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(54) Title: CLAY MINERALS AND THEIR AMENDMENTS TO ACCELERATE SEQUESTRATION OF CARBON FROM OCEAN SURFACE

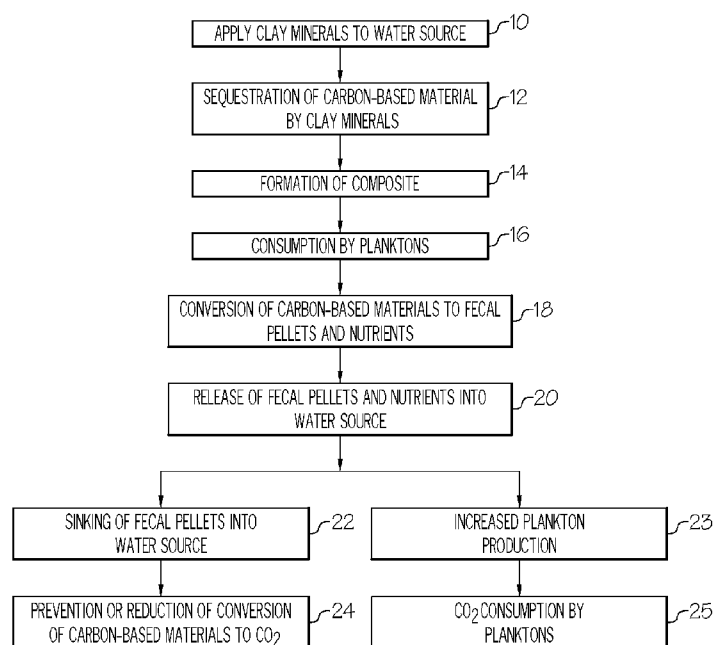


FIG. 1

(57) Abstract: Embodiments of the present disclosure pertain to methods of sequestering carbon-based materials from a water source by applying clay minerals to the water source. The sequestered carbon-based material may include carbon dioxide, organic materials, or combinations thereof. Additional embodiments of the present disclosure pertain to compositions that include clay minerals for sequestering carbon-based materials from a water source. The clay minerals may include isolated clay minerals, purified clay minerals, modified clay minerals, or combinations thereof.

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TITLE**CLAY MINERALS AND THEIR AMENDMENTS TO ACCELERATE SEQUESTRATION
OF CARBON FROM OCEAN SURFACE****CROSS-REFERENCE TO RELATED APPLICATIONS**

[0001] The present application claims the benefit of U.S. Provisional Patent Application No. 63/322,174, filed on March 21, 2022, and U.S. Provisional Patent Application No. 63/398,130, filed on August 15, 2022. The entirety of each of the aforementioned applications is incorporated herein by reference.

BACKGROUND

[0002] The United Nations Framework Convention on Climate Change requires accelerated transition to a low carbon economy, large-scale implementation of carbon capture and sequestration, and massive deployment of atmospheric CO₂ removal technologies. However, harnessing of energy from oil, gas, and coal contributes to about 88% of atmospheric CO₂. Additionally, a transition to a low carbon economy remains in the distant future. Furthermore, at present, there is no technology available to rapidly and cheaply remove atmospheric CO₂. Numerous embodiments of the present disclosure address the aforementioned limitations.

SUMMARY

[0003] In some embodiments, the present disclosure pertains to methods of sequestering one or more carbon-based materials from a water source by applying one or more clay minerals to the water source. In some embodiments, the clay mineral sequesters the carbon-based material from the water source. In some embodiments, the sequestered carbon-based material includes, without limitation, carbon dioxide, one or more organic materials, and combinations thereof. In some embodiments, the sequestered carbon-based material includes carbon dioxide, such as carbon dioxide dissolved in the water source.

[0004] Additional embodiments of the present disclosure pertain to compositions that include one or more clay minerals for sequestering carbon-based materials from a water source. In some embodiments, the clay mineral includes isolated clay minerals, purified clay minerals, or combinations thereof. In some embodiments, the clay mineral includes modified clay minerals. In some

embodiments, the modified clay minerals include one or more additional components, such as phosphorous, aluminum, iron, phyllosilicates, magnesium, alkali metals, alkaline earths, apatite, wavellite, crandallite, dolomite, quartz, or combinations thereof.

[0005] In some embodiments, the compositions of the present disclosure are in the form of a composite. In some embodiments, the composite includes one or more sequestered carbon-based materials and clay minerals.

BRIEF DESCRIPTION OF THE DRAWINGS

[0006] **FIG. 1** illustrates a method of sequestering carbon-based materials in accordance with the methods of the present disclosure.

[0007] **FIGS. 2A-2D** illustrate an experimental setup to evaluate the interaction of clay minerals with carbohydrates (i.e., acid polysaccharides, typically exuded by phytoplankton) and a common bacterium (i.e., *Alteromonas spp.*, a bacterium involved in decomposing particulate organic materials). The different funnels contain sea water, acid polysaccharides, and clay (**FIG. 2A**); sea water, acid polysaccharides, clay, and *Alteromonas spp.* (**FIGS. 2B and 2C**); and sea water, clay, and *Alteromonas spp.* (**FIG. 2D**).

[0008] **FIG. 3** shows that Zeta potential is a measure of the electrostatic interaction between organic molecules dissolved in seawater and clay minerals.

[0009] **FIG. 4A** shows that the concentration of *Alteromonas* in the funnels illustrated in **FIGS. 2A-2D** (i.e., funnels 1-4, respectively) does not change when clay is added.

[0010] **FIG. 4B** shows that *Prochlorococcus* needs ammonium ion to grow, and that the growth shows no impact on the picoplankton when ammonia is added in the growth medium (i.e., fourth set of bars—the dark bar).

[0011] **FIG. 5** shows a carbon sequestration experiment with a mixture of palygorskite and nontronite amended with apatite (a mineral to supply phosphorus—a key nutrient needed in the ocean).

[0012] **FIG. 6** summarizes experimental results demonstrating that clay minerals induce production of Transparent Exopolymer Particles (TEPs) in natural cultures of a diatom (*Thalassiosira weissflogii*).

[0013] **FIG. 7** shows that *Thalassiosira weissflogii* grows, consumes dissolved silica from seawater, and produces TEP in the presence of clay.

[0014] **FIG. 8** illustrates a proposed mechanism for removing atmospheric CO₂ by clay minerals.

DETAILED DESCRIPTION

[0015] It is to be understood that both the foregoing general description and the following detailed description are illustrative and explanatory, and are not restrictive of the subject matter, as claimed. In this application, the use of the singular includes the plural, the word “a” or “an” means “at least one”, and the use of “or” means “and/or”, unless specifically stated otherwise. Furthermore, the use of the term “including”, as well as other forms, such as “includes” and “included”, is not limiting. Also, terms such as “element” or “component” encompass both elements or components comprising one unit and elements or components that include more than one unit unless specifically stated otherwise.

[0016] The section headings used herein are for organizational purposes and are not to be construed as limiting the subject matter described. All documents, or portions of documents, cited in this application, including, but not limited to, patents, patent applications, articles, books, and treatises, are hereby expressly incorporated herein by reference in their entirety for any purpose. In the event that one or more of the incorporated literature and similar materials defines a term in a manner that contradicts the definition of that term in this application, this application controls.

[0017] The United Nations Framework Convention on Climate Change requires accelerated transition to a low carbon economy, large-scale implementation of carbon capture and sequestration, and massive deployment of atmospheric CO₂ removal technologies. However, harnessing of energy from oil, gas, and coal contributes to about 88% of atmospheric CO₂ and a transition to low carbon economy remains in the distant future. The rate of increase of atmospheric C is at 5.1 Pg yr⁻¹ (1 Pg C = 10¹⁵ g C or a billion tons of C; 1 Pg C = 3.664 Pg CO₂) and at present there is no technology available to rapidly and cheaply remove atmospheric CO₂.

[0018] For over two decades, Ocean Iron fertilization (OIF) has been thought to provide a solution that could potentially remove >1 Pg of atmospheric C per year by increasing the phytoplankton biomass (primary productivity) of the ocean. The OIF is based on the observation that while excess nutrients (nitrate, phosphate) are perennially available in a third of the euphotic zone of the ocean, the phytoplankton biomass is low due to a lack of iron—an atmospheric dust delivered essential micronutrient required for the synthesis of chlorophyll. For example, the present-day Southern Ocean

is one such high nutrient low chlorophyll (HNLC) regions that receives little atmospheric dust. Phytoplankton, in particular diatoms, have C: Fe ratios between ~23,000 and ~160,000 or more. Thus, addition of relatively small amounts of iron to HNLC regions would lead to a large sequestration of carbon.

[0019] For instance, the Southern Ocean received ~5 times more iron during the last glacial maximum 20,000 years ago than present-day and likely contributed to removing ~85 Pg C (~50% of the total removed) from the atmosphere. The OIF involves enriching ocean surface with water soluble ferrous iron (Fe^{2+}) and has been shown to induce phytoplankton blooms and atmospheric CO_2 drawdown. If the organic carbon thus produced is transported to a depth of >1000 m, it is effectively removed from exchanging with the atmosphere.

[0020] However, ocean iron fertilization campaigns have revealed the following problems: 1) inefficiency: a) rapid reduction in iron concentration in the euphotic zone following fertilization; b) a majority of particulate organic matter (POM, mainly decaying organisms and excrement) produced in the euphotic zone is re-oxidized to CO_2 by bacteria as it falls through the seawater column; 2) collateral production of N_2O : bacteria also utilize iron in nitrification/denitrification reactions releasing N_2O —a potent greenhouse gas that could exacerbate global warming; 3) possible changes in ecology and marine biogeochemical cycles; and 4) depletion of nutrients. These issues combined with a price-tag that could be as high as \$456 to remove one ton of C have made large-scale ocean iron fertilization quite unappealing.

[0021] Marine biological pumps remove 21.4 ± 5.3 Pg and 9.6 ± 3.6 Pg of atmospheric C yr^{-1} in the form of dissolved organic matter (DOM, carbohydrates, proteins, lipids, other organic molecules) and particulate organic matter (POM, mainly decaying organisms and excrement), respectively. Microbes oxidize over 90% of DOM to CO_2 in the euphotic zone. Similarly, more than 90% of the sinking POM is oxidized to CO_2 by marine biota as it falls through the upper 1000m.

[0022] As such, more effective methods and compositions are required for capturing carbon-based materials from water sources (e.g., capture from and below the marine euphotic zone and sequestering the carbon-based materials at depth). Numerous embodiments of the present disclosure aim to address the aforementioned need through the utilization of clay minerals, which have previously demonstrated effective CO_2 adsorption performance (e.g., Tao et al., Renewable and Sustainable Energy Reviews, Volume 164, 2022, 112536, ISSN 1364-0321).

[0023] Methods of Sequestering Carbon-based Materials

[0024] In some embodiments, the present disclosure pertains to a method of sequestering one or more carbon-based materials by applying one or more clay minerals to a water source. Thereafter, the applied clay minerals sequester the carbon-based materials.

[0025] Carbon-based materials

[0026] The methods of the present disclosure may sequester various types of carbon-based materials. For instance, in some embodiments, the one or more carbon-based materials includes, without limitation, carbon dioxide, one or more organic materials, and combinations thereof.

[0027] In some embodiments, the one or more carbon-based materials includes one or more organic materials. In some embodiments, the one or more organic materials includes, without limitation, carbohydrates, proteins, lipids, polysaccharides, acidic polysaccharides, organic molecules, polymers, exopolymers, hydrocarbons, dissolved organic matter (DOM), particulate organic matter (POM), and combinations thereof.

[0028] In some embodiments, the one or more organic materials includes dissolved organic matter (DOM). DOM generally refers to organic materials that passes through a filter with a pore size of at least 0.22 micrometers. In some embodiments, DOM refers to organic materials that pass through a filter with a pore size of about 0.22 micrometers. In some embodiments, DOM refers to organic materials that pass through a filter with a pore size between 0.22 micrometers and 0.7 micrometers.

[0029] In some embodiments, the one or more organic materials includes particulate organic matter (POM). POM generally refers to organic materials that do not pass through a filter pore size that typically ranges in size from 0.053 to 2 millimeters.

[0030] In some embodiments, the one or more carbon-based materials includes carbon dioxide. In some embodiments, the carbon dioxide includes atmospheric carbon dioxide. In some embodiments, the carbon dioxide includes carbon dioxide dissolved in the water source. In some embodiments, the carbon dioxide includes atmospheric carbon dioxide and carbon dioxide dissolved in the water source.

[0031] Sequestering of carbon-based materials

[0032] The clay minerals of the present disclosure may sequester carbon-based materials through various mechanisms. For instance, in some embodiments, the sequestering occurs by a method that includes, without limitation, adhesion, binding, adsorption, electrostatic interaction, and combinations thereof.

[0033] Application of clay minerals to water sources

[0034] The clay minerals of the present disclosure may be applied to water sources in various manners. For instance, in some embodiments, the application occurs by a method that includes, without limitation, spreading, pouring, sprinkling, spraying, and combinations thereof. In some embodiments, the application occurs by a mechanical mechanism, a manual mechanism, a physical mechanism, a non-natural mechanism, or combinations thereof. In some embodiments, the application occurs by a non-natural mechanism. In some embodiments, the non-natural mechanism includes human intervention.

[0035] Various amounts of clay minerals may be applied to water sources. For instance, in some embodiments, clay minerals may be applied to water sources at amounts sufficient to remove approximately 1 billion tons of carbon-based materials per year from a water source (e.g., an ocean). In some embodiments, clay minerals may be applied to a water source at amounts that range from about 0.01 ton of clay minerals per acre of water source per year to about 1 ton of clay minerals per acre of water source per year. In some embodiments, clay minerals may be applied to a water source at amounts that range from about 0.01 ton of clay minerals per acre of water source per year to about 0.1 ton of clay minerals per acre of water source per year. In some embodiments, clay minerals may be applied to a water source at amounts of at least about 0.01 ton of clay minerals per acre of water source per year. In some embodiments, clay minerals may be applied to a water source at amounts of at least about 0.02 ton of clay minerals per acre of water source per year. In some embodiments, clay minerals may be applied to a water source at amounts of at least about 0.03 ton of clay minerals per acre of water source per year. In some embodiments, clay minerals may be applied to a water source at amounts of at least about 0.04 ton of clay minerals per acre of water source per year.

[0036] Clay minerals

[0037] The methods of the present disclosure may utilize various clay minerals to sequester carbon-based materials. For instance, in some embodiments, the one or more clay minerals includes isolated clay minerals. In some embodiments, the one or more clay minerals includes purified clay minerals. In some embodiments, the one or more clay minerals includes, without limitation, palygorskite, nontronite, illite-smectite, kaolinite, chlorite, and combinations thereof.

[0038] In some embodiments, the one or more clay minerals includes palygorskite. In some embodiments, the one or more clay minerals includes nontronite.

[0039] In some embodiments, the one or more clay minerals include at least a first clay mineral and a second clay mineral. In some embodiments, the first and second clay minerals are present at different weight ratios. For instance, in some embodiments, the first and second clay minerals are present at a weight ratio of 2:1, 3:1, 4:1, 5:1, 6:1, 7:1, 8:1, 9:1, or 10:1.

[0040] In some embodiments, the first and second clay minerals include palygorskite and nontronite. In some embodiments, the palygorskite and nontronite are present at a weight ratio of 4:1.

[0041] In some embodiments, the one or more clay minerals includes modified clay minerals. In some embodiments, the modified clay minerals include by-products or waste products of mining. In some embodiments, the modified clay minerals include clay minerals that have undergone a beneficiation process. In some embodiments, the modified clay minerals that have undergone a beneficiation process contain a lower amount of gangue minerals when compared to clay minerals that have not undergone the beneficiation process. In some embodiments, the gangue minerals include phosphate from phosphate rock. In some embodiments, the modified clay minerals include clay minerals that contain a lower amount of phosphate when compared to unmodified clay minerals.

[0042] In some embodiments, the modified clay minerals include one or more additional components. In some embodiments, the one or more additional components includes, without limitation, phosphorous, aluminum, iron, phyllosilicates, magnesium, alkali metals, alkaline earths, apatite, wavellite, crandallite, dolomite, quartz, or combinations thereof.

[0043] In some embodiments, the one or more additional components includes apatite, wavellite, crandallite, dolomite, quartz, or combinations thereof. In some embodiments, the one or more additional components includes apatite.

[0044] The clay minerals of the present disclosure can have various sizes. For instance, in some embodiments, the clay minerals of the present disclosure have sizes that range from about 0.1 μm to about 10 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes that range from about 0.2 μm to about 5 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes that range from about 0.2 μm to about 1 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 5 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 4 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 3 μm in diameter. In some embodiments, the clay minerals of the present disclosure

have sizes of less than about 2 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 1 μm in diameter.

[0045] Water sources

[0046] The methods of the present disclosure may sequester carbon-based materials from various water sources. For instance, in some embodiments, the water source includes, without limitation, a body of water, an ocean, a sea, a lake, a pond, or combinations thereof.

[0047] In some embodiments, the water source includes a sea. In some embodiments, the water source includes an ocean. In some embodiments, the sequestering of carbon (e.g., carbon removal) occurs in the euphotic zone of the ocean. In some embodiments, the sequestering occurs below the euphotic zone of the ocean.

[0048] Effects of sequestration

[0049] The methods of the present disclosure can have various advantageous effects. Such effects are illustrated in **FIG. 1** and described in more detail herein. For instance, in some embodiments, the clay minerals of the present disclosure can be utilized to sequester carbon dioxide, such as carbon dioxide dissolved in the water source. In some embodiments, the methods of the present disclosure can be utilized to prevent or reduce oxidation of the sequestered carbon-based materials to carbon dioxide. In some embodiments, the prevention or reduction occurs through sedimentation of the one or more carbon-based materials upon sequestration by clay minerals.

[0050] In some embodiments, the sequestering of the one or more carbon-based materials by the one or more clay minerals forms a composite (e.g., steps 10, 12, and 14 in **FIG. 1**). In some embodiments, the composite includes an organoclay composite. In some embodiments, the composite includes a flocculated composite.

[0051] In some embodiments, the composite further includes one or more microorganisms derived from the water source. In some embodiments, the one or more microorganisms are in the form of a biofilm on a surface of the composite. In some embodiments, the one or more microorganisms includes, without limitation, bacteria, heterotrophic bacteria, nanoflagellates, dinoflagellates, ciliates, and combinations thereof. In some embodiments, the one or more microorganisms includes bacteria. In some embodiments, the bacteria includes *Alteromonas spp.* In some embodiments, the composite sediments into the water source.

[0052] In some embodiments, the formed composites are consumed by one or more planktons in the water source (e.g., step 16 in **FIG. 1**). In some embodiments, the one or more planktons includes one or more zooplanktons, such as copepods, krills, and/or salps. In some embodiments, the one or more planktons includes one or more phytoplanktons.

[0053] In some embodiments, the consumed composites are converted to fecal pellets and nutrients by planktons (e.g., step 18 in **FIG. 1**). Thereafter, the planktons release the fecal pellets and nutrients into the water source (e.g., step 20 in **FIG. 1**). In some embodiments, the water source includes sea water.

[0054] In some embodiments, the conversion prevents or reduces oxidation of the one or more carbon-based materials to carbon dioxide. In some embodiments, the conversion prevents or reduces release of carbon dioxide derived from the carbon-based materials by sinking the fecal pellets containing the carbon-based materials into the water source (e.g., steps 22 and 24 in **FIG. 1**).

[0055] The planktons may release various nutrients from the converted composites. For instance, in some embodiments, the released nutrients include, without limitation, silicon, iron, aluminum, manganese, ammonium, metals, and combinations thereof.

[0056] In some embodiments, the released nutrients boost production of planktons (e.g., phytoplanktons) in the water source (e.g., step 23 in **FIG. 1**). Thereafter, the planktons (e.g., phytoplanktons) may consume carbon dioxide and thereby sequester the carbon dioxide (e.g., step 25 in **FIG. 1**). In some embodiments, the planktons (e.g., phytoplanktons) may produce new carbonaceous materials for the sequestration of additional materials from the water source.

[0057] The methods of the present disclosure can have numerous embodiments. For instance, in some embodiments, the methods of the present disclosure include spreading clay minerals onto an ocean to increase the efficiency with which organic carbon-based materials are exported from the euphotic zone of the ocean to greater depths. In some embodiments, the methods of the present disclosure include a step of introducing clay minerals in an ocean to sequester exopolymers and initiate bacterial attachment.

[0058] Compositions

[0059] Additional embodiments of the present disclosure pertain to compositions that include one or more clay minerals. In some embodiments, the one or more clay minerals are operational for sequestering one or more carbon-based materials from a water source.

[0060] In some embodiments, the one or more clay minerals includes isolated clay minerals. In some embodiments, the one or more clay minerals includes purified clay minerals. In some embodiments, the one or more clay minerals includes, without limitation, palygorskite, nontronite, illite-smectite, kaolinite, chlorite, and combinations thereof.

[0061] In some embodiments, the one or more clay minerals includes at least a first clay mineral and a second clay mineral. In some embodiments, the first and second clay minerals are present at different weight ratios. For instance, in some embodiments, the first and second clay minerals are present at a weight ratio of 2:1, 3:1, 4:1, 5:1, 6:1, 7:1, 8:1, 9:1, or 10:1.

[0062] In some embodiments, the first and second clay minerals include palygorskite and nontronite. In some embodiments, the palygorskite and nontronite are present at a weight ratio of 4:1.

[0063] In some embodiments, the one or more clay minerals includes modified clay minerals. In some embodiments, the modified clay minerals include by-products or waste products of mining. In some embodiments, the modified clay minerals include clay minerals that have undergone a beneficiation process. In some embodiments, the modified clay minerals that have undergone a beneficiation process contain a lower amount of gangue minerals when compared to clay minerals that have not undergone the beneficiation process. In some embodiments, the gangue minerals include phosphate from phosphate rock. In some embodiments, the modified clay minerals include clay minerals that contain a lower amount of phosphate when compared to unmodified clay minerals.

[0064] In some embodiments, the modified clay minerals include one or more additional components. In some embodiments, the one or more additional components includes, without limitation, phosphorous, aluminum, iron, phyllosilicates, magnesium, alkali metals, alkaline earths, apatite, wavellite, crandallite, dolomite, quartz, or combinations thereof.

[0065] The clay minerals of the present disclosure can have various sizes. For instance, in some embodiments, the clay minerals of the present disclosure have sizes that range from about 0.1 μm to about 10 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes that range from about 0.2 μm to about 5 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes that range from about 0.2 μm to about 1 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 5 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 4 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes

of less than about 3 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 2 μm in diameter. In some embodiments, the clay minerals of the present disclosure have sizes of less than about 1 μm in diameter.

[0066] In some embodiments, the compositions of the present disclosure are in the form of a composite. In some embodiments, the composite includes one or more sequestered carbon-based materials and the one or more clay minerals. In some embodiments, the composite includes an organoclay composite. In some embodiments, the composite includes a flocculated composite.

[0067] In some embodiments, the composite further includes one or more microorganisms derived from a water source. In some embodiments, the one or more microorganisms is in the form of a biofilm on a surface of the composite. In some embodiments, the one or more microorganisms includes, without limitation, bacteria, heterotrophic bacteria, nanoflagellates, dinoflagellates, ciliates, and combinations thereof.

[0068] **Additional embodiments**

[0069] Reference will now be made to more specific embodiments of the present disclosure and experimental results that provide support for such embodiments. However, Applicant notes that the disclosure below is for illustrative purposes only and is not intended to limit the scope of the claimed subject matter in any way.

[0070] **Example 1. Interaction of clay minerals with carbohydrates and microbes**

[0071] In this Example, Applicant demonstrates that a mixture of clay minerals adsorbs carbohydrates (i.e., acid polysaccharides) dissolved in seawater to form organoclay composites, which flocculate and rapidly settle through a column of seawater. As illustrated in **FIGS. 2A-2D**, Applicant devised an experiment to evaluate the interaction of clay minerals with carbohydrates (i.e., acid polysaccharides, typically exuded by phytoplankton) and a common bacterium (i.e., *Alteromonas spp.*, a bacterium involved in decomposing particulate organic materials). The different funnels in **FIGS. 2A-2D** contain sea water, acid polysaccharides, and clay (**FIG. 2A**); sea water, acid polysaccharides, clay, and *Alteromonas spp.* (**FIGS. 2B and 2C**); and sea water, clay, and *Alteromonas spp.* (**FIG. 2D**).

[0072] **FIG. 3** shows that Zeta potential is a measure of the electrostatic interaction between organic molecules dissolved in seawater and clay minerals. Palygorskite and nontronite are two different types of clay minerals. Interactions between carbohydrates (acid polysaccharides) and palygorskite is physical, while interactions between carbohydrates (acid polysaccharides) and nontronites is

electrostatic. Formation of organo-clay composites and their flocculation takes place in seawater. Settling of the organoclay composites would thus remove dissolved organic materials (DOM) in seawater. The plot shows that, depending on the type and mixtures of clay minerals used, different amounts of carbon can be removed.

[0073] Additionally, Applicant observed that the presence of clay minerals does not adversely impact the growth of a common marine heterotroph (*Alteromonas*) or a key photosynthetic picoplankton (*Prochlorococcus*). In particular, **FIG. 4A** shows that the concentration of *Alteromonas* in the funnels illustrated in **FIGS. 2A-2D** (i.e., funnels 1-4, respectively) does not change when clay is added. Similarly, **FIG. 4B** shows that *Prochlorococcus* needs ammonium ion to grow, and that the growth shows no impact on the picoplankton when ammonia is added in the growth medium (i.e., fourth set of bars—the second bar).

[0074] **FIG. 5** shows a carbon sequestration experiment with a mixture of palygorskite and nontronite amended with apatite (a mineral to supply phosphorus—a key nutrient needed in the ocean). The experiment shows that the clay-apatite mixture removes a substantial amount of carbon due to the formation of organoclay composites (Funnel 1), which is reduced by *Alteromonas spp.* (Funnels 2,3) and sticks to bare-clay (i.e., no organoclay composites) (Funnel 4). In the open ocean, it is expected that formation of organoclay composites and their flocculation would remove DOM and bacteria from the ocean surface. Settling clay minerals coated with organics will be food for zooplankton, which will repackage the carbon and clay into fast settling fecal pellets. Additionally, partial digestion of clays by zooplankton will release nutrients for the phytoplankton.

[0075] **Example 2. Interaction of clay minerals with phytoplankton**

[0076] Experiments show that clay minerals induce production of Transparent Exopolymer Particles (TEPs) in natural cultures of a diatom (*Thalassiosira weissflogii*). The TEPs are highly sticky polymers causing *Thalassiosira weissflogii* to be removed from the water column as flocs. The results are summarized in **FIG. 6**. The *Thalassiosira weissflogii* culture was in an exponential phase when clay was added. The *Thalassiosira weissflogii* cells were not stressed. However, TEP production removed the cells from the water.

[0077] Furthermore, as summarized in **FIG. 7**, treatments with clay leached in seawater show that the trace elements released are not toxic for *Thalassiosira weissflogii*. In particular, the results in **FIG. 7** show that *Thalassiosira weissflogii* grows, consumes dissolved silica from seawater, and produces

TEP in the presence of clay. The *Thalassiosira weissflogii* culture was 15 days old. The culture included stationary and senescent cells.

[0078] Example 3. Interaction of clay mineral flocs with zooplankton

[0079] *Thalassiosira weissflogii* was grown in filtered seawater inoculated with fecal pellets from copepod *Acartia tonsa*, which was fed a diet of *T. weissflogii* + *Rhodomonas* sp. under treatment I and *T. weissflogii* and *Rhodomonas* sp. + clay under treatment II. Results showed that *T. weissflogii* cell counts and chlorophyll a (*Chl a*) concentration increased significantly with the addition of fecal pellets, and that the TEP concentrations were relatively high for fecal pellets +clay compared to fecal pellets without clay and control. The results are summarized in **Table 1**.

Condition	Initial count	<i>T. weissflogii</i> count (cells/ml)	Chlorophyll a count		Nutrients			
			Chla extract	Chla (µg/L)	Si	P	N	N/P
N/A	N/A	161.57	N/A	N/A	1.4	0.74	8.1	10.97
Seawater + Phyto	T1	5134	0.147	0.015	1.96	0.88	15.7	17.82
Seawater + Phyto + FP control	T2	3520	0.140	0.014	2.48	0.69	14.4	20.67
Seawater + Phyto + FP treatment	T3	4293	0.156	0.016	2.15	0.88	14.4	16.32
Seawater + Phyto + FP filtrate treatment	T4	6095	0.177	0.018	2.62	0.74	14.7	19.88

Table 1. Fecal pellets provide nutrients. The experiment duration was 18 hours for 0.75 days. Phyto= Phytoplankton cells (i.e., *T. weissflogii*). FP= Fecal pellets. Chla= Chlorophyll a. Si=Silicate (µM/L). P=Phosphate (µM/L).

[0080] Previous studies have shown that zooplankton (copepods/krills) tend to ingest clay minerals, which are attacked in the zooplankton gut. Zooplanktons release variable amounts of dissolved phosphate, ammonium, and bioavailable iron—all nutrients for the phytoplankton and also excrete clay mineral bearing fecal pellets (POM), which settle rapidly through seawater column.

[0081] Thus, without being bound by theory, and as illustrated in **FIG. 8**, Applicant envisions the following mechanism for removing atmospheric CO₂ by clay minerals: labile dissolved organic matter produced by phytoplankton is bound to clay minerals to form organoclay composites that initiate formation of sinking aggregates, which are then taken up by zooplankton to produce rapidly sinking fecal pellets and nutrients. The cycle is completed when zooplankton released nutrients boost primary production (phytoplankton growth), further sequestering atmospheric CO₂ and producing more DOM.

[0082] By converting DOM to POM, and by significantly reducing oxidation of POM through the upper 1000 m of the ocean, a much larger fraction of organic matter can be sequestered in the deep ocean. Applicant's experiments in this Example show that both of these goals can be accomplished by utilizing clay minerals.

[0083] Without further elaboration, it is believed that one skilled in the art can, using the description herein, utilize the present disclosure to its fullest extent. The embodiments described herein are to be construed as illustrative and not as constraining the remainder of the disclosure in any way whatsoever. While the embodiments have been shown and described, many variations and modifications thereof can be made by one skilled in the art without departing from the spirit and teachings of the invention. Accordingly, the scope of protection is not limited by the description set out above, but is only limited by the claims, including all equivalents of the subject matter of the claims. The disclosures of all patents, patent applications and publications cited herein are hereby incorporated herein by reference, to the extent that they provide procedural or other details consistent with and supplementary to those set forth herein.

CLAIMS

1. A method of sequestering one or more carbon-based materials, said method comprising:
applying one or more clay minerals to a water source, wherein the one or more clay minerals sequesters the one or more carbon-based materials.
2. The method of claim 1, wherein the one or more carbon-based materials is selected from the group consisting of carbon dioxide, one or more organic materials, and combinations thereof.
3. The method of claim 1, wherein the one or more carbon-based materials comprises one or more organic materials.
4. The method of claim 3, wherein the one or more organic materials is selected from the group consisting of carbohydrates, proteins, lipids, polysaccharides, acidic polysaccharides, organic molecules, polymers, exopolymers, hydrocarbons, dissolved organic matter (DOM), particulate organic matter (POM), and combinations thereof.
5. The method of claim 1, wherein the one or more organic materials comprises dissolved organic matter (DOM).
6. The method of claim 1, wherein the one or more organic materials comprises particulate organic matter (POM).
7. The method of claim 1, wherein the one or more carbon-based materials comprises carbon dioxide dissolved in the water source.
8. The method of claim 1, wherein the sequestering occurs by a method selected from the group consisting of adhesion, binding, adsorption, electrostatic interaction, and combinations thereof.
9. The method of claim 1, wherein the applying occurs by a method selected from the group consisting of spreading, pouring, sprinkling, spraying, and combinations thereof.

10. The method of claim 1, wherein the applying occurs by a mechanical mechanism, a manual mechanism, a physical mechanism, a non-natural mechanism, or combinations thereof.
11. The method of claim 1, wherein the one or more clay minerals comprises isolated clay minerals.
12. The method of claim 1, wherein the one or more clay minerals comprises purified clay minerals.
13. The method of claim 1, wherein the one or more clay minerals is selected from the group consisting of palygorskite, nontronite, illite-smectite, kaolinite, chlorite, and combinations thereof.
14. The method of claim 1, wherein the one or more clay minerals comprise at least a first clay mineral and a second clay mineral.
15. The method of claim 14, wherein the first and second clay minerals comprise palygorskite and nontronite.
16. The method of claim 15, wherein the palygorskite and nontronite are present at a weight ratio of 4:1.
17. The method of claim 1, wherein the one or more clay minerals comprises modified clay minerals.
18. The method of claim 17, wherein the modified clay minerals comprise one or more additional components.
19. The method of claim 17, wherein the one or more additional components is selected from the group consisting of phosphorous, aluminum, iron, phyllosilicates, magnesium, alkali metals, alkaline earths, apatite, wavellite, crandallite, dolomite, quartz, or combinations thereof.
20. The method of claim 17, wherein the one or more additional components comprises apatite.

21. The method of claim 17, wherein the modified clay minerals comprise clay minerals that have undergone a beneficiation process.
22. The method of claim 1, wherein the water source comprises a sea.
23. The method of claim 1, wherein the water source comprises an ocean.
24. The method of claim 23, wherein the sequestering occurs below the euphotic zone of the ocean.
25. The method of claim 1, wherein the method prevents or reduces oxidation of the one or more carbon-based materials to carbon dioxide.
26. The method of claim 25, wherein the prevention or reduction occurs through sedimentation of the one or more sequestered carbon-based materials.
27. The method of claim 1, wherein the sequestering of the one or more carbon-based materials to the one or more clay minerals forms a composite.
28. The method of claim 27, wherein the composite comprises an organoclay composite.
29. The method of claim 27, wherein the composite comprises a flocculated composite.
30. The method of claim 27, wherein the composite further comprises one or more microorganisms derived from the water source.
31. The method of claim 30, wherein the one or more microorganisms is selected from the group consisting of bacteria, heterotrophic bacteria, nanoflagellates, dinoflagellates, ciliates, and combinations thereof.
32. The method of claim 30, wherein the one or more microorganisms comprises bacteria.

33. The method of claim 32, wherein the bacteria comprises *Alteromonas spp.*
34. The method of claim 32, wherein the one or more microorganisms is in the form of a biofilm on a surface of the composite.
35. The method of claim 27, wherein the composite sediments into the water source.
36. A composition comprising one or more clay minerals, wherein the one or more clay minerals is operational for sequestering one or more carbon-based materials from a water source.
37. The composition of claim 36, wherein the one or more clay minerals comprises isolated clay minerals.
38. The composition of claim 36, wherein the one or more clay minerals comprises purified clay minerals.
39. The composition of claim 36, wherein the one or more clay minerals is selected from the group consisting of palygorskite, nontronite, illite-smectite, kaolinite, chlorite, and combinations thereof.
40. The composition of claim 36, wherein the one or more clay minerals comprise at least a first clay mineral and a second clay mineral.
41. The composition of claim 40, wherein the first and second clay minerals comprise palygorskite and nontronite.
42. The composition of claim 41, wherein the palygorskite and nontronite are present at a weight ratio of 4:1.

43. The composition of claim 36, wherein the one or more clay minerals comprises modified clay minerals.
44. The composition of claim 43, wherein the modified clay minerals comprise one or more additional components.
45. The composition of claim 44, wherein the one or more additional components is selected from the group consisting of phosphorous, aluminum, iron, phyllosilicates, magnesium, alkali metals, alkaline earths, apatite, wavellite, crandallite, dolomite, quartz, or combinations thereof.
46. The composition of claim 44, wherein the one or more additional components comprises apatite.
47. The composition of claim 43, wherein the modified clay minerals comprise clay minerals that have undergone a beneficiation process.
48. The composition of claim 36, wherein the composition is in the form of a composite comprising one or more sequestered carbon-based materials and the one or more clay minerals.
49. The composition of claim 48, wherein the composite comprises an organoclay composite.
50. The composition of claim 48, wherein the composite comprises a flocculated composite.
51. The composition of claim 48, wherein the composite further comprises one or more microorganisms.
52. The composition of claim 51, wherein the one or more microorganisms is selected from the group consisting of bacteria, heterotrophic bacteria, nanoflagellates, dinoflagellates, ciliates, and combinations thereof.
53. The composition of claim 51, wherein the one or more microorganisms comprises bacteria.

54. The composition of claim 53, wherein the bacteria comprises *Alteromonas spp.*
55. The composition of claim 51, wherein the one or more microorganisms is in the form of a biofilm on a surface of the composite.

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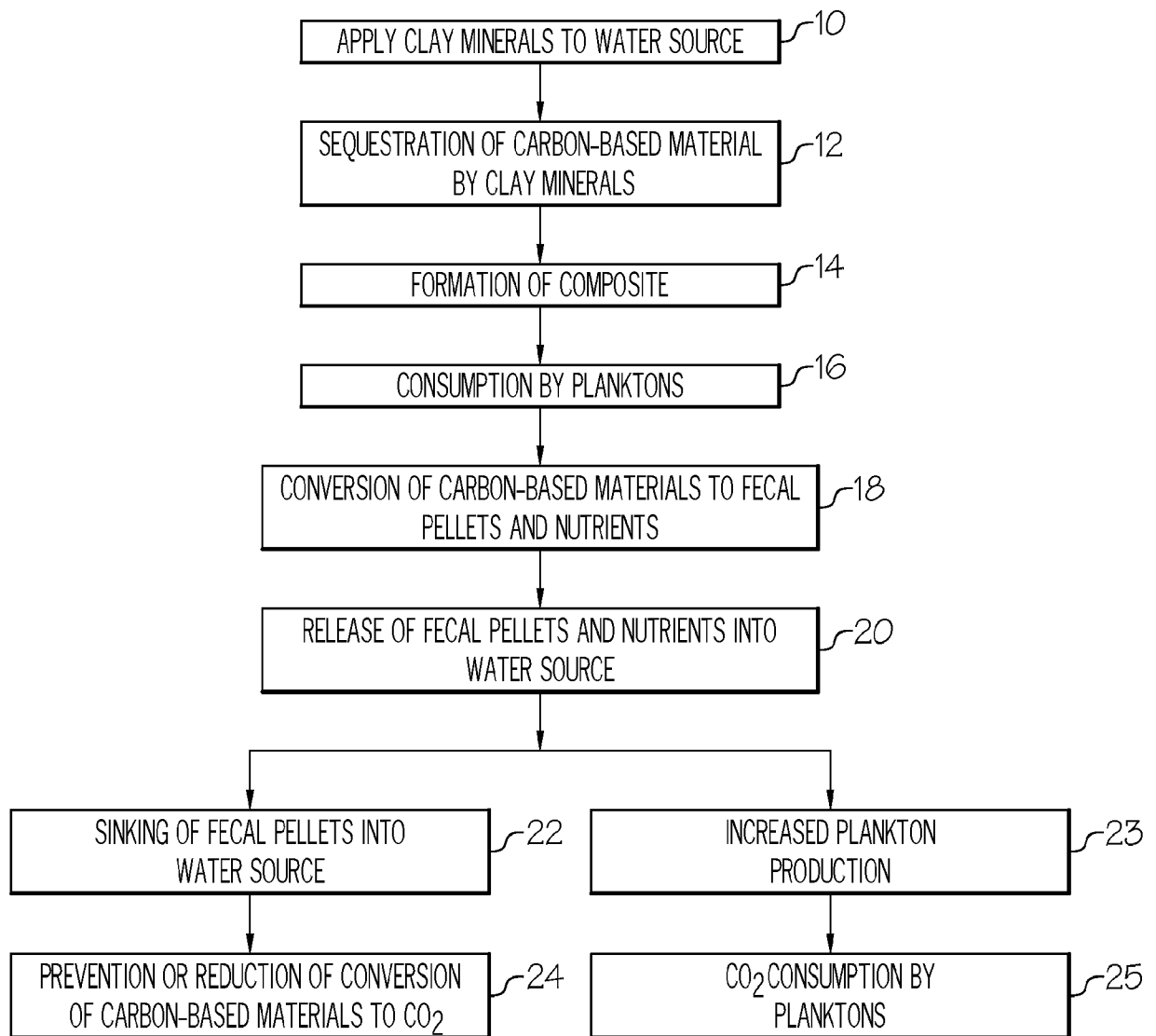


FIG. 1

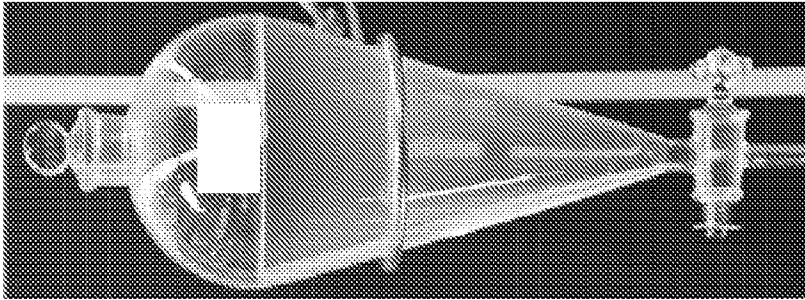


FIG. 2A

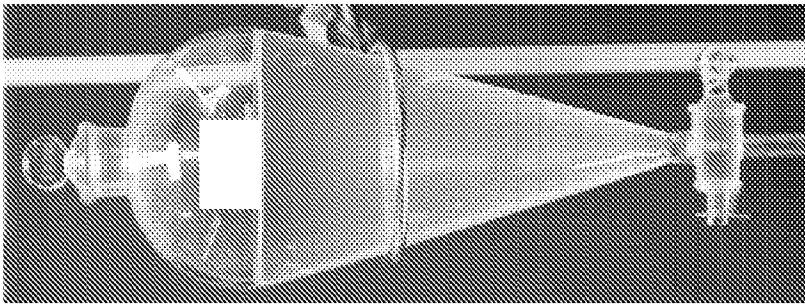


FIG. 2B

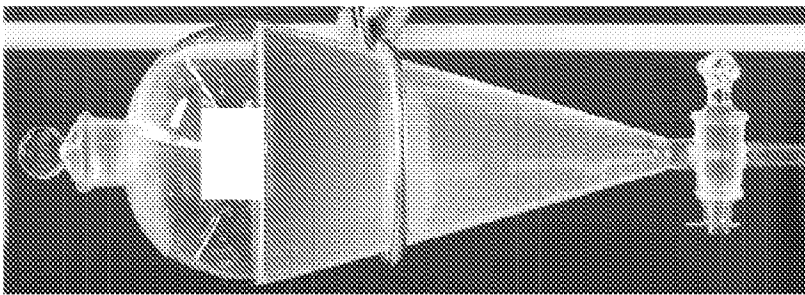


FIG. 2C

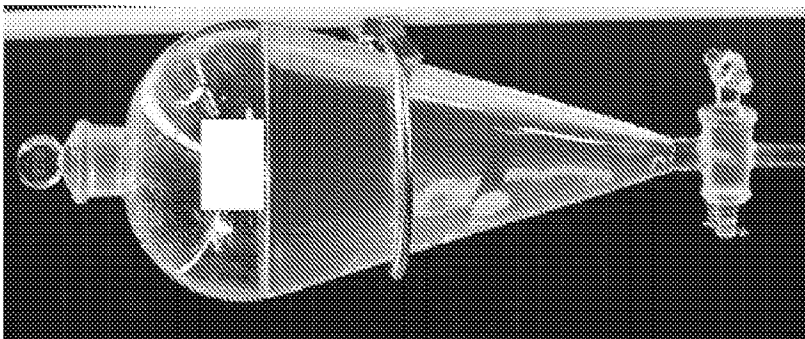


FIG. 2D

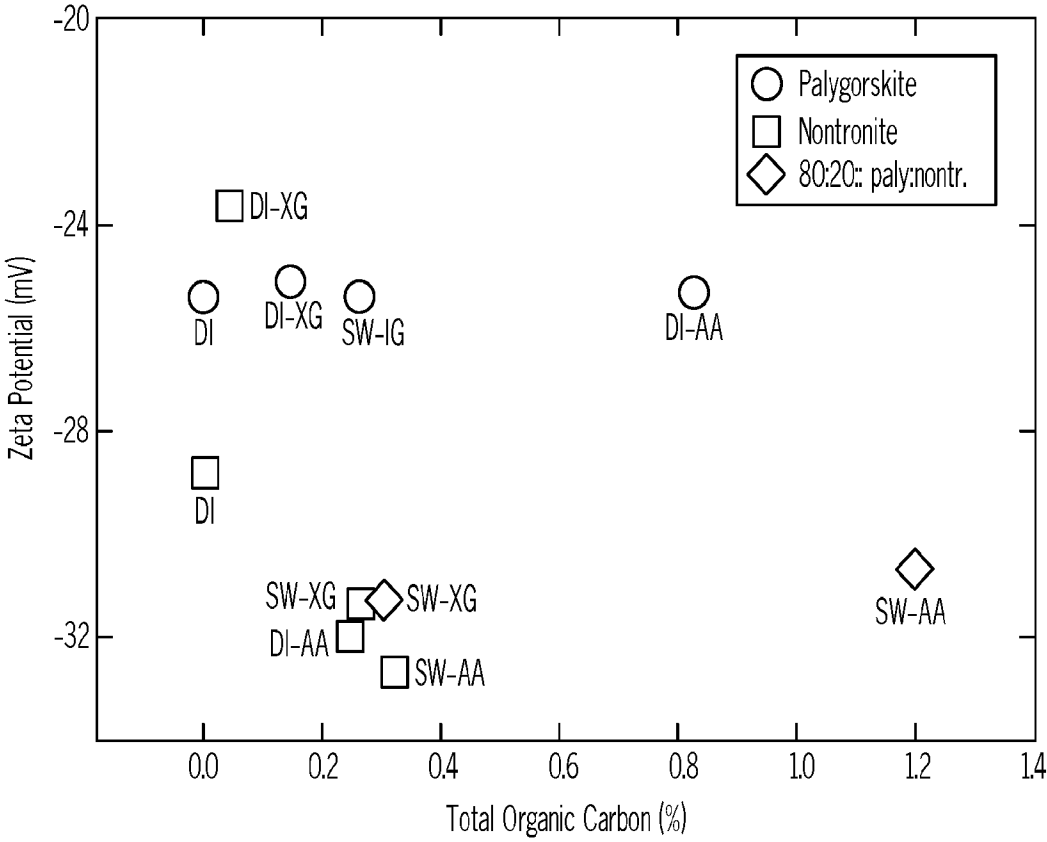


FIG. 3

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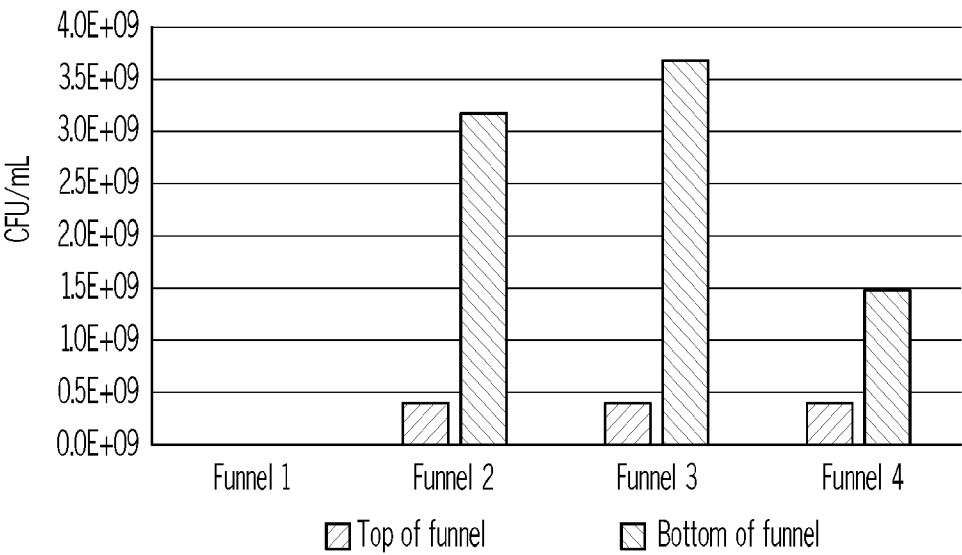


FIG. 4A

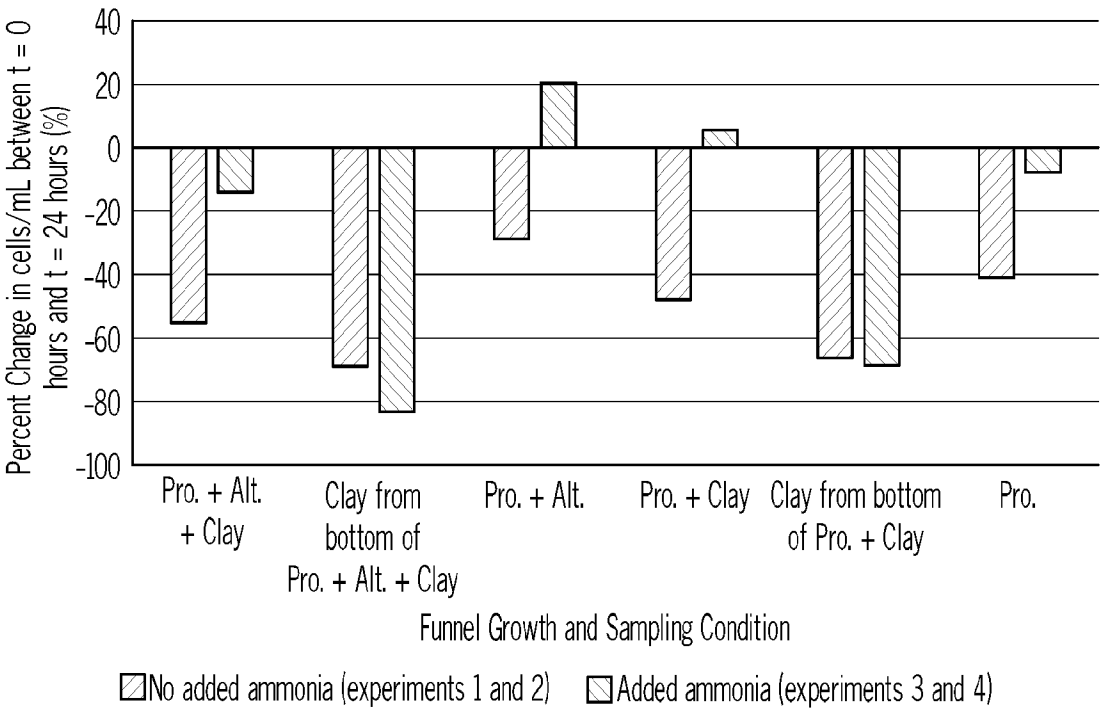


FIG. 4B

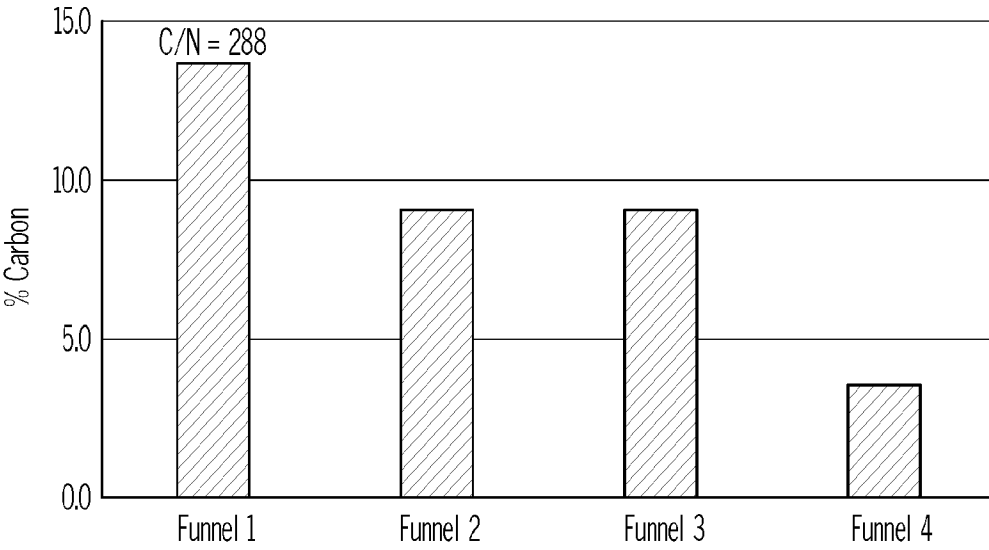


FIG. 5

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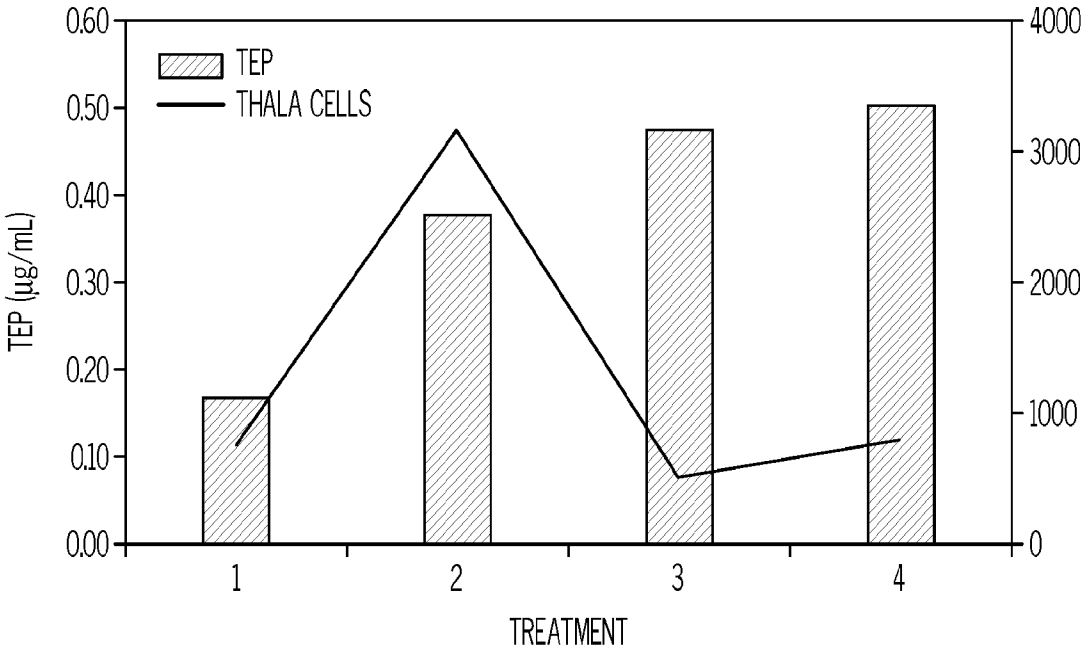


FIG. 6

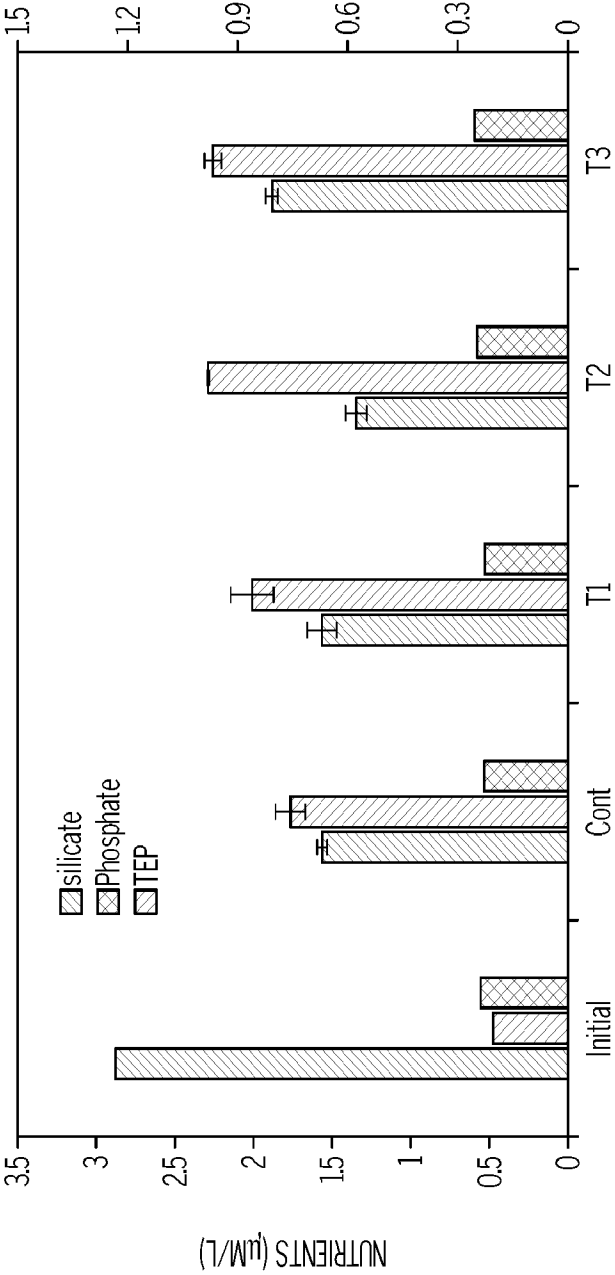


FIG. 7

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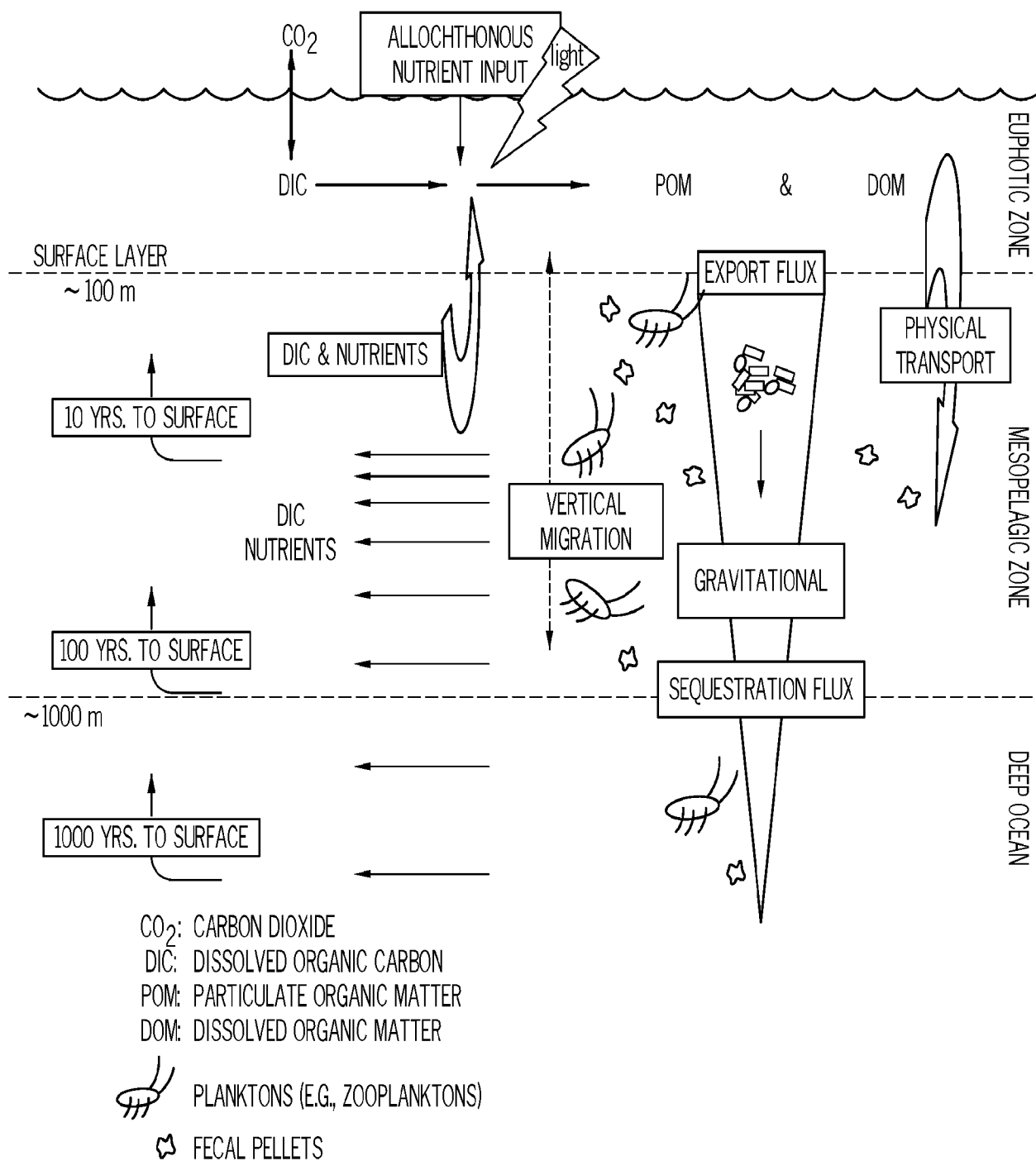


FIG. 8

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 23/15817

A. CLASSIFICATION OF SUBJECT MATTER

IPC - INV. B01D 21/01, C02F 3/34, C02F 1/52, C01B 33/40 (2023.01)

ADD. C02F 1/28, B01D 15/00 (2023.01)

CPC - INV. C02F 1/5236, B01D 21/01, C02F 3/34, C02F 1/52, C02F 3/106, C02F 1/281, C01B 33/40

ADD. C02F 1/28, C02F 2101/30, B01D 2253/11, Y02C 20/40, B01D 2257/504, Y02P 20/151

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

See Search History document

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

See Search History document

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

See Search History document

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WARR et al., "Nontronite-enhanced biodegradation of Deepwater Horizon crude oil by Alcanivorax borkumensis"; Applied Clay Science, Volume 158 (August 2018), pg 11-20 (the entire document, and more specifically: pg 1, col 1, para 1-2; pg 1, col 2, para 1; pg 12, col 1, para 2-3; pg 12, col 2, para 1; pg 13, col 1, para 4; pg 17, col 2, para 4; pg 19, col 2, para 2; table 2; figure 6-7; abstract; title)	1-4, 10-16 and 23-24
X ----- Y	ARAGAW et al., "Synthesis and characterization of Ethiopian kaolin for the removal of basic yellow (BY 28) dye from aqueous solution as a potential adsorbent"; Heliyon, Volume 6, Issue 9 (September 2020), pg 1-17 (the entire document, and more specifically: pg 1, col 1, para 1; pg 1, col 2, para 1; pg 2, col 2, para 3; pg 12, col 1, para 2; pg 15, col 1, para 2-3; figures 8a, 8b, and 8c; table 2; abstract; title)	1, 3, 5, 8, 17-19 and 21 ----- 20
X ----- Y	LIU et al., "CO2 sequestration by mineral trapping in natural analogues in the Yinggehai Basin, South China Sea"; Marine and Petroleum Geology, Volume 104 (June 2019), pg 190-199 (the entire document, and more specifically: pg 193, col 2, para 2-5; pg 195, col 2, para 5; pg 198, col 2, para 2; abstract; title)	1, 3 and 7 ----- 25-26
X	US 4,473,477 A (BEALL) 25 September 1984 (25.09.1984); the entire document, and more specifically: col 1, ln 14-17; col 6, ln 6-20; col 7, ln 56-68; col 8, ln 1-21; figures 8-9; abstract; title	1, 6, 9, 27 and 35
X ----- Y	US 5,342,525 A (ROWSELL) 30 August 1994 (30.08.1994); the entire document, and more specifically: col 1, ln 14-54; col 5, ln 23-48; col 6, ln 16-21; col 7, ln 26-29; col 11, ln 45-47; col 16, ln 3-24; col 17, ln 29-32; col 22, ln 57-63; col 23, ln 1-5; col 24, ln 47-51; abstract; title	1, 22 and 27-32 ----- 33-34

☒ Further documents are listed in the continuation of Box C.☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"D" document cited by the applicant in the international application

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

10 June 2023

Date of mailing of the international search report

JUL 27 2023

Name and mailing address of the ISA/US

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Authorized officer

Kari Rodriguez

Telephone No. PCT Helpdesk: 571-272-4300

INTERNATIONAL SEARCH REPORT

International application No.

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C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	BENSALAH, "Natural and Synthetic Apatites as Adsorbents for the Removal of Azo Dyes from Aqueous Solutions"; Doctoral dissertation submitted in fulfillment of the requirements for the degree of: Doktor der Ingenieurwissenschaften (December 2019), pg i-vi and 1-96 (pg 27, para 1; title; abstract)	20
Y	HARTMANN et al., "Enhanced chemical weathering as a geoengineering strategy to reduce atmospheric carbon dioxide, supply nutrients, and mitigate ocean acidification"; Reviews of Geophysics, Volume 51, Issue 2 (June 2013), pg 113-149 (the entire document, and more specifically: pg 114, col 1, para 3; pg 116, col 2, para 2-3; pg 117, col 1, para 1-2; abstract; title)	25-26
Y	FLEMMING et al., "The biofilm matrix"; Nature Reviews, Microbiology; Volume 8 (September 2010), pg 623-633 (pg 623, col 1, para 1-2; pg 630, col 2, para 3; abstract; title)	33
Y	CHEN et al., "Characterization of a novel biofloculant from a marine bacterium and its application in dye wastewater treatment"; BMC Biotechnology, Volume 17, Number 1 (November 2017), pg 1-11 (the entire document, and more specifically: pg 1, col 1, para 1; pg 1, col 2, para 1; pg 9, col 1, para 1; abstract; title)	34

INTERNATIONAL SEARCH REPORT

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Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:
This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be searched, the appropriate additional search fees must be paid.

Group I: claims 1-35, directed to a method of sequestering one or more carbon-based materials.

Group II: claims 36-55, directed to a composition comprising one or more clay minerals.

The inventions listed as Groups I-II do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

****please see the continuation at the end of this form****

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:
1-35

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- ☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- ☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT
Information on patent family members

International application No.

PCT/US 23/15817

****continuation of Box III (Lack of Unity of Invention)****

Special Technical Features:

Group I requires a method of sequestering one or more carbon-based materials; and applying one or more clay minerals to a water source, not required by group II.

Common Technical Features:

Groups I and II share the technical feature of one or more clay minerals sequestering one or more carbon-based materials; and a water source.

However, these shared technical features do not represent a contribution over prior art, because the shared technical feature is being anticipated by the article "Nontronite enhanced biodegradation of Deepwater Horizon crude oil by *Alcanivorax borkumensis*" to Warr et al. (hereinafter Warr.). Warr teaches one or more clay minerals sequestering one or more carbon-based materials (pg 17, col 2, para 4; pg 19, col 2, para 2; figure 6-7; The effect of adding nontronite-based clay flakes to the crude oil is more precisely determined by plotting the difference between the clay-treated and non-clay treated samples (figures 6 & 7). A clear depletion of alkanes between C13–C28 occurred after clay addition, which is attributable to the enhanced activity of *A. borkumensis* (figure 6). Our laboratory experiments demonstrate that adding nontronite (Fesmectite) based clay flakes to Deepwater Horizon crude oil in the presence of *A. borkumensis* leads to a significant increase in hydrocarbon degradation over an experimental period of 37 days (888 h) when mixed with nutrient-poor seawater. The bacteria population increased five-fold relative to the non-clay bearing control and was accompanied by a 57.7% reduction in alkanes compounds that included pristane and phytane.; and a water source (pg 12, col 1, para 2-3; In this contribution we experimentally investigate the role of nontronite-based clay flakes on the biodegradation of fresh Deepwater Horizon (MC 252) crude oil in an aqueous marine system.).

As the shared technical features were known in the art at the time of the invention, they cannot be considered common technical features that would otherwise unify the groups. Therefore, Groups I-II lack unity under PCT Rule 13.

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 23/15817

****continuation of CPC codes from page 1****
CPC -

ADD. C02F 2103/007, C02F 2103/08