THE UNIVERSITY OF MANITOBA

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROFLORA IN POTATO WASTE WATER

by

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ABSTRACT

The effect of Gamma Irradiation on the aerobic microflora of potato waste water was studied.

The aerobic microbial genera present in potato waste water before and after irradiation with gamma rays were identified. The most fundamental organisms present in the waste at a dosage of 0.0 megarads were micrococci. Organisms of the genus Bacillus also made up a major portion of the microflora. Other organisms such as Aerobacter, Corynebacterium, Erwinia, Sarcina, Streptomyces and Xanthomonas made up only minor portions of the microflora.

After dosages of 0.5 and 1.0 megarads Bacillus species became the predominant organisms and after dosages of 1.5 and 2.0 megarads they became the only organisms.

Most of the microflora was destroyed after dosages of 1.0 megarads.

Work done at various levels of total solids indicated that there was no significant difference in the radiation resistance of organisms at higher levels of total solids.

The addition of sodium hypochlorite and hydrogen peroxide to the waste water proved ineffective in sensitizing the microflora to gamma irradiation.

TABLE OF CONTENTS

	PAGE
INTRODUCTION	1
REVIEW OF LITERATURE	3
THE EFFECTS OF GAMMA IRRADIATION ON MICROORGANISMS	5
(1) General Effects	5
(2) Effect of Irradiation on Vegetative Cells	5
EFFECT OF ENVIRONMENT ON MICROORGANISMS DURING IRRADIATION	7
THE EFFECT OF IRRADIATION ON BACTERIAL SPORES	8
IDENTIFICATION PROCEDURES	10
SCOPE OF THE INVESTIGATION	13
MATERIALS AND METHODS	14
RESULTS	24
A. THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER AT SELECTED GAMMA IRRADIATION DOSAGES	24
B. THE RATES OF DESTRUCTION ON THE AEROBIC MICROBIAL FLORA IN POTATO WASTE WATER BY GAMMA IRRADIATION	29
C. STATISTICAL ANALYSIS	38
Comparison of Total Bacterial Counts in Potato Waste Water at the Eight Different Levels of Total Solids	38
DISCUSSION	40
SUMMARY	47
BIBLIOGRAPHY	49
APPENDIX	53

LIST OF TABLES

TABLE		PAGE
1	Total of Organisms Isolated on Ten Trials at 0.0 Megarads.....	25
2	Total of Organisms Isolated on Ten Trials at 0.5 Megarads.....	27
3	Total of Organisms Isolated on Ten Trials at 1.0 Megarads.....	28
4	Total of Organisms Isolated on Ten Trials at 1.5 Megarads.....	28
5	Total of Organisms Isolated on Ten Trials at 2.0 Megarads.....	29
6	Megarad Temperature Relationship Within the Gamma Cell 220 Irradiation Chamber During the Irradiation of the Potato Waste Water.....	39

LIST OF FIGURES

FIGURE		PAGE
1	General screening method for the identification of gram positive microorganisms isolated from haddock fillets and shucked clams.....	11
2	General screening protocal for the identification of gram negative organisms isolated from haddock fillets and shucked clams.....	12
3	General screening method for the identification of gram positive rods isolated from potatoe waste water.....	18
4	General screening method for the identification of gram positive cocci isolated from potato waste water.....	18
5	General screening method for the identification of gram negative organisms isolated from potato waste water.....	19
6	General screening method for the identification of gram negative organisms isolated from potato waste water.....	20
7	The effect of gamma irradiation on the aerobic microflora of potato waste water containing one percent total solids.....	30
8	The effect of gamma irradiation on the aerobic microflora of potato waste water containing two percent total solids.....	31
9	The effect of gamma irradiation on the aerobic microflora of potato waste water containing three percent total solids.....	32

LIST OF FIGURES CONTINUED

FIGURE		PAGE
10	The effect of gamma irradiation on the aerobic microflora of potato waste water containing five percent total solids.....	33
11	The effect of gamma irradiation on the aerobic microflora of potato waste water containing ten percent total solids.....	34
12	The effect of gamma irradiation on the aerobic microflora of potato waste water containing fifteen percent total solids.....	35
13	The effect of gamma irradiation on the aerobic microflora of potato waste water containing twenty percent total solids.....	36
14	The effect of gamma irradiation on the aerobic microflora of potato waste water containing thirty percent total solids.....	37

INTRODUCTION

INTRODUCTION

The survival and growth of the food industry is becoming very dependent on the solution of its problems of water and waste disposal and or reclamation. Past experience has shown that the quantities of water used per unit of product for most operations are usually excessive and as a result the quantities that are physically, chemically, and microbicaly polluted are larger than necessary. The use of large quantities of water will soon have to cease in order to conserve supplies to permit increased food production in future years.

Furthermore, in order to meet the requirements of the present and future pollution abatement laws, the processing cost of food products will probably increase, due to the cost incurred in waste water disposal. Consequently, if a food processing establishment could reuse its processing water by recycling, the cost of water and the cost of waste disposal could be drastically reduced. One problem associated with the reuse of processing water is the presence of micro-organisms. The elimination of these undesirable organisms is an important criteria in water reclamation. The destruction of micro-organisms in waste water particularly from food

processing industries has not been investigated very much, as there is little information on this subject reported in the literature.

For these reasons a study was initiated to evaluate the efficiency of gamma irradiation in the destruction of micro-organisms present in potato waste water.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Gamma Irradiation

Man has been subjected to irradiation since the beginning of time. Without radiant energy there would be no life on earth. The sun is our primary source of radiation.

Today we think of radiation as streams of radiant energy emitted from a source, transmitted through a space to a receptor in which it is absorbed. Some of the more familiar types of radiation are radio rays, ultra violet rays, and x-rays. We are usually not so familiar with alpha, beta, and gamma rays.

X-rays and gamma radiation although identical in nature, originate in different manners. The emission of an electron from an atom occurs when there is a transition of an electron from an outer shell to a vacancy further within the inner shell and is produced by bombarding a heavy metal target with fast electrons in a man-made accelerator. Gamma radiation is the result of a transition of an atomic nucleus from an excited state to a ground state as in certain radioactive materials Silverman and Sinskey (1968).

Bata radiation, arising from radioactive disintegration, are electrons with a single negative charge and a low mass.

Beta radiation from an isotopic source cannot penetrate materials very deeply but electrons produced in man-made machines can be accelerated to extremely high energies with a subsequent improvement in penetrating ability Silverman and Sinskey (1968).

Alpha rays lack sufficient penetrating ability to be of practical value in the food industry.

Research in the 1940's and 1960's revealed the potential of radiation preservation of foods. It was observed that insects could be killed, micro-organisms could be destroyed, and sprout inhibition in certain root vegetables could be obtained by the use of gamma irradiation, according to the U.S. Department of Commerce (1965).

THE EFFECTS OF GAMMA IRRADIATION ON MICROORGANISMS

(1) General Effects

The concept of direct and indirect effects of ionizing irradiation was developed by Lea (1956) and expanded by Hutchinson and Pollard (1961). The action of ionizing irradiation on a molecule is due to the energy released within the molecule itself in contrast to the indirect action resulting from the diffusion of radicals produced in the adjacent liquid. Therefore, it is seen that direct effects of radiation on microorganisms are effective within the organism and are associated with the target sites. Indirect effects in a sense are still within an organism but inactivate the organism by diffusion to and reacting with a sensitive site. This should not be confused with another type of effect which is a solute effect; that of radicals and other radiation produced compounds formed extracellularly in the media.

(2) Effect of Irradiation on Vegetative Cells

Fran *et al* (1950) investigated the effects of x-rays on cells of *E. coli*, *A. aerogenes*, *S. aureus*,

S. marcesans. They concluded that the percentage of organisms killed was the same regardless of their initial concentration. U.S. Department of the Interior (1960) and Proctor (1960) investigated the destruction of microorganisms in fish products by ionizing radiation of gamma rays. These investigations showed considerable success in extending the shelf life of fish products.

The effects of gamma irradiation on the natural microbial of soil population was investigated by Davis, Sheldon, and Auerbach (1956). The organisms were classified according to the media which supported their growth. They reported that small dosages in the order of 10^3 to 10^4 rads would reduce the number of organisms by at least fifty percent. Tarply et al (1953) reported that molds and non-sporulating bacteria were much less resistant to gamma irradiation than sporulating bacteria. He also found that mixed suspensions of bacterial cultures when irradiated with gamma rays showed slightly increased survival over pure bacterial suspensions. Monib and Zayed (1962) irradiated a sample of loam soil at pH 8.0, supplemented with one percent glucose, at dosages of 0.1 - 1.0 megarads. Their investigation revealed that the destruction of microorganisms in loam soil was directly related to the dosage. However, they

reported that the soil was not free of microorganisms even after treatment with 1.0 megarads of gamma irradiation. Azotobacter was destroyed by 1.0 megarads and was very much reduced in numbers by smaller dosages whereas spores of the genus Clostridium were unaffected by the smaller irradiation dosages and exhibited much more radiation resistance at higher levels of irradiation.

EFFECT OF ENVIRONMENT ON MICROORGANISMS DURING IRRADIATION

Work performed by Gunter and Kohn (1958); Eldjarm and Phil (1961); Kempe (1963); Niven (1958); Proctor *et al* (1955); Shabyj *et al* (1965); Grecz (1965); and Anellis *et al* (1965) showed that the presence of compounds such as glycerol, sodium formate, proteins, and carbohydrates increased the irradiation resistance of microorganisms. Hollaender *et al* (1951); Moos (1952); and Biagini (1953) found organisms to be less sensitive to irradiation when amino acids, proteins, or the mixture of compounds present in nutrient broth were present in the media.

There have been very few studies on the effects of irradiation on microorganisms in their natural habitats, but it is evident from present knowledge that much higher dosages of irradiation are needed to destroy

microorganisms in a liquid environment. Stotzky and Mortensen (1959) observed very little destruction of microorganisms following irradiation of a peaty soil with dosages up to 0.25 megarads. McLaren et al (1957) obtained a complete lethal effect on the microbial flora in a clay loam soil with a dosage of 2.2 megarads.

Tarply et al (1953) reported that young bacterial cells were more sensitive to irradiation than old cells; vegetative more sensitive than bacterial spores and bacteria in the dry state were more resistant than bacteria in aqueous solutions. They also observed that irradiation at low intensity over long periods of time was of no greater efficiency than irradiation at higher intensities over shorter periods of time.

Moos (1953) reported that bacteria in the dry state were much less affected by irradiation than bacteria in solution.

THE EFFECT OF IRRADIATION ON BACTERIAL SPORES

The effects of irradiating various spores in the presence of proteins, nutrient broth, ascorbic acid, and thiourea have been described by Edwards et al (1954); and Morgan and Reed (1954). They found that, although these compounds had an effect, spores in the presence of

these compounds were less susceptible to protective influences than the vegetative cells were.

The organism which dominates most considerations for determining the sterilization dosage required for foods is Cl. botulinum, one of the most radiation resistant spores. Pratt et al (1958) inoculated 10,000 spores per gram of Cl. botulinum type A into foods and found that for sterility dosages of 3.0 megarads in peas, 3.8 megarads in chicken soup, and 4.0 megarads in pork were required. Kempe, Graskoski, and Gillies (1954) reported that while the number of spores of Cl. botulinum varied from 0.4 to 40,000 per gram the dosage required for sterilization of the cooked meat varied from 2 to 4 megarads. Goldblith and Proctor (1956); Westheim (1959; and Thornley (1957), found that in order to sterilize milk and milk products and chicken 4 megarads were required.

Briggs (1960) reported that there is quite a difference in radiation resistance between different species of the genus Bacillus. He reported that some showed non-linear inactivation curves while others exhibited linear inactivation curves. Donnellan and Morowitz (1957) observed linear relationships for the irradiation of some dried species of Bacillus, while some other ones

exhibited a non-linear relationship. Curran (1951) stated that the most resistant spores required about 2,000,000 Roentgens of cathode ray energy for their destruction, while the corresponding value for the vegetative cells required only about one quarter of that energy level.

IDENTIFICATION PROCEDURES

Masurovsky, Voss and Goldblith (1963) studied changes in the microbial flora of Haddock fillets and Shucked Soft Shelled Clams after irradiation with cobolt 60 gamma rays. Their data showed that a decided change occured in the constitution of the microbial population of both produces. The flora changed from mixed prior to irradiation to a predominately gram positive flora after irradiation. The great majority of those surviving microorganisms were: Micrococci, spore-forming Bacillus, and certain yeasts, molds, and Actinomyces. The identification schemes set up by Masurovsky et al (1963) are shown in Figures 1 and 2.

Information relating to the irradiation resistance of microorganisms with regards to total solid concentrations is non-existent.

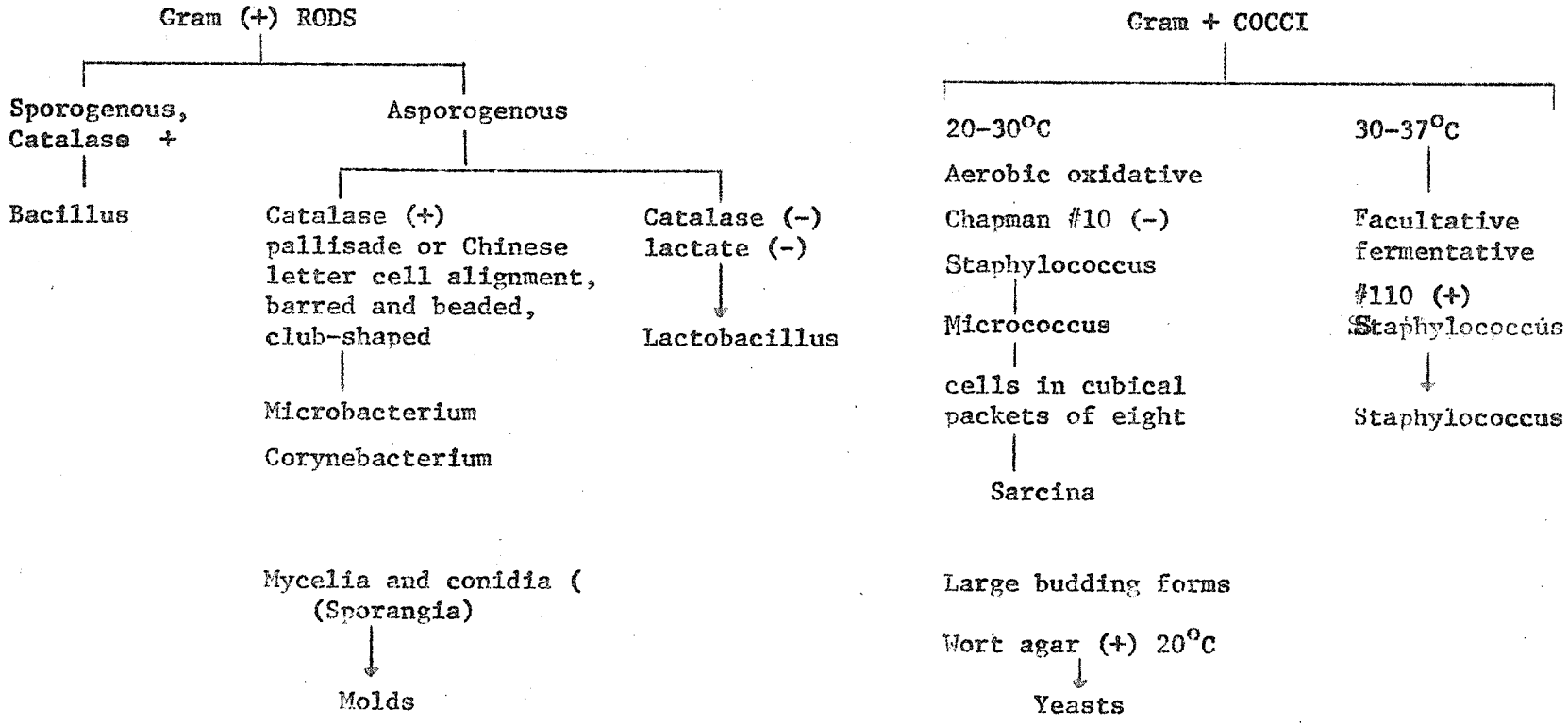


FIGURE 1. General screening method for the identification of gram positive microorganisms isolated from haddock fillets and shucked clams.

SCOPE OF THE INVESTIGATION

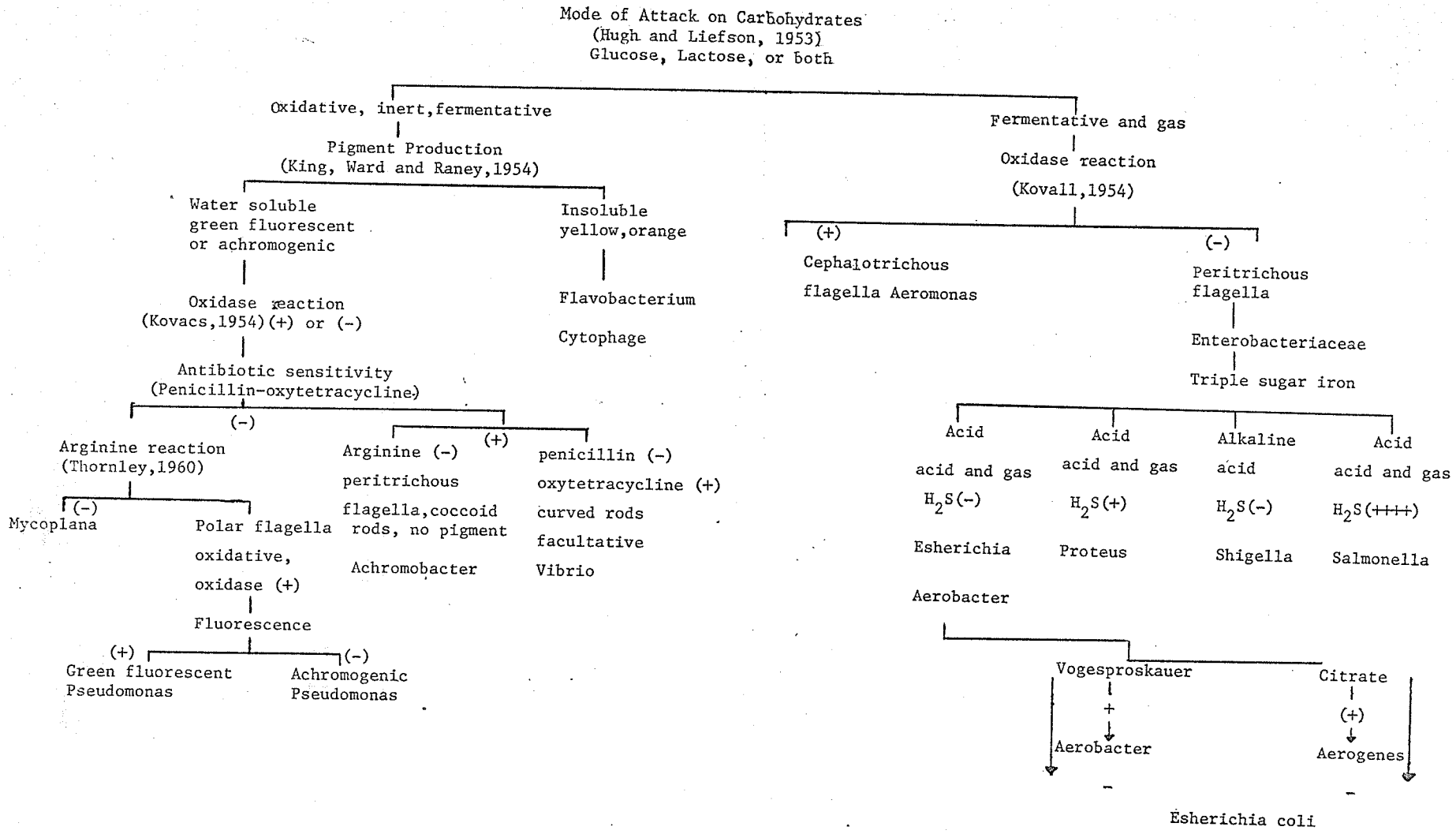


FIGURE 2. General screening protocal for the identification of gram negative organisms isolated from haddock fillets and shucked clams.

MATERIALS AND METHODS

SCOPE OF THE INVESTIGATION

The principle objectives of this study were:

1. To identify the aerobic microbial genera present before and after irradiation with gamma rays in potato waste water.

2. To compare the microbial population of irradiated and non-irradiated waste water of abrasively peeled potatoes.

3. To determine whether various levels of total solids in the potatoe waste water had an effect on the radio-resistance of the microflora present.

4. To determine whether the aerobic microorganisms in the potato waste water could be made more sensitive to gamma irradiation when sodium hypochlorite was added to the potato waste water.

5. To determine whether the aerobic microorganisms in the potatoe waste water could be made more sensitive to gamma irradiation when hydrogen peroxide was added to the potatoe waste water.

MATERIALS AND METHODS

Netted Gem potatoes grown at the University of Manitoba were used throughout this investigation. Four average sized potatoes were peeled by abrasion in a Hobart Abrasion peeler. Four litres of sterile distilled buffered water were used to peel the potatoes. This gave a potato waste water made up of approximately one percent total solids with a pH of 7.1. This waste water was used for microbiological studies. This waste was similar to commercial waste excepting the fact that no micro-organisms were added to the waste through the water supply.

Preliminary trials with several different recovery media such as: nutrient-agars tryptone glucose yeast-extract-agar, trypticase soy agar and soil extract agar were performed. Tryptone glucose yeast extract-agar recovered more aerobic microorganisms than the other media. Hence tryptone glucose yeast extract agar was used in the investigation.

The plates were incubated at 32°C for 48 hours. Preliminary trials indicated that 32oC for 48 hours recovered more microorganisms than 28°C or 32°C temperatures.

The abrasion peeler was sanitized prior to use

with a solution containing 200 ppm available chlorine. The abrasion peeler was then flushed with sterile distilled water. Potato samples were then placed into the peeler and peeled while washing them with four litres of sterile distilled buffered water. The waste water containing the peelings was then collected in sterile containers. Aliquots of 15 ml of waste water were then transferred to a set of five sterile test tubes. One tube was used as the control sample while the other four were subjected to various levels of gamma irradiation. The radiation dosages used were 0.5, 1.0, 1.5 and 2.0 megarads.

The radiation source was a Cobalt 60 Gamma Cell Model 220 supplied by the Atomic Energy Commission of Canada. At the time of the investigation the source delivered an absorbed dosage varying from 1.229×10^6 rads per hour in June of 1968 to 1.0665×10^6 rads per hour in July of 1969. Detailed discussion of the source's calibration, construction of the shield, and various appliances related to the use of the equipment was supplied by the Atomic Energy Commission of Canada Ltd.

The four test tubes containing the samples were placed in the chamber of the Gamma Cell and a sample was removed after it had received the required dosage.

All samples were irradiated at ambient temperature of air atmosphere.

Standard Plate counts according to Standard Methods (1960) were performed on each of the samples and the number of colonies were determined after incubation at 32°C for 48 hours.

Identification of organisms in the control samples and in the samples which were treated with 0.5 megarads of radiation involved the selection of 25 colonies from each of duplicate plates and transferring each to nutrient broth. A total of 50 cultures were prepared from each sample. The colonies to be picked were selected by means of a 5 x 5 grid giving a pattern with 25 intersections. The colony nearest the intersection was picked.

For the samples receiving 1.0, 1.5 and 2.0 megarads respectively, all of the colonies in each plate were picked and identified. The reason being that although there was still growth on the plates the counts were usually less than thirty colonies per plate.

After incubation for 24 hours at 32°C the nutrient broth cultures were streaked onto tryptone glucose yeast extract agar plates. This was done to ensure that pure culturesswould be maintained. Furthermore colony characteristics and catalase tests could be recorded from the

plates. Once the purity of the culture had been established colonies from the tryptone glucose yeast extract plates were picked and transferred into nutrient broth tubes and incubated at 32°C for 18 hours. Gram stains, according to the Manual of Microbial Methods (1957) were then performed on each one of the samples and the staining reaction and the morphology of the culture were recorded. The gram positive catalase positive rods were transferred from the pure culture in nutrient-broth to another tube of nutrient broth and were incubated at 32°C for five days after which smears were made and stains according to the Manual of Microbial Methods (1957) were performed.

By using the identification scheme in Figure 3 the gram positive rod organisms could be identified. All of the Gram negative rods and the Gram positive cocci were inoculated into two tubes of Hugh and Leifson media according to Hugh and Leifson (1953).

Following the Hugh and Leifson test the Gram negative organisms could be identified according to the schemes in Figure 5 and 6. The Gram positive cocci could also be identified according to the identification scheme in Figure 4. For the other portion of the study the waste was collected in the same manner but the percentage of total solids in the waste water was

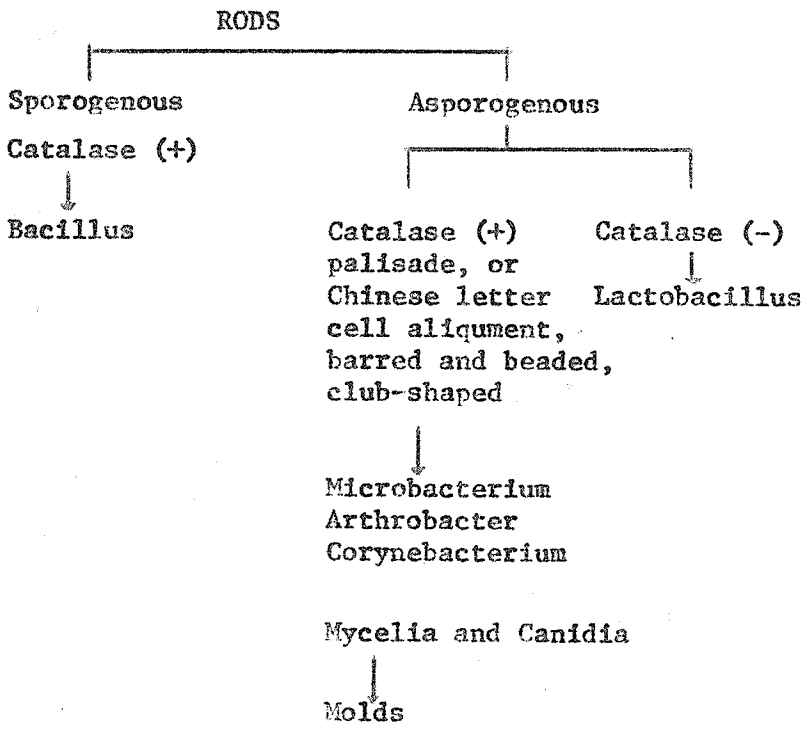


FIGURE 3. General screening method for the identification of gram positive rods isolated from potatoe waste water.

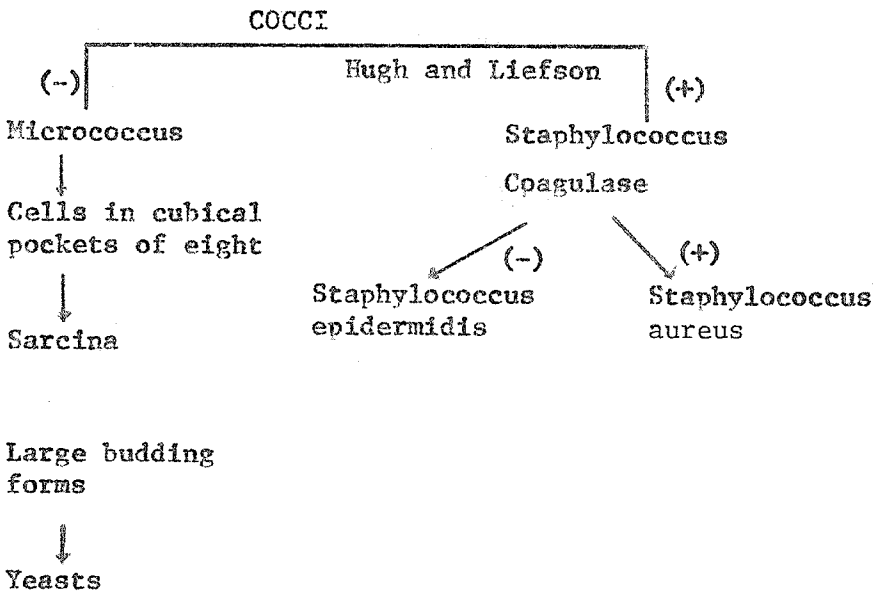


FIGURE 4. General screening method for the identification of gram positive cocci isolated from potatoe waste water.

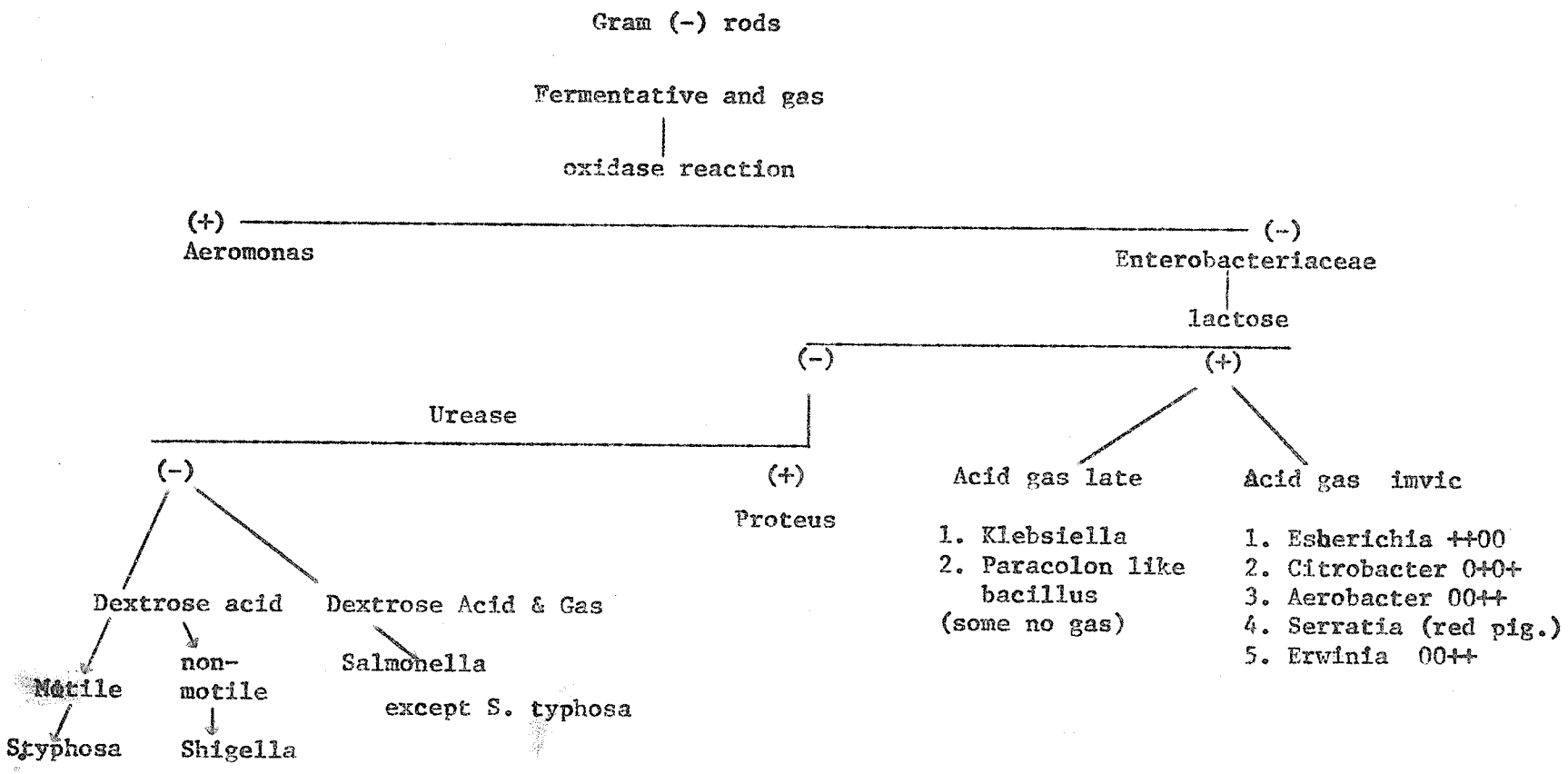


FIGURE 5. General screening method for the identification of gram negative organisms isolated from potatoe waste water.

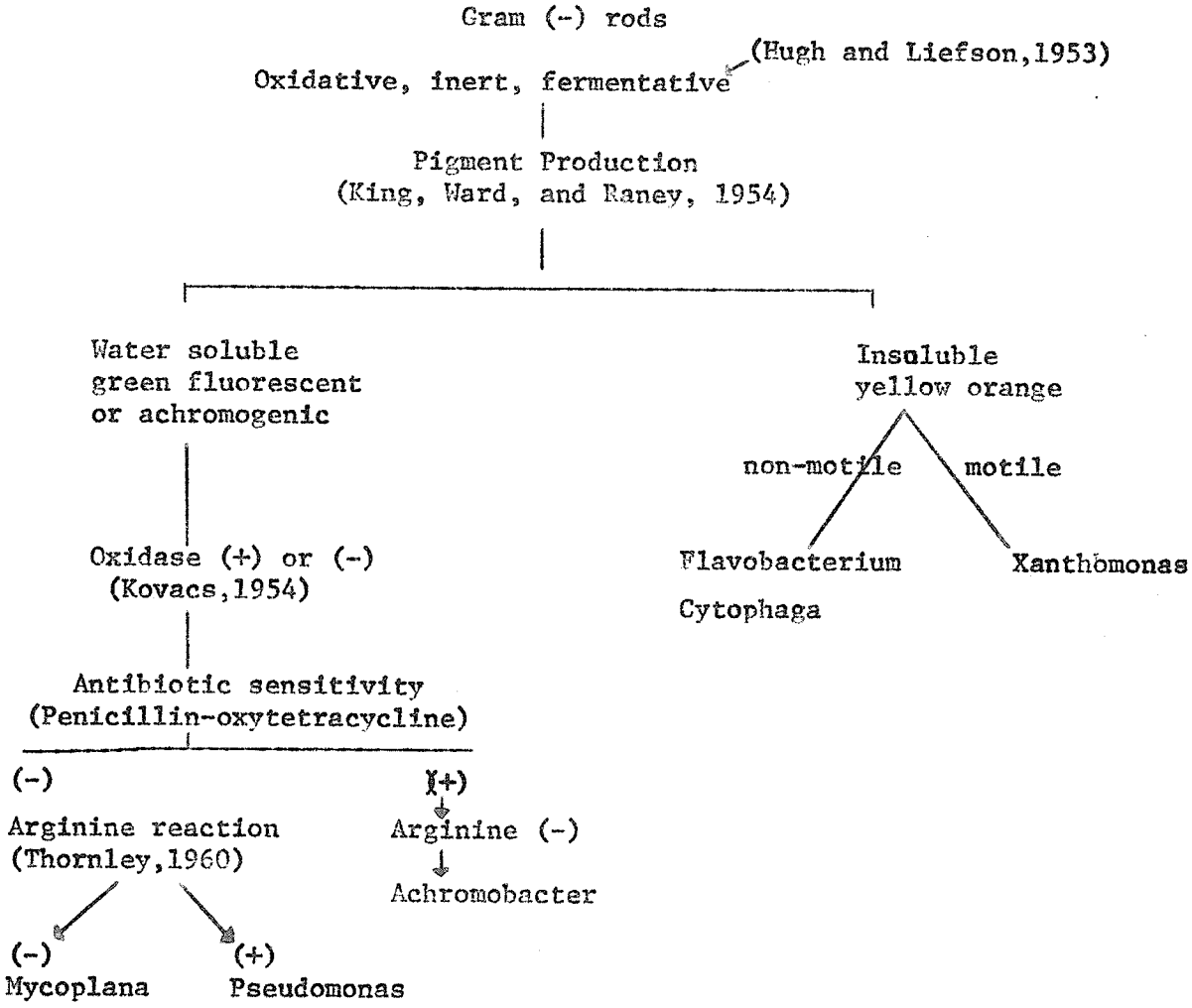


FIGURE 6. General screening method for the identification of gram negative organisms isolated from potatoe waste water.

altered to various levels.

Potatoes from the same source were sliced into thin slices and then frozen in liquid nitrogen. The slices were then freeze-dried in a Virtis laboratory model freeze-drier. After drying these slices were ground into a powder in a small laboratory grinder. The ground potato powder was then stored in sterile glass containers.

The waste collected from the abrasion peeler contained normally approximately one percent total solids. The percentage of total solids could be altered to any desired level by the addition of the potato powder. The desired level was achieved by adding the required amount to 100 grams of potato waste water. Eight levels of total solids were studied and they were as follows: 1%, 2%, 3%, 5%, 10%, 15%, 20% and 30%.

After the total solids level had been adjusted total solids tests were performed on the waste to determine whether proper amounts of potato powder had been added.

Twenty grams of waste water was weighed into dry aluminum dishes. All samples were performed in duplicate. After weighing the dishes were places in a steam bath and the moisture was evaporated. After evaporation the samples were placed in a vacuum oven at

a temperature of 90°C. The vacuum was regulated at 25 inches of mercury. The samples were dried in the oven for 12 hours. After drying the samples were then placed in a dessicator to cool. After cooling the samples were weighed and the total solids were calculated.

After the total solids content of the potato waste water had been adjusted the samples were irradiated in 15 ml aliquots in sterile test tubes in the 220 Gamma Cell at levels of 0.0, 0.5, 1.0, 1.5 and 2.0 megarads of radiation. After irradiation the samples were plated on S.P.C. agar plates. The plates were incubated at 32°C for 48 hours after which the total counts were recorded. The data from this investigation was subjected to a statistical analysis in order to ascertain whether there were any significant differences in the irradiation resistance of the organisms in potato waste water containing different levels of total solids.

An investigation was also performed on the addition of sodium hypochlorites to potato waste water containing one percent total solids.

Standard plate counts according to Standard Methods (1960) were compared on the irradiated potato waste water with no sodium hypochlorite added and irradiated potato waste water with sodium hypochlorite added.

Hydrogen peroxide was also added to potato waste water at levels up to 500 ppm. Again the percent total solids content potato waste water was one percent. Standard plate counts according to Standard Method (1960) were performed on the waste water with hydrogen peroxide added, and controls with no hydrogen peroxide. Also counts on the waste with hydrogen peroxide added coupled with irradiation at dosages of 0.5, 1.0, 1.5 and 2.0 megarads and counts on the waste water with no hydrogen peroxide added, coupled with irradiation dosages of the same level, were compared.

RESULTS

RESULTS

A. THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER AT SELECTED GAMMA IRRADIATION DOSAGES

The total aerobic microbial population of the ten trials of identification at an irradiation level of 0.0 megarads was determined and the results are presented in Table 1. The micrococcus genus was the most predominately occurring organism making up 38.8 percent of the isolated organisms. Organisms belonging to the Bacillus genus were also fairly predominate making up 24.4 percent of the total. The Pseudomonas genus made up 5.6 percent of the total. The genuses of Aerobacter, Corynebacterium, Erwinia, Sarcina, Streptomyces, and Xanthomanas made up 3.2, 2.0, 4.4, 2.0, 3.2 and 3.2 percent respectively. Mold growth at this irradiation level amounted to 1.6 percent of the total. Yeast growth was a little more predominant making up 3.0 percent of total microbial flora. Those organisms appearing least predominantly belonged to the genuses Alcaligines, Arthrobacter, Flavobacterium, Lophomonas, Proteus and Staphylococcus. The percentages of the total made up by these organisms varied from 0.6 percent to 1.4 percent. Of the 500 isolations at this level only one Escherichia genus was found resulting in 0.2 percent of the total.

TABLE 1

TOTAL OF ORGANISMS ISOLATED ON TEN TRIALS
AT 0.0 MEGARADS

Genus	Total No. Isolated	Percentage of Total
Aerobacter	16	3.2
Alcaligines	3	0.6
Arthrobacter	7	1.4
Bacillus	122	24.4
Corynebacterium	10	2.0
Erwinia	22	4.4
Esherichia	1	0.2
Flavobacterium	4	0.8
Lophomonas	4	0.8
Micrococcus	194	38.8
Mold	8	1.6
No growth	16	3.2
Proteus	3	0.6
Pseudomonas	28	5.6
Sarcina	10	2.0
Staphylococcus	5	1.0
Streptomyces	16	3.2
Xanthomonas	16	3.2
Yeast	15	3.0
Total	500	100.00

The total of the organisms isolated on ten trials at an irradiation level of 0.5 megarads is presented in Table 2. A marked difference was seen here in the total percentage made up by each genus from the irradiation level of 0.0 megarads. At a level of 0.5 megarads the predominant genus was Bacillus making up 81.8 percent of the total number of isolations. Mold isolations accounted for 7.2 percent of the total. Micrococcus also occurred fairly consistently and made up 5.0 percent. Aerobacter, Alcaligines, Arthrobacter, Erwinia, and Streptomyces accounted for percentages varying from 0.2 percent to 0.4 percent. A considerable percentage of 4.6 percent did not grow at this level.

The total of the genuses isolated at an irradiation level of 1.0 megarads is shown in Table 3. Of the total of 2.5 isolations here .96 belonged in the genus Bacillus. Mold isolations accounted for 8.379 percent of the total. At this level only one organism was lost in the picking and transferring procedure.

The total of the organisms isolated at irradiation levels of 1.5 and 2.0 megarads are shown in Tables 4 and 5. Of the total number of organisms isolated here 100 percent of them were Bacillus.

TABLE 2

TOTAL OF ORGANISMS ISOLATED ON TEN TRIALS
AT 0.5 MEGARADS

Genus	Total No. Isolated	Percentage of Total
Aerobacter	2	0.4
Alcaligines	1	0.2
Arthrobacter	2	0.4
Bacillus	409	81.8
Erwinia	1	0.2
Micrococcus	25	5.0
Mold	36	7.2
No growth	23	4.6
Streptomyces	1	0.2
Total	500	100.0

TABLE 3

TOTAL OF ORGANISMS ISOLATED ON TEN TRIALS AT 1.0 MEGARADS

Genus	Total No. Isolated	Percentage of Total
Bacillus	196	91.156
Mold	18	8.379
No growth	1	0.465
Total	215	100.000

TABLE 4

TOTAL OF ORGANISMS ISOLATED ON TEN TRIALS AT 1.5 MEGARADS

Genus	Total No. Isolated	Percentage of Total
Bacillus	134	100
Total	134	100

TABLE 5

TOTAL OF ORGANISMS ISOLATED ON TEN TRIALS AT 2.0 MEGARADS

Genus	Total No. Isolated	Percentage of Total
Bacillus	28	100
Total	28	100

B. THE RATES OF DESTRUCTION OF THE AEROBIC MICROBIAL FLORA IN POTATO WASTE WATER BY GAMMA IRRADIATION

The rates of destruction of the aerobic microbial flora in potato waste water at the eight different levels of total solids are shown in the Appendix (Tables 1 to 69 inclusive). The rates of destruction curves are shown in Figures 7 to 14 inclusive. The destruction by gamma irradiation in the potato waste water of the aerobic microbial flora appears to have two distinct rates of destruction.

Figure 7. The effect of gamma irradiation on the aerobic microflora of potato waste water containing one percent total solids.

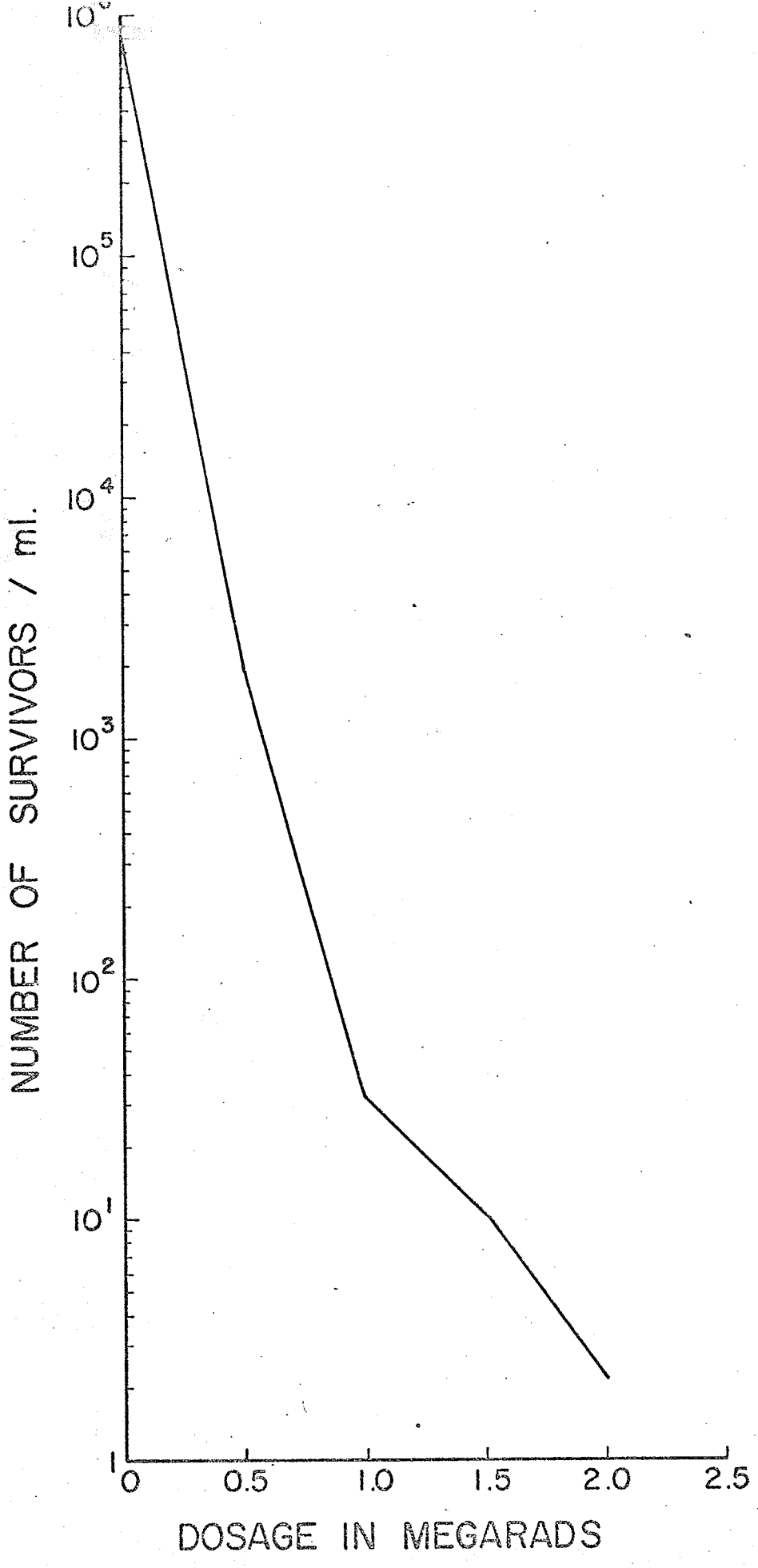

NUMBER OF SURVIVORS / ml.
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

Figure 8. The effect of gamma irradiation on the aerobic microflora of potato waste water containing two percent total solids.

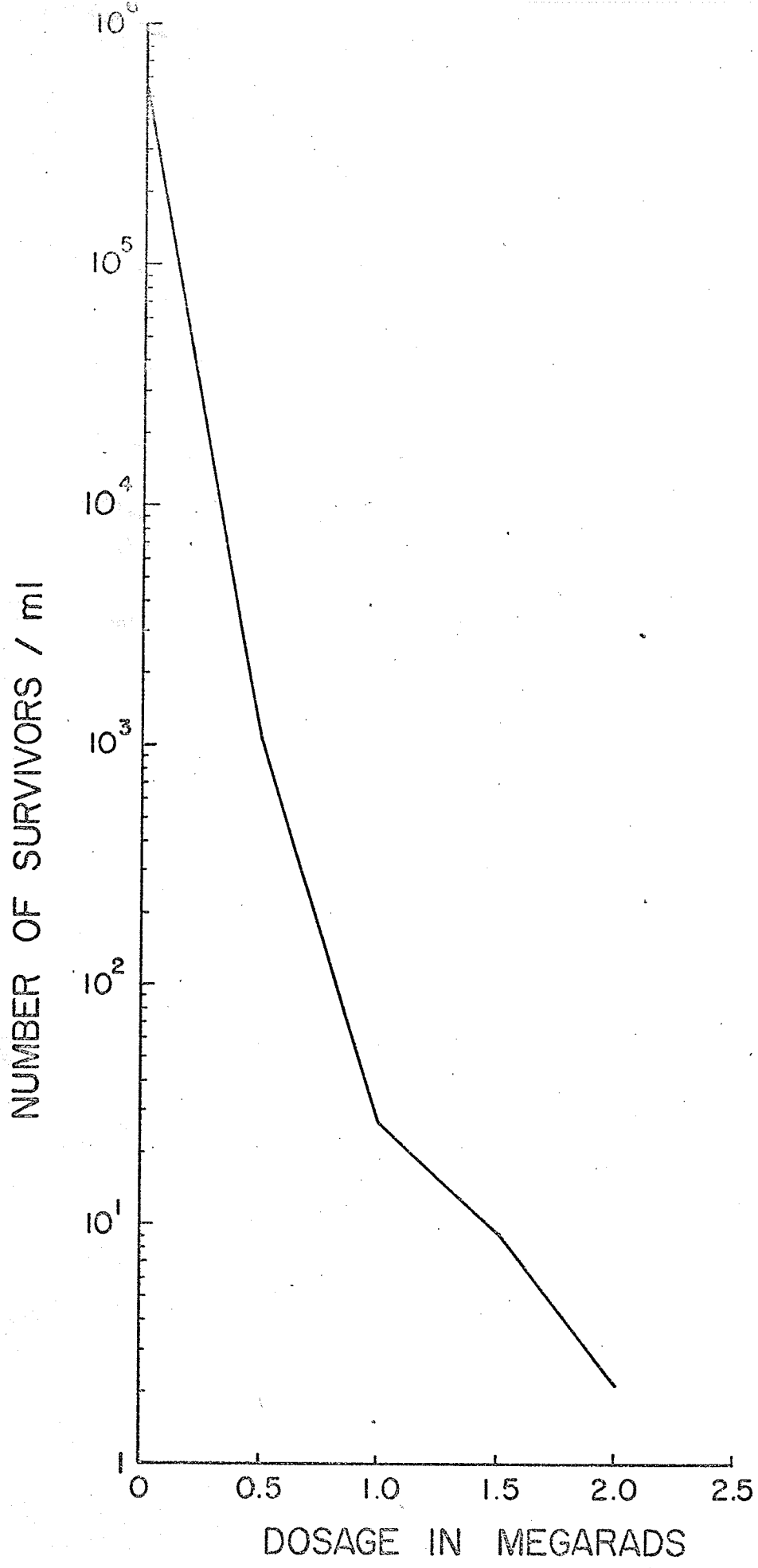

NUMBER OF SURVIVORS / ml
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

Figure 9. The effect of gamma irradiation on the aerobic microflora of potato waste water containing three percent total solids.

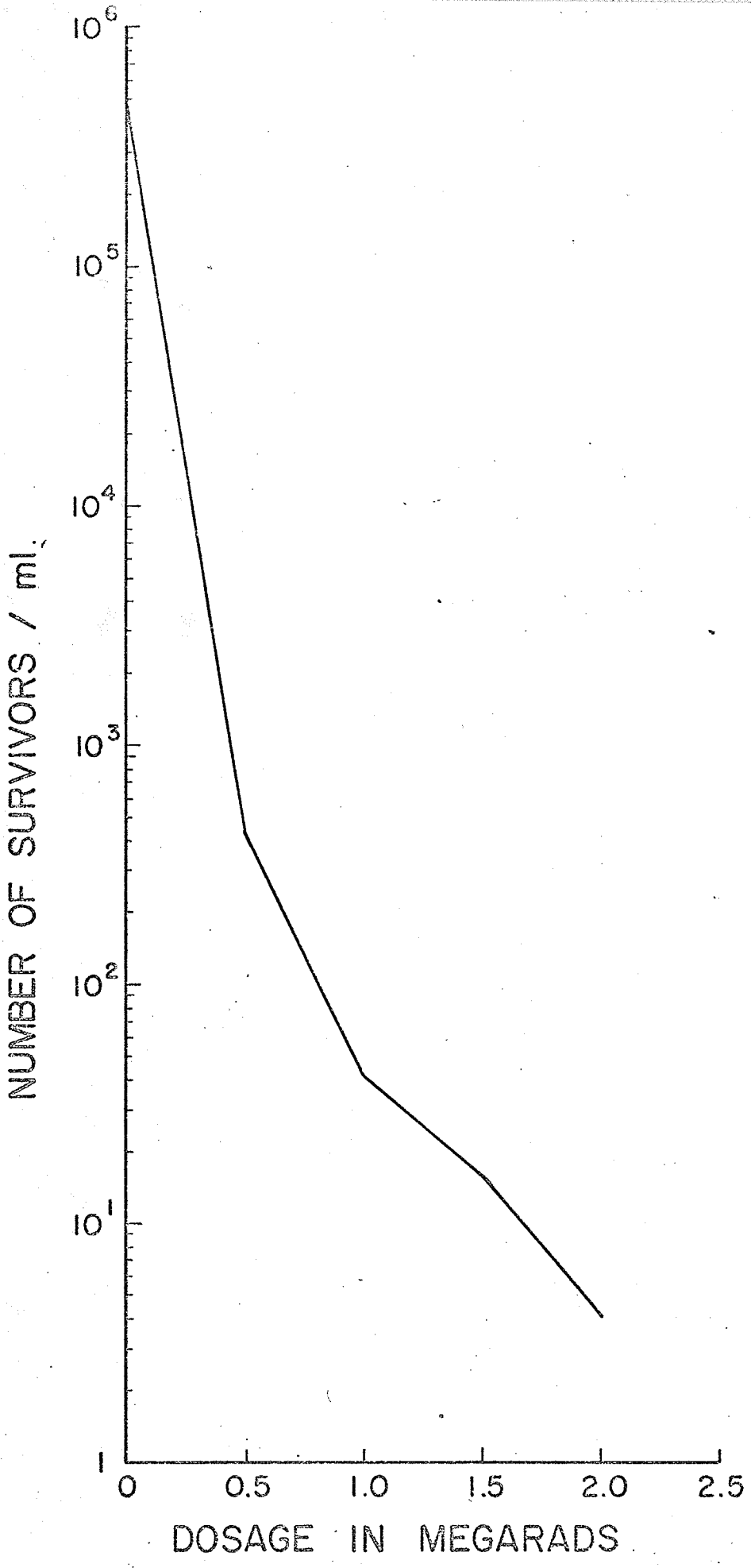

NUMBER OF SURVIVORS / ml.
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

Figure 10. The effect of gamma irradiation on the aerobic microflora of potato waste water containing five percent total solids.

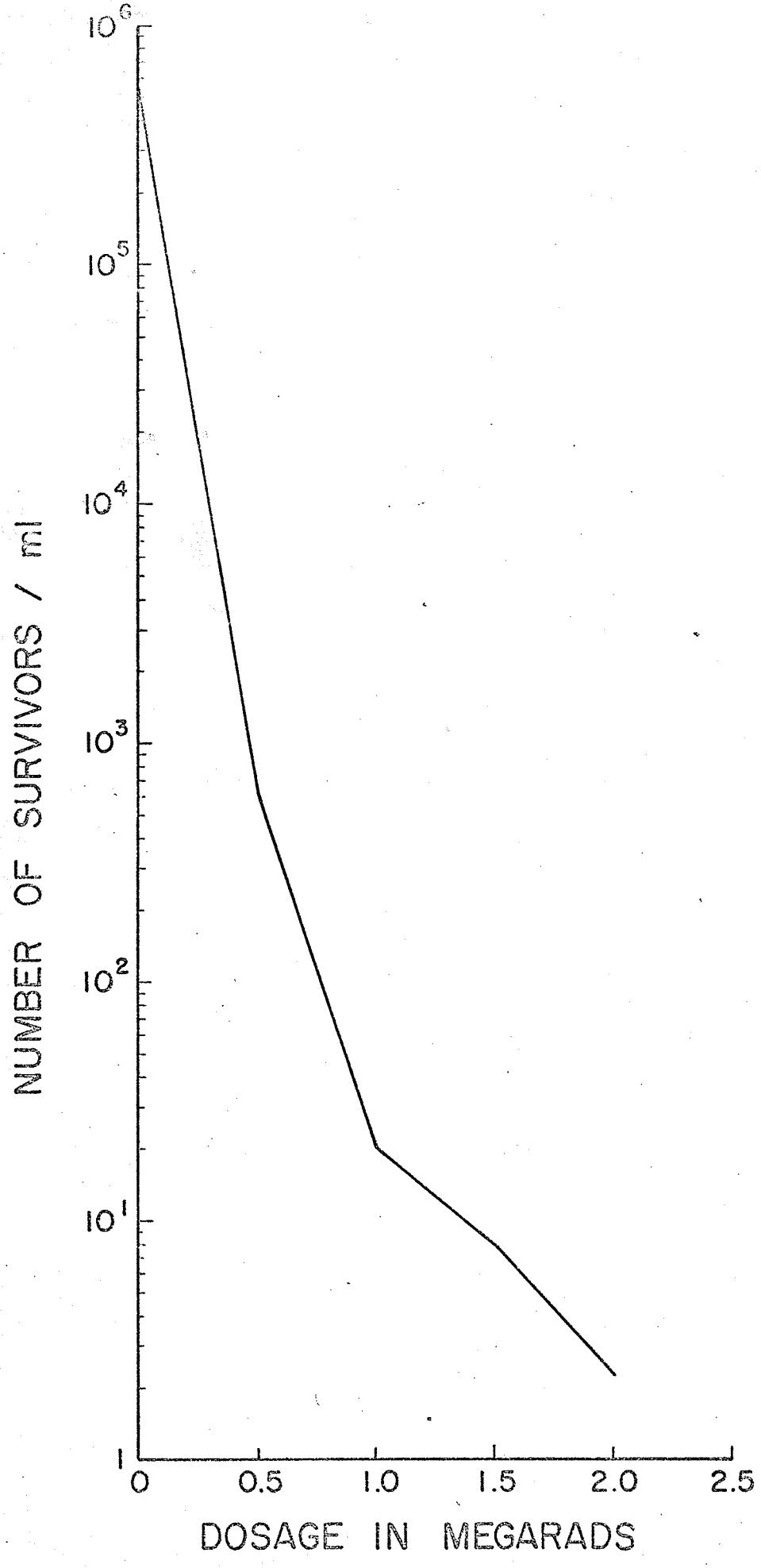

NUMBER OF SURVIVORS / ml
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

Figure 11. The effect of gamma irradiation on the aerobic microflora of potato waste water containing ten percent total solids.

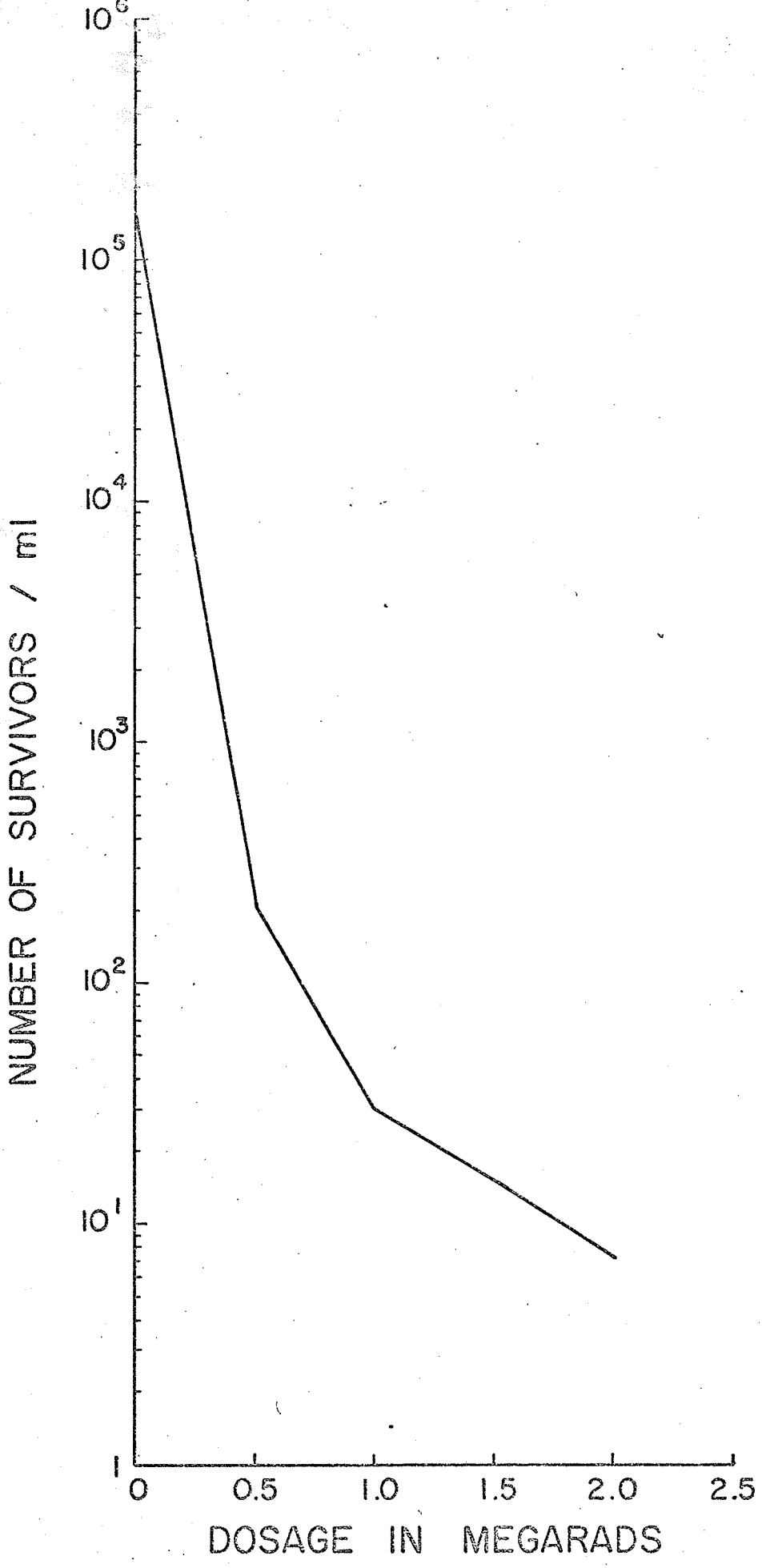

NUMBER OF SURVIVORS / ml
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

Figure 12. The effect of gamma irradiation on the aerobic microflora of potato waste water containing fifteen percent total solids.

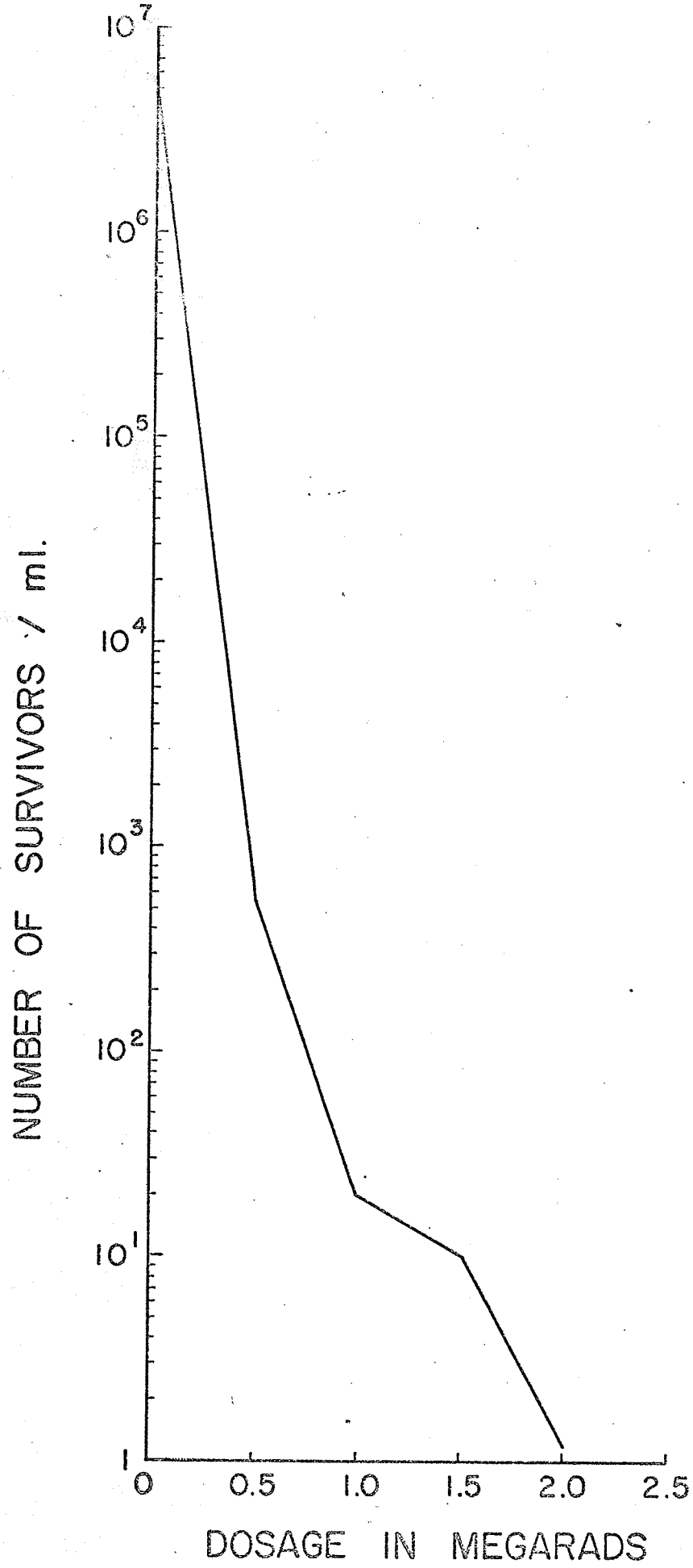

NUMBER OF SURVIVORS / ml.
10^7
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

Figure 13. The effect of gamma irradiation on the aerobic microflora of potato waste water containing twenty percent total solids.

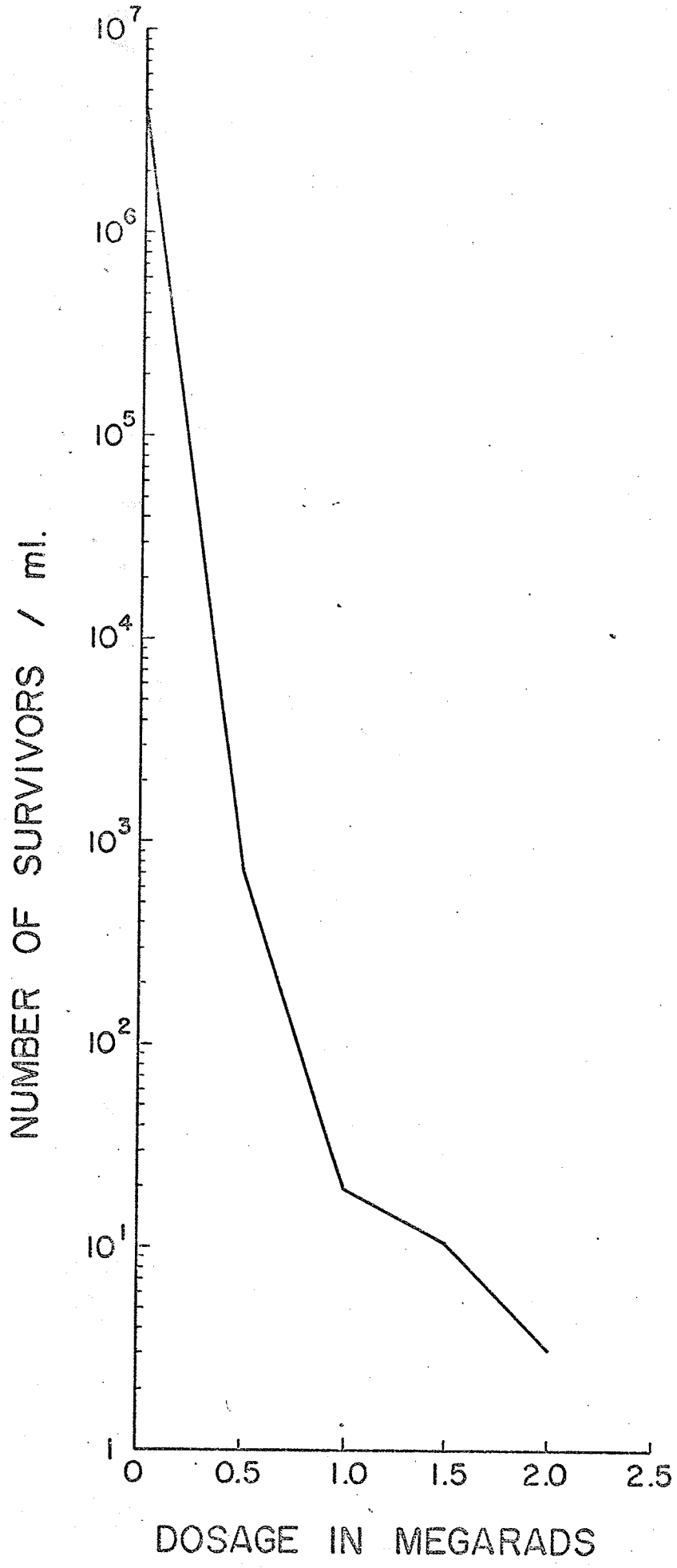
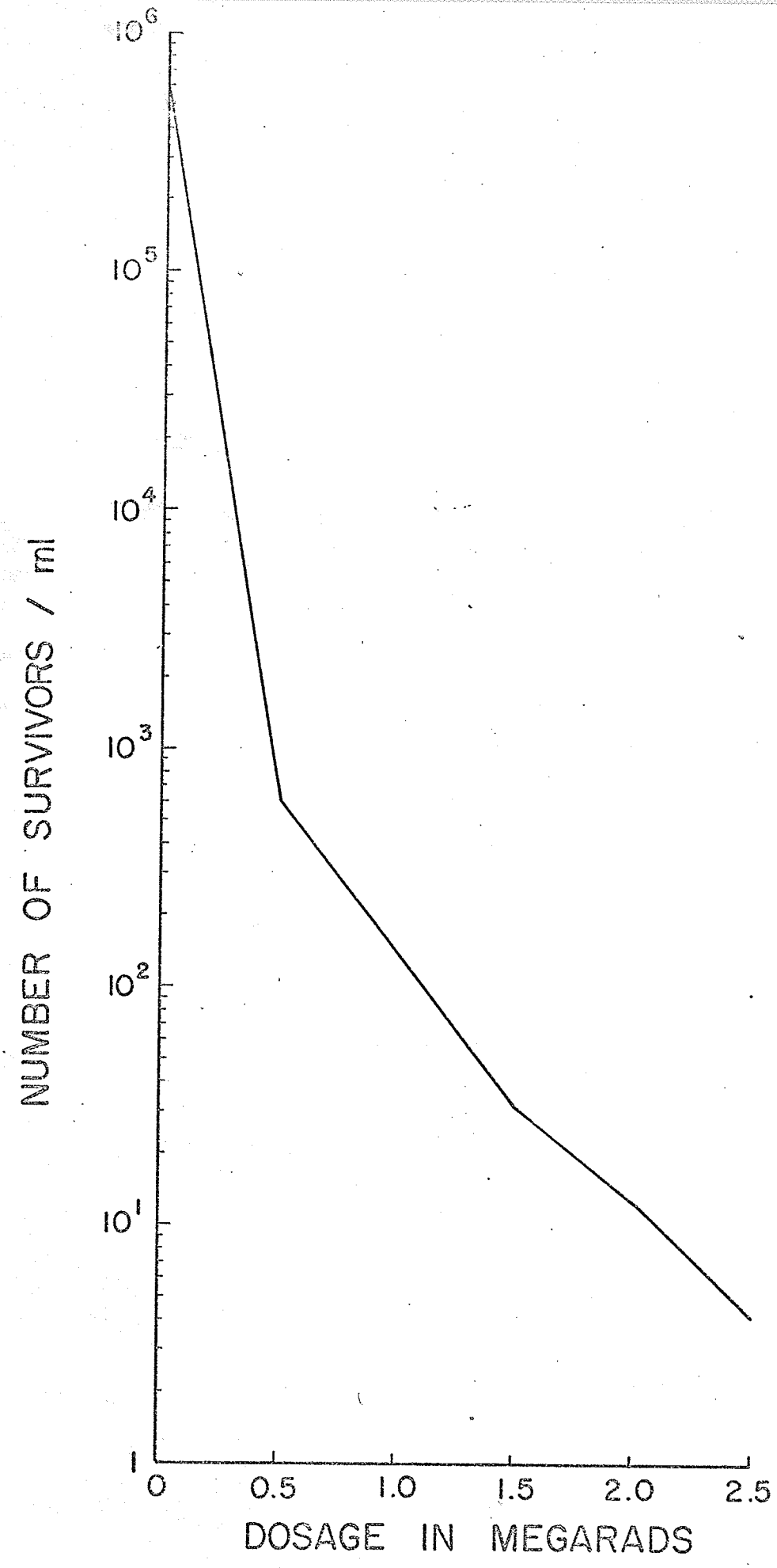

NUMBER OF SURVIVORS / ml.
10^7
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

Figure 14. The effect of gamma irradiation on the aerobic microflora of potato waste water containing thirty percent total solids.


NUMBER OF SURVIVORS / ml
10^6
10^5
10^4
10^3
10^2
10^1
1
0
0.5
1.0
1.5
2.0
2.5
DOSAGE IN MEGARADS

C. STATISTICAL ANALYSIS

Comparison of Total Bacterial Counts in Potato Waste Water at the Eight Different Levels of Total Solids.

An analysis of variance test was performed to determine whether there was a difference in radiation resistance of the organisms when the total solids content of the potato waste water was raised.

The results indicated that there was no significant difference in the radiation resistance of organisms at higher levels of total solids. (Appendix Table 69).

The average temperature at which the potato waste water was irradiated $104^{o}F \pm 1^{o}F$. No attempts were made to control the temperature of the potato waste water during irradiation for it was felt that in commercial practice this would also be the case.

TABLE 6

MEGARAD TEMPERATURE RELATIONSHIP WITHIN THE GAMMA CELL 220 IRRADIATION CHAMBER DURING THE IRRADIATION OF THE POTATO WASTE WATER.

Irradiation Level (Megarads)	Temperature (^{o}F)
0.0	$82^{o}F$
0.5	$103^{o}F$
1.0	$104^{o}F$
1.5	$104^{o}F$
2.0	$104^{o}F$

DISCUSSION

DISCUSSION

This study was initiated to identify the aerobic microbial genera present before and after irradiation, to examine the possibility of utilizing dosages of gamma irradiation to destroy the microflora of potato waste water and also to determine whether various levels of total solids in the potato waste water had an effect on the radio-resistance of the microflora present.

The results obtained in this study revealed that Micrococcus and Bacillus were the most predominant organisms found in the irradiated waste water. This is quite easily explained by the fact that both organisms do predominate in soils. The population of organisms present in potato waste water was derived from the soil in this study because sterile water and sterile equipment were used throughout the study. The other organisms isolated in the study are also found in soil except for the genera Staphylococcus. The presence of Staphylococcus may be explained by the fact that the potatoes had been handled by human hands and may have been contaminated with the organism in this manner.

After the waste had been subjected to an irradiation dosage of 0.5 megarads a marked difference was observed in the total percentages made up by each genera of the population. Bacillus became the predominant

organism present in the population. This is due mainly to the Bacillus genus's ability to form spores. Radio-resistance of spore formers was also observed by Pratt et al (1958), Kempe, Graskoski, and Gillies (1954), Goldblithe and Proctor (1956), Wertheim (1959), and Thornly (1957) in their investigations. The fact that the micrococci made up as large a percentage of the population as observed may be explained by the fact that micrococcus organisms have been found to be the most radio-resistant vegetative cells (Anderson, et al (1956)) Molds also made up a considerable proportion of the total population. This may also be explained by the fact that molds are spore formers and that spores are more radio-resistant than vegetative cells. Moriarity (1950) found certain molds much more radio-resistant than vegetative bacterial cells. The other bacteria present made up only a small proportion of the total population. No radio-resistance characteristics should be contributed to them. It is quite probable that the waste had not been subjected to enough radiation to destroy all vegetative cells.

The genera isolated after 1.0 megarads of irradiation was made up of 91.16 percent Bacillus and 8.38 percent mold. Again Bacillus and molds are spore formers

and their radio-resistance may be explained by Pratt et al (1958), Kempe, Graskoski, and Gillies (1954), Goldblithe and Proctor (1956), Wertheim (1959), and Thornly (1957).

Bacillus was the only genus isolated after 1.5 and 2.0 megarads of gamma irradiation. The ability of this genus to form spores and the resistance of these spores to irradiation would account for this observation.

Molds were not found after dosages 1.5 and 2.0 megarads. This may be explained by the fact that molds form spores mainly as a means of reproduction although they do have the ability to withstand certain environmental hazards such as drought. Bacterial spores are formed, however, mainly as a means of preservation and are able therefore to withstand more severe environmental factors hazardous to the life of microorganisms.

It was also observed during the investigation that on occasion after 1.5 or 2.0 megarads there was no surviving aerobic microflora. This may be explained by the fact that different species of Bacillus have spores with varying degrees of radio-resistance. Briggs (1960) reported that there is a difference in radiation resistance between species of the genus Bacillus. He reported that

some species showed non-linear inactivation curves while others exhibited linear inactivation curves. Donnellan and Morowitz (1957) had similar findings. Curran (1951) stated that the most resistant spores required about 200,000 Roentgens of Cathode ray energy for their destruction while the corresponding value for the vegetative cells required only one quarter of that energy level.

Dunn, Campbell, Fram and Hutchins (1948) irradiated large populations of soil organisms. They found that bacterial spores could be greatly reduced in numbers of 2.0 megarads molds and yeasts were generally destroyed by 1.0 megarads. Vegetative cells were all destroyed at dosages of less than 1.0 megarads of irradiation. Proctor and Goldblith (1951) and Moriarity (1950) published very similar results. The results of this investigation compare well with the findings of these authors.

It should be mentioned that these organisms were irradiated in a liquid medium and that most of the total solids present in the potato waste water was derived from the starch of the potatoes. Work done by Gunter and Kohn (1958), Eldjarm and Phil (1961), Kempe (1963),

Niven (1958), Proctor et al (1955), Shabyj et al (1965), Grecz (1965), and Anellis et al (1965) showed that compounds such as glycerol, sodium formate, proteins, acid carbohydrates increased the irradiation resistance of microorganisms. This may also account for the observation on numbers of genera observed after 0.5 megarads of gamma irradiation. Hollaender et al (1951), Moos (1952), and Biagini (1953) found similar results.

Edwards et al (1954) and Morgan (1954) and Reed (1954) found that the presence of proteins, nutrient broth, ascorbic acid and thiourea in a media increased the radio-resistance of bacterial spores. They noted, however, that spores were less susceptible to protective influence of these compounds than vegetative cells.

An attempt was made during the investigation to sensitize the microflora to irradiation. Sodium hypochlorite and hydrogen peroxide added to potato waste water proved to be ineffective in sensitizing the microorganisms. No attempt was made to sensitize the microorganisms with organic peroxides. This could possibly be done in another study.

The results of this investigation show that most of the microorganisms are destroyed after 1.0 megarads. From a commercial standpoint, waste water that has been

irradiated at this level might be acceptable to be recirculated into the processing system. Also if sensitizers could be added to the waste to make the organisms more susceptible to irradiation dosages of less than 1.0 megarads might be used. The sensitizer of course would have to be a compound that would not interfere with any part of a processing system.

A very important consideration here is the possibility of organisms developing radio-resistance as a result of exposure and recirculation. This could influence the commercial value of such a practice and also the level of irradiation that has to be used to assure that radio-resistance does not occur.

The results of the investigation of various levels of total solids on radio-resistance of micro-organisms indicated that increased levels of total solids did not offer more radio-resistance to the organisms than lower levels. Results of this section did show, however, that the inactivation curves of the micro-organisms were not linear. This has been explained by Lawrence and Block (1968). They found survivor curves to have this distinct characteristic whenever samples contained organisms of two distinct levels of heat resistance. Since organisms of two distinct levels of

of radiation resistance were present in the potato waste water this is probably why the survivor curves appear as they do.

SUMMARY

SUMMARY

This study involved the effect of gamma irradiation on the aerobic microflora of potato waste water. The identification of the aerobic microflora of the potato-waste water at various levels of radiation dosages revealed that at dosages of 0.0 megarads the most fundamental organisms identified were micrococci. Organisms of the genus Bacillus also made up a major portion of the total number identified. Other organisms such as Aerobacter, Corynebacterium, Erwinia, Sarcina, Streptomyces, and Xanthomonas made up only minor portions of the microflora.

After dosages of 0.5 and 1.0 megarads Bacillus species became the predominant organism and after dosages of 1.5 and 2.0 megarads they become the only organisms. Most of the microflora was destroyed after dosages of 1.0 megarads.

The study of the rates of destruction of the aerobic microbial flora of potato waste with total solids levels varying from one to 30 percent subjected to gamma irradiation revealed that there was no significant difference in the radiation resistance of organisms at the higher levels of total solids. There appeared to be two distinct rates of destruction of the microflora explained by the fact that the organisms present had

two distinct levels of radiation resistance.

The addition of sodium hypochlorite and hydrogen peroxide separately to the potato waste water proved to be ineffective in sensitizing the aerobic microflora of the waste to gamma irradiation.

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APPENDIX

TABLE 1

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC BACTERIAL FLORA OF POTATO WASTE WATER AT ONE PERCENT TOTAL SOLIDS

TRIAL 1

Irradiation Level (Megarads)	Total Count Organisms/ml
0.0	6.8×10^6
0.5	3.9×10^2
1.0	1.7×10^1
1.5	0.8×10^1
2.0	0

The organisms identified in Trial 1 were isolated from this source.

TABLE 2

ORGANISMS ISOLATED AT 0.0 (MEGARADS)

TRIAL 1

Genus	No.Isolated	Percentage of Total
Aerobacter	4	8
Bacillus	11	22
Cornybacterium	2	4
Erwinia	6	12
Micrococcus	16	32
Mold	1	2
No growth	3	6
Pseudomonas	3	6
Xanthomonas	2	4
Yeast	2	4
Total	50	100

TABLE 3

ORGANISMS ISOLATED AT 0.5 (MEGARADS)

TRIAL 1

Genus	No.Isolated	Percentage of Total
Bacillus	35	70
Erwinia	1	2
Micrococcus	6	12
Molds	4	8
No growth	4	8
Total	50	100

TABLE 4

ORGANISMS ISOLATED AT 1.0 (MEGARADS)

TRIAL 1

Genus	No. Isolated	Percentage of Total
Bacillus	15	88.23
Mold	2	11.77
Total	17	100.00

TABLE 5

ORGANISMS ISOLATED AT 1.5 (MEGARADS)

TRIAL 1

Genus	No.Isolated	Percentage of Total
Bacillus	8	100
Total	8	100

TABLE 6

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 1

Genus	No. Isolated	Percentage of Total
no growth		

TABLE 7

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 2

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	4.9×10^5
0.5	7.3×10^1
1.0	2.3×10^1
1.5	0.8×10^1
2.0	0.8×10^1

The organisms identified in Trial 2 were isolated from this source.

TABLE 8

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 2

Genus	No.Isolated	Percentage of Total
Aerobacter	3	6
Alcaligines	1	2
Bacillus	12	24
Corynebacterium	2	4
Erwinia	3	6
Micrococcus	20	40
Molds	1	2
Proteus	2	4
Pseudomonas	2	4
Staphylococcus	1	2
Xanthomonas	1	2
Yeasts	2	4
Total	50	100

TABLE 9

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 2

Genus	No.Isolated	Percentage of Total
Aerobacter	2	4
Alcaligines	1	2
Bacillus	32	64
Micrococcus	6	12
Molds	6	12
No growth	3	6
Total	50	100

TABLE 10

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 2

Genus	No.Isolated	Percentage of Total
Bacillus	20	86.95
Mold	3	13.05
Total	23	100.00

TABLE 11

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 2

Genus	No.Isolated	Percentage of Total
Bacillus	8	100
Total	8	100

TABLE 12

ORGANISMS ISOLATED AT 2.0 (MEGARADS)

TRIAL 2

Genus	No.Isolated	Percentage of Total
no growth		

TABLE 13

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 3

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	2.2×10^6
0.5	4.1×10^2
1.0	2.9×10^1
1.5	1.3×10^1
2.0	0.6×10^1

The organisms identified in trial 3 were isolated from this source.

TABLE 14

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 3

Genus	No. Isolated	Percentage of Total
Bacillus	16	32
Corynebacterium	2	4
Erwinia	2	4
Micrococcus	17	34
No growth	1	2
Pseudomonas	5	10
Sarcina	1	2
Streptomyces	3	6
Xanthomonas	3	6
Total	50	100

TABLE 15

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 3

Genus	No.Isolated	Percentage of Total
Bacillus	44	88
Micrococcus	2	4
Molds	2	4
No growth	2	4
Total	50	100

TABLE 16

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 3

Genus	No.Isolated	Percentage of Total
Bacillus	26	89.65
Mold	3	10.35
Total	29	100.00

TABLE 17

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 3

Genus	No.Isolated	Percentage of Total
Bacillus	13	100
Total	13	100

TABLE 18

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 3

Genus	No.Isolated	Percentage of Total
Bacillus	6	100
Total	6	100

TABLE 19

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 4

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	4.7×10^5
0.5	6.1×10^2
1.0	1.9×10^1
1.5	1.2×10^1
2.0	0.3×10^1

The organisms identified in Trial 4 were isolated from this source.

TABLE 20

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 4

Genus	No.Isolated	Percentage of Total
Aerobacter	1	2
Alcaligines	1	2
Arthrobacter	2	4
Bacillus	13	26
Escherichia	1	2
Lophomonas	1	2
Micrococcus	20	40
No growth	3	6
Pseudomonas	4	8
Streptomyces	1	2
Xanthomonas	1	2
Yeast	2	4
Total	50	100

TABLE 21

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 4

Genus	No.Isolated	Percentage of Total
Bacillus	43	86
Micrococcus	3	6
Mold	3	6
No growth	1	2
Total	50	100

TABLE 22

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 4

Genus	No.Isolated	Percentage of Total
Bacillus	18	-
Mold	1	-
Total	19	100.00

TABLE 23

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 4

Genus	No.Isolated	Percentage of Total
Bacillus	12	100
Total	12	100

TABLE 24

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 4

Genus	No.Isolated	Percentage of Total
Bacillus	3	100
Total	3	100

TABLE 25

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 5

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	8.8×10^6
0.5	4.3×10^2
1.0	2.1×10^1
1.5	1.8×10^1
2.0	0.2×10^1

The organisms identified in Trial 5 were isolated from this source.

TABLE 26

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 5

Genus	No.Isolated	Percentage of Total
Aerobacter	2	4
Bacillus	10	20
Corynebacterium	1	2
Flavobacterium	2	4
Micrococcus	18	36
Mold	1	2
No growth	1	2
Pseudomonas	3	6
Sarcina	4	8
Staphylococcus	2	4
Streptomyces	2	4
Yeast	4	8
Total	50	100

TABLE 27

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 5

Genus	No. Isolated	Percentage of Total
Bacillus	39	78
Micrococcus	1	2
Mold	4	8
No growth	6	12
Total	50	100

TABLE 28

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 5

Genus	No. Isolated	Percentage of Total
Bacillus	21	100
Total	21	100

TABLE 29

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 5

Genus	No.Isolated	Percentage of total
Bacillus	18	100
Total	18	100

TABLE 30

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 5

Genus	No.Isolated	Percentage of Total
Bacillus	2	100
Total	2	100

TABLE 31

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 6

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	9.1×10^7
0.5	4.8×10^3
1.0	2.2×10^1
1.5	2.0×10^1
2.0	1.6×10^1

The organisms identified in Trial 6 were isolated from this source.

TABLE 32

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 6

Genus	No.Isolated	Percentage of Total
Aerobacter	4	8
Arthrobacter	1	2
Bacillus	5	10
Erwinia	5	10
Lophomonas	2	4
Micrococcus	25	50
No growth	2	4
Proteus	1	2
Sarcina	1	2
Staphylococcus	1	2
Xanthomonas	3	6
Total	50	100

TABLE 33

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 6

Genus	No.Isolated	Percentage of Total
Arthrobacter	2	4
Bacillus	42	84
Micrococcus	2	4
Mold	3	6
No growth	1	2
Total	50	100

TABLE 34

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 6

Genus	No.Isolated	Percentage of Total
Bacillus	20	90.91
Mold	2	9.09
Total	22	100.00

TABLE 35

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 6

Genus	No.Isolated	Percentage of Total
Bacillus	20	100
Total	20	100

TABLE 36

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 6

Genus	No.Isolated	Percentage of Total
Bacillus	16	100
Total	16	100

TABLE 37

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 7

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	8.1×10^6
0.5	4.0×10^3
1.0	2.1×10^1
1.5	1.0×10^1
2.0	0

The organisms identified in Trial 7 were isolated from this source.

TABLE 38

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 7

Genus	No.Isolated	Percentage of Total
Arthrobacter	1	2
Bacillus	10	20
Erwinia	4	8
Lophomonas	1	2
Micrococcus	26	52
Molds	2	4
No growth	1	2
Sarcina	3	6
Staphylococcus	1	2
Yeasts	1	2
Total	50	100

TABLE 39

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 7

Genus	No.Isolated	Percentage of Total
Bacillus	47	94
Micrococcus	1	2
Mold	1	2
No growth	1	2
Total	50	100

TABLE 40

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 7

Genus	No.Isolated	Percentage of Total
Bacillus	17	80.92
Mold	4	19.08
Total	21	100.00

TABLE 41

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 7

Genus	No.Isolated	Percentage of Total
Bacillus	10	100
Total	10	100

TABLE 42

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 7

Genus	No.Isolated	Percentage of Total
No growth		

TABLE 43

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 8

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	9.3×10^7
0.5	6.1×10^2
1.0	2.8×10^1
1.5	2.2×10^1
2.0	0

The organisms identified in Trial 8 were isolated from this source.

TABLE 44

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 8

Genus	No.Isolated	Percentage of Total
Aerobacter	1	2
Arthrobacter	1	2
Bacillus	11	22
Corynebacterium	1	2
Erwinia	1	2
Micrococcus	22	44
No growth	3	6
Pseudomonas	3	6
Streptomyces	3	6
Xanthomonas	4	8
Total	50	100

TABLE 45

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 8

Genus	No.Isolated	Percentage of Total
Bacillus	48	96
No growth	2	4
Total	50	100

TABLE 46

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 8

Genus	No.Isolated	Percentage of Total
Bacillus	26	92.82
Mold	1	3.59
No growth	1	3.59
Total	28	100.00

TABLE 47

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 8

Genus	No.Isolated	Percentage of Total
Bacillus	22	100
Total	22	100

TABLE 48

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 8

Genus	No.Isolated	Percentage of Total
No growth		

TABLE 49

THE EFFECT OF GAMMA IRRADIATION OF THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 9

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	7.8×10^7
0.5	4.9×10^2
1.0	1.7×10^1
1.5	1.2×10^1
2.0	0.1×10^1

The organisms identified in Trial 9 were isolated from this source.

TABLE 50

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 9

Genus	No. Isolated	Percentage of Total
Alcaligines	1	2
Arthrobacter	1	2
Bacillus	12	24
Corynebacterium	1	2
Flavobacterium	1	2
Micrococcus	22	44
Mold	1	2
No growth	1	2
Pseudomonas	2	4
Sarcina	1	2
Streptomyces	3	6
Xanthomonas	1	2
Yeast	3	6
Total	50	100

TABLE 51

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 9

Genus	No.Isolated	Percentage of Total
Bacillus	43	86
Micrococcus	2	4
Mold	3	6
No growth	2	4
Total	50	100

TABLE 52

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 9

Genus	No.Isolated	Percentage of Total
Bacillus	19	95
Mold	1	5
Total	20	100

TABLE 53

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 9

Genus	No.Isolated	Percentage of Total
Bacillus	12	100
Total	12	100

TABLE 54

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 9

Genus	No.Isolated	Percentage of Total
Bacillus	1	100
Total	1	100

TABLE 55

THE EFFECT OF GAMMA IRRADIATION OF THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

TRIAL 10

Irradiation Level (Megarads)	Total Count (organisms/ml)
0.0	7.3×10^6
0.5	4.8×10^3
1.0	1.5×10^1
1.5	1.1×10^1
2.0	0

The organisms identified in Trial 10 were isolated from this source.

TABLE 56

ORGANISMS ISOLATED AT 0.0 MEGARADS

TRIAL 10

Genus	No. Isolated	Percentage of Total
Aerobacter	1	2
Arthrobacter	1	2
Bacillus	22	44
Corynebacterium	1	2
Erwinia	1	2
Flavobacterium	1	2
Micrococcus	8	16
Mold	2	4
No growth	1	2
Pseudomonas	6	12
Streptomyces	4	8
Xanthomonas	1	2
Yeast	1	2
Total	50	100

TABLE 57

ORGANISMS ISOLATED AT 0.5 MEGARADS

TRIAL 10

Genus	No.Isolated	Percentage of Total
Bacillus	36	72
Micrococcus	2	4
Mold	11	22
Streptomyces	1	2
Total	50	100

TABLE 58

ORGANISMS ISOLATED AT 1.0 MEGARADS

TRIAL 10

Genus	No.Isolated	Percentage of Total
Bacillus	14	93.24
Mold	1	6.76
Total	15	100.00

TABLE 59

ORGANISMS ISOLATED AT 1.5 MEGARADS

TRIAL 10

Genus	No. Isolated	Percentage of Total
Bacillus	11	100
Total	11	100

TABLE 60

ORGANISMS ISOLATED AT 2.0 MEGARADS

TRIAL 10

No growth

TABLE 61

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING ONE PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML				
TRIAL	0.0	0.5	1.0	1.5	2.0
1	2.0×10^5	3.90×10^3	4.5×10^1	1.6×10^1	0
2	7.0×10^5	4.20×10^3	6.0×10^1	0.4×10^1	0
3	2.4×10^5	0.46×10^3	4.2×10^1	2.0×10^1	1.0×10^1
4	6.0×10^5	0.41×10^3	2.9×10^1	0.4×10^1	0
5	7.1×10^5	0.44×10^3	1.8×10^1	1.2×10^1	0.2×10^1
6	3.9×10^5	0.51×10^3	1.6×10^1	1.2×10^1	0
7	6.7×10^5	0.50×10^3	2.1×10^1	0.2×10^1	0
8	5.9×10^5	0.41×10^3	0.4×10^1	0.4×10^1	0
9	7.7×10^5	0.46×10^3	2.1×10^1	1.7×10^1	0.8×10^1
10	6.4×10^5	0.49×10^3	0.4×10^1	0.1×10^1	0.1×10^1
Average	5.51×10^5	1.178×10^3	2.60×10^1	0.92×10^1	0.21×10^1

TABLE 62

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING 5 PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML				
Trial	0.0	0.5	1.0	1.5	0.0
1.	1.9×10^5	3.8×10^3	4.5×10^1	1.6×10^1	0
2.	6.9×10^5	4.0×10^3	6.0×10^1	0.2×10^1	0
3.	22.0×10^5	4.6×10^3	3.8×10^1	1.6×10^1	1.2×10^1
4.	5.9×10^5	0.37×10^3	2.9×10^1	1.1×10^1	0
5.	7.0×10^5	0.35×10^3	2.2×10^1	0.2×10^1	0
6.	3.7×10^5	4.7×10^3	2.1×10^1	1.8×10^1	0
7.	6.1×10^5	0.50×10^3	2.6×10^1	1.2×10^1	0
8.	5.6×10^5	0.39×10^3	1.6×10^1	1.3×10^1	0
9.	7.2×10^5	0.49×10^3	2.2×10^1	1.2×10^1	0.9×10^1
10.	6.7×10^5	0.56×10^3	1.9×10^1	1.4×10^1	0
Average	7.30×10^5	1.98×10^3	2.98×10^1	1.16×10^1	0.21×10^1

TABLE 63

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING 3 PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML									
Trial	0.0		0.5		1.0		1.5		2.0	
1	8.1	x 10^5	3.3	x 10^2	6.6	x 10^1	2.2	x 10^1	0	
2	4.8	x 10^5	3.6	x 10^2	5.3	x 10^1	2.2	x 10^1	0	
3	5.9	x 10^5	3.7	x 10^2	4.2	x 10^1	1.5	x 10^1	1.2	x 10^1
4	8.7	x 10^5	4.9	x 10^2	6.6	x 10^1	2.6	x 10^1	0.3	x 10^1
5	5.7	x 10^5	5.6	x 10^2	4.2	x 10^1	1.9	x 10^1	1.1	x 10^1
6	5.9	x 10^5	7.1	x 10^2	2.7	x 10^1	1.4	x 10^1	1.0	x 10^1
7	7.6	x 10^5	3.9	x 10^2	2.7	x 10^1	1.9	x 10^1	0	
8	3.4	x 10^5	5.0	x 10^2	4.2	x 10^1	0		0	
9	4.9	x 10^5	3.6	x 10^2	1.9	x 10^1	1.4	x 10^1	0	
10	1.8	x 10^5	3.4	x 10^2	2.9	x 10^1	1.7	x 10^1	0.2	x 10^1
Average	5.68	x 10^5	4.41	x 10^2	4.13	x 10^1	1.68	x 10^1	0.38	x 10^1

TABLE 64

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING 5 PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML				
Trial	0.0	0.5	1.0	1.5	2.0
1	1.09×10^5	5.6×10^2	1.6×10^1	0	0
2	1.65×10^5	4.0×10^2	1.7×10^1	0	0
3	1.25×10^5	9.2×10^2	2.1×10^1	1.1×10^1	0
4	14.1×10^5	32.0×10^2	1.6×10^1	1.4×10^1	0
5	15.2×10^5	1.8×10^2	2.9×10^1	2.1×10^1	0
6	14.6×10^5	2.2×10^2	2.9×10^1	0	0
7	3.9×10^5	3.4×10^2	2.1×10^1	0	0
8	1.2×10^5	3.9×10^2	2.1×10^1	1.1×10^1	0.9×10^1
9	3.7×10^5	4.2×10^2	2.7×10^1	2.1×10^1	1.3×10^1
10	0.61×10^5	0.68×10^2	0.7×10^1	0.1×10^1	0
Average	5.73×10^5	6.20×10^2	2.04×10^1	0.79×10^1	0.22×10^1

TABLE 65

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING 10 PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML				
Trial	0.0	0.5	1.0	1.5	2.0
1	1.25×10^5	1.2×10^2	4.2×10^1	1.6×10^1	0.9×10^1
2	1.04×10^5	2.8×10^2	3.9×10^1	2.1×10^1	1.6×10^1
3	0.98×10^5	4.2×10^2	3.5×10^1	2.1×10^1	0.3×10^1
4	3.4×10^5	5.0×10^2	4.2×10^1	2.0×10^1	0.7×10^1
5	4.9×10^5	3.6×10^2	2.7×10^1	1.9×10^1	1.2×10^1
6	0.42×10^5	0.70×10^2	2.9×10^1	2.0×10^1	1.6×10^1
7	0.58×10^5	0.65×10^2	2.1×10^1	1.0×10^1	0.2×10^1
8	0.72×10^5	0.76×10^2	2.3×10^1	0.3×10^1	0.1×10^1
9	0.62×10^5	0.78×10^2	1.6×10^1	0.9×10^1	0.2×10^1
10	0.64×10^5	0.61×10^2	1.1×10^1	0.9×10^1	0
Average	1.46×10^5	2.03×10^2	2.85×10^1	1.48×10^1	0.68×10^1

TABLE 66

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING 15 PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML				
Trial	0.0	0.5	1.0	1.5	2.0
1	18.0×10^6	4.9×10^2	1.6×10^1	1.4×10^1	0
2	9.7×10^6	1.25×10^2	1.7×10^1	1.3×10^1	0.2×10^1
3	6.9×10^6	1.84×10^2	2.1×10^1	0	0
4	1.55×10^6	4.6×10^2	1.6×10^1	1.2×10^1	0.1×10^1
5	1.68×10^6	5.1×10^2	2.4×10^1	2.6×10^1	0
6	0.99×10^6	6.6×10^2	1.7×10^1	0.3×10^1	0
7	1.21×10^6	6.7×10^2	2.9×10^1	1.6×10^1	0
8	1.87×10^6	8.4×10^2	1.9×10^1	1.3×10^1	0.9×10^1
9	0.56×10^6	4.8×10^2	2.2×10^1	1.1×10^1	0
10	1.89×10^6	8.1×10^2	1.6×10^1	1.2×10^1	0
Average	4.44×10^6	5.23×10^2	1.97×10^1	1.00×10^1	0.12×10^1

TABLE 67

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING 20 PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML				
Trial	0.0	0.5	1.0	1.5	2.0
1	180.00×10^6	12.4×10^2	2.1×10^1	1.6×10^1	1.2×10^1
2	0.58×10^6	0.4×10^2	2.1×10^1	0.6×10^1	0
3	220.00×10^6	50.0×10^2	3.8×10^1	0	0
4	1.21×10^6	1.4×10^2	1.4×10^1	0.1×10^1	0
5	1.7×10^6	3.9×10^2	2.0×10^1	1.3×10^1	0
6	5.8×10^6	4.2×10^2	1.2×10^1	1.1×10^1	0.6×10^1
7	1.2×10^6	0.61×10^2	1.6×10^1	1.3×10^1	0
8	1.8×10^6	1.21×10^2	1.4×10^1	1.1×10^1	0.9×10^1
9	0.78×10^6	0.63×10^2	2.1×10^1	1.6×10^1	0
10	0.93×10^6	1.1×10^2	1.6×10^1	1.6×10^1	0.3×10^1
Average	41.4×10^6	7.59×10^2	1.93×10^1	1.03×10^1	0.30×10^1

TABLE 68

THE EFFECT OF GAMMA IRRADIATION ON THE AEROBIC FLORA OF POTATO WASTE WATER CONTAINING 30 PERCENT TOTAL SOLIDS

	IRRADIATION LEVEL (MEGARADS) NUMBER OF ORGANISMS/ML				
Trial	0.0	0.5	1.0	1.5	2.0
1	1.24×10^5	8.9×10^2	3.9×10^1	2.0×10^1	1.6×10^1
2	3.9×10^5	9.9×10^2	4.6×10^1	1.6×10^1	1.3×10^1
3	0.61×10^5	4.8×10^2	2.9×10^1	2.1×10^1	0
4	1.6×10^5	8.4×10^2	2.6×10^1	0.4×10^1	0
5	9.4×10^5	1.25×10^2	1.9×10^1	1.4×10^1	0.4×10^1
6	9.6×10^5	5.1×10^2	2.2×10^1	1.9×10^1	0.3×10^1
7	1.92×10^5	7.2×10^2	6.7×10^1	2.2×10^1	0.1×10^1
8	7.8×10^5	8.8×10^2	1.6×10^1	0.3×10^1	0
9	12.5×10^5	4.3×10^2	2.2×10^1	0.4×10^1	0.4×10^1
10	6.9×10^5	1.27×10^2	1.7×10^1	0	0
Average	5.54×10^5	5.99×10^2	3.03×10^1	1.23×10^1	0.41×10^1

TABLE 69

THE EFFECT OF TOTAL SOLID VARIATION ON THE IRRADIATION RESISTANCE OF THE AEROBIC MICROBIAL FLORA OF POTATO WASTE WATER

Source of Variation	DF	SS	MS	F	SE Diff.
Replications	9	0.636	0.071	2.045	0.038
A	7	0.407	0.058	25.684*	0.027
B	3	2.193	0.731	1.923	0.075
AB	21	1.149	0.055		
Error	279	7.941	0.028		
Total	319	12.327			

Standard error 0.169
Coefficient of variation 0.169

* F values significant

A - total solid levels

B - irradiation levels