

CLAIMS

What is claimed is:

1. A bioremediation system for wastewater treatment comprising:
 - 5 a system wastewater inlet, a system wastewater outlet, an anaerobic reaction vessel, and an aerobic reaction vessel connected by at least one wastewater flow path;
wherein the anaerobic reaction vessel contains an anaerobic reaction solution,
wherein the anaerobic reaction solution contains an anaerobic concentration of at least one anaerobic microorganism in an anaerobic wastewater, wherein the at least one anaerobic
10 microorganism is capable of reducing an anaerobic concentration of metal in the anaerobic wastewater, increasing an anaerobic pH of the anaerobic wastewater, or reducing an anaerobic concentration of organic compounds in the anaerobic wastewater, or a combination thereof; and
wherein the aerobic reaction vessel contains an aerobic reaction solution, wherein the aerobic reaction solution contains an aerobic concentration of at least one aerobic microorganism
15 in an aerobic wastewater, wherein the at least one aerobic microorganism contains a metalloprotein, or is capable of reducing an aerobic concentration of CO₂, or is capable of decreasing an aerobic concentration of metal from the aerobic wastewater, or a combination thereof.
- 20 2. The system of claim 1, further comprising an electrochemical reaction vessel, wherein the electrochemical reaction vessel includes an electrochemical reaction solution, an electrochemical wastewater inlet, an electrochemical wastewater outlet, an anode, a cathode, a power source, and a carbon dioxide source,
wherein the electrochemical reaction vessel is connected by the at least one wastewater
25 flow path to the anaerobic reaction vessel, the aerobic reaction vessel, or a combination thereof.
3. The system of claim 1, wherein the anaerobic reaction vessel further includes an anaerobic wastewater inlet, an anaerobic wastewater outlet, an anaerobic nutrient inlet, and an anaerobic biomass outlet;
30 wherein the aerobic reaction vessel further includes an aerobic wastewater inlet, an aerobic wastewater outlet, an aerobic nutrient inlet, and an aerobic biomass outlet;
wherein the at least one wastewater fluid path connects the anaerobic wastewater inlet to the aerobic wastewater outlet, or the at least one wastewater fluid path connects the aerobic wastewater inlet to the anaerobic wastewater outlet.

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4. The system of claim 1, further comprising an aerobic biomass fluid flow path connecting the aerobic reaction vessel and the anaerobic reaction vessel.
5. The system of claim 2, wherein the carbon dioxide source includes a carbon dioxide inlet, or wherein the power source includes sunlight, a solar power source, an electrical power source, or a combination thereof.
6. The system of claim 2, wherein the power source is configured to provide a voltage of from about 0.7 Volts to about 10 Volts or a current of from about 60 mA to about 100 mA.
7. The system of claim 2, wherein the electrochemical reaction vessel, the anaerobic reaction vessel, the aerobic reaction vessel, or any combination thereof, has a volume of from about 1000 liters to about 1 million liters; or
wherein the anaerobic concentration ranges from about 10^7 to about 10^9 cells per milliliter, or an optical density of about 0.6 to about 1.0; or
wherein the aerobic concentration ranges from about 10^7 to about 10^9 cells per milliliter, or an optical density of about 0.6 to about 1.0;
wherein the electrochemical reaction vessel contains an electrochemical reaction buffer, wherein the electrochemical reaction buffer includes an amount of sodium chloride, sodium hydroxide, sodium carbonate, calcium carbonate, potassium carbonate, potassium chloride, potassium hydroxide, magnesium chloride, or a combination thereof; or
wherein the electrochemical reaction vessel, the anaerobic reaction vessel, and the aerobic reaction vessel each independently includes a vessel wall, wherein the vessel wall includes a cement material, a fiberglass material, a fiber material, a steel material, a natural formation, a plastic material, a gel material, or a combination thereof.
8. The system of claim 1, wherein the at least one anaerobic microorganism is capable of reducing a concentration of sulfate and increasing a concentration of sulfide in the anaerobic wastewater; or wherein the at least one anaerobic microorganism is capable of oxidizing the anaerobic concentration of organic compounds in the anaerobic wastewater; or wherein the at least one anaerobic microorganism is capable of producing sulfide; or wherein the at least one aerobic microorganism includes at least one recombinant aerobic microorganism that expresses at least one protein of at least one aerobic genetic pathway, wherein the at least one aerobic genetic pathway provides to the at least one aerobic microorganism one or more of a resistance to a high concentration of heavy metals, a resistance to a low pH level, a production of one or

more of an alkaline organic molecule, sodium bicarbonate, ammonia, acetate, and an electron donor; and combinations thereof.

9. The system of claim 1, wherein the at least one anaerobic microorganism includes a
 5 microorganism selected from the group consisting of *Desulfovibrio desulfuricans*, *Desulfovibrio vulgaris*, *Desulfovibrio gigas*, *Caldivirga maquilingensis*, *Desulfatibacillum alkenivorans*, *Desulfotomaculum nigrificans*, *Desulfococcus multivorans*, *Thermodesulfovibrio yellowstonii*, *Desulfovibrio aespoeensis*, *Desulfovibrio aerotolerans*, *Desulfovibrio fructosivorans*, *Desulfococcus oleovorans*, *Desulfovibrio aminophilus*, *Desulfovibrio ferrireducens*,
 10 *Desulfovibrio salexigens*, *Desulfovibrio africanus*, *Archaeoglobus fulgidus*, *Desulfococcus biacutus*, *Desulfatibacillum aliphaticivorans*, *Desulfatirhabdium butyrativorans*, *Desulfofaba gelida*, *Desulfovibrio capillatus*, *Desulfosporomusa* spp., *Desulfovibrio acrylicus*, *Desulfovibrio legallii*, *Desulfosarcina alkanivorans*, *Desulfatiferula berrensis*, *Desulfobacula toluolica*, *Desulfovibrio frigidus*, *Desulfofaba hansenii*, *Desulfovibrio senezii*, *Desulfovibrio arcticus*,
 15 *Thermodesulfobium narugense*, *Desulfovibrio burkinensis*, *Thermodesulfobium acidiphilum*, *Thermocladium modestius*, *Caldimicrobium rimae*, *Desulfosarcina ovata*, *Thermodesulfobacterium hveragerdense*, *Desulfovibrio alcoholivorans*, *Desulfovibrio singaporenus*, *Caldimicrobium thiodismutans*, *Desulfosporosinus orientis*, *Desulfovibrio marinus*, *Desulfatitalea tepidiphila*, *Desulfofaba fastidiosa*, *Desulfovibrio ferrophilus*,
 20 *Desulfovibrio bizertensis*, *Desulfovibrio biadensis*, *Desulfatiferula olefinivorans*, *Desulfobacula phenolica*, *Dehalococcoides* spp., *Geobacteria* spp., *Acetobacterium woodi*, *Clostridium ljungdahlii*, *Moorella thermoacethica*, and methanotrophic bacteria, or a combination thereof.

10. The system of claim 8, wherein the at least one aerobic microorganism includes a
 25 microorganism selected from the group consisting of an acidophilic microorganism, *Galdieria Sulphuraria*, a bicarbonate producing species, an alkaline producing species, *Spirulina platensis*, a Cyanobacteria, *Synechococcus elongatus*, *Synechocystis* spp., and a recombinant aerobic microorganism that expresses a higher amount of at least one metalloprotein relative to a control aerobic microorganism.

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11. The system of claim 1, wherein the anaerobic reaction vessel and the aerobic reaction vessel include a sample port, an optical density reader, a turbidity reader, or a combination thereof.

12. The system of claim 2, wherein the electrochemical reaction vessel, the anaerobic reaction vessel, or the aerobic reaction vessel, or any combination thereof, is mounted on or among one or more vehicles; or
- 5 wherein the electrochemical reaction vessel, the anaerobic reaction vessel, or the aerobic reaction vessel, or any combination thereof, is located within 0.3 kilometers of an industrial site and connected to the industrial site by a gas flow path from the industrial site to the carbon dioxide source, or a wastewater flow path from the industrial site to the system wastewater inlet, or a combination thereof; or
- 10 wherein the aerobic reaction vessel further comprises a gas flow path connected to the carbon dioxide source, a gas flow path connected to an air source, or a combination thereof.
13. The system of claim 2, further comprising a pH meter connected to at least one of the system wastewater inlet, the system wastewater outlet, the electrochemical wastewater inlet, the electrochemical wastewater outlet, the aerobic wastewater inlet, or the aerobic wastewater inlet, or a combination thereof; or
- 15 further comprising a heavy metal detector connected to at least one of the system wastewater inlet, the system wastewater outlet, the electrochemical wastewater outlet, the aerobic wastewater inlet, or the aerobic wastewater inlet, or a combination thereof.
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14. A method of treating wastewater comprising:
- providing a bioremediation system, wherein the bioremediation system includes:
- 25 a system wastewater inlet, a system wastewater outlet, an anaerobic reaction vessel, and an aerobic reaction vessel connected by at least one wastewater flow path;
- wherein the anaerobic reaction vessel contains an anaerobic reaction solution,
- 30 wherein the anaerobic reaction solution contains an anaerobic concentration of at least one anaerobic microorganism in an anaerobic wastewater, wherein the at least one anaerobic microorganism is capable of reducing an anaerobic concentration of metal in the anaerobic wastewater, increasing an anaerobic pH of the anaerobic wastewater, or reducing an anaerobic concentration of organic compounds in the anaerobic wastewater, or a combination thereof; and

wherein the aerobic reaction vessel contains an aerobic reaction solution,
wherein the aerobic reaction solution contains an aerobic concentration of at least
one aerobic microorganism in an aerobic wastewater, wherein the at least one
aerobic microorganism contains a metalloprotein, or is capable of reducing an
aerobic concentration of CO₂, or is capable of decreasing an aerobic
concentration of metal from the aerobic wastewater, or a combination thereof;
provided the wastewater entering the anaerobic reaction vessel includes one or more
heavy metals, producing a sulfide by reacting the at least one anaerobic microorganism with a
sulfate compound, and forming at least one metal sulfide compound by reacting one or more of
the heavy metals with the sulfide; and
provided the wastewater entering the aerobic reaction vessel includes one or more heavy
metals, reacting the one or more heavy metals with the at least one aerobic microorganism.

15. The method of claim 14, further including:
measuring a pH level of wastewater entering the system wastewater inlet; and
provided that the pH level ranges from about 4 or lower, flowing the wastewater from the
system inlet into an electrochemical reaction vessel, wherein the electrochemical reaction vessel
includes an electrochemical reaction solution, an electrochemical wastewater inlet, an
electrochemical wastewater outlet, an anode, a cathode, a power source, and a carbon dioxide
source, and the electrochemical reaction solution contains sodium chloride, sodium hydroxide,
and sodium carbonate; and
raising the pH level of the electrochemical reaction solution by forming sodium
hydroxide and sodium bicarbonate; or
provided that the pH level ranges from about 4.1 to 8.0, flowing wastewater from the
system inlet to the anaerobic reaction vessel, the aerobic reaction vessel, or any combination
thereof.

16. The method of claim 14, further comprising forming a biomass in the aerobic reaction
vessel, and feeding an amount of the biomass into the anaerobic reaction vessel; or further
comprising collecting an amount of biomass from the aerobic reaction vessel; or wherein the
anaerobic reaction vessel includes a nutrient inlet, adding an amount of carbon dioxide, an
amount of methane, or a combination thereof through the nutrient inlet.

17. The method of claim 14, further including,

- feeding the wastewater into the anaerobic reaction vessel at a flow rate of from about 50 liters/hour to about 150 liters/hour or more; or
- flowing the wastewater through the anaerobic reaction vessel for a residence time of from about 2 hours to about 9 hours; or
- 5 feeding an anaerobic reaction vessel effluent into the aerobic reaction vessel at a flow rate of from about 50 liters/hour to about 150 liters/hour or more; or
- flowing the wastewater through the aerobic reaction vessel for a residence time of from about 2 hours to about 9 hours; or
- 10 provided the anaerobic wastewater in the anaerobic reaction vessel includes an anaerobic concentration one or more heavy metals, reducing the concentration of the one or more heavy metals in the wastewater by from about 1% to about 30% or more, based on the anaerobic concentration; or
- provided the aerobic wastewater in the aerobic reaction vessel includes an aerobic concentration one or more heavy metals, reducing the concentration of the one or more heavy
- 15 metals in the wastewater by from about 1% to about 30% or more, based on the aerobic concentration.
18. The method of claim 14, provided that wastewater leaving the aerobic reaction vessel, the anaerobic reaction vessel, or the system wastewater outlet has a heavy metal content of 0.1 mg/l
- 20 or more,
- flowing the wastewater to the aerobic reaction vessel, the anaerobic reaction vessel, or any combination thereof.
19. The method of claim 14, wherein the one or more heavy metals includes aluminum, iron,
- 25 zinc, copper, lead, nickel, cadmium, chromium, titanium, vanadium, manganese, cobalt, gallium, germanium, arsenic, zirconium, niobium, and combinations thereof.
20. The method of claim 15, including forming sodium hydroxide and sodium bicarbonate by applying from about 0.7 Volts to about 10 Volts or a current of from about 60 mA to about 100
- 30 mA across the anode and the cathode in the presence of sodium chloride and carbon dioxide.