# EFFECT OF SEED SIZE AND PELLETIZATION ON TOBACCO SEED GERMINATION UNDER VARYING TEMPERATURE REGIMES<sup>1</sup>

By MOHAN GAWANDE<sup>2</sup>, S. C. MOHAPATRA and W. H. JOHNSON<sup>3</sup>

*Nicotiana tabacum* L., cv Speight G-28, seeds were separated into seven size groups (354-400, 400-420, 420-447, 447-473, 473-500, 500-532, 532-563, and 563-595 µm) with the use of an automated sonic silter. Sized seeds were pelletized through different commercial sources to give four different pellet types: Normal, Greenhouse, Asgrow and Royal Siuis. Pelleted and unpelleted seeds were germinated at 25 or 21 C. or under varying day/night temperature regimes: 20/15, 22.5/17.5, 25/20, 27.5/22 and 30/25 C. Under all conditions large seeds fared better than small seeds in terms of both germination time and percentage. This difference was most obvious between seeds representing two extreme size-groups.

Pelletization delayed the germination rate without affecting germination percentage, and appeared to be protective against thermal stress as compared to uncoated seeds. Asgrow and Royal Sluis pellets germinated faster than Normal and Greenhouse pellets whereas the latter two provided greater protection against thermal stress. No interaction could be detected between seed size and pellet type. It appears that the disintegration time for various pellets suspended in free water can be used as a quick and reliable indicator of their germination potential before time consuming studies are undertaken.

#### INTRODUCTION

Seed properties such as maturity (4,12,13,15), morphology (2,22), weight (3,17,18,23,24) and terminal velocity (9,10) have been correlated with the germination potential of tobacco seeds, and the latter has been used for large scale seed sorting (10). Although seed size affects germination of several crop seeds (4,7,8, 11,14,24), only preliminary information is available on its effect on tobacco seed germination (1,20). The present study was undertaken to examine this aspect in greater detail, and to compare various types of tobacco seed pellets. Mechanized seeding of tobacco seeds often requires the use of pelleted seeds, and the coating around the seed could possibly influence its germination physiology.

Present Address: Operations Research Program, NCSU, Raleigh, N.C. 27650. Some observations presented herein have been taken from the unpublished M.S. thesis by this author.

<sup>1</sup>Tobacco Biophysics Laboratory, Department of Biological and Agricultural Engineering, North Carolina State University, Raleigh, N.C. 27650 (Reprint Request Address). Contribution received November 8, 1979, Tob. Sci. XXIV: 49-52, 1980.

## MATERIALS AND METHODS

Seed Sizing and Coating: Nicotiana tabacum L. cv Speight G-28 seeds, procured through the courtesy of the Speight Seed Company, Winterville, N.C. were used throughout this study. The photoblastic property of these seeds did not change when stored for three years at -15C (21). Seed lots procured in 1974 and 1975 were separated to seven size groups using an ATM sonic sifter (Fisher Scientific Co.). One gram of seeds was first separated into three size-groups using 500, 420 and  $354 \mu$  m sieves. In the second step, seeds retained in the 500  $\mu$ m sieve were further separated to three groups using 563, 532, and 500  $\mu$ m sieves. In the last step, seeds retained in the 420  $\mu$ m sieve were separated to three groups using 473, 447, and 420  $\mu$ m sieves. Each step consisted of two runs of 10 minutes each with the cleaning of the sieves between each run. Each run consisted of 5 amplititudes of sonic sifting and 7 amplititudes of mechanical thumping action, and the timing of all operations was programmed through a built-in timer.

Unsized seeds and seeds of selected size groups were coated through the courtesies of Austria Tabak, Vienna, Austria, and Asgrow Seed Company, California. The former provided two types of pellets and the latter one type. These are designated, respectively, as Normal, Greenhouse, and Asgrow pellets (i.e.,

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coated seeds). A new kind of pellet became available in 1976 through the courtesy of the Royal Sluis Company of Holland. These pellets are designated as Royal Sluis. In the absence of complete information on the composition and methods used for various coatings, each pellet type is considered here to represent a different composition and/or procedure.

Germination: Sized or unsized seeds and pelleted or unpelleted seeds were germinated either in the laboratory or in the phytotron (Southeastern Plant Environment Laboratories, NCSU, Raleigh). Laboratory studies were conducted with petri dishes kept under continuous light at constant 25 or 21C incubation temperatures. Tests under continuous darkness were run simultaneously by wrapping selected dishes with black plastic and aluminum foil. Details of this procedure have been described earlier (21).

The phytotron study was conducted in individual control chambers set at selected temperatures (15, 17.5, 20, 22.5, 25, 27.5 or 30 C  $\pm$  0.25 C) and 100% R.H. Petri dishes lined with wet filter papers and each containing 100 seeds were incubated in the above chambers under five day/night temperature regimes: 20/15, 22.5/17.5, 25/20, 27.5/22.5 and 30/25 C. These temperature regimes were obtained by transferring each petri dish to the appropriate chamber twice daily, at 8:00 A.M. and 5:00 P.M., respectively. Treatments were arranged in a completely randomized design with three replicates each. Light received by seeds while transferring the petri dishes between chambers and while making germination counts was sufficient for germination as reported by Mohapatra and Johnson (21).

Chief germination parameters used in this study are germination time and germination percentage. Although radicle emergence is often used as the criterion for germination in physiological studies (19), the seedling stage (i.e., the cotyledonary leaf stage) was used in this study because seedling emergence is the first visible sign of germination in pelleted seeds and under field conditions. The germination time for a seed population depends on whether maximum germination percentage (G<sub>max</sub>) or a submaximal value is used. This was particularly true for Speight G-28 seeds where germination followed a normal distribution with time except for a few seeds which took so long to germinate as to skew the normal distribution curve to the right. To reduce the effects of a few late-germinating seeds on germination time, a submaximal level of germination corresponding to 98% of Gmax (which itself fluctuated between 90 to 99%) was used to determine the germination time T<sub>98</sub>). Time needed for 50% of  $G_{max}(T_{50})$  was also determined because one-half of the seed population germinated at a faster rate than the other half. The concept of  $T_{98}$  and  $T_{50}$  is very similar to that of percentiles in statistics and has been used in various forms by others (16,23).

#### **RESULTS AND DISCUSSION**

Seed Size vs. Germination: Distribution of seed sizes in Speight G-28 seed lots procured in 1974 and 1975 are shown in Figure 1. The size of the sieve which retained the seeds is used for convenience to represent the seed size. However, the actual size(s) of seeds retained by any given sieve would vary between that sieve size and the next bigger size.

Figure 2. The effects of seed size and incubation day-temperature on  $G_{\mbox{max}}$  (A) and Tee (B) of Speight G-28 tobacco seeds.



(Tobacco Science 50)

At a constant 25 C and under continuous light,  $563 \,\mu$ m seeds had a faster germination potential than seeds of other sizes in general and 420  $\mu$ m seeds in particular (**Table 1**). However, seeds of all sizes gave G<sub>max</sub> of 90% or greater provided that sufficient time (approximately 168 hours) was allowed. Under stress, such as continuous darkness and/or 21 C, when the germination of all seeds was affected adversely,  $563 \,\mu$ m seeds also germinated better than  $420 \,\mu$ m seeds (**Table 1**).

A sufficient quantity of seeds of each designated size was not available for the phytotron study. Therefore, the seven sizes were pooled to give three broad size-groups: large (532-595  $\mu$ m), medium (447-531  $\mu$ m) and small (354-446  $\mu$ m). Based on data in **Figure 1**, these three size-groups comprised approximately 35%, 60% and 5%, respectively, of the total seed population.

 $G_{max}$  (Figure 2A) for small seeds was below 90% at all day/ night temperature regimes whereas it was above 90% for unsized, large and medium seeds up to 25/20 C and decreased for unsized seeds at higher temperatures. Small seeds not only had a lower  $G_{max}$ , but also needed a longer time to reach  $G_{max}$  as compared to other seed lots. Thus,  $T_{98}$  (Figure 2B) for small seeds was greater than that for other size-groups at all temperatures except 27.5/22.5. As expected,  $T_{98}$  for all seed groups decreased with an increase in day/night temperature regimes up to 27.5/22.5, but at 30/25 C,  $T_{98}$  for large seeds remained nearly unchanged whereas that for other seeds increased between 30 to 60 hours perhaps due to thermal stress. Thus there are obvious functional differences between the large and small seeds although the physiological basis for such differences is not clearly understood.

Pelletization vs. Germination: Under laboratory conditions (i.e., 25 C and continuous light), Royal Sluis and Asgrow pellets germinated faster than normal and greenhouse pellets (Figure 3), although all pellets eventually gave  $G_{max}$  values greater than 90%. Better performance of the Royal Sluis and Asgrow pellets was associated with the fact that these pellets split within minutes of contact-hydration whereas normal and greenhouse pellets did

Table 1. The effects of light, darkness, seed size, and incubation temperatures on the germination time and percentage of Speight G-28 tobacco seeds. 168 hr. is the total time needed for  $G_{max}$  at 25 C. All germination percentages are adjusted to the nearest whole numbers.

DRUIGNATED NEED DEZE µm	L1GHT				DARKNESS	
	25 C		21	C	25 C	21-2
	1.10	168	144	168	168	168
	hr			hr		
5,6. B	€ C	93	68	82	31	.20
1, (°)	4. j	9,2	60	81	14	я
1.00	22	94	-58	81	7	$\epsilon_{i}$
473	<i>.</i>	93	6,9	78	7	8
447	14	ĢĻ,	-0	78	8	А
4.20	r	90	44	7F:	0	1,
Unsized	44	95	59	78	8	r.,

not split except under the pressure of the emerging plumule (Figure 3) after nearly 168 hours of contact-hydration. Royal Sluis pellets germinated faster than Asgrow pellets (compare developmental stage and number in Figure 3). This may be related to the fact that Royal Sluis pellets are more fragile and thus provided greater seed exposure than Asgrow pellets (Figure 3). When all pellet types were suspended separately in water, the Royal Sluis pellets disintegrated in less than 15 seconds, exposing the seeds in the process. (Figure 3), and Asgrow pellets distintegrated between 25 to 35 seconds. But the Normal and Greenhouse pellets disintegrated only partially during the 24-hour suspension period. These properties closely parallelled the germination behavior of the respective pellets and thus provided a quick and dependable method for predicting the germination potential prior to the time consuming germination studies.

Figure 3. Comparative features of four pellets and unpelleted seeds after 168 hours of imbibition. A-unpelleted seeds, B-Royal Sluis, C-Greenhouse, D-Normal, E-Asgrow. Note: B,C,D,E in the bottom row are corresponding pellets suspended in water to show their disintegration properties. C1 is a  $\times$  30 magnification of a Greenhouse pellet, indicated by arrow in the 'C' row, showing the plumular hook (ph) which breaks the pellet.



(Tobacco Science 51)



Figure 4. The effects of pelletization methods and incubation day-temperature on T98 and T50 of Speight G-28 tobacco seeds.

Pelletization affected only germination time but not Gmax. Under varying alternate temperature regimes and discontinuous light, as expected,  $T_{98}$  and  $T_{50}$  (Figure 4) of unpelleted seeds and all pellets decreased with increase in day/night temperature up to 27.5/22.5 C, these values for Asgrow pellets being lower than Normal and Greenhouse pellets. At 30/25 C,  $T_{98}$  and  $T_{50}$ for unpelleted seeds and to a lesser extent  $T_{50}$  for Asgrow pellets increased considerably, perhaps because of thermal stress, while T<sub>98</sub> for all three pellets and T<sub>50</sub> for Normal and Greenhouse pellets decreased further. Apparently, pelletization provided a certain degree of protection against thermal stress by reducing the rate of temperature change. If so, and as indicated by T<sub>98</sub> and T<sub>50</sub> data (Figure 4), Normal and Greenhouse pellets which enclose the seed longer would be expected to provide greater protection than Asgrow pellets which expose seeds upon hydration. Asgrow pellets are, however, preferred over Normal and Greenhouse pellets from the standpoint of

germination time which is an important consideration for field operations.

Although 20 C has been reported to be the optimum temperature for germination of some tobacco seeds (6), this study showed that a constant 25 C under continuous illumination or 27.5/22.5 C day/night temperature under periodic illumination was optimal in terms of  $G_{max}$  of Speight G-28 seeds. However, the germination time under the latter was nearly twice as long as under continuous light at 25 C without any loss in the final germination percentage.

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