A Comparison of Methods and Blowers For the Purity Analysis Of Kentucky Bluegrass Seed

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CONTENTS

SUMMARY

Three methods for conducting purity anaylsis of Kentucky bluegrass seed were compared. They were: (1) hand method—preliminary separation made with a blower and the final differentiation of questionable florets by hand, (2) standard method—separation with a blower using the same pure seed criteria as for the hand method and (3) climax method—separation with a blower at a level which gives the maximum pure live seed percentage.

Three blowers (the Ottawa, Erickson and Ames) were included in the study. Four laboratories cooperated in the investigation. Variations within laboratories were compared; variations between laboratories were not considered.

More nearly uniform purity analyses and germination results were obtained with the climax method than with either of the other methods. The variability of test results was approximately the same for the hand and standard blowing methods. The hand method required twice as much time as either of the other procedures.

More variable pure seed percentages were obtained with the Erickson blower, when the climax and standard blowing methods were used, than with either of the other blowers. The germination and pure live seed percentages were equally uniform regardless of the blower used.

The data indicate that special procedures for lightweight Kentucky bluegrass seed are unnecessary.

The merits of the climax blowing method suggest that it should be adopted in preference to the hand or standard blowing methods. This procedure also should be applicable to other small-seeded grasses.

A Comparison of Methods and Blowers for the Purity Analysis of Kentucky Bluegrass Seed [1]

BY L. E. EVERSON[2]

Seed analysts and seedsmen have desired to reduce the differences between purity analyses of Kentucky bluegrass (*Poa pratensis*) made by different analysts or different seed laboratories. Variability between germination tests has also caused concern. The purity analysis of Kentucky bluegrass using the "hand" method is tedious and time consuming. Seed analysts would like to reduce the time required to make purity analyses.

This study had three objectives. The first objective was to determine whether the least variable purity and germination results are obtained with the "hand," "standard," or "climax" method of purity analysis. The second objective was to learn which of the three blowers currently in use (the Erickson, Ottawa and Ames) produces the least variable results. Some analysts have presumed that it might be possible to use the "standard" or "climax" blowing methods for purity analysis of heavier seed (20-pound bushel weight or above) but that lighter seed would require special methods. The third objective was to determine whether this assumption is correct.

Only variations within laboratories were compared; variations between laboratories were not considered.

The "hand" method is the procedure outlined for the purity analysis of Kentucky bluegrass in the Association of Official Seed Analysts' "Rules for Seed Testing" (3). Using this method, a blower is employed as an aid in separating the empty florets from the pure seed. However, the final separation of questionable florets is made by visual observation using such tools as tweezers and transmitted light. The Rules[3] also permit the use of a "standard" blowing method[4] which provides a mechanical separation of bluegrass florets into pure seed and inert matter with a seed blower. The standard blowing gives purity results approximately equivalent to those obtained by the hand method. The standard blowing has not been generally used by analysts because they have felt that further study was needed. Early workers also considered the use of the "climax" blowing method. The climax blowing is made with a blower setting that provides somewhat greater air pressure than the standard blowing. As a consequence, the purity percentage is lowered, and the germination is raised. The pure live seed percentage[5] (purity x germination) is maximum $\frac{}{(100)}$ at the climax blowing.

LITERATURE REVIEW

EARLY HISTORY

The "Irish" method was the first recorded procedure used for the purity analysis of small-seeded grasses. All material of any species under consideration was classified as pure seed as long as it resembled a "seed" in external appearance. The word "seed" included seeds, fruits and florets commonly referred to as seeds in the trade. Later, seed analysts decided that it was not scientific to classify undeveloped florets of grasses, or structures in which the caryopses were lacking, as seeds. Accordingly, the definition of pure seed was changed to include only florets or structures containing a caryopsis. In the case of grasses, this was determined by tweezer pressure or by examination over transmitted light. In the purity analysis of Kentucky bluegrass, this is now referred to as the hand method.

VARIATIONS BETWEEN ANALYSTS USING THE HAND METHOD

Musil (9), Porter and Leggatt (13), West (15) and others have agreed that one of the major causes of differences in purity and germination results on Kentucky bluegrass is the presence of immature and shriveled caryopses which are difficult to classify as pure seed or inert material. Porter and Leggatt (13) pointed out that it was

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[1] Project 1083 of the Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa.

[2] The writer wishes to acknowledge the cooperation received from the Federal Seed Laboratory, Beltsville, Md., the New York Seed Laboratory, Geneva, N. Y., and the Purdue Seed Laboratory, Lafayette, Ind., in conducting research herein described. Marche McMahon made the purity analyses for the Iowa State College Seed Laboratory.

[3] The Association of Official Seed Analysts' "Rules for Seed Testing" will be referred to as Rules in the remainder of the bulletin.

[4] "Standard" blowing method—the term "uniform" blowing method is used in the Rules. This term has been confusing. Analysts have preferred to use the term standard blowing or standard blowing method; therefore, the word standard is used in lieu of uniform.

[5] Pure live seed will be abbreviated P.L.S. in the remainder of the text.

possible for a single analyst in a laboratory to attain a satisfactory level of uniformity in purity results but that uniformity rarely extended to the results obtained on samples tested by different laboratories. They felt that poor agreement between tests was caused by the inherent character of the material involved, differences in personal interpretation and faulty operation or design of old type seed blowers. Musil (9) pointed out that analysts may injure caryopses when using tweezer pressure to determine whether florets are empty or have caryopses. Such injury might cause variability in germination results. The Research Committee of the Association of Official Seed Analysts (2) reported the results of referee tests made on Kentucky bluegrass, comparing the hand and standard blowing methods. Twenty-six laboratories took part in the study. The report showed that differences obtained for purity and germination results for the hand method were in excess of what should be expected from samples of a homogeneous lot. Repeated tests on a low purity sample ranged from 60.5 to 82 percent in purity and from 52 to 87 percent in germination. Repeated tests on a high purity sample ranged from 90.9 to 96 percent in purity and from 61 to 91 percent in germination. Porter (12) stated "We have conclusively shown that the differences in purity percentages, obtained by different analysts with representative samples from uniform lots of bluegrass, are far greater than can be accounted for on the basis of natural variation." Aberg *et al.* (1) made a study of the uniformity of tests made by the hand method. Twenty-two laboratories took part in the study. They learned that differences between tests were far greater than should be expected from homogeneous samples. In making purity analyses on a lot of bluegrass seed with an average purity of 92 percent, they found a difference of 5.4 percent between subsample extremes.

TESTING PROCEDURES SUGGESTED TO REPLACE THE HAND METHOD

Direct method. Musil (9) proposed a "direct" method for purity and germination tests on Kentucky bluegrass seed. Using a 0.25-gram sample, bluegrass florets (whether empty or containing caryopses) were separated from all other material. The entire floret portion of the sample was planted for germination. Seeding value was then calculated on the basis of the number of normal seedlings. She compared the direct with the hand method and concluded, "These data show that the plant-producing power of a sample can be determined equally well by either method. The variation in the number of seedlings for repeated tests is approximately the same by either procedure." She found that preparation of seeds for germination by the direct method required only one-fourth to one-eighth the time required for the hand method. As later pointed out by Porter and Leggatt (13), this procedure may be the most accurate measure of seed value because it gives the number of viable seeds per unit weight. However, the requirements of state seed laws make it necessary to determine both a purity and germination percentage and to indicate these values on the label. Any procedure used for the analysis of small-seeded grasses must give this information. Analysts have also criticized the direct method because it has a tendency to favor seed lots with small seeds, since such lots usually produce more seedlings in a laboratory test. It has been pointed out that seedlings from small seeds probably do not have as good a chance for survival in the field as seedlings from large seeds.

Binocular method. Cullinan (5) suggested the "binocular" method for the purity analysis of Kentucky bluegrass. The procedure consisted of examining 0.1 gram of seed under transmitted light with a binocular microscope and separating the bluegrass florets into pure seed and inert material. This effected a considerable saving of time. However, Aberg *et al.* (1) found more variability of purity results for the binocular than for the hand method.

Approximation method. Torpy *et al.* (14) made comparative studies of the "approximation" and standard blowing methods. By the approximation method, a sample was blown at successively increasing air pressures until an examination of the blowings indicated that all empty florets had been removed. The last blowing was examined for fertile florets and these placed into pure seed. The dirt, stems, weeds and other crop seeds were removed from the remaining heavy seed. Torpy *et al.* concluded that more nearly uniform test results were obtained through the use of this procedure than by the standard blowing method. Porter (11) statistically analyzed the data of Torpy *et al.* and concluded that the air pressure used by them for the standard blowing procedure was variable. Porter found that the differences were greater for the approximation than for the standard blowing method.

Standard blowing method. As early as 1935 Brown and Porter (4) recommended the adoption of the standard blowing method. Porter's (10) later investigations showed that essentially a complete separation of empty florets and florets with caryopses could be obtained by the standard blowing when a uniform speed motor and a vertical air blast separator of the Holland type were used. By this method Porter (11) obtained purity results within the range of natural variability. A comparison of the hand and standard blowing methods demonstrated better uniformity for both purity and germination with the standard blowing. Aberg *et al.* (1) concluded from their studies that the standard blowing would give a more nearly correct evaluation of bluegrass seed than the hand method, provide a standard procedure and require less time. Porter (12) recommended that seed analysts adopt the standard blowing procedure for the determination of purity of small-seeded grasses. He maintained that uniform procedures

and consistent results were of primary importance; therefore, theories and concepts should be modified as necessary. He reiterated that the standard blowing procedure was a sounder basis for testing seed because errors of personal judgment were largely eliminated.

Climax blowing method. Leggatt (6) explained that if samples from a seed lot were blown at progressively increasing air pressures and the heavy portions germinated, the P. L. S. percentages obtained would show a series of increasing values up to a maximum or "climax" and remain relatively constant for several air pressure increases before starting to decrease. Leggatt further explained that the only valid estimate of the P. L. S. percentage was the highest point obtainable in such a series. He considered that this figure revealed the true value of the seed and could be interpreted as the number of pounds of viable seed per hundred. Leggatt (7) recommended that consideration be given to the definition of pure seed according to a synthetic sample prepared for the climax blowing. He listed the advantages of the climax blowing as follows: (1) a saving of time, (2) greater uniformity, both between laboratories and between analysts within a laboratory, (3) less necessity for retesting because the heavier blowing produces a sample which is less variable from which to draw seeds for germination, (4) the P. L. S. figure derived by this procedure more nearly reflects the actual number of pounds per hundred than the hand method and (5) less tedium and eye strain for the purity analyst.

Porter and Leggatt (13) asserted that since pure seed is given in percentage by weight, germination in percentage by number, the use of these two percentages in evaluating seed lots is accurate only when the unit weight of the viable and nonviable seeds in the pure seed fraction is approximately the same. Porter and Leggatt stated that we must avoid retaining immature sterile seeds and florets in the pure seed fraction because they cause a decrease in germination that is disproportionate to the increase in purity.

Leggatt (8) described a modification of the climax blowing used by Canadian seed analysts which consisted of one blowing, about 0.4 millimeter less on the Ottawa manometer scale, than the climax blowing. Canadian analysts make one blowing for Kentucky bluegrass samples at the prescribed setting. Weed and other crop seeds are removed from the light and heavy portions. Stems, leafage, sand, etc., from the heavy portion are added to the empty florets and other inert from the light portion to give the total amount of inert matter. Leggatt asserted that this procedure had been used with satisfactory results and had helped speed Canadian testing services.

MATERIAL AND METHODS

The seed laboratories that took part in this study were the Federal Seed Laboratory, Beltsville, Md., the Purdue Seed Laboratory, Lafayette, Ind., the Iowa State College Seed Laboratory, Ames, Iowa, and the New York State Seed Laboratory, Geneva, N. Y. The first three laboratories mentioned made the purity and germination tests; the New York Laboratory divided and distributed the samples.

Three Kentucky bluegrass samples (one each of 18-, 21- and 24-pound bushel weights) were used. Each of these was a composite of seed samples obtained from four seed companies located in different bluegrass seed producing areas of the country. These composite samples were used to gain diversity within the samples without increasing the number of samples to be worked. The three methods compared were the hand, standard and climax.

The performances of the Erickson, Ottawa and Ames blowers were compared. The purpose of a seed blower is to separate light material (i. e., empty florets, chaff, etc.) from the heavier pure seed. How well this is accomplished depends upon the efficiency of the blower and differences in size and weight of the pure seed versus the light material.

Some of the more important features common to all three of the blowers are:

(1) Separations are accomplished by air pressure produced by an electric motor and fan.
(2) For each blower, the 1-gram sample of seed for purity analysis is placed in a cup for blowing. In each instance the base of this cup is made of close mesh screen to hold the seed but to permit passage of the air.
(3) Each blower has a hollow tube (1 to 1½ inches in diameter and 18 to 24 inches long depending upon kind of blower) which is inserted into the seed cup. The tube is held in a vertical position. The separation occurs in this tube.
(4) A seed trap or tray is used to catch the light material blown from the pure seed.

Each of the blowers has certain special features different from the others. The Erickson blower uses a ⅓-horsepower motor. It has a plastic air column with two seed traps set near the top on opposite sides of the tube to catch the light material. The air control is at the top of the tube included with the screen cap that prevents the escape of seed from the top. This blower does not have a pressure chamber; the fan forces air directly into the base of the seed cup.

The Ames blower is dissimilar to the Erickson blower in two important respects. A metal tube is substituted for the plastic tube, and the air pressure is controlled by a worm adjustment at the base of the seed cup. The metal tube prevents seeds from sticking to the sides of the tube because of static electricity. The worm adjustment permits accurate air control.

The Ottawa blower uses a 1/75-horsepower motor. A specially treated glass tube eliminates

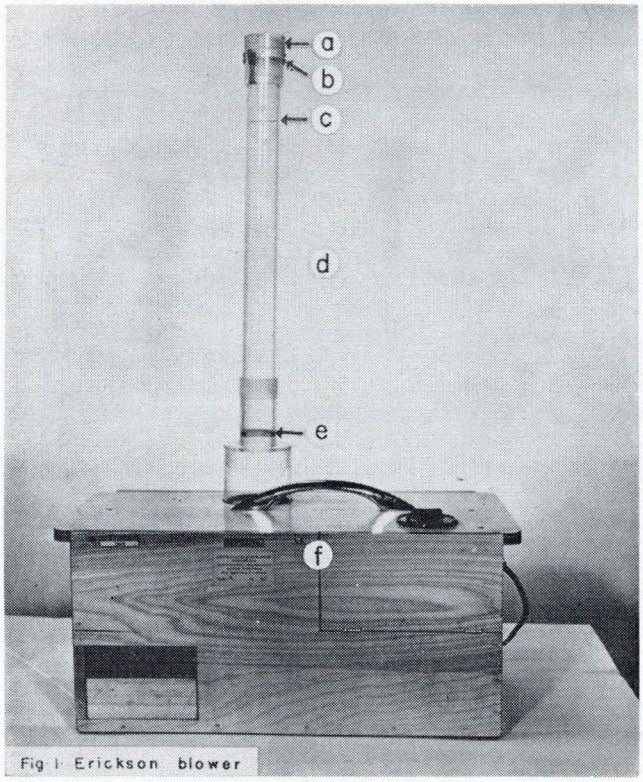

Fig. 1. Erickson blower: (a) air control, (b) screen, (c) inert or seed traps, (d) plastic air column, (e) seed cup, (f) enclosed motor and fan.

static electricity. Instead of seed traps, the light material is blown through the curved, open top into a larger chamber where it settles on a tray or piece of paper which may be removed. The air is controlled by a gate operated by a worm gear. The amount of air pressure entering the bottom of the seed cup is measured with a manometer which is adjustable to offset the effects of barometric pressure and humidity. The Ottawa blower has a pressure chamber which may somewhat offset irregularities of the motor.

The principal features of the three blowers are shown in figs. 1, 2 and 3.

The size of the subsamples used for the purity analysis was between 1.0 and 1.1 grams. The blowing time for the standard or climax method was 3 minutes with the Erickson and Ottawa blowers, and two 1½-minute periods with the Ames blower. Since the trap for catching light material in the Ames blower is small, it is not possible to catch all of the light seed if only one blowing is used. The blowing time for the hand method was the same as normally used by each laboratory in conducting a purity analysis on Kentucky bluegrass. This was probably different at each laboratory because the Rules do not specify a given length of time. Three separations were made on each subsample of seed, regardless of the method used. The pure seed was identified as "separation 1"; the empty florets, classed as inert material, were identified as "separation 2"; and

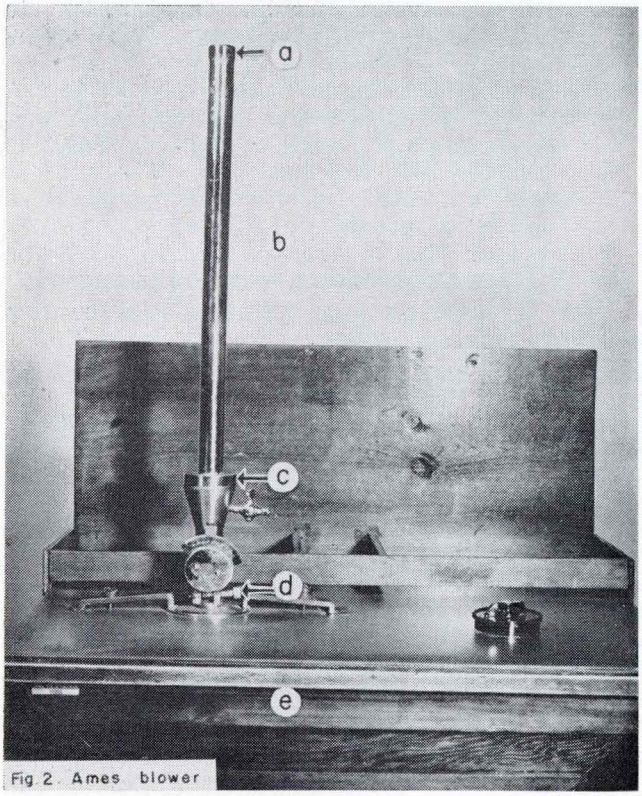

Fig. 2. Ames blower: (a) screen cap, (b) metal air column, (c) seed cup, (d) air control, (e) enclosed motor and fan.

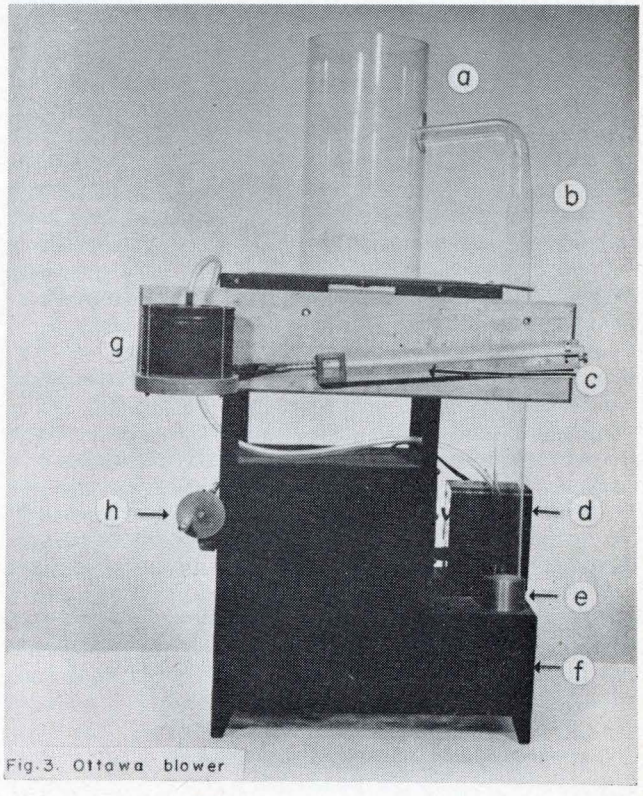

Fig. 3. Ottawa blower: (a) settling chamber, (b) glass air column, (c) manometer scale, (d) motor, (e) seed cup, (f) air pressure chamber, (g) kerosene reservoir for manometer, (h) air control.

material other than pure seed or inert florets (including other crop seeds, weed seeds, stems, dirt, etc.) was identified as "separation 3."

STATISTICAL DESIGN

An analysis of variance of the pure seed percentages was computed for each laboratory and for the three laboratories combined. A split-plot design was used at each laboratory. The whole-plot treaments were bushel weights (3). The sub-plot treatments were the nine combinations of methods (3) and blowers (3).

DIVIDING

The New York State Seed Laboratory divided and shipped the subsamples to participating laboratories. Each seed sample (of a given bushel weight) was handled as follows: 81 individual subsamples (1.0 to 1.1 grams) were divided from each sample. These subsamples were individually sealed in packets and the packets mixed. The 81 subsamples were divided into nine random groups of nine subsamples each. Three groups were randomly assigned to each laboratory to make up the three replicates of that bushel weight. The nine subsamples of each group were then randomly assigned a purity analysis method and blower (three methods x three blowers = nine subsamples). After the dividing had been completed for one sample (bushel weight), the next two were handled in a similar manner. Upon completion of all dividing, the 81 subsamples (nine groups of nine subsamples each) for each laboratory were brought together. Each packet was marked with the method and blower to be used. The order of working the groups was also randomly assigned and written on the packets. The subsamples were then mailed to the three participating laboratories.

PREPARATION OF STAINED SAMPLES

Stained samples had to be prepared which each participating laboratory might use to determine the standard blowing point on each of their three blowers. Seed to be used for the calibration of the standard blowing point was obtained by having the participating laboratories make purity analyses on one sample each of the 18-, 21- and 24-pound bushel weight seed. The hand method, as outlined in the Rules, was used for these purity analyses. Each laboratory returned the pure seed and inert empty florets. The pure seed from all laboratories was mixed together. The inert florets were also mixed. The pure seed was then stained with a 1-percent solution of safranin in alcohol. The inert florets were left unstained. After drying, the stained seed was divided into 0.92-gram samples, and approximately 0.15 gram of empty florets was added to each of these. Six samples were prepared in this manner.

For each of the six samples the point of "minimum error" was determined on the Ottawa blower. The point of minimum error was determined by making the first blowing at a low level and increasing the blowing pressure at each subsequent blowing. At the point of minimum error, 3.2 on the manometer scale, the number of seeds misplaced (heavy stained seeds blown into the light unstained seeds and vice versa) was approximately equal. Each of the six samples was then blown twice at 3.2, and all misplaced seeds were removed and discarded.

The pure seed from the six samples was again bulked and mixed together, and similarly the inert material was mixed. The pure seed was then divided into 0.90 gram samples, and 0.14 gram of empty florets was added to each. The point of minimum error was again checked for each of the six samples (see table 1). Approximately the same number of seeds was misplaced for each sample; therefore, no further adjustment was made.

Determination of the climax blowing point was also necessary before climax stained samples could be prepared for setting the blowers. Eleven 1-gram subsamples were divided from the 18-, 21- and 24-pound samples. These were blown at 0.2 intervals on the Ottawa blower, using successive manometer settings of 3.1 to 5.1. For each subsample the inert empty florets, along with the weeds, other crop seeds, stems, dirt, etc., from the heavy portion were weighed together as a single separation. The pure seed was weighed and the percentage determined. The pure seed from each subsample was divided and half sent to the Purdue Seed Laboratory for germination test. The other half was germinated in the Iowa State College Seed Laboratory. The germination results for each of the three bushel weights at each particular blowing were averaged for the two laboratories and purity and germination curves were drawn (see fig. 4). The blower setting at the maximum percentage of P. L. S. was designated as the climax blowing point (see fig. 5). After determination of the climax blowing point, three 1-gram subsamples were divided from each of the 18-, 21- and 24-pound bushel weight samples. Each of the nine samples was blown at 3.9 with the Ottawa blower. All material, other than bluegrass florets, was removed from the inert empty florets and from the pure seed. The pure seed of all subsamples was bulked together and the inert florets bulked together. The pure seed was then stained with a 0.1-percent crystal violet in alcohol solution and the inert florets left unstained. Preparation of the stained samples for setting the blowers at the climax blowing point was carried out in the same manner as for the standard stained sample, except that the blowing point of 3.9 rather than 3.2 was used.

The procedure employed for calibrating each blower at each participating laboratory for the standard or climax blowing points generally was similar regardless of the blower used. Each blower was calibrated by determining the point of minimum error as described.

Each laboratory recorded the information for

the purity analysis (separations 1, 2 and 3), the germination and the length of time required to work each sample on a standard form to facilitate processing of the data.

RESULTS

STANDARD AND CLIMAX STAINED SAMPLES

Before the samples of stained seed were distributed to each laboratory for calibrating their blowers, the point of minimum error was checked on each sample to be certain it was consistent for all samples. The results obtained on the standard stained samples are recorded in table 1. Since the results obtained for the climax stained samples were similar to those of the standard stained samples, a second table is omitted. For each of the six samples (table 1), the number of unstained seeds left in the stained seed was high when the blower was set at 3.0. The number of stained seeds blown into the unstained seed was high when the blower was set at 3.4. The number of seeds misplaced (heavy stained seeds blown into the light unstained seeds and vice versa) was almost identical at 3.2 for all six samples.

DETERMINING THE CLIMAX BLOWING POINT

Data obtained in determining the climax blowing point were recorded and used to plot the curves shown in figs. 4 and 5. Figure 4 shows the purity and germination curves plotted from the average of the 18-, 21- and 24-pound bushel weight seed.

As the blower settings were increased from 3.1 to 5.1, the purity percentage decreased consistently, forming almost a straight line from 3.1 to 4.5. From 4.5 to 5.1 the decrease in purity was more rapid. The germination curve increased rapidly at the lower blower settings but tended to "level off" at the high settings. The P. L. S. percentages calculated from these curves and recorded in fig. 5 show that the highest percentage was obtained near 3.9. It will be noted that the P. L. S. percentages remained relatively consistent near this point.

Each of the P. L. S. curves are similar even though the samples were widely different in bushel weights. Each curve increased rapidly from a blower setting of 3.1 to 3.5. From 3.5 to 4.3 there was very little change in the pure live seed content. From 4.3 to 5.1 the P. L. S. percentages decreased rapidly. It is evident that the P. L. S. curves obtained for 18-, 21- and 24-pound bushel weight samples responded quite similarly at any given blower setting, since all three curves were almost identical in form.

TABLE 1. OTTAWA BLOWER SETTINGS TO CHECK POINT OF MINIMUM ERROR FOR STANDARD STAINED SAMPLES.

Sample number	Blower setting	Number of stained seeds in unstained seeds	Number of unstained seeds in stained seeds
1	3.0	3	86
	3.2*	15	24
	3.4	32	6
2	3.0	2	122
	3.2*	18	17
	3.4	29	6
3	3.0	4	96
	3.2*	27	20
	3.4	49	1
4	3.0	3	46
	3.2*	22	15
	3.4	59	3
5	3.0	1	101
	3.2*	14	24
	3.4	44	2
6	3.0	2	94
	3.2*	25	16
	3.4	50	3

* Point of minimum error was 3.2 in all cases.

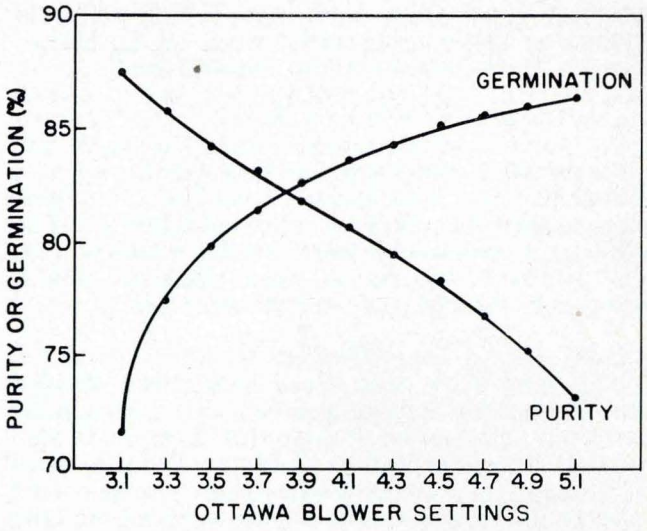

Fig. 4. Purity and germination curves for average of 18-, 21- and 24-pound bushel weight Kentucky bluegrass seed.

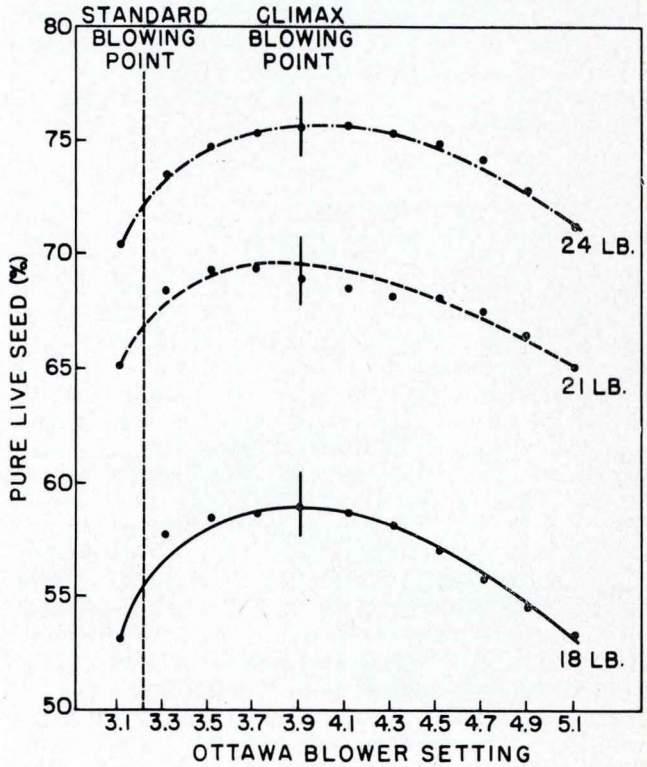

Fig. 5. "Pure live seed" curves for 18-, 21- and 24-pound bushel weight Kentucky bluegrass seed.

TABLE 2. ANALYSES OF VARIANCES FOR PURE SEED, GERMINATION AND PURE LIVE SEED PERCENTAGES. DATA FROM THREE LABORATORIES COMBINED.

Source of variation	Degrees of freedom	Mean squares		
		Pure seed	Germination	Pure live seed
Laboratories (L)	2	30.46**	152.16**	207.39**
Bushel weight (W)	2	51,113.77**	609.99**	4,956.38**
LW	4	1.30	21.43**	73.97
Pooled error (a)	18	1.27	9.12	28.68
Blowers (B)	2	10.04**	44.12**	10.94*
Methods (M)	2	514.64**	973.38**	108.59**
BM	4	1.54**	10.63*	6.04
WB	4	0.46	1.51	1.36
WM	4	33.29**	38.98**	7.60*
WBM	8	0.55	7.61*	4.51
LB	4	1.50**	9.10*	6.21
LM	4	12.63**	51.43**	28.36**
LBM	8	1.67**	10.79**	5.11
LWB	8	0.59	3.68	1.95
LWM	8	0.39	4.05	5.80
LWBM	16	0.40	1.55	2.06
Error (b)	144	0.39	3.49	3.00

* Significant at 5-percent level.
** Significant at 1-percent level.

COMPARISON OF METHODS AND BLOWERS

The analyses of variances for the pure seed, germination and P. L. S. are combined in table 2.

Previous studies have shown differences between the test results of different laboratories. Significant differences for laboratories (see table 2) confirm these differences. Differences between bushel weights were significant. It is obvious that different purity, germination and P. L. S. percentages would be obtained for different bushel weight seed; therefore, further comment is unnecessary.

Significant differences for laboratories x weights were obtained for germination only. This indicates that the laboratories obtained reasonably consistent results for the different bushel weight samples (except for germination).

Differences were obtained for blowers indicating that the results obtained for one blower were not the same as those obtained for another.

The differences between methods were expected because increased air pressure was used for the climax blowing method.

Blowers x methods was significant for pure seed and germination; we can conclude, therefore, that the results with the blowers were, to a certain extent, changed by the methods. We may also conclude that the reverse was true (i. e., the results with different methods were influenced by blowers). Blowers x methods was not significant for P. L. S.

Weights x blowers was not significant, indicating that the differences between bushel weights were reasonably consistent for all three blowers.

Weights x methods was significant; therefore, we may conclude that the results obtained for weights were influenced by the methods used.

Weights x blowers x methods is a three-way interaction. No attempt will be made to interpret three- or four-way interactions.

Laboratories x blowers was significant for the pure seed and germination percentages, indicating that the results obtained for blowers were influenced somewhat by laboratories. We may also conclude that the converse was true. The interaction of laboratories x blowers was not significant for the P. L. S.

Laboratories x methods was significant. We may conclude that the results with methods were influenced, to a certain extent, by the laboratories.

The pure seed, germination and pure live seed means are given in tables 3, 4 and 5. These data are included to permit more critical evaluation of table 2.

An analysis of variance can give the information that the results obtained from three methods were different. An analysis of variance, however, does not indicate which of the methods checked was different from the others or which method had the least variability between tests. Therefore, "tests of uniformity" were made and the data recorded in tables 6, 7 and 8.

The data presented in tables 6, 7 and 8 are pooled results from the three bushel weights and three laboratories. The data in table 6 tabulate variations in purity percentages; table 7, germination; and table 8, P. L. S. To determine if one method produced less variable results than another, the reader should use the variance figure under the method in question as the divisor and the method to be compared as the dividend. If the quotient obtained is greater than the F value given below the table, significantly less variable results were obtained by the method in question.[6] By checking figures horizontally across the table,

[6] For example in table 6, if the figure below hand method opposite the Ottawa blower (0.7085) is divided by the figure under the climax method opposite the Ottawa blower (0.2330) the quotient is 3.04. Checking the F table at 18 degrees of freedom we find that 3.04 is between the 5- and 1-percent levels of significance. This indicates that when the climax method was used with the Ottawa blower, significantly less variable results were obtained than when the hand method was used.

TABLE 3. PURE SEED MEANS. DATA FOR LABORATORIES COMBINED.

Pounds bushel weight	Blower	Method		
		Hand	Standard	Climax
18	Erickson	79.34	78.51	72.82
18	Ames	78.67	77.28	72.11
18	Ottawa	79.70	78.63	72.09
21	Erickson	87.38	87.00	82.62
21	Ames	86.51	86.10	82.76
21	Ottawa	87.32	86.82	83.08
24	Erickson	93.53	93.40	90.60
24	Ames	92.90	92.87	90.67
24	Ottawa	93.69	93.40	90.73

TABLE 4. GERMINATION MEANS. DATA FOR LABORATORIES COMBINED.

Pounds bushel weight	Blower	Method		
		Hand	Standard	Climax
18	Erickson	72.78	77.44	81.61
18	Ames	75.39	76.44	83.33
18	Ottawa	71.39	75.00	82.78
21	Erickson	79.28	81.00	86.06
21	Ames	80.17	82.50	85.56
21	Ottawa	78.50	80.67	85.11
24	Erickson	79.39	81.22	85.67
24	Ames	81.56	82.00	84.50
24	Ottawa	79.00	81.61	84.11

TABLE 5. PURE LIVE SEED MEANS. DATA FOR LABORATORIES COMBINED.

Pounds bushel weight	Blower	Method		
		Hand	Standard	Climax
18	Erickson	57.71	60.80	59.43
18	Ames	59.30	59.07	60.11
18	Ottawa	56.84	58.98	59.08
21	Erickson	69.23	70.48	71.10
21	Ames	69.36	71.03	70.81
21	Ottawa	68.51	71.23	70.72
24	Erickson	72.48	73.80	76.82
24	Ames	73.73	74.33	75.44
24	Ottawa	71.91	73.87	75.17

TABLE 6. VARIANCES FOR PURE SEED PERCENTAGES.

Blower	Degrees of freedom	Testing method: Hand	Standard	Climax	Degrees of freedom	Pooled results for blowers
Erickson	18	0.3819	0.8167	0.6719	54	0.6235
Ames	18	0.5185	0.4552	0.2296	54	0.4011
Ottawa	18	0.7085	0.4063	0.2330	54	0.4493
Pooled results for methods	54	0.5363	0.5594	0.3782	162	0.4913

F values

D. F.	5%	1%
18, 18	2.61	3.57
54, 54	1.80	2.18

TABLE 7. VARIANCES FOR GERMINATION PERCENTAGES.

Blower	Degrees of freedom	Testing method: Hand	Standard	Climax	Degrees of freedom	Pooled results for blowers
Erickson	18	6.6204	5.7964	2.0741	54	4.8303
Ames	18	5.7408	3.2315	2.6667	54	3.8796
Ottawa	18	4.3519	3.4167	3.1574	54	3.6420
Pooled results for methods	54	5.5710	4.1482	2.6327	162	4.1173

F values

D. F.	5%	1%
18, 18	2.61	3.57
54, 54	1.80	2.18

TABLE 8. VARIANCES FOR PURE LIVE SEED PERCENTAGES.

Blower	Degrees of freedom	Testing method: Hand	Standard	Climax	Degrees of freedom	Pooled results for blowers
Erickson	18	8.7389	6.6537	3.1100	54	6.1675
Ames	18	5.1570	4.6022	2.4030	54	4.0541
Ottawa	18	5.8074	11.1608	4.9937	54	7.3206
Pooled results for methods	54	6.5678	7.4722	3.5022	162	5.8474

F values

D. F.	5%	1%
18, 18	2.61	3.57
54, 54	1.80	2.18

any method using a particular blower may be compared with another method using the same blower. Similarly, by checking vertically, any blower may be compared with another blower when the same testing method is used. A small variance figure means less variability of a method or blower than a large variance figure.

The results given in table 6 indicate that more nearly uniform purity results were obtained with the climax method than with the standard or hand method when the Ottawa blower was used. The standard method did not give significantly less variable results than the hand method. The pooled results for methods show a somewhat lower variance for the climax method. Both the Ottawa and Ames blowers gave more nearly uniform purity results than the Erickson blower when the climax method was used.

The data in table 7 indicate that the least variable germination percentages were obtained when the climax blowing method was used to make the purity separations. The results obtained by the standard method were not significantly less variable than the hand method. Uniformity of germination was unaffected by the kind of blower.

The results presented in table 8 indicate that more nearly uniform P. L. S. percentages were obtained with the climax blowing method than with the hand or standard blowing method. The standard blowing method did not produce less variability than the hand method. Significantly less variable P. L. S. percentages were obtained with the Ames blower than with the Ottawa blower.

The amount of time required to work a sample

TABLE 9. MINUTES REQUIRED FOR A KENTUCKY BLUEGRASS PURITY ANALYSIS—AVERAGE OF THREE LABORATORIES

Bushel weight	Testing method: Hand	Standard	Climax
18 lb.	116	59	59
21 lb.	77	37	38
24 lb.	62	29	29
Average of all bushel weights	85	42	42

by the three testing methods is recorded in table 9. It is evident that much time may be saved by use of either the standard or climax blowing method.

DISCUSSION

Only data which may aid in drawing conclusions concerning methods, blowers, weights and time will be considered in this section.

It was noted that variance for blowers x methods was significant for pure seed and germination (i. e., the results obtained with the blowers were changed by the methods and vice versa). Reference should be made to table 2. The variances (table 6) indicate that there were no significant differences between blowers for pure seed when the hand method was used; however, the variance for the Erickson blower was somewhat smaller than for the Ottawa and Ames blowers. Variability of a blower may be completely altered by an analyst; therefore, the differences between blowers, when the hand method was used, better reflects performance of analysts rather than blowers. The results recorded in table 6 show that less variable purity results were obtained with the Ames and Ottawa blowers than with the Erickson blower when the standard or climax methods were used. This is a reversal of the trend obtained with the hand method; therefore, the reason for the significance of blowers x methods in table 2 is apparent.

It was noted that the variance for blowers x methods was not significant for the P. L. S. percentages—even though the interaction of blowers x methods was significant for pure seed and germination percentages. Germination for a given sample usually increases as purity goes down (also, the converse is usually true). P. L. S. percentage is the product of pure seed x germination; therefore, P. L. S. percentage would tend to be more consistent than either the pure seed or germination percentages.

The interaction of weights x methods was significant. The reason for this was apparent in the results recorded for individual bushel weights (not shown in this bulletin). The least variable results were obtained on 24-pound bushel weight seed with the hand method. The least variable results on 18- and 21-pound seed were obtained with the climax method. The reason for less variable results with the hand method for 24-pound seed is that there are very few questionable seeds in the 24-pound seed; therefore, it is relatively easy for an analyst to detect the few seeds which have been misplaced by the blower. Many questionable florets occur in light bushel weight seed; consequently, when the hand method is used, it

is much more difficult for the analyst to obtain uniform pure seed percentages on 18- and 21-pound seed.

If we refer to tables 6, 7 and 8, it will be noted that the pooled results for methods show less variable results for the climax method than either the hand or standard methods. Figures 4 and 5 are helpful in interpreting the reason for more nearly uniform germination and P. L. S. results for the climax method. The germination curve in fig. 4 increased rapidly at the lower blower settings, then tended to "level off." Uniformity of blower performance depends on blower design, accuracy in setting the dial and variability of the electrical supply. Figure 4 demonstrates that germination is markedly affected by a slight change in dial setting at the lower blower settings. Therefore, if an analyst inadvertently set the dial at 3.1 instead of 3.2 (the standard blowing point), a 4-percent change in germination would occur. A similar error in setting near the climax blowing point would affect the germination result less than 1 percent. Variability in the electrical supply would have the same effect.

The P. L. S. percentage is also less affected by a blower setting error at the climax blowing point. The curves in fig. 5 indicate that a change in setting alters the P. L. S. percentages considerably at the lower blower settings but very little near the climax blowing point. This confirms data presented by Leggatt (7).

One of the objectives of this study was to determine whether it was necessary to handle light bushel weight seed differently than heavy bushel weight seed. It is evident from the P. L. S. curves in fig. 5 that different bushel weights responded in a similar manner. Therefore, the climax blowing procedure should be satisfactory for all samples. This was the conclusion reached by Leggatt (7).

CONCLUSION

More nearly uniform test results were obtained with the climax blowing method than with either the standard blowing or hand method. The standard blowing method did not produce better uniformity than the hand method. The results indicate that the germination and P. L. S. percentages are considerably altered by small errors in blower settings near the standard blowing point but are little affected by errors at the climax blowing point.

Less variable pure seed percentages were obtained with the Ottawa and Ames blowers than with the Erickson blower when the standard or climax blowing method was used. This advantage was not apparent for germination or pure live seed percentages.

The data indicate that seed of all bushel weights can be tested by the same method. Special procedures for light weight seed should not be necessary.

Twice as much time was required to test seed by the hand method than by the standard or climax blowing method.

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