Characteristics of Pine Gasification Ash and its Effects on *Chlamydomonas debaryana* Growth

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Large amounts of ash, generated from biomass gasification, often contaminate syngas and the ecosystem. This study showed that the ash obtained from the gasification of pine wood was primarily composed of carbon (15% to 25%), minerals (~21%), and oxygen (52% to 63%), and exhibited low surface area (8.4 to 11.2 m^2/g). The size of ash particles was between 600 nm and 600 µm. Calcium, potassium, and sodium were the three most common mineral elements in the ash. Leaching tests showed that adding ash to water raised the pH value from 5.7 to between 11.2 and 11.5, and, as time progressed, more mineral elements were released from the ash. For growing microalga *Chlamydomonas debaryana* in media containing ashes, no toxicity of pine ash was found.

Keywords: Biomass ash; Gasification; Pine wood; Leaching; Microalgae; Elemental analysis

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INTRODUCTION

Over the past decades, biomass gasification has been considered as a promising renewable energy technology, because of its potential for advanced applications (Kirkels and Verbong 2011). Gasification is an oxygen starved process whereby carbonaceous materials, such as woody products, vegetation, and solid waste, are converted to synthesis gas (syngas). The composition of syngas includes carbon monoxide (CO), carbon dioxide (CO_2), hydrogen (H_2), methane (CH_4), and nitrogen (N_2). Contaminants, such as ammonium (NH_3), hydrogen sulfide (H_2S), tar, water vapor, light hydrocarbon, char, and ash are additional products of syngas (Ransford *et al.* 2013).

Since the average ash content of wood is typically between 1.1 to 4.8 wt.% (Nanda *et al.* 2013), a huge quantity of biomass ash, including bottom ash and fly ash (*i.e.*, fine particles), result from the gasification processes because of the available abundant agricultural residues. Environment protection agencies have strict regulations on the emission for fine particles (US EPA 2013). Currently, the proper disposal for biomass ash is in the form of slurry, as a soil amendment, or as an additive in concrete (Barrows 2011). However, its usage is still limited and no longer satisfies the environmental concerns. New technically and environmentally friendly methods are a necessity, not only for the sustainable development of the gasification industry, but also for the protection of environment (James *et al.* 2012). Recent investigations suggest that the ash of coal combustion can be used as a nutrient for tree fertilizer (Pandey *et al.* 2009), which prompts that biomass ash may also serve as the fertilizer additive. Based on our knowledge, this is the first investigation into the potential utilization of wood ash as a fertilizer for microalgae.

For this paper, pine ash from woody biomass gasification was first characterized. And then a native North Carolina algal species *Chlamydomonas debaryana*, which was considered as a potential source of biofuels and food supplements (Zhang *et al.* 2014), was exposed to ash particles of different sizes, and the growth response was evaluated.

EXPERIMENTAL

Gasification Ash Collection and Fractionation

A fixed-bed downdraft gasifier (Gasifier Experimenters Kit, All Power Labs, Berkeley, CA, USA) was used to process pine wood chips (*Pinus* spp.) that were provided by Beard Hardwoods Inc. (Greensboro, NC, USA). The range of wood chip sizes was 6 to 10 mm. The gasification process was carried out at temperatures > 700 °C. Approximately, 1.0 kg of wood was processed per hour, producing around 2.1 m^3 of gas or 3.1 kW of heat from the burning cycle according to the manufacturer's procedure (All Power Labs 2011).

After each experiment, the pine ash was collected, sealed in plastic buckets, and stored at room temperature until further analysis. Size fractionation of the ash samples was conducted using specification sieves of 30, 50, 100, and 200 mesh, with equivalent opening sizes of 600, 300, 150, and 75 μm, respectively. The size of ash particles was mostly in the micron-scale range. In order to compare the effect of ash particles with that of nano-scale particles, particles of cerium-zirconium oxide (CeO_2-ZrO_2) (< 50 nm, Sigma-Aldrich, Mo, USA) was selected as the reference in this study. The reason of using CeO_2-ZrO_2 was due to its high stability and low bio-toxicity (Wu *et al.* 2007).

Characterization of Ash Particles

The size distribution of the ash particles was evaluated by dynamic light scattering (DLS) using a Zetasizer ZEN3600 analyzer (Malvern Instruments Ltd., Malvern, UK). The moisture content of the ash was determined according to the ASTM E871-82 standard (2006). The total ash content, representing the total inorganic fraction of the sample, was determined according to the ASTM D1102-84 standard (2007).

Elemental analyses for carbon (C), hydrogen (H), and nitrogen (N) were determined using a Perkin-Elmer 2400 CHN/S analyzer (Waltham, MA, USA). A mineral analysis was performed for additional elements (Ca, Na, K, Al, S, Fe, Mg, P, B, Mn, Si, Zn, Pb, Cd, and Cu) using a Varian 710-ES inductively coupled plasma optical emission spectrometer (ICP-OES) (Santa Clara, CA, USA). The oxygen (O) content was calculated by weight difference (Li *et al.* 2015; Yan *et al.* 2015).

Physical nitrogen adsorption was measured using a Micromeritics ASAP 2020 surface area and porosity analyzer (Norcross, GA, USA). The surface area was calculated using the Brunauer-Emmett-Teller (BET) method (Brunauer *et al.* 1938).

The surface topography of the solid residues was measured on a Hitachi SU8000 scanning electron microscope (SEM) (Tokyo, Japan) operating at 10 kV. For morphological observation, microalgae cells were observed using a Zeiss Axio Scope A1 microscope (Jena, Germany, UK).

Fourier transform infrared (FTIR) spectroscopy was performed *via* a Varian 670 FTIR spectrometer (Santa Clara, CA, USA), equipped with a single-bounce diamond attenuated total reflectance (ATR) accessory.

Leaching Tests

For quantifying the mobility of chemicals present in the pine ash in water, the ash was weighed and added into flasks containing 1.0 L of deionized water to make a final ash concentration of 2.0 g/L, and was stored at room temperature. The concentrations of nitrate, phosphorus, and potassium in solution were analyzed according to the LaMotte Smart 3 Colorimeter Manual (LaMotte 2011) over a period of 30 d.

Microalgae Strain and Cultivation Conditions

A green microalga of *Chlamydomonas debaryana* AT24 that was used in this study was originally isolated from the lagoon at the farm of NC A&T. This species showed lower temperature tolerance and higher lipid productivity than *Chlorella vulgaris* (Zhang *et al.* 2014). *C. debaryana* AT24 was cultured with deionized water, Bristol medium, or proteose medium. Bristol medium consists of the following ingredients: $NaNO_3$ (2.94 mM), $CaCl_2.2H_2O$ (0.17 mM), $MgSO_4.7H_2O$ (0.3 mM), K_2HPO_4 (0.43 mM), KH_2PO_4 (1.29 mM), and NaCl (0.43 mM). Proteose medium was made by adding 1 g proteose peptone into 1 L Bristol medium (Rahman *et al.* 2015). To investigate the effects of gasification ash and nanoparticles, the ash and nanoparticles (CeO_2-ZrO_2) were weighed and added into separate Pyrex® Erlenmeyer flasks containing 1.0 L of medium to make a final solute concentration of 2.0 g/L. Then, the flasks were autoclaved for 15 min at 121 °C and 100 kPa saturated steam. Media containing nanoparticles and ash particles (< 75 μm) were sonicated for 30 min. The cultivation of *C. debaryana* AT24 was conducted at room temperature under 150 μmol $m^{-2}s^{-1}$ for 10 to 120 d, and the light was provided by fluorescent light bulbs. The flasks were manually agitated at minimum once per day.

For evaluating the biomass yield, 50 mL of microalgal broth was collected from each flask, and centrifuged at 2600 × G and 20 °C for 15 min. Supernatants were analyzed for the contents of nitrate, phosphorus, and potassium. The biomass yield in grams per liter was used to evaluate the effect of the particles on the algal growth. All experiments and analyses were performed in triplicates.

RESULTS AND DISCUSSION

Characteristics of Ash

Ash samples were separated using woven wire test sieves into four size fractions: 300 to 600 μm (P1), 150 to 300 μm (P2), 75 to 150 μm (P3), and < 75 μm (P4). The total percentage of ash content was 27.2%, 26.9%, 20.8%, and 25.1%, respectively. Then, the P4 ash particles were measured using a DLS and the hydrodynamic diameter of ash particles ranged between 600 and 2000 nm, with a mean diameter of 1342 nm.

Scanning electron microscopy (SEM)

The P4 ash particles were analyzed using SEM (Fig. 1). Compared with the pine wood chips, the ash particles had irregular shapes and were smaller in size. The smallest particles were approximately 600 nm. The results were in good agreement with the measurement made on the basis of DLS.

Elemental analysis, ash content, and BET surface area

Tables 1 and 2 show the elemental analyses, ash content, and BET surface area of four ash fractions. The ash fractions with larger particle sizes contained more unburned

carbon than those with finer particle sizes, while the ash content of each fraction was inversely proportional to the carbon content. The BET surface areas of the ash samples were in a range of 8.4 and 11.2 m^2/g. The difference of BET surface areas among all ash samples, which was mainly due to the selection of measuring points, was not significant.

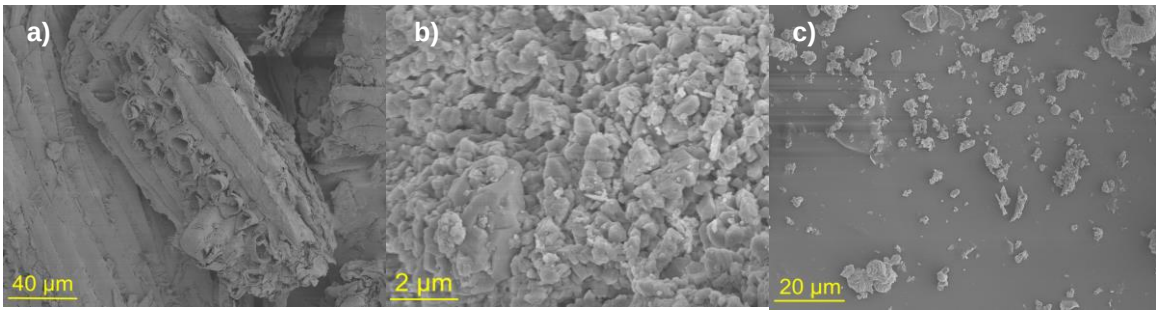

Fig. 1. Scanning electron microscopy (SEM) micrographs: a) pine wood chips, b) and c) P4 particles (< 75 µm)

Table 1. Elemental Composition, Moisture Content, Ash Content, and BET Surface Area

Ash particles*	Elemental composition (wt.%)				Ash content (wt.%)	BET Surface area (m^2/g)	Moisture (wt.%)
	Carbon	Hydrogen	Nitrogen	Oxygen			
P1	25.43	0.81	0.41	52.65	77.7	10.97	1.8
P2	18.71	0.78	0.36	58.95	87.2	10.29	1.6
P3	16.72	0.77	0.28	61.24	87.2	8.45	1.7
P4	15.09	0.81	0.21	63.39	89.8	11.19	1.7
Pine wood chip	52.00	6.06	1.24	39.60	2.9	1.23	3.1

*P1: 300 to 600 µm; P2: 150-300 µm; P3: 75 to 150 µm; P4: 0.7 to 75 µm

Table 2. Mineral Elements Analysis by ICP-OES (mg/kg)

Metal element*	P1	P2	P3	P4	Pine wood chip
Ca	133,031	146,938	146,991	150,864	6,686
Na	13,373	11,909	11,548	10,932	428
K	23,204	19,897	19,129	18,976	2,717
Al	14,016	9,849	8,287	6,318	29
S	8,220	9,082	9,909	11,044	866
Fe	6,091	5,474	4,586	3,778	48
Mg	5,250	5,014	5,018	4,746	270
P	1,499	1,571	1,803	1,844	175
B	1,336	1,424	1,533	1,586	123
Mn	982	806	818	857	BDL
Si	405	283	249	429	128
Zn	405	283	249	429	128
Total (wt.%)	20.7	21.2	21.0	20.5	1.1

*The contents of Pb, Cd, Cr, and Cu were below detection limit (BDL)

The ICP analyses showed that calcium (Ca), potassium (K), and sodium (Na) were the three most common elements overall. Because Ca is usually the most abundant inorganic element in pine wood, Ca comprised approximately half of all inorganic elements detected from the ash samples. The content of heavy metals was relatively low.

Fourier transform infrared spectroscopy analysis

Figure 2 shows the FTIR spectra of the organic functional groups during the progress of feedstock to ash residues. Spectral peaks were interpreted based on previous reports (Silverstein *et al.* 2005; Qian *et al.* 2013). The FTIR of the pine wood chips exhibited -OH groups (broad band between 3600 and 3000 cm^{-1}), aliphatic C-H stretching vibrations (bands of 2921 to 2845 cm^{-1}), asymmetric stretching of C=O in carboxylic groups (1550 cm^{-1}), and the dominant C-O stretch (1030 cm^{-1}) associated with the β-glycosidic bond in cellulose and hemicellulose.

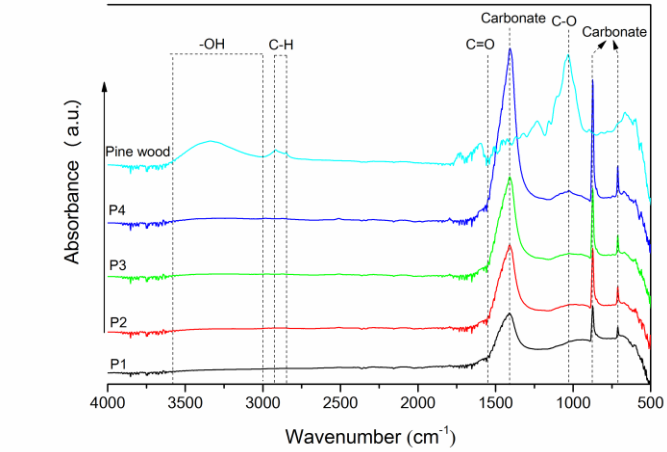

Fig. 2. FTIR spectra of the ash samples and pine wood

The FTIR spectra of ash samples were completely different from that of the pine wood. The disappeared peaks indicated that major components of pine wood decomposed in the gasifier. The peaks at 1400, 870, and 708 cm^{-1} corresponded to carbonate-related bonds.

Leaching Tests

The pH values and concentrations of nitrate (N), phosphorus (P), and potassium (K) were monitored over a period of 30 days. The results of the leaching tests (Table 3) showed that adding ash to water raised the pH value from 5.7 to between 11.2 and 11.5. As time progressed, more chemicals were released from the ash. Nitrate and phosphorus are major nutrients required for algal growth. In addition to nitrate and phosphorus, most other elements shown in Table 2 have the potential to be used as nutrients for algae growth.

Effects of Ash Particles on Algal Growth

C. debaryana was grown for 10 days in deionized water, Bristol medium, and proteose medium containing ash particles, which showed initial pH values of 11.3, 8.3, and 8.4, respectively. The algal biomass yield after a 10-d culture in deionized water was below 0.1 g/L, and its pH was reduced from approximately 11.3 to 10.9. The results indicated that

water containing the pine ash was not a favorable medium for *C. debaryana*. Possible reasons are that high pH may inhibit algal growth, or the release of ions from the ash was limited because of its low solubility.

Table 3. Leaching Tests of Pine Ash in Water

Particle	pH	Chemicals (ppm)	Day 0	Day 10	Day 20	Day 30
P1	11.6	Nitrate	0.6	1.1	1.1	1.2
		Phosphorus	0.55	0.75	0.79	0.93
		Potassium	41	44	52	63
P2	11.5	Nitrate	0.9	0.9	1.2	1.7
		Phosphorus	0.79	0.83	0.85	0.93
		Potassium	36	45	49	56
P3	11.3	Nitrate	0.6	0.75	1.1	1.3
		Phosphorus	0.61	0.63	0.71	0.85
		Potassium	37	50	57	62
P4	11.2	Nitrate	0.4	1.1	1.4	1.5
		Phosphorus	0.22	0.45	0.66	0.75
		Potassium	45	45	51	59

Bristol medium contains only minimal nutrients to keep algae alive, while proteose medium was rich enough to support rapid growth of algae. The starting N:P ratio of Bristol medium and proteose medium were 20:45 and 60:45, respectively (Hill 2014). When growing in Bristol or proteose media for 10-d, no notable inhibition of microalgal growth was observed for the either the nanoparticle (CeO_2-ZrO_2) or the ash fractions, with the exception being P1 (Fig. 3). The particle size of the P1 fraction was between 300 and 600 µm, and large particles may block the light illumination under higher algal cell density.

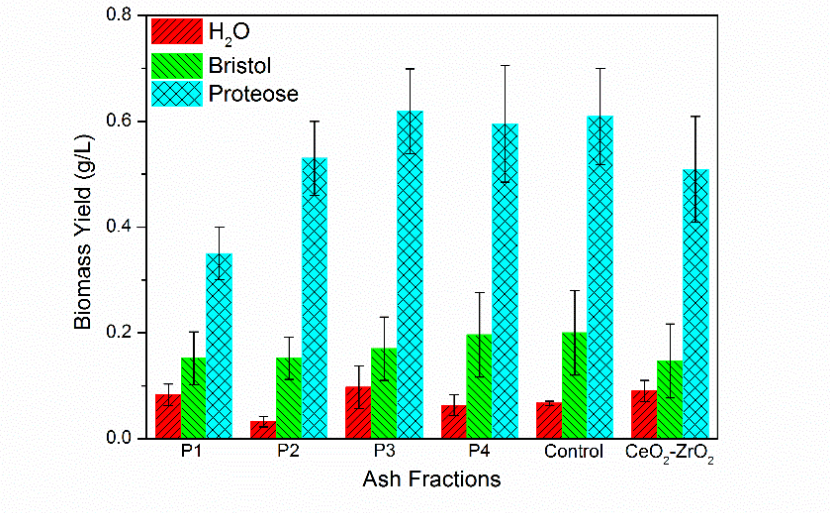

Fig. 3. Biomass yields of *C. debaryana* in media containing ash and nanoparticles after a 10-d culture. Control: media without addition of ash

To study the long-term effect of ash particles on the algal growth, *C. debaryana* cultures were maintained in media containing ash over a 120-d period. As shown in Fig. 4, the algal biomass production from ash media was relatively stable over a long period, while media containing only nanoparticles could not support the long-term growth of algae.

The initial contents of N, P, and K in the Bristol medium were approximately 20 ppm, 25 ppm, and 32.5 ppm, respectively. These nutrients were depleted within 60-d, and then the released ions from pine ash might serve as additional nutrients for algal survival. After 120-d, the concentration of N, P, and K in the Bristol medium were 0.8, 5.9, and 100 ppm, respectively. These results indicated that *C. debaryana* culture could be effective for the bioremediation of ash-containing wastewater. Furthermore, pine ash may be valuable as a fertilizer, providing nitrogen, phosphorus, and other micronutrients.

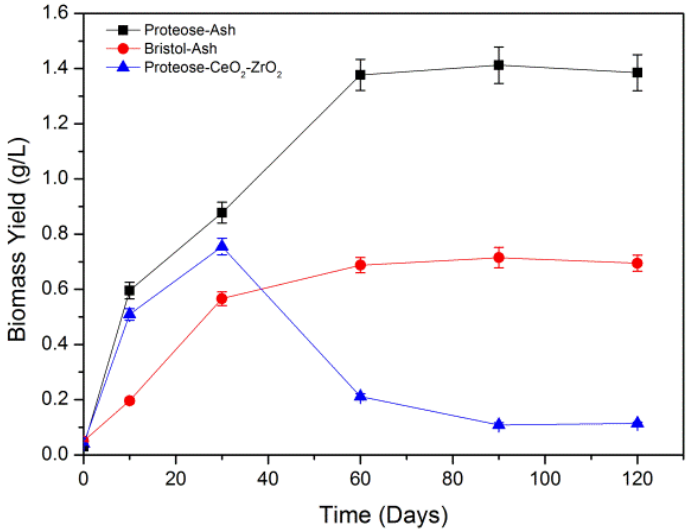

Fig. 4. Biomass yields of *C. debaryana* in media containing ash P4 and nanoparticles during a 120-d culture

Algae are often selected as the model microorganism for environmental assessment (Rai *et al.* 2004; Tripathi *et al.* 2008) because they are easily cultured under challenging growth conditions and are the primary producers in aquatic systems, permitting utilization of population growth (Vocke *et al.* 1980).

The effects of fine particles on algae might be toxic or beneficial. The reasons of toxicity include the dissolved ions, the interaction of nanoparticles with algae, and the entrapment of algal cells (Quigg *et al.* 2013). In this study, the algal cells were surrounded by ash particles or nanoparticles (Fig. 5); however, the size of ash particles is at the micron scale, and the toxicity was not considerable under the experimental conditions.

Syngas generally contains many contaminants, such as fly ash, carbon dioxide (CO_2), tars, sulfur compounds, nitrogen compounds, and alkali metals. The size of the particles that are released from a gasifier is normally in the range of 1 to 100 µm. A wet scrubbing process is typically employed to remove the particles in syngas for industry purposes.

The present results indicated that the particles in syngas produced from pine woods do not have a notable negative impact for the algal species, *C. debaryana*. Algal culture may purify the wastewater generated from the syngas cleaning process and provide positive effects for the ecosystems.

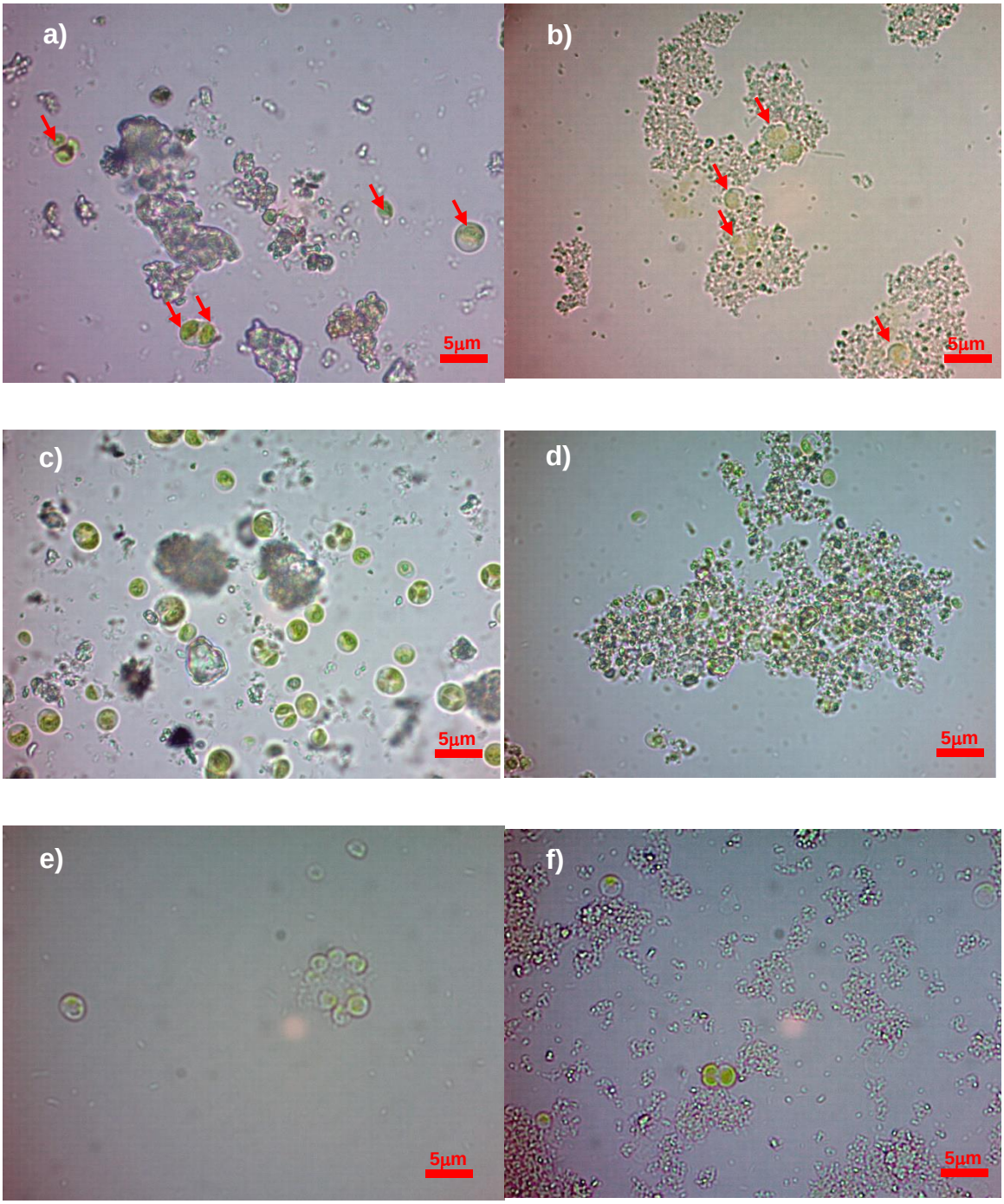

Fig. 5. Microscopic observations of *C. debaryana* in proteose media containing the ash fraction P4 (< 75 µm): a) Day 2, c) Day 7, and e) Day 90; and nanoparticles: b) Day 2, d) Day 7, and f) Day 90. The arrows show the algal cells

CONCLUSIONS

1. Ash obtained from the pine wood gasification process primarily consisted of carbon (15 to 25 wt.%), mineral (approximately 21 wt.%), and oxygen (52 to 63 wt.%), and it had low surface areas (8.4 to 11.2 m^2/g).
2. The size of the ash particles were between 0.6 to 600 μm. The Ca, K, and Na contents were the three most common minerals present in the ash, and Ca comprised approximately half of all inorganic elements detected.
3. Leaching tests showed that adding ash to water raised the pH value from 5.7 to between 11.2 and 11.5, and as time progressed, more chemicals were released from the ash.
4. No toxicity of pine ash was found for growing microalgae, indicating that pine ash might provide a valuable nutrient source for *Chlamydomonas debaryana*.

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