D.U.Quark

Volume 6
Issue 1 *Fall 2021- Spring 2022*

Article 4

3-28-2022

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Recommended Citation

Vietmeier, A. (2022). Generating the Power of Microbes How Microbial Metabolism May Solve Water and Energy Shortages. *D.U.Quark, 6* (1). Retrieved from https://dsc.duq.edu/duquark/vol6/iss1/4

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Generating the Power of Microbes How Microbial Metabolism May Solve Water and Energy Shortages

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D.U.Quark 2022. Volume 6(Issue 1) pgs. 19-28
Published March 28, 2022
Staff Article

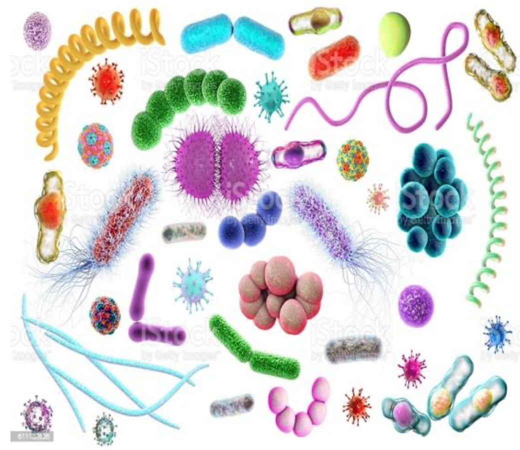
On ancient Earth, water was the most likely place for life to begin, and water is, arguably, one of the most critical and undervalued resources we have [1, 2]. For life on Earth, water is essential, as it makes up 60% of our bodies as the main component of cells, tissues, and organs [3]. Freshwater resources include lakes, rivers, and groundwater [4]. Unfortunately, in the twenty-first century we are facing issues that stand in the way of fresh water *quantity* and *quality.* More than a third of the world's population does not have access to safe drinking water, which is further exacerbated by the waste and uneven distribution of water resources [1, 4]. One way that water can be wasted is through inefficient irrigation of crops and leaky water pipes [1]. Water quality is impacted by increasing pollution due to growing urban populations and industrial waste discharge [1].

Chemical water pollutants can be macropollutants (measured at milligrams per liter), or micropollutants (measured at nanogram or microgram per liter concentrations) [4]. Macro scale water pollutants can be added via human activities such as crop fertilization and irrigation and road maintenance in snowy conditions. When fertilizer macropollutants, such as nitrogen and phosphorus, run off into the watershed, they increase the nutrient concentrations in water, which leads to the production of biomass, depletion of oxygen, and toxic algal bloom formations [4]. The

use of road salts for road maintenance and excessive irrigation of crops lead to an increase in salt concentrations in water, which inhibits crop growth and makes the water non-drinkable [4].

Due to the low concentration of micropollutants, it can be difficult to assess their effect as their toxic impacts are typically subtle chronic effects and not acute [4]. Inorganic pollutants in water include heavy metals, which are especially troubling as they naturally cycle through different chemical oxidation states that are subsequently associated with different toxicity levels [4].

In addition to the pollution of drinking water, we are also experiencing a global energy crisis [5]. Fossil fuels, including oil, gas, coal, and uranium, are projected to be nearly depleted in 55-75 years [5]. The overuse of fossil fuels has also propagated other environmental concerns such as CO_2 emission and thermal pollution, both of which increase the rate of climate change [5]. There is a desperate need for alternative fuel sources as fossil fuel concentrations decrease, global demand for energy increases, and the impacts of human driven climate change becomes more evident [6]. The replacement of fossil fuels with other energy sources can counter greenhouse gas production that is the leading cause of climate change [6]. Alternative greener sources of energy production include solar power, hydropower, and wind power. According to new scientific research, microbes may be another source of green energy.



Microbes are microscopic organisms that can be found just about everywhere. Microbes can include fungi, algae, and bacteria. Microbes adapt to their environment, and in doing so, have evolved a multitude of ways to generate energy to drive their metabolism: a multitude of microbial products and byproducts. Microbes are capable of generating energy, treating contaminated waters, and producing products due to their electron transport and carbon metabolism capabilities [7]. Microbial biomass and microbial biological waste can be used as renewable energy/fuels [6].

TABLE 1 | List of microorganisms producing biofuels or the precursors for biofuel production.

Microorganism	Biofuel	Biofuel yield ($g\ L^{-1}$)	References
Clostridium acetobutylicum	Butanol	3	Lütke-Eversloh and Bahl, 2011
Clostridium thermocellum	Isobutanol	5.4	Lin et al., 2015
Escherichia coli	Butanol	30	Shen et al., 2011
Escherichia coli	Ethanol	25	Romero-García et al., 2016
Saccharomyces cerevisiae	Fatty acids	0.38	Yu et al., 2016
Saccharomyces cerevisiae	Isoprenoid based-biofuel	40	Westfall et al., 2012
Pseudomonas putida	Butanol	0.05	Nielsen et al., 2009
Cryptococcus vishniaccii	Lipids	7.8	Deeba et al., 2016
Zymomonas mobilis	2, 3-butanediol	10	Yang et al., 2016
Zymomonas mobilis	Ethanol	–	Kremer et al., 2015
Caldicellulosiruptor bescii	Ethanol	0.70	Chung et al., 2014
Trichoderma reesei	Ethanol	10	Huang et al., 2014
Yarrowia lipolytica	Fatty acids	55	Beopoulos et al., 2009
Synechococcus sp.	Limonene	0.04	Davies et al., 2014
Synechococcus elongates	1, 3-propanediol	0.28	Hirokawa et al., 2016

Microbial Biofuels

Due to the metabolic diversity of microbes, they are capable of producing biofuels from a variety of substrates or reactants [6]. Biofuel may be a sustainable carbon-neutral energy source that would be compatible with current engine technology [8]. A wide range of bacteria have been found capable of creating a range of biofuels (**Figure 1**) [7]. For instance, bacteria can easily convert sugars into ethanol. Cyanobacteria and microalgae can photosynthetically reduce atmospheric CO_2 into biofuels, while methanotrophs can use methane to produce methanol [6]. Microbial production of ethanol has been reviewed extensively, with the most commonly used organism being *Saccharomyces cerevisiae*, *Zymomonas mobilis*, and *Escherichia coli* [8]. Microbial pathways have been found that can generate molecules that are similar or identical to those found in gasoline [8]. Use of lignocellulosic biomass (wood, agricultural, or forestry wastes) to generate biofuel via microbes involves the conversion of monosaccharides to target molecules [8]. With increased research in regards to the microbial physiology and pathways involved in producing biofuels, microbial engineering can be used to advance the uses of microbes to generate biofuels [8].

Microbial Energy Production

Bioelectrochemical cells, cells that produce a chemical electrical gradient, have gained interest for their applications in bioenergy, bioremediation, and bioelectronics

[9]. Bioelectrochemical cells generate bioenergy from organic biomass in wastewaters [6]. Metabolic potential of microbes through electron transfer in microbial metabolism has a direct relationship to redox reactions that are used to remove contaminants from solution and generate electricity [7]. Microbes have the ability to respire (breathe) metal by using the electrons from environmental metals to power their electron transport chain for energy production [7]. Bacteria that are capable of carrying electrons beyond their cell walls are capable of interacting electrically with their environment [7]. Electrons can either be removed or applied from an external circuit or source [7]. Electrons that move beyond a cell wall can be transferred via soluble mediators or with solid surfaces [7]. To aid the movement of electrons out of the microbial cell and into the environment, microbes have evolved pili, also known as nanowires,with metallic-like conductivity [9]. Nanowires can be used for centimeters long range electron transport, which on a microbial scale is a large distance [9, 10]. Nanowires can also move electrons to and from other species through interspecies electron transfer and insoluble electron acceptors [9]. Microbial fuel cells are devices that generate electricity from microbial catabolism that utilize organic compounds [11]. Microbial fuel cells use integrated anode and cathode chambers [11]. Electrons generated during microbial oxidation of organic compounds are transferred

to the anode of the microbial fuel cell, thereby generating an electrical current and energy (**Figure 2**) [11].

The remediation for contaminated drinking water may help in releasing some of the burden associated with the energy crisis. Several emerging microbial electrochemical technologies utilize interactions between electrodes and bacteria for the production of energy, treatment of waste and wastewater, bioremediation, and the production of valuable products [7]. Microbial fuel cells are an alternative energy source that have an excellent ability to produce electricity while removing pollutants [5]. Microbial fuel cells are promising for sustainable energy production and simultaneous removal of heavy metals from soil and water [5]. Bioremediation is one of the most favored sustainable means of treating contaminated sites, as it is renewable and cost-effective to treat contaminated soils and sediments [10].

electrode reaction using acetate as a substrate [6] and a two-chamber MFC setup (Figure 1) are shown below:

$$\text{Anodic reaction: } CH_3COO^- + 2H_2O \xrightarrow{\text{microbes}} 2CO_2 + 7H^+ + 8e^- \quad (1)$$

$$\text{Cathodic reaction: } O_2 + 4e^- + 4H^+ \rightarrow 2H_2O. \quad (2)$$

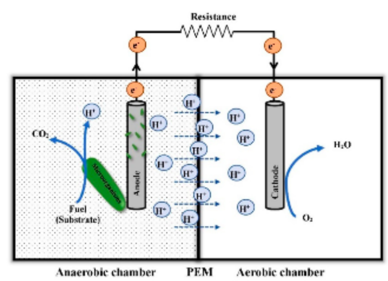


Figure 1. Schematic of a typical two-chamber microbial fuel cell.

One of the challenges for producing biofuels is using microbial factories to generate a large amount of fuel on a comparatively low budget with a greater efficiency as compared to conventional fossil fuels [6]. We have a current need for microbial fuel cells that can be scaled to support widespread and cost-effective applications and a better understanding of the physical properties of potential biofuel molecules [8].

Some biofuel properties still need a better understanding of their sustainability and the purification process [8]. Continued research on metabolic engineering for the production of bulk chemicals and biofuels can hopefully aid in the large-scale use of biofuels and bioenergy in the future [8].

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Microbes

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Biofuel

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Bioenergy

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