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TOXICITY STUDIES OF UNDERGROUND COAL GASIFICATION AND TAR SANDS PROCESSES

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TOXICITY STUDIES OF UNDERGROUND COAL GASIFICATION AND TARSANDS PROCESSES

Progress Report

February 1, 1982 - January 31, 1983

Univ. of Wyoming
Laramie, WY

DOE/EV/04477--T32

Introduction

Underground Coal Gasification (UCG) Technology could make approximately 3.55 trillion tons of coal economically available as an energy source (Glass, 1977). Deposits of deep lignite low grade subbituminuous coal deposits are found in the western United States. Extensive field trial coal gasification experiments by Laramie Energy Technology Center (LETC) at Hanna, Wyoming and Vernal, Utah have been conducted. Samples of process waters for genotoxicity and cytotoxicity studies were provided by LETC from both sites and from the Lawrence Livermore Hoe Creek, Wyoming site.

Although health hazards associated with surface mining are lower for UCG than for deep mining, (Virgona, 1978) the toxicity of waters coproduced with the product gas needs to be assessed.

Condenser water, coproduced with product gas, and removed from the gas stream by condensation, is heavily laden with inorganic and organic solutes. Potential hazards to aquatic biota of untreated condenser water and ground water samples have been determined from samples provided by LETC (Bergman et al., 1979; DeGraeve et al., 1980; Marcus and Bergman, 1980a; Bergman et al., 1981a,b). Chemical analyses of the waters indicated samples were highly alkaline with high phenol and ammonia. Toxicity ranges of 0.01-0.1% dilution for invertebrates and fish was found (Bergman et al., 1981b). Genotoxic effects were assessed in the present study of waters from all field sites in the Ames assay and from Hanna and Utah tarsands in the *Paramecium* bioassay.

Materials and Methods

The cytotoxic and genotoxic procedures are described (Smith-Sonneborn et al., 1981 , 1982).

The following coal process waters were tested in the Ames assay:

1) 80-(Tarsand-1S) 03W-00-C

2) 79-(Hanna-IVB) 02W-03-U

3) 77-(Tarsand-2C) 00W-00-U

4) 80-(Tarsand-1S) 03W-00-C filtered

5) 80-(HOECR) 01W-00-U

6) 79-(Hanna-IVB) 01W-00-U

Samples one, four and six were also tested in the Paramecium assay.

Results

The cytotoxicity of coal gasification process waters from both the Hanna IVB (01W) and Vernal Tarsand field trials indicates sensitivity of these cells at 2% sample dilution for Hanna IV (01W) condensate and 20% for the Tarsand samples. Genotoxicity of the process waters in the Ames assay (Tables 2-7) show increased revertants with both Sample 1 (Tarsand) and Sample 6 (Hanna IVB). The tarsand sample was mutagenic at 100µl in one of four trials with strain TA100 (Table 2); the Hanna IVB (01W) sample was mutagenic at 200µl + S9 in one of two trials with TA98 and at 100µl + S9 in the other trial (Table 7). The sporadic mutagenic response noted in these waters in the Salmonella assay was also found with the Paramecium bioassay. Both the Tarsand and Hanna IVB (01W) (Samples 1 and 6) were mutagenic in Paramecium (Tables 8 and 9).

Discussion

Paramecium was 200 times less sensitive to toxins in the coal process waters than the other bioassays using Daphnia, fathead minnows and rainbow trout (Bergman et al., 1981b). The present studies indicate that both the Paramecium and Ames bioassays show sporadic genotoxic response to the process waters from both Hanna IVB (01W) and the Utah tarsand coal gasification field trial experiments. Previous studies with the mutagenicity of products from coal liquifaction and gasification detected activity associated only with a tar byproduct of coal gasification, a liquification vehicle oil sample and products of the coal hydrogenation process in the Ames assay (Warshawsky et al., 1981). The present study offers additional evidence that byproducts of coal processes can contain genotoxic materials. The results of the present and previous studies indicate the need for some caution in the use and disposal of these waters and suggest that further research on this potential biohazard parallel consideration of further development of this technology.

Table 1

CYTOTOXICITY OF COAL GASIFICATION EXPERIMENTS IN PARAMECIUM

Dilution	Hanna 01W	Tarsand 1S Filtered	Tarsand 1S
1:5	0/16	14/16	Too turbid
1:10	0/16	15/16	Too turbid
1:25	0/16	16/16	Too turbid
1:50	16/16 Very Sick Cells	16/16	12/16
1:100	14/16	16/16	12/16
1:200	14/16	16/16	14/16
Control	16/16	16/16	16/16

Too turbid = too turbid to see cells

Number of surviving cells after 2 hours at 34°C

Table 2

Ames Assay
Genotoxicity of Tarsand 1S 03W-00-C

Number of Revertants

Trial[c]	TA 100 1	TA 100 2	TA 100 3	TA 100 4	TA 100 X̄ ± SEM	TA 98 1	TA 98 2	TA 98 3	TA 98 4	TA 98 X̄ ± SEM
BAP[e] + S9	430[a]	800[a]	246[a]	333[a]	452[a] ± 122	151[a]	279[a]	110[a]	103[a]	161[a] ± 41
BAP[e]	106	102	111	48	92 ± 15	22	31	20	13	22 ± 4
S9	74	110	97	51	83 ± 13	24	29	25	19	24 ± 2
Control	91	114	102	41	87 ± 16	17	23	15	10	16 ± 3
Process Water										
50µl	79	---[b]	71	41	64 ± 12	10	---[b]	15	7	11 ± 2
50µl + S9	47	---[b]	62	69	59 ± 6	24	---[b]	23	17	21 ± 2
100µl	227[a]	67	62	56	103 ± 41	7	---[b]	20	12	13 ± 4
100µl + S9	292[a]	57	66	67	120 ± 57	10	---[b]	24	11	15 ± 5
200µl	2	---[b]	69	24	32 ± 20	0	14	21	7	11 ± 5
200µl + S9	0	---[b]	86	51	46 ± 20	1	25	15	12	13 ± 5
Nitro[d]	1600[a]	1600[a]	1800[a]	---[b]	1666[a] ± 66	159[a]	194[a]	226[a]	---[b]	193[a] ± 19

a = A minimum of 2X control or S9 values were obtained.

b = These dilutions were not conducted during this trial.

c = Each trial value represents a mean count of three plates.

d = 0.15µg/plate of 4-nitroquinoline-N-oxide was used.

e = 4.5µg/plate of benzo(a)pyrene.

Table 3

Ames Assay

Genotoxicity of 79-Hanna IVB 02W-03-U

Number of Revertants

Trial[c]	TA100 1	TA100 2	$\bar{X}$	± SEM	TA98 1	TA98 2	$\bar{X}$	± SEM
BAP[e] + S9	471[a]	800[a]	635[a]	± 165	228[a]	279[a]	253[a]	± 26
BAP[e]	98	102	100	± 2	8	31	19	± 12
S9	96	110	103	± 7	26	29	28	± 2
Control	103	114	108	± 6	6	23	14	± 9
Process Water 50µl	4	—[b]	4		1	—[b]	1	
50µl + S9	9	—[b]	9		9	—[b]	9	
100µl	71	99	85	± 14	8	25	16	± 9
100µl + S9	69	108	88	± 20	23	37	30	± 7
200µl	63	—[b]	63		0	—[b]	0	
200µl + S9	60	—[b]	60		0	—[b]	0	
Nitro[d]	---[b]	1600[a]	1600[a]		---[b]	194[a]	194[a]	

a = A minimum of 2X control or S9 was values were obtained.

b = These dilutions were not conducted during this trial.

c = Each trial value represents a mean count of three plates.

d = 0.15µg/plate of 4-nitroquinoline-N-oxide was used.

e = 4.5µg/plate of benzo(a)pyrene.

Table 4

Ames Assay

77 (Tarsand-2C) OOW-OOU

Trial[c]	TA100 1	TA100 2	TA100 $\bar{X}$ ± SEM	TA98 1	TA98 2	TA98 $\bar{X}$ ± SEM
BAP[e] + S9	381[a]	800[a]	591[a] ± 210	160	279[a]	220[a] ± 60
BAP[e]	90	102	96 ± 6	5	31	18 ± 13
S9	80	110	95 ± 15	23	29	26 ± 3
Control	103	114	109 ± 6	8	23	16 ± 8
Process Water 50µl	54	---[b]	54	4	---[b]	4
50µl + S9	52	---[b]	52	16	---[b]	16
100µl	92	75	84 ± 9	8	17	13 ± 5
100µl + S9	93	82	88 ± 6	22	35	29 ± 7
200µl	60	---[b]	60	3	---[b]	3
200µl + S9	69	---[b]	69	19	---[b]	19
Nitro[d]	---[b]	1600[a]	1600[a]	---[b]	194[a]	194[a]

Number of Revertants

a = A minimum of 2X control or S9 values were obtained.

b = These dilutions were not conducted during this trial.

c = Each trial value represents a mean count of three plates.

d = 0.15µg/plate of 4-nitroquinoline-N-oxide was used.

e = 4.5µg/plate of benzo(a)pyrene.

Table 5

Ames Assay

Genotoxicity of 80-(Tarsand-1S)-03W-00-C (Filtered)

Number of Revertants

Trial[c]	TA100 1	TA100 2	TA100 $\bar{X} \pm$ SEM	TA98 1	TA98 2	TA98 $\bar{X} \pm$ SEM
BAP[e] + S9	369[a]	800[a]	585[a] ± 216	136[a]	279[a]	208[a] ± 72
BAP[e]	123	102	113 ± 11	29	31	30 ± 1
S9	95	110	103 ± 8	36	29	32 ± 4
Control	146	114	130 ± 16	15	23	19 ± 4
Process Water						
50µl	67	---[b]	67	7	---[b]	7
50µl + S9	16	---[b]	16	16	---[b]	16
100µl	108	90	99 ± 9	28	20	24 ± 4
100µl + S9	---[b]	86	86	34	52	43 ± 9
200µl	68	---[b]	68	0	---[b]	0
200µl + S9	39	---[b]	39	5	---[b]	5
Nitro[d]	---[b]	1600[a]	1600[a]	---[b]	194[a]	194[a]

a = A minimum of 2X control or S9 values were obtained.

b = These dilutions were not conducted during this trial.

c = Each trial value represents a mean count of three plates.

d = 0.15µg/plate of 4-nitroquinoline-N-oxide was used.

e = 4.5µg/plate of benzo(a)pyrene.

Table 6

Ames Assay

Genotoxicity of 80-HOECR (01-00-U)

Number of Revertants

Trial[c]	TA100 1	TA100 2	TA100 $\bar{X}$ ± SEM	TA98 1	TA98 2	TA98 $\bar{X}$ ± SEM
BAP[e] + S9	330[a]	800	565[a] ± 235	172[a]	279[a]	226[a] ± 54
BAP[e]	50	102	76 ± 26	8	31	20 ± 12
S9	92	110	101 ± 9	25	29	27 ± 2
Control	80	114	97 ± 17	17	23	20 ± 3
Process Water 50µl	0	---[b]	0	0	---[b]	0
50µl + S9	0	---[b]	0	0	---[b]	0
100µl	109	101	105 ± 4	0	18	9 ± 9
100µl + S9	97	98	98 ± 1	0	31	16 ± 16
200µl	32	---[b]	32	11	---[b]	11
200µl + S9	33	---[b]	33	6	---[b]	6
Nitro[d]	--[b]	1600[a]	1600[a]	125[a]	194[a]	160[a] ± 35

a = A minimum of 2X control or S9 values were obtained.

b = These dilutions were not conducted during this trial.

c = Each trial value represents a mean count of three plates.

d = 0.15µl/plate of 4-nitroquinoline-N-oxide was used.

e = 4.5µl/plate of benzo(a)pyrene.

Table 7

Ames Assay

Genotoxicity of 79-Hanna IVB (01W-00-U)

	Number of Revertants						
	TA100			TA98		
Trial[c]	1	2	$\bar{X}$ ± SEM	1	2	$\bar{X}$ ± SEM
BAP[e] + S9	800[a]	246[a]	523[a] ± 277	279[a]	110[a]	194[a] ± 84
BAP[e]	102	111	107 ± 5	31	20	25 ± 6
S9	110	97	104 ± 7	29	25	27 ± 2
Control	114	102	108 ± 6	23	15	19 ± 4
Process Water						
50µl	---[b]	89	89	---[b]	21	21
50µl + S9	---[b]	100	100	---[b]	34	34
100µl	101	100	101 ± 1	16	22	19 ± 3
100µl + S9	129	81	105 ± 24	67[a]	36	51 ± 16
200µl	---[b]	49	49	---[b]	16	16
200µl + S9	---[b]	75	75	---[b]	75[a]	75[a]
Nitro[d]	1600[a]	1800[a]	1700[a] ± 100	194[a]	226[a]	210[a] ± 16

a = A minimum of 2X control or S9 values were obtained.

b = These dilutions were not conducted during this trial.

c = Each trial value represents a mean count of three plates.

d = 0.15µg/plate of 4-nitroquinoline-N-oxide was used.

e = 5.4µg/plate of benzo(a)pyrene.

Table 8

Paramecium Assay
Genotoxicity of Tarsand 1S 03W-00-C

Percent Lethals and Detrimentals
After Autogamy

Experimental Group	Trial 1	Trial 2	Trial 3
Food	4.7	2.8	1.1
S9	3.4	2.6	2.7
Process Water			
% Dilutions			
1.3	2.9	---[b]	---[b]
1.3+S9	4.1	---[b]	---[b]
1.6	4.7	4.1	---[b]
1.6+S9	7.7[a]	0.8	---[b]
2.0	---[b]	2.5	1.9
2.0+S9	-- [b]	2.7	1.6
2.5	---[b]	---[b]	0.94
2.5+S9	---[b]	---[b]	0.96
BAP[c] + S9	3.3	2.5	3.4
Nitro[d]	63.3[a]	11.8[a]	1.3
Age of Cell	46.3	37.1	26.1

[a]These values are significantly different (P<0.05) from food and S9 only controls.

[b]These dilutions were not conducted in these trials.

[c]Benzo(a)pyrene = 50μg/ml

[d]4-nitroquinoline-N-oxide = 1.9μg/ml

Table 9

Paramecium Assay

Genotoxicity of Hanna IV-B 01W-00-U

Experimental Group	Percent Lethals and Detrimentals After Autogamy			
	Trial 1	Trial 2	Trial 3	Trial 4
Food	2.0	6.3	18.1	7.5
S9 Only	---[b]	4.7	4.6	10.5
Process Water				
% Dilution				
1.0	---[b]	4.8	9.3	---[b]
1.0+S9	---[b]	4.7	9.7	11.8[a]
1.3	3.5	5.3	9.6	5.6
1.3+S9	1.9	2.8	9.3	8.1
2.0	10.9[a]	---[b]	---[b]	---[b]
2.0+S9	1.6	---[b]	---[b]	---[b]
BAP[c] + S9	3.4	4.7	7.1	17.0[a]
Nitro[d]	---[b]	26.4[a]	61.4[a]	19.1[a]

[a]These values are significantly different from food only ($P < 0.05$).

[b]These dilutions were not tested in these trials.

[c]Benzo(a)pyrene = 50μg/ml.

[d]4-nitroquinoline-N-oxide = 1.9μg/ml.

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