**QUANTITATION OF MICROBIAL PRODUCTS AND THEIR EFFECTIVENESS IN
ENHANCED OIL RECOVERY**

QUARTERLY TECHNICAL PROGRESS REPORT

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Objective:

The goals of this project are to determine the growth kinetics and the relationships that exist between microbial growth, microbial product formation and oil recovery, and to develop mathematical models that predict microbial activity in porous materials.

Oil Recovery Related to Microbial Product Formation:

The studies on the influence of microbial growth and product formation on the recovery of residual oil from Berea sandstone cores were continued this quarter. The experiments were carried out at 36 °C using the high pressure core apparatus described previously. Four microbial and nutrient treatments were carried out on a single Berea sandstone core, using the microorganism *Clostridium acetobutylicum* suspended in a glucose-mineral salts medium. The experimental conditions used are listed in Table 1. Microbial growth in the core was monitored by following pore pressure changes.

Table 1: Berea Core Experimental Conditions.

	1st Treatment	2nd Treatment	3rd Treatment	4th Treatment
Nutrient Injection	300 ml	300 ml	300 ml	100 ml
Inoculation	200 ml	250 ml	200 ml	140 ml
Initial Pore Pressure	500 psig	700 psig	750 psig	1000 psig
Max. Pore Pressure	575 psig	880 psig	1050 psig	1090 psig
Incubation Time	12 days	8 days	5 days	8 days
Oil Production	1 ml	1 ml	0 ml	0 ml
Gas Production	27 ml	62 ml	51 ml	83 ml
Effective Permeability	40 md	57 md	55 md	57 md

In the first treatment the pore pressure did not increase during the first 35 hours of incubation. During the next 20 hours the pressure increased by 175 psig. After a further 75 hours of incubation, the pressure decreased, possibly due to the microorganism, *Clostridium acetobutylicum*, switching its metabolism from organic acid production to solvent production which involves the uptake of hydrogen gas. Biochemical analysis of the effluent showed that 16 mM of glucose was degraded with the production of acetate, butyrate, ethanol, butanol and CO₂ (Table 2), products characteristic of sugar metabolism by *C. acetobutylicum*. The effective permeability did not change markedly during this treatment (42 md to 40 md), and only 1 ml (about 3%) of the residual oil was produced.

During the second treatment an increase in pressure was observed after only 22 hours of incubation as opposed to 35 hours for the first treatment. This shorter lag time was probably due to an active microbial population being present from the first treatment. The core pressure again increased by 175 psig over a 36 hour period after which an immediate pressure decrease of 125 psig was observed (Fig. 1). Biochemical analysis of the effluent showed that 60 mM glucose was degraded and a corresponding production of acetate, butyrate, ethanol, butanol and CO₂ was observed (Table 2). Again, only 1ml (3%) of the residual oil was recovered. The effective permeability increased from 40 md to 57 md. The increase in permeability may be due to dissolution of carbonate minerals due to the production of the organic acids.

The lag phase of the third treatment was estimated to be about 3-5 hours. The length of the lag phase was so short that it could not be accurately estimated because the increase in pressure due to microbial gas production overlapped that due to thermal expansion of the injected liquids. Thermal expansion is estimated to account for 150 psig increase in pressure (Fig. 2). The core pressure reached a maximum value after 20 hours of incubation and again decreased by 100 psig during the next 73 hours. No residual oil was recovered during this treatment, and the effective permeability did not change (57 md to 55 md). Biochemical analysis has not been completed on these samples; however, 51 ml of gas was produced during the incubation.

A continual increase in pressure was observed for 20 hours when the fourth batch of nutrients was injected into the core. It is estimated that the increase in pressure due to thermal expansion of the liquid should be complete within 5 hours of incubation. A maximum pressure of almost 1100 psig was seen after 17 hours. After 20 hours incubation, pressure decreased by almost 300 psig over the next 70 hours (Fig. 3). Again no residual oil was recovered, and the effective permeability did not change. Eighty three milliliters of gas were produced.

Work continues on understanding the mechanism of oil recovery using *C. acetobutylicum*.

Table 2: Biochemical analysis of produced brines from the core

Effluent ^a	Glucose mM	Acetate mM	Butyrate mM	Ethanol mM	Butanol mM	CO ₂ mmoles/l of effluent
1st Treatment E1	152.6	27.0	24.0	3.2	5.3	0.52
E2	184.2	9.0	8.0	2.4	2.3	0.35
E3	114.3	0.4	1.6	^b ND	ND	0.25
2nd Treatment E1	19.4	29.0	29.5	7.0	9.6	3.09
E2	114.2	35.0	25.6	7.1	7.3	2.65
E3	66.3	11.4	5.2	1.3	0.9	1.35
E4	9.2	3.0	0.9	ND	ND	0.57

^a1st Treatment: Liquid effluent volume was E1=40ml, E2=43.5ml, E3=50.0ml; all effluent samples contained an oily layer and fine particulates on the liquid surface, except E3. Gas volume was E1=9.0ml, E2=7.5ml, E3=11ml.

2nd Treatment: Liquid effluent volume was E1=41ml, E2=41ml, E3=43ml, E4=47ml; all effluent samples contained fines on the surface except E4. Gas volume was E1=19ml, E2=17ml, E3=14ml, E4=12ml.

The glucose concentrations in the influent of the 1st and 2nd treatment were 166.5 and 111.0 mM respectively.

^b Concentration below detectable limit of the assay used.

Fig.1 Pore Pressure in Treatment #2

Fig.2 Pore Pressure in Treatment #3

Fig.3 Pore Pressure in Treatment #4

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