GROWTH IRREGULARITIES IN HYBRID FREESIAS INDUCED BY X-RAYS

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In recent years much attention has been given to the use of X-rays and radium in the production of new plant and animal types through the effect of these potent rays on the genetic mechanism. In order to test the effect upon Freesias, seeds, flower buds and corms of a recently introduced commercial variety known as Elder's Giant White were subjected to X-rays. The choice of this variety for experimentation was due to the fact that much data concerning its genetics and cytology had already been compiled. Although it is too early to report any observed effect upon the heritable characters of this plant it seemed sufficiently interesting to present some observations made upon irregularities of growth induced by the X-ray exposures.

Corms of similar size and form were carefully selected and these placed in groups of five in small paper bags. Since the corms were of average size (slightly less than five-eighths inch in diameter) it was possible to fold the bags so as to keep the material to be treated in a single layer. In this condition these small parcels of corms were arranged beneath the X-ray tube. The apparatus used was a modern machine normally employed in X-ray therapy and was operated by a professional technician.[1] The first lot of ten groups was placed at twenty inches from the target and exposed in two-minute intervals up to twenty minutes at 105 K.V.—30 M.A. with a 1 mm. aluminum screen. In the second lot no screen was used and the exposure was in three-minute intervals up to twenty-one minutes with a distance from the target of sixteen inches at 105 K.V.—30 M.A. Each group of five corms was planted in a six-inch clay pot and at the same time untreated controls were planted under identical conditions. Careful cultural attention was given throughout the entire growing season. The greenhouse night temperature was held at about forty-eight degrees F. with the usual rise during warm, sunny days.

Due to an irregularity in the storage of the corms from which lot number one was chosen it was decided to abandon this data entirely, therefore the following observations refer only to plants produced by the second lot of corms. This lot was planted on November 15, 1930, and was examined daily. The following representative record of growth was that taken on December 22nd. The measurements of plants were taken from the surface of the soil to the top of the highest leaf.

[1] The author is indebted to Dr. W. E. Pennington for his assistance and in the use of his X-ray equipment. The intensity of the dosages was based upon the helpful suggestions of Dr. A. F. Blakeslee of the Carnegie Institute of Washington.

Control pot: all five showing with heights varying from 3/16 to 1¼ inches.

Pot No. 1 (3 min. exposure): three plants up one 5½ inches, one 4½ inches and one ⅝ inch.

Pot No. 2 (6 min. exposure): due to an injury to one of the corms only four were planted. Two plants up, both 2½ inches.

Pot No. 3 (9 min. exposure): four plants up, two 1¼ inch, two ⅝ inch.

Pot No. 4 (12 min. exposure): four plants up, all about ⅜ inch.

Pot No. 5 (15 min. exposure): four plants up, one ⅞ inch, three ⅜ inch.

Pot No. 6 (18 min. exposure): three plants up, two ¼ inch, one ⅛ inch.

Pot No. 7 (21 min. exposure): two plants up, both 3/16 inch.

Final germination records showed that all corms grew with the exception of those in pots No. 6 and No. 7. In these the final count showed four corms grew in No. 6 and only two in No. 7 Figure one shows the relative growth of this lot and its control on March 28, 1931. As indicated in the size of the buds of No. 1 and No. 2 these pots showed mature blossoms a few days before those on the control plants.

One item that attracts attention in figure one is the greater number of plants in pots numbers 1, 2 and 3 than in the control (N), in spite of the fact that all corms were carefully graded as to size. In the control each corm produced a single plant while as many as five "shoots" were produced by a single X-rayed corm. Although it is not unusual for large corms of Freesia to occasionally produce two or three shoots it was demonstrated by many pots planted from the same stock which supplied the experimental material that only seldom did a corm produce more than a single plant. Some explanation of the increase in number of shoots may be gained from the examination of a Freesia corm with its husk removed. Figure two shows top and side views of a whole corm similar to those used, also a bissected corm. In each of these sketches is shown the buds from which grow the "shoots" just mentioned. Usually the top one germinates while the others are abortive. A careful ex-

Fig. 1. Growth produced by X-rayed corms compared with the control N. Intensity of dosage was graduated from No. 1 to No. 7. Note the increase in number of plants produced by the corms of the lower X-ray dosage.

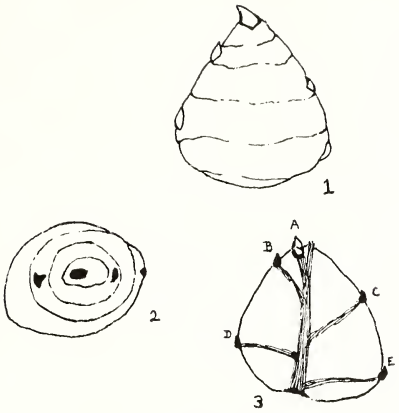

Fig. 2. Sketches of normal Freesia corms. No. 1 is lateral view with husk removed to show primary and axillary buds. Upper view of same corm is shown at No. 2 while No. 3 is the corm bissected to show the above features.

amination showed that not only did this top bud grow in the X-rayed corms but the lateral buds were also stimulated into growth. That this was not alone due to the injury to the primary bud was demonstrated by purposely mutilating corms in removing this upper bud. Such corms seldom produced more than a single shoot from their lateral buds.

As indicated in the early growth records the X-rayed corms that started to germinate immediately after planting made more rapid

Fig. 3. A shows normal leaf besides one taken from a plant produced by an X-rayed corm. B shows the taller plants in the control compared with plants from corms X-rayed nine minutes. Note increase in *number* of plants in the pot containing the X-rayed bulbs.

progress than the controls. It is quite possible that a lower dosage would have given an accelerated growth without noticeable injury. Just what effect X-ray dosage would have upon the corm when it was in the first part of the dormant period has not been determined. As shown in figure one the higher dosages had a distinct retarding effect which very nearly reached the lethal point in No. 7 (XR 18).

In figure 3B the plants from the X-rayed corms lack the upright growth and straight leaves characteristic of this variety as shown on the control (N). Figure 3A is a close-up view of two leaves. These were taken from the plants shown in figure 3B. The plant from pot No. 3 was suggestive of "crepe" cloth or paper and this condition involved the structure of leaf, stem and flower. The condition of the leaves in the pots where the corms had a higher dosage were similar except in the higher degree of deformity while those of the lower dosage were more nearly normal. The irregularity of texture was accompanied by a disturbance in the distribution of chlorophyll producing light and dark areas. As a result of these irregularities in growth the leaves and stems became curled and twisted, and the flowers split and deformed. In spite of these conditions seed was set on the plants in all pots except No. 7 which produced very little more growth than shown in figure 4A.

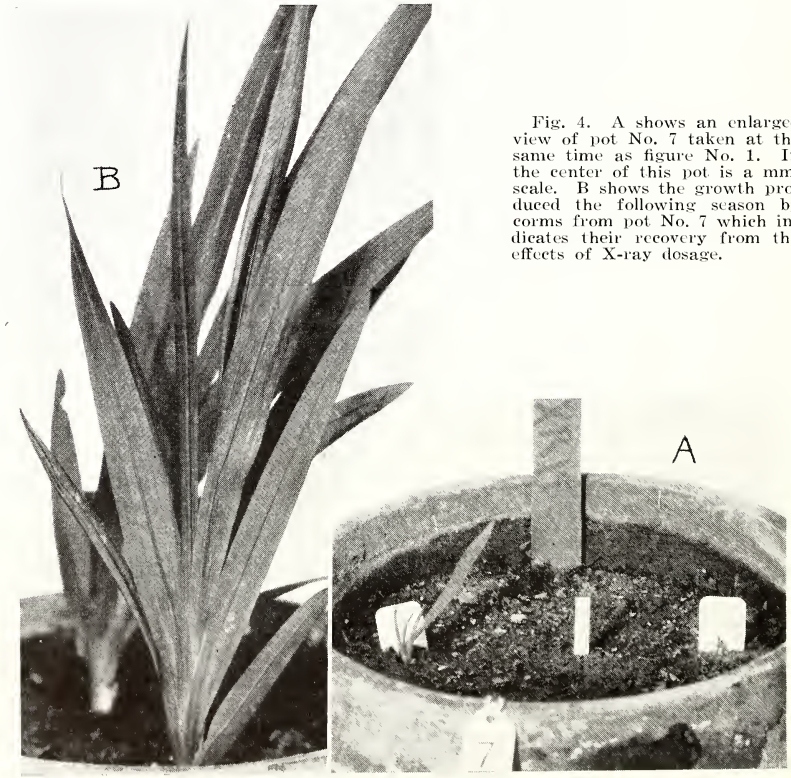

Fig. 4. A shows an enlarged view of pot No. 7 taken at the same time as figure No. 1. In the center of this pot is a mm. scale. B shows the growth produced the following season by corms from pot No. 7 which indicates their recovery from the effects of X-ray dosage.

All plants from the X-rayed corms and their control were kept carefully covered and their blossoms self-pollinated. A record of the seed production, which is never high in this variety, follows: Control pot, 120; pot No. 1, 225; pot No. 2, 186; pot No. 3, 84; pot No. 4, 157; pot No. 5, 58; pot No. 6, 5. The germination of these seeds appeared to be normal, although a record of their growth will not be complete until after the spring of 1933 since seedlings of this variety seldom bloom the first season. To date none of the tiny seedlings show the growth irregularities present in the parent plants.

The resulting corms and cormlets produced in each of the pots described above were carefully saved and planted early in the autumn of 1931, and have made nearly normal growth. Some of the first leaves from the corms of pots No. 5, No. 6 and No. 7 showed some "creping" and curling but this condition has practically disappeared in the later leaves. Figure 4B shows the plants resulting from the two small corms of pot No. 7. This indicated nearly a complete recovery from the effect of the X-ray treatment which was severe enough to kill three of the five corms planted in pot No. 7. Whether any effect will be noted in later growth or blooming of these plants is not known. (Photograph used in 4A was made December 18, 1931).

In addition to the X-raying of the corms both seeds and developing flower buds were subjected to treatment with these rays. In 1930 three lots of seed saved from self-pollinated plants of Elder's Giant White were exposed ten, sixteen and twenty minutes respectively at 105 K.V.—30 M.A., twenty inches from the target with a 1 mm. aluminum screen. These germinated normally with no irregularities noted during the growing season. During the first part of the present season (fall 1931) several plants resulting from corms produced by these seedlings showed leaf irregularities similar to those described above. This condition was found only on plants from the seed that had been exposed sixteen and twenty minutes. It is probable that higher dosage would have produced a pronounced effect uon the seedlings. No explanation can be offered for the irregularities being absent in the seedlings but appearing in plants from corms produced by the seedlings, except that the affected individuals may have been overlooked since the whole lot was quite frail due to the late planting date.

On February 28, 1930, four pots of Freesia plants in bud were exposed four, seven, ten and thirteen minutes respectively at 105 K.V—30 M.A. The flower buds were placed twenty-five inches from the target. A 1 mm. aluminum screen was used. No effect on later growth or blooming was noticed, however the young plants resulting from their seed (by self-pollination of their blossoms) showed leaf deformity similar to that resulting from the X-rayed corms. When the corms produced by the above seedlings were planted this season (fall 1931) they showed only a few of the first leaves to have the characteristic "creping" and deformity. Later leaves appeared to be normal.

Since no histological study was made of the affected plants it was not certain what effect upon the microscopical structures had been produced by exposure to the X-rays. The appearance of the affected leaves

suggested that there was a disturbance in the rate of growth in localized areas, and in the distribution of the chloroplasts or the formation of chlorophyll. The general effect of X-ray treatment in low dosage seemed to result in an acceleration in the rate of growth and in the stimulation into growth of structures that otherwise would have become abortive. With the increase in intensity of the dosage the acceleration was lost and the rate of growth became retarded, approaching the lethal point as in pot No. 7. That the Freesia plant was able to recover from this disturbance of its somatic structure seemed to be indicated when figure 4A and B were compared. Just what this reorganization was that took place during the formation of the corm and subsequent growth after a period of dormancy is not known. No effort has been made in the present paper to deal with any genetic disturbance resulting from the effect of X-rays on the germinal substance of the Freesia.