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(54) Title: BIOMASS TO USEFUL HYDROCARBONS

(57) Abstract: Methods are provided for gasification of cellulosic biomass outside of a termite gut using simulated termite gut cell-to-cell communication.



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FIELD OF THE INVENTION

[0001] This invention relates to conversion of biomass to useful hydrocarbons and, more particularly, to conversion of biomass to useful hydrocarbons utilizing simulated termite gut cell-to-cell communication.

BACKGROUND

[0002] While experts in energy management may dispute the life span of oil reserves, many believe that its depletion will happen within the next half century. As mankind's reliance on, and need for, reliable, sustainable fuel sources increases, researchers are investing increasing amounts of time and money into alternative energy sources.

[0003] Among the various options, use of biomass as a renewable, carbon-based energy source has become a popular idea. Biomass comprises any plant or organic waste that can be used to produce heat, power or fuel. Common examples of biomass are sugar cane and its byproduct, bagasse, corn, switch grass, crop residuals, and animal manure.

[0004] The most common way to convert biomass to energy is burning. The heat generated from burning can be used, e.g., to heat buildings or dry crops, and can be used to create steam for driving steam turbines to generate electricity. However, there are substantial needs for hydrocarbons from biomass that can be used directly, e.g., as fuel. Several publications describe methods for converting biomass to various hydrocarbon chemical intermediates and feed-stocks. Cellulose hydrolysis to glucose and subsequent fermentation has been utilized to produce ethanol and related materials, but overall process economics have not been favorable. US Patent 4,592,762 describes several published processes for gasification of cellulosic biomass.

[0005] Termites have been steadily and effectively converting cellulose to various metabolic products for millenia. Climate researchers have been particularly interested in methane produced in the termite gut. US Patents 5,670,345 and 5,854,032 describe use of bacteria from termite guts for producing humic acid from coal. US Patent 6,143,534 describes use of termite guts for producing methane from coal.

[0006] Yet to our knowledge there is no commercially implemented means of converting biomass to useful gaseous or liquid hydrocarbons. It is estimated that the Gulf Coast makes enough cellulosic materials to provide fuel for much of North America

if commercially viable means for converting the cellulose to such hydrocarbons were available.

[0007] Thus, a need exists for commercially feasible means for converting biomass to useful hydrocarbons.

SUMMARY OF THE INVENTION

[0008] This invention provides methods of converting biomass to useful hydrocarbons. According to this invention, biomass is converted to gaseous methane outside termite gut by using simulated cell-to-cell communication as is done inside termite gut; and the gaseous methane is converted to useful hydrocarbons via means for converting gas to liquids.

DETAILED DESCRIPTION OF THE INVENTION

[0009] The following are provided by this invention. Methods comprising simulating termite gut cell-to-cell communication to form biofilm suitable for forming gaseous methane from biomass outside termite gut. Methods comprising: (a) simulating termite gut cell-to-cell communication to form biofilm suitable for forming gaseous methane from biomass; and (b) using the biofilm to form gaseous methane from biomass outside termite gut; and such methods wherein (b) is conducted at substantially ambient pressure and substantially ambient temperature. Methods comprising: (a) forming gaseous methane from biomass outside termite gut by simulating termite gut cell-to-cell communication; and (b) converting the methane to useful hydrocarbons via process for converting gas to liquids. Methods comprising: (a) identifying termite gut environment capable of converting biomass to methane; (b) simulating the termite gut environment in container outside the termite gut; and (c) converting biomass to methane in the container outside the termite gut. Methods comprising: (a) identifying termite gut environment capable of converting biomass to methane; (b) simulating the termite gut environment in container outside the termite gut; (c) converting biomass to methane in the simulated termite gut in the container; and (d) converting the methane to useful hydrocarbons via process for converting gas to liquids. Methods comprising: (a) obtaining intestinal flora from termite gut environment; (b) simulating cell-to-cell chemistry of the intestinal flora that causes biomass to convert to methane in container outside the termite gut; and (c) converting biomass to methane in the container outside the termite gut. Methods comprising: (a) obtaining intestinal flora from termite gut environment; (b) simulating cell-to-cell chemistry of the intestinal flora that causes

biomass to convert to methane in container outside the termite gut to form biofilm suitable for converting biomass to methane; (c) adding biomass to the container containing the biofilm; and (d) converting biomass to methane in the container.

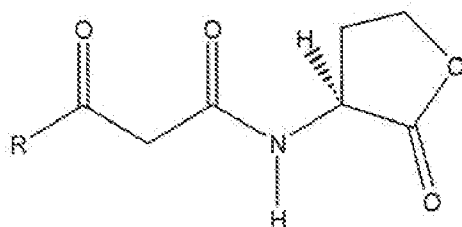
Methods comprising: (a) obtaining intestinal flora from termite gut environment; (b) simulating cell-to-cell chemistry of the intestinal flora that causes biomass to convert to methane in container outside the termite gut; (c) converting biomass to methane in the container outside the termite gut; and (d) converting the methane to useful hydrocarbons via process for converting gas to liquids. Methods comprising: (a) obtaining intestinal flora from termite gut environment; (b) simulating cell-to-cell chemistry of the intestinal flora that causes biomass to convert to methane in container outside the termite gut to form biofilm suitable for converting biomass to methane; (c) adding biomass to the container containing the biofilm; (d) converting biomass to methane in the container; and (e) converting the methane to useful hydrocarbons via process for converting gas to liquids.

[0010] According to this invention, intestinal flora is obtained from termite gut using known or newly developed microbiological investigative techniques. As used herein, "intestinal flora" comprises bacteria and other organisms that live in the intestine (such other organisms may include, without limitation, various protozoans, fungi and yeast). Known or newly developed investigative techniques are used to establish and control biofilm communities that simulate termite gut environment. For example, cell-to-cell signal chemistry of the intestinal flora is identified as is needed to establish and control such biofilm communities. Such biofilm communities are established outside termite gut and are used for converting cellulose to methane. Cellulose and the biofilm can be combined in a container, i.e., both put into the container, for the conversion to methane to occur.

[0011] *In vivo* termite gut intestinal flora comprise a biofilm system that is controlled by cell-to-cell signals such as quorum-sensing signal compounds. In processes of this invention, quorum-sensing signal compounds are simulated and used to construct and inhabit such biofilm systems in an *in vitro* environment. Once such a protection structure is established, the normal operations of the micro-organism community can be harnessed to such biotechnological applications as described within the confines of this document.

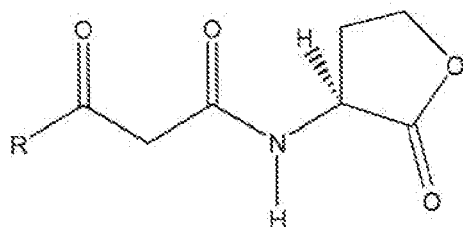
[0012] Some non-limiting examples of biofilm forming compounds are as follows:

Signal "A", acylated homoserine lactone (attachment signal for *Pseudomonas aeruginosa*):



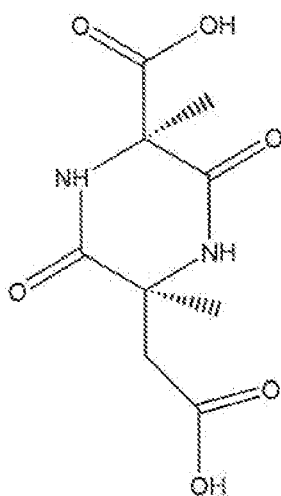
where R is propyl;

Signal "B", acylated homoserine lactone (regulation signal for *Agrobacterium tumefaciens*):



where R is pentyl; and

Signal "C", cyclic peptide (cell-to-cell signal for *Escherichia coli*):



Cyclo (L-Asp-L-Glu)

[0013] Two or more of such signal compounds can be combined to establish biofilm structure. For example, a plurality of signal "A" can be combined to establish biofilm structure, or a plurality of signal "A" and a plurality of signal "B" can be combined to establish biofilm structure. Numerous varieties of combinations of such signal compounds for forming biofilm structure(s) are contemplated by this invention. When two or more of such signal compounds are combined for at least about 30 minutes to about 24 hours, e.g., for about 4 hours, the signal compounds can establish biofilm structure.

[0014] Cell-to-cell signal chemistry of intestinal flora from gut of *Coptotermes* (Formosan-subterranean termites and/or Native-subterranean termites) can be simulated for use in this invention. Cell-to-cell signal chemistry of intestinal flora from other termite sources can also be simulated for use in this invention, e.g., for producing biofilm for use in this invention.

[0015] It is estimated that a 10,000 gallon stirred tank reactor could contain enough termite gut-simulating biofilm to convert 1000 board-feet of cellulose per minute to methane.

[0016] As a side-benefit, as the cellulosic material is "digested" in termite-gut simulating environment, two different streams are produced: (i) product of carbohydrate conversion (i.e., methane), and (ii) essentially unmodified (undigested) lignin (i.e., antioxidant materials). Such antioxidant materials may be used as industrial antioxidant. Examples of applications for this type of antioxidant include antioxidant protection of polymerics (tires, asphalt, etc.).

[0017] Methods of this invention have the advantage of utilizing biomass conversion capability of termite gut outside the termite gut. Additionally, methods of this invention convert biomass to methane at substantially ambient pressure and temperature, adding to the commercial viability of such methods.

EXAMPLE

[0018] The following example is illustrative of the principles of this invention. It is understood that this invention is not limited to any one specific embodiment exemplified herein, whether in the examples or the remainder of this patent application.

[0019] Nests of Formosan-subterranean termites are collected from infested mulch in Louisiana. Termites are removed less than 2 hours after sampling using sterile forceps. The hind-guts of the termites are removed, transferred, and kept at 5°C in

Hungate tubes containing 5 ml medium of Widdel (1980) and gassed with N_2/CO_2 (80/20). The Hungate tubes are transferred into an anaerobic glove box. The gut walls are disrupted in a 5-ml sterilized tissue homogenizer for 5 minutes, then transferred into a 60-ml serum bottle containing 15 ml of Widdel culture medium with the following composition in g/l: KH_2PO_4 , 0.2; NH_4Cl , 0.3; KCl , 0.5; $NaCl$, 1; $CaCl_2 \cdot 2H_2O$, 0.15; $MgCl_2 \cdot 6H_2O$, 0.4; 1 ml/l of trace element solution (Imhoff-Stuckie and Pfennig 1983) and 1 ml/l of resazurin (0.1%, wt/vol) are added. The medium is adjusted to pH 7.0 with KOH and boiled under O_2 -free N_2 . After cooling to room temperature, 20 ml medium is transferred into 60-ml serum bottles inside the glove box. The bottles are stoppered with black butyl rubber closures (Bellco) and outgassed with N_2/CO_2 . After sterilization (110°C, 35 minutes), the following sterile solutions are added to each bottle using one-way syringes: 0.25 ml $NaHCO_3$ (10%, wt/vol); 0.2 ml $Na_2S \cdot 9H_2O$ (4%, wt/vol); 0.2 ml vitamin solution (10%, wt/vol) (Pfennig 1978) and 50 microliters Na_2SeO_3 (0.3%, wt/vol).

[0020] A plurality of quorum sensing signal compounds "A" (as defined herein) are placed in a container. After about 4 hours, the plurality of quorum sensing signal compounds "A" establish a suitable biofilm structure. Then, a portion of the contents of the 60-ml serum bottles containing removed termite hind-guts are removed from the bottles and placed in the container with the plurality of quorum sensing signal compounds "A" and the biofilm structure constructed thereby. After about 4 hours, the quorum sensing signal compounds innate to the termite hind-gut flora adequately overtake the biofilm structure and a biofilm structure emulating that of the termite hind-gut is established.

[0021] The thus-formed biofilm structure emulating that of the termite hind-gut is added to sugar cane biomass and methane is produced.

[0022] While the present invention has been described in terms of one or more preferred embodiments, it is to be understood that other modifications may be made without departing from the scope of the invention, which is set forth in the claims below.

CLAIMS

1. A method comprising simulating termite gut cell-to-cell communication to form biofilm suitable for forming gaseous methane from biomass outside termite gut.
2. A method comprising:
 - (a) simulating termite gut cell-to-cell communication to form biofilm suitable for forming gaseous methane from biomass; and
 - (b) using the biofilm to form gaseous methane from biomass outside termite gut.
3. The method of claim 2 wherein (b) is conducted at substantially ambient pressure and substantially ambient temperature.
4. A method comprising:
 - (a) forming gaseous methane from biomass outside termite gut by simulating termite gut cell-to-cell communication; and
 - (b) converting the methane to useful hydrocarbons via process for converting gas to liquids.
5. A method comprising:
 - (a) identifying termite gut environment capable of converting biomass to methane;
 - (b) simulating the termite gut environment in container outside the termite gut; and
 - (c) converting biomass to methane in the container outside the termite gut.
6. A method comprising:
 - (a) identifying termite gut environment capable of converting biomass to methane;
 - (b) simulating the termite gut environment in container outside the termite gut;
 - (c) converting biomass to methane in the simulated termite gut in the container; and
 - (d) converting the methane to useful hydrocarbons via process for converting gas to liquids.

7. A method comprising:
 - (a) obtaining intestinal flora from termite gut environment;
 - (b) simulating cell-to-cell chemistry of the intestinal flora that causes biomass to convert to methane in container outside the termite gut; and
 - (c) converting biomass to methane in the container outside the termite gut.
8. A method comprising:
 - (a) obtaining intestinal flora from termite gut environment;
 - (b) simulating cell-to-cell chemistry of the intestinal flora that causes biomass to convert to methane in container outside the termite gut to form biofilm suitable for converting biomass to methane;
 - (c) adding biomass to the container containing the biofilm; and
 - (d) converting biomass to methane in the container.
9. A method comprising:
 - (a) obtaining intestinal flora from termite gut environment;
 - (b) simulating cell-to-cell chemistry of the intestinal flora that causes biomass to convert to methane in container outside the termite gut;
 - (c) converting biomass to methane in the container outside the termite gut; and
 - (d) converting the methane to useful hydrocarbons via process for converting gas to liquids.
10. A method comprising:
 - (a) obtaining intestinal flora from termite gut environment;
 - (b) simulating cell-to-cell chemistry of the intestinal flora that causes biomass to convert to methane in container outside the termite gut to form biofilm suitable for converting biomass to methane;
 - (c) adding biomass to the container containing the biofilm;
 - (d) converting biomass to methane in the container; and
 - (e) converting the methane to useful hydrocarbons via process for converting gas to liquids.