March 10, 1964

To the Graduate Council:

I am submitting herewith a thesis written by J. C. Webb entitled "Response of Cottonseed to Audio Frequency Gas-Plasma." I recommend that it be accepted for nine quarter hours of credit in partial fulfillment of the requirements for the degree of Master of Science, with a major in Agricultural Engineering.

John J. McDow
Major Professor

We have read this thesis and recommend its acceptance:

R. B. Stone

Smith Worley, Jr.

Zachary A Henry

Accepted for the Council:

Hilton A. Smith
Dean of the Graduate School

RESPONSE OF COTTONSEED TO AUDIO FREQUENCY GAS-PLASMA

A Thesis

Presented to

the Graduate Council of

The University of Tennessee

In Partial Fulfillment

of the Requirements for the Degree

Master of Science

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by

J. C. Webb

March 1964

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TABLE OF CONTENTS

CHAPTER		PAGE
I.	STATEMENT OF THE PROBLEM	1
	Objectives	3
II.	REVIEW OF THE LITERATURE	4
	Radio Frequencies	4
	Infrared	7
	Visible Light	8
	X-Rays and Gamma Rays	8
	The Glow Discharge	10
III.	THE GLOW DISCHARGE	14
	Types of Discharges	14
	Principles of the Glow Discharge	15
IV.	THE IRRADIATION SYSTEM	22
	The Electrical System	22
	The Irradiation Chamber	25
	The Vacuum System	25
V.	EXPERIMENTAL PROCEDURES	29
	Experimental Design	29
	Treatment of the Experiment Material	29
	System's Characteristics During Test	30
	Germination Procedures	34
	Collection of Data	34
VI.	PRESENTATION AND INTERPRETATION OF THE EXPERIMENTAL DATA	39
	Effects of Irradiation on Germination	39
	Effects of Irradiation on Radicle Development	45

CHAPTER PAGE

Interpretations 51

Suggestions for Future Investigation 56

VII. SUMMARY AND CONCLUSIONS. 58

BIBLIOGRAPHY. 60

APPENDIX 65

APPENDIX A. Definition of Terms 66

LIST OF TABLES

TABLE		PAGE
I.	System Response for Various Currents and Frequencies.	32
II.	Mean Per Cent Germination of Two Day, Four Day, and Abnormal Seedlings for All Treatments and Controls.	40
III.	Mean Per Cent Germination and Radicle Development at Each Frequency Treatment	44
IV.	Mean Per Cent Germination and Radicle Development at Each Current Level.	47
V.	Mean Per Cent Germination After Two and Four Days for Various Currents and Frequencies	48
VI.	Mean Radicle Length, Weight, and Per Cent of Radicles Over One Inch for All Treatments and Controls.	49
VII.	Factorial Analysis of Germination and Radicle Development for All Levels of Current and Frequency.	50

LIST OF FIGURES

FIGURES PAGE

1. Electromagnetic Spectrum Showing Approximate Ranges of the Radiation Based on Figures of International Standards. 5
2. Characteristics of Low Pressure Discharges Showing the Townsend, the Glow, and the Arc Discharge . . 16
3. Details of the Glow Discharge and the Relative Voltage Gradient. 18
4. The Complete Irradiation System 23
5. Block Diagram of Irradiation System. 24
6. The Voltage and Frequency Generating Equipment. . . 26
7. The Electrodes, Needle Valve, and Irradiation Chamber.. 27
8. Gain of the System at Various Frequencies and Currents. 33
9. A Comparison of the Control and the Treated Seed Germination at Two Days. 35
10. A Comparison of the Control and the Treated Seed Germination at Four Days. 37
11. A Contrast of a Normal and An Abnormal Radicle . . . 38
12. Early Germination Verses Frequency at Each Current Level. 41
13. Early and Total Germination as a Function of Frequency. 43
14. Early and Total Germination as a Function of Current. 46
15. Radicle Length and Per Cent of Radicles Over One Inch as a Function of Frequency. 52

FIGURE PAGE

16. Radicle Length and Per Cent of Radicles Over One Inch as a Function of Current. 53

CHAPTER I

STATEMENT OF THE PROBLEM

Man has endeavored to discover the effects of electric energy on living organisms since the discovery of electricity. The advent of each major discovery in the field of electricity has immediately been followed by an intensive effort to apply the new knowledge to the science of biology and to induce physical changes in the characteristics of plants.

There are many ways to apply electric energy to living organisms. One of the simplest methods is to make the organism a part of an electric circuit and pass a current directly through it by applying a potential. A method which was used to apply electric energy to seed was to place the seed in an electric or magnetic field established at radio frequencies. This method subjects the seeds to a strong electric field with a weak magnetic field or to a strong magnetic field with a weak electric field. Another method often used by researchers is the electric discharge. This method subjects the seeds to both high electric and magnetic fields, but not at the same time. Field seed may be subjected to this kind of electromagnetic radiation when a lighting discharge between a cloud and the earth strikes in a field.

Many researchers have reported on these and other methods of treating seed with electromagnetic radiation. Often the results were conflicting and experiments very difficult to repeat. This variation in results was due in part to the wide variation in biological materials. However, a large percentage of this variation results from the lack of control of the electric parameters to which the materials were exposed.

The glow discharge or gas-plasma treatment, which has been used in recent years, is another method of exposing biological materials to

electromagnetic radiation. The principal difference between this method and methods commonly used is that the material is treated at pressures less than atmospheric. This method having the advantage of the electric parameters under the control of the experimenter; therefore, he can control both the current and the electric field intensity. The glow discharge is adaptable to both low and high frequencies; however, most of the research has been done at low frequencies. The parameter combinations of this method are almost unlimited. This field of research is relatively new and it offers great possibilities for future researchers.

In recent years, the glow discharge plasma has been used to irradiate cottonseeds, soybeans, and other field seeds. Some of these treatments have been reported to have a stimulating effect on the seed, thereby, causing earlier germination and earlier radicle development. The above research with the glow discharge was conducted with the time of exposure, pressure, and current as variables and with the excitation frequency held constant. The purpose of the study, as described in this thesis, was to determine the effects of frequency and current on early and total germination and radicle development of cottonseed.

The necessity for obtaining prompt and uniform germination of field seeds after planting to obtain desirable crop yields is generally recognized by farmers. The cost of replanting cottonseed because of poor stands and the cost resulting from overplanting of seed to insure a uniform stand are high operating expenses for the farmer. Thus, any economical method of seed treatment that will aid in earlier and improved germination of field seed will be of general value to the growers of these crops.

I. OBJECTIVES

The objectives of the study were:

1. To determine the effects of radiations from gas-plasma on the early and total germination of machine delinted cottonseed.
2. To determine the effects of radiations from gas-plasma on the radicle development of machine delinted cottonseed.
3. To determine what effects the different excitation frequencies have on early and total germination and on radicle development of machine delinted cottonseed.
4. To determine what effects varying the current intensity have on early and total germination and on radicle development of machine delinted cottonseed.

CHAPTER II

REVIEW OF THE LITERATURE

The effects of electromagnetic radiations on seeds and plant materials have long been the interest of many researchers. The electromagnetic spectrum showing the approximate ranges of radiations from and including radio waves through gamma rays is shown in Figure 1 (24).

I. RADIO FREQUENCIES

Much of the early research in the radiation field was done at low frequencies using a charged network over growing plants to change the electrical state of the atmosphere surrounding the plants. The term used for this type of work was called electroculture (6). Too frequently, the experiments were done by biologists with limited knowledge of the electrical phenomena with which they were dealing, or by physicists or electrical engineers with too little understanding of basic plant physiology (3). In view of the conflicting reports on electroculture experiments, Briggs, et al. (6) of the U. S. Department of Agriculture conducted an extensive study of the problem. This research, which started in the fall of 1907 and ended in 1918, included both field and laboratory experiments. The potential on the charged networks ranged from 30, 000 to 60, 000 volts and the atmospheric current ranged from 0.1 to 1 milliampere per acre. After this extensive study, no significant effects on plant growth were found using electroculture (6).

In recent years studies have been made of the effects of radiofrequency, infrared, visible light, ultraviolet, x-rays, cathode rays, gamma rays, and glow discharge radiation on seed germination and plant growth. In 1950 and 1951 experiments were conducted by Findley

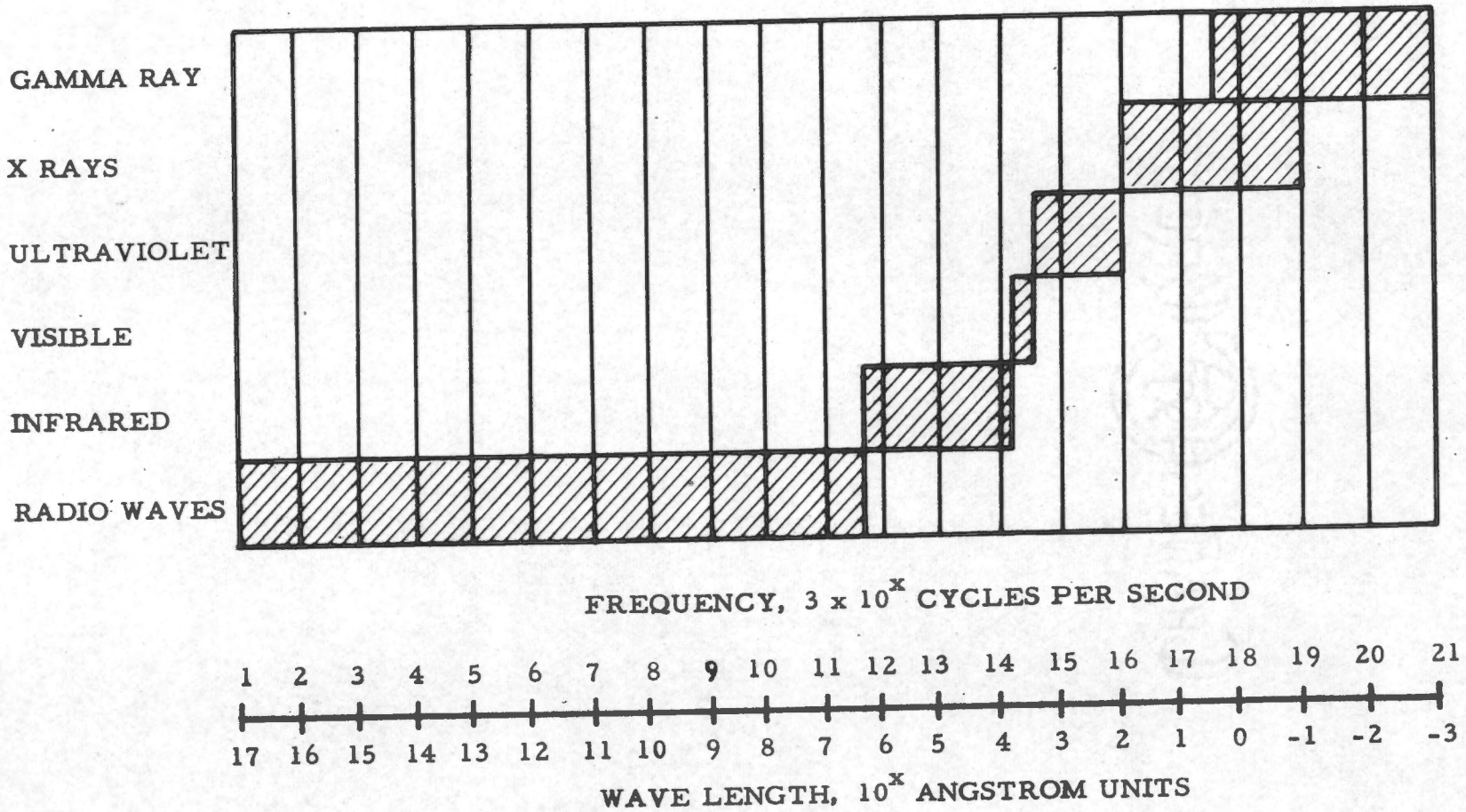


Figure 1. Electromagnetic spectrum showing approximate ranges of the radiation based on figures of international standards.

and Campbell (15) to determine the effects of ultrasonic energy treatments on corn yields. The ultrasonic radiation was produced by a radio frequency generator of the piezoelectric type operating at 400 kilocycles and driving a quartz crystal transducer at the same frequency. The transducer produced ultrasonic radiation in an oil bath, which served to transfer the energy to the treatment chamber. The electric energy prior to conversion to sound energy was approximately 600 watts. Increase in yields were significant in three of the treatments in 1950, but failure to duplicate these results in 1951 suggested to the authors that the differences were due to factors other than the treatment effects.

Metabolic functions which were stimulated by ultrasonic irradiation were reported by See (32) to have increased seed germination, growth, yield of seeds, yield of proteins, carbohydrates, and fats in some seeds. Irradiated grain produced an increase in enzyme activity, particularly in carbohydrates. Stimulation was reported for intensities below five watts/cm^2, but higher levels of irradiation caused inhibition and damage to the seed.

The radiation from radio waves has been used to treat a number of seeds in the past few years. An investigation of low frequency radio waves was conducted by Ginsburg and Cholet (16) on the germination of corn. Exposures from two to seven hours at voltages of 1, 200 to 20, 000 volts per centimeter were used in the experiment. The frequency ranged from 500 cycles per second to 220 kilocycles per second. The results of the experiment showed that there were no significant differences between any of the treatments and the controls. Corn, which was exposed at forty megacycles for five seconds by Nelson and Walker (26), showed a significant increase in germination over the control at two days.

A great deal of research has been done to reduce the hard seed in alfalfa by means of radio-frequency irradiation. Several varieties of

alfalfa were irradiated by Nelson and Wolf (29) at a frequency of thirty-nine megacycles per second and at a field intensity from 2.5 to 5.3 kilovolts per inch. The moisture content of the alfalfa varied from 2.1 to 15.6 per cent. There was an optimum treatment level determined for each variety. The results from these optimum treatments showed a significant decrease in the number of hard seed with a corresponding increase in early germination when compared to the control seed. There was a definite relationship between the field intensity and the moisture content of the seed. The seed at a lower moisture content could be exposed to a higher field intensity without damage, than the seed at high moisture content. This was caused by the higher rate of heat conduction in the high moisture seed. Nelson verified these results in later research (24, 25, 26).

II. INFRARED

Infrared heat treatments were used by Rincher (29) to reduce the number of hard seed in alfalfa, sweetclover, and red clover. The infrared irradiation was applied by direct heat rays from a 110 volt, 250-watt, infrared bulb at a temperature of 220°F. The findings were that the hard seed of alfalfa and red clover could be made permeable by applications of properly regulated dry heat. The hard seed of sweet clover did not respond the same as hard seed of alfalfa and red clover to identical heat treatments.

Seeds of carrots, onions, beets, lettuce, and tomatoes were irradiated with radio frequencies and infrared radiation by Jonas (19). The rate of germination was increased by the radio-frequency treatments and was dependent on the voltage gradient, power and energy input, and the seed temperature. The infrared radiation produced smaller increases in the rate of germination.

III. VISIBLE LIGHT

Low energies of light in the red and far-red regions of the spectrum exert a profound influence on the development of plants (9). The germination and dormancy of many small seeds are affected by such light. All photoperiodic effects such as: the flowering of long-day and short-day plants; and the dormancy of woody plants are controlled by a pigment system which absorbs energy at these wavelengths (9). Lettuce seeds were exposed to red and far-red light waves by Bulter and Norris (9). The seed germination could be alternately stimulated and inhibited depending on whether they were last exposed to red or far-red light. The stimulating effect caused by the red light was counteracted by radiation from the far-red light. The maximum stimulating effect of red light occurs at about 6600 A., and the maximum inhibiting effect of far-red radiation occurs at approximately 7300 A (14). Exposure to light for three to four days during germination, plus the oxidation brought about by using 0.75 per cent sodium hypochlorite, completely broke the dormancy of guayule seed so that freshly harvested achenes gave maximum germination equal to that of seed after eighty-four days in storage (40).

IV. X-RAYS AND GAMMA RAYS

The effects of ionizing radiation on seeds and plants have been studied by many researchers in the past several years. Studies were made in relation to seed germination, plant growth, mutations, lethal doses, chromosome breakdown, and other genetic properties. The studies on seed germination and plant growth have not been as fruitful as some of the other studies. Some increases in germination have been reported when the seeds were exposed to low level treatments, but as the dosages were increased the germination decreased very rapidly.

The effects of x-rays on the germination and growth of seeds have been the subject of much investigation. Most investigators agree that x-rays can cause mutations and that germination and growth decrease as the level of irradiation increases. Cottonseeds were exposed to x-rays and cathode rays by Lambou, et al. (22) to determine the effects of these rays on seed germination. The germination was partially inhibited by the lowest dosage and completely inhibited by the highest dosage. The level of irradiation ranged from 500, 000 up to 2, 000, 000 rep. In another experiment by Bless (5) radish, lettuce, bean, and corn seed were irradiated with x-ray at a voltage of 100, 000 volts and five milliamperes. Bless reported that he found no significant differences in germination between any of the treated seed over the control. Wheat seeds were exposed to soft x-rays by Benedict and Kersten (4). Results of wheat seedlings irradiated for five seconds showed an increase both in diastatic activity and in sugar content, but if irradiated for a longer time there was a divided and progressive decrease in these two substances. The irradiated seeds did not germinate as well as the controls, but those that did germinate grew about two centimeters in length and died. The results indicated that, under the conditions of the experiment, irradiated seeds could not change their stored starch into sugar as readily as did the control seed. Two varieties of soybeans were irradiated with x-rays by Mitrovic, Suput, and Mihajlovic (23) to determine the effects on yield. One variety showed a small increase in yield at low levels of irradiation while the other variety showed a decrease in yield. Three types of annual seeds were exposed to gamma radiation at levels from 109R to 1344R by Elnadl and Sirry (13). The lowest level of irradiation of all three seed types gave a slight increase in germination. but as the treatment level increased the germination decreased. Some seeds are more sensitive to ionizing radiation than others. In an attempt to explain some

of this difference in sensitivity, Hoskinson and Osborne (17) investigated preirradiation soaking and storage of barley seed treated with gamma rays. The results indicated, that with increased time of soaking, radiosensitivity either did not change or it increased. With increasing time of storage, there was an increase followed by a decrease in sensitivity.

V. THE GLOW DISCHARGE

The radiation from the glow discharge cannot be divided into specific portions of the electromagnetic spectrum as was the case in the above types of radiations. The glow discharge is made up of frequencies from several portions of the spectrum which includes radio waves, infrared, visible, and ultraviolet. While the glow discharge process is not new, very little work has been done on seed and plants with it. However, the petroleum industry has used the process rather extensively.

The glow discharge process of thickening or polymerizing lubricants has been known for some time. The process was patented by Alexander de Hemptinne in 1909 and a commercial operation was started in 1910 (44). This process was known by several names and included elektrion, voltal, and voltolization. The products from this process were used as high quality aviation and automotive lubricants. Since this early work, extensive research has been done on both liquids and gases. The energy required for voltolization is greatly influenced by the gas that is present in the glow discharge (44, 38). Certain gases such as chlorine or oxygen react with the product undergoing voltolization, and their effect as ionic media is therefore difficult to determine.

It was not until about 1954 that research was started on irradiating seeds and plant materials with the glow discharge. The methods and equipment for irradiating seed in a glow discharge were first used by

Brown, Stone, and Andrews (8). This equipment was designed to establish a glow discharge plasma in gases at low pressures using a frequency of sixty cycles per second as the excitation frequency. However, with some modification other frequencies could be used if desired. Several types of seeds were irradiated and the effects on some seeds were quite pronounced. One common effect noticed on most seed was that the rate of water uptake increased. Corn which was exposed to a limited amount of radiation germinated faster and more uniformly in a petri dish of free water than did the control. In all cases the seeds were killed if the radiation was made sufficiently intense.

Empire W. R. cottonseeds were exposed to the glow discharge plasma, using sixty cycles per second as the excitation frequency, at five levels of radiation and at a pressure of three millimeters of mercury for three minutes (43). Two day and four day germination counts were made and the radicles were measured after four days. There was a significant increase in the two day germination at all radiation levels over the control seed. There was also a significant increase in the radicle development of the irradiated seed over the control seed. However, the total germination of the highest intensity level of irradiation was significantly lower than the controls. This treatment was sufficiently intense to kill most of the seed.

Seeds of three alfalfa varieties containing high percentages of hard seed were exposed to five levels of infrared, radiofrequency, and gas-plasma treatments (25). All three types of treatments were about equally effective in reducing the hard-seed content and in increasing early germination and emergence. Benefits of all treatments remained after fourteen months in storage. Seedlings from all except overexposed seed samples appeared normal. Water sorption, leachate solution conductivity, and oxygen uptake of seeds were increased about the same amount by the three types of treatments, and were closely related to the degree

of hard seed reduction.

Gas-plasma irradiated milled rice showed a marked increase in the amount of water that could be absorbed (30). A study of the effects of the major operating variables revealed: (1) with the time and pressure constant at five minutes and two millimeters of mercury respectively, maximum changes in hydration characteristics occurred at about 175 milliamperes for Zenith variety and 150 milliamperes for Bluebonnet 50 rice; (2) pressure during treatment had no great effect upon changing the water absorptive capacity of the rice within a pressure range of two to eight millimeters of mercury with a current range of twenty-five to seventy-five milliamperes for five minutes; and (3) any increase in treatment time over forty-five minutes for Bluebonnet variety and seventy minutes for Zenith was inefficient in increasing the amount of water absorption. Rice bran and brown rice were treated with gas-plasma irradiation to determine its effects on oil stability and on certain other physical properties of the petroleum ether solubles by Roseman, et al. (31). The rate of free fatty acid development in the lipids of stored brown rice and in the stored bran separated from rice kernels was much slower in gas-plasma irradiated samples than in controls. Gas-plasma irradiation profoundly changed the chemical and physical characteristics of rice oil from both stored brown rice and stored separated rice bran. The average molecular weight of the oil extracted from irradiated rice bran had increased and the oil was less saturated than that of the controls. Treatments using only heat and vacuum were investigated. These investigations revealed that these changes are affected specifically by the gas-plasma irradiation rather than by the coincidental heat-vacuum effect of irradiation.

Cotton fibers and cottonseeds were irradiated with the plasma of a glow discharge which was established by a sixty cycle power source by Stone (36). The irradiated cotton fibers were rough and stiff and absorbed water very rapidly. Electron micrographs showed broken

surface areas on the cotton fibers and also evidence of primary wall cell damage. Analysis of the wax on the cotton fibers indicated the wax had been degraded as a result of irradiation. The linters on the irradiated fuzzy and machine delinted cottonseed became very water absorbant and when dropped into a container of water, the cottonseed sank to the bottom immediately. The control seed still floated on the surface of the water twenty-four hours after placement. The acid delinted cottonseed following glow discharge irradiation absorbed water over the entire surface of the seed more rapidly than did the control seed. Cotton yarn exposed to gas-plasma irradiation showed an increase in strength of 31 per cent for yarn spun at optimum twist (37). At the lowest twist tested, the irradiated yarn was 75 per cent stronger than the control. The breaking strengths of the irradiated yarns at low twists were greater than the strength of the control at optimum twist.

CHAPTER III

THE GLOW DISCHARGE

I. TYPES OF DISCHARGES

Electrical discharges can be divided into two broad classifications: (a) non-disruptive or silent discharges; and (b) disruptive discharges. The non-disruptive or silent discharges can be further subdivided into the ozonizer, corona, electrodeless, and glow discharge. The disruptive discharge will not be discussed in this thesis.

The ozonizer discharge is established in a container with two electrodes, both separated from the reaction space by a glass wall. The container is ordinarily constructed from glass tubing; however, other forms of containers have been used. Voltages from about five to twenty-five kilovolts are used to operate the ozonizer (38). Discharges are established satisfactorily at fifty cycles per second and up, at either high or low pressures.

A corona discharge occurs when an electric field, around a point or wire, becomes sufficiently high for break down before a spark can propagate across the gas space. The corona discharge occurs at any frequency, including direct current. The voltage that is required to establish this discharge is determined by the physical character of the apparatus (38).

The electrodeless discharge takes place inside a container at low pressures. This discharge resembles the glow discharge; however it will occur only at high frequencies. Then a gas of sufficiently low pressure is subjected to a high frequency electromagnetic field of sufficient intensity, this discharge will be established.

The glow discharge occurs when a sufficient potential is applied between two electrodes at a low pressure. The discharge can be

established at direct current or at high frequencies. The physical aspects of this discharge have been studied and some of the characteristics of the discharge are known under a given set of conditions. Since the glow discharge is of prime importance to the research reported in this thesis, only the principles of this discharge will be discussed in detail.

II. PRINCIPLES OF THE GLOW DISCHARGE

The glow discharge has been studied using known pure gases and direct currents. Some of the findings from these studies will be discussed in this chapter. The discharge takes place between two electrodes in a container at a pressure from approximately 0.01 to 20 millimeters of mercury (44). The nature of the discharge is affected by many factors. Some of these factors are applied voltage, frequency, configuration of the discharge chamber and electrodes, type and pressure of the gas, electrode material, and external circuit conditions.

An idealized discharge characteristics curve is presented in Figure 2. Any one or a combination of the above factors could have a marked effect on any portion of the characteristics curve. The portion of the curve from point A to point B is known as the Townsend discharge. The current in this discharge can be increased only with an increase in the applied voltage. The portion of the curve from point B to point C is the glow discharge, and beyond point C is the arc discharge portion of the curve. The transition from the Townsend to the glow discharge is usually accompanied by a voltage drop, as shown in Figure 2. The Townsend discharge and the glow discharge can take place under the same conditions as shown in the shaded portion of Figure 2 at point B. The transition from the glow discharge to the arc is quite uncertain but takes place at some point beyond point C (10).

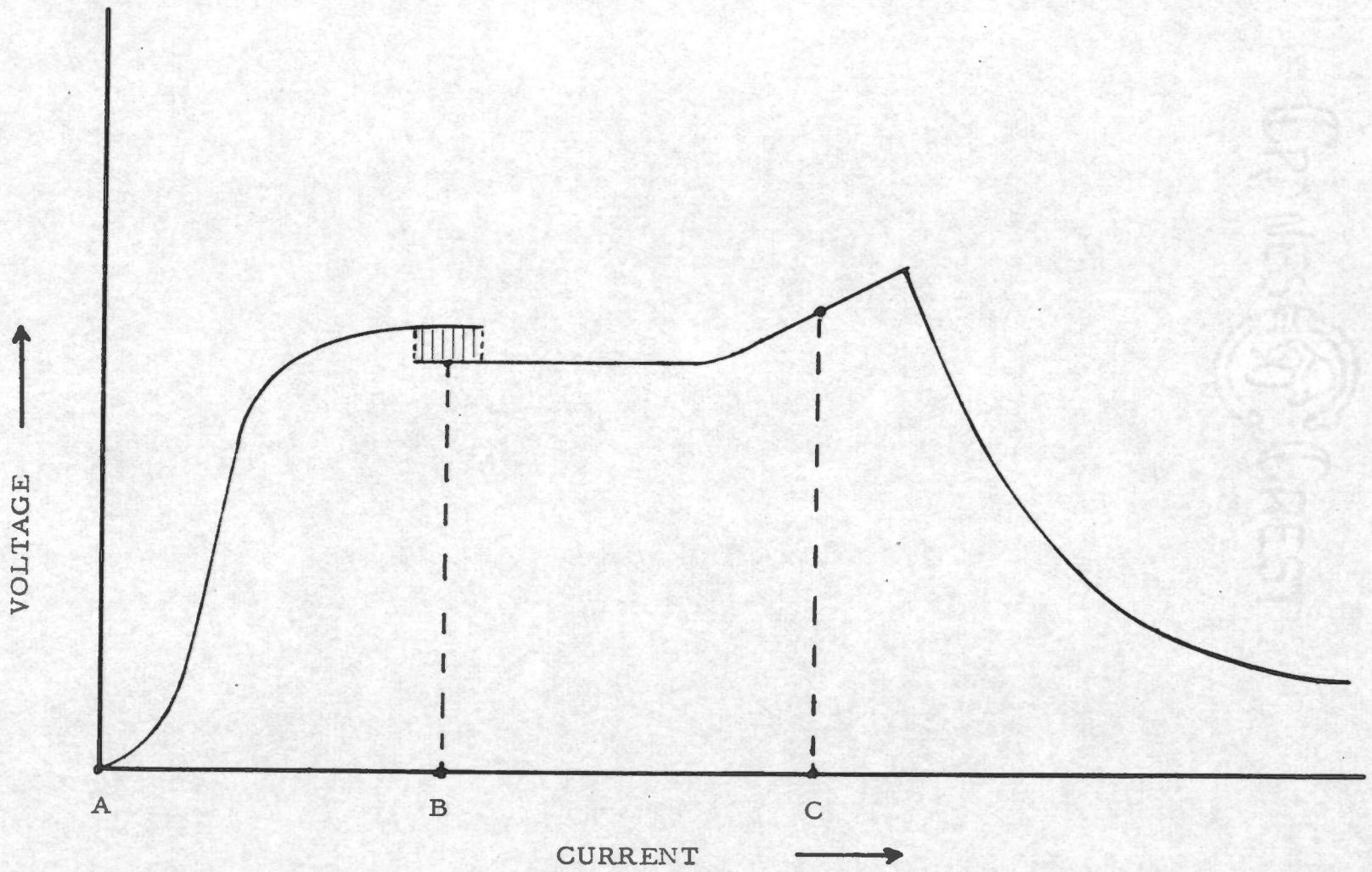


Figure 2. Characteristics of low pressure discharges showing the Townsend, the Glow, and the Arc Discharge.

When the pressure in the discharge chamber is reduced to approximately a millimeter of mercury, depending on the type of gas in the chamber, the discharge consists of alternate dark and light regions. A diagram of the relative position of these regions is shown in Figure 3. Starting with the cathode the regions are as follows: (1) the Aston dark space; (2) the cathode glow, whose length depends on the gas and the gas pressure, in many cases completely masks the Aston dark space; (3) the cathode dark space or sometimes called the Crookes or the Hittorf dark space; (4) the negative glow, the brightest of the glowing regions, is quite long compared with the cathode glow; (5) the Faraday dark space; (6) the positive column, which fills most of the length of the discharge tube; (7) the anode glow; and (8) the anode dark space (10, 39). The last two regions may or may not appear depending on the gas mixture, gas pressure, and on the value of the discharge current. The dark spaces are not absolutely devoid of light, but they are only dark relative to the glowing regions. When alternating voltages are used as the applied voltage, these regions will appear alternately at the cathode and at the anode. These alternations are dependent on the excitation frequency.

The major part of the voltage drop across the discharge occurs across the cathode dark space and is called the cathode fall of potential or cathode drop (39). The relative voltage gradient across the discharge is given in Figure 3. The cathode fall of potential is defined as the difference between the potential of the cathode and the potential in the gas at a distance 'd' from the cathode where 'd' is the thickness of the dark space. The voltage drop depends on the type of gas, the gas pressure, and the work function of the electrode material. The cathode fall is markedly affected by the presence of gaseous impurities, and it increases nearly linearly with the work function of the cathode material. The maintenance of the discharge depends on the emission of electrons from

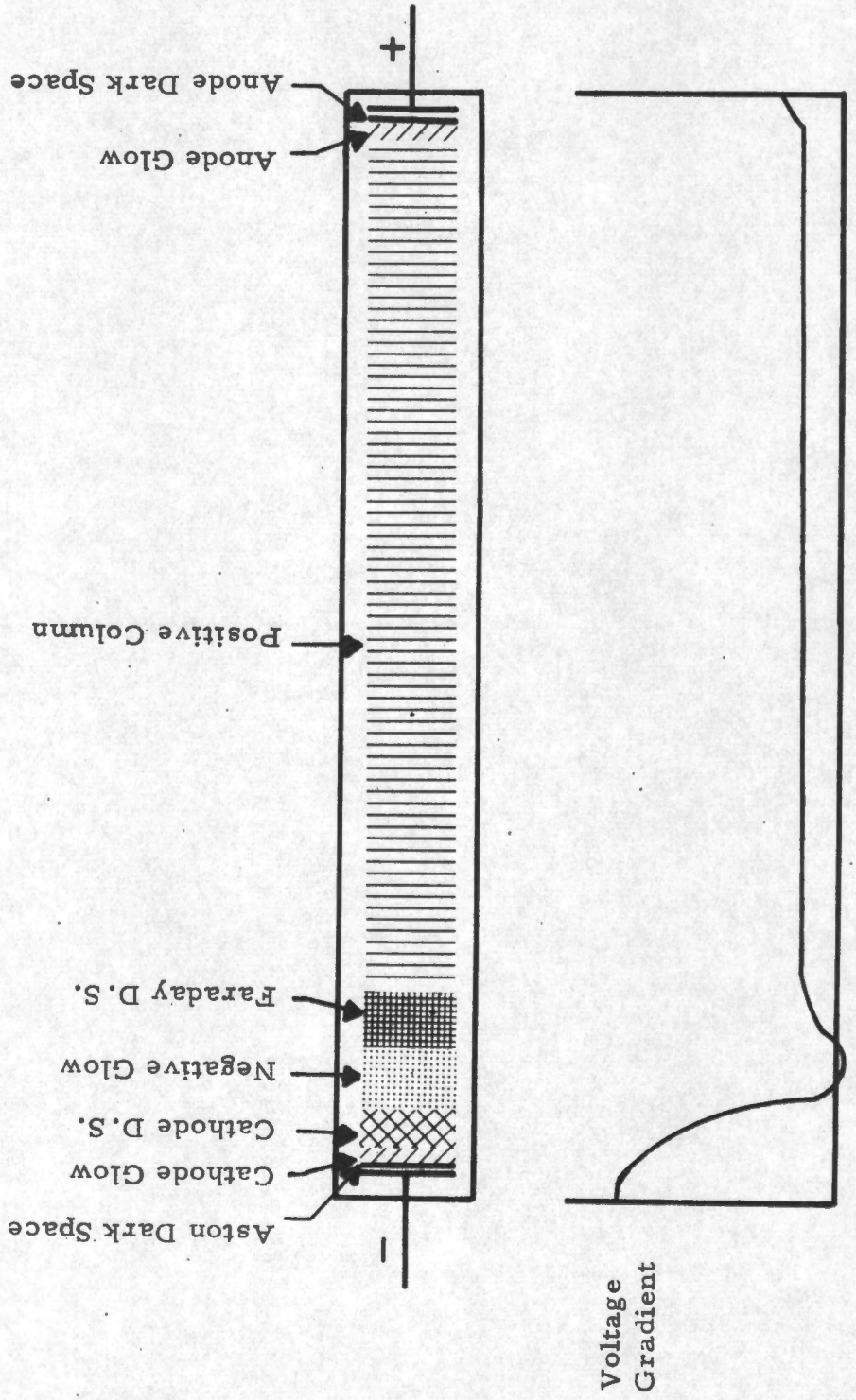


Figure 3. Details of the glow discharge and the relative voltage gradient.

the cathode. Cobine (10) reported that gaseous mixtures caused irregular and inexplicable variations in the cathode drop. The cathode drop is almost independent of the current and the pressure in a normal discharge (39).

The negative glow is the brightest of the glowing regions and is quite long compared to the cathode glow (10, 39). The voltage drop across this region is very small compared to the cathode drop. There is not a sharp visible boundary between the cathode drop and the negative glow. This region is the brightest on the cathode side, and is caused by high-speed electrons coming from the cathode and ionizing the gas molecules. The negative glow is regarded as the seat of ionization which by producing a supply of positive ions prevents the electrons from building up a space-charge, and thereby, keeps the electric force in the glow very low (39). As the electrons travel across the negative glow, they lose energy until not enough energy is left to ionize or produce luminoisty. Because of this slowing down of the electrons, there is no sharp boundary between the negative glow and the Faraday dark space.

In the transition region from the negative glow to the Faraday dark space, the ionizing and excitation effects from electron collisions become less. In the main part of the Faraday dark space, the ionizing and excitation action of electrons are completely absent. The electric field reverses in the transition region between the negative glow and the Faraday dark space. The current through the Faraday dark space is a diffusion current of electrons and positive ions with the electron current to the anode predominating (10).

The positive column of the glow discharge appears at the end of the Faraday dark space. The beginning of the positive column is very sharply defined; and its length depends upon the gas pressure, the distance between electrodes, and the shape of the container in which the discharge is established. The negative glow and the Faraday dark

space will expand at the expense of the positive column as the gas pressure is reduced. If the pressure is sufficiently reduced, the positive column will disappear completely. A similar effect is observed if the electrodes are moved together at a constant gas pressure and constant current, then the positive column decreases in length and finally disappears. However, normally the positive column fills the major portion of the tube. The column is defined as a "typical plasma having equal concentrations of positive ions and of electrons (10)." The glow of the positive column exhibits only the arc spectrum of the gas, and the conductance is maintained by relatively slow-moving electrons. Electron collisions are primarily responsible for maintaining the ionization in the plasma although some photo-ionization is present. There is a continual loss of electrons going on in the positive column caused by: (1) loss by diffusion to the sides of the tube; (2) loss by combination of electrons with neutral molecules to form negative ions; and (3) loss by combination of an electron with a positive ion to form a neutral atom or molecule (39). Under certain conditions of gas, current density, and pressure, alternate dark and bright striations appear in the positive column. These striations may move along the column or may be stationary (10). The gas temperature is greater in the luminous parts than in the dark parts. The temperature of the gas in the positive column is seldom over 100 degrees centigrade, and this indicates that thermal ionization cannot be a factor in the maintenance of the conductance of the glow discharge (10).

Positive ions are produced by the incoming electrons in the transition region between the positive column and the surface of the anode. Some of these ions move toward the positive column and some diffuse radially to the walls of the tube where they are lost by recombination with electrons. This diffusion of ions to the walls of the discharge tube tends to hinder the maintenance of the discharge (10). The walls

and outer boundary of the plasma gain a negative charge, because electrons and negative ions diffuse more rapidly in the tube than do positive ions. This results in a net positive charge in the central portion of the plasma and increases the diffusion of positive ions. An electron space charge exists immediately in front of the anode, and a potential drop occurs at the anode as a result of this electron space charge. This drop of potential usually occurs over a relatively short distance, is difficult to measure, and is subject to uncertainties of interpretation (10).

When an alternating current is used instead of a direct current, each electrode alternately acts as the cathode. The discharge gives the appearance of having a cathode dark space near each electrode with the positive column in the center when the frequency is in the order of sixty cycles per second. The length of the cathode dark space decreases as the frequency is increased. This in turn means a greater potential gradient in this region, which produces electrons having greater velocities (38, 39).

CHAPTER IV

THE IRRADIATION SYSTEM

The irradiation equipment for obtaining a glow discharge in air at low pressures and used in this study was essentially the same as that described by Webb, Stone, and Pate (43). Modifications were made in the frequency generating equipment for the research described in this thesis. The irradiation system consists basically of three sections: a variable frequency and variable high voltage section; a vacuum system equipped for pressure regulation; and a irradiation chamber. The complete apparatus is shown in Figures 4 and 5.

I. THE ELECTRICAL SYSTEM

The variable frequency and the variable voltage section can be divided into four units: (1) the frequency generator; (2) the pre-amplifier; (3) the amplifier; and (4) the high voltage transformers. A Hewlett-Packard test oscillator, model 650 A, was used as the frequency generator. The oscillator has a frequency range from 10 cycles per second to 100 megacycles per second and the frequency response was flat within _ one db in that range, with a maximum output of three volts into a 600-ohm resistive load. The output of the test oscillator was connected into a McIntosh thirty watt audio amplifier model A-116. This amplifier was used as a preamplifier for a Altec Lansing 250 watt amplifier, model A-287 w. The putput from the Altec Lansing amplifier was connected into the output transformers. The output transformers consisted of two 440-cycle transformers with the primary windings connected in parallel, and the secondary windings connected in series. The output of the transformers was connected to two electrodes, one on each end of the irradiation

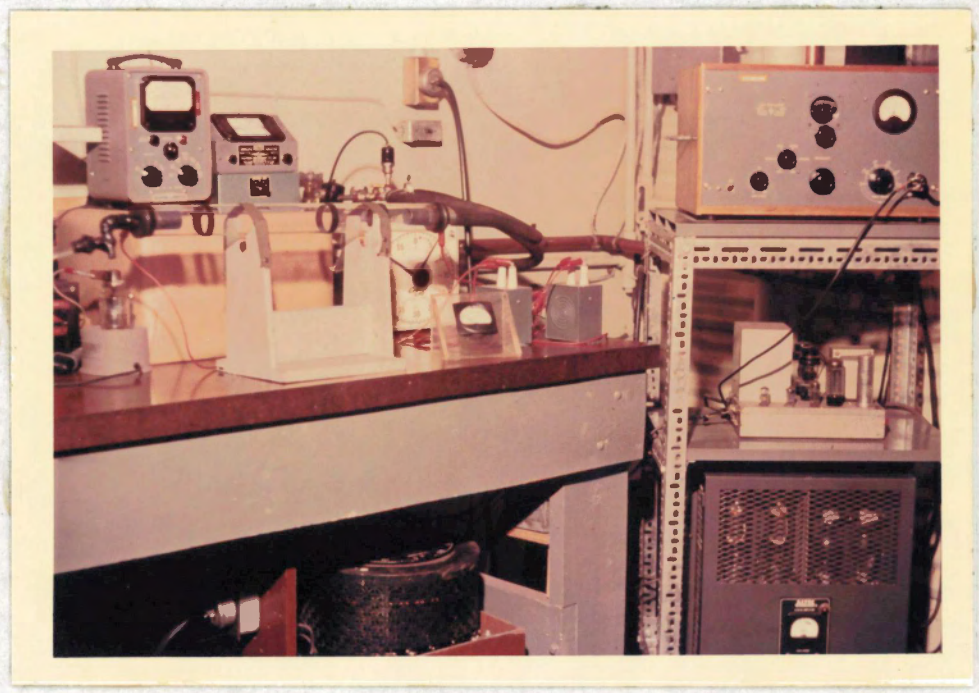


Figure 4. The complete irradiation system.

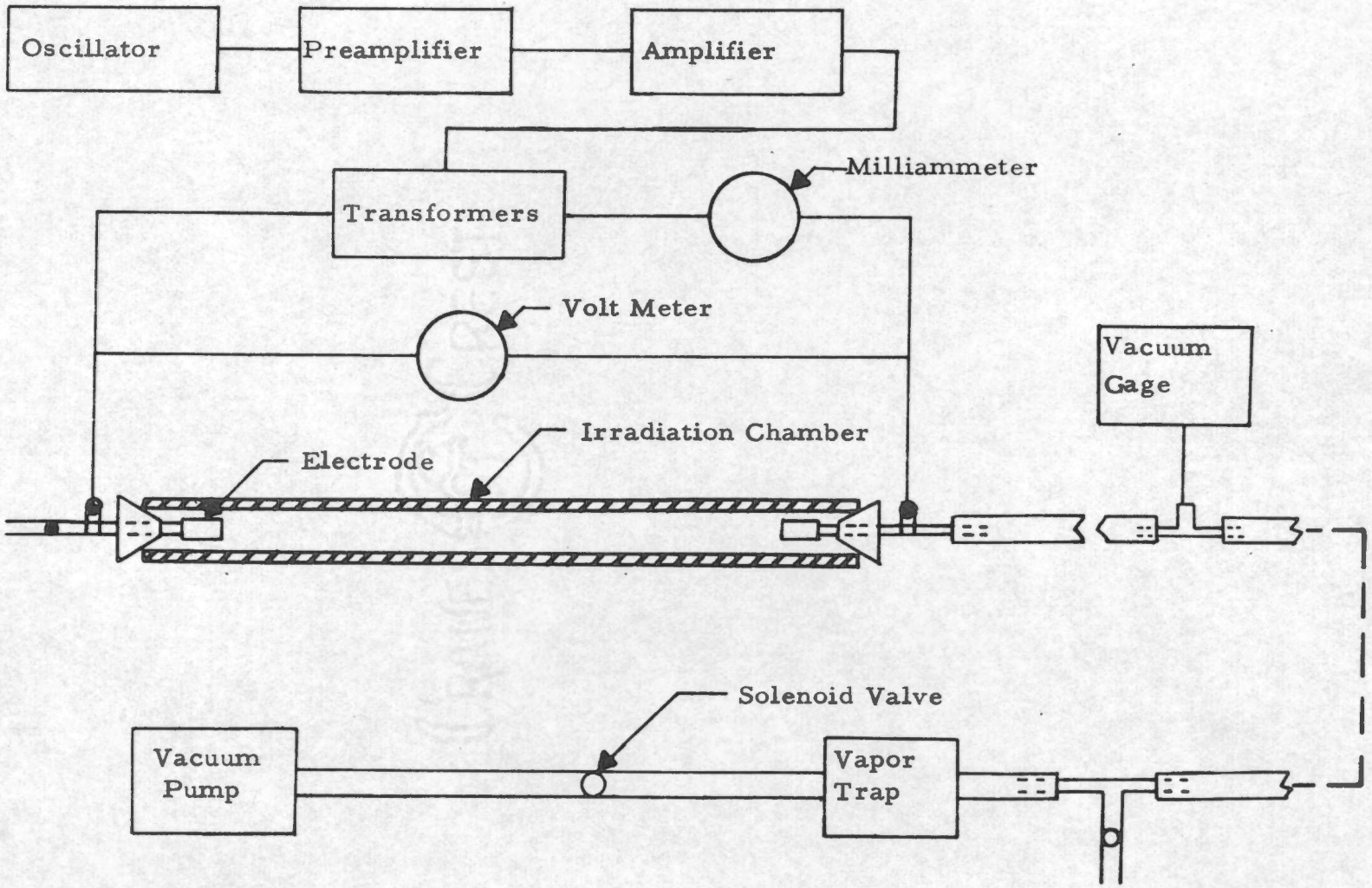


Figure 5. Block diagram of the irradiation system.

chamber. A Hewlett-Packard vacuum tube voltmeter, model 410 B; with a capacitive voltage divider, Hewlett-Packard, model 452 A, was used to indicate the voltage drop across the irradiation chamber. The readings of the vacuum tube voltmeter were compared with the readings of an electrostatic voltmeter, and the readings did not vary over 10 per cent between instruments. A radio-frequency milliammeter was connected in the circuit between the output of the transformers and one electrode to indicate the current flowing through the irradiation chamber. This section of the system is shown in Figure 6.

II. THE IRRADIATION CHAMBER

The irradiation chamber consisted of a borosilicate glass tube forty-eight millimeters inside diameter and twenty-four inches in length, Figure 7. The electrodes consisted of two, three-fourth inch polished black-iron pipe couplings, spaced eighteen inches apart. The glow discharge was established between the electrodes. A three-fourth to a three-eighth inch reducing bushing was fitted in one end of each electrode. A three-eighth inch black-iron pipe nipple was fitted to each reducer and was extended through the rubber stoppers which were used to seal each end of the irradiation chamber. The nipples were used as part of the vacuum system, and they also were part of the electrical system used to transmit current to the electrodes. A needle valve was fitted to one nipple, Figure 7, and pressure regulation was accomplished by adjustments of the needle valve. The other nipple was fitted with a thick wall rubber hose which connected the irradiation chamber to a vapor trap and then to a vacuum pump.

III. THE VACUUM SYSTEM

The irradiation chamber was evacuated and held at a low pressure throughout the irradiation treatment. The vacuum was obtained by using



Figure 6. The voltage and frequency generating equipment.

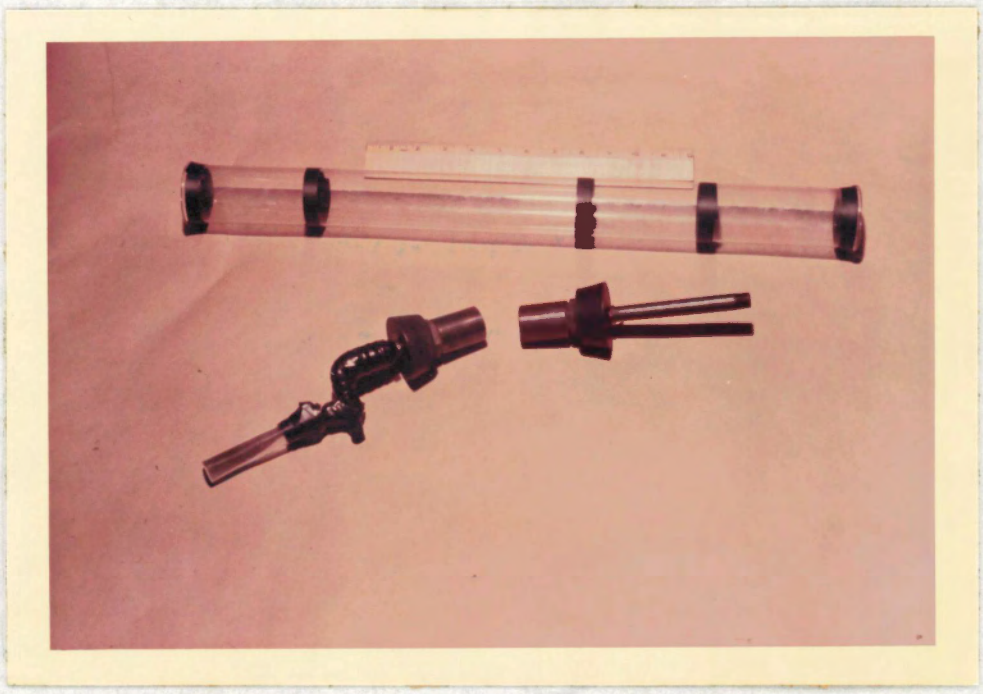


Figure 7. The electrodes, needle valve, and irradiation chamber.

a Beach-Russ model 15 vacuum pump, powered by a one and one-half horsepower General Electric induction motor. A one and one-quarter inch copper pipe was used to connect the vacuum pump to a solenoid valve. Thick wall rubber hose joined the solenoid valve to a vapor trap and the vapor trap to the irradiation chamber. The vapor trap, which was packed with dry ice and methanol, was used to dry the air entering the vacuum pump. A second solenoid valve was placed in the vacuum line on the irradiation chamber side of the vapor trap allowing the irradiation chamber to come up to atmospheric pressure when the first solenoid valve closed. This permitted the irradiation chamber to come up to atmospheric pressure after each treatment without adjustment of the needle valve.

Pressure was measured in the connecting system approximately two feet from the treating chamber. A Hastings Absolute Pressure Indicator, model AP 18, with a matched sensing element was used to determine the vacuum in millimeters of mercury. The sensing element was placed in the vacuum line between the irradiation chamber and the vapor trap. The pressure indicator system was calibrated with a McLeod Gauge. An isolation transformer in the power input to the pressure indicator eliminated interference that might come from the irradiation section. The complete vacuum system is shown in Figure 5, page 24.

CHAPTER V

EXPERIMENT PROCEDURES

The design chosen was considered from the standpoint of obtaining the maximum information from the data. The characteristics of the irradiation equipment were dependent on the treatment of the experimental material. The experimental design, treatment of the experimental material, characteristics of the irradiation equipment, germination procedures, and collection of data are discussed in this chapter.

I. EXPERIMENTAL DESIGN

A factorial experimental design was used, in order that a number of different factors could be investigated simultaneously. The treatments consisted of all possible combinations of the different factors (11). Four current levels, 20, 40, 80, and 120 milliamperes, and five excitation frequencies, 500, 5,000, 10,000, 15,000, and 20,000 cycles per second, were used as treatments. The treatments were chosen to cover the frequency range of the equipment. The total experiment, which consisted of twenty treatments and one set of controls, was replicated eight times.

II. TREATMENT OF THE EXPERIMENTAL MATERIAL

This study was conducted using machine delinted Empire W. R. cottonseed produced in 1960. Before treatment the seeds were kept in a seed storage unit as a temperature of forty degrees Fahrenheit and at a relative humidity of 50 per cent. The moisture content of the seed at the time of treatment was approximately 10 per cent. In

order to assure relatively constant moisture content of the seed at the time of treatment, only small amounts of seed were removed from the storage unit at one time. The seed were allowed to come up to room temperature, approximately seventy-five degrees Fahrenheit, before the treatment began. After the seed were irradiated, they were returned to the seed storage unit to condition for a minimum period of one week. This conditioning period allowed the seed to regain moisture that had been lost during treatment. After the conditioning period, the seeds were ready for the germination test.

Twenty grams of seeds were placed in the center twelve inches of the irradiation chamber between the electrodes, and then they were spread to a single layer on the bottom of the chamber. The treating chamber, containing the seed, was evacuated for one minute before the glow discharge was established. The time required to evacuate the chamber to three millimeters of mercury was between fifteen and twenty seconds. The remaining forty to forty-five seconds were allowed for pressure adjustments, if required. The pressure in the irradiation chamber was adjusted with a needle valve connected to one of the electrodes. All irradiation treatments were of three minutes duration at a pressure of three millimeters of mercury.

III. SYSTEM'S CHARACTERISTICS DURING TEST

The characteristics were determined with the irradiation chamber under normal operating conditions as the load for the amplifier system. The input voltage to the preamplifier and the output voltage, across the irradiation chamber, were measured approximately one minute after the discharge had been established for each treatment. To get comparable data it was important to take these readings at approximately the same time during each treatment, because the rate of outgasing from the seed decreased as the treatment progressed. The change in the

rate of outgasing would cause a gradual change in the gas mixture in the irradiation chamber and; therefore, a change would occur in the ionization potential of the gas. The characteristics for the system are shown in Table I and Figure 8.

The voltage drop across the irradiation chamber was dependent upon the conditions that existed within the chamber. All conditions, except current and frequency, were kept as nearly constant as possible from one treatment to another. The voltage drop varied slightly with current but remained almost constant with frequency as shown in Table I. The voltage drop across the chamber did not exceed 1, 300 root mean square volts in any treatment, as measured with the vacuum tube voltmeter previously described.

The frequency response of the system at each current level was relatively flat over the entire frequency range as shown in Figure 8. Due to the erratic behavior of the discharge, the characteristics were not measured for the twenty milliampere treatments. This erratic behavior was caused by the position at which the discharge occurred on the characteristics curve. The conditions that existed within the chamber caused the discharge, at the twenty milliampere treatment, to take place at the beginning of the glow and at the end of the Townsend discharge region, as shown in the shaded area in Figure 2 (10).

In general, the frequency response of the system was lowest at 500 cycles per second and highest at 5, 000 cycles per second for the 40, 80, and 120 milliampere treatments. However, the system's output voltage was sufficient to establish a glow discharge at all current and frequency combinations.

The gain of the system was more dependent on the current level than on the frequency. The gain varied from approximately 6, 400 at 40 milliampere to 3, 200 at 120 milliampere. There was only a slight variation due to frequency as shown in Figure 8.

TABLE I

SYSTEM RESPONSE FOR VARIOUS CURRENTS AND FREQUENCIES

Frequency cps	Current MA*	Input volts**	Output volts***	Gain****
500	40	0.18	1000	5,500
	80	0.21	960	4,600
	120	0.30	960	3,200
5,000	40	0.17	1100	6,500
	80	0.20	1000	5,000
	120	0.29	1000	3,400
10,000	40	0.17	1050	6,200
	80	0.20	980	4,900
	120	0.27	980	3,600
15,000	40	0.17	1050	6,200
	80	0.20	980	4,900
	120	0.29	980	3,400
20,000	40	0.17	1120	6,600
	80	0.20	1000	5,000
	120	0.29	980	3,400

* Current through the chamber.

** Input to the preamplifier.

*** Voltage drop across the irradiation chamber.

**** $\frac{\text{Output voltage}}{\text{Input voltage}}$

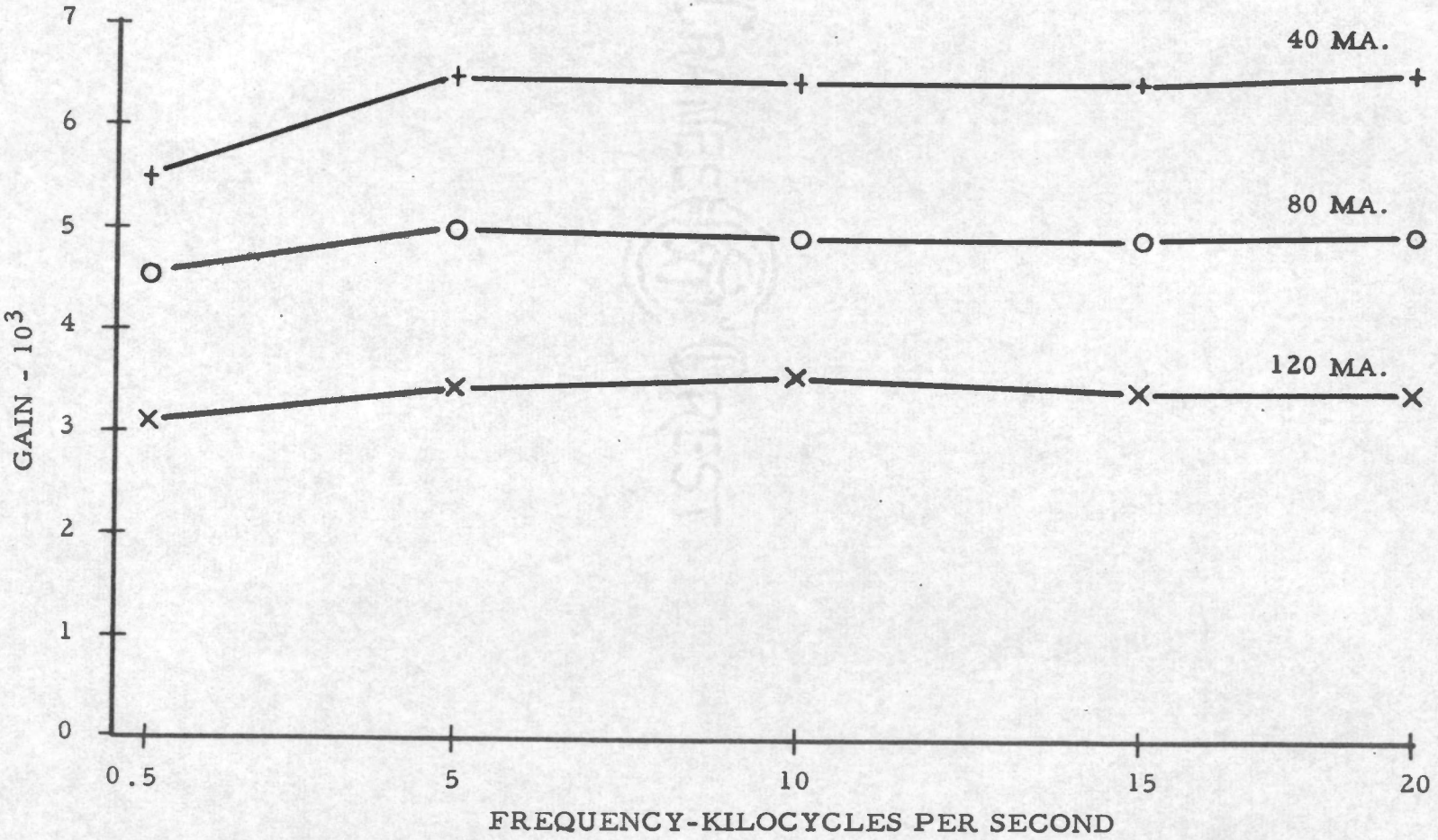


Figure 8. Gain of the system at various frequencies and currents.

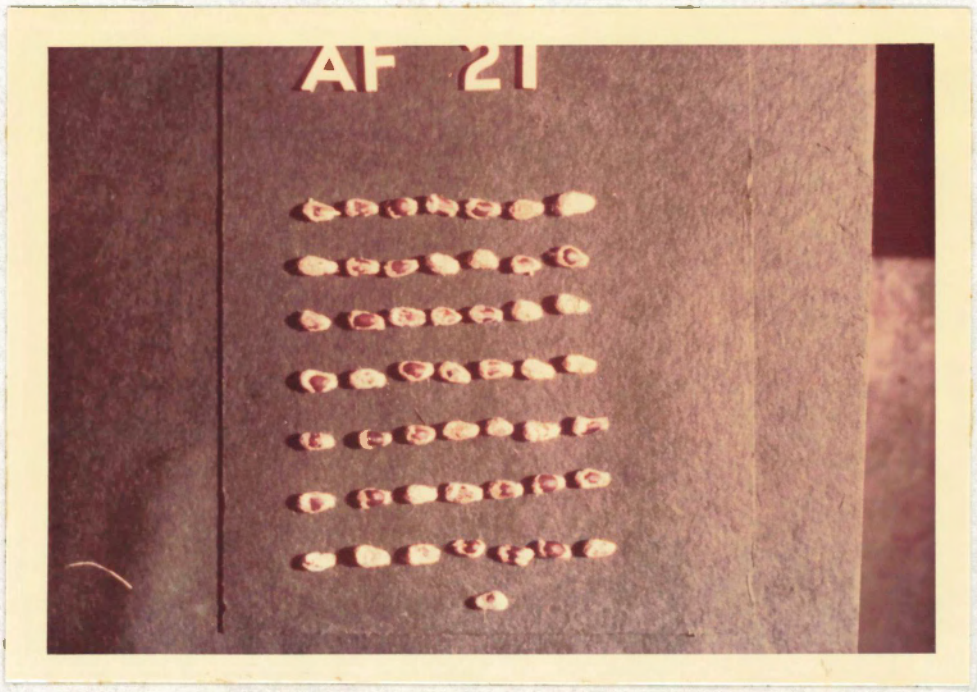
IV. GERMINATION PROCEDURES

A flowing water sheet type seed germinator, similar to that reported by Nelson (28), was used. The water was conditioned to the proper temperature in a tank, pumped to the top of the germinator, and then allowed to flow down the inside back of the germinator. The sheet of water served three purposes: (1) to control the temperature inside the germinator; (2) to prevent temperature stratification; (3) and to hold the relative humidity between ninety-five and one hundred per cent. The high relative humidity was important so that the moisture content of the substrata could be kept as constant as possible during germination. The germinator was equipped with automatic temperature controls which maintained the temperature at sixty-eight degrees Fahrenheit for sixteen hours and eighty-six degrees Fahrenheit for eight hours. Light is not required for the germination of the cotton-seed (40).

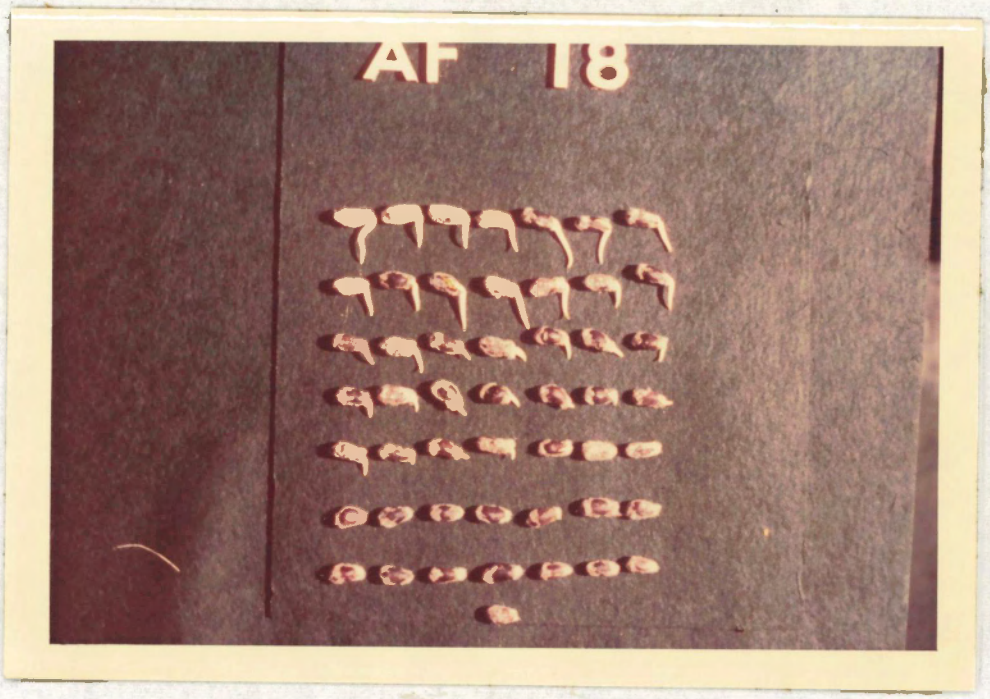
Standard germination towels and blotters were used as the substrata for the germination test. The blotters were used to help maintain a constant moisture level in the substrata. The towels were prewet in a solution of four grams of Ceresan M to five gallons of water. Ceresan M is a seed disinfectant and was used to retard the growth of micro-organisms on the seed. Each replication consisted of fifty seeds per treatment placed inside four folded towels.

V. COLLECTION OF DATA

Germination counts were made at the end of the second and fourth days. The counts on the second day were recorded as early germination and the fourth day counts as the total germination. Figure 9 presents a control and a treated sample after the second day. A comparison of a control and a treated sample after the fourth day is



a - Control



b - Treated

Figure 9. A comparison of the control and the treated seed germination at two days.

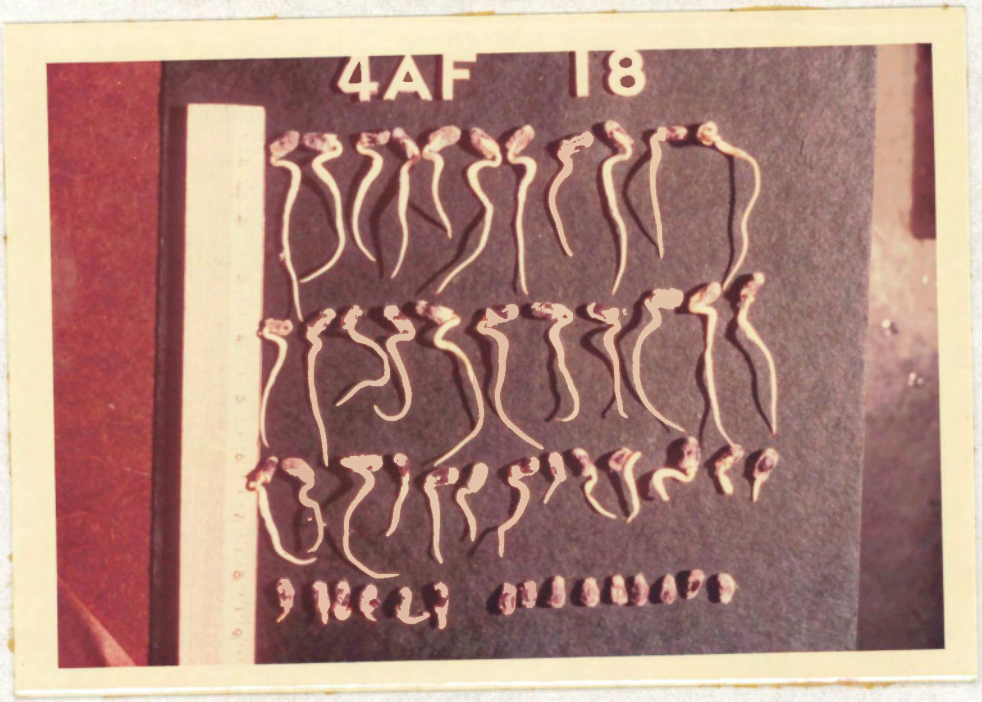
shown in Figure 10. Germinations were recorded as the number of seed germinated. This data was then converted to per cent germination and all analyses were made in per cent germination. The abnormal germinations were handled in a like manner. An example of a normal and an abnormal radicle is shown in Figure 11. Abnormals were so classified in accordance with the Agriculture Handbook Number 30 (42).

In addition to the effects on germination, the effects on radicle development were measured. The radicle development was scored on the basis of the radicle length, weight, and the per cent of radicles over one inch after four days. The radicle length was determined by measuring each radicle having a growth of one-half inch or greater. The one-half inch mark was chosen arbitrarily, due to the necessity of having a sharp dividing line of measurement for classification of germination. The measurements for each replication were totaled and averaged, and this average was used in the analyses.

The radicle weight was determined by removing the radicles at the base of the seedcoats and getting a dry weight of the radicles. After the radicles were removed from the seedcoats, they were placed in a circulating air oven at 220 degrees Fahrenheit for twenty-four hours. At the end of this period of time, the radicles were removed from the oven and weighed.



a - Control



b - Treated

Figure 10. A comparison of the control and the treated seed germination at four days.



Figure 11. A contrast of a normal and an abnormal radicle.

CHAPTER VI

PRESENTATION AND INTERPRETATION OF THE EXPERIMENTAL DATA

The primary objective in seed testing is to obtain accurate and reproducible results regarding the percentage of seeds that can be expected to produce normal plants under favorable conditions (42). Germination testing has become a standard tool for predicting the ability of seeds to produce normal plants (28). In cotton production it is important not only to get normal plants but also to get them as early as possible after planting. Early radicle development is important, because the earlier the radicle gets established in moist soil the less chance of dry weather damage to the plant.

The data obtained during this study were statistically analyzed, and the means were compared using the new Duncan's Multiple Range comparison at the five per cent probability level (35). The results are presented graphically for each treatment where feasible. The exact values of points plotted in the figures are listed in tabular form. Where graphic presentation is not feasible, comparisons are made in tabular form only. Lines joining points on graphs indicate trends and should not be used to predict ordinate values for a given treatment.

I. EFFECTS OF IRRADIATION ON GERMINATION

One objective of this research was to determine if the radiation from the plasma had any effects on early and total germination of cottonseed under germinator conditions. Germination tests revealed that all treatments were effective in producing early germination. The overall treatment comparisons on germination are presented in Tables II and in Figure 12. The mean per cent germination for

TABLE II

MEAN PER CENT GERMINATION OF TWO DAY, FOUR DAY, AND ABNORMAL SEEDLINGS FOR ALL TREATMENTS AND CONTROLS

Treatment*	Means**		
	2-Day Germination	4-Day Germination	Per Cent Abnormals
Control	1.0 a	89.0 def	0.5 a
0.5-20	65.0 de	90.5 f	3.5 abcd
0.5-40	57.5 bcde	87.0 def	4.2 bcd
0.5-80	60.0 cde	85.5 cdef	3.0 abcd
0.5-120	62.0 cde	86.0 cdef	1.7 ab
5-20	58.5 bcde	89.0 ef	1.7 ab
5-40	64.0 cde	87.5 def	3.7 abcd
5-80	60.0 cde	83.0 abcde	5.2 cd
5-120	59.0 cde	83.5 bcde	5.5 d
10-20	59.5 cde	87.0 def	2.5 abc
10-40	61.0 cde	82.0 cdef	2.0 abc
10-80	59.0 cde	85.5 cdef	3.7 bcd
10-120	53.5 bc	78.0 ab	3.7 bcd
15-20	59.0 cde	88.5 def	3.0 abcd
15-40	67.0 e	86.5 def	4.5 bcd
15-80	48.5 b	77.0 a	5.7 d
15-120	61.0 cde	80.0 abc	5.5 cd
20-20	59.5 cde	85.0 cdef	2.0 abc
20-40	59.0 cde	86.5 def	2.2 abc
20-80	53.0 bc	82.5 abcde	2.5 abc
20-120	55.0 bcd	80.0 abc	4.5 bcd

*The first number denotes the frequency in kilocycles per second. The second number denotes the current in milliamperes.

**Means within a column followed by the same letter are not significantly different at the 5-per cent probability level.

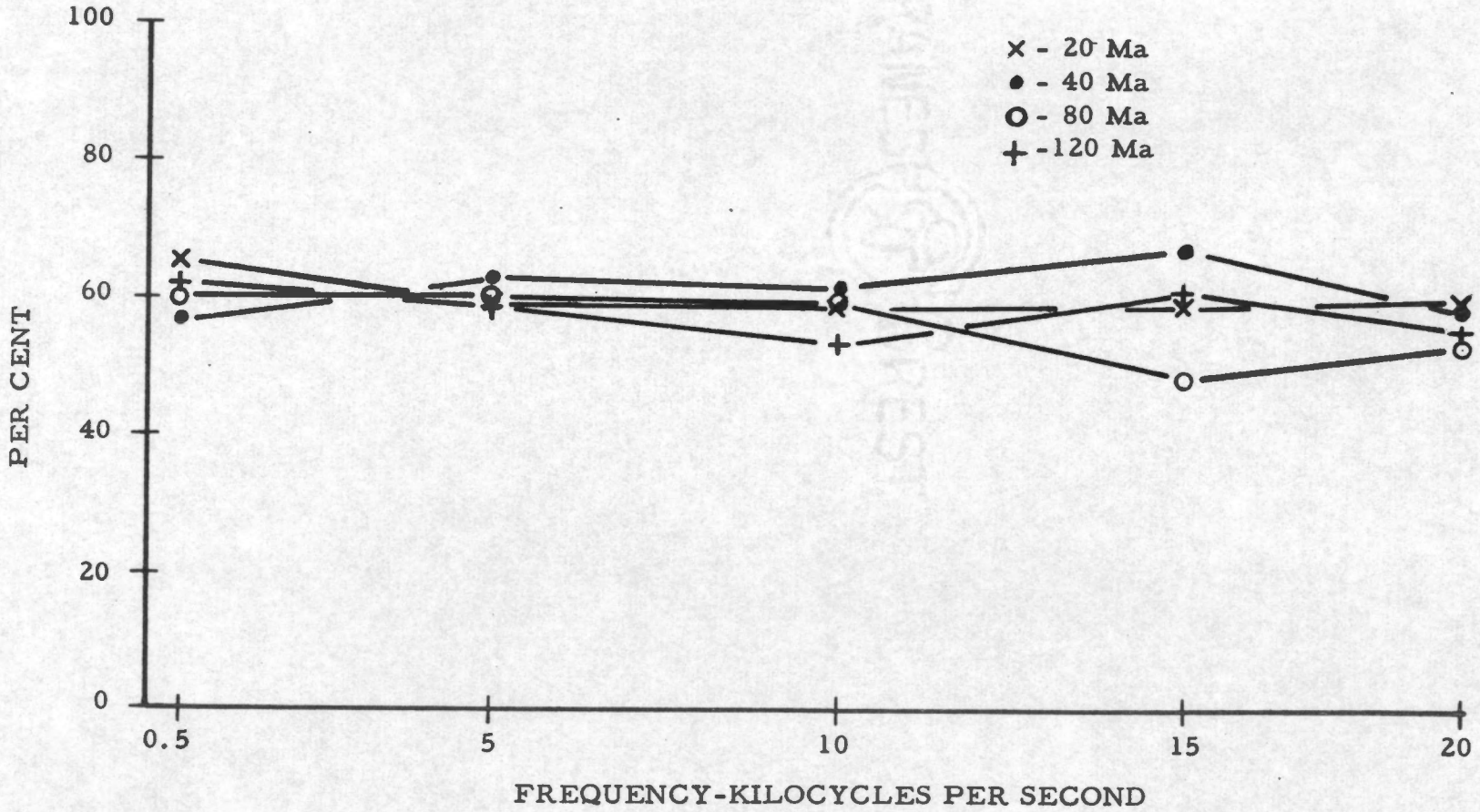


Figure 12. Early germination verses frequency at each current level.

the two day, four day, and abnormal seedlings are presented in Table II, page 40, for all treatments and the controls. All irradiated seeds were significantly better in early germination than the control seed. The control seed has only 1 per cent germination after two days, while the irradiated seed ranged from 48.5 to 67 per cent germination. However, there were differences in early germination caused by the irradiation treatments as shown in Table II, page 40. The greatest differences occurred at fifteen kilocycles. At this frequency, early germination was significantly better at the 20, 40, and 120 milliampere treatments than at the eighty milliampere treatment. The differences between current levels at all other frequencies were not significant. This same frequency and current combination also produced the lowest total germination and the highest per cent of abnormals. The radiation treatments produced no significant beneficial effects on total germination. However, some treatments produced germinations that were significantly lower in total germination than the controls.

The effects caused by the excitation frequencies for the early germination and total germination are shown in Figure 13 and Table III. There were no significant differences in early germination caused by the excitation frequencies. However, the trend in early germination was to decrease as the frequency was increased. Some of the decrease in total germination can be contributed to frequency, because as the frequency increased there was a significant decrease in total germination. The factorial analyses showing the significance for each source of variation for germination and radicle development are shown in Table VII, page 50. The interaction between the frequency and current in both early and total germination was non-significant at the 5 per cent level.

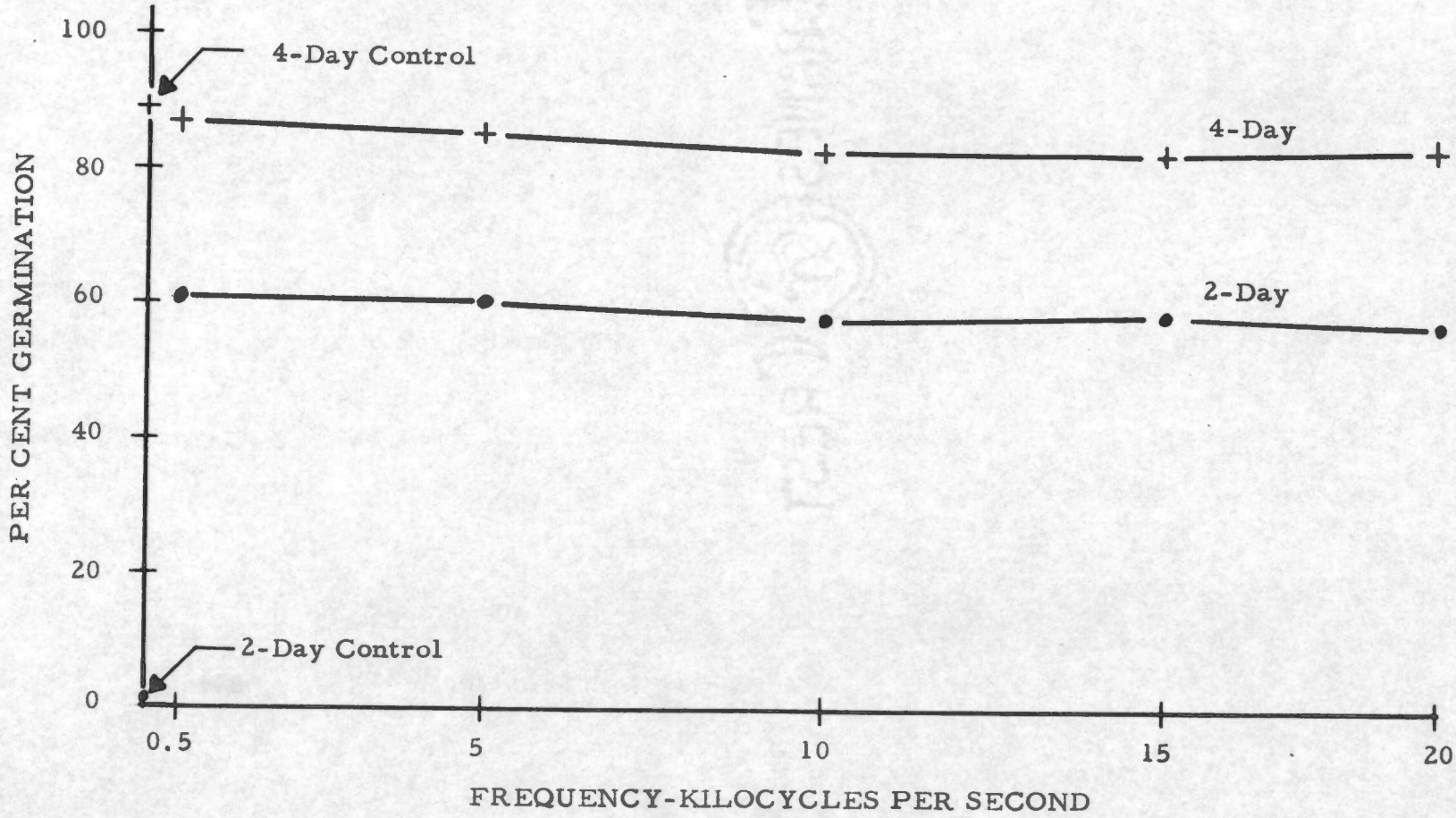


Figure 13. Early and total germination as a function of frequency.

TABLE III

MEAN PER CENT GERMINATION AND RADICLE DEVELOPMENT AT EACH FREQUENCY TREATMENT

Frequency Kilocycles	Per Cent Germination			Radicle Development		
	2-Day	4-Day	Abnormals	Radicle Length-in.	Radicle Weight-mg.	Per cent of Radicles Over 1 inch
0.5	61.3 a*	87.2 a	3.1 a	1.54 a	7.5 a	69.7 a
5	60.2 a	85.7 b	4.1 a	1.54 a	7.5 a	67.8 a
10	58.4 a	83.1 c	3.0 a	1.54 a	7.3 a	67.6 a
15	59.0 a	82.9 c	4.7 a	1.56 a	7.5 a	65.4 a
20	57.0 a	83.5 c	2.8 a	1.54 a	7.4 a	65.5 a

*Means within a column followed by the same letter are not significantly different at the 5-per cent probability level.

The different current levels affected both early and total germination in a manner similar to that of frequency as shown in Figure 14 and Table IV. The differences between current levels in early germination were non-significant, but as the current level increased, there was a significant decrease in total germination. Both the current and frequency were effective in reducing the total germination. The effects on early and total germination of each treatment are presented in Table V.

The per cent abnormals for each treatment and the control seed are presented in Table II, page 40. There was a significant increase in abnormals in some treatments compared to the controls. The abnormals were shorter and larger in diameter than normal radicles and were yellowish in appearance. The general appearance of the abnormal radicles was very similar to those radicles from aged cotton-seed or radicles which had been overly treated with ceresan.

The per cent abnormals were not significantly affected either by frequencies or current levels, Tables III, page 44, and IV. As the current was increased the per cent of abnormal radicles appeared to increase, but this trend was not significant. The interaction between frequencies and currents was non-significant.

II. EFFECTS OF IRRADIATION ON RADICLE DEVELOPMENT

Another objective of this research was to determine if irradiation had any effects on radicle development. The overall treatment comparisons of the radicle development study as measured by radicle growth, radicle weight, and the per cent of radicles over one inch are shown in Table VI. All treatments were effective in producing a significantly longer radicle and a heavier radicle than that of the control seed. The radicle length and weight of the radiated seeds were about twice that of the control seed. There were no significant differences between any

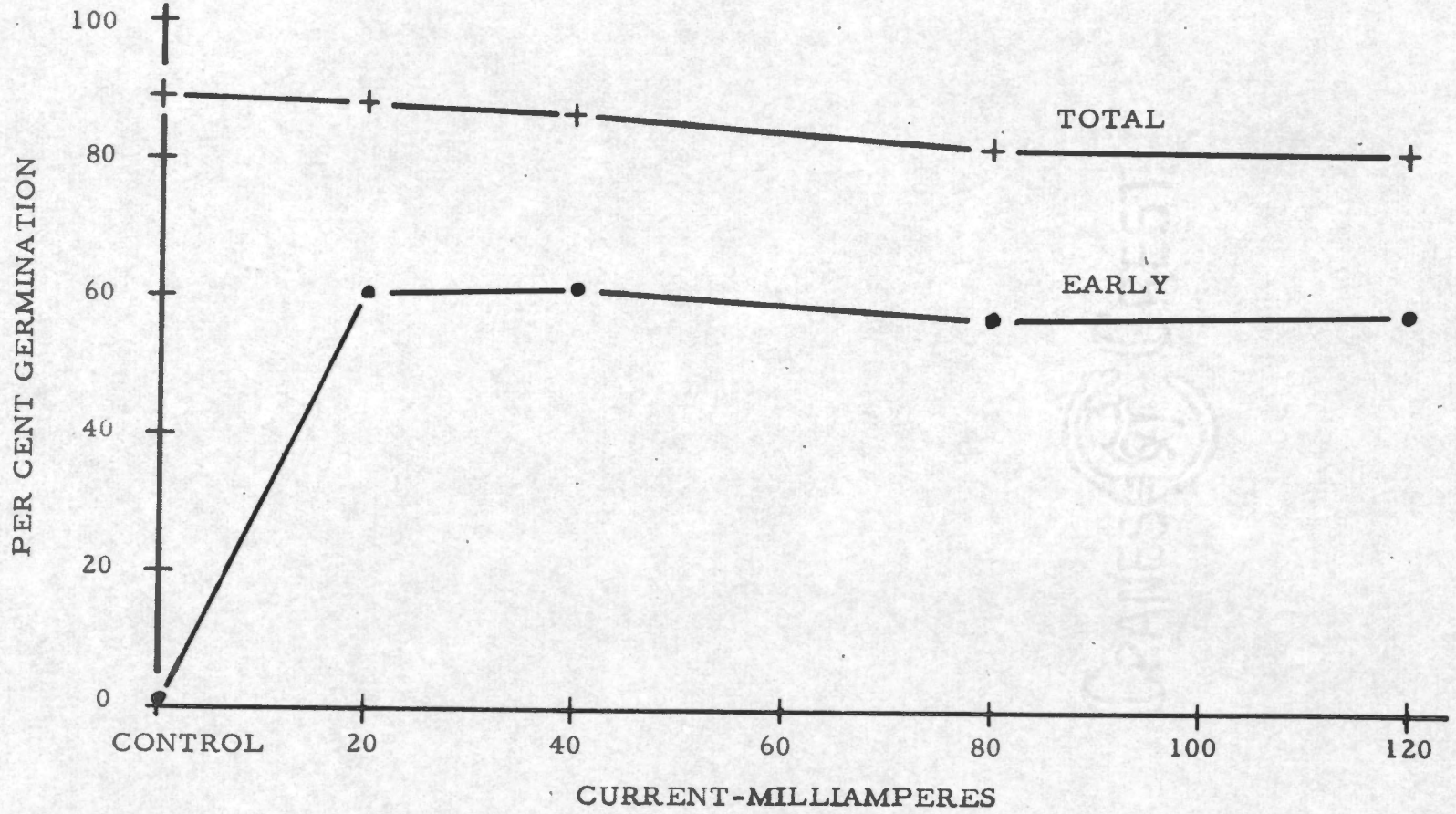


Figure 14. Early and total germination as a function of current.

TABLE IV

MEAN PER CENT GERMINATION AND RADICLE DEVELOPMENT AT EACH CURRENT LEVEL

Current Ma.	Per cent Germination			Radicle Development		
	2-Day	4-Day	Abnormal	Radicle Length-in.	Radicle Weight Mg.	Per Cent of Radicles Over 1 Inch
20	60.4 a*	87.9 a	2.5 a	1.55 a	7.4 a	70.1 a
40	61.8 a	86.4 b	3.3 a	1.54 a	7.5 a	70.5 a
80	57.2 a	82.0 c	4.0 a	1.53 a	7.3 a	63.8 a
120	58.2 a	81.6 c	4.2 a	1.56 a	7.5 a	64.4

*Means within a column followed by the same letter are not significantly different at the 5-per cent probability level.

TABLE V

MEAN PER CENT GERMINATION AFTER TWO AND FOUR DAYS FOR VARIOUS CURRENTS AND FREQUENCIES

Frequency	Current-Milliamperes							
	20		40		80		120	
Kilocycles	2-Day	4-Day	2-Day	4-Day	2-Day	4-Day	2-Day	4-Day
0.5	65.7 cd*	90.5 f*	57.5 abcd	86.7 def	60.0 bcd	85.5 cdef	62.0 bcd	86.2 cdef
5	58.5 abcd	89.0 ef	63.7 bcd	87.5 def	60.2 bcd	82.7 abcde	59.0 bcd	83.5 bcde
10	59.5 bcd	86.7 def	61.5 bcd	85.2 cdef	59.2 bcd	82.2 cdef	53.5 ab	78.5 ab
15	59.0 bcd	88.5 def	67.2 d	86.5 def	48.5 a	77.0 a	61.2 bcd	80.0 abc
20	59.5 bcd	85.0 cdef	59.2 bcd	86.5 def	53.0 ab	82.5 abcde	55.2 abc	80.0 abc

*Means within a two day column for all treatments followed by the same letter are not significantly different at the 5 per cent probability level.

**Means within a four day column for all treatments followed by the same letter are not significantly different at the 5 per cent probability level.

TABLE VI

MEAN RADICLE LENGTH, WEIGHT, AND PER CENT OF RADICLES OVER ONE INCH FOR ALL TREATMENTS AND CONTROLS

Treatment*	Means**		
	Radicle Length-in	Radicle Weight-mg/Radicle	Per cent of Radicles Over 1 in.
Control	0.85 a	3.9 a	19.7 a
0.5-20	1.58 b	7.5 b	72.4 de
0.5-40	1.50 b	7.5 b	69.2 cde
0.5-80	1.53 b	7.6 b	67.7 bcde
0.5-120	1.55 b	7.3 b	69.6 cde
5-20	1.49 b	7.1 b	67.6 bcde
5-40	1.55 b	7.7 b	70.2 cde
5-80	1.55 b	7.6 b	69.4 cde
5-120	1.55 b	7.5 b	64.1 bcd
10-20	1.53 b	7.2 b	70.0 cde
10-40	1.61 b	7.6 b	72.2 de
10-80	1.53 b	7.3 b	61.6 bc
10-120	1.50 b	7.1 b	66.7 bcde
15-20	1.54 b	7.6 b	67.4 bcde
15-40	1.59 b	7.2 b	75.2 e
15-80	1.56 b	7.2 b	58.4 b
15-120	1.57 b	7.7 b	60.9 bc
20-20	1.53 b	7.6 b	73.6 de
20-40	1.44 b	7.0 b	65.5 bcde
20-80	1.50 b	7.0 b	62.1 bc
20-120	1.61 b	7.8 b	60.7 bc

*The first number denotes the frequency in kilocycles per second. The second number denotes the current in milliamperes.

**Means within a column followed by the same letter are not significantly different at the 5-per cent probability level.

TABLE VII

FACTORIAL ANALYSIS OF GERMINATION AND RADICLE DEVELOPMENT FOR ALL LEVELS OF CURRENT AND FREQUENCY

SOURCE OF VARIATION	GERMINATION			RADICLE DEVELOPMENT		
	2-Day	4-Day	Abnormals	Length	Weight	Percent Over 1-Inch
REPLICATIONS	*[a]	N.S.	*	*	*	*
CURRENT	N.S.[b]	*	N.S.	N.S.	N.S.	N.S.
FREQUENCY	N.S.	*	N.S.	N.S.	N.S.	N.S.
CURRENT X FREQUENCY INTERACTION	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

[a]Significant at the 5 per cent probability level.

[b]Non-significant at the 5 per cent probability level.

of the treatments for radicle length or radicle weight. There were differences in the per cent of radicles over one inch after four days between treatments. As in early germination, there were no significant differences among the frequencies or current levels on radicle development as shown in Figures 15 and 16, and Tables III, page 44 and IV, page 47.

Cottonseed irradiated with the gas-plasma germinated earlier and had better radicle development after four days than did the control seed. The total germination was not significantly increased by irradiation. There were no significant differences among frequencies or currents, or a combination of frequency and current, on early germination or radicle development. Another effect observed by the author was that plasma irradiation at frequencies above sixty cycles per second also caused the cotton linters attached to the seed to become water absorbent. The water absorption of the treated linters was very rapid; and the irradiated seeds, when dropped into a container of water, sank to the bottom immediately. The control seeds still floated on the surface of the water twenty-four hours after placement.

III. INTERPRETATIONS

The electron micrographs presented by Stone (36) revealed broken surface areas on cotton fibers which had been exposed to a gas-plasma. These broken surface areas had the appearance of localized high temperature effects. Stone also reported that the waxes on the individual fibers and the primary wall cells were degraded as a result of irradiation. These cotton fibers absorbed water very rapidly. Roseman, et al. (30) reported that irradiation of rice with the gas-plasma and the heating of rice in a vacuum resulted in rapid water uptake about equally for both treatments. A comparison of infrared,

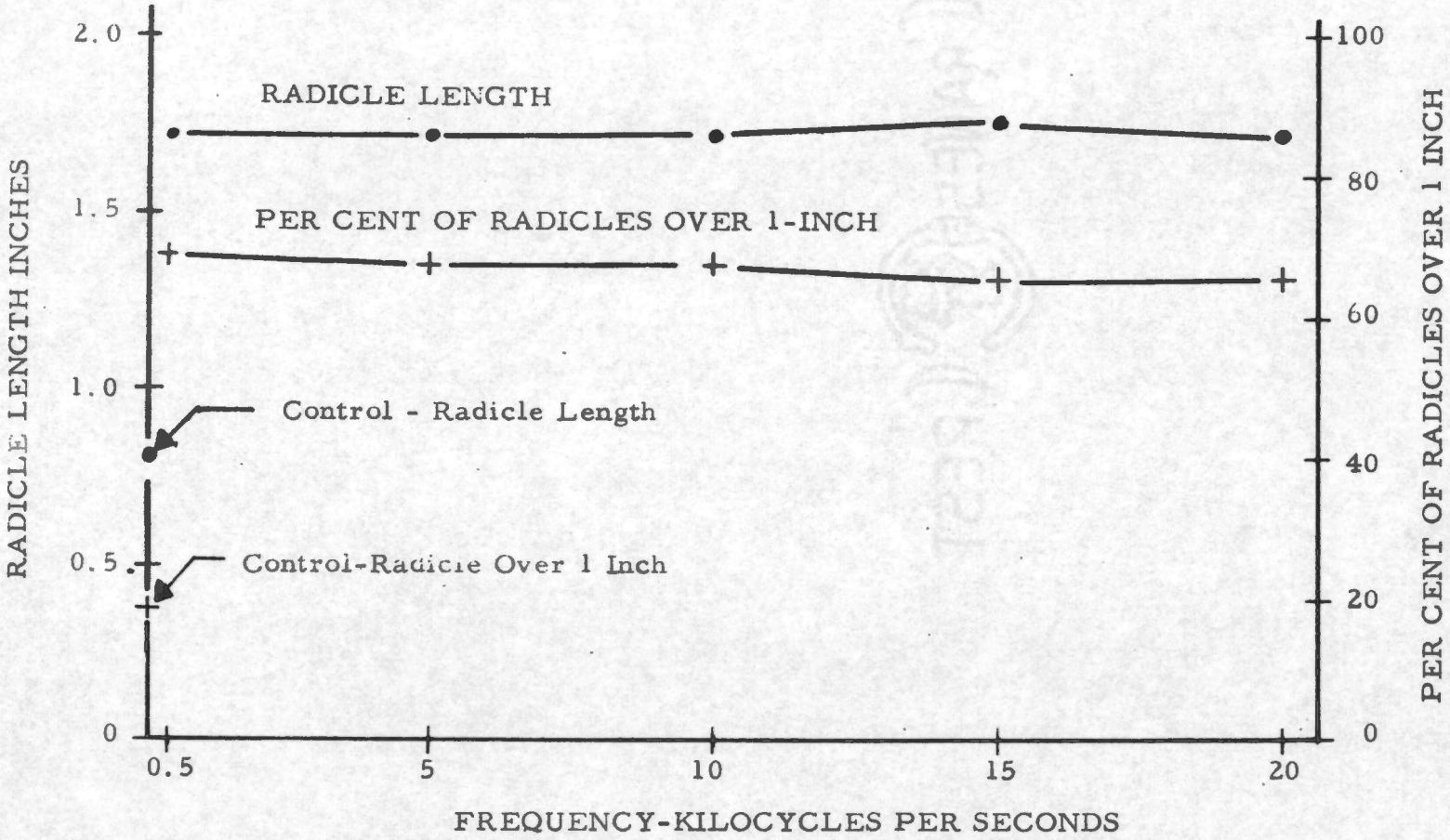


Figure 15. Radicle length and per cent of radicles over one inch as a function of frequency.

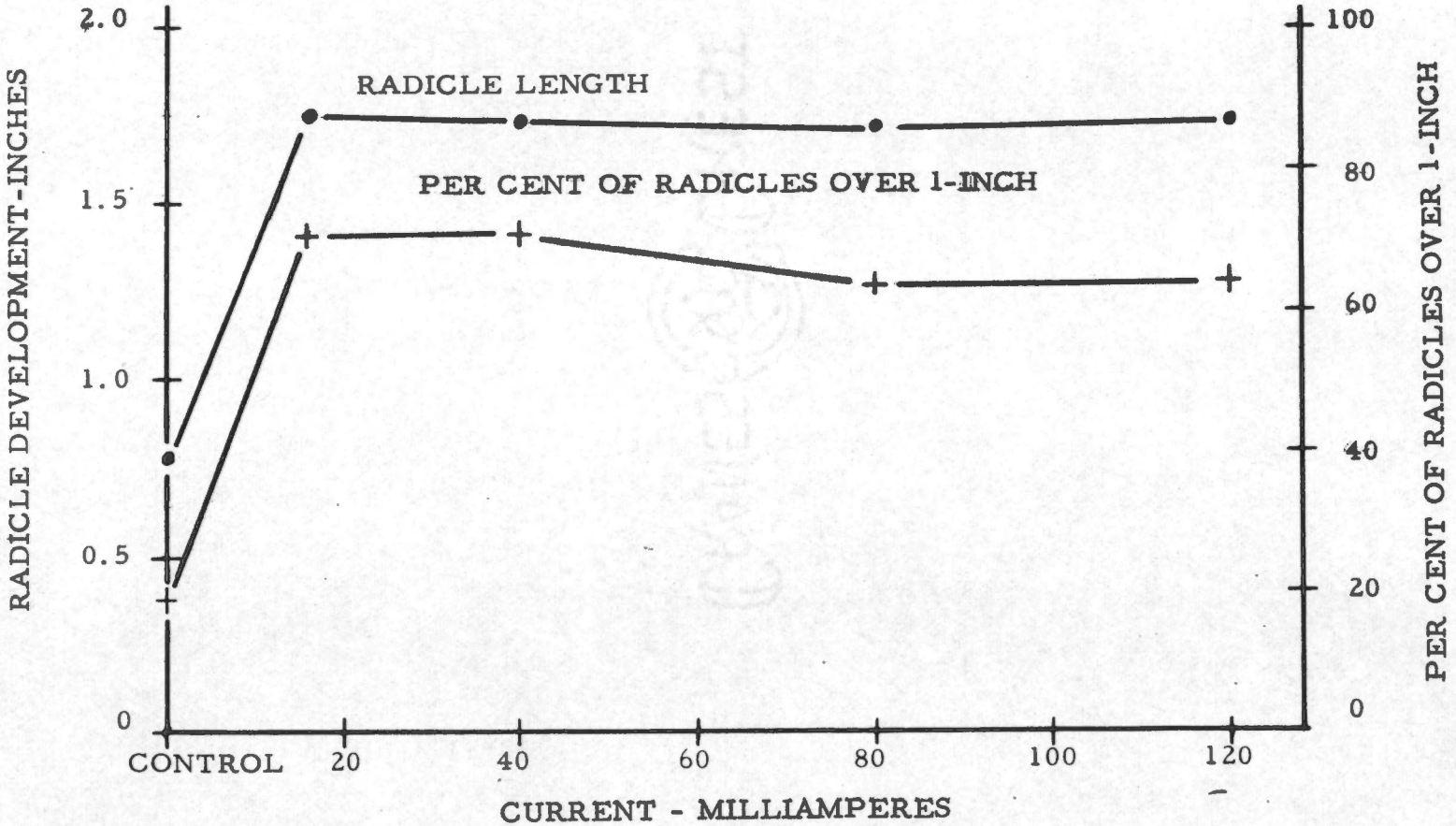


Figure 16. Radicle length and per cent of radicles over one inch as a function of current.

radio-frequency, and gas-plasma irradiations for the reduction of hard-seed in alfalfa was reported by Nelson, et al. (25). The results of the report showed that all three methods of treatments were about equally effective in reducing hard-seed. Heat energy was one common factor in the above methods of radiations. However, radio frequency radiation, infrared radiation or heat and vacuum have not increased the water absorbence of cotton fibers and the linters on cottonseed as have the glow discharge radiation.

The low energy gas-plasma is composed of energies from several regions in the electromagnetic spectrum; including the radio-frequency, infrared, visible light, and ultraviolet regions. The positive column which fills most of the length of the discharge chamber is reported by Cobine (10) to be a typical plasma and is composed of electrons and ions in approximately equal numbers. Some of the energy is lost from the plasma as light and some in electron collisions, but most is lost at the walls of the chamber in recombination of the ions and electrons and in raising the gas temperature. The seeds were not located in the plasma proper at the time of treatment, but were on the wall of the chamber. The upper surface of each seed was exposed directly to the radiation from the plasma; however, the other remaining surface of each seed was shielded from the direct radiation by the chamber wall and other seed. The linters on both sides of the seed absorbed water very rapidly. In view of this observation, it would indicate that the effects noticed on the plasma irradiated cottonseed could be caused by a combination of the energies in the discharge chamber.

An approximation of the minimum energy in the glow discharge can be made on basis of the energy that is required for the first ionization potential of the gas in the mixture with the lowest first ionization potential. The gases in the plasma used in this study consisted of air, gases caused by the outgasing of the seed and rubber

stoppers, and gases that were in the vacuum equipment. Since air was used as the gas medium, nitrogen with a low ionization potential, would be present in the discharge chamber. The energy required for the first ionization potential of nitrogen is 14.53 electron volts (1). This would be the minimum energy in the gas-plasma used in this research.

Based on the data in Table I, page 32, the average energy to which the seed was exposed was estimated by assuming: (1) a unity power factor; (2) uniform radiation from the discharge on the tube wall between the electrodes; and (3) each seed occupied one square centimeter of wall area. The energy was calculated for each current level by using the following formula: $P = (I \times E \times PF)/A$; where I is amperes, E is volts, PF is power factor, and A is square centimeters. The average energy for the 40, 80, and 120 milliampere treatments was 0.058, 0.11, and 0.17 watts per square centimeter or watts per seed respectively. Further investigation and measurements would be necessary to verify these estimations.

The cottonseed as related to germination can be divided into two regions; the micropyle region, the end of the seed where the radicle first appears; and the chalazal region, the large end of the seed. The micropyle region is so constructed that it is usually impermeable to water. The chalazal region allows the necessary moisture for germination to enter the seed, and allows the proper exchange of oxygen and carbon dioxide that is required for germination (33). The faster moisture gets to the seed coat in the chalazal region the faster germination takes place. The linters on the control fuzzy and machine delinted seed do not absorb water very rapidly and thus offer resistance to the entrance of water to the seed coat. Simpson (33) reported that naked cottonseed and cottonseed which had the linters removed with sulfuric acid germinated two to three days earlier than seed with linters.

Webb, Stone, and Pate (43) reported that acid delinted seed which had been irradiated with the gas-plasma germinated no faster than the acid delinted control seed. The degradation of the waxes on the linters of the machine delinted cottonseed caused by the radiation of the gas-plasma allows water to get to the seed coat almost immediately; whereas, several hours are required for moisture to reach the seed coat of the control seed. The availability of moisture to the seed could account for the earlier germination of the irradiated seed over that of the control seed. Earlier germination would cause earlier radicle development. This would not cause an increase in total germination in the treated seed; because as the moisture became available to the control seed, normal germination would occur; whereas, normal germination would occur earlier in the irradiated seed. A decrease in total germination could occur in the irradiated seed if enough water reached the seed coat to block the normal exchange of oxygen and carbon dioxide through the seed coat.(33).

IV. SUGGESTIONS FOR FUTURE INVESTIGATION

There have been very little research reported on the irradiation of gas-plasma as applied to agriculture products. This is a promising field of work and should be explored much more fully. Some future studies in irradiating agricultural seed and plant products with the gas-plasma that might be made are:

1. Current density studies involving irradiation chamber sizes and shapes using the results presented in this thesis as measures of the effects produced.
2. Analyses of the gases in the treating chamber.
3. Separation of the components of the discharge in an effort to find just which or what combinations of energies are causing

the effects noticed in this study.

4. Power and energy determinations throughout the irradiation system.
5. Moisture content studies to show the effects of irradiation with the plasma on seed at different moisture level.
6. Extending the frequency and current range of the treatments.
7. Comparing germinator results with field results.

CHAPTER VII

SUMMARY AND CONCLUSIONS

The purpose of the study described in this thesis was to investigate the effects of radiation from the plasma of a glow discharge on Empire W. R. machine delinted cottonseed. The excitation frequency to the plasma and the current level through the plasma were varied, and the effects caused by these variables were studied. There were five excitation frequencies, ranging from 0.5 to 20 kilocycles, and four current levels, ranging from 20 to 120 milliamperes. The seeds were treated for five minutes at three millimeters of mercury of pressure.

Early germination, total germination, radicle length, and radicle weight were characteristics used to measure the effects of the radiation. Early germination was taken after two days and total germination after four days in the germinator. After total germination was obtained, the radicles were measured, then removed from the seed and dried. This dry weight was used as the radicle weight. The per cent of the seed that germinated and had radicles of one inch or longer was calculated for each treatment.

A significant increase in early germination was obtained at all radiation levels over that of the controls. The per cent of germination of the irradiated seed ranged from 48.5 to 67 per cent with an average of one per cent for the control seed. There was no significant increase in total germination of the irradiated seed over that of the control seed.

No significant differences in early germination or radicle development were caused by varying the frequency or current over the range of treatments. There was a significant increase in radicle length, radicle weight, and the per cent of radicles over one inch of all

irradiated seed over that of the control seed. The radicle length and weight of the irradiated seed were about twice as great as those of the control seed. The per cent of radicles over one inch was about three times greater for the irradiated seed as those of the control seed. All treatments were effective in producing early germination and early radicle development.

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APPENDIX

APPENDIX A

DEFINITION OF TERMS

A. - Angstrom Units

rep. - Roentgen equivalent physical

R. - Roentgen

P. - Power

I. - Current

E. - Volts

PF. - Power Factor

A. - Area

db. - Decibel

Ma. - Milliamperes