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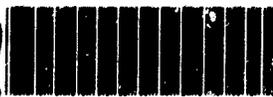
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September 22, 1986

ANGUS

Mr. Louis Borghi
Dynamac Corporation
The Dynamac Building
11140 Rockville Pike
Rockville, MD 20852

Contains No CP

Dear Mr. Borghi:

At long last, I have received the final report on the chronic inhalation of nitroethane. Enclosed is a copy of the report without appendixes, which increase the report to 661 pages, except for page 370 which is the tumor incidence summary page from the pathology appendix.

Please let me know if you wish any further information. Incidentally, Doctors Griffin, Coulston, and Stein are preparing a version of this report for publication in a journal.

Very truly yours,

Allen F. Bollmeier, Jr.
Manager, Regulatory Affairs

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Enclosure

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COULSTON INTERNATIONAL CORPORATION
White Sands Research Center
Final Report
CHRONIC INHALATION OF NITROETHANE
Volume I

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COULSTON INTERNATIONAL CORPORATION

White Sands Research Center

2512 Christina Place

Alamogordo, New Mexico 88310

FINAL REPORT

STUDY NO. 820302

CHRONIC INHALATION OF NITROETHANE

June 9, 1986


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TABLE OF CONTENTS

	Page
1. SUMMARY	1
2. INTRODUCTION	2
3. METHODS AND MATERIALS	3
3.1. Experimental Animals	3
3.2. Animal Husbandry	4
3.3. Experimental Design	5
3.4. Test Material	6
3.5. Chamber Operations	7
3.6. Observations	8
3.7. Pathology	10
3.8. Statistical Analyses	11
4. RESULTS	12
4.1. Conditions of Exposure	12
4.2. General Observations	13
4.3. Body Weights	14
4.4. Hematology	16
4.5. Serum Chemistry	17
4.6. Organ Weights	18
4.7. Pathology	20
5. DISCUSSION	23
5.1. Experimental	23
5.2. Pathology	25
6. CONCLUSIONS	28

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TABLE OF CONTENTS (Continued)

	Page
Table I -- Weekly means of body weights (grams)	29
Table II -- Hematology of rats exposed to nitroethane at 100 ppm or 200 ppm for 2 years	31
Table III -- Serum chemistry of rats exposed to nitroethane at 100 ppm or 200 ppm for 2 years	32
Table IV -- Organ weights of rats surviving to the terminal sacrifice	34
Appendix A -- Chamber Concentration Data	35
Appendix B -- Body Weight Data	39
Appendix C -- Hematology	174
Appendix D -- Serum Chemistry	189
Appendix E -- Organ Weights	212
Appendix F -- Pathology	247

1. SUMMARY

Male and female Long-Evans rats were exposed by inhalation to vapors of nitroethane (NE) at either 100 ppm or 200 ppm, seven hours per day, five days per week for two years. General observations were made daily and body weights were obtained weekly or biweekly.

During the study any rats that were found dead or sacrificed moribund were given a thorough gross examination and tissues retained for microscopic examination. After two years of inhalation of NE, all surviving rats were sacrificed. Blood samples were obtained from selected individuals for hematology and serum chemistry studies. All rats were examined histopathologically.

Exposure of the rats to NE had no pharmacologic effects nor were there any effects on mortality of rats of either sex at either level of exposure. Body weights of both sexes of both exposed groups were slightly less than controls, but lack of a well-defined dose-response relationship suggested the involvement of factors other than just exposure to NE. There were no effects of exposure to NE on hematology. There were no biologically significant effects of exposure to NE on clinical chemistry or on organ weights. There was no significant difference in the non-neoplastic or neoplastic pathology related to exposure to NE.

2. INTRODUCTION

Nitroethane (NE) is an industrial solvent produced in a mixture of nitroparaffins from the vapor phase nitration of propane and purified by distillation. Although the material is a liquid at room temperature, the most likely route of exposure of industrial workers is by inhalation of vapors during various handling procedures.

In the present report, the potential effects of chronic inhalation exposures to NE have been examined. Rats were subjected to a lifetime of exposures to one of two atmospheric concentrations of NE, each of which was well above usual levels of human industrial exposure. At the end of two years of these exposures, all surviving animals were sacrificed and a thorough clinical and anatomic pathological examination was performed on all animals in the study. Particular care was given to an examination of the effects of exposure to NE on the liver, a common target organ for toxic effects of some industrial solvents.

3. METHODS AND MATERIALS

3.1. Experimental Animals

The laboratory animals selected for this investigation were Long-Evans rats having the strain designation of BLU:(LE)BR and were obtained from the Blue Spruce Farms, Altamont, N.Y. After arrival at the laboratory, the animals were quarantined and acclimated to the laboratory conditions.

3.2. Animal Husbandry

During the investigation all animals were individually housed in stainless steel wire mesh cages and were maintained in air-conditioned quarters except during the periods of exposure. A standard laboratory animal diet and water were freely available to the animals except during the daily exposure period. During exposure, food and water were removed from the cages of all animals including controls in order to reduce alimentary exposure to NE.

3.3. Experimental Design

One hundred twenty one (121) male and one hundred nineteen (119) female rats were randomly assigned to the following groups:

<u>Group</u>	<u>Male</u>	<u>Female</u>	<u>NE conc. (ppm)</u>
CONTROL (I)	40	40	0
EXPOSED (II)	40	40	100
EXPOSED (III)	41	39	200

The intent was to assign 40 animals of each sex to each group, but after exposure was begun, it was determined that one animal assigned to group III - female was actually a male. The rats assigned to exposure groups (II and III) were exposed to appropriate concentrations of NE for seven hours daily, five days each week, for two years.

3.4. Test Material

The test material was nitroethane (NE), CAS 000079243, and was supplied by International Minerals and Chemical Corporation (later, ANGUS Chemical Company). The lot number used for the investigation was 2J05-5B and the analysis, as supplied by the manufacturer, was:

Nitroethane	97.92%
Nitromethane	0.01%
2-Nitropropane	2.07%

3.5. Chamber Operations

Vapors of NE were generated by bubbling purified nitrogen through liquid NE in an all-glass vessel maintained in a thermostatted water bath at a temperature of 45°C. Sufficient liquid NE to maintain a constant liquid level in the generator was added automatically.

The exposure chambers were 5 ft. wide, 5 ft. high and 5 ft. deep with tapering conical sections above and below. Chamber contents were exhausted from the lower section. The effluent from the NE vapor generators were conducted to a vortex part of the upper conical section where it was mixed with filtered air-conditioned air. The chambers were operated in the open dynamic mode.

The concentration of NE within the chamber was monitored using a MIRAN IA infra-red gas analyzer. Concentrations were monitored at least three and usually four times each day.

3.6. Observations

The animals were observed daily for general appearance and for signs of pharmacologic, behavioral or other toxic effects of exposure to NE. Moribund animals were sacrificed and these and any animals found dead were subjected to the pathological examination described below.

The animals were weighed weekly during the first six months of the study and at two-week intervals thereafter.

At the time of the terminal sacrifice, blood samples were obtained from 10 male and 10 female rats for clinical laboratory studies of hematology and serum chemistry. Hematologic parameters examined included:

Erythrocyte Count (RBC) - $\times 10^6/\text{mm}^3$
Royco Cell Crit 921 cell counter

Leucocyte Count (WBC) - $\times 10^3/\text{mm}^3$
Royco Cell Crit 921 cell counter

Mean Corpuscular Volume (MCV) - fL
Royco Cell Crit 921 cell counter

Packed Cell Volume (HCT) - %
Royco Cell Crit 921 cell counter

Hemoglobin (HGB) - g%
Cyanmethemoglobin Colorimetric
Method using Chemetrics Analyzer II

Serum chemistry parameters included:

Serum Glutamic-Oxaloacetic transaminase,
also known as serum aspartate amino-
transferase, (SGOT) - U/L
Modified Amador and Wacker Method using
Chemetrics Analyzer II

Serum Glutamic-Pyruvic transaminase,
also known as alanine aminotransferase,
(SGPT) - U/L
Modified Henry Procedure using Chemetrics
Analyzer II

Total Bilirubin (BILI) - mg/dl
Diazo Method using Chemetrics Analyzer II

Total Protein (PROT) - g/dl
Biuret Method using Chemetrics Analyzer II

Blood Urea Nitrogen (BUN) - mg/dl
**Modified Urease Technique using Chemetrics
Analyzer II**

Creatinine (CREAT) - mg/dl
**Modified Fabini and Ertingshauser using
Chemetrics Analyzer II**

Sodium (NA) - meq/l
Flame photometry

Potassium (K) - meq/l
Flame photometry

3.7. Pathology

Full necropsies were performed on all animals found dead or sacrificed moribund and on all remaining animals surviving two years. Each of the animals was given a thorough gross examination and the following organs were weighed: brain (including cerebellum and medulla), liver, kidneys, lungs and heart.

The following organs and tissues were examined, removed and fixed in 10% buffered formalin for processing and preparation of slides for microscopic examination:

liver, heart, lung, artery or aorta, lymph nodes, thymus, spleen, salivary gland, pancreas, kidneys, urinary bladder, mammary glands, trachea, thyroid, esophagus, stomach, colon, intestine, adrenals, eye, pituitary, brain (including cerebrum, cerebellum and medulla), muscle, nerve, bone, bone marrow, skin and subcutis, spinal cord. For males: testes, prostate, epididymas, seminal vesicles. For females: ovaries, uterus, cervix, oviduct.

3.8. Statistical Analyses

a) Chamber concentrations - the daily concentration of NE in each chamber was calculated as the arithmetic mean of the individual concentration measurements. Concentrations for each week of the study were calculated as the mean of the daily concentrations.

b) Body weights - means and standard deviations of the weekly (or bi-weekly) body weights of each sex and group were calculated. Comparisons were made between the control and exposed groups using Student's "t" test.

c) Organ weights and clinical laboratory data - organ weights were expressed as both absolute weight (grams) and weight relative to the whole body weight (% of body weight). Means and standard deviations of each of these parameters and the parameters of clinical chemistry and hematology were calculated. Variances were tested for homogeneity using Bartlett's test, and in cases where the variances proved to be homogeneous, an analysis of variance (ANOVA) was performed. If the ANOVA indicated statistical significance between means at $p=.05$, Duncan's Multiple Range test was used to determine which pairs of means were significantly different.

4. RESULTS

4.1. Conditions of Exposure

The average daily measured concentrations of NE in the air of the exposure chambers is presented in Appendix A (p. 35). This appendix also shows the mean concentrations of NE for each week of the two years duration of the experiment. The grand average of the weekly means of the chamber operated at a nominal level of 100 ppm (Group II) was 99.8 ppm, and of the chamber operated at 200 ppm (Group III) was 199.2 ppm. The altitude of the site of the experiment (Alamogordo, New Mexico) is 1350 meters, and at 25°C, a concentration of 99.8 ppm is equivalent to 263 mg of NE per cubic meter of air. Similarly, a concentration of 199.2 ppm is equivalent to 525 of NE per cubic meter of air.

4.2. General Observations

During the two years of exposure to NE, the rats tolerated the exposures well and did not display pharmacologic or other overt effects of exposure to the nitroparaffin. The number of animals surviving the full two years of exposure to NE was approximately the same among the various control and exposed groups, although the largest number (and percent) of surviving animals was among both male and female rats exposed to NE at 200 ppm. Survival is shown in the table below:

	<u>Group</u>	<u>Initial Number</u>	<u>Number Surviving</u>	<u>Per Cent Surviving</u>
Males	I	40	20	50.0
	II	40	19	47.5
	III	41	24	58.5
Females	I	40	17	42.5
	II	40	17	42.5
	III	39	25	64.1

4.3. Body Weights

Mean body weights of the animals during the study are shown in Table I (p. 29). Individual body weights, and statistical data including means, standard deviation, number surviving at each weighing interval, and Student's "t" test of significance between pairs of means, is presented in Appendix B (p. 39). Generally, after exposure was initiated, the mean body weights of exposed groups of rats was less than the mean body weights of the control groups, although the difference was small. At the selected probability (p) value of 0.05, the "t" test indicated statistical significance throughout the study between Group I (control) and Group II (exposed - 100 ppm) males, although, surprisingly, statistical significance between Group I and Group III (exposed - 200 ppm) males occurred only during weeks 6-15 and occasionally thereafter. Among the females, statistically significant differences were observed throughout the entire study between Group I and Group III but only occasionally between Group I and Group II animals. The lack of a well-defined relationship between body weight and exposure concentration of NE, at least among the males, suggests that factors other than exposure to NE may have been involved. Although every attempt was made to duplicate conditions between exposed and control groups, including removal of food during exposure periods, the control animals were not housed in an exposure chamber during the exposure periods. This small difference in treatment of the groups may have influenced the body weights.

During weeks 88-92, a small decrease in mean body weights was observed among all groups, including controls. Clinical signs observed among the animals, and among other rats in the colony concurrently but not assigned to this study, were consistent with an intercurrent infection of sialodacryoadenitis virus. This condition causes swelling of salivary glands among some individuals, as well as reduced food consumption and weight loss, but is otherwise a mild and self-limiting infection. Mortality is very low and did not appear to influence mortality rates in the present study.

4.4. Hematology

At the termination of the investigation, blood samples were obtained from selected animals for studies of hematology. These data are summarized in Table II (p. 31). Individual animal data are presented in Appendix C (p. 174). The appendix also presents statistical data including mean, standard deviation, number of individuals tested, a test for homogeneity of variances, and, if appropriate, an analysis of variance (ANOVA). At the selected probability level of 0.05, there was no effect of exposure of male or female rats to 100 ppm or 200 ppm of NE on erythrocyte count, packed cell volume, mean corpuscular volume, or hemoglobin. Heterogeneous variances precluded ANOVA calculations of male leucocyte count data if all values were included, but an inspection of the data showed that one outlying value in Group I caused the heterogeneity. If this value is eliminated, the ANOVA calculation is permitted and no significant differences between any pairs of means is indicated. Similarly, a single outlying value in a Group III animal prevented ANOVA calculation of the female leucocyte count data, but if eliminated, allowed the ANOVA calculation which showed no statistically different means.

4.5. Serum Chemistry

Clinical chemistry determinations on blood samples obtained at the termination of the investigation are shown in Table III (p. 32). Individual animal data are shown in Appendix D (p. 189), which also presents mean, standard deviation, number of individuals tested, the test for homogeneity of variance and the ANOVA, if permitted. There was no effect of exposure of male or female rats to either 100 ppm or 200 ppm of NE on glutamic-oxaloacetic transaminase (SGOT), glutamic-pyruvic transaminase (SGPT), total bilirubin, sodium or potassium. In the case of SGOT and SGPT calculations of ANOVA among females, a single outlying value was eliminated from Group III. There was no significant difference between means of total protein among male rats, but at $p=0.05$, there was a slight but significant elevation of total protein among Group III (200 ppm) females as compared to Group I (controls). Elevations of blood urea nitrogen (BUN) of two male rats in Group II (100 ppm) prevented ANOVA calculations, but there was no difference between Group I (controls) and Group III (200 ppm). There was a statistically significant difference in BUN between Group I (control) females and Group III (200 ppm) females, but not between Group I and Group II (100 ppm). Two males in Group II with elevated creatinine levels prevented ANOVA calculation but an examination of the means does not suggest an effect of NE. There was no significant difference between means of creatinine levels among females.

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4.6. Organ Weights

During pathological examination of animals, major organs were weighed. Individual animal organ weights expressed as the absolute weight and as a fraction (percent) of the total body weight are presented in Appendix E (p. 212). Statistical calculations were performed on the organ weight data from animals surviving to the terminal sacrifice. This included determination of mean, standard deviation, a test for homogeneity of variance and ANOVA where applicable. These calculations are also presented in Appendix E and are summarized in Table IV (p. 34). An examination of the data summarized in Table IV does not suggest any biologically significant effect of NE on weights of the major organs. Although heterogeneous variances restricted ANOVA calculations of most liver data, there was no evidence of an effect on liver weights, and it can be said with certainty that there was no enlargement or increase in liver weight resulting from inhalation of NE. There was no statistically significant effect on absolute or relative kidney weights of male rats nor of absolute kidney weights of female rats. With regard to relative kidney weights of the females, at $p=0.05$ there was a statistically significant difference between Group I (controls) and Group II (100 ppm exposed) but not between other groups. However, the difference between the two means is very small; there was no correlation with exposure level, and the finding is considered spurious. Among males, absolute brain weights but not relative brain weights displayed a statistically significant difference between Group I

(controls) and Group II (100 ppm exposed). Among females there were no significant differences in absolute weights, but between Group I (controls) and both exposed groups, there were statistically significant differences at $p=0.05$. Since brain weights are very constant within similar groups of animals, the relative brain weight differences among females may be a reflection of the previously noted differences in total body weight. There were no significant differences in absolute or relative weights of heart or lungs among animals of either sex.

4.7. Pathology

At the completion of two years of exposure to NE, all surviving rats were sacrificed and a complete necropsy was performed on each. Complete necropsies were also performed on all rats that were either found dead or sacrificed moribund. Tissues were examined microscopically and the results of this examination are presented in Appendix F (p. 247). Utilizing electronic data processing techniques, various tabulations of the histopathologic findings were generated. The "Totals" tables (see p. 248) present the findings as fractional numbers, the numerator showing the incidence of the finding and the denominator showing the number of tissues or organs examined. The incidence in the total study is shown followed by the incidence in each treatment group. Furthermore, the "Totals" tables show incidence of findings in both sexes combined and then in each sex separately. The "Totals" tables are also divided into non-neoplastic and neoplastic categories.

As a second means of presenting the data, the findings are listed in "incidence" tables (see Appendix F, Contents, p. 247). The total number of animals in each sex and treatment is shown at the top of the column and the frequency of the finding is shown below followed by the animal identification number of each animal having the finding. The incidence tables are also separated into non-neoplastic and neoplastic findings. A summation table displays the distribution of tumors in several categories.

As a quality assurance measure, the computer also generates

the incidence of "Non-Pathology" findings (pp. 371-372). These include the incidence of autolyzed tissues and tissues which could not be found or identified even after multiple examinations of the container.

As a final means of presenting the histopathologic data, all findings for each animal in the study are listed (pp. 373-661).

An examination of the data in the tables indicated that the expected incidence of age-related degenerative diseases was seen in approximately equal frequencies in all groups. The incidence of nodular hyperplasia and adenomas of the pituitary gland associated with the endocrine target organ response was similar in all groups.

There was an increased incidence of bronchopneumonia in Group I (controls) as compared to Group II (100 ppm exposed) and III (200 ppm exposed).

The total number of tumors (p. 370) in the control group was 46 in males and 46 in females. In treated Group II there were 44 tumors in males and 56 tumors in females, which in treated Group III there were 33 tumors in males and 45 tumors in females. These results indicated that in all treated groups, except Group II females, there were fewer tumors than in the control groups. This increase in tumors in the females of Group II appeared to be related to the increased frequency of pituitary adenoma-endocrine complex related tumors.

In the control rats there were foci of hyperplasia in the liver manifested by compact, small hepatocytes frequently with amphoteric cytoplasm. In other control animals, there were foci

of nodular hyperplasia in which increased number of hepatocytes, frequently with larger hyperchromatic nuclei and prominent nucleoli, were found. Both of these types of focal lesions were multicentric and microscopic and therefore did not present a gross mass lesion. In the control male rats, there were eight (8) instances of focal hyperplasia and four (4) instances of focal nodular hyperplasia of the liver. In the control female rats there was one (1) instance of focal hyperplasia and one (1) instance of nodular hyperplasia (with gross) of the liver.

In Group II there was one (1) male instance of nodular hyperplasia of the liver and no malignancies.

In Group III there were two (2) male rats with nodular hyperplasia of the liver and one (1) hepatocarcinoma in a female rat.

These data do not indicate any significant hepatic pathologic difference between the control and exposed (NE) rat groups, but they do indicate a normal spontaneous risk of hepatic nodules in aged rats.

5. DISCUSSION

5.1. Experimental

In this study of the inhalation of NE by rats, the animals were exposed to either 100 ppm or 200 ppm of NE for seven hours daily, five days per week for two years. This exposure regimen approximates that encountered in industrial settings, and the two year period represents essentially a lifetime of such exposures. Using the published respiratory minute volume of 0.073 l/min for rats weighing 113 grams¹, it can be calculated that the rats were exposed to about 71 mg/kg or 141 mg/kg of NE daily for the exposure levels of 100 ppm or 200 ppm, respectively. These relatively high levels of exposure had remarkably little effect on the animals even though the exposures continued for nearly the entire life-span of the animals. Mortality was not affected by exposure to NE. There appeared to be an effect (slight reduction) on the body weight of the exposed animals, but lack of a clear dose-response relationship made this finding difficult to interpret.

There were no changes in hematologic patterns in the animals exposed to NE nor were there any clearly defined effects on serum chemistry. Of particular interest was a complete lack of an elevation of the serum transaminases. The transaminases frequently become elevated when chemically induced liver injury occurs.

¹-----
Biology Data Book, P.L. Altman and D.S. Dittmer, Eds., FASEB, 1964, p. 220.

The lack of a chemically injurious effect of NE on liver tissue was also demonstrated by failure of NE at either exposure level to induce liver enlargement. In fact, the only observed changes in organ weights, especially relative organ weights, may have been related more to lower whole body weights in exposed groups than to an elevation of the organ weight.

The thorough pathologic examination of tissues and organs, with care and emphasis placed on examination of the liver, provided further evidence of a lack of effect of NE on the animals. The histopathologic examination disclosed the usual age-associated degenerative diseases and the endocrine target organ response to pituitary hyperplasia, also age-related. Nodular hyperplasia of the liver was rare, but it occurred in all groups.

5.2. Pathology

The Nutrition Foundation in the United States recently published (1983) a report of an international expert advisory committee entitled "Relevance of Rodent Liver Hepatoma to Human Carcinogenic Risk." This report pointed out that rat and mouse liver react differently to chemicals regarding the formation of focal histologic changes to hepatocellular carcinoma. They stated that the term "hepatoma" should be used to describe nodular formations and not to call these and other proliferative lesions in the mouse liver cancer or precancerous. These hepatomas rarely, if ever, invade tissues, metastasize or interfere with the life span of the mouse. Most pathologists have considered them to be benign histologically, and called them adenomas or nodules of hypertrophy and hyperplasia.

The argument that only mice and rats and perhaps hamsters should be used in determining carcinogenic potential in a bioassay, because the life span is relatively short and large numbers of animals are available for statistics, is being questioned today. Animals used in carcinogen bioassays should receive the chemical in the manner for which it is intended to be administered and the material should be absorbed, metabolized, excreted or stored and have a pharmacologic or toxicokinetic pattern similar to man. The sites at which toxicity and tumors occur should be similar to those found in man. In evaluation of new chemicals or drugs, there is available a reasonable amount of clinical pharmacokinetic and metabolic information, while in older drugs or chemicals in long

use, there may be information in relation to accidental high-dose exposure in animals or man. Long term, low-dose exposures may have been evaluated to some degree in epidemiologic studies.

Factors which influence laboratory animal spontaneous tumor profiles include species, strain, sex, age, and source of the experimental test animal; dietary and environmental conditions. Also, the qualifications and experience of the study pathologist must be considered as well as diagnostic criteria and nomenclature conventions, including quality assurance and review procedures.

Reading slides in a study as they come along, through deaths or interval sacrifices, is acceptable for preliminary observations. But, before finalizing the report, having identified potential target organs, the diagnostic terms must be defined to establish morphologic criteria -- to provide data for computer and statistical applications.

In old rats, in the liver, one frequently finds

1. Focal bile ductular replication ± ectasia, hyalinization of the wall and/or mild to moderate chronic inflammatory cell infiltration.
2. Scattered hepatocytes that show increase in cytoplasm ± amphoteric tint and large nuclei with increased chromatin and prominent nucleolus(i) indicative of polypoidy.
3. Scattered hepatocytes with multiple nuclei.

Criteria used in classification of nodular hyperplasia and hepatocarcinoma

Pattern:

1. Vascular lobular pattern preserved
2. Focal parenchymal inflammatory cell infiltration
3. Focal necrosis
4. Focal hyperplasia
5. Focal nodular hypertrophy and hyperplasia
6. Marginal compression
7. Independent hepatocyte growth patterns -- tubular, diffuse with angiogenesis
8. Vascular or parenchymal invasion
9. Metastases

Cytology:

- A. Variable, larger nucleus, relative or absolute
- B. Increased chromatin with clumping or coarseness
- C. Nuclear inclusions
- D. Multi nucleated cells
- E. Increased amount of cytoplasm
- F. Vacuolization of cytoplasm
- G. Individual necrosis of hepatocytes
- H. Kupffer cell hypertrophy -- hyperplasia
- I. Increased number of mitoses
- J. Atypical mitoses

The absolute criteria for malignancy by pattern include 7 and 8.

The absolute criteria for malignancy by cytology include J.

Other supporting associate factors of malignancy may include selected markers of dedifferentiation or lack of maturation, such as CEA or markers of angiogenesis.

6. CONCLUSIONS

The following conclusions can be made from this study of inhalation exposure of male and female rats to nitroethane (NE) at 100 ppm or 200 ppm for two years:

1. There were no pharmacologic effects from exposure to NE at either atmospheric concentration.
2. There was no effect on mortality on either sex at either exposure level.
3. Body weights of both sexes of both groups of exposed rats were slightly less than respective controls. Lack of a well-defined dose-response relationship suggested the involvement of factors other than just exposure to NE.
4. There was no effect of exposure of rats of either sex to either level of NE on hematology.
5. Although there were some small differences in certain serum chemistry parameters, and among some individual animals, there were no biologically significant group differences between exposed and control rats of either sex.
6. There were no biologically significant effects of exposure to NE on organ weights.
7. There was no significant difference in the non-neoplastic or neoplastic pathology related to exposure to NE.

TABLE I

Weekly means of body weights (grams) of all rats surviving in each group during the indicated week of the study

Date Weighed	Week of Study	Males			Females		
		I	II	III	I	II	III
Aug 11, '83	0	191	188	195	164	167	161
Aug 21, '83	1	254	242	249	191	195	185
Aug 28, '83	2	288	270	281	203	204	196
Sep 04, '83	3	315	296	306	214	213	206
Sep 11, '83	4	344	319	330	225	224	217
Sep 18, '83	5	360	333	345	231	229	221
Sep 25, '83	6	376	345	358	236	232	225
Oct 02, '83	7	396	368	378	244	242	234
Oct 09, '83	8	415	384	393	250	249	240
Oct 16, '83	9	430	397	407	255	253	244
Oct 23, '83	10	445	409	420	262	259	251
Oct 30, '83	11	460	422	433	269	263	254
Nov 06, '83	12	464	423	437	265	259	256
Nov 13, '83	13	472	439	449	271	267	260
Nov 20, '83	14	479	446	456	274	270	260
Nov 27, '83	15	482	449	457	272	267	258
Dec 04, '83	16	490	457	467	274	269	259
Dec 11, '83	17	502	472	480	281	279	270
Dec 18, '83	18	505	479	487	280	279	271
Dec 24, '83	19	511	485	492	283	282	270
Dec 31, '83	20	517	490	498	286	283	273
Jan 08, '84	21	525	496	506	290	286	275
Jan 15, '84	22	530	501	510	291	288	276
Jan 22, '84	23	532	513	514	291	292	278
Jan 29, '84	24	537	508	522	294	291	280
Feb 05, '84	25	543	513	525	297	295	283
Feb 19, '84	27	552	526	533	301	299	285
Mar 04, '84	29	561	538	547	306	303	288
Mar 18, '84	31	586	546	553	311	306	291
Apr 01, '84	33	578	553	558	316	309	292
Apr 15, '84	35	592	559	565	323	310	294
Apr 29, '84	37	596	566	576	327	314	300
May 15, '84	39	600	563	576	326	314	297
May 27, '84	41	609	574	586	333	322	304
Jun 10, '84	43	610	577	587	335	324	306
Jun 24, '84	45	617	586	591	338	330	310
Jul 08, '84	47	634	592	598	349	336	316
Jul 22, '84	49	639	597	620	352	338	319
Aug 05, '84	51	660	614	631	367	347	326
Aug 19, '84	53	664	620	640	370	353	327
Sep 02, '84	55	670	631	648	375	358	330
Sep 16, '84	57	665	625	647	371	350	326
Sep 30, '84	59	660	614	641	365	344	320

TABLE I (Continued)

Date Weighed	Week of Study	Males			Females		
		I	II	III	I	II	III
Oct 14, '84	61	659	612	636	362	339	314
Oct 21, '84	62	684	640	672	387	369	341
Nov 06, '84	64	692	645	674	393	373	342
Nov 20, '84	66	701	647	663	397	374	341
Dec 04, '84	68	703	644	669	402	378	347
Dec 18, '84	70	705	644	670	408	381	352
Jan 03, '85	72	712	644	670	411	389	357
Jan 15, '85	74	707	641	672	408	385	356
Jan 29, '85	76	708	646	679	412	385	358
Feb 12, '85	78	701	640	667	409	384	355
Feb 26, '85	80	562	644	664	395	385	355
Mar 12, '85	82	653	589	616	392	369	341
Mar 26, '85	84	669	606	626	402	375	352
Apr 09, '85	86	667	616	639	409	383	359
Apr 23, '85	88	688	643	649	414	385	367
May 07, '85	90	683	650	652	418	386	376
May 21, '85	92	686	642	650	418	379	374
Jun 04, '85	94	683	648	648	427	383	383
Jun 18, '85	96	700	654	644	424	390	387
Jul 02, '85	98	699	645	644	423	392	379
Jul 16, '85	100	703	657	656	424	397	381
Jul 30, '85	102	699	651	660	435	377	381
Aug 13, '85	104	686	645	653	439	386	382

TABLE II

Hematology of rats exposed to nitroethane at 100 ppm or 200 ppm for 2 years. Values shown are means \pm one standard deviation.

<u>GROUP</u>	<u>RBC</u> <u>x10⁶</u>	<u>HCT</u> <u>%</u>	<u>MCV</u> <u>fl</u>	<u>HGB</u> <u>g%</u>	<u>WBC</u> <u>x10³</u>
	MALES				
I	7.74 \pm 0.96	37.4 \pm 5.7	48.0 \pm 2.6	15.1 \pm 1.6	13.1 \pm 2.7
II	6.84 \pm 1.55	33.1 \pm 6.5	49.1 \pm 4.7	13.4 \pm 2.8	11.1 \pm 2.2
III	6.86 \pm 1.16	33.3 \pm 5.0	48.7 \pm 2.6	13.6 \pm 2.0	10.3 \pm 4.9
	FEMALES				
I	7.54 \pm 0.73	37.4 \pm 2.4	50.0 \pm 3.4	15.3 \pm 1.2	6.2 \pm 1.3
II	7.58 \pm 0.80	37.3 \pm 2.7	49.4 \pm 2.7	15.2 \pm 1.5	6.7 \pm 1.3
III	7.52 \pm 0.36	36.7 \pm 2.1	49.0 \pm 1.8	15.2 \pm 0.7	8.3 \pm 2.8

TABLE III

Serum chemistry of rats exposed to nitroethane at 100 ppm or 200 ppm for 2 years.
 Values shown are means ± one standard deviation.

<u>GROUP</u>	<u>SGOT</u> <u>J/L</u>	<u>SGPT</u> <u>U/L</u>	<u>T. Bilirubin</u> <u>mg/dl</u>	<u>T. Protein</u> <u>g/dl</u>
MALES				
I	44.9±11.6	18.3± 4.7	0.48±0.11	6.45±0.43
II	44.9± 9.3	19.9± 4.4	0.43±0.12	6.55±0.69
III	49.4±12.8	20.8± 5.8	0.43±0.10	6.76±0.81
FEMALES				
I	50.7±18.8	22.2± 7.8	0.66±0.22	7.55±0.50
II	50.6±16.0	23.7± 6.9	0.59±0.25	8.01±0.59
III	57.5±38.8	27.0±15.1	0.55±0.26	8.18±0.58

TABLE III (Continued)

Serum chemistry of rats exposed to nitroethane at 100 ppm or 200 ppm for 2 years.
 Values shown are means \pm one standard deviation.

GROUP	BUN mg/dl	Creatinine mg/dl	Sodium meq/l	Potassium meq/l
MALES				
I	16.1 \pm 5.0	1.04 \pm 0.30	147.7 \pm 1.5	5.92 \pm 0.79
II	27.6 \pm 26.9	1.32 \pm 0.93	147.5 \pm 2.2	5.74 \pm 0.70
III	16.2 \pm 2.7	0.89 \pm 0.11	147.6 \pm 1.9	5.57 \pm 0.48
FEMALES				
I	14.1 \pm 2.1	0.94 \pm 0.09	146.5 \pm 1.9	5.65 \pm 0.70
II	15.1 \pm 2.6	0.95 \pm 0.13	146.5 \pm 2.2	5.74 \pm 0.59
III	18.7 \pm 4.2	1.07 \pm 0.18	145.6 \pm 2.1	5.65 \pm 0.61

TABLE IV

Organ weights of rats surviving to the terminal sacrifice and which were exposed to nitroethane at 100 ppm or 200 ppm for 2 years. Values are means \pm one standard deviation and are presented as absolute weights and per cent of body weight.

	MALES		FEMALES	
LIVER				
GROUP	Absolute(g)	Relative(%)	Absolute(g)	Relative(%)
I	14.13 \pm 2.42	2.16 \pm 0.35	11.11 \pm 4.50	2.74 \pm 1.01
II	15.01 \pm 3.86	2.51 \pm 0.68	10.02 \pm 1.70	2.84 \pm 0.63
III	14.61 \pm 2.41	2.38 \pm 0.23	9.39 \pm 1.30	2.68 \pm 0.37
KIDNEY				
GROUP	Absolute(g)	Relative(%)	Absolute(g)	Relative(%)
I	4.54 \pm 0.90	0.70 \pm 0.13	2.88 \pm 0.45	0.72 \pm 0.13
II	4.57 \pm 0.63	0.77 \pm 0.18	2.97 \pm 0.32	0.85 \pm 0.20
III	4.60 \pm 0.90	0.76 \pm 0.18	2.83 \pm 0.32	0.81 \pm 0.13
BRAIN				
GROUP	Absolute(g)	Relative(%)	Absolute(g)	Relative(%)
I	2.35 \pm 0.14	0.37 \pm 0.06	2.11 \pm 0.11	0.53 \pm 0.07
II	2.24 \pm 0.08	0.38 \pm 0.06	2.05 \pm 0.14	0.58 \pm 0.09
III	2.27 \pm 0.14	0.38 \pm 0.05	2.07 \pm 0.15	0.59 \pm 0.07
HEART				
GROUP	Absolute(g)	Relative(%)	Absolute(g)	Relative(%)
I	1.82 \pm 0.22	0.28 \pm 0.03	1.38 \pm 0.29	0.34 \pm 0.07
II	1.85 \pm 0.36	0.32 \pm 0.11	1.32 \pm 0.13	0.38 \pm 0.07
III	1.89 \pm 0.36	0.31 \pm 0.05	1.42 \pm 0.27	0.41 \pm 0.11
LUNG				
GROUP	Absolute(g)	Relative(%)	Absolute(g)	Relative(%)
I	3.36 \pm 2.93	0.52 \pm 0.08	2.17 \pm 0.47	0.54 \pm 0.13
II	2.59 \pm 0.29	0.43 \pm 0.07	2.17 \pm 0.27	0.62 \pm 0.15
III	2.78 \pm 0.36	0.46 \pm 0.07	2.20 \pm 0.30	0.63 \pm 0.11

Q.A. INSPECTION STATEMENT

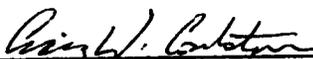
(Reference to 21 CFR 58.35 (b) (7))

Project No. - 828382

WSRC Compound No. - 8815

Type of Study - Chronic Inhalation of Nitroethane

This final study report was reviewed by the WSRC Quality Assurance Unit on June 11, 1986. The study was also inspected on 11/13/83, 2/19/84, 3/5/84, 8/27/84, 9/6/84, 10/21/84, 1/15/85, 1/25/85, 4/23/85, 7/14/85, 7/16/85, 7/21/85, 8/15/85, 8/16/85, 8/19/85 and 8/28/85 by the WSRC Quality Assurance Unit and findings were reported to the Study Director and management.



Director, Quality Assurance Unit
June 11, 1986

0302 INHALATION OF NITROETHANE

TUMOR INCIDENCE -- SUMMATION TABLE

	♂ TOTAL #	♀ TOTAL #	♂ TOTAL #	♀ TOTAL #	♂ # ANIMALS	♀ # ANIMALS	♂ # ANIMALS	♀ # ANIMALS	♂ # ANIMALS	♀ # ANIMALS	♂ # ANIMALS	♀ # ANIMALS	♂ # ANIMALS	♀ # ANIMALS
	# BENIGN	# MALIGNANT	# OF	# OF	# W/ BENIGN	# W/ MALIGN.	# W/ TUMORS	# W/ TUMORS	# OF					
	# TUMORS	# TUMORS	# TUMORS	# TUMORS	# TUMORS	# TUMORS	# TUMORS	# TUMORS	# METASTASIS					
I	♂ N 42 F	♂ N 398 H 4 F	♂ N 78 H 46 F	♂ N 468 H 28 F	♂ N 298 H 4 F	♂ N 78 H 32 F	♂ N 368 H 0 F	♂ N 218 H 0 F						
II	♂ N 33 F	♂ N 398 H 11 F	♂ N 178 H 44 F	♂ N 568 H 25 F	♂ N 308 H 9 F	♂ N 168 H 34 F	♂ N 468 H 27 F	♂ N 418 H 4 F						
III	♂ N 25 F	♂ N 348 H 8 F	♂ N 118 H 33 F	♂ N 458 H 22 F	♂ N 278 H 8 F	♂ N 88 H 30 F	♂ N 358 H 11 F	♂ N 68 H 1 F						

0043