Yonsei Medical Journal
Vol. 28, No. 1, 1987

Physico-chemical Nature and Mutagenic Activity of Ambient Dust in Seoul

Yong Chung

This study deals with the serious pollution of Seoul's ambient air and extends a warning regarding its adverse effects on human health. The collected dust samples exceeded the legal standard (150 $\mu g/m^3$ of total suspended particulates) of ambient air quality. Fine particles, with an aerodynamic diameter of less than 2.5 μm comprised on the average 75.4 percent of the total suspended particulates (TSP). The amount of ether extractable organic matter (EEOM) of the fine particles was found to range from 3.6 to 7.1 percent. Neutral, acidic and basic organics were fractionated. In the neutral fraction, aliphatic compounds (ALP), polyaromatic hydrocarbons (PAH), and polar neutral organic compounds (POC_N) comprised 23.1, 37.8 and 39.1 percent, respectively. Mutagenic activities of the organic fractions were determined by Ames bioassay. PAH showed the most mutagenicity using Salmonella typhimurium *TA98 strain and TA98NR (nitro-reductase activity deficient strain) in the presence of S-9 fractions.*

Key Words: Total suspended particulate (TSP), ether extractable organic matter (EEOM), neutral organic compound, acidic organic compound, basic organic compound, aliphatic compound (ALP), polyaromatic hydrocarbon (PAH), polar neutral organic compound (POC_N), Ames test, *Salmonella typhimurium* TA98, *Salmonella typhimurium* TA98NR (nitro-reductase deficient strain)

Korea is experiencing phenomenal economic growth and unprecedented urbanization. This has inevitably led to a series of disturbances, frictions, disequilibria and other undesirable side effects. The excessive density of people and economic activities in the large cities has resulted in profound changes in environmental quality, transportation patterns, and unbalanced regional growth. Korea's environmental quality management program began in earnest in 1980 when the Environmental Administration was established. There has been much debate concerning environmental pollution. Since the 1962 origination of five year economic development plans, air pollution in the metropolitan and industrial areas has exceeded the legal standards.

The legal standard is 0.05 ppm of sulfur dioxide and 150 $\mu g/m^3$ of total suspended particulates in the ambient air.

Received November 10, 1986
Accepted January 11, 1987
The institute for Environmental Research, Yonsei University, Seoul, Korea.
Address reprint requests to Dr. Y. Chung, Director, the institute for Environmental Research, Yonsei University, Seoul, Korea.
Presented at the 2nd Joint Conference of Air Pollution Studies in Asian Areas. November 18-19, 1986, Kyoto, Japan.

According to the many investigations on air pollution and its hazardous effects, many efforts to control environmental pollution have been at tempted, but the actual causative circumstances cannot always be changed due to reasons of economic growth, population growth and urbanization.

Kwon and Chung, Yonsei University, Seoul, surveyed air pollution in Seoul in 1977, and their results showed high concentrations of air pollutants such as total suspended particulates, heavy metals and benzo(a)pyrene which are known to be harmful to human health (Kwon *et al.* 1978 and 1979). Cha and Kim of Korea University, Seoul, performed the mutagenicity test of air borne particulates in Seoul in 1983 using the Ames' test. The high mutagenicity that they observed could also be very harmful to human health (Cha *et al.* 1983).

Many investigators are predicting a deterioration in both environmental quality and human health unless corrective courses of action against Seoul's air pollution are taken.

The seriousness of the effects of the air pollution compels us to issue warnings to those with chronic deseases such as respiratory deseases, carcinogenesis, mutagenesis and teratogenesis. It is necessary that the analysis of air pollutant components should be under-taken to assess the risk to human health and that adequate antipollution measurements such as the ambient quality standard of pollutants should be revised as well.

The purpose of this study was to investigate the hazardous effects of the components of suspended particulates in the ambient air of Seoul.

METHODS

Sampling of Dust and Measurement of the Physical Properties

A airborne suspended particulates were sampled for 5 or 7 consecutive days a month from January to June, 1985 in the Shin Chon area of Seoul. The sampling point was an office building at the College of Medicine, Yonsei University which is located next to a major traffic road in a residential and commercial area. Each sample was collected for 24 hrs from 9:00 AM to 9:00 AM of the next day.

The dust samples were collected by a Hivol-Cascade impact sampler (flow rate, 400 ℓ/min, Model-130, Kimoto Co. Japan) which divided the dust into fine particles (less than 2.5 μm aerodynamic diameter) and larger particles (greater than 2.5 μm).

The concentration and size distribution based on the ratio between the fine particles and the coarse were measured (Lieback *et al.* 1984; Stahlhofen *et al.* 1979).

Analysis of Chemical Components

Dust samples were extracted with cyclohexane and diethylether (2:8 V/V) (EEOM) and fractionated into basic, acidic and neutral organic compounds using ether and adjusting the pH (Fig. 1).

From the neutral fraction, aliphatic compounds, polyaromatic hydrocarbons (PAH), and polar neutral organic compounds (POC_N) were separated using thin layer chromatography (TLC).

The developing solvent for TLC was n-hexane, cyclohexane, toluene and benzene (8:34:4:52 V/V) (Ketseridis and Hahn 1975; Moriske *et al.* 1982). Monthly samples were combined and represented as one aggregate sample.

Mutagenicity Test

The mutagenicity of each of the organic subfractions was determined using the *Salmonella typhimurium* mammalian microsome bioassay by Ames (Ames *et al.* 1975; McCann and Ames 1976; Maron and Ames 1983).

For the sensitive nitro reductase deficient strain mutagencity test, TA98 and TA98NR (the nitro-reductase deficient strain) were used (Seemayer 1978;

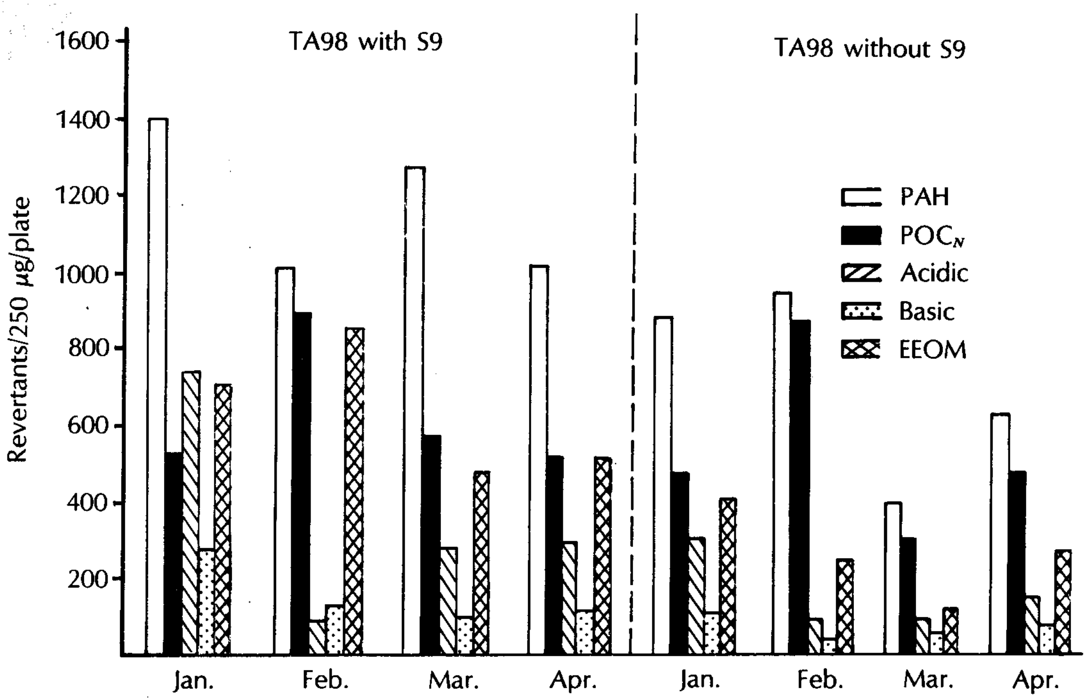

Fig. 1. *Flow diagram for the separation of organic compounds.*

Table 1. Aerodynamic component of suspended particulates in Seoul **(from Jan. to June 1985)**

(Unit: μg/m³)

Period / No. Sample / Item		Jan. 6	Feb. 5	Mar. 5	Apr. 6	May 6	Jun. 8
TSP	ave.	249.2	247.2	204.1	144.7	155.8	163.0
	max.	399.2	472.8	288.1	212.4	240.8	368.2
	min.	102.3	103.0	123.0	77.6	55.6	69.6
Fine particle (<2.5 μm)	ave.	174.2	173.6	145.3	114.0	116.4	124.6
	max.	251.2	336.7	193.2	172.4	180.8	314.9
	min.	72.7	76.4	93.6	64.2	33.5	47.8
Coarse particle (>2.5 μm)	ave.	75.1	73.6	58.8	30.7	39.4	38.4
	max.	148.0	136.1	94.9	48.1	60.0	55.5
	min.	29.6	26.6	29.4	13.4	22.1	21.8
Ratio(%) of fine particle to TSP	ave.	69.9	70.2	71.2	78.8	74.7	76.4
	max.	84.0	74.9	84.0	84.7	82.3	85.5
	min.	71.1	63.6	52.5	72.4	60.3	68.5

Table 2. Organic compounds of fine particles

(Unit: % in Average)

Item / Sample	Jan.	Feb.	Mar.	Apr.	May	Jun.	Ave.
1. EEOM	6.7	7.1	4.2	3.6	4.5	3.9	5.0
2. Neutrals/EEOM	46.7	59.7	48.2	53.4	51.2	46.6	51.0
3. Acidics/EEOM	11.1	9.8	16.9	27.9	18.5	19.5	17.3
4. Basics/EEOM	12.4	15.1	13.5	18.7	17.6	13.8	15.2

Table 3. Component analysis of neutral extracts from fine particles

(Unit: % to the neutrals in average)

Item / Sample	Jan.	Feb.	Mar.	Apr.	May	Jan.	Ave.
ALP	20.7	29.8	20.2	21.5	32.1	24.5	24.8
PAH	42.9	30.8	31.0	39.1	30.6	38.1	35.4
POC_N	36.4	39.4	48.8	39.4	37.3	37.4	39.8

Moriske *et al.* 1985).

The tests were carried out in the presence and absence of S9 mix (50 μg/plate) prepared from rats treated with Arochlor 1254. Each organic fraction was dissolved in dimethylsulfoxide (DMSO) and 250 μg of each was used per plate. Their mutagenicity was compared with those of benzo(a)pyrene in the presence of S9 and of 2-nitrofluorene in the absence of S9.

The revertants per plate were expressed as an average of triplicate plates.

RESULTS

Physical Properties

The total suspended particulates (TSP) or dust samples were separated into fine particles (less than 2.5 μm) and coarse ones as shown in Table 1. TSP concentrations were higher in January, Feburary, and March during the heating season.

The concentration of ambient air fine particles was relatively higher in winter than in spring and summer.

Chemical Composition

Using the fine particles, the ether extractable organic matter (EEOM) was determined, and the neutral, acidic, and basic components of EEOM were also determined as shown in Table 2.

EEOM of the fine particles ranged from 3.6-7.1 percent with an average of 5.0 percent. Seasonally the EEOMs were higher in January and Feburary than in the other months.

The neutral organic compounds comprised 46.6-59.7 percent of EEOM, while the acidics and the basics made up 11.1–29.7 percent and 12.4-18.7 percent of EEOM, respectively.

Aliphatics (ALP), polyaromatics (PAH) and polarorganic compounds (POC_N) were extracted from the neutrals as shown in Table 3.

Table 4. Mutagenic activities of organic fractions

(Unit: Revertants/250 μg/plate)

Strain	S9	250 μg/plate added		Jan.	Feb.	Mar.	Apr.	Ave.
TA98	+	Neutral	PAH	1,400	1,080	1,260	564	1,076
			POC_N	524	878	560	342	576
		Acidic		784	97	166	66	278
		Basic		238	160	99	91	147
		EEOM		765	824	532	162	570
		*Benzo(a)pyrene		364				
		**DMSO		33				
	–	Neutral	PAH	886	970	440	298	649
			POC_N	512	884	389	171	459
		Acidic		360	105	161	61	172
		Basic		126	54	64	47	73
		EEOM		452	362	263	102	295
		*2-Nitro-fluorene		256				
		DMSO		34				
TA98NR	+	Neutral	PAH	1,320	1,456	914	208	975
			POC_N	392	310	258	376	334
		Acidic		–	–	–	–	–
		Basic		–	–	–	–	–
		EEOM		810	688	490	209	549
		Benzo(a)pyrene		450				
		DMSO		35				
	–	Neutral	PAH	985	79	277	241	396
			POC_N	346	292	188	165	248
		Acidic		–	–	–	–	–
		Basic		–	–	–	–	–
		EEOM		462	253	215	99	257
		2-Nitro-fluorene		172				
		DMSO		23				

* 2μg/plate added ** 100μg/plate added

ALP constituted 20.2-32.1 percent of the neutrals, PAH 30.8-42.9 and POC_N 36.4-48.8, respectively.

MUTAGENIC ACTIVITY

Table 4 and Fig. 2 show the mutagenic activities of the organic fractions of the fine particulates which is referred to as respirable dust. PAH was the most mutagenic of the organic fractions tested, and PAH, POC_N, acidics and basics are shown in the order of mutagenicity. The mutagenicity of the organic fractions extracted from EEOM was approximately the same as the half-toxicity of the total samples.

The mutagenic activity of PAHs in the Ames test with the S9 fraction was more sensitive than without the S9 fraction, while no difference in the mutagenicity of POC_Ns in the presence or absence of the S9 fraction was determined.

The mutagenic activites of strains TA98 and TA98NR (nitro-reductase deficient strain) were compared by organic fractions (Table 4); for PAH and EEOM, two strains were shown to have almost the same amount of activity. Concerning POC_N, TA98 was more senstive than TA98NR.

Mutagenicities varied with monthly samples; PAHs

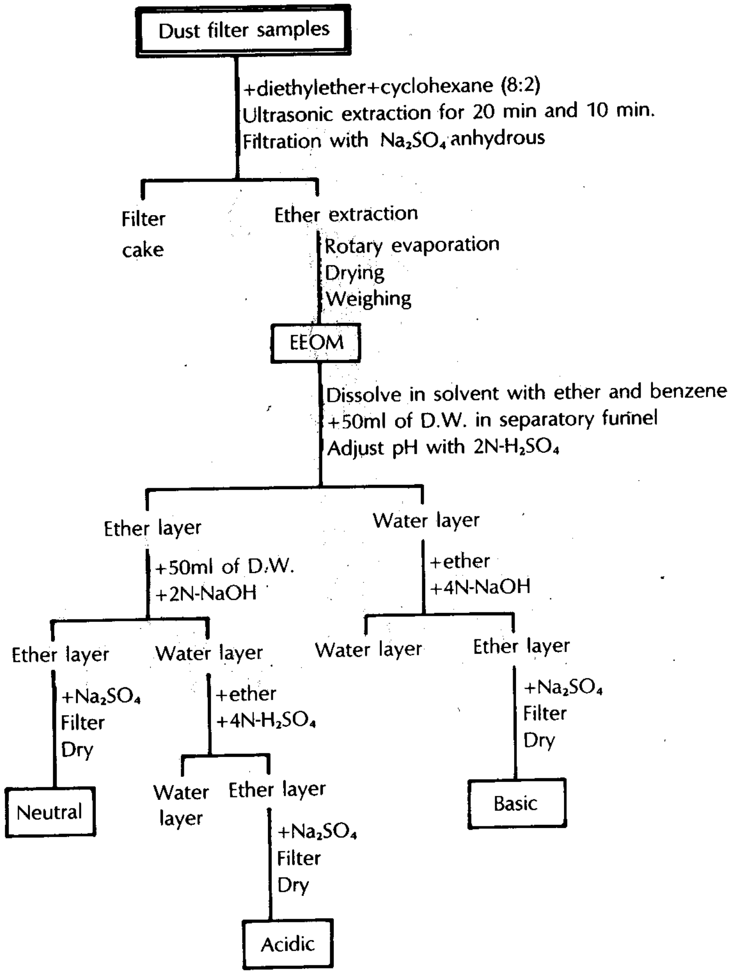

Fig. 2. Mutagenic activities of organic fractions using Salmonella typhimurium TA98 with and without S9.

tested by TA98 with S9 were more mutagenic in the January, Feburary and March samples than in the April sample.

POC_Ns from January, Feburary and March were also more mutagenic than that from April.

The January sample for acidics and basics was the most mutagenic, while the April sample for EEOM was the least mutagenic.

DISCUSSION

Although air pollution is dependent upon many kinds of energy sources, the health hazards due to air pollution are likely comparable in different areas.

Anthracite coal has been used for heating purposes since the 1950's, and this has been one of the sources of suspended particulate air pollution in Seoul.

According to the Environmental Administration's report, with the exception of a residential area, the metropolitan area of Seoul violated the legal environmental standards of air pollution with more than 150 $\mu g/m^3$ of total suspended particulate (TSP). These measurements were collected from April to Auguest 1984 (Environmental Administration).

The TSP of the residential area in this study was also close to the legal environmental standard.

In addition to measuring the concentration of TSP, an analysis of its components is necessary in order to significantly assess the health hazard. The diameter of the aerosol is an important physical characteristic. Fine particles, defined as those with a diameter of less than 2.5 μm, may be deposited in the lungs and penetrate the alveoli by diffusion. There is a bimodal distribution associated with the size of the diameter.

This author surveyed the particle size distribution of ambient air dust in the residential area of Seoul and observed the bimodal distribution of the aerodynamic diameters (Han, Chung and Kwon 1986). Usually organic pollutants are most abundant on the smallest particles (3 μm), the so-called accumulation mode (van Cauwenberghe and van Vaedck 1983).

Many investigators have already indicated that polyaromatic hydrocarbons (PAH) are highly-toxic to human beings as lung cancer agents (Menk *et als.* 1974).

Lao *et al.* (1973) identified more than 70 polycyclic aromatic hydrocarbons (PAH) in air. Some of them have been confirmed as carcinogens and mutagens (Miller 1970; Shubik and Hartwell 1969; Teranishi *et als.* 1975; Kohan and Claxton 1983; Tokiwa *et als.* 1983; Nakagawa *et als.* 1983; MØller and Alfheim 1983; Fukino *et als.* 1982; Alink *et als.* 1983).

This author *et al.* had previously determined the benzo(a)pyrene content of Seoul's ambient dust in 1977 and compared its concentration (5.8-10.9 μg/m³) to that of other foreign cities (Kwon, Chung and Lim 1978). Cha *et al.* (1983) examined the mutagenic activity of methanol extracts from the airborne particulates of Seoul and warned of the hazardous effects to residents health. Nevertheless, although the above results cannot be extrapolated directly to adverse effects on human health, it is important that air pollution should be controlled.

The airborne dust components of Seoul In this study were compared with those of Berlin and Kobe: the Berlin's dust was measured at the Technical University and Free University, Berlin in 1983-1984 (Moriske *et al.* 1985) and Kobe's measured by the Public Health Institute of Kobe in 1975 (Teranishi 1975) as shown in Table 6.

The TSP of Seoul was higher than Berlin's, while the ether extractable organic matter (EEOM) of Seoul was less than Berlin's.

This means Seoul dust has more inorganic components, which might be caused by an emission source of air pollutant, climatological, or topographic condition.

While the neutral fraction from EEOM of Seoul's dust was less than Berlin's, the basics of the Seoul dust was the highest among three areas. Seoul's aliphatics (ALP) were the same as Kobe's and were less than Berlin's. Seoul presented the highest PAH content while Kobe's POC_N was the least.

The dissimilar values of the components suggest different toxicities. When the mutagenicities were compared using the same amount of extract samples (250 μg/plate) and the same strain TA98, the POC_N fraction of Seoul's dust was similar to Berlin's but the PAH of Seoul's dust was much greater as shown in Table 7.

The different results from the Ames test with the same organic fractions could also illustrate that chemical component of the fractions may be different as well.

The Berlin dust, from a domestic area, had higher

Table 5. A comparison of dust components among Seoul, Berlin and Kobe

Item		Seoul	Berlin	Kobe
Period sampled		Jan.-June 1985	March 1983-Jan. 1984	Apr.-May 1975
TSP	(μg/m³)	55.6–472.8	19.3–153.2	
EEOM	(range %)	3.6– 7.1	4.9– 47.9	
		5.0	19.2	
Neutrals to EEOM	(range %)	46.6– 59.7	62.6– 95.5	54.7
	(ave. %)	51.0	85.0	
Acidics to EEOM	(range %)	9.8– 27.9	2.4– 28.9	23.6
	(ave. %)	17.3	12.3	
Basics to EEOM	(range %)	12.4– 18.7	0.4– 28.8	1.9
	(ave. %)	15.2	4.3	
ACP to Neutrals	(range %)	20.2– 32.1	8.5– 57.6	25.3
	(ave. %)	24.8	34.3	
PAH to Neutrals	(range %)	30.8– 42.9	3.4– 36.4	11.8
	(ave. %)	35.4	18.0	
POC_N to Neutrals	(range %)	36.4– 48.8	28.4– 62.8	17.6
	(ave. %)	39.8	47.4	

Table 6. A comparison of mutagenicity among dust components from Seoul, Berlin and Kobe

(Unit: Revertants/250 μg/plate)

Strain	Organics	Seoul	Berlin	Kobe
	EEOM	162– 824	116– 567	147
	POC_N	342– 878	188–1,027	
TA98	PAH	564–1,400	33– 365	
+S9	Acidic	66– 784	33– 607	101
	Basic	91– 238	16– 236	147
	EEOM	102– 452	42– 262	
	POC_N	171– 884	54– 555	118
TA98	PAH	298– 970	0– 19	192
–S9	Acidic	61– 360	72– 372	
	Basic	47– 126	11– 84	
Remark		ether extracts	ether extracts	benzene extracts

concentrations of 2-fluorenaldehyde, anthrone, anthrachinone, and 2-nitrofluorene. The compound primarily identified from the coal-firing emissions in Berlin was 2-fluorenone, while in the heavily trafficed sampling area, acenaphthenchinone, anthrachinone, benzanthrone and 1-nitropyrene also were identified (Moriske 1985).

Energy sources, combustion devices and conditions of combustion should be analyzed and based on the identification of the pollutants, the appropriate conditions should be employed to control the emission of the hazardous compounds.

It is suggested that in the future the particulate matter emitted from different sources such as automobiles, petroleum burners, anthracite burners etc. from different areas should also be analyzed for chemical components and tested for mutagenicity.

ACKNOWLEDGEMENTS

Author expresses sincerely gratitude to professor Rüden H. and Dr. Moriske HJ, Institute für Allgemeine Hygiene der Freien Universitut Berlin, W-Germany.

REFERENCES

Alink GM, Smit HA, Vanhoudt JJ, Kolkman JR, Boleij JSM: Mutagenic activity of airborne particulates at non-industrial locations. *Mutation Res 116:21-34, 1983*

Ames BN, Mc. Cann J, Yamasaki E: Methods for detecting carcinogens and mutagens with the *Salmonella/mammalian* microsome mutagenicity test. *Mutation Res 31:347-364, 1975*

Cha CW, Kim YW, Kim JS: Study on mutagenicity of the air pollutants in Seoul. *J Kor Env Preserv Assoc 4:167-179, 1983*

Environmental administration, Korea government: *Air pollution and its control*, Environmental Preservation (Annual report) 1984, p. 192.

Fukino H, Mimura S, Inoue K, Yamane Y: Mutagenicity of airborne particles. *Mutation Res 102:237-247, 1982*

Han EJ, Chung Y, Kwon SP: Particle size distribution of suspended particulates in the atmosphere of a Seoul residential area. *Korean J of Preventive Med. 19:130-136, 1986*

Ketseridis G, Hahn J: Bestimmung der organischen Bestandteile von Aerosolpartikeln aus Reinluft. *Z Analy Chem. 273:257-261, 1975*

Kohan M, Claxton L: Mutagenicity of diesel-exhaust particle extract, 1-nitropyrene and 2.7-dinitrofluorenone in *Salmonella typhimurium* under various metabolic activation conditions. *Mutation Res 124:191-200, 1983*

Kwon SP, Chung Y, Lim DK: Study on benzo(a)pyrene of suspended particulate in atmosphere of Seoul City. *Korean J Preventive Med 11:65-75, 1978*

Kwon SP, Chung Y, Lim DK: Heavy metals of the suspended particulate in atmosphere of Seoul city. *Korea J Preventive Med 12:49-55, 1979*

Lao RC, Thomas RS, Oza H, Dubois L: Application of a gas chromatograph-mass spectrometer-data processor combination to the analysis of the polycyclic aromatic hydrocarbon content of airborne pollutants. *Anal Chem 45:908-915, 1973*

Lieback JU, Bauer HW, Rüden H: Uber eine geeignete Korngrößenfraktionierung von Schwebstäuben aus medizinisch-lufthygienischer Sicht. *Staub-Reinhalt Luft 44:221-225, 1984*

McCann, Ames BN: Detection of carcinogens/mutagens in the Salmonella/microsome test. Assay of 300 chemicals: Discussion. *Proc Nat Acad Sci USA 73:950-954, 1976*

Maron DM, Ames BN: Revised methods for the *Salmonella* mutagenicity test. *Mutation Res 113:173-215, 1983*

Menk HR, Casagrande JT, Henderson BE: Industrial air pollution possible effect of lung cancer. *Science 183:210-212, 1974*

Miller JA: Carcinogenesis by chemicals, an overview-G.H.A. *Cancer Res 30:559-576, 1970*

Moriske HJ, Block I, Schleibinger H, Rüden H: Polare neutrale organische Verbilndunger (POC_N) in Stadtaerosol: Chemische Charakerisierung und mutagene Wirksambeit in Abhängigkeit verschiedener Quelleinflusse. *Zbl Bakt Hyg I Abt Orig B 181:240-271, 1985*

Moriske HJ, Wullenweber M, Ketseridis G, Rüden H: Untersuchungen zur mutagenen Wirksamkeit von polycyclischen aromatischen Kohlenwasserstoffen (PAH) und polaren organischen Verbindungen (POC_N) im Stadtaerosol. *Zbl Bakt Hyg I Abt Orig B 176:508-518, 1982*

MØller M, Alfheim I: Mutagenicity of air samples from various combustion sources. *Mutation Res 116:35-46, 1983*

Nakagawa R, Kitomori S, Horikawa K, Nakashina K, Tokiwa H: Identification of dinitropyrenes in diesel-exhaust particles. Their probable presence as the major mutagens. *Mutation Res 124:201-211, 1983*

Seemayer N: *Die Zellkultur als Testsystem "in nitro" zum Nachweis von kanzerogenen Stoffen*. Gas. z. Förderung der Lufthygiene und Silicoseforschung e.v., Düsseldorf, Jahresbericht Bd. *1:65-79, 1978*

Shubik P, Hartwell JL: *Survey of compounds which have been tested for carcinogenic activity*, US Department of Health, Eduction and Welfare Public Health Service, ed JA Peters, Supplement 2, 1969

Stahlhofen W, Eckard B, Gethard J, Heyder J, Stuck B: Measurement of the exhalathoratic, tracheobronchial and alveolar deposition of aerosol particles in human respiratory tract. 7. Knof. d. Gesellsch. f. Aerosolforschung, Kongreßbericht, Düsseldorf 234-235, 1979

Teranishi K, Hamada K, Watanabe H: Quantitative relationships between carcinogenicity and mutagenicity of polyaromatic hudrocarbons *Salmonella typhimurium* mutants. *Mutation Res 31:97-102, 1975*

Idid: Mutagenicity in *Salmonella typhimurium* mutants of benzendsoluble organic matter derived from air-borne particulate matter and its five fractions. *Mutation Res 56:273-280, 1978*

Tokiwa H, Kitomors S, Nakagawa R, Horikawa K, Matamala L: Demonstration of a powerful matter. *Mutation Res 121:107-116, 1983*

Van Cauwenberghe K, van Vaeck L: Toxicological implications of the organic fraction of aerols: a chemist's view. *Mutation Res 116:1-20, 1983*