Environmental Mutagenesis, Soil Studies

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ABSTRACT

Organic extracts of street soil samples from industrial, commercial and residential areas in Cainta, Makati, Mandaluyong, Manila, Marikina, Parañaque, Pasig, Quezon City, San Juan and Valenzuela were subjected to genotoxicity studies.

None of the samples exhibited direct DNA damaging potential as measured by the Rec Assay, indicating that the organic extracts of the street soil samples were not genotoxic before metabolic activation.

After metabolic activation, organic extracts of street soil samples from industrial areas of Cainta, Mandaluyong, Manila and Quezon City showed appreciable genotoxicity. The same observation was made of organic extracts from street soil samples from commercial areas of Cainta, Mandaluyong, Manila, Pasig, Quezon City and San Juan. Appreciable genotoxicity was also observed of organic extracts from street soils of residential areas in Makati, Marikina and San Juan. These were revealed by the host-mediated assay.

Chromosome breaking effects were observed in organic extracts of industrial areas in Cainta, Mandaluyong and Quezon City and also in commercial areas in Cainta, Mandaluyong, Pasig, Quezon City and San Juan. The same observation was made of

organic extracts from street soils of residential areas in Makati, Marikina and San Juan.

These observations were correlated with benzo(a)pyrene content of these street soil samples.

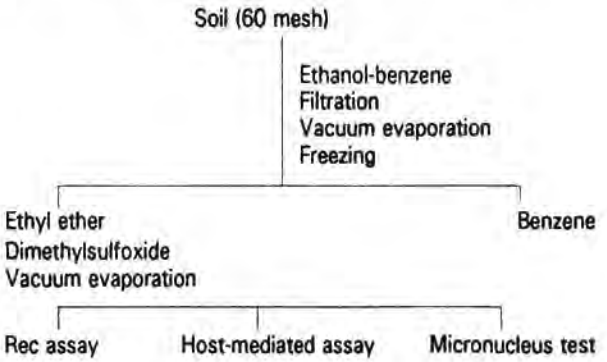
INTRODUCTION

About 75% of air pollution in Metro Manila is contributed by emissions from more than 500,000 motor vehicles operating daily (Garcia, 1987). A number of these vehicles are diesel powered which can expose the urban population to diesel exhausts which contain polycyclic aromatic hydrocarbons. Polycyclic aromatic hydrocarbons possess human carcinogenic potential. These polycyclic aromatic hydrocarbons can accumulate in street soils. Analysis of organic extracts from street soil samples can reveal the extent of mutagen pollution in industrial, commercial, and residential areas around Metro Manila.

Pollution of the environment with mutagens is of great concern. Mutagens not only induce cancer but also cause birth defects. Mutagens can also induce genetic disorders which can be transmitted from one generation to the next.

METHODOLOGY

Street soil samples were collected from ten sites in each residential, industrial, or commercial area in each district in Metro Manila. The street soil samples were sieved (60 mesh) and subjected to the following extraction procedure.



The Rec assay (Kada, *et. al.*, 1980) was used to determine the genotoxicity potential before metabolic activation.

The host-mediated assay was utilized to determine genotoxicity after metabolic activation (Legator and Gabridge, 1969). The micronucleus test was employed to study chromosome breaking effects (Schmid, 1977).

Benzo(a)pyrene was extracted from street soil samples using the method of Perolie and Bulzon (a). Ten gram samples of street soils were extracted with bexone-acetone mixture (1:1) for 17 hours in a Soxhlet extraction apparatus. The solvent extract was reduced to a small volume by a rotary evaporator. The method of thin layer chromatography on silica gel was used to separate the benzo(a)pyrene spot. The spot was scraped and eluted with 6 N NHO_3. The fluorescent intensity of the solution was measured at 545 nm using an excitation wavelength of 525 nm.

RESULTS AND DISCUSSIONS

Table 1 shows the ten districts around Metro Manila where the street soil samples were taken.

Table 1. Metro Manila districts included in the study.

DISTRICT	NUMBER DESIGNATION
Cainta	I
Makati	II
Mandaluyong	III
Manila	IV
Marikina	V
Parañaque	VI
Pasig	VII
Quezon City	VIII
San Juan	IX
Valenzuela	X

Table 2 indicates that none of the street soil sample extracts from industrial, commercial, and residential areas of these ten districts exhibited direct DNA damaging potential. This is interpreted to mean that the organic extracts of street soil samples are not genotoxic before metabolic activation. No zones of inhibition were observed even with the Rec strain of *B. subtilis* which does not possess a recombination repair system.

Table 2. Direct DNA damaging potential of organic extracts of street soil samples (Rec assay).

	ZONE OF INHIBITION (mm)					
	Industrial		Commercial		Residential	
	Rec+	Rec−	Rec+	Rec−	Rec+	Rec−
I	0	0	0	0	0	0
II	0	0	0	0	0	0
III	0	0	0	0	0	0
IV	0	0	0	0	0	0
V	0	0	0	0	0	0
VI	0	0	0	0	0	0
VII	0	0	0	0	0	0
VIII	0	0	0	0	0	0
IX	0	0	0	0	0	0
X	0	0	0	0	0	0

Positive control 4NQO	Rec+	10.40 ± 1.76
	Rec−	13.95 ± 1.36
Negative control	Rec+	0
	Rec−	0

Table 3. Mutagenicity potential after metabolic activation of organic extracts from street soil samples using the Host Mediated Assay with his G46 as indicator organism.

		MUTATION FREQUENCY (+SD)		
DISTRICT	CODE	Industrial Value ±SD	Commercial Value ±SD	Residential Value ±SD
Cainta	I	11.09 ±0.07	3.34 ±0.01	4.09 ±0.06
Makati	II	1.11 ±0.11	1.91 ±0.23	11.09 ±0.12
Mandaluyong	III	9.41 ±0.67	9.65 ±0.12	1.14 ±0.91
Manila	IV	8.98 ±0.17	4.56 ±0.54	2.45 ±0.98
Marikina	V	1.89 ±0.18	1.78 ±0.16	8.19 ±0.87
Parañaque	VI	1.96 ±0.18	2.04 ±0.18	1.09 ±0.07
Pasig	VII	1.76 ±0.16	7.98 ±0.76	1.97 ±0.68
Quezon City	VIII	8.33 ±0.16	7.98 ±0.56	2.22 ±0.98
San Juan	IX	1.87 ±0.78	8.87 ±0.45	8.69 ±0.76
Valenzuela	X	1.98 ±0.78	1.19 ±0.09	1.07 ±0.34

Positive control	(Benzo(a)pyrene	10.07 ± 1.13
Negative control	(Distilled water)	1.34 ± 0.56

The data in Table 3 depicts appreciable genotoxicity after metabolic activation of organic extracts from street soil samples of industrial districts in Cainta, Mandaluyong, Manila, Quezon City and San Juan. Appreciable mutagenicity after metabolic activation was exhibited by organic extracts from street soil samples from residential areas of Cainta, Makati, Marikina and San Juan.

Chromosome breaking effects (Table 4) were observed in organic extracts of street soil samples from:

1. Industrial areas of Cainta, Mandaluyong and Quezon City
2. Commercial areas of Cainta, Mandaluyong, Pasig, Quezon City and San Juan
3. Residential areas of Makati, Marikina, and San Juan

Table 4. Chromosome breaking effects of organic extracts from street soil samples using the micronucleus test.

		NO. OF MICRONUCLEATED POLYCHROMATIC ERYTHROCYTES PER THOUSAND					
DISTRICT	CODE	Industrial Value	±SD	Commercial Value	±SD	Residential Value	±SD
Cainta	I	10.01	±1.11	8.92	±0.98	1.09	±0.09
Makati	II	1.03	±0.04	1.11	±0.76	6.98	±0.09
Mandaluyong	III	12.04	±1.45	11.65	±0.98	0.87	±0.18
Manila	IV	2.34	±0.86	2.33	±0.78	1.18	±0.78
Marikina	V	0.96	±0.08	0.98	±0.78	8.91	±0.98
Parañaque	VI	1.16	±0.98	1.23	±0.76	1.04	±0.06
Pasig	VII	1.14	±0.98	8.78	±0.79	1.05	±0.07
Quezon City	VIII	9.66	±0.45	5.33	±0.08	2.22	±0.17
San Juan	IX	0.98	±0.15	7.89	±0.11	9.02	±0.19
Valenzuela	X	0.89	±0.03	0.99	±0.08	0.95	±0.06
Positive control		(Benzo(a)pyrene)		16.33	± 1.22		
Negative control		(Distilled water)		1.93	± 0.09		

Benzo(a)pyrene concentration was appreciable in the following:

1. Industrial areas in Cainta, Mandaluyong, and Quezon City

2. Commercial areas in Mandaluyong, Pasig, and San Juan

3. Residential areas of Makati, Marikina, and San Juan

Benzo(a)pyrene is only one of the polycyclic aromatic hydrocarbons present in diesel exhausts. It is metabolized to a bay region carbocation which alkylates DNA (Selkirk, 1984). It is a well-known mutagen and carcinogen.

Benzo(a) pyrene was found in the organic extracts of sieved street soil samples, of those that exhibited genotoxicity after metabolic activation and chromosome breaking effects. This is a consequence of pollution from automotive exhausts especially from diesel exhausts.

Table 5. Benzo(a)pyrene content of street soil samples.

		BENZO(A)PYRENE ug/g soil		
DISTRICT	CODE	Industrial Value ± SD	Comercial Value ± SD	Residential Value ± SD
Cainta	I	3.11 ± 0.87	0.37 ± 0.09	0.44 ± 0.12
Makati	II	1.77 ± 0.04	1.02 ± 0.06	2.20 ± 0.67
Mandaluyong	III	3.12 ± 0.67	5.20 ± 0.96	1.56 ± 0.19
Manila	IV	1.68 ± 0.89	1.12 ± 0.76	1.34 ± 0.65
Marikina	V	1.83 ± 0.34	0.92 ± 0.13	3.14 ± 0.45
Parañaque	VI	0.81 ± 0.13	0.22 ± 0.05	1.88 ± 0.65
Pasig	VII	1.28 ± 0.56	2.39 ± 0.43	0.40 ± 0.03
Quezon City	VIII	2.21 ± 0.12	1.72 ± 0.26	0.78 ± 0.13
San Juan	IX	1.18 ± 0.02	3.11 ± 0.14	2.39 ± 0.87
Valenzuela	X	1.01 ± 0.13	1.21 ± 0.12	0.81 ± 0.14

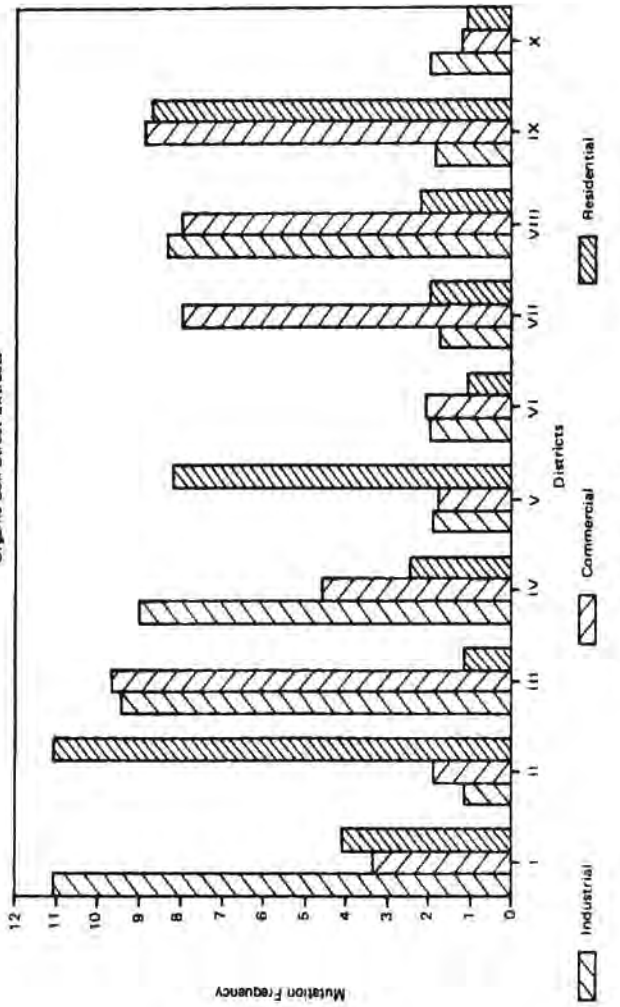


Fig. 1. HMA mutation frequency.

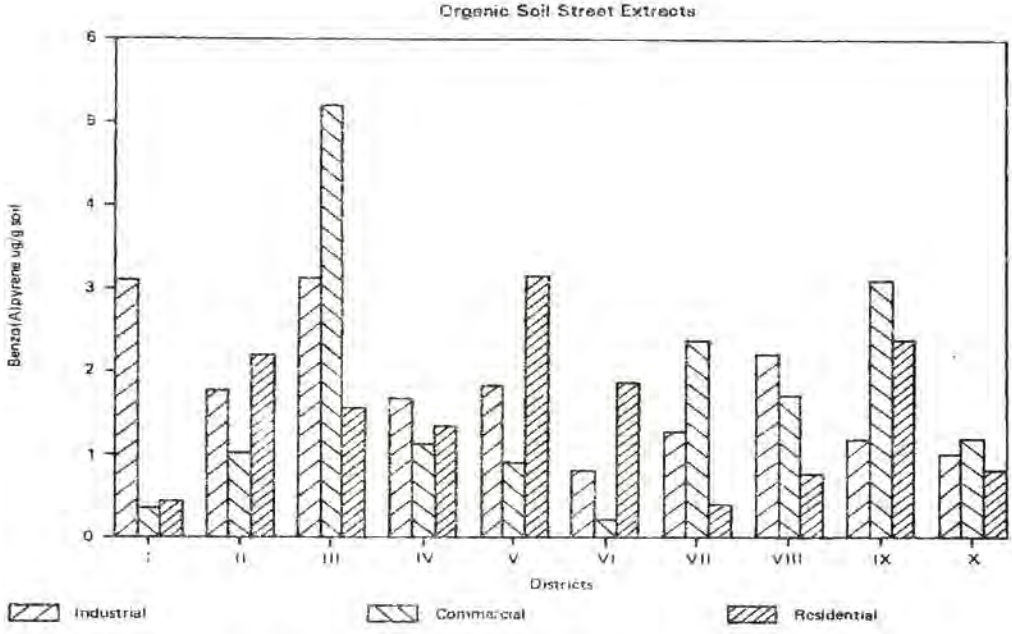


Fig. 3. Benzo(a)pyrene content of soil street samples.

CONCLUSION

Benzo(a)pyrene was detected in street soil samples that were extracted with organic solvents. These organic extracts showed mutagenic activity after metabolic activation and chromosome breaking effects.

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