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**ENVIRONMENTAL INDICATORS FOR SUSTAINABLE PRODUCTION OF ALGAL  
BIOFUELS**

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June 24, 2014

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22 ABSTRACT

23           For analyzing sustainability of algal biofuels, we identify 16 environmental indicators  
24 that fall into six categories: soil quality, water quality and quantity, air quality, greenhouse gas  
25 emissions, biodiversity, and productivity. Indicators are selected to be practical, widely  
26 applicable, predictable in response, anticipatory of future changes, independent of scale, and  
27 responsive to management. Major differences between algae and terrestrial plant feedstocks, as  
28 well as their supply chains for biofuel, are highlighted, for they influence the choice of  
29 appropriate sustainability indicators. Algae strain selection characteristics do not generally affect  
30 which indicators are selected. The use of water instead of soil as the growth medium for algae  
31 determines the higher priority of water- over soil-related indicators. The proposed set of  
32 environmental indicators provides an initial checklist for measures of biofuel sustainability but  
33 may need to be modified for particular contexts depending on data availability, goals of  
34 stakeholders, and financial constraints. Use of these indicators entails defining sustainability  
35 goals and targets in relation to stakeholder values in a particular context and can lead to  
36 improved management practices.

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## 40 **1. Introduction**

41 Sustainability considerations influence the development of alternative sources of energy,  
42 including algal-based bioenergy. Algae hold promise as a future source of liquid fuel in part  
43 because of anticipated sustainability benefits such as the use of degraded, non-agricultural land  
44 (Gao et al. 2012, NRC 2012), high productivity per land area (Clarens et al. 2010), potential net  
45 greenhouse-gas (GHG) emissions benefits (Sander and Murthy 2010), and potential use of  
46 wastewater as a nutrient source (Woertz et al. 2009, Craggs et al. 2012). However, technologies,  
47 scenarios, and supply chains are still under development, and sustainability costs and benefits are  
48 influenced by the choice among many options (e.g., open pond versus photobioreactor, the latter  
49 being a closed device for generating biological products that uses sunlight or sugars for energy).

50 Progress toward sustainability can be estimated using indicators, which represent  
51 environmental or socioeconomic elements of sustainability (NRC 2010a, McBride et al. 2011).  
52 The focus of this paper is on environmental indicators of sustainable biofuel production.

53 The evaluation and selection of environmental sustainability indicators for algal biofuels  
54 have not kept pace with those activities for other feedstocks. Indicators of the sustainability of  
55 bioenergy pathways have been proposed by many institutions and researchers [e.g., Roundtable  
56 on Sustainable Biomaterials (RSB 2010), Global Bioenergy Partnership (GBEP 2011), McBride  
57 et al. (2011)] and are under development by others such as the International Organization for  
58 Standardization (ISO 2010). Most indicators, principles, and standards for bioenergy have  
59 focused on terrestrial, vascular plant feedstocks such as corn, switchgrass, and forest products  
60 (CSBP 2012). Some compilations of indicators and standards mention algae in the context of  
61 potential risk from genetically modified organisms (RSB 2010, Fritsche 2012). The U.S.

62 National Research Council (NRC 2012) published potential environmental impact and resource  
63 requirement metrics for the sustainable development of algal biofuels and listed the most  
64 important potential sustainability concerns but did not identify the most likely benefits or a  
65 practically measurable set of environmental sustainability indicators. Hence, technology  
66 development for algal biofuels is moving rapidly in the absence of clear means to define and  
67 quantify its sustainability.

68 A practical set of sustainability indicators is needed for algal biofuel processes and site-  
69 specific applications for several reasons. Indicators can be used to compare effects of different  
70 circumstances under which biofuels are produced, including different initial conditions.  
71 Alternatively, algal biofuel systems may be compared with business-as-usual fossil gasoline  
72 (Harto et al. 2010) or alternative diesel systems (Dinh et al. 2009, Harto et al. 2010). Indicators  
73 can be used to screen technologies for feasibility. Furthermore, indicators may be used to help  
74 with facility siting (Venteris et al. 2014). And indicators may be used as an early warning signal  
75 of changes in the environment (Cairns et al. 1993, Dale and Beyeler 2001) of an algae system or  
76 of system collapse. They can also be used to diagnose the cause of a problem.

77 A set of practical environmental sustainability indicators for bioenergy was proposed by  
78 McBride et al. (2011) to include six categories: soil quality, water quality and quantity, air  
79 quality, GHG emissions, biodiversity, and productivity. The indicators and indicator categories  
80 were science-based, considered many national and international efforts, and were intended to  
81 apply to a wide range of bioenergy systems, pathways, locations, and management practices, as  
82 well as feedstocks. The focus was on feedstock production—annual and perennial plants and  
83 residues from agriculture, forestry and related industry. Even for vascular feedstocks, the  
84 generic set of indicators developed by McBride et al. (2011) requires the adjustment of indicators

85 for some contexts (Efroymson et al. 2013), particularly for applications with limited budgets.  
86 While GBEP does not address the applicability of their 24 sustainability indicator categories to  
87 particular feedstocks, some are implicitly mentioned (e.g., harvest levels of wood resources), and  
88 algae is not (GBEP 2011).

89         Some analyses have considered how indicators apply to specific feedstocks. For  
90 example, Dale et al. (2013a) previously considered the applicability of a generic list of  
91 sustainability indicators (McBride et al. 2011) to *Eucalyptus*. They found that sustainability  
92 issues were consistent with those of other terrestrial feedstocks, but that the prioritization of  
93 environmental concerns was specific to *Eucalyptus*, with invasiveness and water use being  
94 particularly important for that feedstock. Though not addressed by this study, social acceptance  
95 was also important to sustainability of *Eucalyptus* for biofuel.

96         This analysis identifies environmental sustainability indicators that pertain to the majority  
97 of algal biofuel systems. The evaluation is based on how well salient characteristics of those  
98 biofuel systems, algae cultures, and strain selection characteristics match candidate indicators  
99 and selection criteria for indicators. This manuscript also discusses the indicator set in the  
100 context of future technology development. A wide variety of algal biofuel supply chains are  
101 under development with more than 60 pathways proposed (NRC 2012). We focus on eukaryotic  
102 photoautotrophic microalgae and cyanobacteria as feedstock organisms and consider the entire  
103 supply chain. The key question addressed in this manuscript is which environmental indicators  
104 of sustainability are especially important for biofuels produced from algae.

105

## 106 **2. Approach**

107 To select sustainability indicators for algal biofuels, we consider the broad range of  
108 indicators that have been recommended for bioenergy. Large sets of indicators, such as those  
109 recommended by GBEP and RSB, are examined. Special emphasis is placed on indicators  
110 proposed by McBride et al. (2011), which represent a focused, scientifically based, and practical  
111 set that were selected from a broad range of sources. We consider differences between algal  
112 biofuel and terrestrial biofuel systems and between the biology and production methods for algae  
113 and vascular plants. Algae strain selection characteristics are also part of analysis, for they lead  
114 to particular sustainability benefits or concerns or an emphasis on particular indicators. We  
115 examine indicators in six environmental categories—soil quality, water quantity and quality,  
116 biodiversity, air quality, and productivity. Indicators are selected based on specific criteria  
117 discussed below.

118

### 119 *2.1. Criteria for indicator selection*

120 The criteria for selecting sustainability indicators for algal biofuels include the following  
121 characteristics, as defined by Cairns et al. (1993), Dale and Beyeler (2001), and Catford et al.  
122 (2012).

- 123 1) Practical. Indicators should be straightforward and inexpensive to measure or simulate.
- 124 2) Widely applicable. Indicators that are only applicable to a small subset of algal biofuel  
125 pathways are not considered.
- 126 3) Predictable in response. For example, an indicator of biodiversity must consistently  
127 respond to a change in biodiversity.

- 128 4) Anticipatory of future changes. Adequate warning of a culture crash can lead to  
129 preventive management interventions and hence is particularly important for  
130 productivity.
- 131 5) Independent of scale. Indicators that are independent of temporal and spatial scale are  
132 more generally applicable to sustainability assessments, but some environmental  
133 indicators (e.g., tropospheric ozone) violate this criterion. Also, for many indicators (e.g.,  
134 water quality, biodiversity), it is not advisable to aggregate values from inside and  
135 outside ponds.
- 136 6) Responsive to management. Whereas temperature and light could be indicators of  
137 productivity, they cannot be effectively managed in open-pond systems.
- 138 7) Sufficient and non-redundant when considered collectively. Indicators should not be  
139 strongly correlated.

140 In addition, past data should be available in consistent units (Cairns et al. 1993). For example,  
141 Catford et al. (2012) eliminate indicators of invasion diversity and evenness indices that have  
142 been measured inconsistently across past studies. However, an advantage of the incipient  
143 development of algal biofuel facilities is that selected indicators can be measured consistently in  
144 the future.

145

### 146 **3. Differences between algae and terrestrial bioenergy supply chains**

147 Differences between algae and terrestrial plant feedstocks, as well as their supply chains  
148 for biofuel, influence the choice of appropriate sustainability indicators (Table 1). Algal biofuel  
149 production interacts with aspects of the environment across the entire supply chain (Figure 1),  
150 Algal biofuel supply chains differ somewhat from other bioenergy supply chains. For example,

151 crop protection methods are different (Table 1). Interactions between feedstock production  
152 systems and environmental variables differ between open pond systems and closed  
153 photobioreactors (Table 2). The magnitude of environmental effects may be greater during  
154 construction and decommissioning of open ponds for algae than for terrestrial bioenergy crops  
155 because of the change from land to water and back. As with other bioenergy systems, water  
156 quantity and air quality are affected throughout the supply chain (Figure 1).

157         Feedstock selection is the first step in the supply chain. Algae are selected or genetically  
158 modified based on characteristics that favor productivity, survival or other aspects of  
159 sustainability, such as a lack of known toxin production (Table 3). Characteristics related to  
160 environmental sustainability include CO<sub>2</sub>-absorbing capacity, limited nutrient requirements, and  
161 ability to flourish in brackish or saline water.

162         The use of water, nutrients, and CO<sub>2</sub> is different for algae and terrestrial feedstocks. The  
163 majority of water used in algae production is for growth media rather than for biomass, as in  
164 vascular plants. Many algal biofuel systems can use brackish or briny ground water or seawater  
165 rather than freshwater, and much of the water may be recycled, as little is incorporated in  
166 biomass. Unlike vascular plants, algae do not extract nutrients or water from local soil. Algae  
167 have the potential to remove nutrients from wastewater (Cai et al. 2013). Carbon dioxide is  
168 needed as an input for phototrophic algal systems, and collocation with CO<sub>2</sub> sources may be  
169 needed (Roberts et al. 2013).

170         Extreme weather events may affect terrestrial crops and aquatic algal biofuel crops and  
171 their environmental effects differently, but they lead to similar potential for crop loss. Drought  
172 can affect both terrestrial crops and open-pond algae with regard to the need for irrigation and  
173 replacement of evaporated water, respectively. Storms can cause slow leaks, overtopping of

174 ponds, or sudden releases of pond water, and these losses of nutrients and biomass can have  
175 environmental effects on adjacent waters and aquatic biota (Gressel et al. 2013).

176         The timing of harvest is different for algae and terrestrial crops. High algal biomass  
177 growth rates lead to more frequent (and sometimes continuous) harvesting compared to  
178 terrestrial feedstock systems (Milledge and Heaven 2013). In temperate climates, algae have a  
179 seasonal production pattern that affects the biofuel system and sustainability requirements. As a  
180 result, surplus biomass is available in the fall when temperatures drop (Behnke 2013). This  
181 biomass can be transformed to a commercially viable coproduct such as defatted animal feed  
182 (NRC 2012, Behnke 2013) or digested and applied to land as fertilizer (Frank et al. 2012).

183         Unlike terrestrial crops, algae in photobioreactors rarely interact with the surrounding  
184 ecosystem. In contrast, algae in open ponds are part of the ecosystem in several ways, as they  
185 are connected through air and sometimes linked by pathways to ground water or surface water,  
186 although liners are intended to disconnect the organisms from soil. Mammals and birds can visit  
187 these ponds and ingest and subsequently disperse their contents.

188         Most biofuel feedstocks tend to have the same sustainability implications as farming for  
189 food or fiber or growing wood for timber. However, algal biofuels might have different  
190 occupational hazards, such as a potential for toxin production or emission of harmful particulates  
191 if biomass is dried (Table 1). These hazards are more common in industrial processes than in  
192 crop production.

193         The storage and transport of algae and terrestrial feedstock are very similar. Therefore,  
194 these processes do not have unique implications for indicator selection for algal biofuels.

195         Options for conversion processes and transport of fuel are generally similar for algae and  
196 terrestrial bioenergy feedstocks, but the emphases can be different. The choice between

197 conversion processes that use wet algae (e.g., hydrothermal liquefaction, which can also be used  
198 for terrestrial crops) and dry algae can affect interactions with air quality. One conversion  
199 process that is used for algae but is different from terrestrial processes is the direct secretion of  
200 ethanol by live algae (Luo et al. 2010). There has been more emphasis on drop-in fuels produced  
201 by algae than for terrestrial crops. Drop-in fuels make pipeline transport possible and can  
202 obviate the need for blending (see Figure 1).

203 Algal-based fuels may be different in structure, impurities, and manufacturing process  
204 from other biofuels and petroleum fuels. Hence algae-based fuels may result in different  
205 effluents or emissions from those of competing fuels. Refineries for algal biofuels have the  
206 potential to produce biodiesel, green diesel, green gasoline, aviation fuel, ethanol, methane, and  
207 many coproducts (Pienkos and Darzins 2009). The likeliest coproducts with a large commercial  
208 market are animal feedstuffs (NRC 2012).

209 Most contemporary sustainability assessments of algal biofuels would occur prior to  
210 commercial development and therefore evaluate future scenarios. This emphasis on the future is  
211 similar to that of cellulosic sustainability assessments but different from analyses of corn grain  
212 ethanol and soybean diesel, for which commercial development is ongoing. Hence, the  
213 sustainability implications of cellulosic and algal-based biofuels are based on demonstration  
214 biofuels facilities, uses of the biomass for other purposes, or models.

215

#### 216 **4. Indicators of sustainability of algal biofuels**

217 Our analysis of sustainability of algal biofuels identifies 16 indicators that fall into six  
218 categories: soil quality, water quality and quantity, air quality, GHG emissions, biodiversity, and  
219 productivity (Table 4). These indicators were selected using the criteria presented above to be a

220 minimum, practical, and scientifically based set and are described below. Additional indicators  
221 that are applicable in particular contexts, have insufficient information about importance in algal  
222 biofuel systems generally, or may be applicable in the future, depending on technology  
223 development, are presented in Table 5.

224

#### 225 *4.1. Indicators of soil quality*

226 Soil quality is an important sustainability category for terrestrial bioenergy feedstocks  
227 such as crops that draw nutrients from the soil, and petroleum, for which exploration and  
228 production can contaminate soil. Soil quality affects productivity of vascular bioenergy crops  
229 and ecosystems but not algae used for biofuels. The main linkages of algal biofuels to soil  
230 quality are via short-term excavation for construction and ultimate decommissioning. Erosion is  
231 minimal because flat lands are preferred for algal biofuel facilities (Table 1), although berms can  
232 erode if they are not lined (Lundquist et al. 2010). Thus, many indicators, including soil organic  
233 carbon, total nitrogen, and extractable phosphorus (McBride et al. 2011), are not major  
234 determinants of sustainability of algal biofuel production as they are for biofuels from terrestrial  
235 feedstocks (Table 1). The NRC (2012) did not include soil quality as an important determinant  
236 of sustainable development of algal biofuels. However, aspects of soil quality, such as salinity  
237 and bulk density, are worthy of consideration, and waste disposal and comparative studies with  
238 other fuels are worthy of discussion.

239 Local soil salinization could occur when briny ground water is pumped to the surface for  
240 use in open ponds or photobioreactors or when water overtops saline ponds. The footprint of  
241 brine scars where oil drilling occurs can last many decades (Jager et al. 2005, Parish et al. 2013).  
242 Similarly, salinization of soil and water is a sustainability concern for agriculture in the Central

243 Valley of California (Schoups et al. 2005). GBEP recommends that in places where soil  
244 salinization is a hazard, soil electrical conductivity (EC) should be measured, for example, using  
245 USDA's electrical conductivity test (USDA 2001, Chapter 5; GBEP 2011). However, soil  
246 salinization indicators would be of low priority for most locations because of its small footprint.

247 In contrast to soil nutrients, bulk density, another measure proposed by McBride et al.  
248 (2011), is an important indicator relevant to subsoils below liners after ponds are removed or  
249 filled in (Table 1). For situations where there is a high risk of soil compaction, bulk density  
250 could be measured according to USDA's bulk density test (USDA 2001, Chapter 4) following  
251 decommissioning. Changes in bulk density could affect future productive capacity of the soil  
252 and hence are proposed to be part of the minimum set of sustainability indicators (Table 4).

253 Nutrient levels in soil could be affected if soil is amended with anaerobically digested  
254 algae (Table 5). The extent and frequency of such applications are uncertain, so soil nutrient  
255 measurements are not recommended at the current time.

256 In comparative studies of algal biofuel with biofuel from other sources or with petroleum  
257 diesel, many soil quality variables may be important to measure. In these comparisons the  
258 percentage of land for which soil organic carbon is maintained or improved (GBEP 2011) could  
259 provide useful information.

260

#### 261 *4.2. Indicators of water quantity*

262 The importance of water quantity indicators for the sustainability of algal biofuels is clear  
263 from the requirement of large volumes of nutrient-containing water as growth media (Murphy  
264 and Allen 2011), water for separation processes employed for biomass harvesting and fuel  
265 extraction (Luo et al. 2010), and water sometimes used for spray-cooling of photobioreactors

266 (NRC 2012). Because significant water volume is not used to build biomass, much of it can be  
267 recycled. However, evaporation is significant in open pond systems (NRC 2012, Talent et al.  
268 2014) (Table 1).

269 Consumptive water use is water withdrawal and loss through evaporation, runoff, or  
270 incorporation into a product. It is the only resource requirement indicator that we propose for  
271 algal biofuels (Table 4). Consumption or withdrawal is useful for evaluating water-use  
272 efficiency of particular technologies or pathways (GBEP 2011). For example, the direct  
273 secretion of ethanol without harvesting and extraction avoids significant water usage (Luo et al.  
274 2010). NRC (2012) proposes that indicators of the sustainability of freshwater requirements for  
275 growth of algae include consumptive freshwater use (kg water/kg fuel produced) and energy  
276 return on water invested (mJ/L) (Mulder et al. 2010).

277 Consumptive water use alone does not capture water quantity sustainability relative to  
278 local availability (NRC 2012). For this reason GBEP (2011) suggests that water withdrawals be  
279 expressed as a percentage of total actual renewable water resources or as a percentage of total  
280 annual water withdrawals. The alternatives that we recommend are 1) to interpret the  
281 consumptive water use indicator for algal biofuels with respect to water use for other local  
282 activities and 2) to add minimum base flow and peak storm flow as indicators of water quantity  
283 (see McBride et al. 2011) (Table 4). These indicators incorporate the spatial and temporal  
284 context of water usage. Consumptive water use is not as important for algal biomass production  
285 when brackish or saline waters are used (Table 1).

286 All water quantity indicators are influenced by evaporation. Even where briny or  
287 brackish waters are used, increasing salt content may necessitate additions of freshwater  
288 (Venteris et al. 2013, Talent et al. 2014).

289

### 290 *4.3. Indicators of water quality*

291 Water quality of effluents from algal biofuel facilities and receiving waters is influenced  
292 by the source of the water, nutrients and other amendments, and by the efficiency of nutrient use.  
293 Depending on the purpose of a sustainability assessment, either total nutrient concentrations in  
294 water bodies or nutrient mass exported, which represents the contribution of the algal biofuel  
295 system, may be important sustainability indicators. The quality of the culture water is not  
296 typically an environmental sustainability issue.

297 Four generic water quality indicators for bioenergy are concentrations of nitrate, total  
298 phosphorus, suspended sediment, and herbicide concentration in streams, as well as the loadings  
299 of these chemicals and materials exported to streams (McBride et al. 2011). Nutrient measures  
300 are recommended for algae production (Table 4), because slow leakage to groundwater or  
301 surface water may occur through ponds to many ecosystems, and breaching of pond berms  
302 would be a rare but real possibility that could lead to eutrophication of neighboring waters (Table  
303 1). If treated wastewater is used as a nutrient source, downstream concentrations of nutrients in  
304 streams may be positively affected by algae cultivation, but the risks to productivity from  
305 variable water chemistry and added microbes have yet to be overcome at large scale (Shurin et  
306 al. 2013). Recycling of nutrients and algae would also affect water quality (Murphy and Allen  
307 2011).

308 Some common indicators of water quality would not be very pertinent to algal biofuels.  
309 Algal cultures should not be a source of significant suspended sediment, because ponds are  
310 usually located at a distance from surface waters; they are located on relatively flat land  
311 (Benemann et al. 1982, Darzins et al. 2010, Wigmosta et al. 2011); there is no tilling of soil; and

312 excess biomass is not released to natural waters (Table 1). Herbicide concentrations would only  
313 be important sustainability indicators if herbicide-resistant strains are used, so we do not include  
314 them in the proposed set (Table 5). As algal biofuels move toward commercial development,  
315 antibiotics or antiseptic agents may become important crop protection chemicals (Table 5).

316 Because algae may be grown in coastal waters or saline or brackish groundwater (Table  
317 1), salinity of ground water or surface water will sometimes be an important sustainability  
318 indicator (Table 4), as recommended by the NRC (2012) and proposed for this minimum set of  
319 indicators. For example, Araujo et al. (2011) found that *Chaetoceros gracilis*  
320 (Heterokontophyta) and *Tetraselmis tetrathele* (Chlorophyta) are among the many species that  
321 can grow in saline water and produce high levels of lipids for biodiesel (see strain selection  
322 characteristics, Table 3). Unintentional leakage from open ponds or injection of saline waste into  
323 the ground could lead to the possible salinization of ground water or surface water in some  
324 environments.

325 The importance of measuring other contaminants of natural waters that potentially  
326 originate from algae cultivation systems is, as yet, unknown. Preliminary studies have measured  
327 metals in algae cultures originating from produced waters and soils with high elemental  
328 background levels (Sullivan 2013), but the significance of these metals for human health or  
329 ecological risk is unclear (Table 5). Toxins potentially produced by unfamiliar strains or  
330 opportunistic cyanobacteria should be monitored (Table 5). However, the ability to detect  
331 unknown toxins from less familiar strains is uncertain. Pathogens infecting algal cultures do not  
332 need to be monitored outside of algal cultures, because the source of these pathogens would be  
333 neighboring soils or waters.

334 Harvesting processes could raise water quality issues, depending on the methods used.  
335 Harvesting methods can include sedimentation, flotation, flocculation, centrifugation and  
336 filtration, or combinations of these (Uduman et al. 2010, Milledge and Heaven 2013). While  
337 most methods do not have implications for water quality, flocculation may require chemicals that  
338 would need to be measured in effluents or possibly streams (Table 5). Potential flocculants  
339 include inorganic chemicals such as aluminum and iron salts, synthetic organic polymers, and  
340 natural inorganic and organic products (Milledge and Heaven 2013, Vandamme et al. 2013).  
341 Algae cultivated in brackish water and seawater tend to require higher flocculant concentrations  
342 than freshwater species (Sukenik et al. 1988). Because it is uncertain if flocculation will be a  
343 dominant harvesting method in the future and which flocculants will dominate, we do not  
344 propose flocculant water quality indicators for algal biofuels.

345

#### 346 *4.4. Indicators of GHG flux*

347 GHG flux associated with algal biofuel occurs at every step of the supply system. To  
348 determine net GHG emissions of these pathways, many factors need to be considered. CO<sub>2</sub> can  
349 be added from flue gas, reducing power plant emissions (Kadam 1997, Orfield et al. 2014).  
350 Losses of CO<sub>2</sub> from open ponds influence net emissions (Table 1). Although CO<sub>2</sub> can be  
351 temporarily sequestered from industrial processes by algae (Menetrez 2012), the decomposition  
352 rate of waste biomass is also pertinent (Fernandez et al. 2012).

353 Processes in the biofuel supply chain that demand high energy input can lead to  
354 comparable CO<sub>2</sub> emissions. Stirring cultures is a power-intensive (Stephenson et al. 2010) and  
355 therefore a CO<sub>2</sub>-emitting process. Similarly, moving the water to and from the dewatering step,  
356 as well as thermal drying, is energy- and CO<sub>2</sub>-intensive (Frank et al. 2012, Weschler et al. 2014).

357 CO<sub>2</sub> is also related to nutrient demand (Clarens et al. 2011) and productivity (Frank et al. 2012).  
358 Frank et al. (2012) found that calculations of net GHG emissions were highly dependent on  
359 biogas production parameters, including “yields from digesters, yields from gasification, fugitive  
360 emissions, nutrient recovery rates, and electrical efficiency of the [Combined Heat & Power]  
361 generator.”

362 Fugitive methane and N<sub>2</sub>O may also be emitted during the cultivation process. Emissions  
363 from open ponds have not been studied (NRC 2012). Methanogenesis is possible from anaerobic  
364 cultures, especially if they crash, but the process is expected to be rare. N<sub>2</sub>O emissions have  
365 been measured from *Nannochloropsis salina* (Eustigmatophyceae) under a nitrogen headspace  
366 (Fagerstone et al. 2011), and *Nannochloris* (Chlorophyta) in coastal open-pond systems have  
367 been found to have high emissions of N<sub>2</sub>O during senescence (Florez-Leiva et al. 2010). But  
368 emissions are expected to be low under aerobic conditions.

369 Frank et al. (2012) estimated methane and N<sub>2</sub>O emissions from anaerobic digestate solids  
370 used as crop fertilizer, based on the Intergovernmental Panel on Climate Change (IPCC)  
371 proportions for organic fertilizer. IPCC (2010) acknowledges that emissions factors vary widely  
372 based on region, climate, and soil chemistry. The estimates for fugitive methane and N<sub>2</sub>O for  
373 algal biofuels were 14% and 23% of the whole pathway GHG emissions, respectively (Frank et  
374 al. 2012). Emissions from catalytic hydrothermal gasification processes may be lower than those  
375 from anaerobic digestion (Frank et al. 2012).

376 Other options for waste disposal can affect net GHG emissions. For example, Luo et al.  
377 (2010) assumed that annual disposal of cyanobacteria biomass would be via deep well injection,  
378 which could result in a slight net GHG reduction for the photobioreactor system.

379 GHG emissions indicators also reflect land-use change that would be attributable to algal  
380 biofuel systems. Land converted to algal biofuels is expected to include industrial brownfields,  
381 rangelands, deserts, abandoned or unproductive farmland, dredge spoil islands, or other coastal  
382 areas (NRC 2012). Depending on the CO<sub>2</sub> storage associated with the baseline land condition,  
383 the algal biomass production system may increase sequestration (e.g., if the prior land use was a  
384 brown field or desert with little vegetation) or decrease it (e.g., in the unlikely case that the  
385 previous land cover was forest).

386 Carbon-dioxide-equivalent emissions is a commonly endorsed and scientifically based  
387 indicator for tracking net GHG emissions. This indicator accounts for the 100-year global  
388 warming potential of methane being 25-34 times that of CO<sub>2</sub> (IPCC 2007, Shindell et al. 2009)  
389 and of nitrous oxide being 300 times that of CO<sub>2</sub> (NRC 2010b). This indicator is highly  
390 adaptable to changes in technology, because all GHG emissions can be translated into these  
391 units.

392 Under large-scale commercial development, changes in albedo and potential effects on  
393 local weather conditions should be studied, as well as GHG emissions. Recent papers show that  
394 tradeoffs between carbon sequestration and local warming or cooling from albedo are an  
395 important research area (Jackson et al. 2008), including research on bioenergy crops (Georgescu  
396 et al. 2013).

397

#### 398 *4.5. Indicators of biodiversity*

399 Algal biofuel production could affect aquatic or terrestrial biodiversity. Two general  
400 biodiversity indicators that have been proposed for sustainability of bioenergy include presence  
401 and habitats of taxa of special concern (McBride et al. 2011). “Taxa of special concern” can

402 encompass valued, invasive, or undesirable species, genera, or functional groups. Here we  
403 discuss the biodiversity of the algae culture itself as well as the aquatic and terrestrial  
404 biodiversity of the surrounding landscape.

405         For most pond cultures, an indicator of pond diversity is not necessary, as maintaining  
406 diversity in pond cultures will rarely be an environmental goal. In many algal biofuel systems, a  
407 monoculture is desired, but invasion by other algae, bacteria, zooplankton, and other organisms  
408 is likely (see section on productivity). In some biofuel systems, cultures of algae could be  
409 diverse, with select combinations of strains of algae decreasing risk from grazers (Mayfield et al.  
410 2013), or multiple species (Stockenreiter et al. 2012) or trophic levels (Smith et al. 2010)  
411 potentially increasing productivity.

412         Moreover, monitoring the presence of feedstock species at a distance from open ponds is  
413 not a priority unless nonnative species or strains are used (Gressel et al. 2013). Many eukaryotic  
414 microalgae and cyanobacteria are cosmopolitan in their spatial distributions (Hoffman 1994,  
415 1996), so their dispersal through air (Grönblad 1933), soil, or via animal vectors (see references  
416 in NRC 2012) from ponds should not affect biodiversity (Table 1).

417         However, if there is a breach in a pond or photobioreactor and a large quantity of algae  
418 and nutrients are released to aquatic ecosystems, then some algal taxa may bloom, potentially  
419 causing changes in the native community. Measures of abundance are superior to measures of  
420 occupancy as indicators of invasiveness or blooming of algae, and abundance of the introduced  
421 species or strain is recommended as an indicator of aquatic biodiversity (Table 4). Relative  
422 measures of alien species richness (Catford et al. 2012) are not recommended in this case,  
423 because for monocultures only one introduced species would be of concern. In addition to

424 monitoring the abundance of algae, we recommend the presence or absence of valued (e.g., rare)  
425 aquatic species as an indicator.

426         The indicators “presence of taxa of special concern” and “habitat area of taxa of special  
427 concern” for the particular context are appropriate indicators for effects on terrestrial species  
428 (Table 4). Terrestrial habitat displacement or fragmentation effects can result from the  
429 infrastructure of ponds, photobioreactors, and buildings for conversion and storage. These  
430 displacement effects are typical of any industry. Moreover, wildlife may drink from algal  
431 biofuel ponds, with potential toxic effects to individuals from metals, salinity, or toxins from  
432 opportunistic cyanobacteria (Kotut et al. 2010). Population demographic effects are also  
433 possible if migrants change their trajectories because of a new water source. Following the  
434 measurement of these indicators of biodiversity, more detailed measurement and analysis of  
435 effects may be needed.

436

#### 437 *4.6. Indicators of air quality*

438         Air quality indicators relate to regional human health, occupational health, or ecosystems.  
439 Air emissions can occur during feedstock production, processing, and transportation and use.  
440 McBride et al. (2011) recommended a suite of four indicators, namely tropospheric ozone,  
441 carbon monoxide, total particulate matter less than 2.5  $\mu\text{m}$  diameter and total particulate matter  
442 less than 10  $\mu\text{m}$ . The NRC Committee on Sustainable Development of Algal Biofuels (NRC  
443 2012) suggested that air quality indicators may include concentrations of volatile organic  
444 compounds (VOCs) and odorous secondary metabolites for open pond systems; particulates for  
445 active drying processes; air concentrations of solvent used for extraction processes; and  
446 particulates, hydrocarbons and acid gases for pyrolysis, if used (NRC 2012). We propose that

447 concentrations of odorous chemicals be considered a social sustainability indicator rather than an  
448 environmental sustainability indicator, so they are not included here. GBEP (2011) recommends  
449 consideration of NO<sub>x</sub> and SO<sub>2</sub>, as well as large and small particulates. The GREET model  
450 estimates emissions of six EPA criteria pollutants: CO, VOCs, nitrogen oxides, sulfur oxides,  
451 PM10 and PM2.5 (Frank et al. 2011a), without a judgment about their relative importance  
452 compared to other measures. Aerosols and acid gases have also been considered (NRC 2012).

453 Evidence supporting the selection of particular indicators of air quality for algal biofuels  
454 is varied, with some chemicals actually measured and others assumed to be important based on  
455 emissions from natural ponds containing algae, tailpipe emissions from other biofuels, and  
456 preliminary scientific results (see Appendix 1). Few studies of air emissions from algal biofuels  
457 are available, but one study of emission rates for a marine vessel operating on 50% hydrotreated  
458 algae diesel [and 50% ultra-low sulfur diesel (ULSD)] suggests that total particulate matter less  
459 than 2.5 µm in size (PM2.5) is an appropriate sustainability measure, as well as NO<sub>x</sub> and CO  
460 (Khan et al. 2012). All were reduced when the fuel blend was used, compared to the ULSD.

461 The selection of particular air quality indicators depends on the exact pathway and supply  
462 chain for algal biofuel (Appendix 1) and the purpose of the assessment. Particulates are  
463 important to measure if drying biomass is part of the fuel pathway and are always important for  
464 end-use, but they are less important at the conversion step if crude oil is extracted from wet algae  
465 (e.g., Moreno 2013). Ozone is a useful integrative air quality indicator because it is formed by a  
466 reaction of sunlight with nitrogen oxides and hydrocarbons and removes aldehydes. However, it  
467 is not easy to attribute ozone to particular vehicle and fuel sources, because it may be formed at a  
468 distance away from the source. Thus, the purpose of the sustainability assessment will determine  
469 whether ozone is a useful indicator.

470           Some indicators apply primarily at the local or occupational scale (e.g., toxins, VOCs).  
471 VOCs have been detected as emissions from open ponds (personal communication from Paul  
472 Zimba in NRC 2012). These chemicals may also be emitted from solvents used in extractions  
473 (e.g., toluene or hexane for upgrading the product following hydrothermal liquefaction, Liu et al.  
474 2013). No evidence suggests that combustion of algal biofuels produces VOCs in greater  
475 quantities than non-algal biofuels.

476           We propose that air quality indicators for algae include tropospheric ozone, carbon  
477 monoxide, total particulate matter less than 2.5  $\mu\text{m}$  diameter and total particulate matter less than  
478 10  $\mu\text{m}$  (Table 4). More research is needed to understand whether VOCs should be selected as an  
479 air quality indicator for algal biofuels (Table 5).

480

#### 481 *4.7. Indicators of productivity*

482           Productivity is a measure of the efficiency of biofuel production, and it may also be an  
483 economic or environmental measure. Aboveground net primary productivity, defined as the net  
484 flux of carbon from the atmosphere to the aboveground parts of green plants per unit time, is an  
485 environmental sustainability measure for biofuel derived from vascular plants, because of its  
486 relationship with photosynthesis and respiration (McBride et al. 2011). Aboveground net  
487 primary productivity sometimes includes algae (e.g., Ewe et al. 2006), but the term  
488 “aboveground” implies that there are roots belowground. Primary productivity is also related to  
489 secondary productivity, or the efficiency of generation of biomass of consumers in an ecosystem.  
490 For photosynthetic organisms, yield of biomass (and ultimately, fuel) is related to primary  
491 productivity. As with biodiversity and other indicators, it is important to assess both the  
492 productivity of algae and productivity of the neighboring and displaced ecosystems.

493           The productivity of algae is influenced by many abiotic environmental conditions,  
494 including temperature (Waller et al. 2012), light (Wondraczek et al. 2013), and wind-blown  
495 materials in arid or semi-arid areas that become sediment in open ponds and that constitute ash in  
496 conversion processes (J. Sullivan, Los Alamos National Laboratory, pers. comm. May 2013;  
497 Sayre 2013). Neutral lipid production by some strains is enhanced under nitrogen limitation (Li  
498 et al. 2011). Biotic conditions such as microbial community structure and the abundance of  
499 predators, pathogens, and self-shading by other algae also affect productivity (Kazamia et al.  
500 2012, Shurin et al. 2013). Whether productivity of algae represents an environmental indicator  
501 relates to the extent to which algal biofuel cultures are part of the ecosystem, which is  
502 determined by how the efficiency of production relates to other environmental variables and  
503 whether algae are available for consumption.

504           Another linkage between productivity and environmental sustainability is the relationship  
505 with land area. Algae cultures grown for biodiesel are anticipated to use a small fraction of the  
506 land area required to produce biodiesel by vascular plants (Groom et al. 2008, Clarens et al.  
507 2010). This environmental benefit can be quantified with a productivity indicator that has land  
508 area in the denominator.

509           Clearly, the primary productivity of algae in photobioreactors is not related to many  
510 environmental variables other than net GHG emissions (which can be measured more directly)  
511 and therefore is not as important a measure of environmental sustainability in closed systems as  
512 it is for terrestrial feedstocks (Table 1). The primary production associated with closed systems  
513 would not be related to secondary production in most contexts. Algae productivity would  
514 typically be more related to economic sustainability than environmental sustainability.

515           The feasibility of open pond cultivation (and to a lesser extent, cultivation in closed  
516 photobioreactors) is highly dependent on controlling contamination and culture collapse (Gao et  
517 al. 2012, Letcher et al. 2013) through crop protection (Smith and Crews 2014). Potential agents  
518 of collapse include zooplankton predators, viruses, bacteria, fungi, and competitive algae. The  
519 frequency, extent, and duration of culture collapses may be measurable or predictable, affecting  
520 yield. The density of particular pathogens or parasites or their DNA may be an early warning  
521 sign of culture collapse (e.g., Letcher et al. 2013), but the most important pathogens to measure  
522 in each region for each desired monoculture are unknown. Some researchers are measuring  
523 environmental conditions and metagenomes of algal samples from collapsed ponds to develop  
524 probes that may serve as early warning indicators of collapse (Lane 2013). A suitable surrogate  
525 for pathogens or their genomes is the density of algae or chlorophyll and, ultimately, the rate of  
526 change of that value through time. The frequency of reversion of genetically modified algae will  
527 also affect yield, but, when commercial-scale applications are deployed, this potential issue  
528 should be resolved.

529           For the ecosystem outside of the algae culture, aboveground net primary productivity is  
530 an appropriate sustainability indicator. GBEP (2011) proposed a somewhat different but related  
531 indicator, productive capacity of the land and ecosystems. Both indicators would be applicable  
532 to terrestrial productivity of algae production locations after the cessation of production.

533           We propose that current and past productivity of an algal biofuel system be measured as  
534 yield of carbon per land area (Table 4), but we acknowledge that the yield of fuel from these  
535 fairly isolated feedstock systems represents economic sustainability more than environmental  
536 sustainability. Because of the potential for crashes of algae cultures in open ponds, pathogen  
537 densities are important measures of future productivity in these systems, but which pathogens are

538 most important to measure in specific locations is still uncertain (Table 5). Aboveground net  
539 primary productivity is an important indicator for neighboring or displaced ecosystems.

540

#### 541 *4.8. Indicators of CO<sub>2</sub> resource requirements*

542 Resource inputs are an important aspect of sustainability if the resource is finite and is in  
543 decline, if the resource is being used at a different rate from replenishment, or if resource  
544 availability limits the potential locations of proposed facilities. We have discussed water  
545 quantity indicators in the context of regional supply. And we previously considered the  
546 depletion of non-renewable energy resources to be an indicator of socioeconomic sustainability  
547 effects, rather than an environmental indicator (Dale et al. 2013b). However, no generic  
548 sustainability indicator scheme for bioenergy has proposed CO<sub>2</sub> availability as a sustainability  
549 indicator, because it is pertinent only to algae. The NRC (2012) proposed mass of CO<sub>2</sub> required  
550 per liter of fuel produced and mass of CO<sub>2</sub> required per tonne dry biomass of algae as  
551 sustainability indicators, based on the units of nutrient requirements recommended by GBEP  
552 (2011).

553 Algae can fix CO<sub>2</sub> to produce biomass with greater efficiency and speed than terrestrial  
554 plants (Pienkos and Darzins 2009). They require about 2 g of CO<sub>2</sub> per g biomass produced  
555 (Pienkos and Darzins 2009) or 3.7 to 5.5 kg CO<sub>2</sub> per liter of algal oil (Pate et al. 2011).  
556 Supplemental CO<sub>2</sub> may be needed to reach productivities that are economically competitive  
557 (NRC 2012), and CO<sub>2</sub> may be the most limiting nutrient for algae. One potential source of CO<sub>2</sub>  
558 is power plant flue gas (Kadam 1997, Orfield et al. 2014). Another is natural repositories in the  
559 earth (Liu et al. 2013). Still another could be sodium bicarbonate (Pate et al. 2011).

560 CO<sub>2</sub> requirement is only a useful sustainability indicator if it varies with the biofuel  
561 supply chain and can be reduced with specific management practices. The solubility of carbon  
562 dioxide in water varies with temperature and pH, and the rate of CO<sub>2</sub> exchange between air and  
563 water depends on the surface area and turbulence of the water. Different systems will be more or  
564 less efficient in their use of CO<sub>2</sub>, within a small range. An alternative sustainability indicator  
565 would be supplemental, non-recycled CO<sub>2</sub> required/L of fuel produced, suggesting that the use of  
566 CO<sub>2</sub> produced by a power plant is more sustainable than purchased bicarbonate. An additional  
567 qualitative indicator might be the presence or absence of flue gas within a certain distance of an  
568 algal biofuel facility.

569 However, these components of sustainability could be captured either in GHG emissions  
570 indicators or in profitability, a socioeconomic sustainability indicator category (Dale et al.  
571 2013b). Aside from cost, CO<sub>2</sub> is not a regionally limiting nutrient. And we do not believe that  
572 the efficiency of CO<sub>2</sub> use can be controlled much by management practices. Therefore, we do  
573 not propose an environmental sustainability indicator related to CO<sub>2</sub> use.

574

## 575 **5. Discussion and conclusions**

576 We have proposed a practical, scientifically-based set of 16 environmental sustainability  
577 indicators for algal biofuels. The indicators may be used in concert with models and frameworks  
578 for comparing algae scenarios with each other, comparing them with other transportation fuel  
579 systems (Frank et al. 2012), and using them for other sustainability purposes. Eventually, these  
580 indicators may be used to set sustainability targets and to develop recommended management  
581 practices for algal biofuel systems.

582 Indicators were selected to be practical, widely applicable, predictable in response,  
583 anticipatory of future changes, independent of scale (where possible), and responsive to  
584 management. Clearly, there are compromises among selection criteria (Niemi and McDonald  
585 2004). Tradeoffs commonly relate to the usefulness versus the cost of information, the quality of  
586 the information versus the ease of measurement, and the specificity versus the generality of the  
587 indicator (Cairns et al. 1993, Catford et al. 2012).

588 The set of sustainability indicators for algal biofuels is very similar to the generic set  
589 proposed for bioenergy by McBride et al. (2011), with indicators proposed in each of six  
590 categories: soil quality, water quantity and quality, biodiversity, GHG emissions, air pollution,  
591 and productivity. Many indicators, such as CO<sub>2</sub>-equivalent emissions, are important to measure  
592 for all fuel production systems, whereas others, such as salinity, are only important for some  
593 algal biofuel systems. Although photobioreactor systems are different in structure and  
594 environmental connectivity from open-pond systems, the sustainability indicators are generally  
595 the same, though they may be prioritized differently for particular assessments. An examination  
596 of some of the main criteria for selecting algal strains suggests that few of those characteristics  
597 influence whether a sustainability indicator is chosen; instead they have more influence on the  
598 importance of the indicator. Concerns about genetically modified organisms differ in intensity  
599 from those of unmodified organisms, but it is not clear that effects will differ in kind.

600 The similarity of this set of sustainability indicators to a generic set of indicators for  
601 bioenergy means that most of the factors that need to be measured are not dependent on the  
602 obvious differences between algae and vascular plants or between the dominant supply chain  
603 steps or on the algal traits that are selected. For this reason, most of these indicators should not  
604 change as technologies narrow to a set that is commercially viable.

605           Nonetheless, there are a few differences between these indicators and those that have  
606 previously been recommended for bioenergy (McBride et al. 2011). Regular monitoring of soil  
607 nutrients, suspended sediment in streams, and herbicide loadings to streams are not usually  
608 necessary for algae production; water quality indicators should include salinity if saline water is  
609 used; and aquatic biodiversity indicators should include species richness for streams and  
610 abundance of potentially invasive algae.

611           Because of the nascent technology development for algal biofuel systems, research is  
612 needed on other environmental factors before some candidate indicators can be proposed or  
613 eliminated. These include toxins, metals, flocculants, and crop protection chemicals in water as  
614 indicators of water quality; volatile organic compounds as an indicator of air quality; and  
615 pathogen densities as an indicator of productivity.

616           It is challenging to propose generic sustainability indicators for algal biofuels because  
617 assessment purposes are not generic (Efroymson et al. 2013), and it is uncertain which  
618 technologies will prevail in the future. Most current algal biofuel systems, especially those using  
619 strains with high oil content, produce feedstock in open ponds (Menetrez 2012), but it is unclear  
620 whether open-pond systems or photobioreactors will become dominant. Hence indicators for  
621 open-pond and photobioreactor systems and for saline and freshwater systems are included in the  
622 proposed set. However, components of the biofuel pathway (e.g., drying biomass, anaerobic  
623 digestion and disposition of waste) will influence the sustainability indicators that are selected  
624 for particular assessments.

625           In contrast, some aspects of the biofuel system will not influence sustainability indicator  
626 selection. Conversion processes will probably not affect the selection or measurement of

627 sustainability indicators, unless they alter other steps of the supply chain (e.g., hydrothermal  
628 liquefaction not requiring a drying step or air quality indicators for that step).

629         The purpose for sustainability assessment typically determines the system boundaries for  
630 conducting the analysis. Measurements related to algal productivity would focus on the biofuel  
631 system itself, but biodiversity is usually measured in streams or terrestrial ecosystems.

632         There is significant overlap between environmental and socioeconomic sustainability  
633 indicators (Dale et al. 2013b). The overlap relates to relationships between productivity and  
634 profitability, water and air quality and human health (part of social well-being), and resource use  
635 and conservation. We have focused on environmental sustainability indicators but have  
636 sometimes discussed them in the context of socioeconomic effects. Including socioeconomic  
637 indicators in a proposed minimum set would provide a more comprehensive picture of  
638 sustainability of algal biofuels as deployed in particular contexts.

639         The proposed set of environmental sustainability indicators is a starting point for  
640 assessing sustainability of algal biofuels systems. The set of indicators will need to be modified  
641 for particular situations, and measurement protocols and interpretations of indicators must be  
642 specific to the context of the assessment (Efroymson et al. 2013). To use these indicators,  
643 sustainability goals and targets need to be defined in relation to stakeholder values and concerns  
644 for a particular algal biofuel system. Some indicators may be constrained by data availability.  
645 The next step is to use these indicators to develop appropriate management practices for algal  
646 biofuel systems.

647

648 **Acknowledgments**

649 Tanya Kuritz, Esther Parish, and Kristen Johnson provided helpful reviews of earlier  
650 drafts. We thank Matt Langholtz, Jeri Sullivan, Kitt Bagwell, Tanya Kuritz, Gary Saylor, Ed  
651 Frank, and Mark Wignosta for useful discussions. We thank Kristen Johnson and Dan Fishman  
652 of DOE for insights and project sponsorship. This research was supported by the U.S.  
653 Department of Energy (DOE) under the Bioenergy Technologies Office (BETO). Oak Ridge  
654 National Laboratory is managed by UT-Battelle, LLC, for DOE under contract DE-AC05-  
655 00OR22725.

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Table 1. Characteristics of algae and algal biofuel supply chain compared to vascular terrestrial feedstocks and their supply chains, and consequences for selection of environmental sustainability indicators

<b>Property of algal biofuel</b>	<b>Consequence for sustainability indicator</b>
No local soil resource use	Soil nutrient indicators not important
Large quantities of water used as culture media with evaporation from open ponds	Water quantity indicators important
Some algae grown in salt or brackish water	Salinity important water quality indicator; consumptive water use may be less important an indicator
CO <sub>2</sub> supplements needed	This CO <sub>2</sub> factored into greenhouse gas emissions indicator
Low slope lands required with no tilling	Sediment loading less important
Productivity of ponds susceptible to crashes	Pond crash frequency and presence or densities of responsible organisms are candidate indicators
Crop protection methods different	Indicators of chemicals other than herbicides (e.g., fungicides) may be needed

Photobioreactors (PBRs) not interacting with ecosystem

Toxins produced by algae may be occupational hazards

Breaches from natural disasters possible

Many algae cosmopolitan (broad range)

Blooms are important concern

Frequent harvesting needed because of high growth rates

Different air pollutants emitted from different production and logistics processes<sup>a</sup>

Fuels may differ in structure and manufacturing process

Variety of potential supply chains

Productivity in PBRs not ecosystem-related

Indicator (e.g., toxin) measurable/predictable at local scale

Timing of indicator measurement important

Presence of algae often not a useful indicator of invasion or biodiversity

Abundance more useful than presence as indicator of potentially invasive species

System-specific harvesting process and fate of waste important determinants of indicators

Air quality indicators tailored to supply chain

Air quality indicators custom fit to product

Practical indicators applicable to most supply chains

Commercial-scale development in the future<sup>b</sup>

Indicators should be able to be modeled

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<sup>a</sup> For example, some production processes may emit volatile organic compounds, while others may not. If biomass is dried, particulates are an important indicator, but if wet extraction is used, particulates are not an important indicator.

<sup>b</sup> This is also applicable to cellulosic feedstocks.

Table 2. Comparison of primary environmental variables differing between open and closed cultivation systems

<b>Parameter</b>	<b>Open ponds</b>	<b>Photobioreactor</b>
Land area	Higher	Lower
Water requirement	Higher	Lower
Loss of added CO <sub>2</sub>	Higher	Lower
Productivity	Lower	Higher
Cleaning of container	Not needed	Required
Contamination risk	Higher	Lower

Table 3. Characteristics that are desired for new strains of algae to be used to produce biofuels (based on Jones and Mayfield 2011, Araujo et al. 2011, NRC 2012, Gressel et al. 2013)

High photo-conversion efficiency
Rapid and stable growth
Ability to absorb light in inverse proportion to culture density
High lipid content (for biodiesel)
Easy production and high value of coproducts
High CO <sub>2</sub> -absorbing capacity
Limited nutrient requirements
Genetic stability
No detectable toxins
Ability to flourish in brackish, briny, or wastewater
Robustness toward shear stresses in photobioreactors
Competitiveness against wild native strains in open ponds
Resistance to predators, viruses, fungi in open ponds
Resistance to crop protection chemicals (algaecides, herbicides, antibiotics, antiseptics, etc.)

Tolerance to temperature variations, pH, salinity
Harvestability (e.g., sedimentation rate, self-flocculation ability)
Capability for secretion of hydrocarbons by live organisms
Extractability (influenced by cell volume, cell wall thickness, toughness)
Digestibility

Table 4. Set of 16 proposed generic environmental indicators for sustainability of algal biofuels, as derived from many national and international recommendations for sustainability indicators, criteria, and standards for bioenergy.

Category	Indicator	Units	Reference that discusses methods used to collect data
Soil quality	Bulk density	g/cm <sup>3</sup>	Doran and Jones 1996
Water quantity	Peak storm flow	L/s	Buchanan and Somers 1969
	Minimum base flow	L/s	Buchanan and Somers 1969
	Consumptive water use (incorporates base flow)	feedstock production: m <sup>3</sup> /ha/day; biorefinery: m <sup>3</sup> /day	Feedstock production: calculated from flow measurements. Biorefineries: reported total water withdrawn used as proxy.
Water quality	Nitrate concentration in streams (and export)	concentration: mg/L; export: kg/ha/yr	Rice et al. 2012
	Total phosphorus (P) concentration in streams (and export)	concentration: mg/L; export: kg/ha/yr	Rice et al. 2012
	Salinity	Conductivity (no units)	Rice et al. 2012
Greenhouse gases	CO <sub>2</sub> equivalent emissions (CO <sub>2</sub> and N <sub>2</sub> O)	kgC <sub>eq</sub> /GJ	Spreadsheet models (e.g., GREET; Frank et al. 2011a,b)
Biodiversity	Presence of taxa of special concern	Presence	Various methods exist depending on taxa selected.

	Habitat of taxa of special concern	ha	Various methods exist depending on taxa selected (e.g., Turlure et al. 2010)
	Abundance of released algae	Number/L	Initially calculated from known biomass in culture and estimated release rate or estimated using genetic markers
Air quality	Tropospheric ozone	ppb	Combination of sources and methods necessary, for example: EPA Mobile Source Observation Database, Community Multiscale Air Quality model (for example: Appel et al. 2007), reports from biorefineries, collation of vehicle use with emissions data per fuel type (for example: Gaffney and Marley 2009).
	Carbon monoxide	ppm	
	Total particulate matter less than 2.5µm diameter (PM <sub>2.5</sub> )	µg/m <sup>3</sup>	
	Total particulate matter less than 10µm diameter (PM <sub>10</sub> )	µg/m <sup>3</sup>	
Productivity	Primary productivity or yield	gC/L/year or based on chlorophyll a	Berkman and Canova 2007

Table 5. Set of ancillary environmental indicators for sustainability of algal biofuels that are applicable in particular contexts, have insufficient information, or may be applicable in the future, depending on technology development.

Category	Indicator	Units	Reference that discusses the methods used to collect data	Applicability to algal biofuels
Soil quality	Total organic carbon (TOC)	Mg/ha	Doran and Jones 1996	Applicable if digested algae are mixed with soil as a means of waste treatment
	Total nitrogen (N)	Mg/ha	Bremner and Mulvaney 1982	Applicable if digested algae are mixed with soil as a means of waste treatment
	Extractable phosphorus (P)	Mg/ha	Nelson et al. 1953	Applicable if digested algae are mixed with soil as a means of waste treatment
Water quality	Suspended sediment concentration in streams (and export)	Concentration: mg/L; export: kg/ha/yr	Rice et al. 2012	Applicable only during construction
	Herbicide concentration in streams (and export)	Concentration: mg/L; export: kg/ha/yr	Rice et al. 2012	Applicable only to herbicide-resistant strains
	Metals	Concentration; mg/L	EPA 1994	Not enough information available yet to determine if particular metals should be monitored

	Toxin concentration in cultures	Concentration; mg/L	e.g., FWR 1994	May be necessary for unfamiliar strains or if blooms of opportunistic cyanobacteria occur
	Crop protection chemicals (e.g., antibiotic, disinfectant)	Concentration, mg/L	Methods specific to chemical	Not enough information available yet to determine if particular chemicals will be used broadly
	Flocculants	Concentration, mg/L	Methods to be determined and specific to flocculant	Applicable only where flocculants are used; not enough information yet to determine if these chemicals will be used broadly or released to natural waters
Air quality	Volatile organic compounds	Concentration, g/m <sup>3</sup>	EPA 1999	More research is needed
Productivity	Pathogen densities	Number of cells or particles/L for desired species or indicator species	Methods dependent on pathogen, e.g., Brenner et al. 2010	Some pathogens may be important to measure in some cultures

Appendix 1. Expected and actual air emissions from algal biofuel production and use and evidence. NA is not applicable.

Stage of biofuel production	VOCs	aerosols	sulfate	NH <sub>3</sub>	PM2.5	PM10	NO <sub>x</sub>	CO	acetaldehyde
Open pond cultivation	Expected based on Gschwend et al. (1985), Zuo et al. (2012), Shaw et al. (2010); 45 VOCs identified (Zimba 2012, NRC 2012)	Expected to include algae, nutrients, products of reactions of SO <sub>2</sub> , NO <sub>x</sub> , NH <sub>3</sub> , VOCs (NRC 2012)	NA	NA	NA	NA	NA	NA	NA
Drying	NA	NA	NA	NA	May include fine particulates (NRC 2012)	May include coarse particulates (NRC 2012)	NA	NA	NA
Extraction	Expected, such	NA	NA	NA	NA	NA	NA	NA	NA

	as hexane or other extractants (Demirbas 2011, Lardon et al. 2009, Gong and Jiang 2011)								
Pyrolysis	NA	NA	NA	NA	Possible but not characterized (NRC 2012)	Possible but not characterized (NRC 2012)	Possible but not characterized (NRC 2012)	Possible but not characterized (NRC 2012)	NA
Anaerobic digestion	NA	NA	NA	Possible, but likely recycled	NA	NA	NA	NA	NA
Use of bioethanol	Reduced production for E85 (EPA 2002a)	NA	Reduced production for bioethanol (EPA	NA	Reduced production for bioethanol (EPA	Reduced production for bioethanol (EPA 2002a)	Reduced production for bioethanol (EPA 2002a)	Reduced production for E85 (EPA 2002a)	Higher emissions from bioethanol (EPA 2002a)

			200a2)		2002a)				
Use of biodiesel	NA	Reduced production for non-algae biodiesel (EPA 2002b)	Reduced production for non-algae biodiesel (EPA 2002b)	NA	Reduced production from blend in marine vessel (Khan et al. 2012)	Reduced emission from non-algae biodiesel (EPA 2002b)	Reduced production from blend in marine vessel (Khan et al. 2012)	Reduced production from blend in marine vessel (Khan et al. 2012)	NA

## Figure Captions

Figure 1. Stages of common algae biofuel supply chains, elements within those stages, and categories of environmental effects that often represent major effects for each element. A blank box indicates that the category is not appreciably affected by that element of the supply chain.