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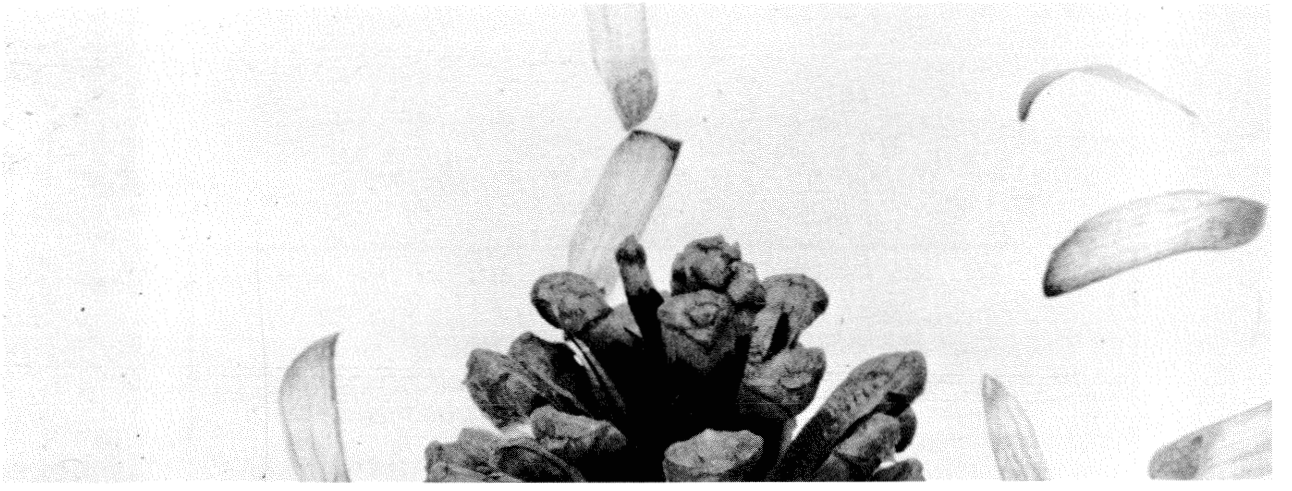

CONE ANALYSIS OF SOUTHERN PINES A GUIDEBO

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USDA-Forest Service
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and
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PREFACE

The information in this publication was presented at two Cone Analysis Workshops organized by Dr. Earl W. Belcher, Jr., Director of the Eastern Tree Seed Laboratory, during November 1976 at Macon, Georgia. These workshops were presented by personnel from the Southeastern Area, State and Private Forestry, and the Southeastern Forest Experiment Station. The authors of this workbook are those who developed the workshop and conducted training sessions.

This guidebook contains the information essential for cone analysis and interpretation of the results. Cone Analysis Service (CAS) is also available at cost from the Eastern Tree Seed Laboratory at Macon, Georgia.

CONE ANALYSIS OF SOUTHERN PINES
A Guidebook

by
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I. INTRODUCTION

Southern pine tree improvement programs require an ample supply of improved **seeds,**[1] but production from so pine **seed orchards** has often been disappointing. If high production is to be maintained, yields must be monitore causes of seed losses must be identified. Techniques for determining seed efficiency were first used for red pine, *resinosa* Ait., by Lyons (**1956**). Bramlett (**1972**b, **1974**) modified and further developed a procedure, known as **analysis,** for evaluating production efficiency in southern pine seed orchards.

Cone analysis provides information needed to evaluate seed production and seed orchard management. Actua yield of individual **cones** is compared to the potential seed yield. Productivity can then be expressed in terms of se ficiency. One can determine in which stages of seed development certain losses occur, and the types of seed failures identified and quantified.

This guidebook outlines the basic cone-analysis procedure, the factors involved in losses of seed, and the interpre and value of the results. It is a stepwise guide for those who wish to conduct their own cone analyses. More detail cussions may be found in the publications listed in the Literature Cited section. Tree improvement workers who want to analyze cones themselves can obtain this service at cost from the Eastern Tree Seed Laboratory. This guic will help them understand the analytical results they receive.

[1]Terms in boldface type are defined in a glossary at the end of this guidebook.

II. CONE AND SEED DEVELOPMENT AND MORPHOLOGY

A basic knowledge of cone and seed development and morphology is essential to understand cone analysis.

Pine trees produce two types of **strobili**: male or pollen-producing **catkins** and female or ovule-producing c primordia of the **female cones** are initiated in the summer. The following spring **female flowers** emerge in an e tion from protective bud scales on the tips of new shoots (fig. 1A). The female flowers are composed of many s scales spirally attached at right angles to a central axis. Pairs of **ovules** originate as small protuberances at the scales (fig. 1B). Not all scales produce functional ovules; only those in the central region of the cone have the p produce ovules and eventually seeds. These are called **fertile scales.** The lower scales and those at the tip of the infertile and never bear seeds **(infertile scales).**

At the time of **pollination,** a single layer of cells **(integument)** covers the ovule. Pollination occurs when t **grains** are carried by the wind from the male catkins to the female flowers. Pollen grains enter the ovule (fig. 1C an opening (the **micropyle)** in the **seedcoat.** Once in contact with the ovule, pollen grains germinate and **pol** grow from the pollen chamber into the ovule tissue **(nucellus)** (fig. 1D). The pollen tube develops slowly durin growing season, and the winter is over before it reaches the mature egg cell of the ovule. For more than 1 ful ovule remains only a fraction of the size of a fully developed seed. During the year after pollination, the female fl called **conelets.**

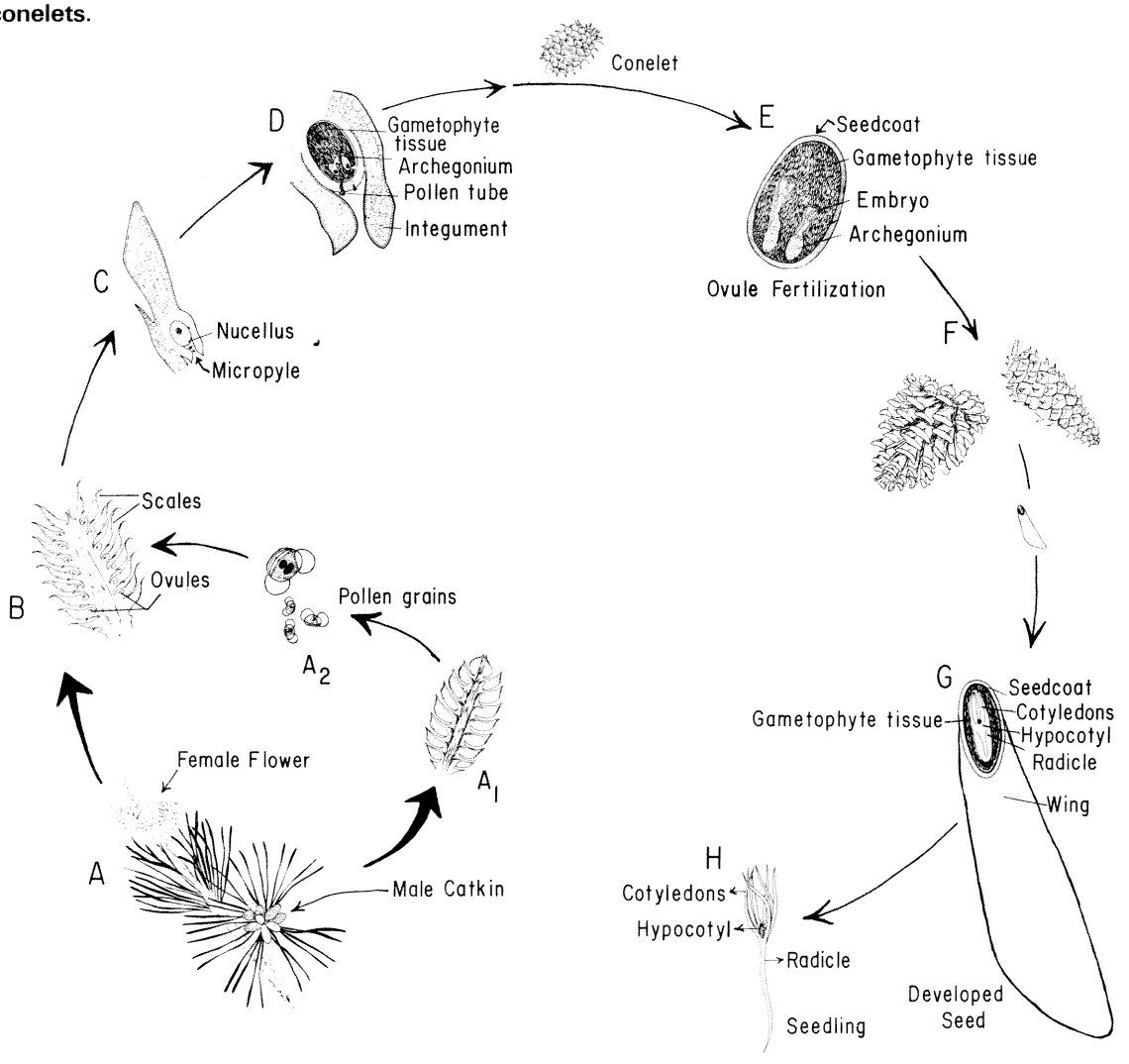


Figure 1.–Pine seed formation. Drawing adapted from one provided by Carolina Biological Supply Company.

When vegetative growth starts in the second spring, the ovule also resumes development and enlarges. From 12 tc months after pollination, depending on pine species, the pollen tubes grow far enough into the ovule to reach special structures **(archegonia)** containing the egg cells (fig. 1E). Sperm cells from the pollen tubes fertilize the eggs, and emb (2N tissues) form. The gametophyte tissue surrounds the embryo and provides the nutritional reserves for the seed tissues). During this time, the conelet enlarges rapidly to form a full-sized cone. Seeds are full sized at the tim **fertilization,** but seeds and cones continue to mature until late summer or early fall.

A mature, fully developed seed (fig. 1G) has an embryo composed of immature needles **(cotyledons),** immature s **(hypocotyl),** and an immature root **(radicle).** The gametophyte tissue enveloping the embryo provides the food reserve **germination.** The hardened seedcoat protects the seed.

The sequence of pine seed development – from flower primordia formation to seed fall – is illustrated in figure **2.** development spans parts of three growing seasons.

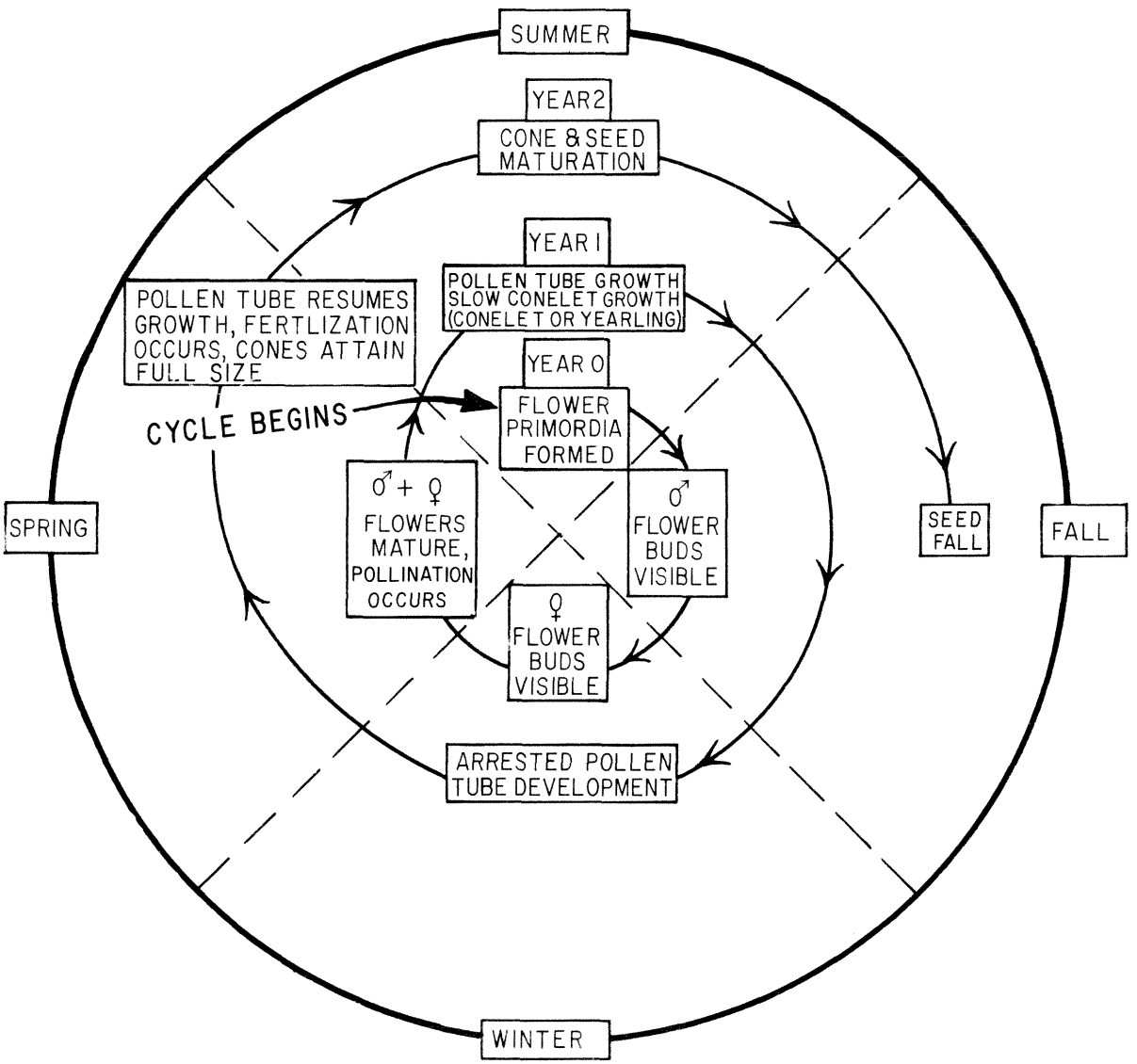


Figure 2.–The cycle for flowering and seed production in southern pines.

III. GENERAL GUIDELINES FOR CONE SELECTION

The method and number of cones to select for analysis depend on the analytical objectives, the planting de variation within the orchard.

Overall performance of the orchard can be evaluated by selecting cones from throughout the orchard. Exclude damaged cones, but do include cones of all sizes that are normally harvested and extracted. Such a bulk samp used for comparisons with other orchards or with other years for the same orchard. A sample of from **25** to **100** orchard is normally sufficient to evaluate overall seed efficiency. However, with such selection, no conclusior reached about individual **clones.**

If seed efficiencies must be estimated for individual clones, cones have to be collected separately from speci From limited data, it appears that **10** or more cones per clone are required. If individual **ramets** are sampled withi then three to five cones per ramet will probably be necessary. Additional research and experience will provide a be for specific cone sample numbers.

How well the sample represents orchard conditions depends on the variation within the orchard. Morpholog such as cone length, number of scales, and **seed potential** vary relatively little, and only small samples are requi timate these traits with specified reliability. Seed-yield traits, however, are more difficult to estimate because mu tors (pollination, insects, etc.) contribute to the variation. Seed and ovule losses must be attributed to specific identify problems in orchard management, and such losses can be highly variable. Since seed losses are l management is good, seed-yield traits can be estimated from relatively small cone samples in a high-yield managed orchard. The very orchards where problems exist and where information needs are greatest are the mo to assess and require the largest cone samples.

IV. RECOGNIZING ABORTED OVULES AND SEEDS

In order to understand the details of the cone analysis procedure, it is first necessary to be able to disti developed seed from an **aborted ovule.** Ovule abortion may occur during the first or second year of developme and second-year aborted ovules can be distinguished from developed seeds fairly easily by the methods give section.

FIRST-YEAR ABORTED OVULES

This class of ovule abortion occurs on fertile scales during the conelet stage – before cone enlargement begi second growing season. The seed wing develops normally, and these ovules have been called "wings withou Rudimentary, nonfunctional ovules may develop with wings on some of the larger infertile scales. Infertile scale recognized by their narrow bases (fig. **3**). The nonfunctional ovule never has the capacity of becoming a seed anc as a small dark spot at the base of the wing (Lyons **1956**). First-year aborted ovules are noticeably larger than rud ovules (figs. **4**, **5A** and **5B**). Some infertile scales at the base of a cone may produce wings with no evidence c rudimentary ovule. The smallest infertile scales at the very base of the cone may not produce wings.

SECOND-YEAR ABORTED OVULES

Ovules that abort during the second growing season are always larger than those that abort during the first y may have a partially developed seedcoat (figs. **4B** and **5C**). These ovules are usually considerably smaller than d seeds, because they generally abort early in the second year of development. Some ovules abort in late spring or ea mer; these may be as large in outline as fully developed seeds but their seedcoats are collapsed. These ovules are tly necrotic or shriveled and usually are attached to the cone scale with resin (fig. **5D**).

DEVELOPED SEEDS

Developed seeds are full sized and have complete seedcoats (figs. **3C** and D; **4C**; **5E**, right, and F). These can b in specific classes based on their internal appearance on a radiograph (X-ray picture).

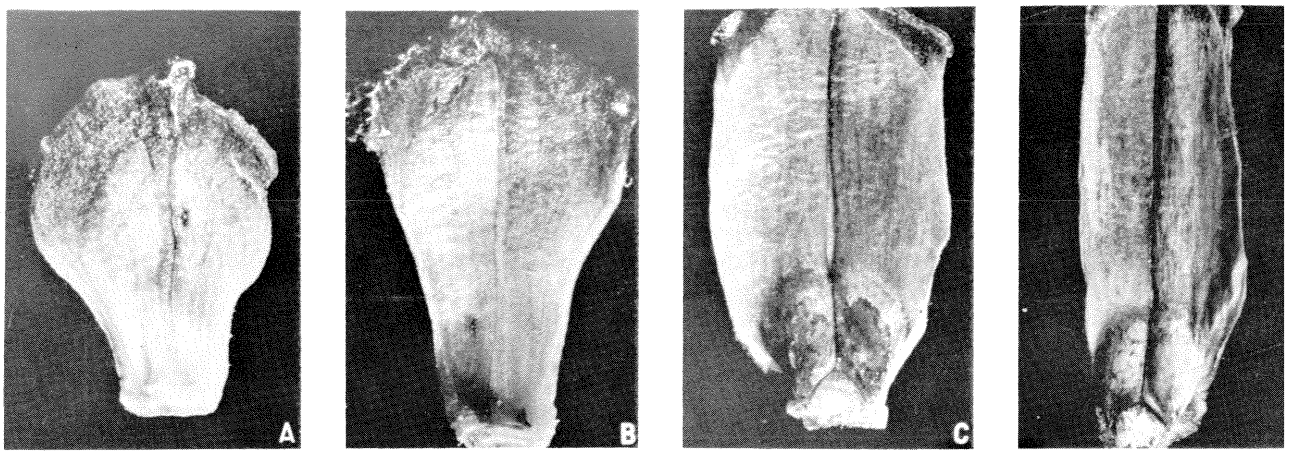


Figure 3.–Cone scales: (A & B) infertile scales with narrow base and no functional ovules, (C & D) fertile scales containing developed seeds with mature seedcoat.

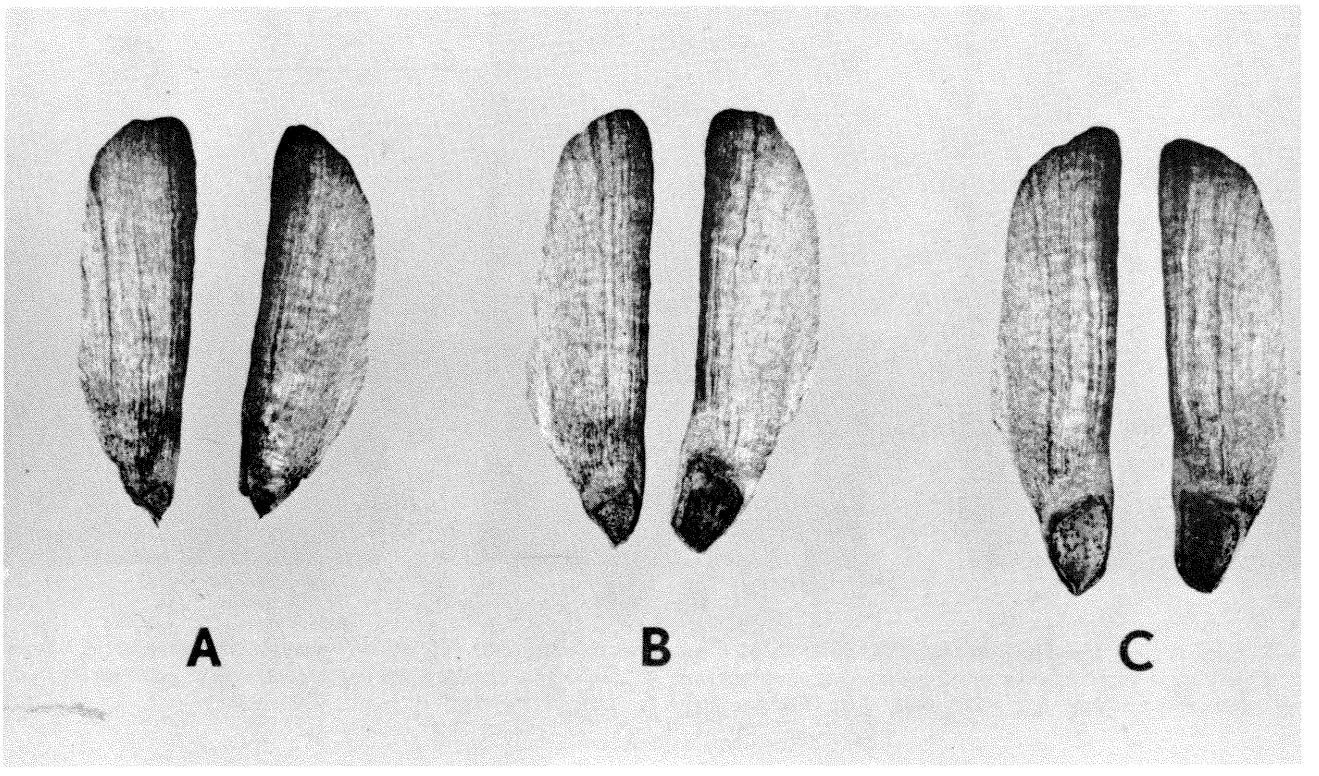


Figure 4.–Classification of aborted ovules and developed seeds: (A) first-year aborted ovules, (B) second-year aborted ovules, and developed seeds.

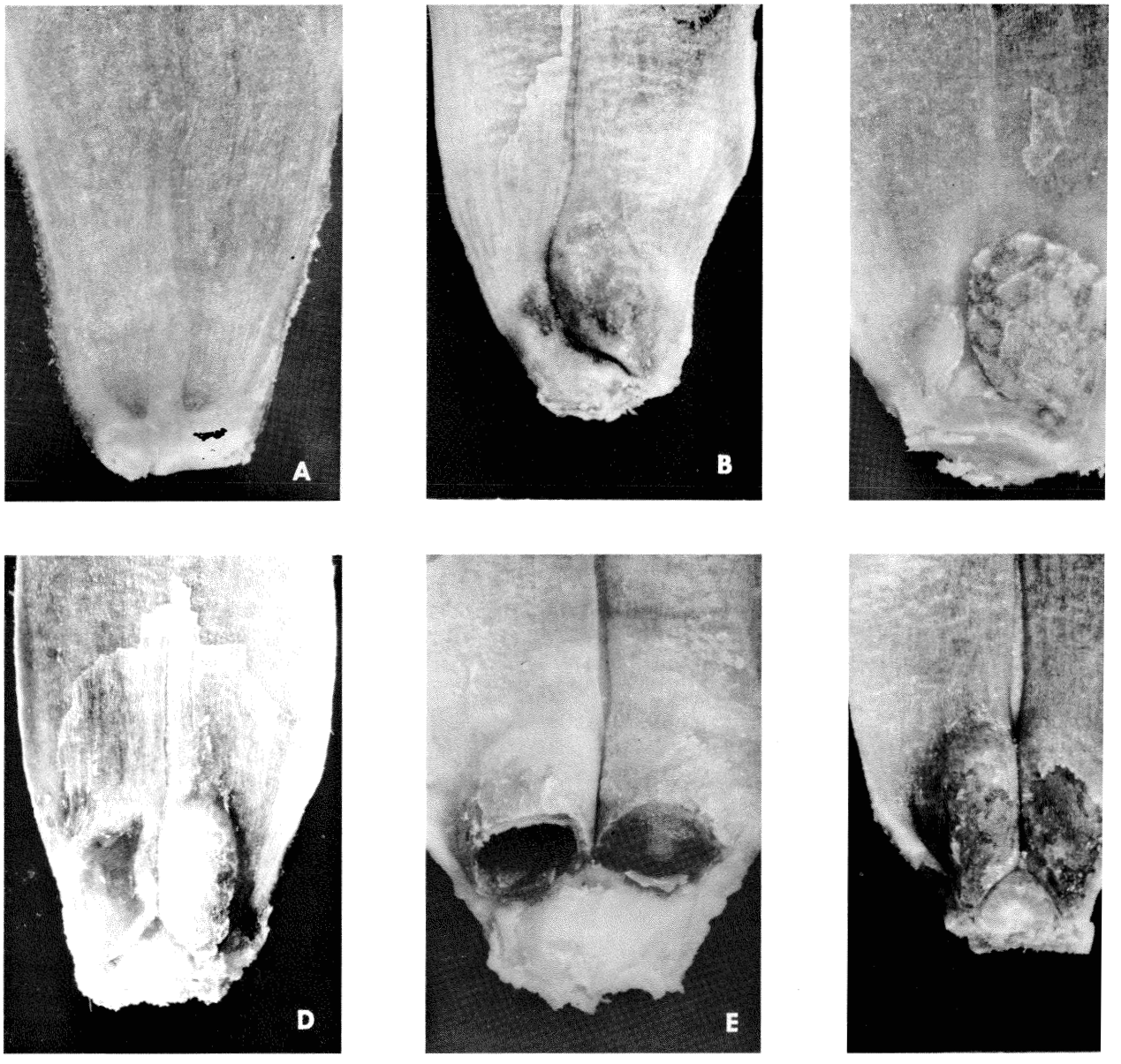


Figure 5.–Classification of ovules and seeds: (A) rudimentary ovules from infertile scale; (B) left, first-year aborted ovule; rigl ovule; (C) left, early second-year aborted ovule; right, normal ovule; (D) left, typical seedbug induced second-year aborted ov normal ovule; (E) sliced seed: left, empty seed; right, filled seed; (F) fully developed seeds.

V. CONE-ANALYSIS PROCEDURE

In cone analysis, all loose seeds and aborted ovules are extracted from the cone by normal procedures. Then cone sca are systematically removed and all the remaining ovules and seeds are collected. The number of scales classed as fer (fig. 3) is used to determine the biological potential of the cone to produce seeds. Ovules and seeds extracted and dissec from the cone are then classified by type (figs. 4 and 5). The seed efficiency of each cone is the number of filled seeds a percentage of the biological potential. The causes for seed losses can be identified and quantified on radiographs. The tent of seed losses to each cause indicates where corrective measures might be taken to reduce seed mortality.

EQUIPMENT NEEDS

Cone analysis requires little specialized equipment. Essential needs are:

1. Paper collection bags for individual cones (bag size depends on tree species).
2. Cone-drying facility (90°-105° F [32°-40° C] and relative humidity below 50 percent).
3. Weighing balance accurate to 1 gram.
4. Variable-speed 3/8-inch electric drill mounted in a stationary horizontal position and adjusted to run at the slow speed.
5. Drill bits of various sizes (inches): 1/4, 5/16, 3/8, 7/16, 1/2, 5/8.
6. Grafting knife.
7. Leather work gloves.
8. Metric ruler.
9. Containers for soaking cones.
10. Germination supplies and equipment.
11. X-ray supplies and equipment.

PROCEDURE

The stepwise procedure for cone analysis is presented in a flow chart in figure 6. Bits of data that should be recorded indicated in the chart; the columns mentioned are those on the sample data sheet in the Appendix. Some morpholog traits are considered optional (shaded columns on data sheet) and are not necessary for computation of the most import seed and cone characteristics.

Two cone-analysis procedures are presented. These are designated "Procedure A" and "Procedure B." During collection of data, there is one basic difference in the two procedures. With Procedure A, the extracted and dissec developed seeds are combined (steps 9A and 15A – columns 46-61). With Procedure B, the extracted developed se (step 9B – columns 46-61) are kept separate from the dissected developed seeds (step 15B – columns 62-77) from cone. Each procedure has certain merits and limitations, which are summarized below.

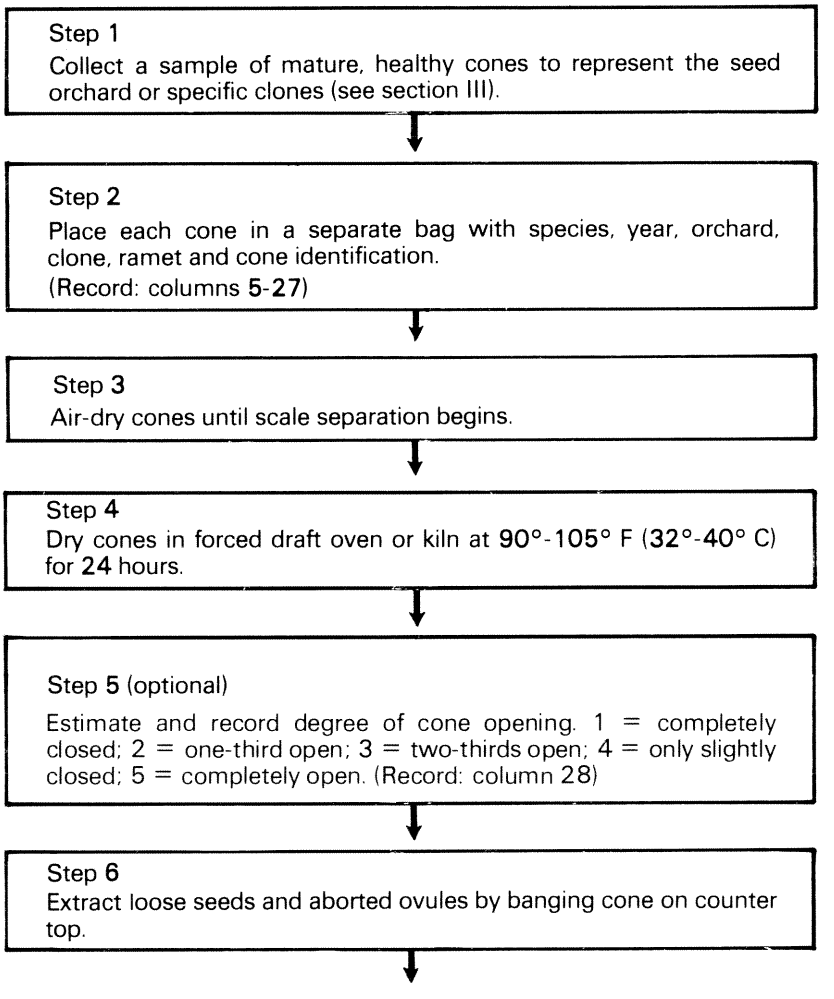
Procedure A. – This procedure evaluates the yield and the extraction and germination efficiencies of all the seeds in mature cone. The developed seeds extracted from the cone by normal procedures are combined with the developed se recovered by removing the cone scales (dissection). All seeds are then radiographed and classified as described in sect VI. The advantage of the procedure is that only a single radiograph is required for each cone. Once the cone analysi begun on a particular cone, the extraction, dissection, and seed classification can be completed at one time. Also, primary emphasis is on the number of seeds actually produced in the cone, rather than just on those seeds extracted fr the cone under laboratory conditions. This procedure is simplest for small organizations doing their own cone analysis. C mination percentage using Procedure A is based on the ratio of the number of germinated seeds to the total numbe filled seeds produced by the cone.

Procedure B. – This is the procedure currently being used by the Eastern Tree Seed Laboratory for its Cone Ana vice (CAS). In this procedure the seeds extracted after forced drying are radiographed, and tests of their germin begun before the remaining steps in the analysis are completed. Because germination testing is the step that rec most time, this procedure can substantially reduce the number of weeks needed to complete the analysis. This particularly valuable when many cones are to be analyzed.

The germination percentage in Procedure B is the percentage of the extracted filled seeds that produce a norm ing. Extraction efficiency by this procedure is the percentage of all filled seeds in the cone that were extracted af (Karrfalt and Belcher 1977).

Before starting cone analysis, the user must decide which procedure to follow. Once a procedure is decided steps and computations indicated for that procedure must be followed.

Figure 6.–Flow chart for the steps in cone analysis. At several points steps are different for Procedure A than for Pro



Step 7

Separate developed seeds from aborted ovules (see section IV).

Step 8

Count and record number of extracted, developed seeds. (Record: columns **43-45**)

Step 9A

Place developed seeds in a labeled envelope.

Step 9B

a. Place developed seeds in a labeled envelope for X-ra examination.

b. Radiograph seeds in envelope.

c. Begin germination procedures (see step **18**).

d. Classify seed images on radiograph (see section VI). (Re cord: columns **46-61**)

Step 10

Separate first-year aborted ovules from second-year aborted ovules (see section IV). Record only number of second-year aborted ovules. (Record: columns **41-42**). Discard all aborted ovules.

Step 11

Soak cone in water until scales close.

Step 12 (optional)

Measure cone length to nearest millimeter. (Record: columns **29-31**)

Step 13

Dissect cone: beginning at base of cone, remove lower scales with a knife point (fig. **7**). As **cone axis** is exposed, use a drill with bit diameters of **1/4** inch to **5/8** inch to remove cone axis. This facilitates scale removal. Alternate drilling (**1** inch or less at a time) and scale removal (fig. **8**). Match drill bit diameter with that of the cone axis.

Step 14

Separate scales into groups: (1) lower infertile scales, occurring at the base of the cone (figs. 3A, B and 5A); (2) fertile scales, occurring in the central two-thirds of the cone and having two functional ovules or developed seeds (figs. 3C and D); and (3) upper infertile scales, occurring at the apex of the cone and not having the capacity to produce seed.

Step 15A

a. Take developed seeds from removed cone scales and add to previously extracted seeds in the envelope (step 9A).

b. Radiograph envelope containing seeds.

c. Classify seed images on radiograph (see section VI). (Record: columns 46-61)

Step 15B

a. Take developed seeds from removed cone s and place in separate envelope.

b. Radiograph envelope containing seeds.

c. Classify seed images on radiograph (see se VI). (Record: columns 62-77)

Step 16

Count any remaining second-year aborted ovules and add to number previously extracted (step 10). (Record: columns 41-42)

Step 17

Determine the number of first-year aborted ovules by subtracting the total number of second-year aborted ovules plus the total number of developed seeds from two times the number of fertile scales on the cone. (Record: columns 38-40)[2]

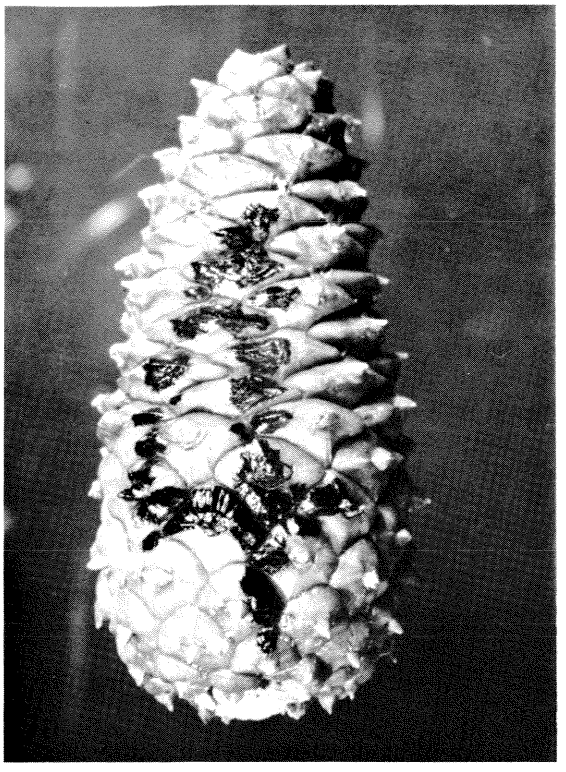
Step 18

Place all developed seeds in germination test using standard germination testing procedures.

Step 19

Summarize data and compute important seed production indicator values (see section VII).

[2]*First-year aborted ovules may be counted as the scales are removed by observing the location of the scale where the seed is produced. S first-year aborted ovules do not have the depression formed in the scale where the seed was located.*



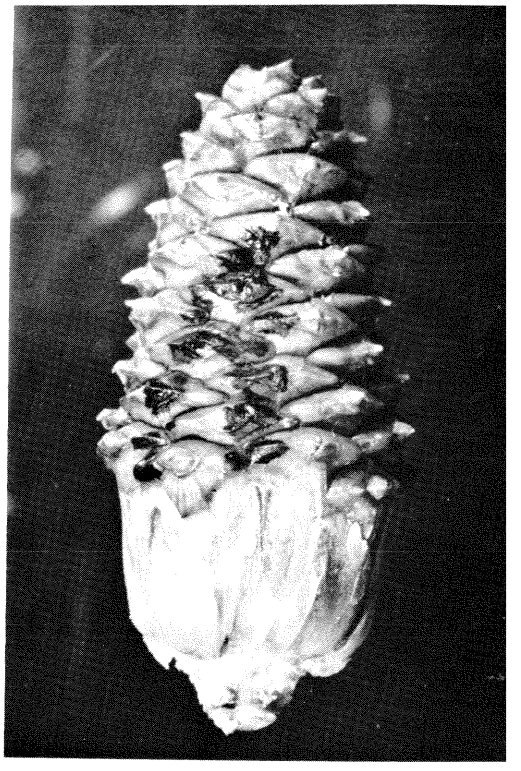


Figure 7.–(A) Shortleaf pine cone at harvest. (B) Same cone with lower infertile scales removed. Remaining scales are all fertile e for a few infertile scales at the cone tip.

Figure 8.–Cone scale removal. Scale removal is alternated with drilling out short distances of the cone axis.

VI. CAUSES OF OVULE AND SEED LOSSES

After first-year aborted ovules and second-year aborted ovules have been separated from developed seeds, the for seed losses can be identified. The cause for abortion of an ovule often must be inferred from the time at which a occurred. Developed seeds are classified by the appearance of the embryo and gametophyte tissue on a radiograph Classes of ovules and seeds are defined, and causes for losses are summarized in tabular form in the Appendix.

FIRST-YEAR ABORTED OVULES

There are two causes of ovule abortion in the first year – lack of pollen and seedbug damage.

Lack of viable pollen. – Pollen is an obvious requirement for the production of healthy cones and viable seeds 1962). For production of viable seeds, two pollination requirements must be met. Pollen must be available when flowers are receptive, and it must germinate and grow into the ovule tissue. Female flower receptivity begins wl opening between the scales is large enough for pollen to enter and extends from a few days to more than a week. O sca'es have closed, additional pollen cannot enter the ovule. If no pollen is present or if the pollen fails to germin ovule aborts early in the first year of cone development (Bramlett and Johnson 1975; McWilliam 1959; Sarvas 196

Seedbug feeding. – Feeding by nymphs of the leaffooted pine seedbug, *Leptoglossus corculus* (Say), often cause tion of a high percentage of ovules (DeBarr and Kormanik 1975; DeBarr and Ebel 1973; DeBarr and others 1975). destroyed by seedbugs cannot easily be distinguished from ovules aborting from lack of pollen.

Losses to seedbugs can be estimated by protecting some cones in screened cages and comparing yields of pr with unprotected cones. With this technique, the impact of seedbugs can be separated from losses to physiological vironmental factors.

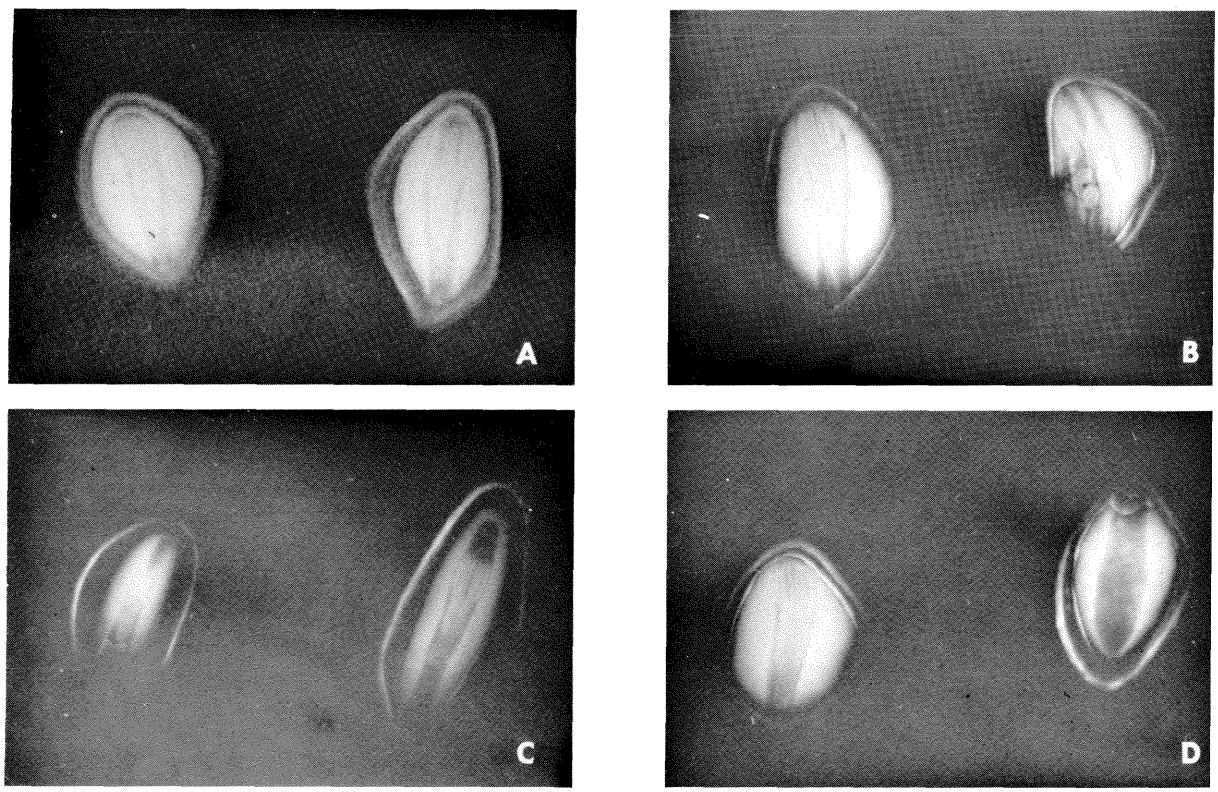
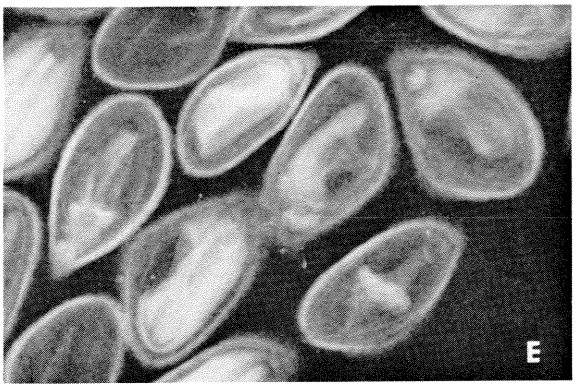
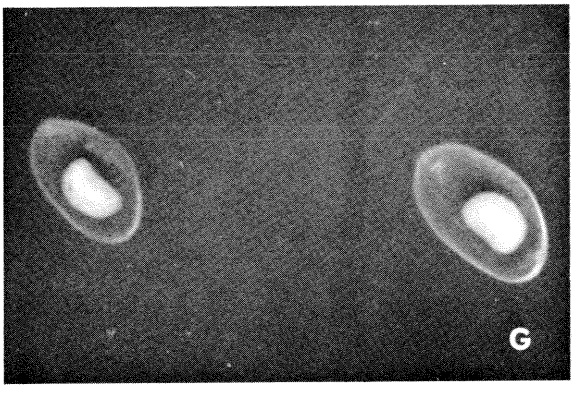
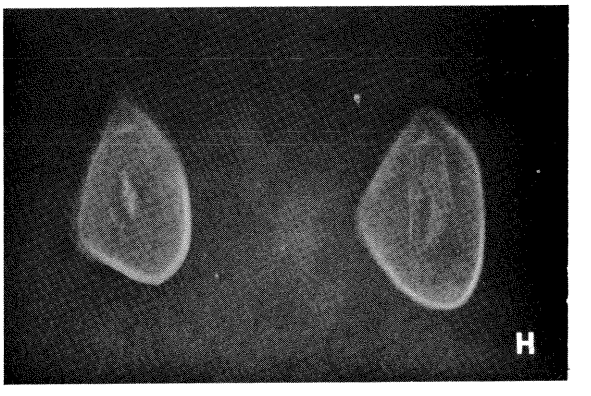
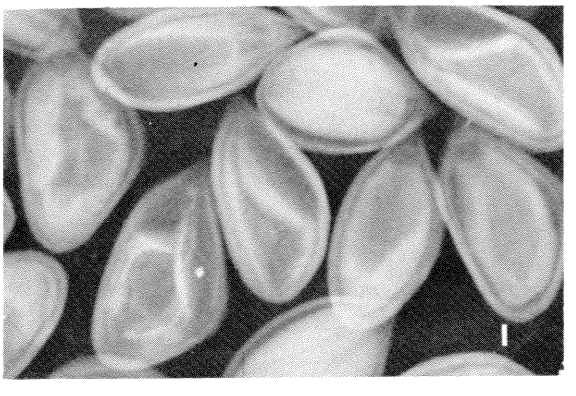


Figure 9.–Classification of radiographic images of developed seeds: (A) filled (potentially sound) seeds; (B) mechanically c seeds; (C & D) malformed seeds–C, incomplete gametophyte; D, distorted embryo (left); missing embryo (right); (continued)









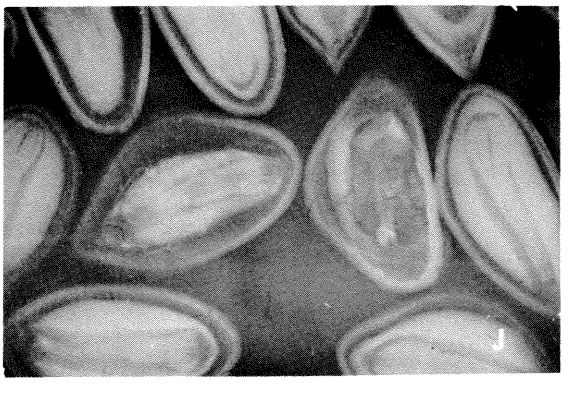


Figure 9 (continued).–(E) damaged by seedbug; (F) destroyed by seedworm; (G) destroyed by seed chalcid; (H) empty seeds and (I & J) damaged by fungi.

SECOND-YEAR ABORTED OVULES

Seedbug feeding. – In extensive studies of shortleaf *(P. echinata* Mill.) and loblolly *(P. taeda* L.) pines (DeB slash pine *(P. elliottii* Engelm. var. *elliottii)* (DeBarr and others **1975**), and Virginia pine *(P. virginiana* Mill.) (B Moyer **1973**), second-year ovule abortion has clearly been shown to result almost entirely from feeding by th pine seedbug in early summer (fig. **10**). These ovules appear resinous, collapsed, or necrotic on the mature (figs. **4B**; **5C**, left; and **5D**, right).

Developmental problems.–Some ovules stop developing during the second year after they have developed These "miniature seeds" may be only slightly larger than first-year aborted ovules or up to one-half the norr seeds of the species. These are all empty. The cause of this type of ovule mortality is unknown, but it appare from a block in the normal developmental sequence prior to fertilization. The frequency of second-year aborted to developmental problems has been quite low in production seed orchards.

DEVELOPED SEEDS

Filled, partially filled, and empty developed seeds are readily distinguishable on radiographs. In addition, seve damage can be identified on radiographs (fig. **9**).

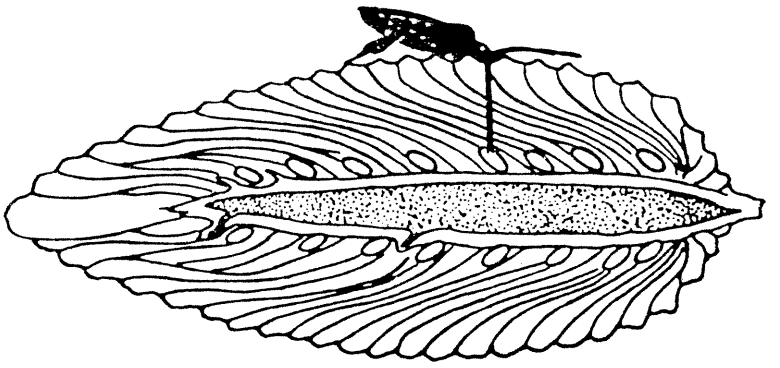


Figure 10.–Diagram of a cone showing the leaffooted pine seedbug penetrates a nate layers of cone scales and seed wing puncture seeds lying along the cone (From DeBarr 1970)

Filled Seeds

Filled seeds (potentially sound seeds) are those that have healthy undamaged gametophyte tissue, a norm and no evidence of insect or fungal damage. These seed structures are readily distinguished on a radiograp Potentially sound seeds may be mechanically damaged at any stage of extraction (fig. **9B**). Since the primary cone analysis is to judge production efficiency in the seed orchards, mechanically damaged seeds should be filled.

Partially Filled Seeds

Seedbug.–The leaffooted pine seedbug and the shieldbacked pine seedbug, *Tetyra bipunctata* (H.-S.), feed year cones, damaging and destroying seeds (DeBarr **1967**). Damage to fully developed seeds can often be ic radiographs by the appearance of the gametophyte tissue (DeBarr **1970**). Typical seedbug feeding removes shaped portions of gametophyte tissue (fig. **9E**). Extended feeding on a single seed often removes most of the ga tissue and embryo, leaving only a shriveled remnant in the seedcoat.

Seedworm.–Three species of seedworms, *Laspeyresia* spp., infest seeds of the southern pines (Ebel and others 1975 Since seedworm-damaged seeds generally are stuck to the cone scales with excrement and silk webbing, most are detec ted and counted as the individual cone scales are dissected during cone analysis. The few seeds included in the extracte sample are easily detected on the radiograph (fig. 9F). Seedworm larvae are not present in the seeds, and the exit holes ar large. The gametophyte tissue has been replaced by larvel excrement and boring frass that looks granular on the radiograp and often is formed into concentric rings (DeBarr 1970).

Seed chalcid.–A seed infested by a seed chalcid, *Megastigmus atedius* Walker, lacks gametophyte tissue and contains single larva that is readily visible on the radiograph (fig. 9G). To date, the only pine growing in the Southern United State known to be infested by this seed chalcid is eastern white pine *(P. strobus* L.) (Speers 1975).

Fungi.–Fungal damage is recognizable on radiographs as (1) cloudy or hazy semicircular areas generally at the margin c the gametophyte tissue (fig. 9I), (2) partial to complete disorganization of both the gametophyte and embryonic tissues (fi 9J), and (3) all gametophyte tissue and embryo shrunken or "mummified." Fungal damage may have the same appearanc as advanced seedbug damage on the radiograph. Dissection and close examination of infected seeds have revealed fungi i the gametophyte tissues, embryo, or both. The mechanisms of fungal penetration are not known, but the presence of fun in seeds is known to reduce the germination percentage and to increase damping-off of seedlings (Miller 1976).

Malformed seeds.–Seeds with incomplete embryos, distorted embryos, or incomplete gametophytes are classed a malformed. Missing or malformed embryos show up clearly on radiographs (fig. 9C and D). These malformations hav physiological causes. In some malformed seeds, the gametophyte tisse does not completely fill the seedcoat and no inte nal insect, disease, or mechanical damage is evident (fig. 9C). In these seeds, approximately one-half to three-quarters c the normal amount of gametophyte tissue is normally present. Seeds with incomplete gametophytes occur in cones ha vested before maturity. Malformed seeds occur at a low frequency in most lots and rarely exceed 1 to 2 percent for mo trees.

Empty Seeds

Seeds that contain only a remnant of gametophyte or embryo tissue (fig. 9H) are classed as empty. In the trade, empt southern pine seeds are often called "pops." There are two known causes of empty seeds. First, each pine parent carrie some recessive lethal genes (Bramlett and Pepper 1974; Franklin 1969). When two recessive lethal genes for the sam trait are present, the embryo dies and the gametophyte tissue does not develop. The result is an empty seedcoat with on a remnant of the dead embryo present. Second, seedbug feeding during the early stages of embryo and gametophyt development will produce a completely empty seedcoat. The gametophyte tissue is completely destroyed by digestive er zymes of the insect.

VII. SUMMARIZING DATA

To interpret the information obtained from the cone analysis, the data must be summarized. The summary should b tabulated according to trees, clones, or other categories of interest. For the following items, a numerical average can b computed:

Cone opening (CO)
Length (LT)
Lower infertile scales (LI)
Filled seeds (FL)
Malformed seeds (MS)
Seedbug-damaged seeds (SB)

Upper infertile scales (UI)
Fertile scales (FS)
First-year aborted ovules (A1)
Second-year aborted ovules (A2)
Extracted seeds (ES)
Seedworm-destroyed seeds (SW)
Seed chalcid-destroyed seeds (SC)
Fungal-damaged seeds (FN)
Empty seeds (EM)
Germinating number of seeds (GN)

From the basic summary of the raw data, the key indicators of seed performance are calculated as shown b
bers in parentheses are the columns on the suggested data form in the Appendix.

Indicator **Computation**

1. **Seed potential (SP) = 2 x Fertile scales** *or* **2 x (34-36)**

2. **Total developed seeds (TS) = Seedbug-damaged seeds + seedworm-destroyed seeds chalcid-destroyed seeds + fungal-damaged seeds + malformed seeds + empty seeds + filled (46-47 and 62-63) + (48-49 and 64-65) + (50-51 and 66-67) + (52-53 and 68-69) + (54-55 ar + (56-58 and 72-74) + (59-61 and 75-77)**

3. **Percent developed seeds =** $\frac{\text{Total developed seeds}}{\text{Seed potential}} \times 100$ *or* $\frac{TS}{SP} \times 100$

4. **Percent filled seeds =** $\frac{\text{Filled seeds}}{\text{Total developed seeds}} \times 100$ *or* $\frac{(59\text{-}61 \text{ and } 75\text{-}77)}{TS} \times 100$

5. **Percent known insect-damaged seeds =**
$$\frac{\text{Seedbug-damaged seeds + seedworm-destroyed seeds + seed chalcid-destroyed seeds}}{\text{Total developed seeds}} \times 100 \textit{ or}$$
$$\frac{(46\text{-}47 \text{ and } 62\text{-}63) + (48\text{-}49 \text{ and } 64\text{-}65) + (50\text{-}51 \text{ and } 66\text{-}67)}{TS} \times 100$$

6. **Percent malformed seeds =** $\frac{\text{Malformed seeds}}{\text{Total developed seeds}} \times 100$ *or* $\frac{(54\text{-}55 \text{ and } 70\text{-}71)}{TS} \times 100$

7. **Percent fungal-damaged seeds =** $\frac{\text{Fungal-damaged seeds}}{\text{Total developed seeds}} \times 100$ *or* $\frac{(52\text{-}53 \text{ and } 68\text{-}69)}{TS} \times 100$

8. **Percent first-year aborted ovules =** $\frac{\text{First-year aborted ovules}}{\text{Seed potential}} \times 100$ *or* $\frac{(38\text{-}40)}{SP} \times 100$

9. **Percent second-year aborted ovules =** $\frac{\text{Second-year aborted ovules}}{\text{Seed potential}} \times 100$ *or* $\frac{(41\text{-}42)}{SP} \times 100$

10. **Seed efficiency (SE) =** $\frac{\text{Total filled seeds}}{\text{Seed potential}}$ *or* $\frac{(59\text{-}61 \text{ and } 75\text{-}77)}{SP} \times 100$

11. Extraction efficiency (EE) =

Procedure A: $\frac{\text{Extracted developed seeds}}{\text{Total developed seeds}}$ *or* $\frac{(43\text{-}45)}{\text{TS}} \times 100$

Procedure B: $\frac{\text{Extracted filled seeds}}{\text{Total filled seeds}}$ *or* $\frac{(59\text{-}61)}{(59\text{-}61 \text{ and } 75\text{-}77)} \times 100$

12. Germination percentage (GP) =

Procedure A: $\frac{\text{Germinated seeds}}{\text{Total filled seeds}} \times 100$ *or* $\frac{(78\text{-}80)}{(59\text{-}61 \text{ and } 75\text{-}77)} \times 100$

Procedure B: $\frac{\text{Germinated seeds}}{\text{Extracted filled seeds}} \times 100$ *or* $\frac{(78\text{-}80)}{(59\text{-}61)} \times 100$

13. Seedling efficiency = $\frac{\text{Seed efficiency x Extraction efficiency x Germination percentage}}{10{,}000}$ *or* $\frac{\text{SE x EE x G}}{10{,}000}$

VIII. DATA INTERPRETATION AND UTILITY

The purpose of cone analysis is to evaluate seed production and to identify when and why potential seeds are lost. Tabl 1 shows typical values for key indicators in a well-managed seed orchard. It also shows values that may indicate need fc corrective action.

The seed potential defines the biological limit for the number of seeds produced by each cone. Thus, each tree specie has an average seed potential and a range of observed values based upon the number of fertile scales per cone. For exam ple, the seed potential for cones from a slash pine seed orchard averaged 170. Loblolly pines had an average seed potenti of 155, and Virginia and shortleaf pines had averages of 88 and 87 (Bramlett 1974). Variation in seed potential is relativel small within specific clones. Seed potentials in a given orchard, however, may differ from the average for the specie depending on the particular clones in the sample.

Aborted ovules are potential seeds that die before the formation of a normal seedcoat. For all the southern pines, ovul abortion can be a serious cause of low seed yields. The two known causes of first-year aborted ovules (lack of pollen an seedbug damage) are influenced by the orchard age. In young orchards, female flowering of many clones usually begin before male catkin production is adequate to ensure complete pollination. Unless there is a supply of pollen from su rounding stands, many ovules die within 2 months from lack of viable pollen. In contrast, older seed orchards have a abundant pollen supply but also often have large seedbug populations that cause extensive first-year ovule abortion in un protected conelets. Conclusive separation of insect from pollination problems requires comparison of full seed yields fror cones with and without screened cages that exclude insects.

Second-year aborted ovules are, for practical purposes, a direct indication of seedbug feeding. Other agents seldor cause second-year abortions exceeding 1 to 2 percent of the seed potential. Second-year ovule abortion may reach 50 pe cent, but seedbug control may be warranted when the loss approaches 10 percent of the seed potential. To preven second-year ovule abortion, insecticide applications must be timed to reduce populations of leaffooted pine seedbugs du

ing the spring when the conelets are rapidly enlarging to full-sized cones. Seedbug feeding on full-sized con empty or damaged seeds rather than aborted ovules.

Total developed seed is simply the number of normal, full-sized seeds in each cone. Obviously, the orchard strives to maximize developed seed production. With adequate pollination and effective insect control, the devel yields should approach 80 to 90 percent of the seed potential.

Yields of developed seeds are meaningless without considering seed quality. Using radiography, the actual filled seeds per cone is determined. Then seed efficiency is expressed as the ratio of filled to potential seed. Thu ficiency measures the productivity of a cone in relation to its biological capacity. The average seed efficiency for an orchard is the single most important measure of seed production. It can be used to compare the relative perfc two pine species with different seed potentials. For example, if a shortleaf pine produces an average of 45 filled of a potential of 90, the seed efficiency would be 50 percent. However, 45 filled seeds in a slash pine cone with of 180 indicates a seed efficiency of only 25 percent.

TABLE 1. – KEY INDICATORS FOR DATA INTERPRETATION

Cone characteristic and threshold level (percent)[1]	Interpretation
1. First-year aborted ovules	
0-10	Good management practice
11-19	Evaluate possible insect or pollination problems
20+	Identify and correct insect or pollination problems
2. Second-year aborted ovules	
0-5	Good management practice
6-9	Evaluate seedbug control program
10+	Increase seedbug control
3. Percent filled seeds	
85+	Good management practice
50-84	Evaluate possible seedbug or inbreeding problems
Below 50	Identify and correct seedbug or inbreeding problems
4. Insect-damaged seeds	
0-10	Good management practice
11-19	Evaluate insect problem and control program
20+	Improve or increase control program
5. Seed efficiency	
55+	Good management practice
35-54	Evaluate causes of seed loss
Below 35	Identify causes of seed loss and begin correction
6. Extraction efficiency	
90+	Good management practice
Below 90	Identify and correct improper conditions or immature cone collections
7. Germination percentage	
90+	Good management practice
80-89	Identify cause (May be: undetected seedbug damage or mishandling c
Below 80	Identify and correct problem (May be: improper germination, undetect seedbug damage, mishandling of seed)
8. Seedling efficiency	
50+	Good management practice
26-49	Identify cause – poor germination, immature cones
Below 25	Identify and correct cause

[1] Each organization may wish to modify these thresholds to meet its targets.

The biological maximum for seed efficiency may be as high as 80 percent for individual cones, but values this high a seldom observed under seed-orchard conditions. Seed efficiencies greater than 70 percent have occurred when scree wire cages were used to protect slash pine cones from insects (DeBarr and others 1975).

Seed efficiency values of 35 to 54 percent indicate that some potential seeds are being lost. Yields could be increase by identifying the specific causes of seed losses and prescribing corrective procedures within economic constraints. See efficiencies of less than 35 percent indicate excessive losses. The causes should be identified and the problems correcte as soon as possible.

Developed seeds not classified as "filled" are worthless. Thus, filled seed percentage simply expresses the ratio of fille to developed seeds. Filled seed percentages seldom exceed 90 percent in commercial operations, but greater values ha been reported with controlled pollinations or effective seedbug control (Bramlett and Pepper 1974; Merkel and othe 1976). Wind-pollinated cones from orchards should average 85 percent or more filled seeds. Lower values indicate loss in some or all of the classes identified from the radiographs. Individual-loss classes exceeding 10 percent warrant correcti measures.

Insects, particularly seedbugs, destroy a large percentage of the developed seeds. In cone analysis, losses are attribute to seedbugs, seedworms, and seed chalcids. Usually, some seeds classed as "empty" are also the result of seedbu feeding. Therefore, as recognized seedbug losses increase, the number of empty seeds often rises (DeBarr 1974b; DeBa and Ebel 1973).

Malformed seed losses (incomplete gametophyte tissue, distorted embryo, missing embryo) seldom exceed 10 perce of the developed seeds in well-managed orchards. Incomplete gametophytes apparently result from retarded developme or inadequate nutrition. Cones collected before maturity produce seeds with incomplete gametophytes. Malformed see occur in sample lots at such low frequencies that these types of losses are of only minor importance.

Fungus damage to southern pine seeds has only recently been recognized as a potential problem. The importance fungal damage is undetermined. Part or all of the seeds from cones with external symptoms of fungal damage may be i fected by fungi. Corrective measures are not yet available to prevent seed destruction by fungi.

The causes of empty seeds are difficult to identify. Lethal genes in the embryo, seedbugs, and possibly fungi cau empty seeds. In wind-pollinated seeds from an orchard, as many as 15 percent of the developed seeds may be empty. Th value would reflect some effects of self-pollination within the clones in the orchard (Kraus 1975); the proportion of emp seeds is normally high after self-pollination. Lethal genes in embryos cannot be eliminated easily. This genetically controll system minimizes inbreeding and favors outcrossing in pines.

If more than 15 percent of the developed seeds are empty, seedbug damage should be suspected and control prograr should be reevaluated. Seedbug damage can be reduced with insecticides.

Extraction efficiency measures the ease with which developed seeds are removed from the cones. It is the percentage developed seeds extracted by kiln or air-drying the cones. It should be noted that this extraction is done in a laboratory that the results are not necessarily the same as would be obtained by commercial extraction. Percentages below 90 i dicate poor extraction technique or immature cones.

Germination percentage measures the viability of seeds classified as "filled" (potentially sound). This value norma averages 90 percent or better. Lower germination percentages may indicate improper classification of some seeds in t lot (Rowan and DeBarr 1974). Germination percentages are useful for comparing the viability of collections from differe trees or from subsequent years.

Seedling efficiency is simply the product of the three efficiency values – seed efficiency, extraction efficiency, and g mination percentage. It reflects the total yields of the cone, seed, and seedling crops from the orchard. If any one of the ficiency values is low, the overall efficiency of the orchard is reduced. A maximum realistic goal for production orchards is seedling efficiency of about 55 percent. Such would be the case if seed efficiency = 70 percent, extraction efficiency = 9 percent, and germination percentage = 90 percent.

Cone analysis does not estimate all seed losses; it only measures seed losses from surviving cones. Cone losses frc the time of flower initiation to cone maturity are not considered. Often 50 percent or more of the seed crop is lost duri this period. By combining cone crop life tables with cone analysis, the total seed production for an orchard can be evaluat (Fatzinger 1975; DeBarr and Barber 1975; Bramlett 1972a; Ebel and Yates 1974).

IX. FOR ADDITIONAL INFORMATION

For help with known insect problems or assistance in identifying insect problems, contact:

STATE FOREST ENTOMOLOGIST

Alabama Forestry Commission
513 Madison Ave.
Montgomery, AL 36104
205/832-5897

Arkansas Forestry Commission
3821 W. Roosevelt Rd.
Little Rock, AR 72204
501/371-1736

Florida Division of Forestry
Collins Building
Tallahassee, FL 32304
904/488-7936

Georgia Forestry Commission
P.O. Box 819
Macon, GA 31202
912/744-3241

Kentucky Division of Forestry
207 Holmes St.
Frankfort, KY 40601
502/564-4496

Louisiana Forestry Commission
P.O. Box 1628
Baton Rouge, LA 70721
504/389-7121

Mississippi Forestry Commission
908 Robert E. Lee Building
Jackson, MS 39201
601/354-7124

N.C. Dep. of Natural Resources
P.O. Box 27687
Raleigh, NC 27611
919/733-4141

Oklahoma Forestry Division
Capitol Building
Oklahoma City, OK 73105
405/521-3886

S.C. State Commission of Forestry
P.O. Box 21707
Columbia, SC 29202
803/758-2261

Tennessee Division of
2611 West End Ave.
Nashville, TN 37203
615/741-3326

Texas Forest Service
P.O. Box 310
Lufkin, TX 75901
713/632-7761

Virginia Division of For
P.O. Box 3758
Charlottesville, VA 229
804/977-6555

FOREST INSECT AND DISEASE MANAGEMENT ZONE OFFICE

Forest Insect & Disease Management
Seed Orchard and Nursery Insects
P.O. Box 5895
Asheville, NC 28803
704/258-2850 x625

Forest Insect & Disease Management
Seed Orchard and Nursery Insects
2500 Shreveport Highway
Pineville, LA 71360
318/445-6511

For questions on the use of pesticides, consult:
Pesticide Specialist
Resource Protection
Suite 706
1720 Peachtree Rd., NW
Atlanta, GA 30309
404/881-3734

For further information on cone analysis or on the service supplied, contact:
Eastern Tree Seed Laboratory
P.O. Box 819
Macon, GA 31202
912/744-3311

X. GLOSSARY

Many of the definitions given below are based on those in the "Glossary for Forest Tree Improvement Workers" (Snyder 1972) ar "Seeds of Woody Plants in the United States" (USDA Forest Service 1974).

ABORTED OVULE:

FIRST-YEAR: Any potential seed that aborts (ceases development) during the first growing season or conelet stage of pine see production.

SECOND-YEAR: Any potential seed that aborts (ceases development) during the second growing season before fertilization or see coat hardening.

ARCHEGONIUM: Female sex organ in the ovule of pine.

CATKIN: The male strobilus which produces pollen.

CLONE: Group of genetically identical plants produced by vegetatively propagating a single plant over one or more generations; a complished by grafting, rooting, air-layering, or tissue culture. Compare ortet, ramet.

CONE: The female reproductive structure of pines, consisting of a central axis supporting bracts, each of which subtends a scale bea ing naked seeds.

CONE ANALYSIS: A systematic procedure to evaluate the seed production potential and efficiency of pine cones.

CONE AXIS: Central core of the cone.

CONELET: Immature female cone of pines from time of female "flower" scale closure after pollination until the initiation of rap development of the cone a few months before maturity.

COTYLEDONS: First leaves developed in the embryo of a seed. They become functional leaves after germination; immature leaves.

FEMALE CONE: See cone.

FEMALE FLOWER: The pine female strobilus (cone) prior to pollination.

FERTILE SCALE: A cone scale that is capable of producing seed; generally includes most scales in the upper one-half to two-thirds the cone.

FERTILIZATION: The union of the egg (1N) within the ovule and the sperm (1N) from the pollen tube to form the embryo (2N).

FILLED SEED (POTENTIALLY SOUND): Mature ovule containing an embryo and nutritive tissue enclosed in layers of protective tissu (seedcoat); capable under suitable conditions of development into a plant similar to the one that produced it.

GAMETOPHYTE TISSUE: Haploid material (1N) surrounding the embryo; provides nutritional reserves for the seed of pine.

GERMINATION: Sufficient development of plant parts from a seed to indicate a potential for developing into a normal plant.

HYPOCOTYL: All of the axis of an embryo or stem of a seedling between the cotyledons and the radicle; an immature stem.

INFERTILE SCALE: A pine cone scale incapable of producing seed; located at the base and apex of the cone.

INTEGUMENT: A single layer of tissue enclosing the pine ovule and nucellus. After fertilization, it develops into the seedcoat.

MICROPYLE: Small opening in the integument of an ovule through which the pollen grain normally passes to reach the pollen chan ber.

NUCELLUS: Mass of thin-walled cells that composes the central and main part of the body of an ovule; it is surrounded by the integ ment.

OVULE: A female structure which contains an egg cell and develops into a seed.

POLLEN GRAIN: The winged male fertilizing structure.

POLLEN TUBE: An elongate outgrowth of the germinating pollen grain.

POLLINATION: The transfer of pollen to the receptive female flower.

POTENTIALLY SOUND SEED: Seed filled with apparently undamaged tissue on which viability has not been tested.

RADICLE: Portion of the axis of an embryo from which the root develops; immature root.

RAMET: Independent member of a clone.

SEED: "In conifers" the structure formed after fertilization; initially consists of seedcoat, embryo, and gametophyte storage tissue.

SEEDCOAT: Protective layer on a seed derived from an integument.

SEED ORCHARD: A plantation consisting of clones or seedlings from selected trees and cultured for early and abundant see production.

SEED POTENTIAL: Two times the number of fertile cone scales or the maximum number of seeds a cone is capable of producing.

STROBILUS: Male or female fruiting body of pines.

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XII. APPENDIX

CONE ANALYSIS DATA CARD CODING AND COLUMN NUMBERS

ITEM	CODE	COLUMN	
Test number	TN	1-4	Identification Input
Year	YR	5-6	
Species	SP	7-8	
Orchard	OR	9-10	
Clone	CL	11-17	
Ramet	RA	18-25	
Cone number	CN	26-27	
Cone opening	CO	28	Optional Data
Cone length	LT	29-31	
Lower infertile scales	LI	32-33	
Upper infertile scales	UI	37	
Fertile scales	FS	34-36	
First-year aborted ovules	A1	38-40	
Second-year aborted ovules	A2	41-42	
Extracted seeds	ES	43-45	Developed Seeds
Seedbug-damaged seeds	SB	46-47,62-63	
Seedworm-destroyed seeds	SW	48-49,64-65	
Seed chalcid-destroyed seeds	SC	50-51,66-67	
Fungal-damaged seeds	FN	52-53,68-69	
Malformed seeds	MS	54-55,70-71	
Empty seeds	EM	56-58,72-74	
Filled seeds	FL	59-61,75-77	
Germinated seeds	GN	78-80	

CONE ANALYSIS DATA FORM

| | | |

CLASSIFICATION OF OVULES AND SEEDS

Class	Columns on data card: Procedure A	Columns on data card: Procedure B	Identity[1]	Cause	Comment
A. Aborted ovules					
1. First-Year	38-40	38-40	Small undeveloped ovules with normally developed wing (V)	a. Lack of viable pollen b. Seedbug feeding	Requires caged cones to establish cause
2. Second-Year	41-42	41-42	Flattened seed often resinous (V)	a. Seedbug feeding b. Developmental problems	Usually due to seedbug attack
B. Developed seeds					
1. Filled	59-61	59-61 75-77	Full-sized seed with complete seedcoat (V), embryo, and a gametophyte that fills the seedcoat (R)	--	Includes mechanically damaged filled seed; does not change classification
2. Partially filled					
Seedbug-damaged	46-47	46-47 62-63	Irregular areas of missing gametophyte tissue (R)	Seedbug feeding	--
Fungal-damaged	52-53	52-53 68-69	Cloudy or hazy semicircular areas at margin of gametophyte tissue, disorganization or shrunken or mummified gametophyte tissue (R)	Fungi	--
Malformed	54-55	54-55 70-71	Missing or damaged embryo or gametophyte tissue does not fill seedcoat (R)	Physiological	Early cone harvest will cause incomplete gametophyte tissue
3. Empty					
Seedworm-destroyed	48-49	48-49 64-65	Seed filled with granular material (R) Entry-exit hole in seedcoat (V)	Seedworm feeding	--
Seed chalcid-destroyed	50-51	50-51 66-67	Larva in seedcoat (R)	Seed chalcid feeding	Limited to eastern white pine seed
Empty	56-58	56-58 72-74	Seedcoat empty or containing a shriveled remnant of tissue (R)	a. Seedbug feeding b. Recessive lethal genes	Requires caged cones to establish cause

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