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Comparison of seed conditioning procedures for *Eucalyptus grandis W.* Hill ex Maiden using seed sizing, specific gravity table and general blower

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Summary

Eucalyptus grandis seed cleaning is particularly difficult due to the small size of the seeds. When large seed quantities are involved, the usual method of screening followed with an air blower is too time consuming resulting in inefficiencies and bottlenecks in the process. Different seed cleaning processes are analyzed to evaluate their effectiveness in removing the chaff from the seeds. The combination of seed sizing with the specific gravity table gives the best results, averaging a 47.3% increase in the number of germinants. Although screening before the specific gravity table gives better results, with an 20.4% increase in the number of germinants, it is not significantly different from using the specific gravity table before screening. Germination was higher with screening after gravity table than with the gravity table alone at both the upper and lower positions of the deck.

Introduction

Seed conditioning is an important step in seed processing and has a great influence in seedlot quality (Stein *et al.,* 1974; Edwards, 1980; 1993). In the cleaning process, different operations are adjusted to consider specific seed or fruit characteristics such as size, shape and density (Belcher *et al.,* 1984; Simak, 1984; Vieira *et al.,* 1989; Bergsten, 1993).

"Eucalyptus seeds" are a mixture of fertile seeds, sterile structures, unfertilized ovules, and fruit structures, the last three components are known as "chaff' (Piotto, 1994). Size and specific gravity of fertile seeds and chaff are frequently similar making separation difficult. Seed purity is normally as low as 10 to 15% for Rose gum, *Eucalyptus grandis* W. Hill ex Maiden.

After the screening process, seed blowers are frequently the next step which separates the lighter weight infertile seed from the fertile seeds. But, when seedlots become too large for air blowers to handle the capacity in a timely manner, additional equipment needs to be considered. Cavalcanti and Gurgel (1973) used a vibratory separator to clean up to 100 kg of fruit and seeds per hour, yielding 10 kg of clean seed.

E. grandis seeds are produced in southern Florida, USA, for an increasing market and a more efficient way of cleaning large quantities is needed. The current method of screening and using an air blower to clean seeds takes up too much of time, resulting in inefficiencies and bottlenecks in the process. A small table top air blower being used is named the General Blower and is designed to separate chaff from very small size seeds, but can only handle a small amount of seed with each operation; thus, making the machine very inefficient for cleaning large seedlots. Using the specific gravity table would increase yield and speed up the process, but because the equipment is expensive, an investigation was initiated comparing the machine to standard methods.

Material and methods

Germination tests

Germination tests were used as an indicator of success and X-rays were taken to evaluate the separation of the chaff from the seeds. The image was enlarged two times in the X-ray machine so the image could be viewed without magnification equipment.

Germination tests quantified the response to each procedure. A germination test was conducted on the original seedlot to compare with the seed cleaning treatments. Blue blotter paper was laid over Kimpak® and wetted with 70 ml of tap water in each dish. A 0.10 gram seed sample was sprinkled into each of 4 dishes. Germination dishes were placed in a germination room set at 20° Celsius (68°F) for 16 hours of darkness and 30° Celsius (86°F) with 8' hours of light according to International Seed Testing Association Rules of Seed Testing (The Australian Tree Seed Centre uses a constant 25°C. for this species). Germination counts were taken every 2 to 4 days ending the count around the 10th day. Germination was recorded when the radicle emerged. Cotyledons appeared around the 3rd to 5th day.

Seedling counts are relative and are indicative of the procedure's success : the higher the seedling count the more successful the procedure. The actual number is not important, just its' relationship to the other seedling counts. It is usually infeasible to count individual seeds with this species; the seeds are too small to distinguish from the chaff with the naked eye. Germination is not calculated by percentage for *Eucalyptus seeds,* because a specific number of seeds are not planted per dish but planted by weight. The number of germinants in the figures and tables represent the average number of germinants over 4 germination dishes per 0.10 gram sample.

Seed conditioning

A seedlot of 725.08 grams of *Eucalyptus grandis* was used to compare the effectiveness of the specific gravity table with and without seed screening. Hand held screens were used to size the seeds. Four approaches were studied:

1) seed sizing alone;
2) specific gravity table alone;
3) specific gravity table followed by seed sizing;
4) seed sizing followed by specific gravity table and seed blower.

Figures accompany each procedure outlining each step (figures 1 to 4). From past experience with the species's seed and trial and error the most appropriate screens, 6 x 40 and 6 x 50, were selected. Screen numbers represent the number of wire per inch.

For experiments 2, 3 and 4, an Oliver 30 specific gravity table with a rectangular linen deck was used. The linen's weave prevented the microscopic seeds from falling through the deck but did not impede air flow. Deck area was cut in half with a board running lengthwise and the deck was tilted at its maximum slant. Air was blown up through the deck; thereby, stratifying the seed with light material floating on top of the heavier material. Values of air speed and control operations are indicated further. Shaking the deck in eccentric circles caused the heavier material to move up to the highest point of the slant while the lightest material remained at the lower end. Shake and air used simultaneously caused the material to move down the deck toward the chute end where the material fell through chutes into containers. Cut gates, located at the deck's end, directed the material into three chutes.

For experiment 4, an air blower named, the General Blower, was included in the conditioning process. The General Blower consists of an air tunnel .where forced air is blown up through the seeds, resulting in the lightest weight material moving up the column into a stainless steel cup on top of the machine.

1) Seed sizing alone (Method 1)

A seed sample (0.40 grams) was extracted from the seedlot for the original germination test before *any* cleaning was begun, and a small seed sample (approximately 100 grams) was withdrawn to be screened with hand held screens. Screen 4 x 40 was tried first but screen 6 x 40 yielded the best separation between the chaff and the seeds. An additional screen 6 x 50 was used to further upgrade the sample. Figure 1 illustrates the procedure.

Seeds that fell through the 6 x 40, 6 x 50 and 6 x 60 screen were labeled as trash. A microscope was used to view the seeds for determination of screening effectiveness. Chaff was a lighter color than the seed, so color was used as another indicator of separation.

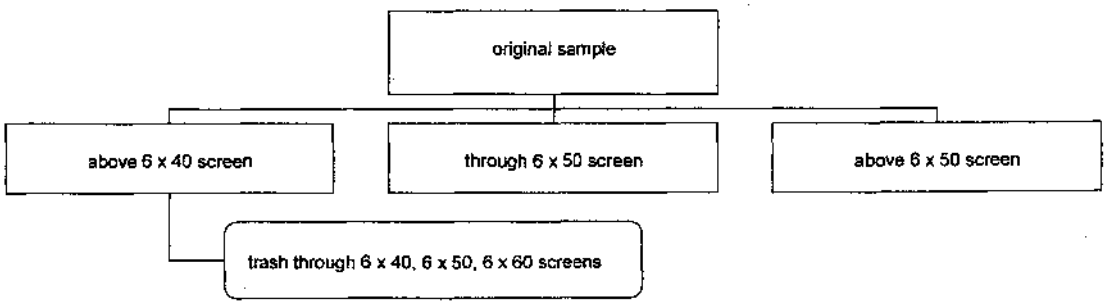

Figure 1. Seed sizing by hand held screens (6 x 40, 6 x 50 and 6 x 60 wires per inch). (Method 1)

2) Specific gravity table alone (Method 2)

The majority of the seedlot (approximately 720 grams) was run over the specific gravity table without sizing. Seeds from the sizing procedure were added back to the sample for gravity table procedures 2, 3, and 4. A board was laid on top of the deck reducing the

rectangular deck area by half. The machine was run at 383 rpm with air speed of 2 on the electrostat 60 controller. The gravity table deck was set at maximum slant; thereby, allowing the seeds to move up the table while the lighter chaff floated down to the lowest part of the table. Due to the small size of the seedlot, the seeds were not allowed to fall off the table for fear of losing the microscopic seeds. After the separation, the seed on the table was divided horizontally into upper and lower decks and vertically into chute end and middle of the deck resulting in 4 subsamples: 1 - middle upper; 2 - middle lower; 3 - upper chute end; 4 - lower chute end (figure 2). The majority of the seeds were located in the middle upper part of the deck.

3) Specific gravity table followed by seed sizing (Method 3)
Seeds (approximately 705 grams) from the gravity table divisions described in the previous procedure were run through 6 x 40 inch screen. Seeds that fell through the screen were labeled as trash (figure 3).

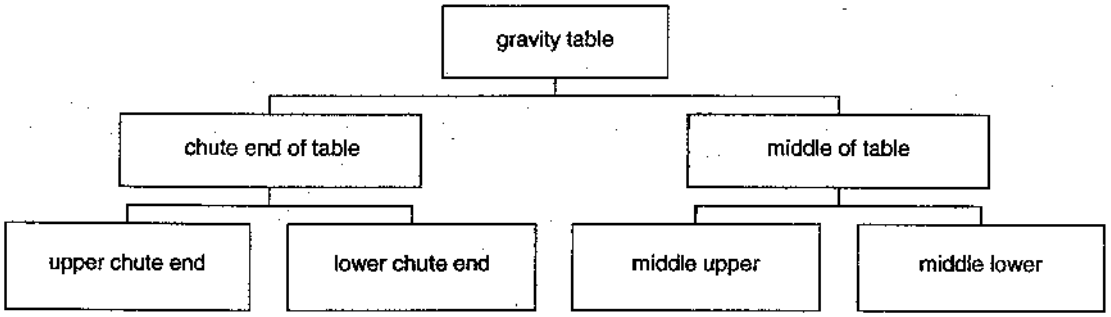

Figure 2. Specific gravity table alone (383 rpm, air speed of 2 on the electrostat 60 controller, table deck at maximum slant). (Method 2)

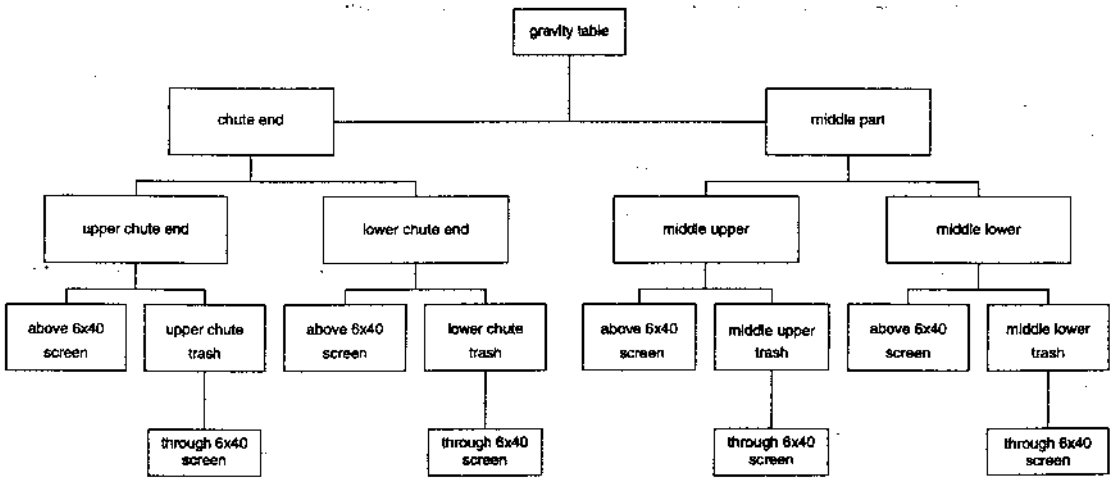

Figure 3. Specific gravity table followed by seed sizing (6 x 40 wires per inch screen). (Method 3)

4) Seed sizing followed by specific gravity table (Method 4)

Seeds (approximately 695 grams) were screened through 6 x 40 and 6 x 50 screens before performing the weight separation which is the usual procedure when using the specific gravity table. Seeds from each screen size were divided into two sections and labeled upper and lower to represent their position on the table deck (figure 4). Seeds that did not pass through both screens and were in the lower part of the specific gravity table, were rerun over the specific gravity table again in an attempt to remove more bad seed. Seeds were subdivided horizontally on the table and labeled as lower-upper and lower-lower.

To remove more chaff, the General Blower was used on the upper and lower seed samples from the gravity table. Side vents of the blower remained closed leaving only the front aperture open to admit air. A couple of teaspoons of seeds were placed in the bottom cup of the blower each time the machine was run. The front aperture was gradually opened to 13, 15 for seeds from the upper part of the table deck and a front opening of 10 for seeds from the lower part of the table deck and allowed to operate for about 1 to 2 minutes. Seeds that blew up into the column and landed in a cup on top of the machine did not germinate as well as the seeds that remained in the bottom of the blower.

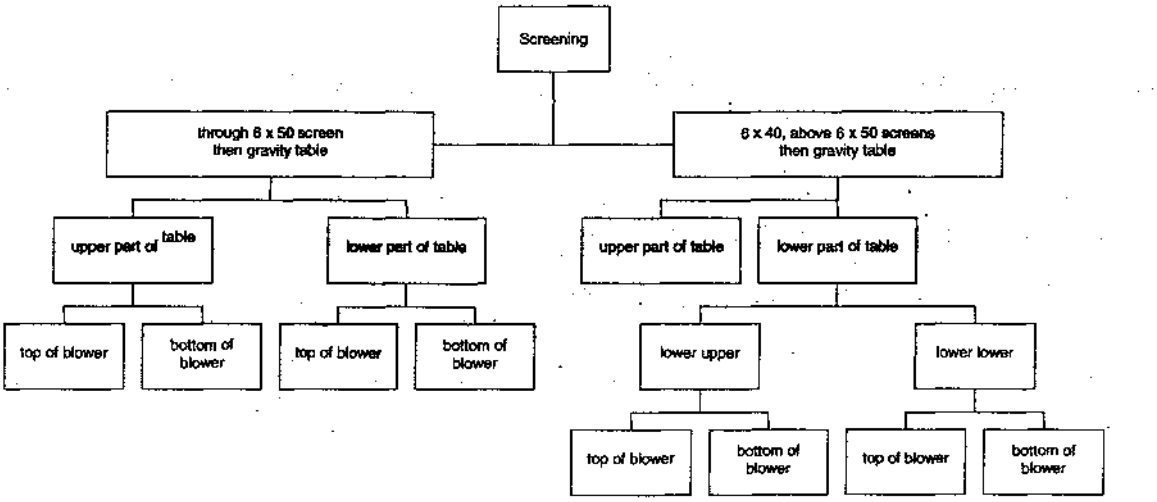

Figure 4. Seed sizing by hand held screens (6 x 40 and 6 x 50 wires per inch) followed by using of a specific gravity table (383 rpm, air speed of 2 on the electrostat 60 controller, table deck at maximum slant). (Method 4)

Statistical analysis

An analysis of variance using SAS General Linear Models- Type III sum of squares and a Duncan's means comparison test were performed following Steel and Torrie (1986). The alpha level used was P = 0.01. A logarithmic transformation of data was done for the analysis to correct for heterogeneity of variances. The analysis evaluated differences in the average number of germinants from different steps within each cleaning method, and compared the average number of germinants between methods 1 through 4. Chaff, labelled as trash, was not in the analysis since it did not have any interest in terms of the cleaning results. The two best steps of each cleaning method were compared and analysed to determine the best step within each method.

Results

Table 1 presents the results of the analysis of variance and mean comparison test for the different steps within each method of seed cleaning. Table 1 presents the value of F - statistic and probability at a significance level of 1%. Steps with the same letter are not significantly different from each other. The average number of germinants from the germination tests for the different cleaning methods is also presented.

Table 1. Results of the statistical analysis, comparing all steps of each method. *F* – statistic from ANOVA for each cleaning method (M1 to M4), and group subsets from mean comparison test for steps within each method.

Method	M1: Sizing w/o gravity table	M2: Gravity table, no sizing	M3: Gravity table then sizing	M4: Sizing, gravity table, and blower
F - statistic	52.153 (Prob < 0.0001)	405.723 (Prob < 0.0001)	27.426 (Prob < 0.0001)	72.520 (Prob < 0.0001)
Group subsets comparison; number	a-Original - 49	a-Lower chute end-5	a-Middle lower, above 6x40-90	a- through 6x50, lower + top-13
	a-Through 6x50-66	a-Middle lower-6	a-Lower chute end, above 6x40-111	b- 6x40, above 6x50, lower lower-21
	b-Above 6x50-104	b-Middle upper-130	b-Middle upper, above 6x40,-156	b- 6x40, above 6x50, lower lower + top-23
	c-Above 6x40-163	b-Upper chute end-134	b-Upper chute end, above 6x40-197	c- through 6x50, lower-53
				c- 6x40, above 6x50, lower upper + top-65
				cd- through 6x50, upper + top-69
				de- 6x40, above 6x50, lower upper + bottom-104
				e- 6x40, above 6x50, lower lower + bottom-133
				e- 6x40, above 6x50, upper-150
				f- through 6x50, lower + bottom-198
				f- through 6x50, upper + bottom-227

Table 2. The two steps with the highest number of germinants from each method were analysed and compared with the other methods to ascertain significant statistical differences:

Method	*Steps*
Method 1 (M1)	Above 6 × 50; above 6 × 40
Method 2 (M2)	Middle upper; Upper chute end
Method 3 (M3)	Middle upper, above 6 × 40; Upper chute end, above 6 × 40
Method 4 (M4)	Through 6 × 50, lower + bottom; Through 6 × 50, upper + bottom

Results are represented in figure 5 showing the average number of germinants for 2 steps with each method. Steps with the same letter were not significantly different.

From the means comparison test, the steps broke out into 4 groups. `M4: Through 6 x 50, upper + bottom', 'M4: Through 6 x 50, lower + bottom' and `M3 Upper chute end, above 6 x 40' had the highest average number of germinants. Ml above 6 x 50 was significantly different from all the other steps (table 2). Step 'Ml above 6 x 40' was not significantly different from the `M4: Through 6 x 50, lower + bottom', `M3 middle upper chute, above 6 x 40' and `M3 Upper chute end, above 6 x 40' (table 1). In Method *3, the* number of germinants from the `upper chute end, above 6 x 40' was not significantly different from the M3: `middle upper, above 6 x 40' step. Seed sizing after the specific gravity table raised the average number of germinants by 22 to 63 seedlings from Method 3.

As more steps are added to the conditioning process, the average number of germinants increases from method 1 to 4.

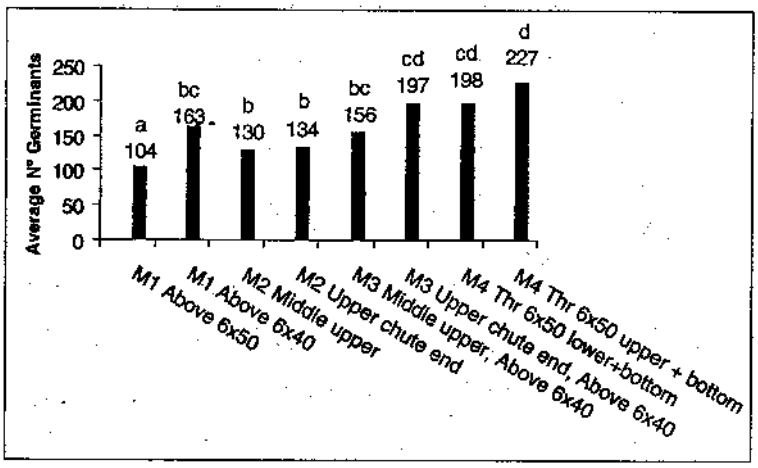

Figure 5. Average germination counts of the two best steps for each seed cleaning method. Different letters above bars indicates means that are statistically different (P = 0.01) using Duncan's multiple range test. M1 to M4 indicates the four methods into analysis (M1: Sizing without gravity table; M2: Gravity table, no sizing; M3: Gravity table then sizing; M4: Sizing, gravity table, and blower).

The original sample weight was 725.08 grams and the final total weight was 680.24 grams, resulting in only a 6% loss of 44.84 grams between the original and cleaned sample. Seeds were used for germination tests and the rest, mostly chaff, blew away during the cleaning process. The final weight of pure seed was 106.21 grams and the trash weighed 574.03 grams: a proportion of 16% seed to the total weight. Hodgson's research reported that ***E. grandis*** produced 20% viable seed by weight (Boland ***et al., 1980).***

Results within method

Seed sizing alone (Method 1)

In seed sizing without specific gravity table (figure 1), 4 steps were analysed with 3 treatments being significantly different from each other. Screen size, above 6 x 40, had the highest average number of germinants, 163 (table 1). The original sample averaged 49 germinants per dish, which was significantly less than the seeds that did not fall through

the screens. Screening separated the smaller seed and chaff from the seedlot resulting in higher number of germinants. The above 6 x 40 seeds were bigger than the above 6 x 50 seeds and yielded a significant difference of 59 germinants.

Specific gravity table alone (Method 2)

In the specific gravity table alone procedure, figure 2, table position was analyzed with two combinations of steps: `Lower end + Middle lower' and `Middle upper + Upper chute end'. There were about 127 more germinants from the upper part of the deck than from the lower part of the deck and they were significantly different. Table position at `Upper chute end' had the highest germination 134, but was not significantly different from the `Middle upper' 130, so the horizontal location of the seeds on the table did not influence the number of germinants. The heaviest seeds moved up the slanted table while the lighter seeds moved down, thus separating the seeds by vigor (figure 2). Specific gravity table positions `Middle upper' (130) and `upper chute end' (134) gave better results than 'lower chute end' (5) and `middle lower (6) and were the best positions to maximize germination.

Specific gravity table followed by seed sizing (Method 3)

The gravity table then seed sizing procedure was analysed with 4 different steps and the upper deck position was significantly different from the lower end of the deck. This is the same result as demonstrated in use of the gravity table without seed sizing (Method 2), except sizing the seeds after the gravity table operation resulted in an increase of 26 to 63 germinants from the upper deck and an increase of 84 to 96 germinants from the lower deck. Table position `Upper chute end, above 6 x 40' had the highest average number of germinants but was not significantly different from the `Middle upper, above 6 x 40', so the seeds' location on any part of the upper part of the table did not influence the number of germinants.

Table position `lower chute end, above 6 x 40' and middle lower above 6 x 40' were not significantly different from each other, so the seeds' location on any part of the lower part of the table did not influence the number of germinants.

Seeds that fell through the 6 x 40 inch screen had a much lower number of average germinants than seeds that did not fall through the screen for all areas of the deck and were labeled as trash (figure 3).

Seed sizing followed by specific gravity table (Method 4)

Finally, the seed sizing then gravity table procedure was analysed resulting in 6 combinations of steps. Combining seed sizing, the gravity table, and the General Blower yielded the highest number of germinants per dish. Step `through 6 x 50, upper + bottom', had the highest average number of germinants, 227, and was not significantly different from step `through 6 x 50, lower + bottom', 198, but was significantly difference from all the other steps (table 1). The General Blower was added as an additional step to remove more chaff and yielded the highest average number of germinants for all procedures. At this point most of the seeds were separated from the chaff so that the seeds could be becounted for individually and a traditional germination test could be performed.

Seeds from the upper part of the gravity table and screened with sizes 6 x 40 and 6 x 50 would have had a higher number of germinants if run through the General Blower. A much larger number of germinants were realized when seeds from the lower part of the gravity table deck was further cleaned with the General Blower. More chaff was removed leaving a greater proportion of seeds to be planted in the germination tests.

Discussion

Eucalyptus grandis seed cleaning is particularly difficult due to the small size of the seeds. Several conditioning methods have been used to remove chaff from *Eucalyptus* seeds. Walters and Geary (1989) used standard sieves to sift the chaff from the seeds resulting in 85% pure seed (species not identified). Purity percent was increased with an air blower that blew away more chaff. The number of mesh per square inch required to separate seeds for *E. maculata* was 12 (77 per cm^2), for *E. citriodora* was 14 (90 per cm^2), for *E. botryoides* 26 (168 per cm^2), *E. alba 30* (194 per cm^2), and for *E. paniculata, E. robusta, E. saligna* and *E. tereticornis 32* (206 per cm^2) (Cavalcanti and Gurgel 1973). With Endecotts sieves of gauge 500 μm, Edmundo Navarro de Andrade in Brazil greatly increased seed purity and reduced the seedlot weight by 50% (Hodgson 1974). The Australian Tree Seed Centre uses a mesh aperture of 1.2 mm for conditioning *E. grandis* seeds. Hodgson (1977) used sieve sizes 500 pm, 600 pm, and 710 pm to clean *E. grandis* seeds and the germination for each size was 96, 99, and 88% respectively. As the seeds became smaller in the seedlot, the germination decreased. The smallest seeds that passed through the sieves became mixed with the chaff and the seeds germinated and grew slowly resulting in poor survival of the germinants.

The principles of seed conditioning are demonstrated with these results. Equipment used in tandem in the proper order upgrades seed quality and increases germination. Seed sizing alone gave slightly better results than the gravity table alone, but the difference was not statistically significant. Without the specific gravity table, the largest seeds that did not fall through screen 6 x 40 had comparable germination rates with the gravity table alone, method 2, and gravity table then sizing, method 3. Seed sizing alone eliminated the smallest chaff but did not remove chaff the same size as the seeds. This chaff was included in the germination text, so fewer seeds were planted per dish, which resulted in lowering the number of germinants for method 2.

Specific gravity table without sizing, method 2, did not produce the as many germinants as when sizing was included (method 3 or 4). Because size and weight were confounded, the specific gravity table could not detect a difference between seed size and seed weight simultaneously. The heavier seeds and chaff moved to the upper part of the deck while the lighter seeds and chaff moved to the lowest part of the deck. Methods 2, 3, and 4 had a higher average number of germinants. Normally, heavy seeds have higher germination than light seeds. The average number of germinants was higher when seed sizing was included with the specific gravity table process, resulting in a 47.3% increase in germinants.

Germination counts were higher with screening after gravity table than with the gravity table alone at both the upper and middle positions of the deck, resulting in a

33.7% increase in the number of germinations. Although the screening before the gravity table gave better results, with an increase of germinants of 20.4%, the differences were not significantly different from the upper deck portions in method 2.

Since sizing the seeds after the gravity table did not increase the efficiency of the specific gravity table, it is traditionally performed before the specific gravity table operation. Eliminating or reversing steps in the process may save time, but decreases equipment efficiency and does not maximize germination.

Seed sizing in conjunction with the specific gravity table separated more chaff and poor germinating seeds from the seedlot demonstrating that the specific gravity table can separate small differences in weight when seedlots are homogeneous. As the seedlot became more subdivided in the process, a point was reached where the amount of seed was too small to run the specific gravity table efficiently, so the General Blower was included, because this blower is designed to work with small seeds and small quantities. The General Blower had its' greatest effect on seeds from the lower part of the gravity table's deck because it was able to further separate the good germinating light seeds from the poor germinating light seeds. Had there been sufficient seed in the lower part of the table, the light seeds could have been run over the table again probably resulting in the same effect as the General Blower.

The specific gravity table is a piece of equipment that can handle large quantities of seed in a short period of time, and is designed to upgrade seed quality through the removal of trash and seed in a degraded condition; thereby, improving germination. It separates seed by weight and is most effective in increasing germination when the seed is uniform in size. In Zambia, Guldager (1973) used a specific gravity table to clean E. *grandis* seeds resulting in purity and germination percentages of 95%; a bulked seedlot had a 97% germination. Running seeds over the deck of the specific gravity table five times did not lead to seed degradation (Guldager, 1973).

There were significant differences in number of germinants from the horizontal positions on the table's deck but no significant differences between the vertical placement. The heavier seeds from the upper table deck had higher number of germinants than from the lower table deck. It did not matter if the seed was in the middle of the table or at the chute end. Upper and middle deck positions had the highest number of germination while the lower end and middle lower deck positions had the lowest number of germinants. Vieira *et al.* (1989) with *E. grandis* also reports similar results.

Unlike blowers and aspirators, the specific gravity table is designed to handle large quantities of seeds in a short time period; therefore, it is an effective method for conditioning *E. grandis* seeds. Specific gravity table efficiency is increased when the seedlot is homogeneous. Even though, the specific gravity table is an expensive piece of equipment the expense would be justified by the large increase in purity and. germination.

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