Research article

The effects of feedstock pre-treatment and pyrolysis temperature on the production of biochar from the green seaweed *Ulva*

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ABSTRACT

Green seaweeds from the genus *Ulva* are a promising feedstock for the production of biochar for carbon (C) sequestration and soil amelioration. *Ulva* can be cultivated in waste water from land-based aquaculture and *Ulva* blooms ("green tides") strand millions of tons of biomass on coastal areas of Europe and China each year. The conversion of *Ulva* into biochar could recycle C and nutrients from eutrophic water into agricultural production. We produce biochar from *Ulva ohnoi*, cultivated in waste water from an aquaculture facility, and characterize its suitability for C sequestration and soil amelioration through biochemical analyses and plant growth experiments. Two biomass pre-treatments (fresh water rinsing to reduce salt, and pelletisation to increase density) were crossed with four pyrolysis temperatures (300 —750 °C). Biomass rinsing decreased the ash and increased the C content of the resulting biochar. However, biochar produced from un-rinsed biomass had a higher proportion of fixed C and a higher yield. C sequestration decreased with increasing pyrolysis temperatures due to the combination of lower yield and lower total C content of biochar produced at high temperatures. Biochar produced from un-rinsed biomass at 300 °C had the greatest gravimetric C sequestration (110—120 g stable C kg^{-1} seaweed). Biochar produced from un-pelletised *Ulva* enhanced plant growth three-fold in low fertility soils when the temperature of pyrolysis was less than 450 °C. The reduced effectiveness of the high-temperature biochars (>450 °C) was due to a lower N and higher salt content. Soil ameliorated with biochar produced from pelletised biomass had suppressed plant germination and growth. The most effective biochar for C sequestration and soil amelioration was produced from un-rinsed and un-pelletised *Ulva* at 300 °C. The green tide that occurs annually along the Shandong coastline in China generates sufficient biomass (200,000 tons dry weight) to ameliorate 12,500 ha of soil, sequester 15,000 t C and recycle 5500 t N into agriculture. We provide clear parameters for biochar production to enable the beneficial use of this biomass.

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1. Introduction

Biochar is a charcoal-like material that is produced through the combustion of biomass in a low oxygen atmosphere — a technique known as slow pyrolysis (Lehmann and Joseph, 2009). Biochar has a high C content that can be resistant to degradation, and so is considered to be "stable" for centuries to millennia, thus providing a form of C sequestration (Lehmann and Joseph, 2009). In addition, biochar can improve the fertility of soils by altering soil properties such as water holding capacity and through the retention of

nutrients on the surface of the biochar particles (Lehmann and Joseph, 2009). Consequently, the amelioration of soils with biochar can greatly improve the efficiency of fertilizer use, the yield of crops, and the re-vegetation of soils for landscape rehabilitation, while also increasing the C content of soils (reviewed in Beesley et al., 2011; Jeffery et al., 2011; Spokas, 2010).

While biochar has recognized roles in C sequestration and the amelioration of soil, both the stability of biochar C and the agronomic utility of biochar are strongly influenced by the properties of the feedstock and the conditions under which the biochar is produced (Bird et al., 2015). It is possible that not all of the benefits of biochar application can be maximized simultaneously (Jeffery et al., 2013). For example, if the feedstock has a high ash content this can alter the rate of decomposition of the biomass during pyrolysis and

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the chemical pathways by which biochar is formed, with concomitant effects on C sequestration (White et al., 2011). Additionally, high pyrolysis temperatures are typically predicted to yield biochar with a higher fixed C content (McBeath et al., 2015) but this can be at the expense of its agronomic benefits (Zhao et al., 2013). The production of biochar must therefore be conducted with the intended application in mind and it is important to quantify tradeoffs between C sequestration and agronomic utility for a range of feedstock and production conditions (Abiven et al., 2014; Jeffery et al., 2013).

Seaweeds are a novel feedstock for biochar production (Bird et al., 2011; Roberts et al., 2015a) that can be sourced from ocean farms (Roberts et al., 2015b), land-based cultivation (De Paula Silva et al., 2008), or from seaweed blooms that occur in eutrophic waters (Merceron et al., 2007). Biochar produced from seaweed has a lower C content (~30–35%) than those produced from ligno-cellulosic ("woody") biomass (typically >70%), but high concentrations of exchangeable trace elements and macro-nutrients (N, P, Ca, Mg, K, Zn and Mo) that are essential for plant production (Roberts et al., 2015b). Consequently, seaweed biochar can outperform ligno-cellulosic biochars, particularly in low fertility soil (Bird et al., 2012). Green seaweeds from the genus *Ulva* are particularly promising as they can be cultivated in waste water from land-based aquaculture and so sequester and recycle nutrients for terrestrial crop production (Lawton et al., 2013). *Ulva* is also a bloom forming ("green tide") seaweed that is an issue in eutrophic water bodies, most famously along the Shandong coast in China (Wang et al., 2015), and in Brittany, France (Merceron et al., 2007). The annual *Ulva* bloom along the Shandong coastline alone can result in millions of tons of *Ulva* being beached (Liu et al., 2013). The annual production of cultivated seaweeds is in the vicinity of 20 million tons (FAO, 2012), so this green tide represents a significant resource. The conversion of green tide biomass into biochar would serve the dual purposes of reducing the volume of material to be removed from coastal areas and would valorize a nuisance biomass.

While seaweed biochar improves the growth of crops in low fertility soils (Bird et al., 2012) it does have limitations. Seaweed biochar has a high exchangeable Na content which may have negative effects on soil microbes in anything other than sandy soils (Bird et al., 2012; Domene et al., 2015; Rajkovich et al., 2012). The availability of Na can be controlled by manipulating the conditions under which the biochar is produced. Na can volatilize from biomass at high temperatures (Lane et al., 2015), or the Na content of seaweed can be reduced through a fresh water rinse of the biomass before pyrolysis (Neveux et al., 2014). By removing salts, fresh water rinsing can also increase the C, N and P content of the feedstock, potentially making it a more suitable material for C sequestration and soil amelioration. Finally, the fact that seaweed is a brittle feedstock may result in losses of surface-supplied biochar in agronomic settings, and is a barrier to the use of biochar in plant nurseries where bulky materials are required for soil aeration (Dumroese et al., 2011). Pelletisation of biomass prior to pyrolysis can improve the pyrolysis of low density and brittle feedstock and reduce the loss of biochar when applied to soil (Kim et al., 2014). Biochar produced from pelletised biomass may also be more recalcitrant and sequester C more effectively (Manyà, 2012; Sigua et al., 2014).

In this study we examine how biomass pre-treatment (fresh water rinsing and pelletisation) interact with pyrolysis temperatures to influence the properties and utility of biochar produced from seaweed. We consider how these factors alter the composition of seaweed biochar, and its ability to sequester C and improve the production of plants in soils. We do so through a combination of biochar characterisation and plant growth experiments in a low

fertility soil ameliorated with biochar.

2. Methods

2.1. Biomass cultivation and biochar production

The green seaweed *Ulva ohnoi* was grown in 50 m High Rate Algal Ponds (HRAPs) at the MBD Energy seaweed cultivation facility within the Pacific Reef Fisheries Ltd. prawn farm in northern Queensland, Australia (19°28′46″ S, 147°29′18″ E). The HRAPs are operated with discharge water from the prawn farm via two sand filters that convert dissolved organic nitrogen (DON) into the dissolved inorganic N (DIN) required for the seaweed. The water and biomass are circulated in the HRAPs with a paddle wheel and the biomass is harvested by draining the water through coarse mesh baskets. The *U. ohnoi* was cultivated between February–April, 2015.

Four feedstocks for biochar production were prepared from *U. ohnoi* – un-manipulated seaweed, fresh water rinsed seaweed, pelletized seaweed and pelletized fresh water rinsed seaweed. To make the fresh water rinsed seaweed, a subset of the drained *U. ohnoi* was washed in fresh water following the methods of Neveux et al. (2014). Briefly, *U. ohnoi* was loosely packed into a 60 L plastic tub (~100 g biomass L^{-1}). The tub was filled with fresh water and stirred for 60 s, then removed and drained and the process repeated two more times with new water. This technique reduces the ash content of the seaweed by approximately 50% (Neveux et al., 2014). The rinsed and un-rinsed biomass were spread on plastic trays and dried overnight in a solar kiln at 60 °C. Half of the rinsed and un-rinsed dried biomass was then converted into pellets using a Nova N-Micro Pellet Mill. The pellet mill pulverizes dried *U. ohnoi* with a hammer mill then sieves the biomass (<3 mm). The sieved biomass is mixed with a small amount of water to attain 10% moisture content. This biomass is then extruded through a rotating pellet die to produce pellets with a diameter of 6 mm and an average length of 25 mm. The pelletizing process increased the bulk density of the dried seaweed from 0.033 ± 0.001 g cm^{-3} to 0.707 ± 0.004 g cm^{-3}. The remaining biomass was left in its natural "flake" morphology.

The four feedstocks were converted into biochar at one of four temperatures (300, 450, 600 and 750 °C). The flakes or pellets were placed in a sealed stainless steel retort inside a muffle furnace that was pre-heated to the target temperature. The retort was purged with N_2 gas at 4 L min^{-1} during pyrolysis. The biomass was left at the target temperature for 60 min and the retort was removed from the furnace and cooled under continued N_2 flow. The biochar was weighed to calculate yield (% of original feedstock). Three independent samples were produced for each combination of biomass pre-treatment and pyrolysis temperature. A constant mass of feedstock (80 g) was included in the biomass and pellet treatments to ensure the biochar yield from a constant feedstock mass could be compared across the treatments.

2.2. Biochar characterisation

The yield of biochar (%) was recorded for each sample. The elemental profile (C, H, O, N, and S) of the biomass and biochar were analysed with an elemental analyser (OEA Laboratory Ltd, United Kingdom). The stability of C in each biochar sample was determined through the Edinburgh method (Crombie et al., 2013; Cross and Sohi, 2013). Briefly, once the C content of each sample had been determined, a sufficient quantity of each sample was weighed into a 15 ml sample tube to give a biochar sample containing 0.1 g C. Each sample was then mixed with 7 ml of a 5% H_2O_2 solution and capped with aluminium foil until bubbling stopped (2–3 min). The foil covering was removed and the sample tubes were placed in a

shaker cabinet (50 rpm) at 80 °C and left for 48 h, or until dry. The samples were reweighed to calculate the mass of biochar lost during oxidation, and the residue was analysed for final C content as described above. The stability of C in each sample is expressed as the percentage of the original C remaining after oxidation, and is determined based on the mass loss during oxidation and the initial and final C contents of the biochar samples according to Equation (1):

$$C_{Stable} = \left(\frac{M_f \times C_f}{M_i \times C_i} \right) \times 100 \qquad (1)$$

where: C_{Stable} is the stable C content (%) that is resistant to oxidation; M_i is the initial mass (g) of the biochar sample; M_f is the final mass (g) of biochar after oxidation; C_i is the initial C content (%) of the sample; and C_f is the final C content (%) of the biochar after oxidation. In addition to this measure of relative C stability, the C sequestration potential of each sample was also calculated based on the amount of C in the original seaweed feedstock that was converted into a stable form during slow pyrolysis, according to Equation (2):

$$C_{SeqPot} = Y \times \left(\frac{C}{100} \right) \times \left(\frac{C_{Stable}}{100} \right) \qquad (2)$$

where: C_{SeqPot} is the C sequestration potential of each biochar (g stable C kg^{-1} *Ulva* feedstock); Y is the yield of biochar (g biochar kg^{-1} feedstock); C is the C content of the biochar (%); and C_{Stable} is the stable C content (%), as determined using Equation (1).

2.3. The effects of biochar on the germination and growth of Raphanus sativus

A laboratory experiment was conducted to test the effects of the biochars on the germination and growth of the radish *R. sativus*. Each biochar was mixed into a sandy loam (Bedrock Landscaping, Townsville Australia) at 8.5 g kg^{-1}. This stocking density is sufficient to achieve the greatest benefits of algal biochar on radish growth (Roberts et al., 2015a). The biochar-soil mixtures were placed into seedling trays and five radish seeds were planted in each cell of the tray. The soil-biochar mixtures were wet to water-holding capacity (50 ml deionized water per replicate) and excess water was left to drain through the soil and was collected in a vial beneath each cell. The electrical conductivity (EC) of all leachates were measured and the trays were placed into a light and temperature-controlled culture cabinet with a 12 h light: 12 h dark photoperiod (30 μmol photons m^{-2} s^{-1}, 25 °C). Each cell was re-wet with 20 ml of deionized water five times weekly and the EC of the leachates were measured to monitor the leaching of salts from each treatment. After 7 d the germination of radishes was recorded and the experiment was sampled to measure the length of the hypocotyl of each plant.

The results of the laboratory experiment demonstrated that the flake biochar produced at 300–600 °C was the most suitable for soil amelioration. The biochar produced from pelletised biomass and all biochars produced at 750 °C (regardless of pre-treatment) either impaired germination or reduced the production of plant biomass (see results section "*3.3. The effects of biochar on the germination and growth of Raphanus sativus*" for a detailed summary of the results). Consequently, an outdoor experiment was conducted to demonstrate the effects of *Ulva* biochar produced at 300–600 °C from flake biomass on longer-term radish growth. The rinsed and un-rinsed *Ulva* flake biochar samples produced at 300–600 °C were mixed into the sandy loam at 8.5 g kg^{-1} and added to 100-mm diameter pots. Five radish seeds were planted in each pot and the

soil was wet to water-holding capacity (200 ml per pot). The pots were left for 30 d in a greenhouse and watered every second day to prevent desiccation. Germination stabilized after 10 d, so all pots were thinned back to one plant per pot. The radishes were harvested after 30 d, dried at 60 °C and the dry weights of the above and below ground biomass was recorded. No fertilizer was added to any of the treatments. A control treatment receiving no biochar was also included with 4 replicate pots for each treatment.

2.4. Data analysis

The yield and C sequestration potential of the biochars, and the growth and germination of radishes in the laboratory experiment, were contrasted with a three factor Analysis of Variance (ANOVA), including the factors "rinsing" (rinsed vs. un-rinsed), "biochar" (flake vs. pellet) and "temperature". As only flake biochar samples were included in the pot trial, the production of radish biomass was contrasted using a two-way ANOVA including the factors "rinsing" and "temperature". The assumptions of normality and heterogeneity of variance were tested with residual histograms and scatterplots of residuals vs. estimates, respectively (Quinn and Keough, 2002).

3. Results

3.1. Biochar production and characterisation

The rinsing of the *Ulva* biomass reduced the ash content of the biomass from 28.6% to 14.7%. Both the pyrolysis temperature and the biomass rinsing pre-treatment had a strong effect on the yield of biochar from *U. ohnoi*. The yield of biochar decreased with increasing temperature and this was true for rinsed and un-rinsed biomass when pyrolysed in the form of both flakes and as pellets (Fig. 1). Overall, the un-rinsed flake *U. ohnoi* had a mean yield of 62.4% at a HHT of 300 °C, decreasing to 34.2% at a HHT of 750 °C (Fig. 1a). The un-rinsed pelletised biomass had a maximum yield of 62.9% at 300 °C, decreasing to 35.1% at 750 °C (Fig. 1b). The yield of biochar produced from rinsed *U. ohnoi* was 5–10% lower than the equivalent biochar produced from un-rinsed biomass across the thermo-sequence (Fig. 1). There was also a slight, but statistically significant, effect of pelletisation on biochar yield. Biochar produced from *Ulva* pellets had a 1–5% higher yield than biochar produced from *Ulva* flakes across the thermo-sequence (Fig. 1). The only exception was biochar produced from rinsed biomass at 300 °C, which had a slightly higher yield as flake (Fig. 1).

The EC of the leachates was higher from un-rinsed biochar than rinsed biochar, and this was true for both the flake and pellet treatments (Fig. 2). In addition, the EC of the leachates from un-rinsed *Ulva* flake biochars tended to be higher than from un-rinsed pellet biochars (Fig. 2). With respect to pyrolysis temperature, the EC of leachates from the un-rinsed biochars increased with pyrolysis temperature from 300 to 450 °C, and stabilized thereafter (Fig. 2). In contrast, the EC of leachates from rinsed biochar produced from *Ulva* flake and pellets decreased with increasing pyrolysis temperatures (Fig. 2).

Biomass rinsing and pyrolysis temperature had a strong effect on the elemental composition of the biochar (C, H, O, N and S). Biochar produced from rinsed biomass had a higher C content across the pyrolysis thermo-sequence than the biochars produced from un-rinsed biomass (Table 1). The biochar produced from rinsed biomass had C contents from 41.0 to 43.5% for flake and 29.6–39.6% for pellets (Table 1). In contrast, the biochar produced from un-rinsed biomass had C contents from 24.9 to 30.9 for flake and 23.4–29.2% for pellets (Table 1). In all cases, the C content of the biochar samples decreased slightly with increasing pyrolysis
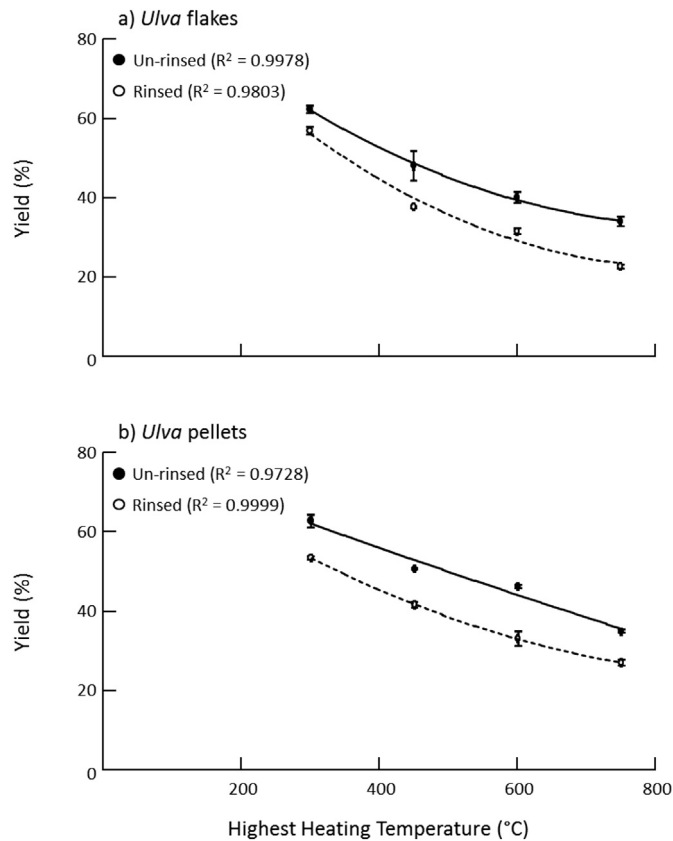
Fig. 1. The effect of biomass rinsing and highest heating temperature on the yield of biochar produced from *Ulva* (a) flakes and (b) pellets. Solid and dashed lines show data for un-rinsed and rinsed biomass, respectively. All data are means (%) ± S.E. (n = 3). There were significant effects of pelletisation ("Pelletisation": $F_{1, 32} = 9.76$, P = 0.004; flake > pellet), rinsing ("Rinsing": $F_{1, 32} = 200.09$, P < 0.001) and temperature ("Temperature": $F_{3, 32} = 347.85$, P < 0.001) on biochar yield.

Fig. 2. Electrical conductivity (mS cm^{-1}) of pore water leachates from soils containing biochar produced from *Ulva* biomass as a) flakes and b) pellets. Solid lines show data for un-rinsed biomass and dashed lines show data for rinsed biomass. All data are means (mS cm^{-1}) ± standard error (n = 4).

temperature (Table 1). The biochars produced from rinsed biomass had a higher N content, ranging from 3.0 to 6.7% in the rinsed biochar and 2.4–4.7% in the un-rinsed biochar (Table 1). The N content of all biochars decreased as pyrolysis temperatures increased, with the highest N content found in the rinsed flake biochar produced at 300 °C (Table 1). The H and O content followed a similar pattern to C and N, being higher in biochar produced from rinsed biomass, and decreasing with increasing pyrolysis temperature (Table 1).

3.2. Biochar C sequestration potential

The proportion of stable C in the biochar samples increased with pyrolysis temperature ("Temperature": $F_{3, 32} = 64.29$, P < 0.001) and was slightly higher in the biochar produced from unrinsed biomass ("Rinsed": $F_{1, 32} = 12.613$, P = 0.001). All of the biochars had a mean C stability in excess of 95% when produced at 750 °C (Fig. 3a). In contrast, all biochar samples produced at 300 °C had a mean C stability of 50–60%, regardless of biomass pre-treatment (Fig. 3b). The stable C content was calculated as the product of the C content (g kg^{-1}) and the proportion of stable C to give a measure of stable C (g kg^{-1}) in each sample. The biochar produced from rinsed biomass had a slightly higher stable C content than the biochars produced from un-rinsed biomass at all temperatures (Fig. 3c, d; "Rinsed": $F_{1, 32} = 65.58$, P < 0.001). There was also a significant interaction between the pelletisation of biomass and temperature on the stable C content of the resulting biochars
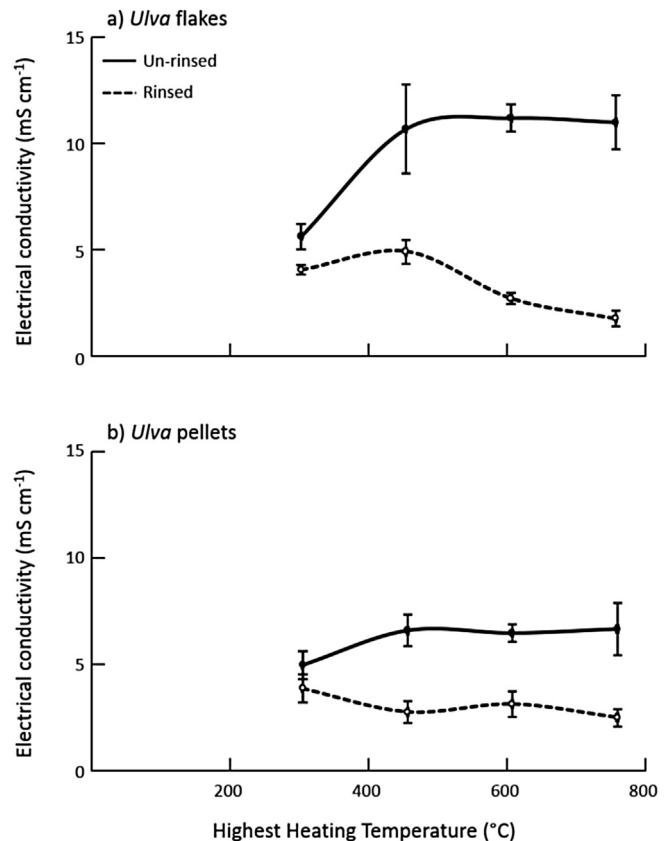
("Biochar × Temperature": $F_{3, 32} = 5.52$, P = 0.004). The stable C content of biochar produced from *Ulva* flake increased with the pyrolysis temperature, from 174 to 209 g kg^{-1} in the un-rinsed and rinsed flake biochars produced at 300 °C, to 246 and 389 g kg^{-1} in the biochars produced at 750 °C (Fig. 3c). In contrast, the pyrolysis temperature had no effect on the stable C content of the biochar produced from pelletised feedstock (Fig. 3d).

Finally, a direct comparison of the C sequestration potential as a function of feedstock mass was determined by including the yield of biochar under each pyrolysis condition. The yield of biochar (g kg^{-1} of feedstock) was multiplied by the stable C content of each sample (Fig. 3e, f). This measure of C sequestration showed that there was no effect of biomass rinsing, nor pelletisation, on the ability of the biochars to sequester C ("Rinsed": $F_{1, 32} = 0.35$, P = 0.557; "Biochar": $F_{1, 32} = 0.12$, P = 0.735). There was, however, a decrease in C sequestration potential with increasing pyrolysis temperature ("Temperature": $F_{3, 32} = 12.97$, P < 0.001). The conversion of 1 kg of rinsed and un-rinsed *Ulva* flake at 300 °C yielded 109–120 g of stable C in biochar form, decreasing to 84–89 g of stable C at 750 °C (Fig. 3e). The conversion of 1 kg of rinsed and un-rinsed *Ulva* pellets at 300 °C yielded 117–131 g of stable C in biochar form, decreasing to 76–80 g at 750 °C (Fig. 3f).

3.3. The effects of biochar on the germination and growth of Raphanus sativus

There was a significant interaction between the biochar treatment (flake vs pelletised) and the pyrolysis temperature on the germination of radish seeds in the laboratory experiment

Table 1
The elemental profile (C, H, O, N, and S) of biochar produced from *Ulva* biomass at a range of pyrolysis temperatures. All data are mean values ± standard error (n = 3). A Permutational Analysis of Variance (PERMANOVA) found a significant effect of biomass rinsing ("Rinsing": Pseudo-$F_{1,\ 32}$ = 48.09, P < 0.001) and pyrolysis temperature ("Temperature": Pseudo-F3, 32 = 48.90, P < 0.001) on the elemental composition of the biochars. There was no effect of pelletisation ("Pellet": Pseudo-$F_{1,\ 32}$ = 2.93, P = 0.08), nor any significant interactions between the factors.

| Rinsing	Biochar	Temperature (°C)	C (%)	H (%)	O (%)	N (%)	S (%)
Unrinsed	Flake	300	30.9 ± 1.1	3.4 ± 0.1	23.7 ± 0.7	4.7 ± 0.2	5.7 ± 0.8
		450	25.2 ± 1.4	2.1 ± 0.3	21.8 ± 1.6	3.3 ± 0.2	7.2 ± 0.2
		600	26.4 ± 0.3	1.1 ± 0.2	16.3 ± 1.8	2.9 ± 0.1	6.6 ± 0.3
		750	24.9 ± 0.4	0.8 ± 0.1	12.7 ± 0.6	2.4 ± 0.1	4.1 ± 0.2
	Pellet	300	29.2 ± 1.9	3.0 ± 0.2	23.3 ± 0.5	4.5 ± 0.4	5.7 ± 0.3
		450	25.8 ± 0.3	1.6 ± 0.1	19.6 ± 0.1	3.5 ± 0.1	6.6 ± 0.3
		600	23.8 ± 1.5	0.8 ± 0.1	16.4 ± 0.2	2.6 ± 0.2	6.0 ± 0.2
		750	23.4 ± 0.4	0.7 ± 0.1	11.3 ± 0.6	2.4 ± 0.1	3.9 ± 0.5
Rinsed	Flake	300	43.5 ± 0.2	4.5 ± 0.1	26.7 ± 0.8	6.7 ± 0.2	4.9 ± 0.5
		450	41.7 ± 0.4	2.7 ± 0.2	24.9 ± 0.4	5.5 ± 0.4	7.2 ± 0.4
		600	42.2 ± 0.3	1.6 ± 0.1	21.3 ± 0.4	4.9 ± 0.1	7.6 ± 0.7
		750	41.0 ± 2.9	1.1 ± 0.2	15.9 ± 1.6	4.1 ± 0.3	4.2 ± 1.0
	Pellet	300	39.6 ± 0.4	4.0 ± 0.1	26.0 ± 0.3	6.1 ± 0.1	6.0 ± 0.4
		450	35.8 ± 0.2	2.4 ± 0.2	25.4 ± 0.4	4.7 ± 0.1	6.5 ± 0.8
		600	34.0 ± 3.8	1.0 ± 0.1	17.5 ± 2.5	3.7 ± 0.5	6.0 ± 1.1
		750	29.6 ± 4.1	0.8 ± 0.1	13.3 ± 0.5	3.0 ± 0.4	3.7 ± 0.5
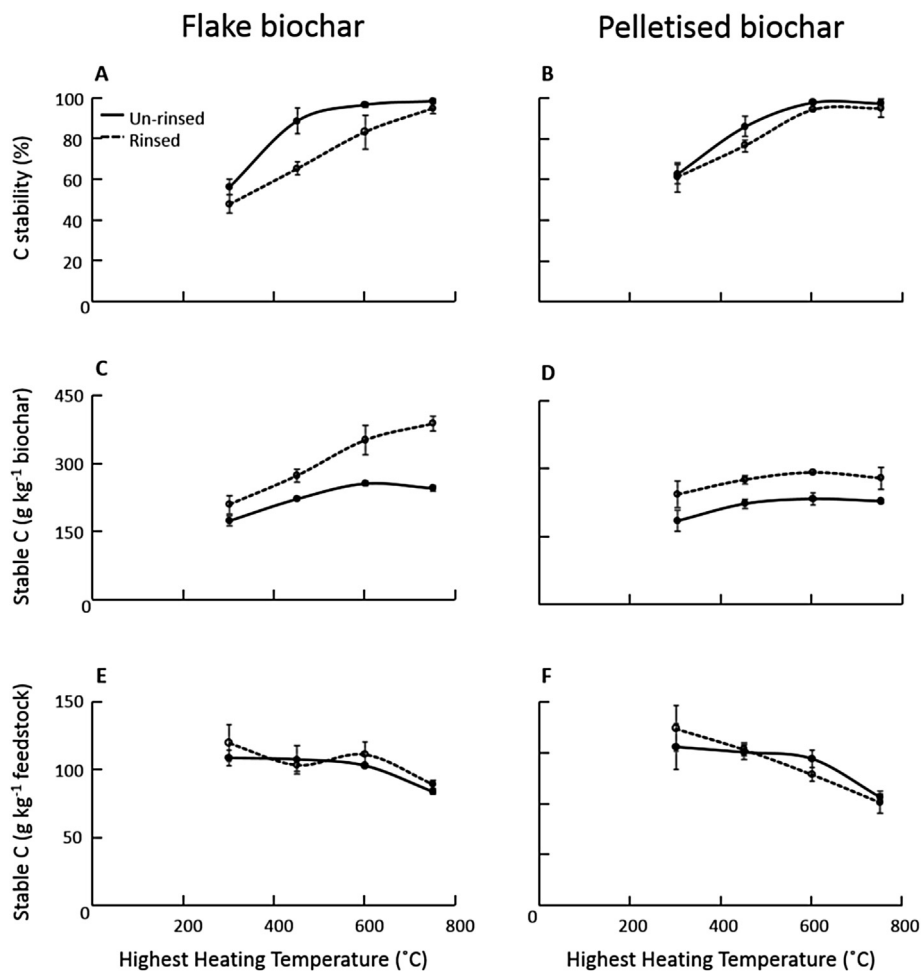
Fig. 3. The effect of biomass pre-treatment (rinsing and pelletisation) and pyrolysis temperature on the C sequestration properties of biochar produced from *Ulva ohnoi*. Panels A–B show the C stability (%) of each biochar as determined by the Edinburgh assay. Panels C–D show the stable C content of each biochar (g kg^{-1} biochar), which is the product of the C content and the C stability metric. Panels E–F show the stable C content of each biochar as a function of the original feedstock (g kg^{-1} feedstock), which is the product of the yield (g kg^{-1} feedstock), and the stable C content of each biochar. Solid and dashed lines show data for biochar produced from un-rinsed and rinsed biomass, respectively. All data are means ± SE (n = 3).

("Biochar × Temperature": $F_{3,\ 48}$ = 11.44, P < 0.001), but no significant effect of biomass rinsing ("Rinsing": $F_{1,\ 48}$ = 1.29, P = 0.260).

Ulva flake biochar produced at 300–600 °C supported the highest germination of radishes of 62–80%, in comparison to mean

germination of 45% in the no-biochar control (Fig. 4a). However, the germination of radish seeds decreased in treatments receiving *Ulva* flake biochar produced at 750 °C, with a mean germination of approximately 20% which was lower than germination in the no-biochar control (Fig. 4a). Treatments receiving biochar produced from *Ulva* pellets produced at 300–450 °C had lower rates of germination than the flake biochars produced at the same temperature (Fig. 4b). Germination of radishes in soils receiving the biochar pellets produced at 600–750 °C were variable but in most cases did not differ to the no biochar control (Fig. 4b). The pyrolysis temperature did not influence the germination rate of radishes grown in soils receiving biochar pellets (Fig. 4b). While variable, the hypocotyl growth data showed a general declining trend in growth with increasing pyrolysis temperatures (Fig. 4c, d).

The total production of radish biomass followed the same pattern as the germination data. There was an interaction between biochar (flake vs pellets) and pyrolysis temperatures ("Biochar × Temperature": $F_{3, 48} = 10.37$, $P < 0.001$). The production of radish biomass was greatest in pots receiving flake biochar produced from rinsed and un-rinsed biomass pyrolysed at 300–600 °C and all treatments receiving this biochar had higher plant

production than the no-biochar control (Fig. 4e). Overall the treatments receiving the flake biochar produced at 300–450 °C ranged from 102 to 137 mm of plant growth per pot, in comparison with 38 mm in the no-biochar control (Fig. 4e). As for germination, the total production of radish biomass in the soils mixed with flake biochar produced at 750 °C was lower, averaging 12–20 mm of plant growth per pot (Fig. 4e). There was no significant difference in total production of radish biomass in pots receiving biochar from pelletised biomass (Fig. 4f). The treatments receiving pelletised biochar ranged from 18 to 81 mm of plant production per pot (Fig. 4f). On the basis of these results, only the rinsed and un-rinsed flake biochars produced at temperatures between 300 and 600 °C were included in the outdoor pot trial, as these treatments were the only ones to consistently deliver greater germination and plant production than the no-biochar control.

While the radish productivity data were variable in the pot trial, the results confirmed the results of the laboratory experiment, with flake biochar produced at 300–450 °C supporting higher plant production than the no biochar control (Fig. 5). There was a significant effect of pyrolysis temperature on the production of plant biomass ("Temperature": $F_{2, 18} = 3.65$, $P = 0.047$), but no effect of
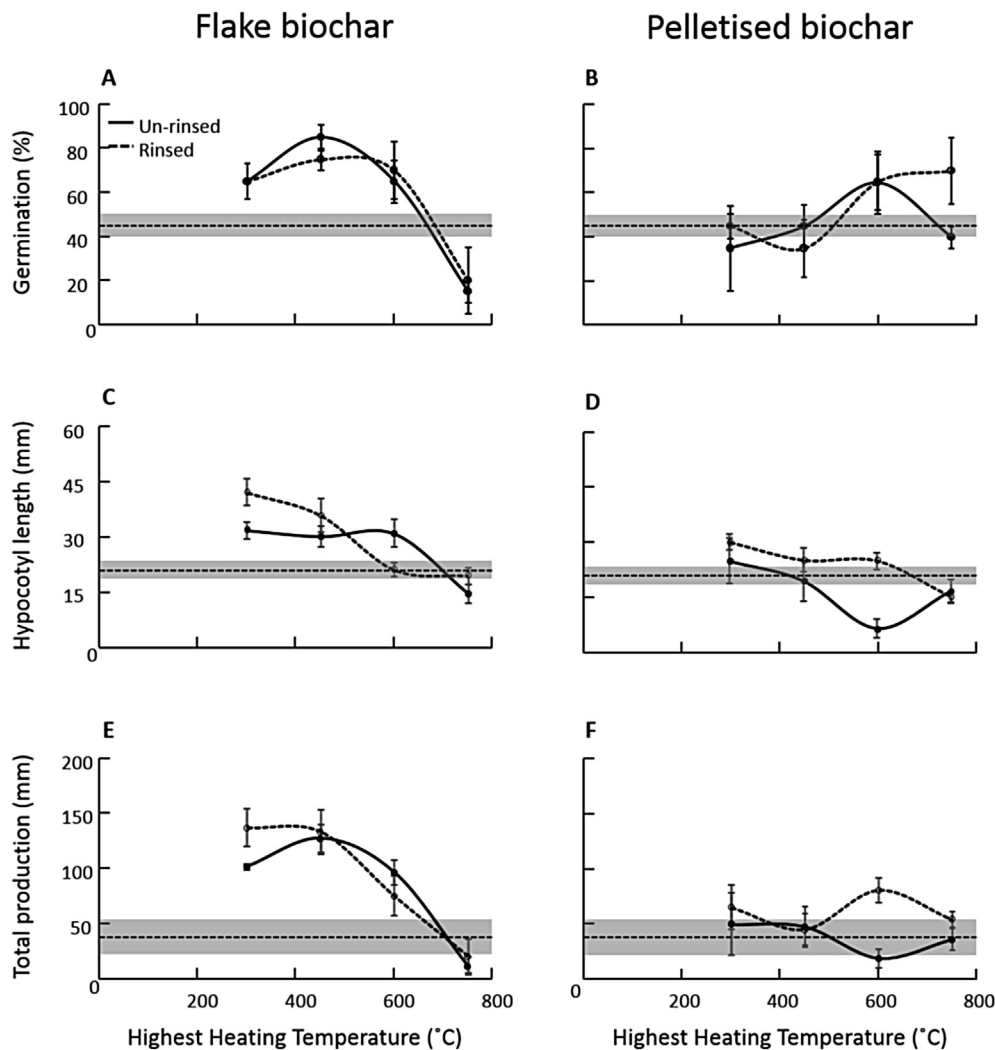

Fig. 4. The effect of biomass pre-treatment (rinsing and pelletisation) and pyrolysis temperature on the production of plants grown in biochar-amended soils. Panels A–B shown the germination of radishes (%), panels C–D show hypocotyl growth (mm) and panels E–F show total production of radishes (the product of germination and hypocotyl length). Solid and dashed lines show data for biochar produced from un-rinsed and rinsed biomass, respectively. The horizontal dotted line and grey shading shows the mean and SE of radish germination, growth or production in the control treatments without added biochar. All data are means ± SE (n = 4).
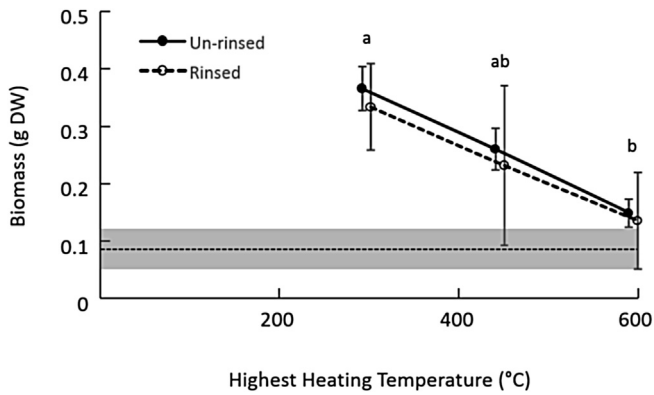
Fig. 5. The production of plant biomass (g dry weight) by radishes grown in biochar-amended soils in the outdoor pot trial. Solid and dashed lines show data for biochar produced from un-rinsed and rinsed biomass, respectively. The horizontal dotted line and grey shading shows the mean and SE of radish production in the control treatments without added biochar. All data are means ± SE (n = 4).

rinsing ("Rinsing": $F_{1, 18} = 0.15$, $P = 0.701$), nor an interaction between the two factors ("Temperature × Rinsing": $F_{2, 18} = 0.008$, $P = 0.992$). Plants grown in soils amended with biochar produced at 600 °C had a significantly lower production of biomass than those grown in soils amended with biochar produced at 300 °C (Fig. 5). The plant production in soils amended with biochar produced at 450 °C was intermediate (Fig. 5).

4. Discussion

While extensive research over the past decade has demonstrated that biochar can be used to sequester C in soils and improve soil fertility, few studies acknowledge that there are often direct trade-offs between the ability of biochar to sequester C and the utility of biochar as a soil ameliorant (Jeffery et al., 2013). We demonstrate here that while biomass pre-treatment and biochar production temperatures strongly influence the suitability of seaweed biochar for soil amelioration and C sequestration, it is possible to simultaneously maximize these benefits of biochar application using *U. ohnoi* as a feedstock.

The most direct measurement of C sequestration by biochar is the efficiency at which the predominantly labile C-content of the feedstock is converted into a stable form in biochar. The existing paradigm is that higher pyrolysis temperatures produce biochar with both a higher total C content and proportion of stable C (Jeffery et al., 2013). While this is true for most ligno-cellulosic feedstocks, few studies have considered high-ash biochars, such as those produced from seaweeds. We found that the percentage of total C that is "stable" in *Ulva* biochar (defined here as the fraction of the total C content that is resistant to oxidation) increased with temperature, but the gravimetric C sequestration (g stable C kg^{-1} feedstock) decreased. As temperatures increase there is a rapidly decreasing yield of biochar coupled with a slight reduction in the total C content. This combination of reduced yield and C content at higher temperatures means that more of the original C in the feedstock is sequestered at lower pyrolysis temperatures, despite the lower percentage of stable C in those biochars. In short, more stable C per unit of feedstock was sequestered in biochar when *U. ohnoi* was processed at 300 °C than at higher temperatures.

Similar patterns occur for some ligno-cellulosic feedstocks. Low ash feedstocks (e.g. pine and oak, <20% ash) have an increasing stable C content with increasing temperature, while higher ash feedstocks (e.g. rice husk, >20% ash) have a decreasing stable C content with increasing temperature (Enders et al., 2012). Even

minute quantities of salts in a feedstock can reduce the activation energy of the material, which promotes biochar formation at lower temperatures (White et al., 2011). The mineral content of high-ash biomass suppresses the cracking of high molecular weight hydrocarbons which promotes cross-linking reactions leading to char formation (Enders et al., 2012; White et al., 2011).

Our data demonstrate that, for high-ash feedstock, lower pyrolysis temperatures result in a biochar with greater agronomic utility. The N content of the *Ulva* biochars decreased with increasing temperature. This has also been demonstrated for giant reeds (Zheng et al., 2013), freshwater macroalgae (Roberts et al., 2015c) and waste water biosolids (Hossain et al., 2011). The EC of soil-biochar leachates also increased with pyrolysis temperature for the un-rinsed feedstock, demonstrating a higher concentration of soluble salts in the biochars as temperatures increased. While we have previously shown that metals (including Al, Cu and Zn) become less leachable from biochar with increasing pyrolysis temperatures (Roberts et al., 2015b; Roberts et al., 2015c), there was no reduction in the leaching of salts across the thermo-sequence we tested in this study. Pyrolysis temperatures in excess of 1000 °C are required for the volatilisation of dominant ions (e.g. Na) that form the ash component in algal feedstock (Lane et al., 2015). Therefore, the reduced agronomic utility of the higher-temperature biochars is likely due to a reduction in available N and an increase in the leaching of non-essential salts into soil-biochar pore waters.

A somewhat surprising result was that biochar produced from pelletised biomass had a lower efficacy as a soil ameliorant than the flake biochars. The pellets may have physically impaired the emergence of plant seedlings. There was also a lower EC in the leachates from soils amended with un-rinsed pellets relative to the flake, indicating the exchangeable elements on the surface of the biochars were more readily leached from biochar with the flake morphology. This may have meant the essential trace elements and macro-nutrients required for plant growth were less available in treatments receiving pelletised biochars due to the compaction of the material and the reduced surface area available for exchange. While it is known that the micro-topography of biochar can have strong effects on cation exchange capacity (CEC), the retention of water and nutrients, and the agronomic performance of biochar, few studies have explicitly compared the performance of biochars with differing gross-morphologies (Liang et al., in press). Our data show that this is an important consideration, as pelletised biochar consistently underperformed despite all other variables being held constant. Recent research suggests this may be due to interactions between smaller biochar particles and soil processes. Small biochar particles tend to stimulate soil enzyme and microbial activity to a greater extent, and have a greater liming effect in acidic soils, than large biochar particles (Liang et al., in press). Overall, the most effective biochar was produced from un-rinsed *Ulva* processed as flake at a pyrolysis temperature of 300 °C. This biochar sequestered 110 g stable C kg^{-1} *U. ohnoi* and tripled the production of radish biomass, relative to the no biochar control. This provides a clear framework for the production of biochar from cultivated and bloom-forming *Ulva* biomass.

Up to 2 million tonnes of *Ulva* biomass is beached in green tide events across the globe each year. The largest example of this phenomena has occurred along the coastline of the Shandong province of China since 2007, where 1–2 million tonnes of *Ulva* washes ashore each spring (Wang et al., 2015). The green tide is linked to the settlement of *Ulva* propagules on nets used to cultivate the red seaweed *Porphyra*. Approximately 4000 t of *Ulva* propagules are removed from the *Porphyra* nets each year and aggregate in the Yellow Sea, where they undergo rapid growth before washing ashore (Wang et al., 2015). The Brittany coast of France experiences a similar green tide, with 100,000 t of *Ulva* biomass beached

annually (Smetacek and Zingone, 2013). The 2 million tonnes of beached *Ulva* represents 200,000 t dry biomass (a 10:1 fresh: dry ratio) (Chung et al., 2011), so a biochar yield of 125,000 t per annum is feasible (62% yield for un-rinsed flake, 300 °C). This could ameliorate at least 12,500 ha of soil (10 t ha^{-1}) and return 15,000 t of stable C to soil annually (120 g kg^{-1} stable C). In addition, 5500 t N would be returned to soils for crop production. *Ulva* can also be cultivated in the effluent from finfish and invertebrate aquaculture, achieving productivities of up to 75 t ha^{-1} yr^{-1} in managed cultures while treating nutrient-rich effluent from aquaculture farms (Mata et al., 2015). This production of biomass would yield 47 t biochar ha^{-1} of seaweed cultivation each year, which would be sufficient to ameliorate at least 5 ha of soils, sequester 5.5 t C and recycle 2.2 t N into crop production per ha of seaweed cultivation. Clearly the conversion of *Ulva* biomass into biochar offers a promising pathway for recycling of dissolved nutrients from aquaculture and other coastal activities into soil ameliorants for agricultural crop production, while also sequestering C in soils.

Acknowledgements

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