## Radiographing as a Method of Observing Some Seed Characters in Monogerm Sugar Beet Fruits' G. J. HOGABOAM<sup>2</sup>

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A sugar beet fruit is made up of a lignified and suberized mass within which are one to several seeds. Seed examination has been a problem since time-consuming dissection of the fruits has been necessary. The X-ray techniques developed to detect insect infestations in wheat (2,3,4) " proved to be an answer to the seed examination problem for many types of research. Wiksten (5) recently reported the use of X-ray to study stratification and seed quality in Scotch pine and Norway spruce.

In 1955, E. F. Patton of the General Electric Company's' X-ray department provided a radiogram of 3 lots of sugar beet fruits, two multigerm (many-seeded) and one monogerm (one-seeded). Since sugar beet seeds are oblate spherical and are oriented in various planes within the multigerm fruit, the size and shape of all seeds cannot be determined with a single radiograph. However, Grimm (1) in 1958 by X-ray examination was able to predict rather accurately the subsequent germination of whole and segmented multigerm fruits. By mounting the monogerm sugar beet fruits with the "seed cap" down, the seed of all fruits lie in one plane making possible a study of seed characters in the radiograph (Figure 1). This technique was of value in comparisons of seed and fruit size and their consequent effect on the speed of germination<sup>a</sup>.

Seeds from each of 19 plants of a monogerm variety were studied. Since monogerm fruits are approximately oblate spheroid, they were screened through a round-hole screen for fruit diameter and then through a slotted screen for fruit thickness. Categories were made by plant source for fruit diameter and thickness in preparation for radiographing. Since the required X-ray exposure might vary with fruit thickness, fruits with similar thicknesses were mounted on the same template for radiographing. The templates 13" x 16", were prepared from 1/8" tempered fiberboard by cutting 48 - 1" x 13/," openings. Strips of cellophane tape were placed on the lower side of the template across the

<sup>3</sup> Trade and company names are mentioned for identification only and do not constitute recommendation by United States Department of Agriculture.

A manuscript is in preparation by Hogaboam, G. J. and F. W. Snyder on influence of size of fruit and seed on germination of a monogerm sugar beet variety.

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<sup>&</sup>lt;sup>8</sup> Numbers in parentheses refer to literature cited.



Figure 1.—Portion of the original radiograph of monogerm sugar beet fruits. An empty fruit containing no viable seed may be seen in the lower left corner. A fruit containing two seeds is marked by the arrow. Light areas represent resistance to X-ray penetration, thus the seeds appear light. The ligneous inner layer of the seedballs is outlined as light at the edges where the layer is vertical. This is especially noticeable with empty seedballs.

openings. The fruits to be radiographed were then mounted in rows on the tape in these openings. As a check on distortion and as a standard for measurement, size gauges were placed in three random positions on each template.

Because a very soft X-ray is needed to differentiate the seed within the sugar beet fruit and show details of seed structure, General Electric's low-voltage X-ray unit, model LC-90,<sup>4</sup> was used. Very good results<sup>6</sup> were obtained on 14" x 17" Industrial type M film using 3 and 5 milliamperes, 15 and 25 K.V.P., with exposures varying from 11/2 to 21/2 minutes, with a 30" focal film distance.

The template holding the fruits was placed in contact with the film for radiographing. The duration of radiographic exposure depended upon the detail desired. Within limits, the greater the duration the greater the detail within the seed in the radiograph. Since the outer portions of the sugar beet fruit are suberized and the inner portions surrounding the seed lignified, a shorter exposure would show the suberized material of the fruit at the expense of seed detail.

The true-seed diameter (Figure 2) can be measured either by projection of the radiograph or by a direct measurement of the contact film. When the projection method is used, a size gauge must also be radiographed and projected to give a basis for measurement. Direct measurement of the radiographed size gauges revealed very little distortion. Since dark film from long exposure does not project well, a relatively short exposure must

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be used at the expense of some seed detail (Figure 3). Each template opening was cut from the film and mounted in  $2'' \ge 2''$  slides for projection. Projected size gauges and seeds were then compared.

The direct measurement method is preferred since darker radiograms with more detail (Figure 4) can be used. With this method, a mirror is used to reflect bright light up through the radiograph and the seed diameter is measured directly with a size gauge.



Figure 2.—Radiograph of excised true seeds of sugar beets showing differences in shape. Negative reproduction.



Figure 3.—Radiograph of monogerm sugar beet fruits made with an X-ray exposure of 1½ minutes at 3 M.A. and 15 K.V.P. A film with this exposure is suitable for projection. More details of the fruit are visible but details within the seed cavity are difficult to observe. Negative reproduction.



Figure 4.—Radiograph of monogerm sugar beet fruits made with an X-ray exposure of 2 minutes at 5 M.A. and 25 K.V.P. Such a radiograph is suitable for direct observation over a bright light. Only the most lignified portions of the fruit are visible but details of the seed are clearly visible. Note the empty fruit in the third row from the bottom, fourth from the left. Fruits containing two seeds are located on the left side in the top and bottom rows. The third and fifth fruit from the left in the third row from the bottom contain bump-like growths in addition to a viable seed within the cavity.

The germination performance and seedling appearance indicated that the exposure of the dry seeds to the roentgen level employed had no perceptible deleterious effects.

Some of the observations follow:

- 1. Seed from each plant source examined contained some seedless fruits. One lot had as few as 2% seedless fruits whereas another had as many as 35%.
- 2. Sixteen of the 19 monogerm plants had some fruits which contained 2, 3, or 4 seeds in the seed cavity. The percentages of such fruits varied from 0.3 to 23.8.
- 3. A few twin embryos were found.
- 4. Occasional bump-like growths occurred in the seed cavity along with a well-developed seed (Figure 4).
- 5. The shape of the seed varied from oblong to round, when viewed from the top (Figure 2).
- 6. The perisperm of some seeds was not fully developed although the embryo had developed normally.
- 7. One seed cavity contained a fully developed perisperm but an underdeveloped embryo.
- 8. When mounting the fruits on the templates, striking differences between plants were noted in the size of the openings (vascular trace scars) into the seed cavity at the point of attachment of the fruit.

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