Methanogenic Conversion of CO₂ Into CH₄

A Potential Remediation Technology for Geologic CO₂ Storage Sites

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Units and Acronyms

Units

‰	Parts per mil (thousand)
Bbl	Barrel
Bcf	Billion cubic feet
°C	Degrees Celsius
°F	Degrees Fahrenheit
Ft	Feet
Gt	Giga tonnes
Lbs	Pounds
km	Kilometer
m	Meter
M	Molar
mD	Millidarcy
mW/m ²	Milliwatts per square meter
Mcf	Thousand cubic feet
Mi ²	Square miles
MMcfd	Million cubic feet equivalent per day
ppm	Parts per million
R _o	Vitrinite reflectance
t	Metric tonne
Tcf	Trillion cubic feet

Acronyms

ARI	Advanced Resources International, Inc.
CBM	Coalbed methane
CH ₄	Methane
CO ₂	Carbon dioxide
EOR	Enhanced oil recovery
GHG	Greenhouse gas
k	Permeability
SBIR	Small Business Innovation Research
TD	Total depth
USDOE	US Department of Energy

Executive Summary

This SBIR project evaluated the potential to remediate geologic CO₂ sequestration sites into useful methane gas fields by application of methanogenic bacteria. Such methanogens are present in a wide variety of natural environments, converting CO₂ into CH₄ under natural conditions. We conclude that the process is generally feasible to apply within many of the proposed CO₂ storage reservoir settings. However, extensive further basic R&D still is needed to define the precise species, environments, nutrient growth accelerants, and economics of the methanogenic process. Consequently, the study team does not recommend Phase III commercial application of the technology at this early phase.

Overall Concept. Methanogens, which belong to the Euryarchaeota kingdom of Archaea, obtain energy for growth by reducing carbon dioxide to methane with hydrogen gas or formate as an electron-donating reductant. CO₂-destroying methanogenesis takes place on a vast scale in a diversity of settings in the Earth's crust -- including those similar to proposed geologic sequestration sites -- generating billions of tons of methane annually. Consequently, natural processes support our overall concept of employing methanogens to remediate CO₂ sequestration sites.

Methanogen Screening. Of the 98 known species of methanogens that were evaluated, six were selected as the potentially the best candidates for CO₂-to-CH₄ conversion under conditions present in typical geologic sequestration sites. These include: *Methanosarcina vacuolata* and *barkerii*; *Methanococcus vanniellii*; and *Methanobacterium uliginosum*, *ivanovii*, and *subterraneum*. Screening factors included temperature, salinity levels and composition, and pH. Reservoir pressure, which is related to depth, does not interfere with methanogen growth; in fact, high pressure only accelerates methanogenic reactions.

Biochemistry. Methanogens employ two separate pathways to convert CO₂ into methane: either the methyl group of acetate or the reduction of carbon dioxide with electrons from hydrogen or formate. Acetate, carbon dioxide, hydrogen, and formate are the major substrates for the methanogenic group. The hydrogen-producing acetogenic group is necessary to further metabolize butyrate and propionate to substrates for the methanogenic group. Such "interspecies electron transfer" is fortunate, in that the hydrogen-producing and hydrogen-consuming groups are interdependent. We conclude that maintaining suitable biochemical conditions for methanogenesis

at CO₂ sequestration sites is feasible.

Potential Application. Our preliminary analysis of US reservoirs indicates that up to 84 Gt of CO₂ storage capacity has suitable environmental conditions and could be remediated using methanogen technology. This represents about 40 years of current power generation emissions. Of this total, perhaps 20 Gt of CO₂ storage capacity exists in high-graded, onshore, high-temperature settings with the best potential for methanogenic remediation. It appears generally feasible to engineer the application of methanogen technology to sequestration reservoirs based on existing petroleum reservoir exploitation and management technologies. For example, injection well drilling, completion and operation technologies developed for water-flooding and enhanced oil recovery, with limited modifications, could be applied to inject cultured methanogens and their chemical substrates. However, specific advanced horizontal drilling, field-positioned incubators, and batch mixing technology still needs to be developed.

St. Johns Dome Study Site. The St. Johns Dome is a large (1,800 km²) anticline located in eastern Arizona and western New Mexico. CO₂ is trapped within the Permian Supai Fm and overlain by evaporite deposits (anhydrite, gypsum) and shales which act as cap rocks. The field is relatively shallow, with depth to CO₂ ranging from about 300 to 750 m. It contains an estimated 730 million t (13.9 Tcf) of CO₂ in a free gas state. Reservoir architecture is complex, with multiple, vertically dispersed reservoirs consisting of sandstone, siltstone, and vuggy dolomite that are separated by thin, impermeable anhydrite seals. CO₂ concentration averages 92% along with 6.6% nitrogen, 0.2% argon, and 0.6% helium. CO₂ at St. Johns appears to be largely igneous in origin.

Ridgeway Petroleum Corp., the former field operator, drilled 21 CO₂ production wells since discovering the field in 1994. At present the field is shut in due to limited local market demand and lack of a CO₂ pipeline. The production wells were drilled with air or with a fresh water and starch-based mud to avoid formation damage and were completed in one or more of the three CO₂-bearing zones (Ft. Apache, Amos Wash, and Upper Abo/Granite Wash) at an average depth of 850 m. The wells were completed using amine carbon gauze fiberglass or conventional carbon steel casing lined with high-density polyethylene (HDPE) to minimize corrosion. The St. Johns wells were relatively inexpensive to drill and complete (\$300,000/well) compared with wells at much deeper CO₂ fields.

Our initial evaluation indicates that the geologic and reservoir conditions at St. Johns Dome

may be conducive to maintaining suitable biochemical conditions for methanogenesis of various species for the conversion of CO₂ into CH₄. The six methanogen species selected as the potentially best candidates for conversion of CO₂ into methane under conditions present at St. Johns Dome. The selection was based on the ability of these species to tolerate a variety of environmental conditions and their simplicity of requirements for growth and CO₂ reduction to methane. The most promising methanogen species are: *Methanosarcina vacuolata* and *barkerii*; *Methanococcus vanniellii*; and *Methanobacterium uliginosum*, *ivanovii*, and *subterraneum*.

Future Research. Future research into methanogen application for remediating CO₂ storage fields could focus on isolating individual species within the Methanobacteriaceae and Methanosarcinaceae families with greatest potential for application to CO₂ storage sites. Other topics include further laboratory experimentation with accelerating methanogen growth under varying temperature, pH, and nutrient conditions.

1.0 Introduction

1.1 Overview

Combustion of fossil fuels increases atmospheric CO₂ levels at such high rates that natural geologic sequestration processes may not be fast enough to avoid severe effects on the global climate. “Engineered” geologic sequestration in subsurface rock strata is one of the more promising methods for rapidly reducing the input of CO₂ into the atmosphere. The U.S. Department of Energy (USDOE) has committed substantial R&D funding to investigate and demonstrate such technologies.¹ The essential strategy is to separate CO₂ from anthropogenic sources and store it underground in geologic settings.

To sequester the large volumes of CO₂ needed to make meaningful reductions in greenhouse gas (GHG) emissions, the United States likely will need to rely heavily on depleted oil and gas fields and saline aquifers as sequestration sites. Deep coal seams also offer significant CO₂ sequestration potential, although CO₂ can cause swelling in coals, raising injectivity issues that do not exist with high-permeability depleted oil and gas fields.

Arguably, CO₂ sequestration should not be viewed as “waste disposal.” Naturally occurring CO₂ fields are common and relatively stable geologic phenomena, and in some cases have been exploited as low-cost sources of CO₂ for enhanced oil recovery and other industrial uses.² However, many environmental protection groups do not share this benign view of geologic sequestration. Without a strategy for removing or remediating long-term CO₂ storage sites, the innovative R&D funded by USDOE on geologic sequestration may never achieve the public acceptance needed for large-scale implementation by industry.

Geologic sequestration also has small but potentially significant inherent safety risks and disadvantages. Long-term geologic storage of CO₂ -- if implemented on a global scale would create hundreds of sites containing large volumes (perhaps 1,000 Gt) of CO₂ at high reservoir pressures. These sites would present a risk of leakage and require long-term monitoring.³

This USDOE Small Business Innovation Research (SBIR) project evaluated the potential for breakthrough biological technologies that could permanently transform mature geologic sequestration sites back into methane deposits. The study focused on naturally occurring bacterial

consortia that are known to convert CO₂ into CH₄ (“methanogens”). The long-term strategy of the research would be to replicate what nature has performed over long geologic time periods at innumerable natural gas fields, but at a much faster rate using engineered approaches. Such technology could help ensure public acceptance of geologic sequestration by providing USDOE with a strategy for ameliorating sequestration sites. In the process, it may also create new and readily exploitable methane gas deposits.

One of the most important CO₂ processes active in the earth’s crust is the bacterially mediated conversion of CO₂ into methane. This process has been documented in soils,⁴ lake and ocean sediments,^{5,6,7,8} and also in deeply buried sediments and commercial natural gas accumulations.^{9,10} In fact, some of the world’s largest natural gas deposits were entirely generated by bacterial conversion of CO₂. Examples are the Santa Barbara and related gas fields in the Po Valley Basin in northern Italy, which collectively amount to 10 trillion cubic feet (Tcf) of nearly pure methane,¹¹ as well as the world’s largest gas field (Urengoy in West Siberia) with 270 Tcf of pure methane likely of bacterial origin.¹²

Bacterial conversion of CO₂ to methane is one of the most effective CO₂ sequestration processes. It not only removes CO₂ from the atmosphere, it could also generate new methane deposits, thereby helping to meet the nation’s rapidly increasing demand for clean fuels. Most methanogens require only a simple solution of inorganic nutrients, CO₂, hydrogen (or formate) and the absence of oxygen. Methanogens can thrive under extreme conditions, such as total darkness and a broad range of pressures, temperatures, and other extreme environmental conditions. For example, CO₂-reducing methanogens can tolerate high salinities of more than 1.5 M NaCl. Yet, methanogenesis – particularly within geological formations -- remains poorly understood.

Advanced Resources International, Inc. (ARI), along with research partners Professor James G. Ferry of Pennsylvania State University and Dr. Martin Schoell of GasConsult, Inc., conducted this SBIR project. We evaluated the bacterial conversion of CO₂ to methane as an unconventional “breakthrough” strategy for disposing of CO₂. We also studied our industrial partner’s CO₂ field site in Arizona to evaluate the critical catalysts and pathways for methanogenesis.

1.2 Anticipated Public Benefits

Methanogen technology is a new field that offers several potentially high-reward benefits for

the USDOE-industry geologic sequestration program. First, we anticipate our project could help to demonstrate the potential for this unique biological technology to remediate future geologic CO₂ sequestration sites, addressing an environmental concern. The second potential benefit is to help provide the basis for an engineered process to convert sequestered CO₂ back into a valuable resource – methane. If achieved cost effectively, this could provide future generations with a clean resource from current CO₂ waste gases, while reducing current global warming potentials from point sources. It could provide both an economic incentive to sequester carbon dioxide as well as a “green” alternative to pure disposal.

In the commercial sector, methanogen technology could aid utilities and other point source CO₂ generators to meet potential emission restrictions. It also could help the U.S. Government to comply with international protocols and treaties on GHG emissions. Consider the potential: some current methane fields were sourced by conversion from carbon dioxide; many oil and gas fields have seals that have been reasonably effective for millions of years; and technology exists to inject into and produce gases from reservoirs. Industry must only identify candidate reservoirs and tailor bacteria to be efficient conversion catalysts. We believe that our early investigations in this area could help lay the foundation for such a breakthrough.

2.0 Methanogen Biochemistry

In this section, we summarize the key results from our biochemical studies of methanogenic consortia. The most significant result is that we have defined the physico-chemical conditions required to sustain methanogens. We also identified the most promising methanogen species for potential use in the geologic conversion of CO₂ into methane.

2.1 Ecology, Phylogeny, and Biochemistry of Methanogens

The first step of our study was to define the unique ecology, phylogeny, and biochemistry of methanogenic bacteria. This analysis provides the foundation for evaluating the feasibility methanogens to convert CO₂ sequestration sites into methane.

a) Ecology. The biological production of methane -- methanogenesis – is an important component of the global carbon cycle that is essential for life on earth. The cycle is divisible into

organisms requiring oxygen (aerobic) and organisms that function in the absence of oxygen (anaerobic). In the anaerobic world, atmospheric carbon dioxide is fixed by plant photosynthesis into cell carbon. Oxygen-requiring microorganisms (aerobes) decompose the photosynthetic organisms and others that consume them (organic matter) back into carbon dioxide.

However, a significant portion of this organic matter is deposited in environments where oxygen is unavailable (anaerobic world). Here, microbes metabolizing without oxygen (anaerobes) decompose the organic matter to methane. Environments where methanogenesis occurs are diverse and vast, with an estimated billions of tons of methane released annually. Recent evidence suggests that CO₂-reducing methanogens are abundant in the deep subsurface, leading to the proposal that the recent discovery of methane on Mars is biological in origin.

The process of methanogenesis requires a consortium of at least three interacting metabolic groups of obligatory anaerobic microbes (Figure 1). The fermentative group decomposes cellulose and other complex molecules to volatile carboxylic acids and hydrogen gas. Only acetate, carbon dioxide, hydrogen, and formate are the major substrates for the methanogenic group; thus, the hydrogen-producing acetogenic group is necessary to further metabolize butyrate and propionate to substrates for the methanogenic group.

The methanogenic group (methanogens) employs two separate pathways in which methane derives from either the methyl group of acetate (reaction 1) or the reduction of carbon dioxide with electrons from hydrogen or formate (reactions 2a and 2b). At least two-thirds of the total methane produced derives from the acetate and the remainder from the reduction of carbon dioxide.

The hydrogen-producing acetogenic group metabolizes a limited inventory of growth substrates and relies on the fermentatives to provide them, whereas the methanogens rely on both these groups to supply their growth substrates (Figure 1). The production of hydrogen gas is a thermodynamically unfavorable reaction under standard conditions of equimolar reactants and products; therefore, the thermodynamically favorable removal of hydrogen gas by the carbon dioxide

reducing methanogens is essential for growth of hydrogen-producing acetogenic microbes. This important principle of methanogenesis is termed "interspecies electron transfer".

Thus, the hydrogen-producing and hydrogen consuming groups are interdependent, and the process of methanogenesis is best described as involving a "consortium" rather than a "food chain" of metabolic groups. In most environments the rate-limiting step in methanogenesis is not the terminal step of methane formation but the steps involved in the production of acetate and hydrogen gas. The interspecies transfer of hydrogen is facilitated by the formation of microbial communities structured with hydrogen-producing species adjacent to hydrogen-consuming species in microbial flocs or aggregates.

Formate also has been proposed to replace hydrogen gas in interspecies electron transfer.¹³ Formate and hydrogen gas can be interconverted by formate hydrogen-lyase enzyme systems that are documented in species from the hydrogen-producing acetogenic group. Although formate and hydrogen gas are likely in equilibrium in most methanogenic habitats, several factors such as the bicarbonate concentration and pH, are likely to have an effect on this equilibrium. Thus, the relative contributions of hydrogen and formate to interspecies electron transfer are yet to be determined.

In marine sediments that contain upwards of 20-30 mM sulfate from seawater, sulfate reducing anaerobes replace methanogens and sulfide is the terminal product of the degradation of organic matter.¹⁴ Low rates of methanogenesis by reduction of carbon dioxide and conversion of acetate occur concurrently with sulfate reduction; however, the sulfate-reducing bacteria have a greater affinity for hydrogen gas and acetate than do the methanogens. Thus, much of the methane generated in marine environments originates from methylated amines that are prominent in estuarine and marine environments and by oxidation of hydrogen generated from geothermal sources.

Figure 1 : Anaerobic microbial food chain (consortium) converting complex organic matter to methane in freshwater environments. Solid lines represent the major routes of methanogenesis.

b) Phylogeny. Early investigations of methanogens identified unique ether lipids rather than typical glycerol ester lipids and cell wall components which are not found in all other procaryotes and eucaryotes which led to the present three-domain (Archaea, Bacteria, and Eucarya) concept of phylogeny based on 16S rRNA sequences. The Archaea are procaryotes composed of methanogens, extreme halophiles, and extreme thermophiles with other characteristics that distinguish them from all other procaryotic (Bacteria domain) organisms.

The Archaea comprise four kingdoms (Euryarchaeota, Crenarchaeota, Korarchaeota, and Nanoarchaeota). The methanogens are classified in the Euryarchaeota and are further subdivided

into five orders. With very few exceptions, strains in the orders Methanobacteriales, Methanomicrobiales, Methanococcales, and Methanopyrales obtain energy for growth by reducing carbon dioxide to methane with hydrogen gas or formate. Figure 2 illustrates an updated methanogen phylogeny.

The Methanobacteriales contains the family Methanothermaceae which is distinguished from other members of this order by their high temperature optima for growth of between 83 and 88°C. *Methanothermobacter thermoautotrophicus* (*Methanobacterium thermoautotrophicum*) in the family Methanobacteriaceae is one of the most intently studied of all the methanoarchaea, and for which the genome has been sequenced. The Methanococcales contain coccoid marine species, all of which are slightly halophilic and most chemolithotrophic. Most strains in this order are mesophilic; however, a few thermophilic species are represented such as *Methanococcus jannaschii* for which the genome is sequenced. Some species in the order Methanomicrobiales reduce carbon dioxide using alcohols. Nearly all species of Methanomicrobiales require acetate as a source of cell carbon and have complex nutritional requirements. The order Methanopyrales contains a single rod-shaped species, *Methanopyrus kandlerii*, growing at or above the boiling point of water.

Figure 2 : Methanogen Phylogeny¹⁵

Finally, the order Methanosarcinales comprises two families Methano-sarcinaceae and Methanosaetaceae, which are the only families of methanogens able to utilize acetate as an energy source which accounts for most of the methane produced in nature. Members of the Methanosarcinaceae are the most metabolically versatile of all methanogens with respect to carbon and energy sources. Certain species of this family are able to utilize at least seven substrates for carbon and energy generation. This diversity is reflected in the size of the genome of *Methanosarcina acetivorans* and *Methanosarcina mazei*, which is more than twice the size of the genomes of *M. thermoautotrophicus* and *M. jannaschii*.

c) Biochemistry of the reduction of carbon dioxide to methane. The steps shown in Figure 3 are common to the two major pathways for methane production in nature, carbon dioxide reduction and fermentation of acetate. The two pathways differ primarily in the manner by which a methyl group is generated and passed to the co-factor tetrahydromethanopterin (H_4MPT) to form $\text{CH}_3\text{-H}_4\text{MPT}$. Conversion of $\text{CH}_3\text{-H}_4\text{MPT}$ to methane is essentially the same in both pathways. Step 1 in Figure 3 is catalyzed by N^5 -methyltetrahydromethanopterin:coenzyme M methyltransferase, which is an integral membrane-bound complex that also functions to generate a sodium ion gradient across the membrane coupled to the methyl transfer reaction. The enzyme as characterized from *Methanothermobacter marburgensis* and *Methanosarcina mazei*, contains a corrinoid cofactor, cobalamine (5'-hydroxybenzimidazolyl cobamide), which accepts the methyl group from $\text{CH}_3\text{-H}_4\text{MPT}$ in the first of two partial reactions catalyzed by the enzyme. The second partial reaction involves transfer of the methyl group from $\text{CH}_3\text{-cobalamine}$ to coenzyme M (HS-CoM) producing $\text{CH}_3\text{-S-CoM}$.

Figure 3 : Carbon dioxide reduction and fermentation of acetate, steps common to all pathways for methanogenesis.

The methyl-coenzyme M reductase catalyzes step 2 in Figure 3. The electron donor is coenzyme B (CoB) and the heterodisulfide CoM-S-S-CoB is produced in addition to methane. Heterodisulfide reductase catalyzes the final step in Figure 3, releasing the active sulfhydryl forms of the cofactors. Heterodisulfide reduction is coupled to formation of an electrochemical proton gradient that drives ATP synthesis catalyzed by an A₁A₀-type ATP synthase.

For most CO₂-reducing species, the six electrons needed for carbon dioxide reduction to CH₃-H₄MPT derive from the oxidation of either hydrogen gas or formate as follows:

Reaction 3, catalyzed by the Ech hydrogenase, is endergonic given the low partial pressures of hydrogen gas in the native environment; however, it is proposed that the Ech hydrogenase utilizes the electrochemical proton gradient to drive the reaction. Coenzyme F₄₂₀-dependent hydrogenases (reaction 4) and a formate dehydrogenase (reaction 5) have been characterized (20). Coenzyme F₄₂₀ is an obligate two-electron carrier involving hydride transfer. Six reactions are required for reduction of carbon dioxide to CH₃-H₄MPT (reactions 6-10).

In the first reaction (reaction 6) carbon dioxide is attached to methanofuran (MF) as the carbamate (5) and reduced to the formyl level by formylmethanofuran dehydrogenase with ferredoxin as the electron donor. Reaction 7 is catalyzed by formylmethano-furan:tetrahydromethanopterin formyltransferase. Reaction 8 is catalyzed by N⁵,N¹⁰-methenyltetrahydromethanopterin

cyclohydrolase comprised of two identical subunits with no prosthetic groups. Two mechanistically distinct *N*⁵,*N*¹⁰-methylenetetra-hydromethanopterin dehydrogenases catalyze reaction 9, one that oxidizes reduced coenzyme F₄₂₀ (reaction 9a) and the other that oxidizes H₂ (reaction 9b). The F₄₂₀-dependent enzyme is composed of one type of subunit, either as a hexamer or octamer, with no detectable prosthetic group. The other dehydrogenase is essentially a hydrogenase that contains iron in an unknown configuration and a cofactor of unknown composition.

Carbonic anhydrase catalyzing reaction 11 is also present in CO₂-reducing species:

where it is likely to be essential for methanogenesis and growth, particularly chemolithoautotrophic species. These microbes have a high demand for CO₂ in both catabolic and anabolic reactions, suggesting that carbonic anhydrase may be essential to deliver CO₂ to the cell and concentrate it in the vicinity of CO₂-utilizing enzymes. Recently, a gamma class of carbonic anhydrase from a methane-producing species was shown to utilize iron in the active site.

2.2 Screening Methanogen Species for Sequestration Conversion

The next step was to conduct an exhaustive review of methanogen consortia to provide the basis for identifying the most promising species for further study. We constructed a searchable data base of the 98 species described at the time of the study. The data base features nutritional and environmental factors that influence the growth of each species in subterranean environments likely to be targeted for CO₂ storage. These methanogen groups are described below, while their growth requirements are discussed in Section 2.3.

a) *Methanobacteriaceae*. With thirty species in four genera, *Methanobacteriaceae* is the largest and most diverse families of methanogenic *Archaea*. Cells range from coccoid, to filamentous rods, with most species being coccobacilliary or short rods. The cell wall structure is very similar to that of Gram-positive bacteria, except that pseudomurein replaces muramic acid as the predominant peptidoglycan polymer.

b) *Methanothermaceae*. The family *Methanothermaceae* contains but a single genus,

Methanothermus, which comprises only two species. Cells are rod-shaped and motile by bipolar flagellar tufts. The cellular envelope is doubly layered, consisting of an inner pseudomurein layer similar to that of the family *Methanobacteriaceae*, and an outer S-layer. *Methanothermus fervidus* cells are straight rods that occur in pairs or short chains. *Methanothermus sociabilis* cells are curved rods growing in large aggregates.

c) *Methanococcaceae* is another small family of methanogens containing only two genera and five species. Cells are irregular cocci, occurring singly and in pairs, motile by a polar tuft of flagella, and form a protein cell wall or S-layer. The family *Methanocaldococcaceae* comprises only two genera and six species, differentiated primarily by their optimum growth temperatures. Like the *Methanococcaceae*, cells are motile, irregular cocci occurring singly or in pairs.

d) *Methanocaldococcaceae* comprises two genera and six species, differentiated primarily by their optimum growth temperatures. Cells are motile, irregular cocci occurring singly or in pairs.

e) *Methanomicrobriaceae* is one of the larger and more diverse families of methanogens, with 23 species in seven genera. Cell morphology ranges from cocci to short rods to the disc- or plate-shaped cells of the genus *Methanoplanus*. Cell walls are made of proteins and do not contain either peptidoglycan or pseudomurein.

f) *Methanocorpusculum* is the lone genus containing four species that comprise the family *Methanocorpusculaceae*. Cells are irregular cocci and occur singly or in aggregates. Cell walls are an S-layer of hexagonally arranged units, and all species are motile by flagella (apart from *Methanocorpusculum labreanum*).

g) *Methanospirillaceae* is one of two families of methanogens that comprises only a single genus and species. *Methanospirillum hungatei* cells are symmetrically curved rods that form wavy filaments. The cells are motile by polar tufts of flagella. The cell wall consists of an S-layer of non-glycosylated polypeptides, and is enveloped by a rigid paracrystalline outer sheath that may encase several cells.

h) *Methanosarcinaceae* is a large family of methanogens comprising eight genera and 24 species. Cells are coccoidal or pseudosarcinal forming a protein cell wall devoid of peptidoglycan or pseudomurein, and in some cases are surrounded by a heteropolysaccharide sheath to form large

aggregates.

i) ***Methanosaetaceae***, comprising only two species within a single genus, are nonmotile, straight rods that grow in chains and sometimes long filaments. The cell envelope, unique among *Methanosaeta* species, is enclosed in a tubular sheath and separated by structures called spacer plugs.

j) ***Methanopyraceae***, the final family of methanogenic *Archaea*, consists of but a single genus and species. The cells of *Methanopyrus kandleri* are motile rods that grow singly or in chains. The cell wall consists of pseudomurein and cells are also sheathed by a “fuzzy” coat of unknown composition.

Of the 98 species evaluated, six were selected as the potentially the best candidates for conversion of CO₂ into methane under conditions present in typical sequestration sites. The selection was based on the ability of these species to tolerate a variety of environmental conditions and their simplicity of requirements for growth and CO₂ reduction to methane.

Methanosarcina vacuolata

Methanosarcina barkerii

Methanococcus vanniellii

Methanobacterium uliginosum

Methanobacterium ivanovii

Methanobacterium subterraneum

2.3 Nutritional/Environmental Controls on Methanogen Growth & Metabolism

The next study task involved defining the nutritional and environmental controls on methanogen growth and metabolism. These factors are discussed below for the species previously identified and described in Section 2.2.

a) ***Methanobacteriaceae*** produce energy by reducing CO₂ with H₂, except for the species of the genus *Methanosphaera*, which produce methanol with H₂. Many species also use formate or secondary alcohols to reduce CO₂. Most species of the genera *Methanobacterium* and *Methanothermobacter* are autotrophic, utilizing ammonium (some species are also capable of fixing dinitrogen) and sulfide as nitrogen and sulfur sources, and most also require trace elements such as tungsten and nickel. Species of *Methanosphaera* and *Methanobrevibacter* are heterotrophic,

requiring acetate and sometimes vitamins for growth. Most *Methanobacteriaceae* species can be considered enteric organisms, as they have been isolated from the digestive tracts and feces of animals, as well as sewage sludge digesters. All species are mesophilic except those of the genus *Methanothermobacter*, which are thermophilic. Nearly all species are neutrophilic, having an optimum pH of 6.8 to 8.0. However, there are two acidophilic species of *Methanobrevibacter* and two alkalophilic species of *Methanobacterium*. Of particular interest is *Methanobacterium subterraneum*, which is not only alkalophilic, but also the only species which is halo-tolerant, forms aggregates, and of all known methanogens, the only one to be isolated from granitic ground water.

b) *Methanothermaceae*. *Methanothermus sociabilis* and *M. fervidus* produce energy solely by reducing CO₂ to methane using hydrogen gas. Both organisms are autotrophic, but organic supplements such as yeast extract may stimulate growth. Both species were isolated from solfataric water sources, are neutrophilic, and (as the name implies) are hyperthermophilic with temperature optima of 83 to 88°C.

c) *Methanococcaceae* reduce CO₂ to methane through use of H₂ gas and formate as electron donors. All species are autotrophic, with the exception of *M. voltae*, which requires acetate and amino acids. Species of the genus *Methanococcus* can utilize sulfide and elemental sulfur as their sulfur sources and ammonium as their nitrogen source. Furthermore, *M. maripaludis* is also capable of fixing nitrogen gas. And the impressive species *Methanothermococcus thermolithotrophicus* can grow on nearly any sulfur and nitrogen source including: sulfide, elemental sulfur, thiosulfate, sulfite, sulfate, ammonium nitrate, and nitrogen gas. All species of *Methanococcaceae* were isolated from marine or estuarine environments with the *Methanothermococcus* found in geothermally heated sea sediments or deep sea hydrothermal vents. Despite these habitats, most species are only halotolerant, preferring to grow at lower salinities.

d) *Methanocaldococcaceae* likewise utilize CO₂ as the sole carbon and energy source, but they are only able to reduce it with hydrogen gas -- not formate. Ammonium and sulfide serve as the nitrogen and sulfur sources. Members of the genus *Methanocaldococcus* also require selenium. All species of *Methanocaldococcaceae* were isolated from deep-sea hydrothermal vents, are thus hyperthermophiles, with all species exhibiting growth temperature optima of 80 to 88°C. These organisms are slightly acidophilic and prefer salt concentrations near seawater levels (35,000 ppm). The second genus, *Methanotorris*, contains only a single species that is separated from all other *Methanocaldococcaceae* by the absence of both motility and the requirement for selenium, and by

being significantly more halotolerant (NaCl range: 0.45% to 7.2%) than other species.

e) ***Methanomicrobiaceae*** reduce CO₂ to form methane using hydrogen gas and formate; many species are also capable of using secondary alcohols. Nearly all species are heterotrophic, with acetate being the most common nutritional requirement. *Methanomicrobiaceae* have been isolated from a wide range of anaerobic habitats and the diversity of their physiological optima reflect this. While most species are mesophiles, two are thermophilic: *Methanoculleus thermophilicus* isolated from the effluent channel sediment of a nuclear power plant, and *Methanogenium frittonii* isolated from lake sediment. There is also a single psychrotrophic species, *Methanogenium marinum*, isolated from a lake in Antarctica, and a single psychrophilic species, *Methanogenium frigidum*, isolated from sediments in Alaska. *M. frigidum* is also one of two halophilic organisms, the other being *Methanocalculus halotolerans*, which was isolated from an oil field well head. There are also three halotolerant organisms: *Methanoculleus submarinus* from deep sea sediments in the Nankai Trough, *Methanofollis tationis* from a sulfataric field, and *Methanocalculus chunghsingensis* from a marine water aquaculture fish pond.

f) ***Methanocorpusculum*** grows by reducing CO₂ into methane using H₂, formate and (in *M. parvum* and *M. bavaricum*) secondary alcohols as reductant. All species are organotrophic, requiring acetate, peptones, or rumen fluid to serve as carbon and nitrogen sources. All species are mesophilic and neutrophilic. The type species *M. parvum* was isolated from an anaerobic sour whey digester inoculated with sewage sludge. *M. bavaricum* and *M. sinense* were isolated from the industrial waste water from a sludge facility and distillery, respectively, while *M. labreanum* was isolated from surface sediments at the La Brea Tar Pits.

g) ***Methanospirillaceae*** uses hydrogen gas and formate as electron donors for reducing CO₂ to methane. *M. hungatei* fixes N₂ and autotrophic growth is possible, with yeast extract and peptones to stimulate growth. It is mesophilic with temperature optimum of 37°C and neutrophilic with a very narrow pH range for growth (6.6 to 7.4). *M. hungatei* was isolated from sewage sludge, while a recently discovered strain (TM20-1) appears to be a new species isolated from a rice paddy field.

h) ***Methanosarcinaceae*** grow and produce methane by dismutating methyl compounds such as methanol, methyl amines, and methyl sulfides. Acetate, CO₂, and CO also can be utilized by some species. Ammonium and sulfide serve as the major nitrogen and sulfur sources, with many

species being autotrophic; however, cofactor supplements such as biotin are required by some. Species of *Methanosarcinaceae* have been isolated from very diverse anaerobic habitats and exhibit a wide range of physiological optima. Most species are mesophilic. However, *Methanococcoides thermophila*, which was isolated from a 55°C anaerobic digester, is thermophilic. Conversely, *Methanococcoides burtonii* (Ace Lake, Antarctica) and *Methanosarcina baltica* (Baltic Sea) are psychotrophic. Since many species have been isolated from marine environments, they prefer salinity near that of sea water. But *Methanohalobium evestigatum* (saline lagoon) is an extreme halophile which grows only at extreme NaCl concentrations higher than 15.2%, with an optimum of 25.1%.

i) *Methanosaetaceae* uses acetate solely as its substrate for methanogenesis. Ammonium and sulfide provide its nitrogen and sulfur sources. Both of the species in this genus require cofactor supplements in the form of biotin or vitamins. *Methanosaeta concilii* (from anaerobic thermophilic bioreactor sludge) is thermophilic with an optimum temperature of 55°C, slightly acidophilic with a pH optimum of 6.0, and grows better in the presence of trace metals.

j) *Methanopyraceae* form energy and methane by reducing CO₂ with hydrogen gas. It is autotrophic with ammonium and sulfide serving as nitrogen and sulfur sources. *M. kanderi*, which was isolated from hydrothermally heated deep sea sediment, is a hyperthermophile with optimum temperature of 98°C. It has a pH optimum of 6.5 and an optimum NaCl concentration of 2.0%. Its complete genome has been sequenced, demonstrating that it is a unique family with affinities for the *Methanococcales* and *Methanobacteriales* groups.

2.4 Electronic Database of Methanogen Species

Penn State constructed a searchable database of the characteristics for the 98 methanogenic species published as of the study date. These data consist of each species' genus, type strain, family, order, morphology, cell arrangement, cell size, motility, methanogenesis substrates, isolation location, and the references needed to determine this data. Whenever possible or necessary, the mol % G + C, Gram reaction, temperature range, temperature optimum, pH range, pH optimum, [NaCl] range, [NaCl] optimum, nutritional requirements, minimum generation time, and former classifications, were also reported, as well as any other information pertinent to the growth of the organism. The program Access is a database program that allows for the quick referencing of files.

After the file is opened, the data can be shown in a number of ways, the simplest being a table. Under the table button on the left hand side, the constructed table can be opened by clicking on Methanogen Review. Once the table is opened, records can be sorted or filtered. By right clicking on any column header, you can sort the column ascending (alphabetic from A-Z in the case of text or from lowest to highest in the case of numbers) or descending (alphabetic from Z-A in the case of text or from highest to lowest in the case of numbers). There are two ways to filter records, by selection, and by form. By selection shows only those records with the same value as the highlighted cell in the table. Filtering by form allows you to not only pick a specific value but also to set ranges that you want to view.

For example, if you only want to view species of the genus *Methanosarcina*, then simply highlight the genus cell of any *Methanosarcina* species and click the filter by selection button on the toolbar (filter with a lightening bolt). This should give you nine records, each corresponding to a single organism. Now if you only want to view thermophilic organisms of the genus *Methanosarcina* then you will have to filter by form so click on the filter by form button (filter with a table). First under the genus column, select *Methanosarcina* from the pull down menu if it isn't already selected. Second, under the optimum temperature column, type in ">49". This will give you the only thermophilic species of *Methanosarcina*, *Methanosarcina thermophila*. All filters can be removed at any time by clicking the remove filters button on the toolbar (just a filter). Each column can be sorted or filtered in the same fashion.

Because Access tables can often be too large to print out (as is the case for this table), it allows for a variety of ways for reporting the data that is more visually appealing. The first is the query which simply allows you choose which columns of the table you wish to view and omits all the remaining data. The second is called the form, which places all the data for a single record in one column. The database is formatted to show two records per page. The last is the report, which arranges all the data for a single record to fit on one page; however, this often leaves large amounts of blank space. The tables can also be exported to Excel by clicking export under the file menu, and then selecting one of the Excel versions under the "save as type" pull down menu.

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3.0 Screening USA Reservoirs for Application of Methanogens

3.1 Introduction

The next task involved screening USA reservoirs to identify those with generally appropriate conditions that could be suitable for the methanogenic conversion of CO₂ at sequestration sites that would host these organisms.

The applicability of methanogen technology to the universe of U.S. oil and gas reservoirs has not previously been assessed. This is a daunting task that could consume many man-years of effort. Given budget limitations, we modified a useful (but simplified) global data base of CO₂ sequestration potential that ARI previously developed for a study conducted for the IEA Greenhouse Gas R&D Programme.¹⁶

3.2 Methodology

ARI's petroleum province data base started with estimates of ultimate oil and gas recoverable resources by individual area, which had been developed by the U.S. Geological Survey during the past decade.¹⁷ We then conducted a literature review and compiled estimates of average reservoir depth, oil gravity (viscosity), and temperature, none of which are in the USGS data base. Obviously, actual field conditions vary widely within each petroleum province; our numbers are meant to reflect average conditions.

Using the petroleum province data base, we estimated volumetric storage capacities for CO₂ within each petroleum province, assuming the ultimate petroleum resource is discovered, developed and produced. Our estimate is therefore inclusive; some of this potential would only become available after decades of development. For oil reservoirs, our estimate assumed a volumetric ratio of 6 Mcf CO₂ per barrel of enhanced oil recovery (EOR) for miscible enhanced oil recovery (comparable to actual Permian basin ratios) and a 10:1 ratio for immiscible basins (i.e., shallower, low-gravity oil reservoirs).

For natural gas reservoirs, we estimated CO₂ sequestration capacity based on empirical CO₂/CH₄ ratios. These vary widely with depth from about 1.4 Mcf CO₂ : 1.0 Mcf CH₄ at a depth of

4,000 m to a 3.1: 1.0 ratio at a depth of 1,000 m, due to the varying compressibility (z-factors) of CO₂ vis-à-vis CH₄. For both oil and gas reservoirs, we conservatively assumed that 25% of the storage capacity simply could not be filled with CO₂, due to reservoir complexity, field edge effects, or other factors.

3.3 Results

The results of our screening analysis are summarized in Table 1.

Table 1 : Analysis of CO₂ sequestration potential in US petroleum basins

a	B	C	D	E	F	g	H	i	j	K	l	m
	USA Petroleum Province	Location	Ultimate Recovable Resources			Reservoir Properties				Sequestration		Methanogen Potential
			Oil (BB)	Gas (Tcf)	Total (BBOE)	On/ Off Shore	Avg. Depth (m)	Oil Gravity (API)	Temp deg F	Potential CO ₂ @75% Vol (Tcf)	Vol (Gt)	
1	Anadarko Basin	Central	5.4	107.0	23.2	On	2500	35	173	134.7	7.1	High
2	Permian Basin	Southwest	40.9	109.0	59.1	On	2800	35	188	131.1	6.9	High
3	Louisiana-Miss. Salt	Southeast	9.6	57.5	19.2	On	2500	35	173	72.4	3.8	High
4	East Texas Basin	Southeast	12.0	49.5	20.3	On	2300	35	163	65.8	3.5	High
5	SW Wyoming	W-Central	1.3	30.3	6.4	On	3000	40	198	35.2	1.9	High
6	Arkoma Basin	Central	1.0	17.4	3.9	On	2500	35	173	21.9	1.2	High
7	Rocky Mtn Def. Belt	W-Central	0.6	13.8	2.9	On	2500	35	173	17.4	0.9	High
8	Bend Arch-Fort Worth	S-Central	6.2	13.5	8.5	On	2500	35	173	17.0	0.9	High
9	Wyoming Thrust Belt	W-Central	1.5	14.5	3.9	On	3000	40	198	16.9	0.9	High
10	Sacramento Basin	West	0.2	12.0	2.2	On	2200	40	158	16.5	0.9	High
11	Wind River Basin	N-Central	0.7	4.3	1.5	On	3000	40	198	5.0	0.3	High
12	Southern Oklahoma	Central	3.5	3.7	4.1	On	2500	35	173	4.6	0.2	High
13	Nemaha Uplift	Central	3.1	3.2	3.7	On	2500	30	173	4.1	0.2	High
14	Sedgwick Basin	Central	1.0	2.2	1.4	On	2200	32	158	3.0	0.2	High
15	Big Horn Basin	N-Central	3.1	2.2	3.5	On	3000	40	198	2.6	0.1	High
16	Cherokee Platform	Central	2.3	1.6	2.6	On	2200	32	158	2.1	0.1	High
	Subtotal High Potential		92.6	441.8	166.2					550.3	29.1	High
17	N. Alaska	Northwest	20.5	83.0	34.3	On	2000	25	148	120.1	6.4	Mod
18	Palo Duro Basin	S-Central	4.0	48.4	12.1	On	1800	35	139	74.6	3.9	Mod
19	San Juan Basin	Southwest	1.7	40.0	8.4	On	2000	30	148	57.9	3.1	Mod
20	Ventura Basin	Southwest	4.5	10.4	6.3	On	1600	25	129	17.3	0.9	Mod
21	Michigan Basin	N-Central	2.2	10.4	3.9	On	2000	38	148	15.0	0.8	Mod
22	Los Angeles Basin	Southwest	9.9	8.5	11.3	On	1500	30	124	14.7	0.8	Mod
23	Uinta-Piceance	W-Central	1.9	6.7	3.0	On	1600	30	129	11.1	0.6	Mod
24	Denver Basin	W-Central	1.4	5.0	2.3	On	1800	35	139	7.7	0.4	Mod
25	Williston Basin (US)	N-Central	2.9	3.0	3.4	On	2000	35	148	4.3	0.2	Mod
26	Santa Maria Basin	Southwest	1.8	2.1	2.1	On	1500	23	124	3.7	0.2	Mod
27	Cambridge Arch	Central	3.2	0.9	3.3	On	2000	30	148	1.3	0.1	Mod
28	Illinois Basin	E-Central	4.3	0.7	4.4	On	1500	25	124	1.2	0.1	Mod
	Subtotal Mod. Potential		58.3	219.1	94.8					328.9	17.4	Mod
29	Western Gulf	S-Central	48.7	254.1	91.1	On/Off	2200	35	158	347.9	18.4	Low
30	Gulf Cenozoic OCS	S-Central	12.2	232.3	50.9	Off	2300	40	163	308.8	16.3	Low
31	San Joaquin Basin	Southwest	15.7	15.0	18.2	On	1200	20	109	30.2	1.6	Low

32	Southern Alaska	Northwest	2.2	9.5	3.8	On/Off	1200	40	109	19.1	1.0	Low
33	Powder River Basin	N-Central	4.8	4.8	5.6	On	1300	20	114	9.4	0.5	Low
	Subtotal Low Potential		83.5	515.7	169.5					715.4	37.8	Low
	TOTAL		234	1177	431					1595	84	All

a = ranking b = petroleum province c = location in USA d = estimated ultimate recoverable oil resources (billions of barrels) e = est ultimate natural gas resources (trillion cubic feet) f = total oil & gas (billion barrels) g = location (onshore/offshore) h = average depth (m) i = average oil gravity (°API) j = average temperature (°F) k = estimated ultimate CO₂ sequestration potential (Tcf) l = (billion t)

USA Potential Application. Our preliminary analysis of US reservoirs indicates that up to 84 Gt of CO₂ storage capacity has suitable environmental conditions and could be remediated using methanogen technology. This represents about 40 years of current power generation emissions. Of this total, perhaps 20 Gt of CO₂ storage capacity exists in high-graded, onshore, high-temperature settings with the best potential for methanogenic remediation. It appears generally feasible to engineer the application of methanogen technology to sequestration reservoirs based on existing petroleum reservoir exploitation and management technologies. For example, injection well drilling, completion and operation technologies developed for waterflooding and enhanced oil recovery, with limited modifications, could be applied to inject cultured methanogens and their chemical substrates. However, specific advanced horizontal drilling, field-positioned incubators, and batch mixing technology still needs to be developed.

The next step was to estimate the potential for applying methanogen technology in each petroleum province. We first screened out offshore fields as low potential, on the basis that offshore fields generally are too costly to support EOR due to the large numbers of injection wells required (note: this could change in future with horizontal drilling, but offshore EOR remains extremely rare and high-cost).

Next we grouped the petroleum provinces by reservoir temperature, an important environmental control on methanogens. None of the provinces exceeded the maximum temperature for methanogen growth, thus all are technically feasible locations. However, methanogenic reactions take place much faster at higher temperatures. We grouped the provinces into three temperature categories: less than 120°F, 120-150°F, and hotter than 150°F.

Based on location (onshore vs. offshore) and reservoir temperature, we estimated that 29.1 Gt of geologic CO₂ storage capacity would have “high” potential for remediation using methanogen

technology. Another 17.4 Gt of CO₂ storage capacity exists in provinces with “moderate” methanogenic potential. Finally, some 37.8 Gt of capacity is characterized as “low” potential for methanogenic remediation. Together, the total 84 Gt of CO₂ storage potential is of national significance, representing approximately 40 years of total power plant emissions in the USA at current emission rates. (Large additional potential for CO₂ storage also exists in saline aquifers, but was not assessed here because methanogen technology is likely to be restricted initially to well-defined and well-contained oil and gas fields where growth may be modulated.)

Technology for applying methanogens at geologic sequestration sites is likely to be adapted from commercial injection technologies developed for waterflooding and enhanced oil recovery at mature oil fields. Figure 4 shows a conceptual methanogen injection scheme, where methanogen cultures, nutrients, and co-factors are injected into the CO₂ storage reservoir using low-cost water-injection wells. Advanced horizontal pinnate drilling is becoming more cost-effective and could be utilized to better distribute agents, particularly in low-permeability reservoirs (Figure 5).¹⁸

Figure 4 : Injection strategies for methanogen remediation of CO₂ sequestration sites.

Figure 5 : Horizontal wells could more effectively distribute methanogenic agents.

One of the key challenges for successfully engineering methanogen conversion will be identifying the “sweet spots” in a reservoir where geologic and hydrologic conditions will favor methanogenesis. We expect the hydrology of sequestration reservoirs will be a key control on methanogen activity: bacterial CO₂ conversion is most intense where fresh water mixes with basinal brines.

4.0 St. Johns CO₂ Dome Field Site

4.1 Introduction

Industrial partner Ridgeway Petroleum Corp. provided access to and information on the St. Johns CO₂ Dome field site for this project. St. Johns Dome is a naturally occurring carbon dioxide field in eastern Arizona and western New Mexico containing approximately 13 Tcf of high-purity (mostly >90%) CO₂ within clastic and fractured reservoirs about 500 m deep.

Figure 6 shows the location of the St. Johns field and its CO₂ production wells. Two large coal-fired power plants are located close to the St. Johns field. The Tucson Electric Power Co. 760-

MW Springerville generating station is located near the center of St. Johns field. TEP recently constructed an 800-MW expansion to bring total generating capacity to 1,560 MW. Also, north of St. Johns field, the Salt River Project operates the 780-MW Coronado generating station, which started up in 1979. Ridgeway has discussed potential joint geologic sequestration projects with the owners of these power facilities, including the possibility of injecting flue gas CO₂ into the St. Johns reservoirs. However, currently there are no firm plans for such a project.

St. Johns Dome is a recently discovered field, thus the reservoir is relatively unaffected by development and remains largely in its original pristine condition. Gas composition is relatively simple (mostly CO₂ with minor impurities). Reservoir biochemistry probably is not overly complex, as it might be in a depleted oil field. ARI recently evaluated the St. Johns site as part of a separate project supported by USDOE to evaluate Natural Analogs for Geologic Sequestration.¹⁹ The data base – which contains all of the geophysical logs, core descriptions and analyses, well completion configurations, and geochemical data available for the field – greatly facilitated our methanogen study.

4.2 Field Description

St. Johns Dome is a large (1,800 km²) anticline located on the southern Colorado Plateau. CO₂ is trapped within the Permian Supai Fm and overlain by evaporite deposits (anhydrite, gypsum) and shales which act as cap rocks. The field is relatively shallow, with depth to CO₂ ranging from about 300 to 750 m. It contains an estimated 730 million t (13.9 Tcf) of CO₂ which, due to its relatively shallow depth averaging about 500 m, is stored in a free gas state. Reservoir architecture is complex, with multiple, vertically dispersed reservoirs consisting of sandstone, siltstone, and vuggy dolomite (porosity 11-20%, permeability 0.5->100 mD) that are separated by thin, impermeable anhydrite seals (k<0.01 mD).

The CO₂ concentration at St. Johns varies from 83-99%, averaging 92%. Other constituents include nitrogen (6.6%), argon (0.2%), and commercially significant quantities of helium (0.6%). Carbon isotope (delta¹³CO₂) values were uniform at three wells across the field (-3.8‰), suggesting that the CO₂ was generated from a single or well-mixed multiple source and that internal barriers and compartmentalization are minimal. Heat flow is relatively high (>100 mW/m²) over the St. Johns field, suggesting convective flow. The CO₂ origin at St. Johns appears to be largely igneous.

Figure 6 : Surface elevation and well location map for the St. Johns Dome

Figure 7 : Sample composite log from St. Johns Dome

The cap rock at St. Johns Dome consists of multiple but thin, mainly anhydrite rocks. Although a reasonably good seal for preserving commercial quantities of CO₂ and helium, CO₂ is also present in the overlying Permian Glorieta Sandstone and San Andres Limestone. This CO₂ may have entered these formations by gradual seepage through the cap rock matrix or by overspill and lateral migration, or along fault planes. On the other hand, the presence of helium -- a light and small molecule particularly prone to leakage -- in high concentrations (up to 1.1%) indicates that the cap rock is sealing over the north flank of the field.

4.3 Regional Geology

Regionally, the St. Johns Dome region is located on the southern edge of the Colorado Plateau, in the Transition Zone with the Basin and Range and Rio Grande Rift tectonic provinces. St. Johns lies on the southeastern Mogollon Slope, which comprises Precambrian crystalline rocks, sedimentary rocks of Paleozoic, Mesozoic, and Cenozoic age, and Cenozoic volcanic rocks.²⁰ The entire sequence was uplifted several thousand meters during Tertiary time, accompanied by minor northward tilting, as indicated by the presence of Cretaceous marine strata above 2,000 m elevation.

Structurally, the Defiance Uplift is a prominent and long-lived structural high that influenced deposition into the Mogollan Slope (where St. Johns is located) during Paleozoic and later time. Cambrian, Devonian, and Mississippian strata that are present over most of the Colorado Plateau wedge out in the St. Johns area; either they were never deposited or were eroded.

The surface geology at the St. Johns Dome reflects the Colorado Plateau – Rio Grande Rift transition. The oldest rocks exposed at the surface are Triassic Moenkopi and Chinle Fms and Cretaceous sedimentary rocks to the north of the field, representing the Mogollon Slope on the southern edge of the Colorado Plateau. Tertiary intrusive igneous rocks occur south of St. Johns Dome in the White Mountains. Extensive Tertiary volcanic related to the Springerville Volcanic Field as well as Tertiary sedimentary rocks occur throughout the field area.

The main CO₂-bearing strata are within the Permian Supai Formation, although CO₂ also occurs locally in the overlying Permian Glorieta Sandstone and San Andres Limestone as well as the Cretaceous Moenkopi Fm. The Supai Fm ranges from 425 to 580 m (1,400 to 1,900 ft) thick at St.

Johns Dome and rests directly on Precambrian basement. It consists primarily of siltstone and sandstone, with local carbonate and evaporite beds that may act as seals to vertical CO₂ migration. The Supai formed in a coastal desert eolian to sabkha depositional setting.²¹

The St. Johns field is defined by a broad, asymmetrical anticline that trends northwest, parallel to regional tectonic trends. Maximum vertical closure amounts to about 250 m. Structural dip is almost imperceptible, only 0.5 degrees across the northeastern flank; this partly explains why such a large anticline went untested until 1994. A major northwest-trending, high-angle reverse fault (referred here as the “St. Johns Fault”) parallels the anticline. The fault and fold appear to be related and likely formed from a common east-northeast to west-southwest compressional event. This fault, and smaller related splays, may have influenced the sourcing and trapping of carbon dioxide at St. Johns.

The St. Johns fault/fold set, with its eastward vergence, appears to be a typical structure formed by the Late Cretaceous Laramide Orogeny. The structural fabric of the St. Johns – Springerville area is dominated by numerous sub-parallel, northwest-trending elements comprising folds, faults, joints, lineaments, travertine outcrops, and subsurface contours. Aeromagnetic data suggested a basement fault and positive closure directly below the Antelope Valley anticline; this became the basis for discovery of the much larger and more continuous St. Johns fault.

The St. Johns field is located on the southeast side of the Holbrook basin, in an area largely free of isostatic residual gravity trends. The Springerville Volcanic Field is prominent as a low-density anomaly, perhaps caused by upwelling of relatively hot, low-density igneous rocks. Regionally, the gravitational fabric of basement trends NE-SW, which is perpendicular to the NW-SE trend of the St. Johns fault and dome. The Defiance Uplift, Paleozoic high, is prominent. Overall, the gravity data suggest that deep basement structures are less important in defining the structural history of the St. Johns Dome than the more surficial Laramide folding.

Microseismicity. Geothermal areas such as St. Johns are often associated with shallow microseismic activity, i.e., multiple small events rather than large single events. No shallow (<5 km) microseismic events were detected in the St. Johns area during a relatively short (2-week) survey which employed 19 seismographs on 20-km spacing in the Clifton and Springerville-St. Johns areas.²² However, a longer survey would be needed to monitor long-term activity.

Direct-Current Resistivity and Telluric Current Measurements. Surveys of the geothermal potential of the Springerville area determined that low DC resistivity values associated with high TDS in spring water north and south of Springerville.²³ Telluric current surveys found a low-resistivity zone southwest of Springerville, reflecting conductive rock at anomalously shallow depths: only 12 km deep compared with 24 km elsewhere in the region. This anomaly also correlates with a gravity low several hundred km² in size. The anomaly was interpreted as a crustal scale hot spot that heats water, causing high geochemical temperatures and high TDS.

Geothermal Gradient. The St. Johns – Springerville area is well known for hydrothermal activity and considered prospective for geothermal energy development. Late Tertiary and Quaternary travertine deposits are commonly associated with local structures near St. Johns, particularly anticlines, where they form cones or coalesced cones or cap hills. Deposition apparently has ceased and these travertine deposits may represent a waning phase of formerly more extensive geothermal activity.²⁴

A geothermal exploration project tested heat flow in 18 water wells in the Springerville-Alpine area during the late 1970s.²⁵ The shallow (150-450 m) boreholes were TD'd in the Supai Fm. The wells penetrated sandstone and interbedded basalts and cinder, along with copious formation water. Heat flow was logged at 5-m intervals. Geothermal gradients in the wells ranged from 46 to 60°C/km, significantly above the regional average of about 22.5°C/km.²⁶ Thermal conductivity of representative drill cuttings was measured to calculate heat flow. These wells defined a low-amplitude heat flow high (maximum $Q > 120 \text{ mW/m}^2$) in the southern portion of Ridgeway's CO₂ field, as well as a smaller maximum of similar magnitude just north and west of the field.

4.4 Stratigraphy

The stratigraphic sequence at St. Johns Dome comprises Precambrian basement, Permian evaporites and clastics, Triassic limestone, Cretaceous sandstone and shales, and Tertiary volcanic and sedimentary rocks. The following description of the stratigraphy at the field is based on log analysis, such as for the Ridgeway State 22-1X well (Figure 7) and on core analysis performed by Ridgeway on selected wells.

Precambrian Granite/Granodiorite forms the crystalline basement at St. Johns. A total of 13 m of whole conventional core was sampled from the Ridgeway State #9-21 well at a drill depth of 808 to 821 m (2,650.5 to 2,693 ft). Although, the granite is not now considered to be a prospective CO₂ reservoir at St. Johns, it was selected for coring because similar fractured basement provides the reservoir for Arizona's largest oil field. The granite is fractured and rubblized in core and weathered to dark reddish brown to dark pinkish gray color, reflecting its extreme alteration by low-grade metamorphic mineralogic replacement.

Core analysis of the granite showed porosity averages about 10%. Permeability averages about 20 mD, but ranges widely from 0.03 to 147 mD. The principal pore types are fracture and dissolution pores, although most of the tectonic fractures have been effectively healed by dolomite/rhodochrosite deposition.

Permian Supai Formation is the main CO₂-bearing formation at St. Johns Dome. From its base, the Supai Fm consists of the Amos Wash, Big A Butte, Fort Apache, and Corduroy Members. Most of the high-quality CO₂ zones are located in the Big A Butte and Amos Wash Members. In New Mexico, the Supai Fm correlates with the Yeso and underlying Abo Fm. The Supai Fm consists largely of red to reddish-brown clayey siltstone and mudstone, with significant evaporitic gypsum and anhydrite. Some halite is present in the northwest portion of St. Johns, but it is much thinner (<10 m) compared with more than 300 m encountered in the center of the Holbrook basin 50 km to the west.

Amos Wash Member of the Supai Fm (also known as Granite Wash) consists of reddish brown sandy to gravelly conglomerate, fine-grained sandstone, and minor shaly siltstone. Anhydrite nodules and caliche are common locally. Core analysis indicates porosity averaging 6.6%, formed by reduced intergranular pores and micropores. Permeability averaged 1.4 mD, but ranged widely from 0.01 to 7.6 mD. The productive zone near the base of the Amos Wash is called the Riggs Zone by Ridgeway. A generally non-productive basal zone called the Upper Abo underlies the Riggs Zone.

Big A Butte Member of the Supai Fm comprises reddish brown siltstone and very fine-grained sandstone, with minor gray-brown dolomite and five thin but laterally persistent anhydrite beds. The base of the Big A Butte is defined by a 0.7-m thick gray, argillaceous limestone. The Big A Butte ranges from about 80 to 120 m thick. Conventional core plug

porosities of the fine-grained clastic beds ranged from 4.7 to 30.3% and permeability from <0.01 to as high as 1,619 mD.

Fort Apache Member is a distinctive carbonate marker unit near the top of the Supai Fm that can be traced throughout central and east-central Arizona. The base of the Fort Apache Member can be readily identified on well logs by a distinctive gamma ray shift. The Member comprises an upper anhydrite unit and lower vuggy-to-cavernous dolomite unit that is up to 30 m thick at St. Johns Dome. West of St. Johns near Winslow, it thins to a 3-m thick carbonate with some anhydrite and gypsum. The Fort Apache Member is 20 to 30 m thick. Siltstone core plugs tested from 3.1 to 26.7% porosity and 0.01 to 0.78 mD permeability, while the dolomite ranged from 3.5 to 30.1% porosity and 0.01 to 233 mD permeability.

Corduroy Member ranges from 180 to 230 m thick and consists of very fine grained red-bed clastics interbedded with thin anhydrite and carbonate beds. Locally, the base of the Corduroy sometimes has friable water sands.

Permian Glorieta Sandstone is the main aquifer in the St. Johns Dome area. It is correlative with the better known Coconino Sandstone, but lacks the large-scale cross bedding of the latter; the Salt River Lineament forms the boundary between these two formations. The Glorieta Sandstone is white to gray, very fine- to fine-grained, well-sorted, and friable. The Glorieta is 50 to 80 m thick. Some water wells in the area produce carbonated water, suggesting (but not proving) that some CO₂ has made its way through the cap rock at St. Johns. A second explanation is that CO₂ has overfilled the capacity of the St. Johns Dome, allowing CO₂ to enter the overlying Glorieta Sandstone; this explanation does not require leakage through the cap rock. Testing of gas geochemistry in the water wells is needed to resolve the origin of CO₂ in the Glorieta Sandstone.

Permian San Andres Limestone is the eastern equivalent of the Kaibab Formation found in the Grand Canyon west of the Salt River Lineament that marks the transition. The San Andres is generally 80 to 110 m thick in the St. Johns Dome area. It consists of gray-brown, finely crystalline limestone. It is locally fractured and cavernous in the St. Johns area. Lost circulation during drilling is common. Correlating the top of the unit is impeded by solution collapse breccia.

Tertiary to Quaternary Tufa. Many terraces, mesas and buttes in the St. Johns area (e.g., near Lyman Lake) are capped by tufa deposits.²⁷ These tufa-capped buttes, consisting of the mineral

travertine (CaCO₃), are probably erosional remnants of a formerly more extensive and continuous deposit. The presence of soil, sand or gravel within the carbonate suggests a pedogenic (rather than caliche) origin. Surveys of the St. Johns area using a portable gamma-ray spectrometer found only normal (<6 ppm) uranium levels. Given erosion rates of roughly 30 cm per thousand years in the region, and the position of the travertine up to 60 m above the currently active CO₂ seepage areas, travertine deposition has been active for at least several hundred thousand years.²⁸

4.5 Reservoir Geology

The St. Johns field produces CO₂ from Permian Supai Formation, which consists primarily of fine-grained non-marine sandstone, with intercalated siltstone, anhydrite, and dolomite. Well depths are relatively shallow (200 to 700 m) and CO₂ exists as free gas in the reservoir.

Table 2 provides a summary of reservoir properties at three of the CO₂ wells. Net pay thickness ranged from 3.4 m in the Riggs Zone to over 25 m in the Amos Wash Member. Formation porosity ranges from 11-20%. Permeability varies widely, averaging approximately 10 millidarcies. Initial bottomhole pressures generally were below the hydrostatic gradient (0.3 psi/foot) in all formations. Reservoir temperatures ranged from 30°C in the Ft. Apache Member to 49°C in the deeper Riggs Zone.

Table 2 : Average Reservoir Properties for CO₂ Production Zones at St. Johns

Formation	Permeability (mD)	Porosity (%)	Net Pay Thickness (m)	Bottomhole Pressure (psi)	Bottomhole Temperature (C)
Ft. Apache	0.5 – 20	13	12.2	400	30
Amos Wash	0.5 – 100	20	25.3	480	31
Riggs Zone (L. Amos Wash)	2	11	3.4	690	49

The representative Ridgeway State 22-1X well produces from the lower portion of the Big A Butte Member, within the Permian Supai Formation. The Big A Butte Member in this well has three thin anhydrite layers which act as intra-reservoir seals. Analysis of core from the lowermost perforated zone in this well (depth 495 m or 1625 feet) showed significant porosity (26.5%) and permeability to gas (92.2 mD).

4.6 Cap Rock Geology

There are no cores or detailed cuttings description for the overlying cap rock at St. Johns Dome. However, there is useful information about the internal evaporite seals within the reservoirs. The main sealing cap rocks are evaporite minerals within the Supai Fm itself, consisting of anhydrite and gypsum; minor muddy (not clear) halite totals less than 20 m thick. This complex evaporate deposit apparently provides the main seal for CO₂ storage at St. Johns.

Multiple impermeable cap rocks are present at St. Johns field, in the form of anhydrite and shale beds in the Fort Apache and Big A Butte Members. These barriers trap carbon dioxide within multiple permeable zones. Depth to the top of the Supai Fm ranges from minimum 250 m near the St. Johns fault, while maximum overburden thickness approaches 1,000 m in the south, due to the combined effect of south-dipping structure and rising topography. The St. Johns Dome lies on the eastern margin of the Holbrook basin, which contains extensive Permian salt deposits.²⁹ Overlying the CO₂ sequence and providing additional seal are the uppermost Yeso Member, Upper Permian San Andres Limestone, and the Lower Triassic Moenkopi, Shinarump, and Chinle Formations.

4.7 CO₂ Origin and Trap

CO₂ isotope and noble gas geochemistry -- particularly the isotopic composition and abundances of He, Ne and Ar -- have established the origin of CO₂ at St. Johns to be magmatic (mantle derived).^{30,31} An independent reservoir analysis commissioned by Ridgeway in 1999 estimated a total resource of 780 billion m³ (14.8 Tcf) of original gas in place at the St. Johns field. Of this resource in place, approximately 730 billion t (13.9 Tcf) is estimated to be CO₂, 1.8 billion m³ (64 Bcf) is helium, and the balance primarily nitrogen. Recoverable CO₂ reserves are an estimated 420 billion t (8 Tcf). Most of the wells at the field have been temporarily abandoned and could not be sampled. Overall, gas composition averages about 92% CO₂, 6.6% nitrogen, and 0.6% helium, along with minor methane and argon. Heavier hydrocarbons, H₂S, and CO are scarce.

Stable carbon isotopes provide insight into the origin of CO₂ at St. Johns field, as well as compartmentalization (or the lack thereof in this case) within the field. Delta ¹³CO₂ values were quite consistent across the field and different stratigraphic intervals. The relative uniformity of delta ¹³CO₂ at around -3.8‰ suggests that there are few internal barriers in the field to lateral mixing and no significant compartmentalization within the reservoir. This uniformity was not seen in the wide range in CO₂, helium, or nitrogen content.

4.8 CO₂ Drilling Operations

Ridgeway Petroleum Corp., the former field operator, has drilled 21 CO₂ production wells since discovering the field in 1994. At present the field is shut in due to limited local market demand and lack of a CO₂ pipeline. Ridgeway had planned to construct a 987-km (617-mi) pipeline from the St. Johns field to a delivery point near Bakersfield, California. The CO₂ would be used mainly for immiscible enhanced oil recovery in the relatively heavy depleted oil fields in the southern San Joaquin Valley.

The production wells were drilled with air or with a fresh water and starch-based mud to avoid formation damage and were completed in one or more of the three CO₂-bearing zones (Ft. Apache, Amos Wash, and Upper Abo/Granite Wash) at an average depth of 850 m. Typically, a surface casing of conventional carbon steel 24.4-cm (9 5/8-in) to 34-cm (13 3/8-in) in diameter was first cemented into place at a depth of about 30-40 m (100-130 ft) in the Quaternary, Triassic Chinle, Moenkopi or other outcropping units to shut off potential fresh-water zones. Next, an intermediate

casing string of 22-cm (8 5/8-in) pipe was cemented to a depth of about 210-400 m (700-1300 ft). The base of the intermediate casing was generally near the top of the Supai Fm, often in the Glorieta Sandstone or San Andres Limestone, but occasionally partway into the Corduroy Member of the Supai.

The wells were completed using 4.5-inch diameter casing, consisting of amine carbon gauze fiberglass or conventional carbon steel lined with high-density polyethylene (HDPE) to minimize corrosion. St. Johns Dome has the advantage of relatively shallow depth, permitting the use of corrosion-resistant amine carbon gauze casing. This material is feasible at pressures below about 2,000 psia, equivalent to about 1,500 m depth. In the western (Arizona) portion of the field the casing is amine carbon gauze fiberglass. But in the eastern region (New Mexico), state regulations required the casing to be conventional carbon steel lined with high-density polyethylene (HDPE). The St. Johns wells were relatively inexpensive to drill and complete (\$300,000/well) compared with wells at much deeper CO₂ fields.

Most wells (14 out of 19) were simply completed without hydraulic fracture stimulation. Five of the wells (22-1X, 11-21, 9-21, 9-22-29, and 10-16-31) were stimulated using an unrecorded volume of water and approximately 45 t (100,000 lbs) of sand. Three of the wells were hydraulically fractured in two zones (Riggs and Big A Butte zones). The other two wells employed a single frac (Riggs or Big A Butte).

4.9 Potential Application of Methanogens to St. Johns Dome CO₂ Field

Methanogens employ two separate pathways to convert CO₂ into methane: either the methyl group of acetate or the reduction of carbon dioxide with electrons from hydrogen or formate. Acetate, carbon dioxide, hydrogen, and formate are the major substrates for the methanogenic group. The hydrogen-producing acetogenic group is necessary to further metabolize butyrate and propionate to substrates for the methanogenic group. Such "interspecies electron transfer" is fortunate, in that the hydrogen-producing and hydrogen-consuming groups are interdependent. Other environmental screening factors for methanogens include temperature, salinity levels and composition, and pH. Reservoir pressure (related to depth) does not interfere with methanogen growth. In fact, high pressure only accelerates methanogenic reactions.

Our initial evaluation indicates that the geologic and reservoir conditions at St. Johns Dome

may be conducive to maintaining suitable biochemical conditions for methanogenesis of various species for the conversion of CO₂ into CH₄. Testing and further evaluation could be conducted under future R&D programs. Given the embryonic phase of methanogen conversion technology, actual Phase III commercial application appears to be some decades in the future.

Methanogen Species. Of the 98 species evaluated, six were selected as the potentially the best candidates for conversion of CO₂ into methane under conditions present at St. Johns Dome. The selection was based on the ability of these species to tolerate a variety of environmental conditions and their simplicity of requirements for growth and CO₂ reduction to methane. The most promising methanogen species are: *Methanosarcina vacuolata* and *barkerii*; *Methanococcus vannielii*; and *Methanobacterium uliginosum*, *ivanovii*, and *subterraneum*.

a) **Methanosarcinaceae** grow and produce methane by dismutating methyl compounds such as methanol, methyl amines, and methyl sulfides. Acetate, CO₂, and CO also can be utilized by some species. Ammonium and sulfide serve as the major nitrogen and sulfur sources, with many species being autotrophic; however, cofactor supplements such as biotin are required by some. Species of *Methanosarcinaceae* have been isolated from very diverse anaerobic habitats and exhibit a wide range of physiological optima. Most species are mesophilic. However, *Methanococcoides thermophila*, which was isolated from a 55°C anaerobic digester, is thermophilic. Conversely, *Methanococcoides burtonii* (Ace Lake, Antarctica) and *Methanosarcina baltica* (Baltic Sea) are psychotrophic. Since many species have been isolated from marine environments, they prefer salinity near that of sea water.

c) **Methanobacteriaceae** produce energy by reducing CO₂ with H₂, except for the species of the genus *Methanosphaera*, which produce methanol with H₂. Many species also use formate or secondary alcohols to reduce CO₂. Most species of the genera *Methanobacterium* and *Methanothermobacter* are autotrophic, utilizing ammonium (some species are also capable of fixing dinitrogen) and sulfide as nitrogen and sulfur sources, and most also require trace elements such as tungsten and nickel. Species of *Methanosphaera* and *Methanobrevibacter* are heterotrophic, requiring acetate and sometimes vitamins for growth. All species are mesophilic except those of the genus *Methanothermobacter*, which are thermophilic. Nearly all species are neutrophilic, having an optimum pH of 6.8 to 8.0. Of particular interest is *Methanobacterium subterraneum*, which is not only alkalophilic, but also the only species which is halo-tolerant, forms aggregates, and of all known methanogens, the only one to be isolated from granitic ground water.

Nutritional and Environmental Controls. The main growth factors are free hydrogen, formate or secondary alcohols to reduce CO₂, moderate pH (6.8 to 8.0). Of these conditions, only temperature and pH are suitable at St. Johns Dome. Hydrogen, formate or secondary alcohols, methanol, methyl amines, and methyl sulfides would need to be added to support methanogen growth. Acetate, CO₂, and CO also can be utilized by some species. Certain trace elements (nickel, tungsten) or yeast extract also would need to be added.

5.0 Phase III Commercial Application Potential

Methanogenic conversion of CO₂ into CH₄ is still at an embryonic pre-commercial phase of research and development. Considerable basic R&D will need to be conducted to evaluate the technical and economic feasibility of this process. For this reason, the ARI consortium does not recommend a Phase III commercial application proposal at this time.

High-potential topics for future R&D into methanogenic conversion of CO₂ could include:

- Isolating individual species within the Methanobacteriaceae and Methanosarcinaceae families with greatest potential for application to CO₂ storage sites.
- Further laboratory experimentation with accelerating methanogen growth under varying temperature, pH, and nutrient conditions.

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