THE EFFECTS OF STORAGE UPON THE TOXICITY OF OIL REFINERY EFFLUENTS

By

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1954

Submitted to the Faculty of the Graduate School of
the Oklahoma State University
in partial fulfillment of the requirements
for the degree of
MASTER OF SCIENCE
May, 1960

THE EFFECTS OF STORAGE UPON THE TOXICITY

OF OIL REFINERY EFFLUENTS

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PREFACE

The toxicity bioassay is becoming an important tool for evaluating industrial effluents. Pollution laws generally require that effluents may not have a detrimental effect on aquatic life. The toxicity bioassay is a means of evaluating the effects of effluents upon aquatic life.

The collection, handling, and storage of effluent samples for testing presents problems to all technicians who are involved in toxicity bioassay. Governmental enforcement agents and industrial waste control personnel alike are interested in the solution of problems which arise in use of the toxicity bioassay.

The assistance and guidance of Dr. Troy C. Dorris is hereby acknowledged. Grateful appreciation is expressed to Dr. W. H. Irwin and Dr. R. W. Jones for their suggestions. Dr. R. D. Morrison gave helpful suggestions on the design of the experiment and the statistical analysis. Thanks are extended to those individuals that assisted in the collecting of test animals.

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CHAPTER I

INTRODUCTION

The study reported here provides information on the effects of short- and long-term storage on toxicity of oil refinery effluents of different degrees of toxicity. The study was conducted at Oklahoma State University from July 5, 1958 to October 18, 1959.

The Aquatic Biology Laboratory of the Zoology Department, Oklahoma State University, is engaged in an extensive program of investigations of oil refinery effluents. A study sponsored by the Oklahoma Oil Refiners' Waste Control Council required information on the effects of short periods of storage upon toxicity. Research sponsored by the National Institutes of Health for a continuous-flow bioassay of oil refinery effluents required information on the effects of long-term storage upon the toxicity of oil refinery wastes.

Clemens and Summers (1954) found that the toxicity of oil refinery wastes changed when stored at full strength in glass containers at room temperature. They found that the toxicity decreased with storage. They found no apparent correlation between pH and the toxicity of wastes. No further fish kill resulted after 48 hours under test conditions. However, experience in the Oklahoma State University laboratories had indicated tht in some cases effluents did not change under storage.

CHAPTER II

MATERIALS AND PROCEDURES

Characteristics of Effluents

Final effluents of four levels of toxicity were studied. Effluent I was toxic at a concentration of approximately 7 percent by volume. This effluent contained wastes from a catalytic poly-unit, a catalytic cracker, a platformer, a vis-breaker vacuum still, two crude distillation units, and the normal refinery operating processes. No information on the chemical composition of this effluent was available.

Effluent II was toxic at a concentration of approximately 18 percent. This effluent contained effluents from a crude unit, two coking units, a catalytic cracker, a catalytic reformer, a thermal reformer, and a compound house.

TABLE I

CHEMICAL CHARACTERISTICS OF EFFLUENT II

Date	Sulfides ppm.	Phenols ppm.	NH_3 ppm.	pH	I.O.D. ppm.	D.O. ppm.	Total Oils ppm.
August, 1958	0.0	11.3	10.0	--	3.0	0.0	326

Effluent III was toxic at a concentration of approximately 65 percent. This effluent was taken from a point part way along a series of holding ponds and contained effluents from a crude unit, a thermal cracker, a catalytic cracker, an alkylation unit, and a vapor-recovery unit.

TABLE II

CHEMICAL CHARACTERISTICS OF EFFLUENT III

Date	Sulfides ppm.	Phenols ppm.	NH_3 ppm.	pH	C.O.D. ppm.
August, 1959	0.0	0.5	23.0	9.0	190
September, 1959	0.0	450.0	17.0	8.9	313
October, 1959	0.0	2.5	25.0	8.9	189

Effluent IV was obtained from a holding pond. This effluent contained waste waters from desalters, atmospheric, vacuum-distillation, and topping crude units, thermal cracking, catalytic reforming and polymerization, H. F. alkylation, lube-oil solvent treating, Perco, doctor treating, and dewaxing units.

TABLE III

CHEMICAL CHARACTERISTICS OF EFFLUENT IV

Date	Sulfides ppm.	Phenols ppm.	NH_3 ppm.	pH	C.O.D. ppm.
July, 1959	0.08	0.7	20.0	7.6	202
August, 1959	0.06	1.6	18.0	7.2	177
September, 1959	0.08	2.1	26.0	7.5	265

Methods

Short-term storage of one to three days was required in the shipment of all oil refinery effluents to Oklahoma State University, Stillwater, for bioassay tests on the Oklahoma Oil Refiners' Waste Control Council Project. The need to store effluents for periods of approximately one month was anticipated for the National Institutes of Health continuous-flow bioassay study. Bioassay tests were run with each effluent at periods throughout the duration of storage to gain information on the effects of storage upon the toxicity of oil refinery effluents.

All refinery effluents tested were stored in five-gallon polyethylene bottles which were filled to the top to exclude air and then stoppered or capped. To increase uniformity, the effluent samples were pumped into a large drum for mixing before being drained into the storage containers. The effluent samples were stored at a temperature of about 75° F. Effluent samples were collected in several bottles. Bottles were chosen at random for each test within the series. In all tests an effort was made to place effluent from each bottle in each test container. No effluent remaining from the previous test in a partially emptied bottle was used in a later test. Each bottle of effluent was shaken before using to insure uniformity of the effluent.

All test animals used were fathead minnows (*Pimephales promelas*, Rafinesque) raised in ponds at Oklahoma State University. All fish were held in the laboratory for at least ten days prior to use as suggested by Doudoroff et al. (1951). During this time specimens that were diseased, injured, abnormal, undernourished, or showed breeding characteristics were discarded. All holding tanks were treated with terramyecin to prevent outbreak of a bacterial infection, "tail rot." The procedures on feeding the test animals set up by Doudoroff et al. (1951) were followed. The weight of all fish in a test container did not exceed 1 gram per liter. If, as happened a number of times, more than 10 percent of the test animals became unfit for testing purposes, the entire series of tests was discontinued. A control of 10 to 20 fish taken at random from the stock population was placed in dilution water and observed simultaneously with each test.

Tap water from Lake Carl Blackwell was dechlorinated by aeration for at least 24 hours previous to use for dilution of the effluents. The water had been tested for a year previous to use and was found to be fairly constant in chemical content.

The test containers were of polyethylene. Each test container was filled with 10 liters of dilution water. Test concentrations were then made by removing the appropriate amounts of water and replacing them with effluent. In testing effluents I and II, each concentration was replicated twice. In testing effluents III and IV, the number of replications was increased to four and schedule of testing modified to facilitate analysis of any changes that might occur. Five fish were used in each test container. All tests in the series I and II were maintained for a 96-hour period. Because of the lack of space resulting from the increase in replication in tests III and IV, and because previous experience had shown the 48-hour test yielded about the same results as the 96-hour test, all tests in the series on effluents III and IV were terminated after 48 hours. All tests were performed under 75° F. temperature conditions. Hydrogen-ion concentration measurements were made with an electric Beckman pH meter. Dissolved oxygen concentrations were determined with a Bausch and Lomb Spectronic 20 spectrophotometer.

The oxygen concentration in all tests was observed closely during the tests. Only test concentrations which remained above 1 ppm. dissolved oxygen were used in analysis of effluent toxicities. This concentration was usually achieved by the high initial dissolved oxygen content of the dilution water and the constant diffusion of oxygen from the air into the test medium. Test concentrations which dropped below the minimum allowable level of dissolved oxygen were treated by the addition of bubbles of pure oxygen (Hart et al., 1949; Doudoroff et al., 1951). Three test concentrations fell below the 1 ppm. minimum despite the added oxygen.

Measurement of Toxicity

Median tolerance limits (TL_m) are used in the analysis of the data. These values represent concentrations of effluents that would cause a 50-percent mortality to test animals, (Doudoroff et al., 1951). The values were determined by plotting data on mortality of more than 50 percent and less than 50-percent on semi-logarithmic graph paper, then using a straight-line interpolation to estimate the concentration of effluents that would cause a 50-percent mortality.

Statistical Analysis

The derived median tolerance limits were treated statistically. The experiments were designed as randomized complete blocks. An analysis of variance table was computed. The calculated F (Fisher) value of treatments was compared with the tabulated F (Fisher) value. A probability statement was made for each effluent.

CHAPTER III

RESULTS AND DISCUSSION

Oil refinery effluent I had an initial median tolerance limit of approximately 7 (TL_m^{96} 7). No significant change in toxicity was found to occur in a 30-day test period. The pH did not change during this period.

Oil refinery effluent II had an initial median tolerance limit of approximately 18 (TL_m^{96} 18). No significant change in toxicity took place in this effluent during a 30-day test period. No change in pH occurred during the 30-day test period.

Oil refinery effluent III had an initial median tolerance limit of approximately 65 (TL_m^{48} 65). There was no significant change in toxicity during the first four days of storage. Between the fourth and eighth days of storage the toxicity of this effluent decreased. No change in the pH of the effluent took place during the 32-day test period. An intensification in the color of the effluent was noticed at the beginning of the eighth day of storage.

No statistical information was obtainable from effluent IV because the effluent did not produce a sufficient number of tests in which 50 percent or more of the fish were killed. Under such circumstances no measure of the median tolerance limit was obtainable and statistical analysis could not be applied properly to the test data. It was noted that a larger number of fish survived in the later tests of this effluent than in the earlier tests. The longer the effluent was stored the closer the pH approached neutrality. From these observations it is suggested that this effluent decreased in toxicity during the period of storage.

Conclusions

Certain tentative conclusions may be drawn from the data presented here (Figure 1). When stored at room temperature without access to air, effluents of high initial toxicity (TL_m^{96} 7 and TL_m^{96} 18) show no change in toxicity over a storage period of 30 days. Effluents of intermediate toxicity (TL_m^{48} 65) undergo a change in toxicity after being stored a short time. It appears that effluents of very low toxicity (high TL_m) may also change during the period of storage.

Roberts (personal communication) studied the populations of algae and bacteria in a series of oil-refinery-effluent-holding ponds. He found few kinds of smaller organisms in large numbers in the earlier high-toxicity portion of the pond system. In the lower-toxicity ponds later in the series he reported finding many kinds of larger organisms in smaller numbers.

The low-toxicity effluents used in the present study were taken at points along series of effluent holding-ponds. It is believed that the low toxicity of these effluents was the result of biological action on effluents of initially high toxicity. In taking the samples the organisms in them were also taken. Undoubtedly some of these organisms continued to function under the storage conditions thereby reducing the toxicity of the effluents throughout the storage period.

Effluents of high toxicity were taken from locations where they were subject to little biological action. The organisms present in these effluents apparently were not capable of producing significant changes in toxicity during the storage period.

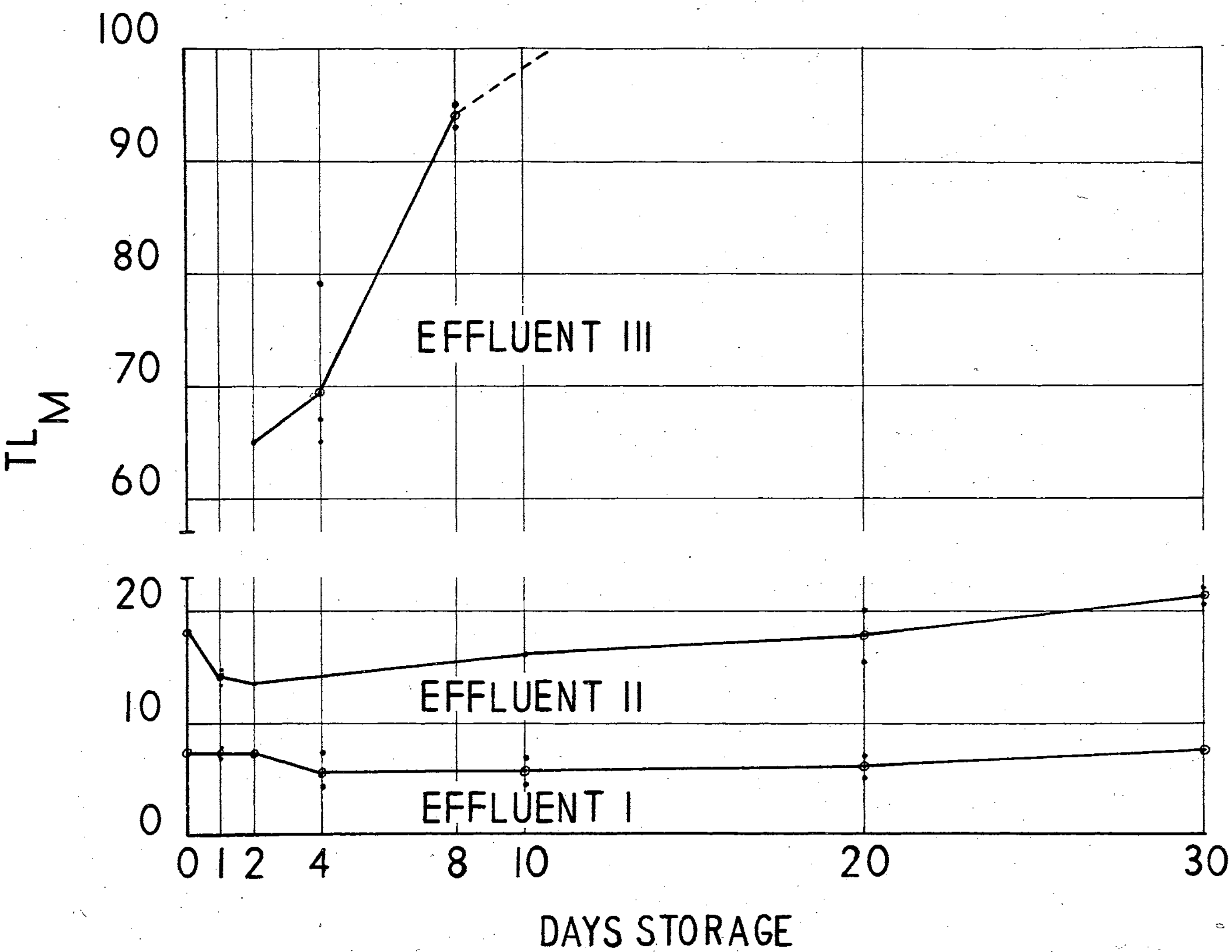

Figure 1. The effect of storage upon the toxicity of three oil refinery effluents. .------replication value. O------mean value. Dotted line is extrapolated TL_m value.

CHAPTER IV

SUMMARY

1. A study was made to determine the effects of storage upon the toxicity of oil refinery effluents.

2. Oil refinery effluents of four levels of toxicity were studied.

3. The results of bioassay tests performed are presented.

4. Where possible a median tolerance limit (TL_m) was determined.

5. The median tolerance limits (TL_m) were treated statistically.

6. Oil refinery effluent I (TL_m 7) did hot change in toxicity during the 32-day storage period.

7. Oil refinery effluent II (TL_m 18) did not show a significant change in toxicity during the 30-day storage period.

8. Oil refinery effluent III (TL_m 65) decreased in toxicity during the fourth and eighth days of the 32-day storage period.

9. Oil refinery effluent IV did not kill a sufficient number of test animals to be treated statistically. There was evidence that the effluent decreased in toxicity during the storage period.

CHAPTER V

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APPENDIX A

TOXICITY BIOASSAY RESULTS

TABLE IV

TOXICITY BIOASSAY I, OCTOBER 4, 1958

	% Conc. Effluent	Number of Test Animals		Survival 24 Hour		Survival 48 Hour		Survival 72 Hour		Survival 96 Hour		TL_m^{96}		Dissolved Oxygen ppm		pH	
		A	B	A	B	A	B	A	B	A	B	A	B	init	end	init	end
#1 - Initial	10	5	5	0	0	0	0	0	0	0	0	7.4	7.4	6.2	4.7	8.4	8.4
1:50 P.M.	5.6	5	5	5	5	5	5	5	5	5	5			5.7	5.7	8.4	7.7
October 4, 1958	3.2	5	5	5	5	5	5	5	5	5	5			6.0	6.4	8.3	7.8
	1	5	5	5	5	5	5	5	5	5	5			7.4	6.8	8.2	7.9
#1 - CONTROL		5	5	5	5	5	5	5	5	5	5			8.1	7.0	8.1	7.9
#2 - 24 Hours Storage	10	5	5	0	0	0	0	0	0	0	0	7.0	7.4	5.0	6.0	8.4	8.4
2:30 P.M.	5.6	5	5	4	5	4	5	4	5	4	5			5.9	4.1	8.4	7.6
October 5, 1958	3.2	5	5	5	5	5	5	5	5	5	5			6.0	4.9	8.3	7.8
#2 - CONTROL		5	5	5	5	5	5	5	5	5	5			6.4	4.9	8.1	7.8
#3 - 48 Hours Storage	10	5	5	0	0	0	0	0	0	0	0	7.4	7.4	5.7	5.6	8.5	8.5
4:00 P.M.	5.6	5	5	5	5	5	5	5	5	5	5			6.0	5.0	8.5	7.6
October 6, 1958	3.2	5	5	5	5	5	5	5	5	5	5			6.6	3.9	8.4	7.6
#3 - CONTROL		5	5	5	5	5	5	5	5	5	5				6.4	8.1	7.8
#4 - 96 Hours Storage	10	5	5	0	0	0	0	0	0	0	0			5.1	5.1	8.4	8.4
2:30 P.M.	5.6	5	5	0	5	0	5	0	5	0	5	7.4	4.3	6.8	A-2.3	8.4	A-8.0
October 8, 1958															B-6.4		B-7.7
	3.2	5	5	5	5	5	5	5	5	5	5			7.4	6.4	8.4	7.8
#4 - CONTROL		5	5	5	5	5	5	5	5	5	5						
#5 - 10 Days Storage	10	5	5	0	0	0	0	0	0	0	0			5.3	4.7	8.6	8.6
8:30 A.M.	5.6	5	5	1	5	1	4	1	4	1	4	4.5	7.0	5.6	A-7.4	8.5	A-7.9
October 14, 1958															B-6.6		B-7.9
	3.2	5	5	5	5	5	5	5	5	5	5			5.9	4.5	8.4	7.7
#5 - CONTROL		5	5	5	5	5	5	5	5	5	5						
#6 - 20 Days Storage	10	5	5	0	0	0	0	0	0	0	0			5.9	6.0	8.5	8.5
1:10 P.M.	5.6	5	5	2	4	2	4	2	4	2	4	5.1	7.0	6.2	5.6	8.5	7.8
October 24, 1958	3.2	5	5	5	5	5	5	5	5	5	5			7.0	7.2	8.4	8.0
#6 - CONTROL		5	5	5	5	5	5	5	5	5	5			7.2	8.0	8.3	8.0
#7 - 30 Days Storage	10	5	5	0	0	0	0	0	0	0	0	7.4	7.4	6.0	5.4	8.4	8.4
3:30 P.M.	5.6	5	5	5	5	5	5	5	5	5	5			6.2	7.0	8.4	7.9
November 3, 1958	3.2	5	5	5	5	5	5	5	5	5	5			6.8	7.6	8.3	7.9
#7 - CONTROL		5	5	5	5	5	5	5	5	5	5			7.6	7.8	8.1	8.0

TABLE V

TOXICITY BIOASSAY II, JULY 5, 1958

	% Conc. Effluent	Number of Test Animals		Survival								TL_m 96		Dissolved Oxygen ppm		pH	
				24 Hour		48 Hour		72 Hour		96 Hour							
		A	B	A	B	A	B	A	B	A	B	A	B	init	end	init	end
#1 - Initial 2:30 P.M. July 5, 1958	32	5	5	0	0	0	0	0	0	0	0			6.4	1.9	8.5	7.3
	18	5	5	5	3	3	3	3	2	3	2	20	16	6.2	1.5	8.5	7.4
	10	5	5	5	5	5	5	5	5	5	5			6.8	2.2	8.4	7.5
#1 - CONTROL		5	5	5	5	5	5	5	5	5	5			7.4	3.3	8.3	8.2
#2 - 24 Hour Storage 2:00 P.M. July 6, 1958	32	5	5	0	0	0	0	0	0	0	0			3.9	3.5	8.3	7.5
	18	5	5	5	5	3	1	1	0	1	0	14.5	13.5	6.0	A-1.3 B-3.3	8.3	A-7.5 B-7.6
	10	5	5	5	5	5	5	5	5	5	5			6.0	1.2	8.2	7.5
#2 - CONTROL		5	5	5	5	5	5	5	5	5	5			7.2	4.6	8.2	7.9
#3 - 48 Hour Storage 2:00 P.M. July 7, 1958	32	5	5	1	4	0	0	0	0	0	0			4.6	3.6	8.6	7.7
	18	5	5	5	5	3	4	0	2	0	2	13.5	--	6.0	A-1.3 B-0.7	8.5	A-7.5 B-7.4
	10	5	5	5	5	5	5	5	5	5	5				0.0	8.4	7.5
#3 - CONTROL		5	5	5	5	5	5	5	5	5	5			7.0	5.2	8.2	7.7
#4 - 10 Day Storage 2:00 P.M. July 15, 1958	32	5	5	0	0	0	0	0	0	0	0			5.3	A-3.1 B-2.6	8.5	A-7.6 B-7.5
	18	5	5	5	5	1	2	1	2	1	2	--	16	6.0	A-0.0 B-1.3	8.4	A-7.4 B-7.4
	10	5	5	5	5	5	5	5	5	5	5			7.2	5.1	8.4	7.5
#4 - CONTROL		5	5	5	5	5	5	5	5	4	5			7.8	1.2	8.3	7.6
#5 - 20 Day Storage 7:00 P.M. July 28, 1958	32	5	5	2	5	1	0	1	0	1	0			5.0	A-2.7 B-7.8	8.3	A-7.2 B-7.3
	18	5	5	5	5	5	4	5	3	2	3	15.5	20	6.4	A-1.6 B-3.0	8.2	A-7.3 B-7.3
	10	5	5	5	5	4	5	4	5	4	5			7.2	4.1	8.2	7.2
#5 - CONTROL		5	5	5	5	5	5	5	5	5	5			7.6	1.2	8.1	7.4
#6 - 30 Day Storage 2:15 P.M. August 12, 1958	32	5	5	2	3	0	1	0	1	0	1			5.3	A-7.2 B-7.2	8.1	A-7.3 B-7.2
	18	5	5	5	4	4	3	4	3	4	3	22	20.5	6.8	2.6	8.1	7.2
	10	5	5	5	4	4	3	4	3	4	3			7.4	1.8	8.1	7.2
#6 - CONTROL		5	5	5	5	5	5	5	5	5	5			8.3	1.4	8.0	7.3

TABLE VI

TOXICITY BIOASSAY III, SEPTEMBER 14, 1959

Test	% Conc. Effluent	Number of Test Animals A	B	C	D	Survival 24-Hour A	B	C	D	Survival 48-Hour A	B	C	D	TL_m 48 A	B	C	D	Dissolved Oxygen initial A	B	C	D	Dissolved Oxygen end A	B	C	D	pH initial A	B	C	D	pH end A	B	C	D
#1 - 2 Days Storage	75	5	5	5	5	0	3	4	1	0	1	3	0	65	65	>75	65	1.1	1.0	.6	.7	2.8	5.0	5.6	3.7	7.4	7.4	7.4	7.4	7.2	7.4	7.5	7.2
9:30 A.M.	56	5	5	5	5	5	5	4	5	5	4	3	5					2.4	2.3	2.2	2.1	3.6	4.1	4.2	4.6	7.5	7.5	7.5	7.5	7.3	7.4	7.5	7.4
September 16, 1959	32	5	5	5	5	5	5	5	5	5	5	5	5					4.5	4.5	4.5	4.2	2.9	3.6	3.4	3.8	7.6	7.6	7.5	7.6	7.4	7.4	7.4	7.4
#1 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					7.8				3.7	3.1	3.9	4.7	8.0				7.4	7.4	7.4	7.5
#2 - 4 Days Storage	100	5	5	5	5	1	1	1	0	0	0	0	0	79	65	67	67	0.0	0.0	0.0	0.0	5.7	5.1	5.4	3.7	7.3	7.3	7.4	7.3	7.4	7.4	7.5	7.1
6:30 P.M.	75	5	5	5	5	4	3	4	2	3	1	1	1					0.9	1.1	0.5	0.6	4.9	2.9	3.9	5.1	7.4	7.4	7.4	7.4	7.4	7.4	7.4	7.5
September 18, 1959	56	5	5	5	5	5	5	5	5	5	4	5	5					2.1	2.7	2.2	2.1	4.7	4.7	5.7	4.9	7.5	7.5	7.4	7.4	7.5	7.4	7.5	7.5
#2 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					7.4				5.4				8.1				7.5			
#3 - 8 Day Storage	100	5	5	5	5	4	4	3	3	3	2	3	2	>100	93	>100	95	0.0	0.0	0.0	0.0	7.4	8.9	6.0	8.3	7.4	7.4	7.4	7.3	7.5	7.7	7.4	7.5
9:15 P.M.	75	5	5	5	5	3	5	5	5	3	4	4	5					0.8	1.1	0.6	0.7	6.8	8.3	9.8	10+	7.4	7.4	7.3	7.3	7.4	7.6	7.6	7.6
September 22, 1959	56	5	5	5	5	5	5	5	5	5	4	5	5					2.1	2.7	2.4	2.2	8.0	7.8	6.6	7.0	7.4	7.3	7.4	7.4	7.6	7.5	7.5	7.4
#3 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					7.2				3.1				7.9				7.5			
#4 - 16 Day Storage	100	5	5	5	5	5	5	5	5	4	4	4	5	>100	>100	>100	>100	0.0	0.0	0.0	0.0	5.7	7.4	6.6	5.4	7.3	7.4	7.3	7.4	7.6	7.4	7.5	7.4
9:00 A.M.	75	5	5	5	5	5	5	5	5	4	5	4	5					1.2	1.2	0.8	1.2	5.7	6.2	5.9	5.1	7.4	7.4	7.3	7.3	7.4	7.4	7.4	7.4
September 30, 1959																																	
#4 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					7.2				3.4				8.1				7.6			
#5 - 32 Day Storage	100	5	5	5	5	5	4	5	5	5	4	5	5	>100	>100	>100	>100	0.0	0.0	0.0	0.0	8.0	7.2	6.6	7.0	7.2	7.2	7.2	7.3	7.5	7.5	7.5	7.6
10:15 A.M.	75	5	5	5	5	5	5	5	5	5	5	5	5					1.3	1.4	1.2	1.4	7.6	5.9	5.1	7.2	7.3	7.3	7.3	7.2	7.7	7.5	7.5	7.5
October 16, 1959																																	
#5 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					7.6				5.4				8.0				7.5			

TABLE VII

TOXICITY BIOASSAY IV, AUGUST 19, 1959

Test	% Conc. Effluent	Number of Test Animals				Survival 24-Hour				Survival 48-Hour				TL_m^{48}				Dissolved Oxygen initial				Dissolved Oxygen end				pH initial				pH end			
		A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D
#1 - 2 Day Storage	100	5	5	5	5	3	2	5	2	3	1	5	0	>100	87	>100	--	0.9	0.6	1.2	0.7	+	* +	+	0.0	7.8	7.8	7.9	7.9	7.3	7.3	7.3	7.3
8:45 A.M.	75	5	5	5	5	5	4	4	4	4	4	4	4					2.7	2.9	2.2	2.8	+	+	+	+	8.0	7.9	7.9	7.9	7.5	7.5	7.5	7.5
August 21, 1959																																	
#1 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					6.4				+				8.2				7.8			
#2 - 4 Day Storage	100	5	5	5	5	5	3	4	1	4	3	4	1	>100	>100	>100	90	-	-	* -	-	3.4	4.4	5.4	5.0	7.9	7.9	7.9	7.9	7.3	7.4	7.4	7.4
3:00 P.M.	75	5	5	5	5	5	5	5	4	5	5	5	4					-	-	-	-	5.7	6.2	5.6	5.1	7.9	7.9	7.9	7.9	7.6	7.5	7.5	7.5
August 23, 1959																																	
#2 - CONTROL		5	5	5	5	5	5	4	5	5	5	4	5									5.7	4.4	5.9	5.4	8.3				7.9	7.7	7.9	7.9
#3 - 8 Day Storage	100	5	5	5	5	4	5	4	4	2	5	4	3	93	>100	>100	>100	0.7	2.3	1.1	1.3	3.5	3.0	4.1	3.3	7.8	7.8	7.8	7.8	7.3	7.3	7.4	7.4
9:15 A.M.	75	5	5	5	5	4	4	5	5	4	4	5	5					2.3	2.1	1.6	2.7	4.4	3.7	4.4	2.9	7.8	7.8	7.7	7.8	7.4	7.4	7.4	7.3
August 27, 1959																																	
#3 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					6.6				6.0	5.9	5.0	6.0	8.1				7.7	7.8	7.7	7.8
#4 - 16 Day Storage	100	5	5	5	5	5	4	5	5	5	4	5	5	>100	>100	>100	>100	0.3	0.4	0.3	0.3	4.9	4.4	5.4	6.2	7.6	7.6	7.6	7.7	7.4	7.3	7.4	7.4
8:45 A.M.	75	5	5	5	5	5	5	5	5	5	5	5	5					1.7	2.2	1.9	1.3	5.3	3.6	5.1	5.0	7.6	7.7	7.7	7.7	7.4	7.3	7.3	7.4
September 4, 1959																																	
#4 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	3					6.2	6.0	6.4	6.4	4.4	3.7	5.1	2.7	8.0	7.9	8.0	8.0	7.6	7.6	7.6	7.5
#5 - 32 Day Storage	100	5	5	5	5	5	5	5	5	4	3	5	5	>100	>100	>100	>100	0.0	0.0	0.0	0.0	2.9	4.1	3.9	4.0	7.3	7.3	7.3	7.3	7.4	7.5	7.5	7.4
8:30 P.M.	75	5	5	5	5	5	5	5	5	5	5	5	4					0.0	0.6	0.4	0.7	3.9	4.2	3.6	3.3	7.3	7.4	7.4	7.4	7.4	7.4	7.3	7.3
September 20, 1959																																	
#5 - CONTROL		5	5	5	5	5	5	5	5	5	5	5	5					7.4				1.7				8.0				7.4			

*Visual Evaluations
- less than 1 ppm
+ greater than 1 ppm

APPENDIX B

STATISTICAL ANALYSES

TABLE VIII-A

BIOASSAY I, OCTOBER 4, 1958, TL_m^{96}

Replication	Time of Storage						
	Initial	24 hrs.	48 hrs.	96 hrs.	10 days	20 days	30 days
A	7.4	7.0	7.4	7.4	4.5	5.1	7.4
B	7.4	7.4	7.4	4.3	7.0	7.0	7.4

TABLE VIII-B

BIOASSAY I, ANALYSIS OF VARIANCE

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F.
Total	13	17.34		
Time	6	7.53	1.26	.787
Replications	1	.20	.20	
Error	6	9.61	1.60	

Tabular F. = 4.28 at 5% Level
Calculated F = .787

The null hypothesis, that storage does not affect the toxicity of oil refinery effluents, is not rejected.

TABLE IX-A

BIOASSAY II, JULY 5, 1958, TL_m^{96}

Replication	Time of Storage					
	Initial	24 hrs.	48 hrs.	10 days	20 days	30 days
A	20	14.5	13.5	---	15.5	22
B	16	13.5	---	16	20	20.5

TABLE IX-B

BIOASSAY II, ANALYSIS OF VARIANCE

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F.
Total	9	90.02		
Time	5	60.27	12.05	1.21
Replications	1	.02		
Error	3	29.73	9.91	

Tabular F. = 9.01 at the 5% Level
Calculated F = 1.21

The null hypothesis, that storage does not affect the toxicity of oil refinery effluents, is not rejected.

TABLE X

BIOASSAY III, SEPTEMBER 14, 1959, TL_m^{48}

Replication	Time of Storage 2 days	4 days	8 days	16 days	32 days
A	65	79	>100	>100	>100
B	65	65	93	>100	>100
C	>75	67	>100	>100	>100
D	65	67	95	>100	>100

TABLE XI

BIOASSAY IV, AUGUST 19, 1959, TL_m^{48}

Replication	Time of Storage 2 days	4 days	8 days	16 days	32 days
A	>100	>100	93	>100	>100
B	87	>100	>100	>100	>100
C	>100	>100	>100	>100	>100
D	--	90	>100	>100	>100

VITA

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Master of Science

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