pty Ltd, Humansdorp, South Africa). They were planted at a density of 16 plants per square metre, in raised beds (1.0 × 1.0 m, with a soil depth of 0.7 m), that were filled with the amended soils. Each soil amendment treatment was randomly assigned to three raised beds, such that the treatments were replicated three times with a replicate consisting of a single raised bed.

They were irrigated once a day, except during rain or directly thereafter (Laboski & Peters 2012), and received a total of 377 mm of water over the 13-week growth trial (275 mm irrigation and 102 mm of rain). Plants were irrigated via a drip irrigation system receiving a maximum of 5 mm of water per irrigation to minimise leaching (Laboski & Peters 2012).

**DATA COLLECTION**

A sample of the un-digested and anaerobically digested WAS and algae that were applied to the soil were subject to elemental analysis (inductively coupled plasma mass-spectrometry and X-ray fluorescence) at an independent laboratory (Central Analytical Facilities, Stellenbosch University, South Africa).

At the beginning of the trial, ten plants were randomly taken from the population of seedlings that were used for the experiment, and were weighed to determine the mean wet starting mass (0.1 g). After five weeks, the plants were ready for harvesting, when all the large, fully expanded leaves from each plant were removed and the wet weight weighed (0.1 g). This was repeated every two weeks until the experiment was terminated, after 13 weeks. At the end of the trial, all the above-ground biomass was harvested and wet weight weighed (0.1 g).

At the beginning of the trial three plants were randomly chosen and used for leaf chemical analysis. These plants were not used in the experiment due to the destructive nature of the sampling. At the end of the trial leaves from each plot were collected and used for chemical analysis. All samples were analysed for aluminium, calcium, copper, iron, manganese, magnesium, nitrogen, phosphorus, potassium, sodium and zinc concentrations at an analytical laboratory (inductively coupled plasma mass-spectrometry; Cedara, Department of Agriculture, Kwa-Zulu Natal, South Africa).

Daily temperature and rainfall data were recorded using a rainfall gauge and a thermometer (Hanna, HI 991300, United Kingdom) which were situated next to the experimental area.

Infiltration rate was measured in each replicate at the beginning and end of the trial. Infiltration rates were determined using a 30-cm-diameter ring infiltrometer placed 15 cm into the soil with 12 cm protruding above the surface. Five litres of water were poured into the ring infiltrometer and the time taken for the water to drain into the soil was recorded (ISO 2017). Infiltration rate was then calculated (Equation (1), ISO 2017):

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\text{Infiltration rate (cm/min)} = \frac{\text{volume of water added/surface area}}{\text{time}}
\] (1)

Air-filled porosity (AFP), bulk density and water holding capacity were measured in each replicate at the beginning and end of the trial (ISO 2017). The apparatus used was a 110 mm plastic pipe with an end cap that had four 3.0 mm holes drilled into it. The pipe was bored into the soil to get an undisturbed soil sample. A gauze was placed over the top of the vessel and submerged in water to just above the surface of the soil, for 60 min. The holes in the bottom were then sealed and the vessel was moved into a tray, where the holes were unblocked. The vessel was left to drain for 30 min and the amount of water collected was measured. Air-filled porosity was calculated using Equation (2) (ISO 2017). Directly after the AFP test the vessel was placed in a drying oven at 105 °C and allowed to dry for a minimum of 24 h, until a constant mass was achieved. Water holding capacity was calculated using Equation (3)
and bulk density was then calculated using Equation (4) (ISO 2017).

Air filled porosity (%) = \((\text{volume drained}/\text{volume of soil}) \times 100\)

Equation (2)

Water holding capacity (%) = \(((\text{wet weight} – \text{dry weight})/\text{volume}) \times 100\)

Equation (3)

Bulk density (g/cm\(^3\)) = \(\text{dry weight}/\text{volume}\)

Equation (4)

Soil aggregate stability was measured, in each replicate, at the beginning and end of the experiment using five grams of 2–5 mm aggregates (Le Bissonnais 1996). Samples were placed in distilled water and allowed to stand for ten minutes. The distilled water was then removed with a pipette and the aggregates transferred onto a 0.05 mm sieve which was immersed in ethanol and shaken five times with a gentle regular helical rotation movement. The >0.05 mm aggregates on the sieve were collected, dried at 40 °C, and then gently sieved using a column of six sieves: 2.00, 1.00, 0.50, 0.20, 0.10, and 0.05 mm (Le Bissonnais 1996). The aggregate stability was represented by the mean weight diameter (MWD) of aggregates and was calculated using Equation (5) (Le Bissonnais 1996): Mean weight diameter = \(\sum (d \times m)/100\)

Equation (5)

where \(d\) was the mean diameter between the two sieves (mm) and \(m\) was the weight fraction of aggregates remaining on the sieve (%).

The chemical analysis of the soil was determined at the start of the trial (\(n = 5\)). At the end of the trial, a sample was taken from the bed of each replicate for soil chemical analysis. Soil samples were sent to a commercial analytical laboratory and analysed for pH, cation exchange capacity (CEC), carbon, calcium, copper, potassium, phosphorus, sodium, magnesium, manganese, nitrogen and zinc (Ambic-2-extractable and KCl-extractable; Cedara, Department of Agriculture, Kwa-Zulu Natal, South Africa). The sodium adsorption ratio (SAR) of the soil was calculated using Equation (6), where sodium, calcium, and magnesium are expressed in milliequivalents per litre (meq/l), obtained from a saturated paste soil extract (Qadir et al. 2003):

\[\text{Sodium adsorption ratio} \quad \text{Na}^+ \div \sqrt{\frac{\text{Ca}^{2+} + \text{Mg}^{2+}}{2}}\]

Equation (6)

Samples of anaerobically digested and un-digested WAS or algae applied to the soil were tested for Escherichia coli. Similar analyses were repeated on soil and leaf samples taken from each replicate every four weeks at the Ibhayi Brewery laboratories (IS 17994 method; ISO 2014).

**STATISTICAL ANALYSIS**

The experimental design allowed for: (1) a multifactor analysis of variance (ANOVA) where the treatments included two soil-amendments (algae and WAS), both of which were either subject to AD or left un-digested; and (2) a one-way ANOVA that included a comparison of the four treatments described above and a fifth inorganic-fertiliser treatment that acted as the reference-control. If no significant interactions were observed between soil-amendment factor and/or AD pretreatment factor (multifactor ANOVA), then the statistical analysis generated from the one-way ANOVA/Kruskal–Wallis ANOVA was used. All analyses were carried out at \(p < 0.05\) and, when differences were found, a Tukey’s multiple range analysis was used at \(p < 0.05\). Data collected over the course of the trial were compared using multifactor repeated measures or one-way ANOVA or a non-parametric Mauchly’s sphericity test, if ANOVA assumptions were not met \((p < 0.05)\). All data were checked for equality of variance and for the normal distribution of the residuals using Levene’s test and a Shapiro–Wilk plot of the residuals, respectively. If the assumptions were not met, then the data were log or square-root transformed and checked for equal variance and normal distribution of residuals. If the assumptions were still not met, a non-parametric Mann–Whitney \(U\) test or a Kruskal–Wallis ANOVA was used to compare the data between treatments. All analyses were performed using a statistical software package (Statistica Version 10, StatSoft Inc., Tulsa, USA). Statistical analysis on pH data was done using hydrogen ion concentration.
RESULTS

WAS and algal biomass applied to the soil were within the heavy metal limit for application to agricultural land (Table 1; DWAF 1997). WAS biomass had a chromium concentration between 243.5 and 223.3 mg/kg, which was below the land application limit. Algal biomass had a four-times higher chromium concentration (1,043.7 – 1,057.1 mg/kg) than WAS biomass (Table 1). Mercury concentrations in algal and WAS biomasses were below 3.5 mg/l. All results came back negative for the presence of E. coli in algae, sludge, soil and on the plant leaves.

Harvested Swiss chard biomass was not influenced by an interaction between fertiliser treatment and time (repeated measures ANOVA, $F_{(12,30)} = 0.85$, $p = 0.6045$) or by an interaction between biomass type (algae vs WAS) and AD pre-treatment (repeated measures multifactor ANOVA, $F_{(3,24)} = 1.54$, $p = 0.2288$). However, there was a significant difference in biomass harvested between fertiliser treatments, with a higher average biomass harvested from algae and WAS fertiliser treatments when compared with the inorganic-fertiliser treatment (Figure 1; repeated measures ANOVA, $F_{(4,10)} = 48.62$, $p < 0.0001$). The mean fortnightly yield from the inorganic-fertiliser treatment was 3.45 ± 0.89 kg/m$^2$ while algae and WAS treatments had a similar yield of 5.08 ± 0.73 kg/m$^2$ (Figure 1).

The chemical concentration of plant leaves was not influenced by an interaction between factors of biomass type (WAS vs algal) and AD pre-treatment (multifactor ANOVA, $p > 0.05$). Similarly, no significant differences were observed in leaf elemental concentration between biomass type and AD pre-treatment (multifactor ANOVA, $p > 0.05$). There was also no significant difference in the nitrogen concentration of Swiss chard leaves cultivated under the five fertiliser treatments at the end of the trial (Table 2); although the leaf nitrogen concentration in all the algae and WAS treatments increased significantly from the start to the end of the trial, those of the inorganic-fertiliser treatment did not increase significantly compared with those from those at the start (Table 2; Kruskal–Wallis, $p > 0.65$, $F_{(5,20)} = 11.80$, $p = 0.0376$). The potassium and phosphorus concentrations of Swiss chard plants were similar between all fertiliser treatments (Table 2; Kruskal–Wallis, $p > 0.05$). The iron concentration in leaves from the inorganic-fertiliser treatment was significantly lower than that of leaves from the algae and WAS treatments (Table 2; Kruskal–Wallis, $p > 0.05$). The iron concentration in leaves from the algae or WAS treatments subject to the algae or WAS fertiliser treatments (Table 2). Except for iron, there was no significant difference in all the other nutrient (calcium, magnesium, sodium, zinc, copper, manganese and aluminium).

### Table 1

| Parameter          | Limit          | Inorganic fertiliser | Algae  | AD-algae | WAS   | AD-WAS |
|--------------------|----------------|----------------------|--------|----------|-------|--------|
| Nitrogen (g/kg)    | N/A            | 68                   | 53.9   | 51.3     | 52.5  | 50.4   |
| Phosphorus (g/kg)  | N/A            | 42                   | 15.5   | 13.2     | 20.5  | 21.4   |
| Potassium (g/kg)   | N/A            | 208                  | 5.4    | 6.6      | 4.6   | 3.5    |
| Cadmium (mg/kg)    | 15.7           | –                    | 0.6    | 1.8      | 2.6   | 0.9    |
| Cobalt (mg/kg)     | 100.0          | –                    | 6.5    | 4.0      | 2.0   | 2.5    |
| Chromium (Cr$^{3+}$) (mg/kg) | 1,750.0 | –                   | 1,043.7 | 1,057.1 | 243.5 | 223.3 |
| Copper (mg/kg)     | 50.5           | 22                   | 38.1   | 41.6     | 36.3  | 48.8   |
| Mercury (mg/kg)    | 10.0           | –                    | <3.5   | <3.5     | <3.5  | <3.5   |
| Nickel (mg/kg)     | 200.0          | –                    | 59.2   | 63.1     | 45.4  | 55.3   |
| Lead (mg/kg)       | 50.5           | –                    | 10.9   | 19.7     | 10.3  | 21.4   |
| Arsenic (mg/kg)    | 15.0           | –                    | 2.0    | 8.6      | 3.2   | 7.6    |
| Selenium (mg/kg)   | 15.0           | –                    | 0.3    | 1.3      | 0.8   | 1.4    |

Anaerobically digested algae (AD-algae); anaerobically digested WAS (AD-WAS).
concentrations in the leaves between all five fertiliser treatments (Table 2; one-way ANOVA/Kruskal–Wallis, \( p > 0.60 \)). The final aluminium, zinc, copper and phosphorus concentrations of the leaves were significantly lower than their starting concentrations, for all fertiliser treatments (Table 2; Kruskal–Wallis, \( p < 0.05 \)).

The physical properties of the soil were not influenced by an interaction between factors of biomass source and AD pre-treatment (multifactor ANOVA, \( p > 0.05 \)). After 13 weeks, there was also no significant difference in the bulk density, porosity, water holding capacity, infiltration rate and mean weight diameter of soils subject to the five...
fertiliser treatments (Table 3; one-way ANOVA/Kruskal–Wallis, \( p > 0.05 \)). The combined mean porosity, water holding capacity and mean weight diameter of the soils were 33.90\% \pm 0.71\%, 22.57\% \pm 0.72\% and 1.57 \pm 0.27 mm, respectively (Table 3).

All measured soil chemical concentrations were not influenced by an interaction between biomass type and AD pre-treatment (multifactor ANOVA, \( p > 0.05 \)). The zinc concentration of the soil was significantly higher in the algae-fertilised soils when compared with WAS-fertilised soils (Table 4; multifactor ANOVA, \( F_{(1,8)} = 85.51, \ p = 0.0001 \)). No other significant difference was observed between soil chemical concentration and biomass source or AD pre-treatment step (multifactor ANOVA, \( p > 0.05 \)).

The soil pH, exchangeable acidity and CEC were similar between all fertiliser treatments (Table 4; one-way ANOVA/Kruskal–Wallis, \( p > 0.05 \)). The soil phosphorus and potassium concentrations were higher in inorganic-fertilised soils compared with algae or WAS fertiliser soils (Table 4; one-way ANOVA, \( F_{(5,14)} = 28.35, \ p < 0.0001 \)). After 13 weeks, the calcium, copper, magnesium and manganese concentrations of the soil were similar between all five fertiliser treatments (Table 4; one-way ANOVA/Kruskal–Wallis, \( p > 0.05 \)). Soil fertilised with inorganic fertiliser had a higher potassium concentration than soils fertilised with algae or WAS (Table 4; one-way ANOVA, \( F_{(5,14)} = 4.20, \ p = 0.0156 \)). The SAR and sodium concentrations were significantly higher in algae and WAS fertiliser soils when compared with soils fertilised with inorganic fertiliser (Table 4; one-way ANOVA, \( p < 0.05 \)). Algae and WAS fertilised soils had a similar sodium concentration and SAR with a combined mean of 952.17 \pm 34.89 mg/kg and 3.53 \pm 0.13, respectively (Table 4). Inorganic fertiliser and WAS-fertilised soils had a similar zinc concentration (Table 4; one-way ANOVA \( p > 0.05 \)).

At the end of the trial, the carbon (54.46 \pm 0.92 g/kg) and nitrogen (3.42 \pm 0.05 g/kg) concentrations of soils fertilised with algae or WAS were significantly higher than those of soils fertilised with inorganic fertiliser (C: 50.01 \pm 1.43 and N: 3.12 \pm 0.06 g/kg; one-way ANOVA, \( p < 0.05 \)). For both elements, the concentrations of soils fertilised with AD-WAS or algae were significantly lower than those of soils fertilised with undigested WAS or algae (multifactor ANOVA, \( p < 0.05 \); Table 5).

### DISCUSSION

Biosolids have the potential to supplement or replace commercial fertilisers as they contain nitrogen, phosphorus and various micro-nutrients needed to support plant growth (Singh & Agrawal 2008). In this study, the yield of Swiss chard plants cultivated in soil fertilised with WAS or algae was significantly higher than the yield from inorganic-fertiliser treatments. Similarly, the yields of various crops fertilised with WAS were similar to crops fertilised with a commercial fertiliser (Chitdeshwari et al. 2002; Warman & Termee 2005; Singh & Agrawal 2008). However, barley fertilised with sewage sludge had a lower yield when compared with inorganic fertiliser, but the addition of inorganic potassium to the sludge increased the yield of barley, indicating that the sludge was deficient in potassium (Miah et al. 1999). The present study shows that WAS and algae can be utilised as an inorganic fertiliser replacement when applied to the soil at the same nitrogen loading rate and can even increase crop yield.

| Parameter                  | Start      | Algae      | AD-algae   | WAS        | AD-WAS     | Inorganic-fertiliser | F/H value | P value |
|----------------------------|------------|------------|------------|------------|------------|----------------------|-----------|---------|
| Bulk density (g/cm³)       | 1.42 \pm 0.01 | 1.41 \pm 0.06 | 1.44 \pm 0.03 | 1.35 \pm 0.06 | 1.38 \pm 0.03 | 1.42 \pm 0.04 | 2.303     | 0.0764  |
| Porosity (%)               | 36.17 \pm 0.67 | 33.48 \pm 1.57 | 34.08 \pm 2.19 | 36.61 \pm 1.62 | 33.75 \pm 0.88 | 31.55 \pm 0.72 | 1.020     | 0.4301  |
| Water holding capacity (%) | 22.43 \pm 0.68 | 23.01 \pm 0.98 | 20.49 \pm 1.35 | 22.79 \pm 1.65 | 24.99 \pm 2.49 | 21.58 \pm 0.99 | 0.947     | 0.4691  |
| Infiltration rate (cm/min) | 0.80 \pm 0.02 | 0.82 \pm 0.11 | 0.92 \pm 0.10 | 0.95 \pm 0.04 | 0.72 \pm 0.03 | 0.82 \pm 0.05 | 8.367     | 0.2127  |
| Mean weight diameter (mm)  | 1.56 \pm 0.09 | 1.55 \pm 0.04 | 1.67 \pm 0.05 | 1.49 \pm 0.05 | 1.56 \pm 0.06 | 1.59 \pm 0.10 | 0.788     | 0.5782  |

Anaerobically digested algae (AD-algae), anaerobically digested WAS (AD-WAS).

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Table 3 | The mean (±standard error) starting and final physical characteristics of soils subject to the different fertiliser treatments (one-way ANOVA/Kruskal–Wallis, \( p < 0.05 \))
Table 4 | The mean (± standard error) chemical characteristics of soils subject to the different fertiliser treatments

| Parameter                        | Start   | Algae   | AD-algae | WAS     | AD-WAS  | Inorganic-fertiliser | F/H value | P value |
|----------------------------------|---------|---------|----------|---------|---------|----------------------|-----------|---------|
| pH                               | 7.60 ± 0.03a | 7.63 ± 0.051 | 7.53 ± 0.041 | 7.65 ± 0.031 | 7.64 ± 0.051 | 7.57 ± 0.051 | F = 0.67 | 0.6502  |
| Exchangeable acidity (cmol(+)/kg) | 0.09 ± 0.00a | 0.09 ± 0.011 | 0.09 ± 0.011 | 0.10 ± 0.011 | 0.09 ± 0.011 | 0.10 ± 0.00a | H = 1.69 | 0.8900  |
| CEC (cmol(+)/kg)                 | 26.52 ± 0.34a | 27.97 ± 0.1251 | 29.57 ± 0.3891 | 25.90 ± 0.451 | 28.23 ± 0.791 | 24.22 ± 0.361 | H = 10.23 | 0.0691  |
| Phosphorus (mg/kg)               | 37.50 ± 0.21a | 55.93 ± 0.0303b | 57.26 ± 0.0307b | 58.37 ± 0.0379b | 56.21 ± 0.0236b | 79.79 ± 0.0361c | F = 28.35 | <0.0001 |
| Potassium (mg/kg)                | 618.00 ± 17.87a | 691.33 ± 52.27a | 651.33 ± 19.38a | 653.33 ± 3.80a | 648.33 ± 3.29a | 826.63 ± 50.60b | F = 4.20  | 0.0156  |
| Calcium (g/kg)                   | 4.49 ± 0.04a | 4.79 ± 0.008a | 4.64 ± 0.0071 | 4.51 ± 0.0012 | 4.75 ± 0.0091 | 4.64 ± 0.00151 | F = 1.78  | 0.1812  |
| Copper (mg/kg)                   | 1.16 ± 0.08a | 1.33 ± 0.0012a | 1.17 ± 0.0071 | 1.27 ± 0.0012 | 1.43 ± 0.0012 | 1.20 ± 0.0061 | F = 1.22  | 0.3499  |
| Magnesium (mg/kg)                | 518.80 ± 17.06a | 544.00 ± 16.46a | 516.67 ± 22.591 | 506.67 ± 0.9351 | 515.67 ± 29.541 | 517.00 ± 18.011 | F = 0.37  | 0.8632  |
| Manganese (mg/kg)                | 30.80 ± 0.97a | 51.67 ± 0.291b | 47.33 ± 0.19b | 48.67 ± 0.145b | 46.33 ± 0.0636b | 53.00 ± 0.0351b | H = 12.28 | 0.0311  |
| Sodium (mg/kg)                   | 774.80 ± 13.66a | 1,001.67 ± 38.97b | 943.67 ± 33.07b | 940.67 ± 39.87b | 922.67 ± 27.65b | 782.00 ± 13.32a | F = 12.72 | 0.0001  |
| Sodium absorption ratio          | 2.91 ± 0.04a | 3.66 ± 0.0013b | 3.51 ± 0.0013b | 3.54 ± 0.0012b | 3.40 ± 0.0013b | 2.90 ± 0.0008a | F = 11.31 | 0.0002  |
| Zinc (mg/kg)                     | 4.16 ± 0.12a | 14.83 ± 0.41b | 12.53 ± 0.92b | 5.10 ± 0.005a | 5.53 ± 0.046a | 4.10 ± 0.010a | H = 15.27 | 0.0093  |

Anaerobically digested algae (AD-algae), anaerobically digested WAS (AD-WAS), cation exchange capacity (CEC).

Values in the same row represented by a different superscript symbol (a, b) represent significantly different treatment means (one-way ANOVA/Kruskal-Wallis, p < 0.05).
Heavy metals can accumulate in the leaf tissue of plants and render them unsafe for consumption when WAS is used as a fertiliser. Of the heavy metals tested, iron was the only metal present at a higher concentration in Swiss chard leaves from WAS- and algae-fertilised treatments when compared with the inorganic-fertiliser treatment. A single application of WAS to the soil, at 112 t/ha dry weight, increased the copper, zinc, iron and chromium concentrations of snap beans and flax plants (Dowdy et al. 1978; Tsakou et al. 2002). Land application of sludge has been shown to increase the foliar cadmium, chromium, copper, iron, manganese and zinc concentrations of agricultural crops (Hernández et al. 1991; Warman & Termeer 2005; Singh & Agrawal 2008). The acidity of the soil influences the accumulation of heavy metals in crop tissue due to their increase in solubility as pH decreases; therefore it is advisable only to apply waste sludge to soils with a pH above 7.0 (Hernández et al. 1991; DWAF 1997). The accumulation of heavy metals in plant tissue varies between plant species, with Swiss chard assimilating more soil iron into its leaves than cabbage, kale, potato, red beet and cauliflower (Itanna 1998; Itanna 2002). The land application of algae and WAS from a brewery effluent treatment process increased the foliar iron concentration, however the concentration was still well within the limits for human consumption. The foliar heavy metal concentration of crops grown in sludge-fertilised soils needs to be continuously monitored if practiced commercially as there is a possibility of heavy metal contamination, especially in acidic soils.

Biosolids have the ability to improve the soil’s physical characteristics and are thus commonly used for soil reclamation. There was no difference in the physical properties of the soil receiving algae, WAS or inorganic fertiliser. The literature reports an improvement in the soil’s physical properties when sludge is applied (Tsadilas et al. 1995; Singh & Agrawal 2008; Diacono & Montemurro 2010). The organic matter added to the soil from WAS improved the bulk density, porosity and water holding capacity of the soil, after one year (Ojeda et al. 2003; Diacono & Montemurro 2010). Organic matter concentration of the soil increases when sludge is applied to soil and this aids in stabilising soil structure by increasing inter-particle cohesion within aggregates and increasing their hydrophobicity, thus increasing soil structural stability (Diacono & Montemurro 2010). The improvement in soil structural stability is normally only noticed one to two years after the application of sludge (Sodhi et al. 2009; Tejada et al. 2009). During this study no differences were noticed in the soil’s physical characteristics and structure due to the short period of the trial (13 weeks). However, after prolonged application (>two years) an increase in the soil’s physical structure would be expected (Ojeda et al. 2003; Tejada et al. 2009).

When applying sludge to soils the possibility of contaminating them with heavy metals exists. Of the heavy metals analysed, zinc was the only element that was higher in algae-fertilised soils when compared with soils fertilised with inorganic fertiliser. Various authors have reported an increase in soil iron, copper, zinc, manganese nickel, chromium, cadmium and lead concentrations when WAS is applied to soil (Hernández et al. 1991; Singh & Agrawal 2008; Lu et al. 2012). Zerzghi et al. (2010) applied anaerobically digested WAS to desert soils for 20 years and reported no detrimental effect on the soil’s heavy metal content or soil fertility. Soils containing a high clay content decrease the mobility of heavy metal into plant tissue, however once all absorption sites are saturated this benefit is redundant (Wuana & Okieimen 2011). It is important to apply sludge from WWTPs to soil below the permitted application rate of 8.0 t/ha/y and to ensure that its heavy metal content is always within the limits for land application (DWAF 1997; EPA 1999). If this is carried out, in conjunction with regular soil monitoring procedures, the land

Table 5 | The mean (±standard error) carbon and nitrogen concentrations of soils fertilised with the algae and WAS treatments

| Parameter   | Algae          | AD-algae      | WAS           | AD-WAS        | F<sub>lev</sub> value | P value |
|-------------|----------------|---------------|---------------|---------------|-----------------------|---------|
| Carbon (g/kg) | 58.70 ± 1.04<sup>a</sup> | 54.82 ± 1.05<sup>b</sup> | 57.03 ± 0.81<sup>a</sup> | 55.29 ± 0.76<sup>b</sup> | 9.221                | 0.0161  |
| Nitrogen (g/kg) | 3.64 ± 0.04<sup>a</sup>  | 3.47 ± 0.06<sup>b</sup>  | 3.66 ± 0.04<sup>a</sup>  | 3.53 ± 0.06<sup>b</sup>  | 7.569                | 0.0250  |

Anaerobically digested algae (AD-algae), anaerobically digested WAS (AD-WAS).

Superscripts (a, b) in the same row represent significantly different treatment means (multifactor ANOVA, p < 0.05).
application of sludge from WWTPs should not have any detrimental effect on the soil’s chemical fertility (Zerzghi et al. 2010).

Sodium contamination of agricultural soils is the leading cause of rendering soils unsuitable for agriculture (Qadir et al. 2003; Muyen et al. 2011). In this study, the sodium concentration and SAR of soils fertilised with algae or WAS were significantly higher than those of the soils fertilised with inorganic fertiliser. Karami et al. (2012) applied various organic wastes (animal manure, waste sludge and plant derived organic wastes) to soils and, after six months, they recorded a decrease in the soil’s SAR. This was due to an increase in soil magnesium and calcium concentration and not a decrease in soil sodium concentration (Karami et al. 2012). The land application of animal manures to calcareous soils with a sodium concentration greater than 0.9 g/l resulted in an increase in soil sodium concentrations (Bernal et al. 1995). The application of sewage sludge to potting soil that was planted with olive trees resulted in an increase in soil sodium concentrations, SAR and decreased plant growth due to osmotic and sodium stress (Gascó & Lobo 2006). The application of brewery-effluent-grown algae and WAS to land increased the soil’s SAR, which could be accompanied by a deterioration in the soil’s structural stability and crop yield.

In South Africa there are no sodium guidelines or limits for the disposal of waste sludge on agricultural land. In this study, the soil’s sodium concentration increased when WAS or algae was applied to it. Future research should monitor the influence of WAS on the soil’s SAR and sodium concentration, as prolonged application could lead to soil salinisation, rendering it unsuitable for agriculture (Gascó & Lobo 2006; Muyen et al. 2011). Thus, regulations on the application rates of sludge to agricultural soils should also consider limit values for sodium (not only heavy metals) to avoid the accumulation of sodium in the soil and consequent deterioration in the soil’s fertility.

The use of inorganic fertilisers in agriculture results in the leaching of nitrate and ammonia, which can lead to eutrophication of surrounding water bodies (Withers et al. 2014). After only 13 weeks, the nitrogen concentration of soils fertilised with algae or WAS was significantly higher than that of soil fertilised with inorganic fertiliser. At the beginning of the trial all fertilisers were applied at the same application rate. Nitrogen is removed from the soil by plant uptake, by leaching into surrounding water bodies or by transformation into a gaseous form (N2, NH3, NO, NO2, N2O; Withers & Lord 2002; Withers et al. 2014). More nitrogen was assimilated into plant biomass in the algae- and WAS-fertilised treatments as plants grown in these treatments had a greater biomass production and similar leaf nitrogen content when compared with inorganic fertilised plants. Therefore, a greater portion of nitrogen must have left the soil via leaching or outgassing to the atmosphere in inorganic-fertiliser treatments when compared with WAS- or algae-fertilised treatments.

The amount of nitrogen leached from soils is dependent on the form of nitrogen supplied, the crop utilisation rate and the irrigation regime, while the mass of nitrogen lost to the atmosphere is influenced by soil temperature, oxygen concentration, moisture and the amount of available organic carbon and nitrogen (Bremner 1997; Signor & Cerri 2015). Nitrogen leaching is predominant in carbon-deficient soils containing more inorganic nitrogen than crops can utilise (Bremner 1997; Signor & Cerri 2015). The nitrogen in the inorganic-fertiliser treatment was present in the form of ammonia and nitrate, whereas in the WAS and algae treatments it was present as ammonia and other organic compounds (Hygrotech Pty Ltd, South Africa). Since ammonia and nitrate are highly soluble in water (Hollister et al. 2013), more nitrogen was probably lost to leaching from the inorganic-fertiliser treatment when compared with the algae- and WAS-fertilised treatments where 50–60% of the nitrogen applied to the soil was in an insoluble particulate form. Walsh et al. (2012) reported greater nitrate and ammonia leaching from a sandy clay loam fertilised with ammonium nitrate when compared with anaerobically digested sludge. However, Schröder et al. (2010) concluded that the extent of nitrogen leaching from grassland soils was not influenced by the nitrogen source (organic vs inorganic), the main factor being balancing nitrogen supply with crop demand. More nitrogen was retained in the WAS- or algae-fertilised soils than in the inorganic-fertiliser soils because all nitrogen supplied by the inorganic fertiliser was in a soluble form (ammonia and nitrate) while 50–99% of the nitrogen supplied by the WAS and algae was in an insoluble form.

Sludge-derived organic fertiliser can be advantageous over inorganic fertilisers as a portion of its nitrogen is in an insoluble form and only becomes available to plants after...
microbiological decay in the soil, thus resulting in less nitrogen being leached into surrounding water bodies (Walsh et al. 2012; Withers et al. 2014). Future studies should determine the effect wastewater derived sludge has on the metabolic community structure and activity of a soil and collect and analyse the leachate from the soil to determine the amount of nitrogen lost via leaching and to the atmosphere.

During AD, insoluble particulate bound nitrogen is converted to ammonia, which can be utilised by plants (Tchobanoglous et al. 2003). There was no difference in the crop yield and chemical leaf concentration of Swiss chard plants grown in soils fertilised with anaerobically digested or non-anaerobically digested algal or WAS biomass. Heat-dried WAS that was previously subject to AD had a higher concentration of plant available inorganic phosphorus than non-anaerobically digested heat-dried WAS (Lyberatos et al. 2004). The yield of Zea mays from sludge fertiliser treatments was higher in soils fertilised with AD-WAS when compared with soils fertilised with untreated WAS (Warman & Termeer 2005). The application of sewage sludge increased the soil’s microbial activity, respiration and enzyme activities, indicating the soil’s microbes are able to degrade the sludge into constituents that are available for plant uptake (Maiti et al. 1992). The microbial community present in the soil was able to degrade insoluble particulate bound nutrients into soluble plant-available nutrients, resulting in no difference in crop yield between soils fertilised with anaerobically digested or non-anaerobically digested biomass (Antolín et al. 2005).

Anaerobic digestion is able to recover a portion of the carbon in waste biomass into a fuel source (methane) before it is applied to the soil. The carbon concentration of soils fertilised with AD sludge or algae was significantly lower than soils fertilised with undigested sludge or algae. During AD the carbon content of organic matter is converted into methane and carbon dioxide, thus decreasing its carbon concentration (Tchobanoglous et al. 2005). Anaerobic digestion is a recommended pre-treatment step prior to the land disposal of waste sludge as the process can decrease its odour and pathogen count, as well as the greenhouse gas emissions from further decomposition in the soil (EPA 1999; Warman & Termeer 2005; Singh & Agrawal 2008). The cost of construction, operation and maintenance must be weighed up against the benefit of biogas recovery for an anaerobic pre-treatment step to be viable. The AD stabilisation step is beneficial as it allows the recovery of energy from waste solids, does not have any negative effects on plant growth or soil fertility and decreases the amount of carbon applied to the soil.

CONCLUSION

These waste products can be utilised as an inorganic fertiliser replacement when applied to the soil at the same nitrogen loading rate and can even increase crop yield. The nitrogen applied to the soil from algal and WAS biomass appeared to leach out of the soil less than the nitrogen (ammonia and nitrate) supplied by inorganic fertilisers. These biomasses offer a good alternative to inorganic fertilisers as the slow nutrient release will aid in reducing the nutrient contamination conventional inorganic fertiliser-based agriculture has on surrounding water bodies. Anaerobic digestion is a viable pre-treatment step for sludge and algal biomasses as it recovers a portion of the carbon as an energy source and does not decrease their quality as fertilisers. The application of algae generated from a brewery effluent treatment system increased the soil sodium concentration, so it is recommended that regulations on the application rate of sludge to agricultural land should consider the limit values for sodium to avoid its accumulation in the soil and deterioration in soil fertility.

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DATA AVAILABILITY STATEMENT

All relevant data are available from an online repository or repositories: https://drive.google.com/drive/u/0/folders/11cQMEncGsvljfwW-PftTR6uER17kGCq.
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