was as follows: To an acidic solution of 15 ml of water were added 0.006437 moles (4.0000 g) of thallic chloride and 0.01288 moles (3.4970 g) of mercuric chloride. The solution was stirred and then subjected to vacuum desiccation over concentrated sulfuric acid for a period of 24 hr.

In this preparation, as in all the others, the ratio of 2 moles of thallic chloride to 1 mole of the metallic chloride was utilized in deciding what weights to use.

The results were successful, yielding the two corresponding double salts. Both were very hygroscopic and as a result were kept in a vacuum desiccator or in an airtight container. The octochlorodithallate of barium was white in color and formed monoclinic crystals. Its formula is $2TlCl_3 \cdot BaCl_2 \cdot 6H_2O$. Monoclinic crystals were also very noticeable in the white double salt of divalent mercury. Its formula was proved to be $2TlCl_3 \cdot HgCl_2 \cdot 12H_2O$.

Although these new double salts of thallium may not have any immediate value, it is hoped that they will eventually serve to disclose more of the chemistry of the "duckbill" of the elements.

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## Effects of High-Voltage Cathode-Ray Irradiation on Cottonseed

**M. G. Lambou,[1] R. Y. Mayne, B. E. Proctor, and S. A. Goldblith**

*Southern Regional Research Laboratory,[2] New Orleans, Louisiana, and Food Technology Laboratories, Massachusetts Institute of Technology, Cambridge*

High voltage x-rays and cathode rays have been shown to destroy pure cultures of molds and bacteria at minimum dosages of 1,000,000 and 1,500,000 roentgen-equivalents-physical (rep), respectively (*1*). This information suggested a practical application of ionizing radiations in the sterilization of foods and possibly seeds. Ground beef (*2*), fish slices (*3*), and other foods (*4*) have been irradiated, with the desired results. The reports on these foods indicated, however, that at dosages necessary for sterilization, enzymes in foods are slightly affected.

The effects of ionizing radiations have been recorded for many species of seeds. When applied to wheat (*5*), tomato (*6*), and mung beans (*7*) excessive irradiation has resulted in injury manifested as reduced and delayed germination, inhibition of growth, and, in some instances, complete loss of viability. Similar unfavorable reactions were observed when cottonseed was first irradiated with x-rays in 1933 (*8*). Since then, the sterilizing effects of combined ultraviolet and infrared rays have been noted (*9*).

This paper describes the adverse effect of high-voltage cathode rays on germination and growth of cottonseed when these rays are applied in dosages of sufficient strength to destroy associated microorganisms. Cathode rays were produced by a pressure-insulated, electrostatic generator of the Van de Graaff type, operating at 3 mev and capable of emitting a continuous supply of monoenergetic electrons in a magnetically focused beam (*10*). The dosages of cathode rays are expressed in terms of rep as described by Evans (*11*).

Two varieties of cottonseed were used in these investigations: a Stoneville 2B variety, crop year 1949, of 16.7% moisture and 2.24% free fatty acids contents; and a Delfos 651 variety, crop year 1948, of 11% moisture and 0.4% free fatty acids contents. The lot of Stoneville 2B seed was subdivided into 7 samples which were heat-sealed in polyethylene tubing. One sample was set aside as the control, and the remaining 6 received dosages of high-voltage cathode rays ranging from 500,000 to 3,000,000 rep in increments of 500,000 rep. The Delfos 651 variety was subdivided into 9 samples. One unirradiated sample was used as the control. The 4 remaining pairs of samples were irradiated at increasing dosages of cathode rays as follows: 500,000; 1,000,000; 1,500,000; and 2,000,000 rep, respectively. All treated lots of seed were irradiated in a single layer—the thickness necessary for effective exposure with a single dose. Irradiation was conducted from one side only.

The effects of ionizing radiations on cottonseed were measured in terms of moisture and free fatty acids contents, total and internal microbial populations, germinability, and growth. Methods used for the determinations of moisture and free fatty acids contents were those recommended by the American Oil Chemists' Society (*12*). The numbers of microorganisms per gram of whole seed were determined as published previously (*13*), except that the plating medium used for the growth of bacteria was tryptone glucose extract agar. Except where otherwise indicated, internal microflora were observed by planting 200 acid-delinted surface-sterilized (0.1% $AgNO_3$) seeds from each sample on a modified Czapek's medium and recording the percentages of seeds showing visible evidence of the growth of microorganisms. In addition, the numbers of seeds germinating on the agar were noted. Separate germination tests were made according to the standard method (*14*); counts were taken on the sixth and ninth days, no additional sprouts being observed after the sixth day. Growth measurements of the sprouts were recorded as their lengths in millimeters at 9 days.

When applied to the Stoneville 2B variety of cottonseed, high-voltage cathode rays at 1,500,000 rep inactivated the microflora in the seeds (Table 1). Ex-

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[2] One of the laboratories of the Bureau of Agricultural and Industrial Chemistry, ARA, USDA.

TABLE 1

EFFECT OF HIGH-VOLTAGE CATHODE RAYS ON INTERNAL INFECTION OF STONEVILLE 2B COTTONSEED

| Dosage of cathode rays (rep) | Seeds* infected with | | | Total infected (%) |
|---|---|---|---|---|
| | Molds (%) | Bacteria (%) | Molds and bacteria (%) | |
| None | 58 | 0 | 36 | 94 |
| 500,000 | 92 | 4 | 4 | 100 |
| 1,000,000 | 36 | 0 | 4 | 40 |
| 1,500,000 | 0 | 0 | 0 | 0 |
| 2,000,000 | 0 | 0 | 0 | 0 |
| 2,500,000 | 4† | 0 | 0 | 4† |
| 3,000,000 | 4† | 0 | 0 | 4† |

* Samples of 50 seeds each were used for the analyses, 5 seeds/plate.

† Probably a plate contaminant; molds were not of the same genera as appeared on the other plates.

posure to ionizing radiation at the dosages used did not increase the quantity of free fatty acids present or reduce the moisture content. It was impossible to gauge definitely the effect of cathode rays on viability, because of the extremely poor quality of this seed.

The effect of increasing dosages from 500,000 to 2,000,000 rep of high-voltage cathode rays on the Delfos 651 variety of cottonseed of good quality confirmed the data obtained for the Stoneville 2B seed with respect to microorganisms, free fatty acids, and moisture. Because few microorganisms were originally present in the Delfos lot, a dosage of 1,000,000 rep was successful in reducing the total count from approximately 1500 to less than 10 microorganisms/g whole seed. The lowest dosage—i.e., 500,000 rep—appeared to have destroyed the internal microflora in the few seeds found infected (1%).

Germinability decreased regularly as the dosage of ionizing radiation increased from 500,000 to 1,500,000 rep. Irradiation with 2,000,000 rep destroyed viability in all the seeds (Table 2). They did not germinate on agar at room temperature or between moistened towels at alternating temperatures of 20° and 30° C. Trends for germination on agar and rolled towels were similar though slightly higher on the agar because of the additional ions of nutritive value present in the medium (*15*).

TABLE 2

EFFECT OF HIGH-VOLTAGE CATHODE RAYS ON VIABILITY OF A DELFOS 651 VARIETY OF COTTONSEED

| Dosage of cathode rays (rep) | Germination count | |
|---|---|---|
| | On agar* (%) | On rolled towels† (%) |
| None | 47.0 | 66.7† |
| 500,000 | 51.5 | 47.2 |
| 1,000,000 | 17.0 | 8.5 |
| 1,500,000 | 3.8 | 0.0 |
| 2,000,000 | 0.0 | 0.0 |

* Acid-delinted seed, surface-sterilized with 0.1% aqueous $AgNO_3$; 200 seeds/sample planted on Czapek's solution agar containing 0.1% $KH_2PO_4$ instead of $K_2HPO_4$ and 1% dextrose instead of sucrose, pH 5; stored at room temperature. Germinated seedlings were counted at 8 and 12 days; no change in count occurred after 8 days.

† Standard germination test: 4 × 100 seeds used except where value is marked †; in this case only 75 seeds were available.

All seedlings shown in Fig. 1 were selected as representative of the average length of the sprouts after 9 days. Growth of the seedlings was partly inhibited by the lowest dosage of high-voltage cathode rays, 500,000 rep, and completely inhibited by 1,500,000 rep (Fig. 1, seedlings 2 and 4, respectively). Fig. 1 (4 and 5) are dehulled seeds that had received dosages of cathode rays at 1,500,000 and 2,000,000 rep, respectively. These seeds had obviously imbibed water, were swollen, but did not germinate. They were typical of all the sound-appearing seeds on the towels (standard germination test).

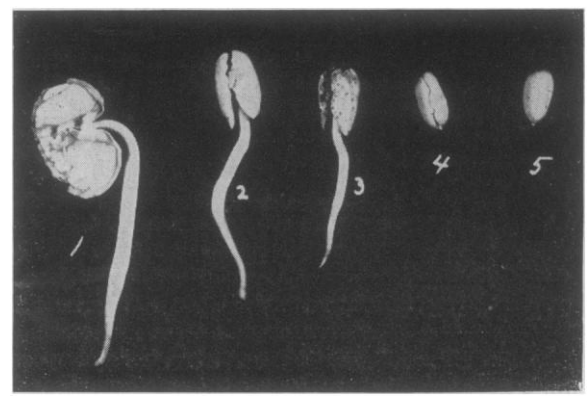


FIG. 1. Nine-day-old sprouts from cottonseed exposed to increasing dosages of high-voltage cathode rays, as follows: 1—none; 2—500,000 rep; 3—1,000,000 rep; 4—1,500,000 rep; and 5—2,000,000 rep.

As in the case of other materials, irradiation of cottonseed with high-voltage cathode rays significantly reduced the numbers of microorganisms present on and in the seed. This was accomplished without bringing about any changes in the moisture and free fatty acids contents of the seed, but was accompanied by a reduction in germination count and an inhibition of growth. The data showing increasing inhibition of germination and growth of cotton seedlings with increasing dosages of high voltage cathode rays—a pattern of response demonstrated by cottonseed and other seeds exposed to x- and γ-rays—suggest that one or more enzymes associated with the normal phenomena of germinating and growing have been inactivated by the rays.

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# Influence of Nutrition upon Appearance of Tumors in *Tu$^{50j}$* Stock of *D. melanogaster*

Sidney Mittler

***Department of Biology,***
***Illinois Institute of Technology, Chicago***

*Tu$^{50j}$* is a recessive mutation which appeared upon inbreeding Chicago wild *D. melanogaster,* and it contributes to the production of melanotic growths in the abdomen of the fly. Recently Burdette (*1*) has shown that *tu$^{48}$* reared on a medium deficient in yeast produced fewer tumors than those raised on re-yeasted medium, and since then Begg and Robertson (*2*), in an attempt to produce a completely synthetic medium for *D. melanogaster,* have reported that an unknown alkali-soluble factor from yeast must be present for growth. Hence, to determine the penetrance of *tu$^{50j}$* the influence of nutrition must be controlled. To be certain that all the nutritive factors (*2*)—chiefly amino acids and vitamins—came from the yeast and not the medium (usually cornmeal-molasses), a survey was made of yeasts that could live on a vitamin- and amino acid-free medium, as shown in Table 1. Included in this medium are the essential elements with which some of the yeasts are able to synthesize their own protoplasm. It is primarily a glucose and ammonium sulfate medium, plus several trace elements.

Twenty-seven species of yeasts from 14 genera were tested for growth on the vitamin- and amino acid-free medium (Table 2); 12 were found capable of living on the minimal medium. The yeasts were transferred twice onto vitamin- and amino acid-free medium to make sure that none of the nutritive ingredients were carried over from the complete medium in which the yeasts were originally grown. To prevent overcrowding and to maintain a standard of comparison, a pair of flies from a highly inbred stock of *tu$^{50j}$* were placed on the minimal medium inoculated with yeast. It is believed that practically all the nourishment that the larvae obtained came from the yeast on the medium. The penetrance of *tu$^{50j}$* reared on yeasts that do require vitamins and amino acids on a minimal medium is presented in Table 3. It is quite evident from the results that *tu$^{50j}$* reared on yeasts growing in the minimal medium produced fewer tumors than those in *tu$^{50j}$* reared on *S. cerevisiae,* the yeast most commonly used in Drosophila work, grown on the cornmeal-molasses medium, for *S. cerevisiae* cannot grow on the minimal medium (Table 2). Hence, the larvae in the control series obtained some nourishment from the medium, for the substances essential for the growth of Drosophila are present both in the yeast and in the medium. There are probably as many variations in medium used to rear *D. melanogaster* as there are research workers who study them. It is difficult to separate the effect of the medium and *S. cerevisiae* upon the penetrance of *tu$^{50j}$*. However, one can compare the penetrance of the tumor gene when the flies

TABLE 1

VITAMIN- AND AMINO ACID-FREE MEDIUM

| Ingredient | Amount |
|---|---|
| Agar | 15 g |
| $C_6H_{12}O_6$ | 30 " |
| $KH_2PO_4$ | 1 " |
| $NaKC_4H_4O_6$ | 8 " |
| $(NH_4)_2SO_4$ | 2 " |
| $CaCl_2$ | 0.5 " |
| NaCl | .5 " |
| $MnSO_4$ | .5 " |
| $MgSO_4$ | .5 " |
| $FeSO_4$ | 0.5 " |
| $H_2O$ | 1000 ml |

TABLE 2

ABILITY OF YEASTS TO LIVE ON COMPLETE MEDIUM AND ON VITAMIN- AND AMINO ACID-FREE MEDIUM

| Yeast | Vitamin- and amino acid-free | M–Y Complete medium |
|---|---|---|
| *Candida monosa* | – | + |
| *C. pseudotropicalis* | – | + |
| *C. mesenterica* | – | + |
| *C. sorbosa* | + | + |
| *Endomyces magnusii* | + | + |
| *Hansenula anomala* | + | + |
| *H. saturnus* | + | + |
| *Rhodotorula sanniei* | – | + |
| *R. aurantica* | – | + |
| *R. gracilis* | + | + |
| *R. glutinis* | + | + |
| *R. suganii* | – | + |
| *Pichia membranaefaciens* | + | + |
| *Debaryomyces globosus* | + | + |
| *D. membranaefaciens* | + | + |
| *D. disporus* | – | + |
| *D. matruchoti* | – | + |
| *D. guilliermondii* | – | + |
| *Kloeckera apiculata* | – | + |
| *Zygosaccharomyces lactose* | – | + |
| *Schizosaccharomyces versatilis* | – | + |
| *S. fragilis* | – | + |
| *Torulopsis utilis* | + | + |
| *Nadsonea fulvescens* | + | + |
| *Endomycopsis fibuliger* | – | + |
| *Geotrichium* | + | + |
| *Saccharomyces cerevisiae* | – | + |