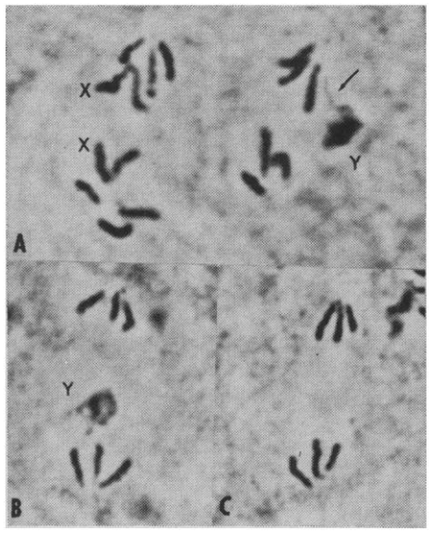


Fig. 1. (*A*) Two anaphase-II divisions, possibly in sister cells. The X-bearing cell (left) shows normal anaphase separation of the V-shaped X chromosome and of the three rod-shaped autosomes; the dot-like autosome is also visible at both poles. In the Y-bearing cell (right) the autosomes have undergone normal anaphase disjunction, but the Y chromosome exhibits the "degenerate" condition characteristic of "sex ratio." Evidence of some centromeric activity of the Y chromosome in this cell is indicated by the arrow. (*B*) A Y-bearing cell in anaphase II showing normal anaphase behavior of the autosomes and the "degeneration" of the Y chromosome. (*C*) A cell in anaphase II; no sex chromosomes are present. The material at upper right is a portion of the chromosome complement of another cell.

parture from normality. The course of meiosis is usually as follows: the X and Y chromosomes pair and separate normally in the first division, but in the second division the Y chromosome loses its characteristic appearance and appears as a chromatin mass which usually shows no centromeric activity at anaphase (Fig. 1, *A* and *B*). In some individuals this "degeneration" of the Y occurs early in metaphase II, while in others it is not seen until the autosomes have initiated their anaphase movement. Occasionally the aberrant behavior of the Y is seen as early as anaphase I. In accord with this observation, some second-division cells lack a sex chromosome (Fig. 1*C*) and their sister cells contain both a normal X chromosome and a Y chromatin mass. These cells would result from inclusion of the Y into the cytoplasm of the X daughter cell at the end of the first division. In three males, analyses of complete anaphase-II cysts were made; in each case the 32 X-bearing cells were normal and the 32 Y-bearing cells had only autosomes at the poles, with the Y-mass remaining at the equator. In none of the males examined was there any indication that all second-division cells contained an X chromosome.

It should be emphasized that the features of meiosis just described were found in both the laboratory and wild stocks. All the lines had three inversions in the right arm of the "sex ratio" X chromosome, these inversions corresponding to those figured by Dobzhansky and Epling (*11*). Counts were made on sperm bundles at spermatid and mature sperm stages in "sex ratio" and control males; "sex ratio" males had a full complement of 128 sperms in each bundle, with no indications of degeneration or abnormal development.

These observations lead to a simple explanation for the "sex ratio" effect in terms of the regular nonfunctioning of two of the products of meiosis: preferential movement of the X chromosome to the functional pole at anaphase I ensures an all-female progeny. Although it remains to be proved that the abnormal behavior of the Y chromosome is not responsible for the inactivation of the non-X-bearing sperm, it is possible to present a strong argument against a direct role of the degenerating Y chromosome. Schultz (*12*) has shown that sperms bearing neither an X nor a Y chromosome are functional, producing viable but sterile XO males. In our material the frequency of nondisjunction of the sex chromosomes at the first division is sometimes unusually high (up to 10 percent), but the progeny class which would correspond to this event, the XO males, does not appear with any appreciable frequency. It seems preferable to regard the occasional males that occur in some cultures as resulting from the rare failure of the X chromosome to become oriented toward the functional pole.

In connection with this study we also examined "sex ratio" in *D. athabasca* (*13*), another species of the *obscura* group. The features of meiosis were identical with those described for *D. pseudoobscura*, with the X and Y chromosomes disjoining regularly at first anaphase and the Y "degenerating" in the second division. Novitski (*14*) had previously shown that the "sex ratio" X chromosome of *D. athabasca* carries three inversions which bear some resemblance to those of *D. pseudoobscura*. Apparently "sex ratio" has enjoyed long-term involvement in the genetic systems of the *obscura* complex.

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8 February 1965

## Seed Dispersal Velocity in Four Dwarfmistletoes

Abstract. *By means of a high-speed photographic technique, the initial velocities of seeds were studied as they were expelled from the fruits of four Colorado dwarfmistletoes*: Arceuthobium douglasii, A. campylopodum *f.* cyanocarpum, A. vaginatum *f.* cryptopodum, *and* A. americanum. *Velocities of the seeds of the latter two species averaged 2600 centimeters per second and were significantly greater than those of the first two, which averaged 2200 centimeters per second. The initial velocity of 526 seeds of the four dwarfmistletoes averaged 2400 centimeters per second.*

The dwarfmistletoes (*Arceuthobium* spp.), which are one of the most serious agents of disease of western coniferous forests, have explosive fruits for seed dissemination. Local dispersal of the plants is almost exclusively due to this mechanical seed expulsion. As part of our studies of the biology of *Arceuthobium*, we measured the velocities of seeds immediately after they were expelled from the fruit.

Each *Arceuthobium* fruit contains a

Table 1. Some characteristics of fresh *Arceuthobium* seeds.

| Species | Seed size | | | Seed weight (mg) | Seed volume ($mm^3$) | Specific gravity |
|---|---|---|---|---|---|---|
| | *N** | Length (mm) | Width (mm) | | | |
| *A. cyanocarpum* | 35 | 2.0 | 0.9 | 0.9 | 0.6 | 1.5 |
| *A. douglasii* | 22 | 2.4 | 1.1 | † | 1.4 | † |
| *A. cryptopodum* | 30 | 2.7 | 1.1 | 2.3 | 1.6 | 1.4 |
| *A. americanum* | 30 | 2.4 | 1.1 | 2.0 | 1.4 | 1.4 |

* Number of seeds on which measurements were made. † Not determined but presumably similar to that for *A. americanum*, which has seeds of comparable size.

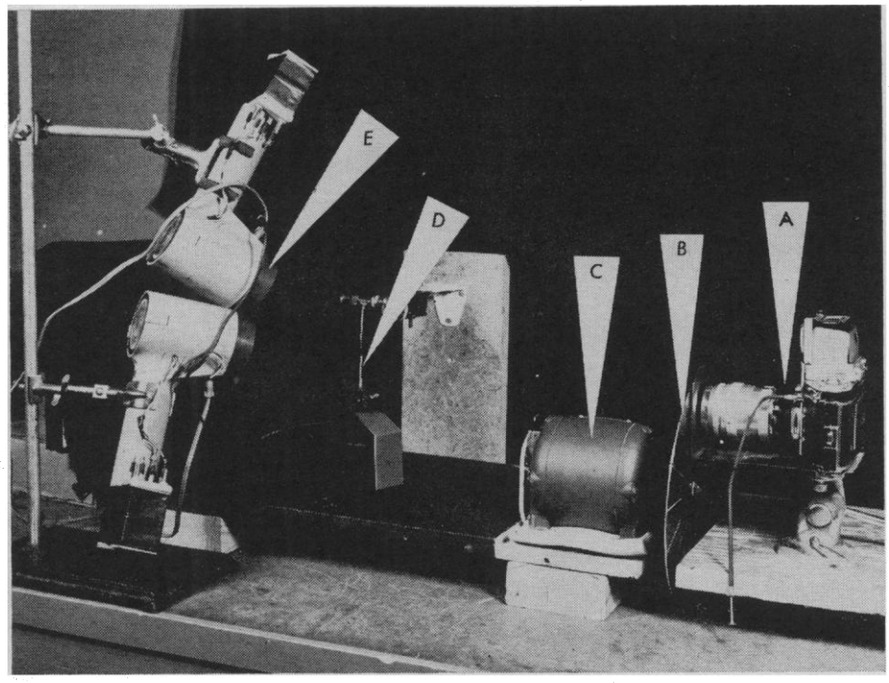


Fig. 1. Apparatus used for photographic study of *Arceuthobium* seed velocity. *A*, Camera; *B*, disc with 16 slits; *C*, motor for rotating disc; *D*, pendulum on which mature fruit was placed; and *E*, flash unit.

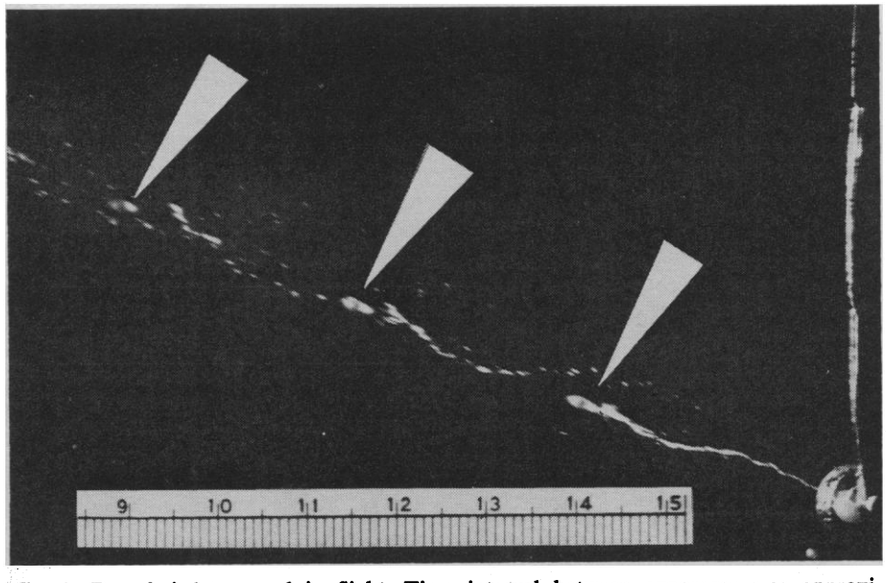


Fig. 2. Dwarfmistletoe seed in flight. Time interval between exposures was approximately 1/1000 second. The distance traveled by the seed between exposures is indicated by the millimeter rule. Seeds are blurred because they travel about one-third of their length during the slit exposure period of 33 μsec.

single semifusiform seed (*1*). When the fruit is ripe, the pedicel is elongated and recurved so that the fruit apex points downward. An abscission zone develops between the tip of the pedicel and the base of the fruit. High hydrostatic pressure develops in a layer of viscin cells surrounding the seed, and when the fruit is severed from the pedicel the exocarp of the fruit rapidly contracts and expels the seed upward.

Hawksworth (*1*), in a preliminary study of *Arceuthobium vaginatum* f. *cryptopodum*, estimated that the initial velocity of the seed was 1370 cm/sec (45 feet per second). This figure, however, was an indirect calculation based primarily on the average height of seeds expelled directly upward and their measured terminal velocity. Since the calculation was based on average values, no statistical evaluation was possible. Hinds *et al.* (*2*), in a photographic study of seed discharge in *Arceuthobium*, showed that tumbling in the vertical plane begins soon after discharge; on the average, seeds had turned 90 degrees when they had traveled about 6 cm from the fruit. The discovery that the seeds tumble in flight raised some question as to the accuracy of the previous velocity calculations, so it was apparent that a more direct method of velocity determination, in which many seeds could be measured, was desirable.

Seed velocity was studied photographically (*3*). The equipment used was a 35-mm single-lens reflex camera equipped with an 85-mm lens on a 100-mm extension tube, a rotating disc which allowed light penetration at known time intervals, and a pendulum device for triggering an electronic flash unit (Fig. 1). A mature fruit with the pedicel end pointing into the area to be photographed was placed on the base of a pendulum 8 cm long (*2*). Seed ejection was initiated by gently heating the fruits from below. When the seed was ejected from the fruit, the recoil of the exocarp caused the pendulum to swing back to a contact point and trigger the strobe unit. The assembly was used in a dark room with the camera shutter open. The total exposure period was thus determined by the length of the light flash. A disc with 16 slits (each 1 mm wide) rotating at 60 revolutions per second was placed in front of the camera lens. The rotation rate of the motor was checked electronically several times during the experiment. Slit exposures (of 33 ×

Table 2. Initial velocity of seeds of four species of *Arceuthobium*.

| Species | No. of seeds | Velocity* (cm/sec) |
|---|---|---|
| *A. cyanocarpum* | 131 | 2130 ± 30 |
| *A. douglasii* | 125 | 2230 ± 30 |
| *A. cryptopodum* | 146 | 2540 ± 40 |
| *A. americanum* | 124 | 2610 ± 20 |

* Mean ± standard error.

$10^{-6}$ second) were made every 104 × $10^{-5}$ second. The length of the flash period for the strobe unit used was approximately 5 × $10^{-3}$ second, so this resulted in from three to four exposures of a seed in each frame (Fig. 2). The distance traveled in the known time interval was measured and the seed velocity calculated. The field of view of the camera allowed about the first 10 cm of seed flight to be photographed.

Four Colorado dwarfmistletoes were studied: (i) *Arceuthobium americanum* Nutt. ex Engelm., a parasite of *Pinus contorta* Dougl.; (ii) *A. campylopodum* Engelm. f. *cyanocarpum* (A. Nels.) Gill (here abbreviated *"cyanocarpum"*) on *Pinus flexilis* James; (iii) *A. vaginatum* (Willd.) Presl f. *cryptopodum* (Engelm.) Gill (here abbreviated *"cryptopodum"*) on *Pinus ponderosa* Laws.; and (iv) *A. douglasii* Engelm. on *Pseudotsuga menziesii* (Mirb.) Franco. Ten to twenty branches bearing dwarfmistletoe plants with mature fruits were cut, the ends were placed in water, and the branches were transported to the laboratory. Usually, velocity measurements were made on the same day as collection, and only the most mature fruits were used. Measurements were obtained from the 1963 and 1964 seed crops for each dwarfmistletoe.

Size, weight, volume, and specific gravity of seeds of the four dwarfmistletoes studied are given in Table 1. Initial velocity measurements of 526 seeds of the four species are given in Table 2. There were no statistically significant differences between the 1963 and 1964 measurements for any of the four dwarfmistletoes, so data for the 2 years were combined for each species. Velocities ranged from 2100 cm/sec for *A. cyanocarpum* to 2600 cm/sec for *A. americanum*, and averaged 2400 cm/sec for all seeds measured. The difference in velocities between seeds of *cyanocarpum* and of *douglasii* was not significant, nor was that between seeds of *cryptopodum* and of *americanum*. However, the difference between the group with the lower velocity (*cyanocarpum* and *douglasii*, 2200 cm/sec) and the group with the higher velocity (*cryptopodum* and *americanum*, 2600 cm/sec) was significant at the 1 percent level of probability.

The initial velocities are thus considerably greater than the velocity of 1370 cm/sec previously estimated indirectly *(1)*. That the previous estimate was so low can probably be accounted for by the use of a formula in which no allowance was made for the seeds tumbling in flight, and in which airflow around the seeds was assumed to be laminar—that is, the frictional force was assumed to be proportional to the seed velocity. Recent calculations based on the assumption of an equivalent sphere (with the same terminal velocity as mistletoe seeds) showed that the Reynolds number calculated from the initial velocity of the mistletoe seed was well beyond the critical velocity for the transition from laminar to turbulent flow about the seed. The initial resisting force calculated for a turbulent flow was of the order of 15 times that for laminar flow.

As is evident from the data in Tables 1 and 2, there is no direct relationship between seed velocity and seed size. *Arceuthobium douglasii* and *A. americanum* have seeds of the same size, but the former is in the group having a low velocity while the latter is in the group with a high velocity.

The two dwarfmistletoes in the group with high velocity are larger plants (the shoots usually being 7 to 12 cm high) than those in the group with a low velocity (shoots 2 to 5 cm high). Possibly the water regime of the larger mistletoes enables them to build up higher hydrostatic pressures within the fruits, thus resulting in greater seed velocities.

We do not have sufficient data to compare seed velocity with horizontal distance of seed flight for the various dwarfmistletoes. However, the longest horizontal distance of seed flight that we have measured (14.6 m, *2*) is for *A. cryptopodum*, a species in the group having a high velocity.

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## Control of Glutamine Synthetase in the Embryonic Retina in vitro

Abstract. *Glutamine synthetase activity in the neural retina of the chick embryo increases sharply during terminal differentiation of this tissue. This characteristic increase can be reproduced in cultures of retinal tissue fragments from late embryos. A similarly sharp increase can be elicited precociously in younger retina by culturing in medium with adult serum. Both the precociously elicited and the later increase in enzyme activity require continuous protein synthesis; both can be suppressed during the first 24 hours of culture by blocking RNA synthesis or by removing the adult serum. Subsequently, the increase in enzyme activity becomes progressively less dependent on RNA synthesis and on the continuous presence of adult serum. This transition is attained more rapidly in the older retina. The data suggest a progressive stabilization of the enzyme-forming system during differentiation.*

Recent work on regulatory mechanisms in microorganisms has stimulated interest in the availability of embryonic systems in which gene function and the appearance of differentiation products might be experimentally manipulated and modified. This report summarizes further features of a system previously shown *(1, 2)* to have certain advantages for such work: control of glutamine synthetase *(3)* in the neural retina of the embryonic chick in vitro and in vivo in relation to the differentiation of this tissue.

Rudnick and Waelsch *(4)* found that glutamine synthetase (GS) activity in the neural retina of chick embryos is at low levels until about the 17th day of incubation, after which time the activity increases very rapidly until af-