

An Evaluation of the Feasibility of Combining CO_2 Flooding Technology with MEOR Technologies in order to Sequester CO_2

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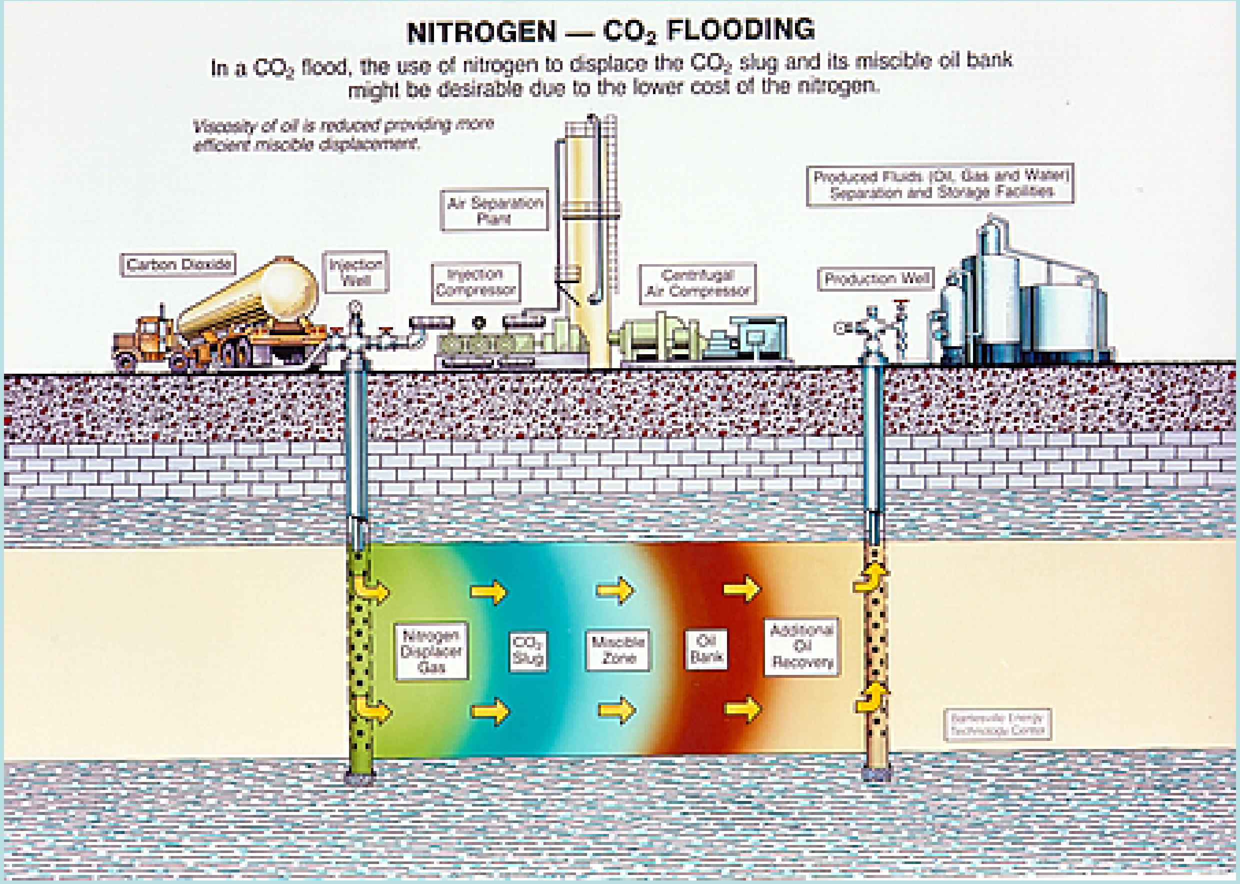
Objectives

- ✓ To revitalize archived microbial isolates from a previous DOE funded MEOR Project.
- ✓ Screen those cultures for the ability to produce methane, saponifiable membrane lipid types, and protein production
- ✓ Using radio-labeled CO_2 determine where it will go and to what extent.

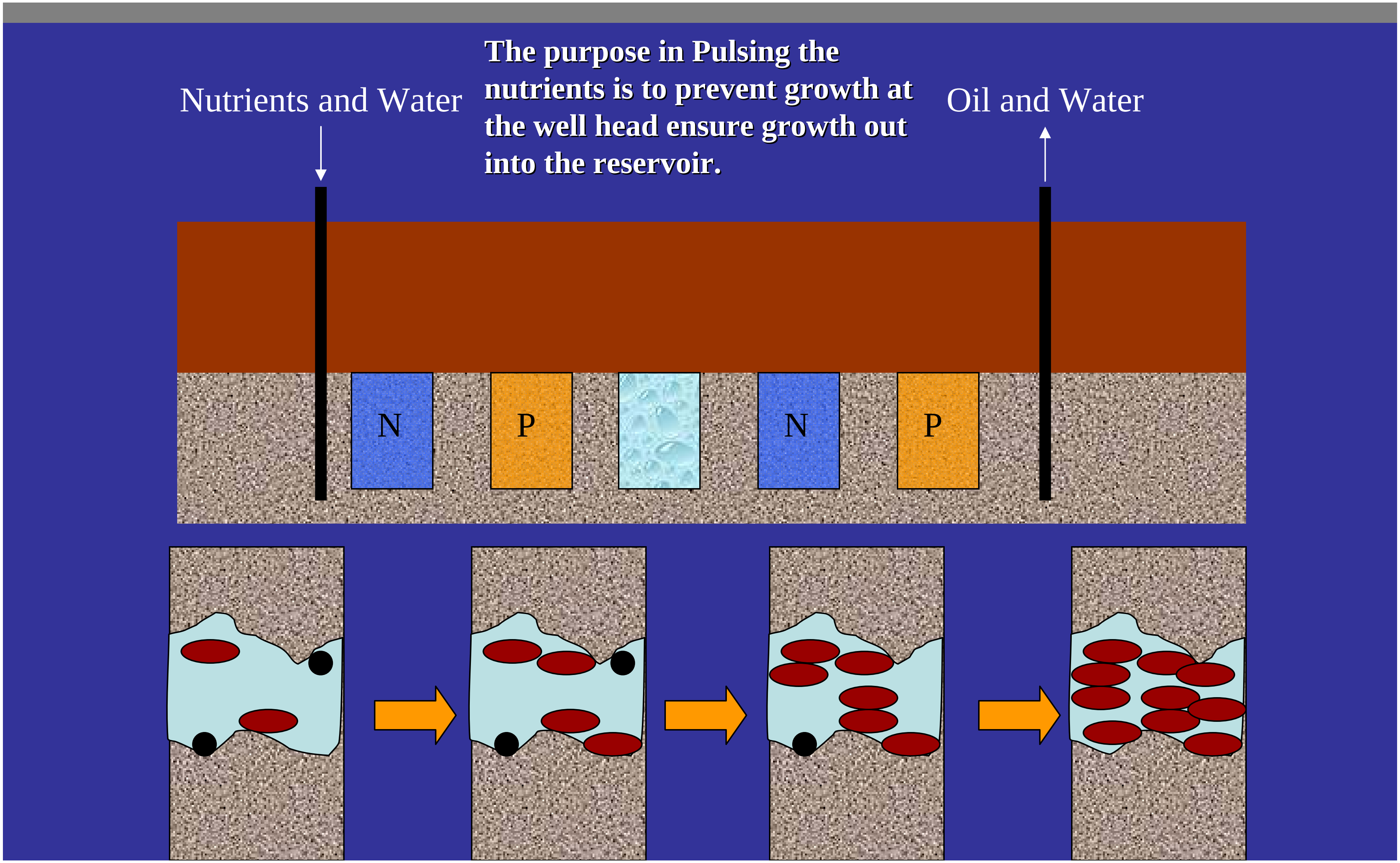
CO_2 Flooding



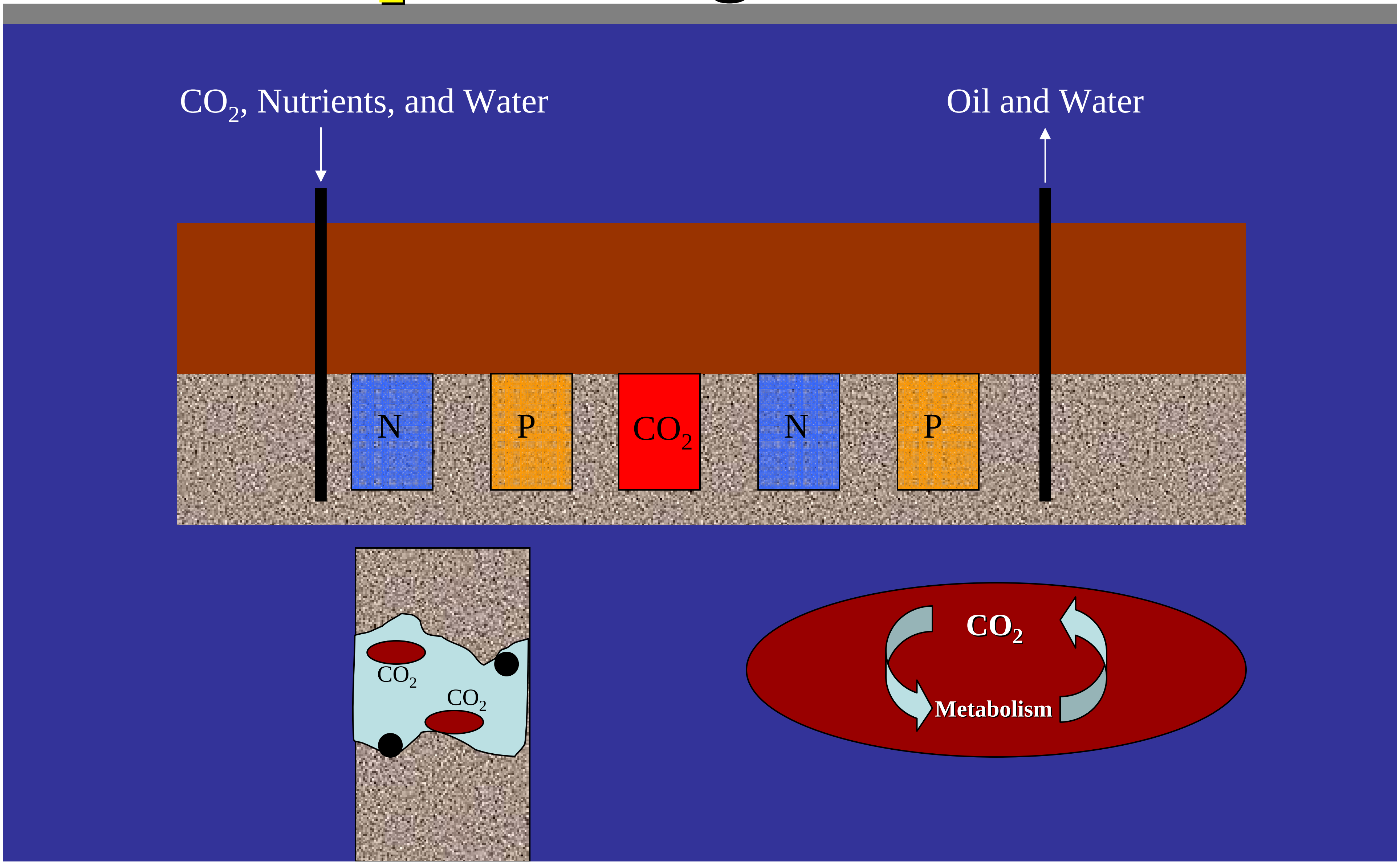
http://www.netl.doe.gov/technologies/oil-gas/EP_Technologies/Images/EP_Pages/EOR_CO2Flood.jpg

MEOR

(MSU Process)



Coupling
CO_2 Flooding & MEOR



Products of CO_2 Metabolism

- CO_2 is combined with acetyl CoA to form Malonyl CoA
  - Fatty acid production
  - Reversed to produced Amino Acids
- CO_2 is combined H_2 to produce Methane
- H_2 is provided from the metabolism of hydrocarbons
  - Microorganisms produce alcohols, aldehydes, acids thereby providing enough NADPH to reduce CO_2
  - Some organisms produce H_2 as a means of regenerating NAD
- The reducing power for converting the CO_2 comes from the hydrocarbons or any reduced metals present in the reservoir.

Methods

- 1st Step in the project was to begin the revitalization of archived cultures.
  - Began reanimation of cultures by slowly increasing the temperature to 30°C
  - Transfer the cultures to new medium
  - Since these cultures were incubated anaerobically they were very slow growing.
- Successfully revitalized 20+ isolates

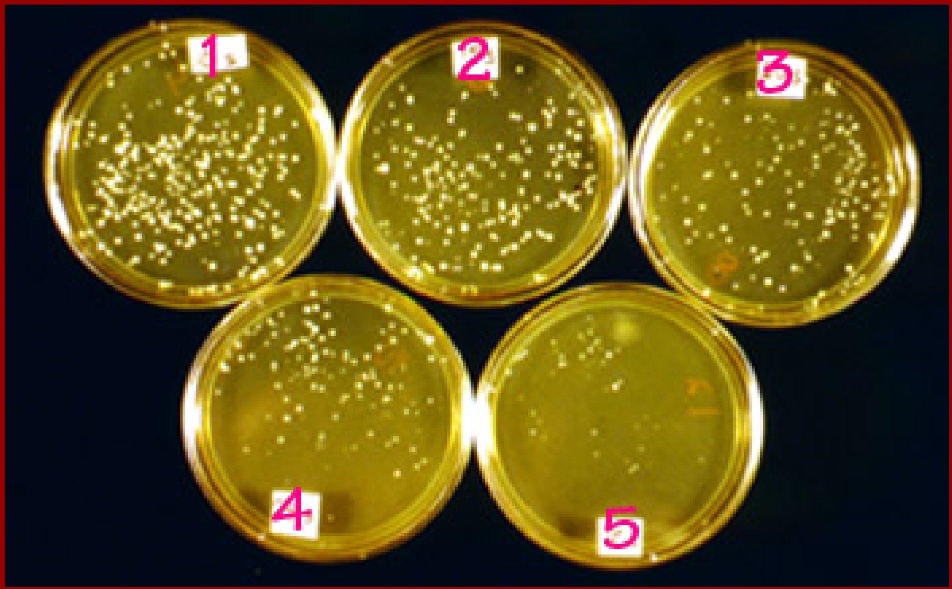
Methods

- 2nd Step in the project was to determine the potential chemicals that CO_2 could be converted into.
  - Methane
    - GC-TCD analysis of the headspace
  - Lipids (i.e. Fatty acids)
    - Bligh-Dyer Extraction
    - Transesterification RXN to produce FAME
    - GC FID analysis
  - Protein
    - Lowry Method

- All conducted using non-labeled CO_2

Results

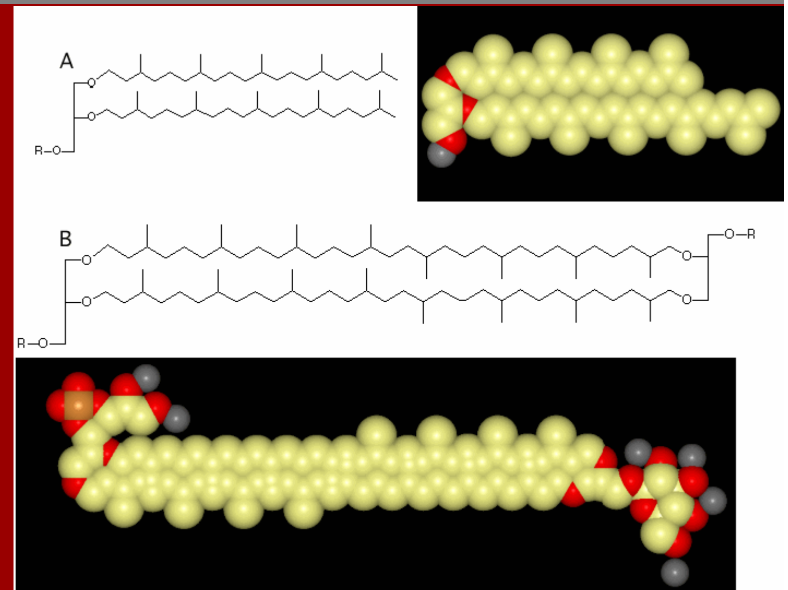
- **Methane Production**



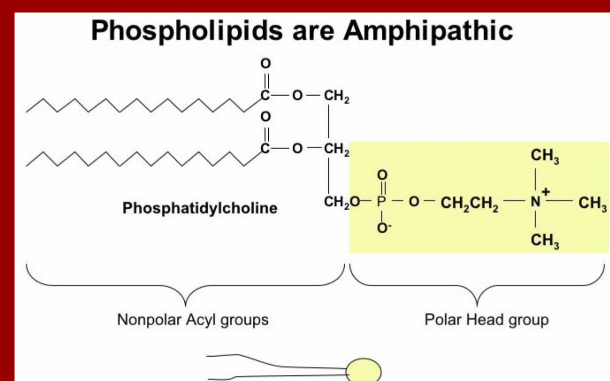
Culture #	Medium + Carbon Source	% Produced	
		CO_2 %	CH_4 %
2	Tryptic Soy Agar	6.54	0.00
29	Tryptic Soy Agar	11.21	0.00
43	Tryptic Soy Agar	12.97	0.00
75	Tryptic Soy Agar	8.53	0.00
1	Tryptic Soy Agar	10.28	0.00
5	Tryptic Soy Agar	9.23	0.00
26	Tryptic Soy Agar	8.53	0.00
52	Tryptic Soy Agar	12.45	0.00
56	Tryptic Soy Agar	13.78	0.00
70	Tryptic Soy Agar	6.03	0.00
43	MSM + Hexadecane	1.35	1.61
53	MSM + Hexadecane	1.35	0.81
82	MSM + Hexadecane	1.36	0.81
88	MSM + Hexadecane	1.34	1.62
29	MSM + Hexadecane	1.35	1.62
78	MSM + Hexadecane	1.41	1.61
64	MSM + Hexadecane	1.38	1.62
85	MSM + Hexadecane	1.37	0.81
14	MSM + Crude Oil	1.35	0.81
64	MSM + Crude Oil	1.32	1.61
2	MSM + Molasses	9.26	0.00
88	MSM + Molasses	5.08	0.00

Results

- Lipid Analysis
- Type of lipids present
  - Ether
    - Archae
  - Ester
    - Phospholipids
    - Bacteria



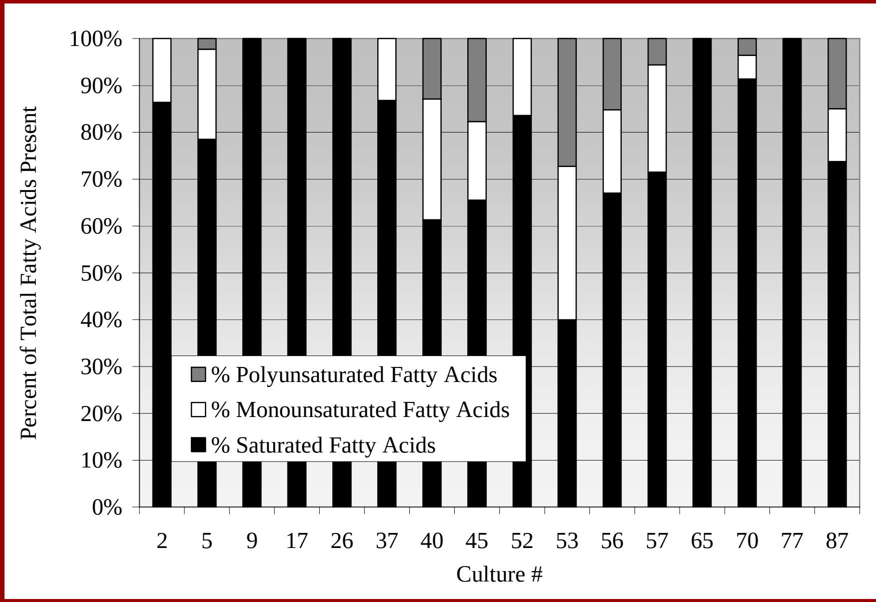
http://www.bact.wisc.edu/Microtextbook/images/book_4/chapter_2/2-69.gif



http://www.langara.bc.ca/biology/mario/Assets/phospholipids.jpg

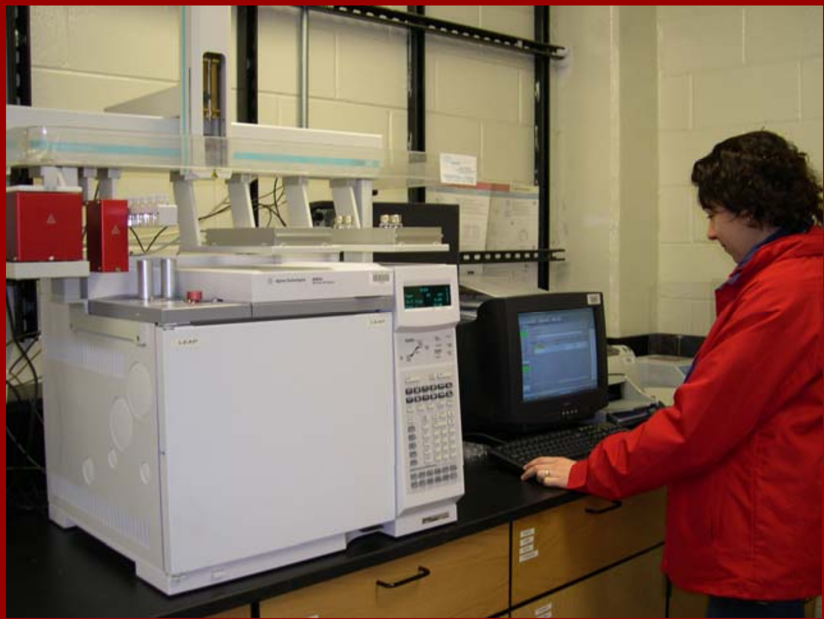
Results

- 5 cultures (9, 17, 26, 65, and 77)
  - Had no unsaturated fatty acids.
  - Not Common in Nature
- Most all cultures had greater than 60% saturated fatty acids
- Culture 53 had greater than 60% unsaturated fatty acids.



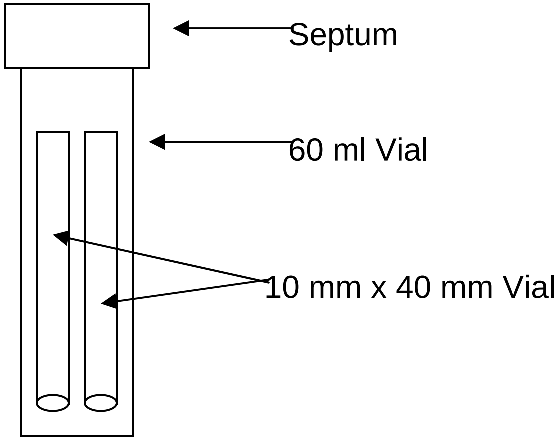
Results

- Protein was below detectable limits
- Decision made to wait until radioisotope studies



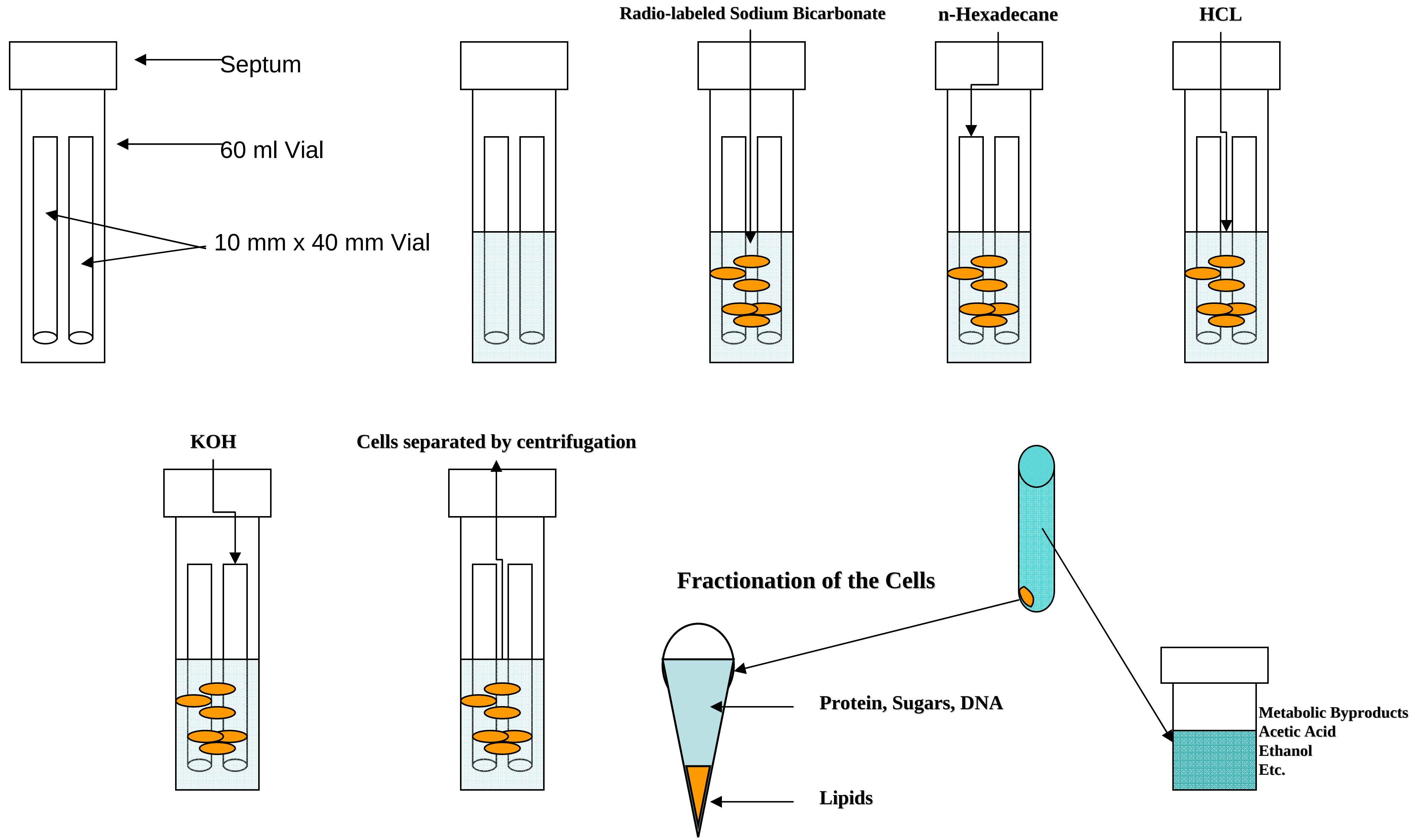
Methods

- 3rd Step in the project was to determine fate of CO_2 in biological systems using radio-labeled CO_2
  - Methane
    - N-hexadecane trap
  - Unused CO_2
    - KOH Trap
  - Lipids & Protein
    - Separated using Bligh-Dyer Extraction method

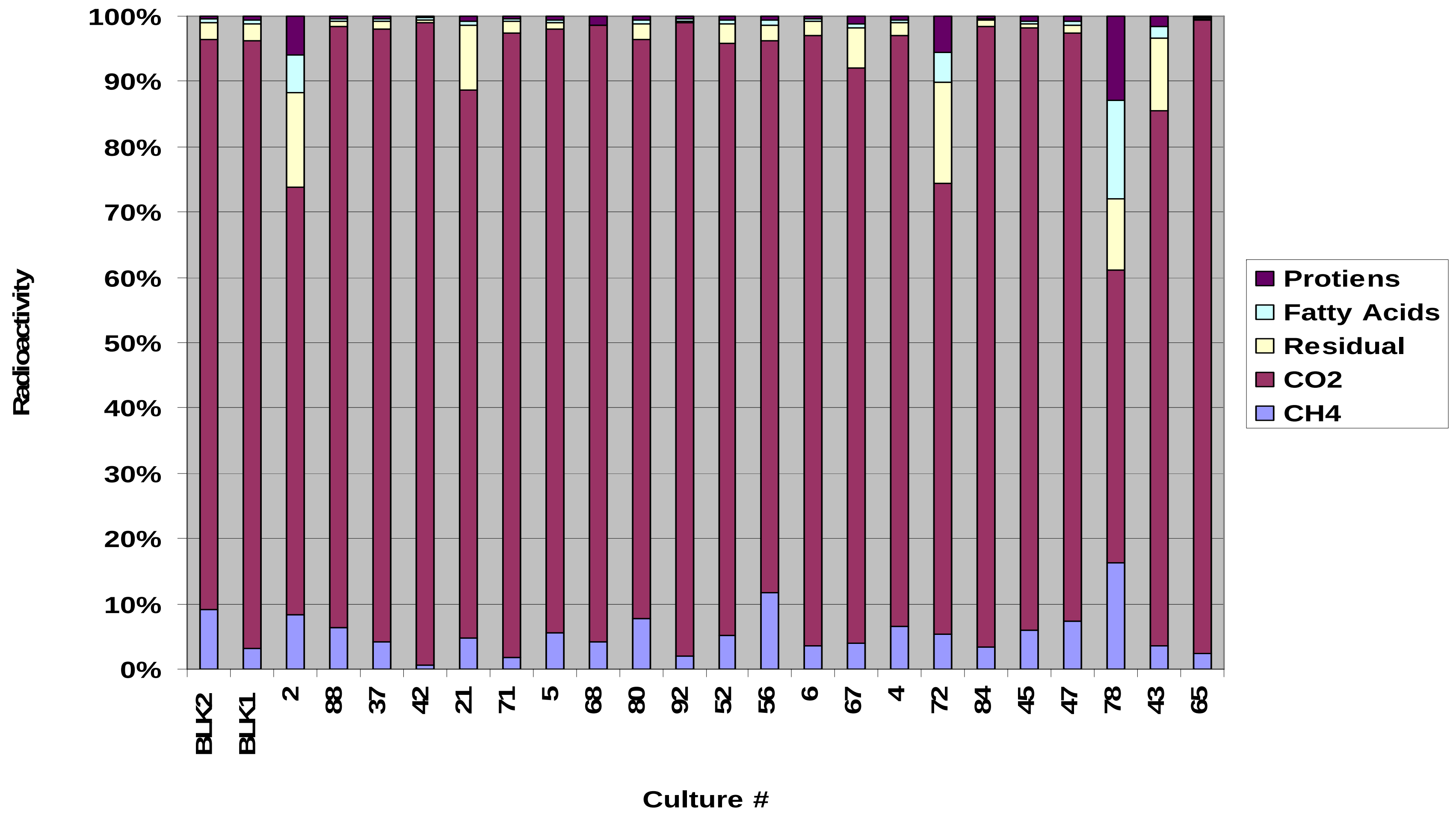


- All conducted using C^{14}-labeled sodium bicarbonate

Methods


Radio-labeled Sodium Bicarbonate
n-Hexadecane
HCL
Septum
60 ml Vial
10 mm x 40 mm Vial
KOH
Cells separated by centrifugation
Fractionation of the Cells
Protein, Sugars, DNA
Lipids
Metabolic Byproducts
Acetic Acid
Ethanol
Etc.

Results



100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Radioactivity
BLK2
BLK1
2
88
37
42
21
71
5
68
80
92
52
56
6
67
4
72
84
45
47
78
43
65
Culture #
Protiens
Fatty Acids
Residual
CO2
CH4

Summary

- **Cultures isolated responded positively to revitalization efforts.**
- **These cultures were capable of producing saponifiable lipids and proteins.**
- **Some of the cultures were capable of producing methane.**
- **Preliminary results using ^{14}C indicate these cultures are capable of biologically converting CO_2 into cellular material**

Future Efforts

- Continue to identify where the CO_2 is being incorporated using ^{14}C sodium bicarbonate.
  - Since these were only initial experiments, they are currently being repeated.
  - Use this information to determine if there is any appreciable amount of CO_2 converted to cellular material

Acknowledgements

- Jack Carmicheal
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Thank You!
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