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Germination and seedling growth response of refined and standard bedding plant seed to temperature and vapor pressure deficit

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by

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INTRODUCTION

High germination percentages and vigorous seedling growth and development are essential for the production of high quality bedding plants. Traditionally, bedding plants have been produced in flats containing approximately 6 cm of soil. Seeds were sown in furrows, covered with soil, and placed into the greenhouse for germination. After germination, crowded seedlings competed for light, water, and nutrients. Germination percentages were difficult to determine because the exact number of seeds sown was unknown. Separation of seedlings for transplanting into marketable packs required intensive hand labor and provided an additional opportunity for seedling damage. Once transplanted, the surviving seedlings grew to marketable size and were sold to the consumer.

Many growers recently incorporated plug-production systems into their bedding plant production schemes. Seed is machine sown singly into specially designed flats containing 200 to 648 individual cells. Each seed has a specific soil volume in which to grow, and this eliminates the majority of the competition faced by the seedlings in traditional greenhouse flats. Transplanting from plug flats into marketable packs is done at approximately the same time as transplanting from traditional flats, but the time required to produce saleable packs is greatly reduced with plug-grown seedlings. The decrease in time may result from less transplanting stress because the entire plug is inserted into the pack without disruption of the root system, and there is less direct handling of the seedling.

Germination of each seed is of particular importance to plug growers. Germination counts of 70% or less have been observed in single-sown plug flats of many bedding-plant annuals. This may be due to a low quality seed source, a less-than-optimal germination environment, improper seed storage, or mechanical damage to the seed. In response to these problems in seed germination and seedling establishment, many companies have developed their own procedures to clean and select seed that is characterized by increased germination and more vigorous seedling growth.

Refined seed is derived by thoroughly cleaning bulk seed lots and by separating seed according to weight, size and density, but the cleaning and separation procedures vary among seed species and seed companies. The separated seed fractions are more uniform physically than the original bulk lot. These fractions are tested for germination and vigor; and those with the highest percentage of viable seedlings are designated refined seed. If refined seed could eliminate low quality seed as the cause of low germination, then environmental factors such as temperature and moisture could be studied more accurately.

The objectives of this research were: 1) to study the effect of temperature on germination of refined and standard seed of selected cultivars of petunias and impatiens, 2) to study the effect of temperature and vapor pressure deficit (VPD) on germination, growth, and flowering of 2 petunia cultivars, and 3) to determine if several cultivars of refined and standard seed of petunia react similarly under the same environmental conditions.

SECTION I. GERMINATION OF REFINED AND STANDARD SEED OF SELECTED PETUNIA AND IMPATIENS CULTIVARS

Germination of refined and standard seed of selected petunia and impatiens cultivars

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ABSTRACT

Refined (High Energy™ seed, Ball Seed Co.) and standard seed of 2 cultivars each of petunia and impatiens were germinated in petri dishes at 20, 25, and 30°C. Germination counts, number of seeds unable to shed their seed coats, number of abnormal seedlings, and number of moldy seeds were recorded. Percentage germination and germination rate were calculated from daily germination data. There were no differences between refined and standard seed in percentage germination, germination rate, number of abnormal seedlings or number of moldy seeds. Temperature had a significant effect on the rate of germination of both species. As temperature increased, the rate of petunia seed germination also increased. Impatiens seed germination rate increased as temperature increased from 20 to 25°, but it decreased dramatically at 30°.

INTRODUCTION

Rapid, uniform seed germination is essential for the production of high-quality bedding plants, and germination of each seed is of particular importance for bedding plants produced in plug flats. Germination counts of 70% or less have been observed in single-sown plug flats of petunia and impatiens. This may be due to low-quality seed, less-than-optimal germination environment, improper seed storage, or mechanical damage to the seed. In response to these problems in germination and seedling establishment, many seed companies have developed their own procedures to clean and select seed that is characterized by increased germination and more vigorous seedling growth.

Refined seed is derived by thoroughly cleaning bulk seed lots and by separating seed according to weight, size, and density, but the cleaning and separation procedures vary among seed species and seed companies. The separated seed fractions are more uniform physically than the original bulk lot. These fractions are tested for germination and vigor, and those with the highest percentage of viable seedlings are designated refined seed. In most cases, a positive correlation of seed size to seed performance has been found. Previous research has found that large seed germinated more rapidly and at higher percentages than small seed and that the larger seeds produced more vigorous seedlings (3). If refined seed could eliminate the seed as the cause of low germination, then other factors contributing to the production of vigorous seedlings could be studied.

The optimum germination temperature for most seed lies in the range of 15 to 30^{o}C (2). Because specially constructed germination areas are used by most bedding-plant producers, studying the effects of a range of temperatures on seed germination provides useful information for selecting sowing dates and scheduling successive crops (5). Seed vigor has been defined as that condition of active good health and natural robustness in seeds which, upon planting, permits germination to proceed rapidly and to completion under a wide range of environmental conditions (7). Refined seed, characterized as producing more vigorous seedlings, should perform better than the standard seed over the selected temperature range if this definition for seed vigor is true. The objectives of this research were to study the effect of temperature on germination of refined and standard seed of petunias and impatiens in petri dishes.

MATERIALS AND METHODS

Standard and refined seed (High Energy™ seed, Ball Seed Co., West Chicago, IL) of *Petunia* x *hybrida* 'White Cascade' and 'Blue Flash' and *Impatiens wallerana* 'Super Elfin Salmon' and 'Super Elfin White' were tested at temperatures of 20, 25, and 30°C because these temperatures bracket the recommended ranges for germination of petunias and impatiens (1). Plastic petri dishes 9 cm in diameter were lined with 8 sheets of absorbent toweling (Kimpack, Kimberly-Clark Corp., Rosewell, GA), and the toweling was covered with 1 sheet of Whatman No. 1 filter paper. Ten ml of deionized water (>18 megohm resistance) were added to each petri dish. Twenty-five seeds of each cultivar and type (standard and refined) were placed on top of the moistened filter paper, and the petri dishes were closed, sealed with plastic film (Parafilm, American Can Co., Greenwich, CT), and placed in growth chambers.

Chambers were maintained at constant temperatures of 20, 25, and 30°C (+/- 0.5°) with continuous fluorescent light at 135 μmole s^{-1} m^{-2}. Germination counts were taken daily for 15 days. Seed was considered germinated when cotyledons were expanded completely. Germination rate was determined by Maguire's equation with a larger number signifying a faster rate (4). At 15 days, the number of abnormal seedlings and number of moldy seeds were recorded, along with the number of seeds unable to shed their seed coats (no-sheds). Abnormal petunia seedlings were translucent and grew horizontally, whereas abnormal impatiens seedlings grew horizontally and had deformed shoots with stunted roots.

The experiment was conducted using a split-plot design where temperature was the whole-plot treatment arranged in a randomized block design. The three growth chambers were the blocks and each temperature was replicated twice in each block. The split-plot treatments were a factorial combination of two species, two cultivars, and two seed types. Data for species were analyzed separately, because when species were compared significant differences existed for all parameters.

RESULTS AND DISCUSSION

Both petunia cultivars showed no significant differences between seed types (refined and standard) for any of the criteria measured (Table 1). Impatiens also showed no significant differences between seed types for germination at 15 days, germination rate, and number of abnormal seedlings (Table 1). Statistically, type did show significant differences for number of moldy seeds and number of no-sheds (Table 1). Because the occurrence of moldy seeds was very low, this difference cannot be considered biologically significant. The frequency of occurrence of no-sheds was not consistently higher in one type of seed. 'Super Elfin Salmon' refined seed failed to shed seed coats more frequently (47.8%) than standard seed (37.3%), but 'Super Elfin White' standard seed failed to shed seed coats more frequently (33.1%) than the refined seed (22.0%). Because there were few significant differences between the refined and standard seed, the values for these seed types were pooled for the analysis of the temperature effects.

Germination percentage at day 15 for both petunia cultivars showed no significant response to temperature, whereas the percentage germination of impatiens increased as temperature rose from 20 to 25^{o}C, but decreased as temperature increased from 25 to 30^{o} (Fig. 1). The quadratic component of the germination percentage response to temperature was significant (1% level), suggesting 25^{o} as the optimal germination temperature for these cultivars.

Temperature had a profound effect on the germination rate of both species and their respective cultivars (Table 1). As temperature increased, the rate of germination increased for both petunia cultivars (Fig. 2). The linear and quadratic components of the germination rate response to temperature were analyzed. Both the linear and the quadratic component were significant at the 1% level, and this may explain the plateau effect on petunia germination rate as the temperature approached 30°. As temperature increased from 20° to 25°, germination rate increased for both impatiens cultivars, but, as temperature increased from 25° to 30°, germination rate decreased in a manner similar to the percentage germination at 15 days (Compare Figs. 1 and 2). The quadratic component of the germination rate response to temperature was significant (1% level), and this curve again suggested that germination rate was maximized at 25°.

The number of moldy seeds for both cultivars of both species did not show a significant response to temperature (Table 1). Abnormal seedlings did show a significant response to temperature for both petunia and impatiens, but again this difference cannot be considered biologically significant because the numbers were very small. No-sheds did show a significant response to temperature for impatiens but not for petunias. Number of no-sheds for both impatiens cultivars dropped as temperature increased from 20° to $25^{\circ}C$ ('Super Elfin Salmon', 9.58 to 6.67 seeds, and 'Super Elfin White', 3.83 to 2.58 seeds). As temperature increased from 25° to 30°, no-sheds increased significantly ('Super Elfin Salmon' rose to 15.7 seeds, and 'Super Elfin White' rose to 14.3 seeds).

Refined seed did not prove different from standard seed on any of the criteria measured except no-sheds for impatiens cultivars where refined seed had fewer no-sheds for one cultivar but more no-sheds for the other cultivar. No differences were found for germination percentage or germination rate for either species or cultivars, and this suggests that refined seed from these lots does not warrant the extra refinement processes and the possible increased price. Research on the comparison of standard and refined seed of other species, cultivars, and seed lots may show an increase in germination percentage or germination rate, and additional studies that verify these results should be conducted.

Temperature consistently affected the rate of germination. Petunias achieved the same germination percentage at each temperature, but the rate increased as the temperature increased. This agrees with previous research that showed that temperature only affected the rate and not the germination capacity (3). It has been suggested that the germination capacity (total amount of seed able to germinate) is identical under all conditions if the seed is capable of germination at all (3). The decrease in germination percentage at $30^{o}C$ in impatiens is identical to results obtained in research using 10 cultivars of impatiens where every cultivar showed a similar decrease as temperature rose from 25^{o} to 30^{o} (6). At 25^{o}, the germination rate and germination percentage are maximized, and the number of no-sheds are minimized for both impatiens cultivars. At 30^{o}, petunias have the highest germination rate, while the germination percentage is statistically the same at all temperatures. Until more information can be accumulated, the results of this research suggest that impatiens seed should be germinated at 25^{o}. Individual growers will have to decide if the increased germination rate, for petunia seed, at higher temperatures would offset the increased cost of heating the germination area.

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Table 1. Sums of squares and single degree of freedom tests for linear and quadratic relationships among temperature for selected petunia and impatiens cultivars

Source	df	Impatiens					Petunias				
		Day 15	Rate	Moldy	Abnormal	No-shed	Day 15	Rate	Moldy	Abnormal	No-shed
Chamber	2	115.36**	21.18*	9.33	0.78	2.19	50.19*	167.9**	2.53	0.69	21.44
Temperature	2	3080.19**	712.84**	5.58	7.86**	1433.44**	29.19	1345.68**	1.19	7.19**	1.03
Linear	1	1963.01**	265.08**	5.39*	3.50	816.75**	0.75	1228.97**	0.02	0.10	0.35
Quadratic	1	1116.09**	447.75**	0.25	4.33	618.03**	28.52**	118.37**	1.17	7.08*	0.67
Error a	13	333.89	188.91	49.21	15.97	775.10	211.72	280.68	39.72	38.85	62.60
Cultivar	1	420.50**	93.26**	1.68	2.72**	253.13**	256.89**	337.43**	12.50*	5.01**	86.68**
Type(Cultivar)	2	2.22	5.86	25.69**	2.06	130.81**	24.72	65.08	2.50	1.14	1.36
Temp x Cultivar	2	171.75**	50.92**	3.86	5.53**	57.33	0.36	8.37	0.58	2.53	0.53
Error b	4	24.6	15.13	11.22	1.94	92.44	22.78	41.66	11.33	1.23	2.22

*,**Indicates significance at the 0.05 and 0.01 levels, respectively.

Figure 1. Influence of temperature on germination percentage of petunia and impatiens seed

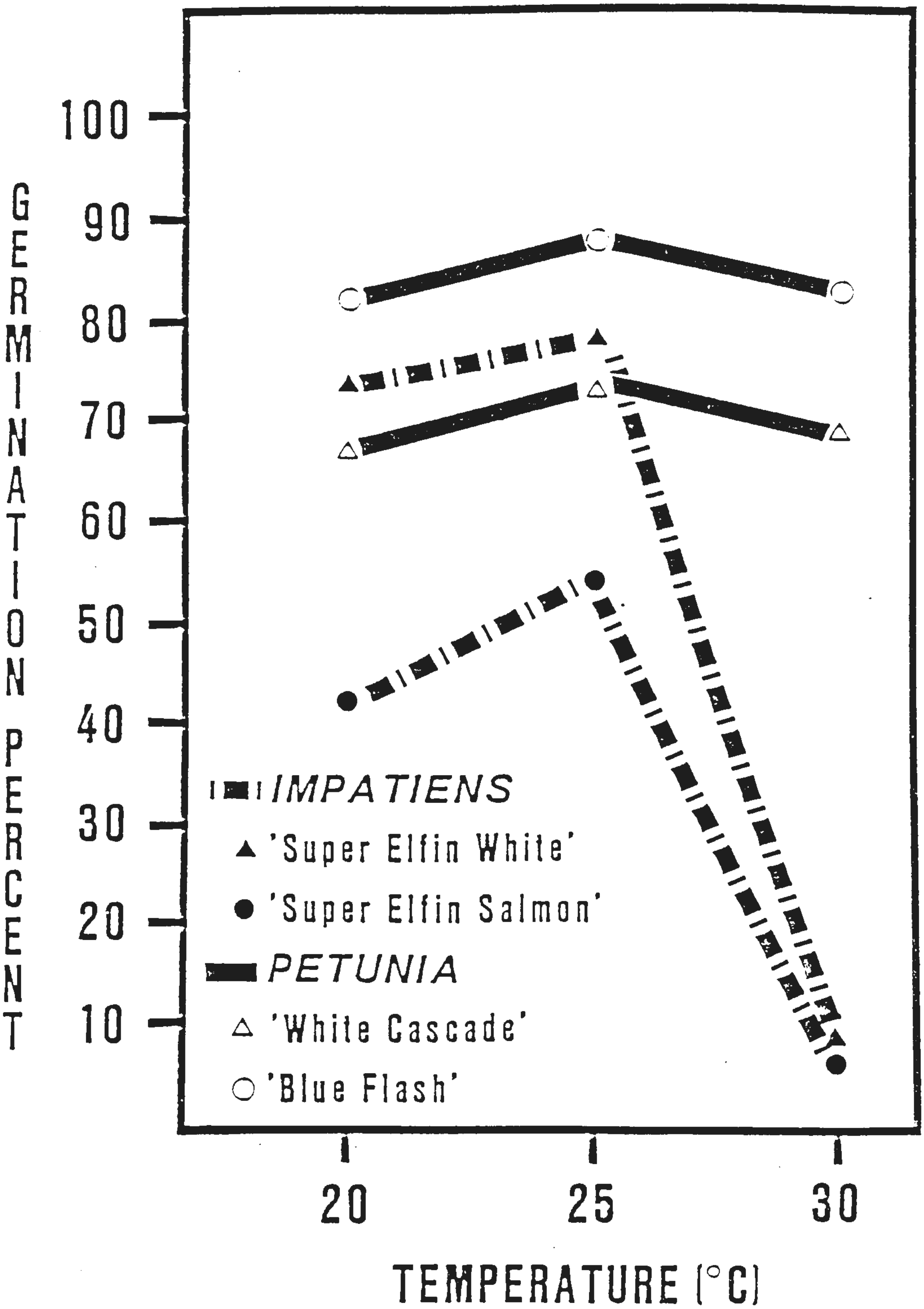
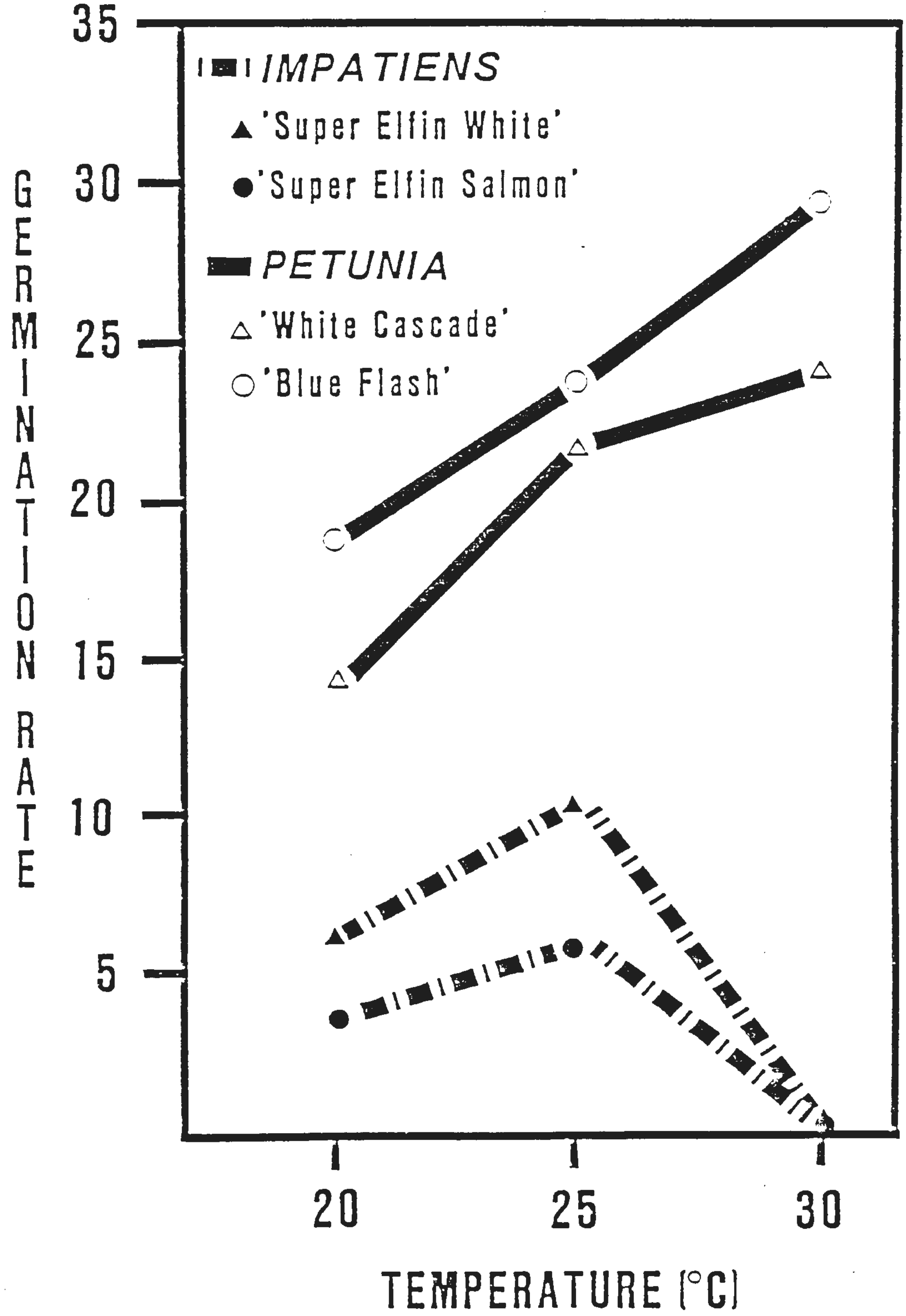

100
90
80
70
60
50
40
30
20
10
GERMINATION PERCENT
IMPATIENS
'Super Elfin White'
'Super Elfin Salmon'
PETUNIA
'White Cascade'
'Blue Flash'
20
25
30
TEMPERATURE (°C)

Figure 2. Influence of temperature on germination rate of petunia and impatiens seed


35
30
25
20
15
10
5
GERMINATION RATE
IMPATIENS
'Super Elfin White'
'Super Elfin Salmon'
PETUNIA
'White Cascade'
'Blue Flash'
20
25
30
TEMPERATURE (°C)

SECTION II. GERMINATION ENVIRONMENT EFFECTS ON REFINED AND STANDARD SEED GERMINATION, GROWTH, AND FLOWERING OF *PETUNIA X HYBRIDA* (VILM.) 'BLUE FLASH' AND 'WHITE CASCADE'

Germination environment effects on refined and standard seed germination, growth, and flowering of *Petunia* x *hybrida* (Vilm.) 'Blue Flash' and 'White Cascade'

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ABSTRACT

Refined (High Energy™ seed, Ball Seed Co.) and standard seed of 2 cultivars of Petunia x hybrida were germinated under 9 different environments. The environments studied were a factorial combination of 3 temperature levels and 3 vapor pressure deficit (VPD) levels. Germination counts were taken daily and, at 15 days after sowing, seedlings were evaluated. Growth data including height, diameter, plant fresh weight, shoot fresh weight, and shoot dry weight, were taken 2, 4, and 6 weeks after sowing. Days to flowering and dry shoot weight at first flowering also were recorded. Temperature consistently affected the germination rate with the rate increasing significantly with each increase in temperature. VPD was most notable for its effects on growth with 0.80 and 1.33 kPa (6 and 10 mm Hg, respectively) treatments resulting in larger, heavier plants than those grown at 1.87 kPa (14 mm Hg). The interaction of temperature and VPD was significant for germination percentage and germination rate. Germination percentage was largest at 25°C with a VPD of 1.87 kPa (25°/1.87) and lowest at 20°/1.87 for both cultivars studied. Germination rate was statistically the same at all VPDs within a temperature except for 20° where a VPD of 1.87 kPa was significantly lower than 0.80 or 1.33 kPa. Refined seed of one cultivar showed an increased germination percentage (82.6 versus 78.1), and increased germination rate (92.8 versus 86.1).

INTRODUCTION

Essential for the production of high quality bedding plants is rapid, uniform seed germination. Satisfactory levels of moisture, aeration, temperature, and in some cases light are required for seed germination (4,11,23). Soil moisture in deficiency or excess is probably the most common stress encountered by germinating seeds (17). Water is essential for enzyme activation, breakdown, translocation, and use of stored food reserves (4). The germination medium must be moist enough at all times to supply needed moisture to the seeds and seedlings, but excessive moisture will limit aeration (9,17). Upper layers of soil are the first to be moistened, but also the first to dry by evaporation into the air or percolation to lower levels within the medium (11). Seeds sown on the soil surface are very vulnerable to drying; therefore, the relative humidity of the atmosphere is very important. Moisture tension that the seed perceives is a result of the vapor pressure deficit at the soil surface. If the medium is kept uniformly moist but free of standing water, it has a relative humidity of 100%. If the relative humidity of the atmosphere is less than 100% a vapor pressure deficit is created at the soil surface. Relative humidity changes with temperature as does vapor pressure deficit (VPD), and therefore, the same vapor pressure deficit can be achieved at different temperatures by adjusting the relative humidity (Table 1).

Complex changes occur in the seed during germination, and these changes involve temperature-dependent metabolic events. Therefore, it is not surprising to find a close dependence of germination on temperature (12,4). In many cases, the germination rate increases steadily with increasing temperatures up to an optimum, which usually lies between 15° and 30°C (5,12,15). Response to temperature depends on species, variety, growing region and duration of time from seed harvest (4). In studies with tomatoes, low soil temperatures resulted in reduced and delayed germination (8). Oats showed no significant differences in total germination at 5 temperatures studied (24). Temperatures in excess of 30°C were unfavorable for germination of *Lepidium virginicum* (22). Optimum temperature for germination may be defined as that temperature giving the greatest percentage germination in the shortest period of time (4). Studies on wheat, soybeans, and oats showed that optimum temperatures gave greater opportunity to compare the vigor of seed lots (24).

The most important factor influencing seed vigor of wheat is seed size (7,10,19). In most cases, a positive correlation exists between large seed and germination percentage (12). These differences are more pronounced in early growth stages when the seedling is dependent on food reserves of the seed. Contrasting studies have shown that smaller seed germinated and grew faster than larger seed (3,6). Size graded broccoli seed produced more uniform transplants than bulk seed (21).

Bedding plant seed producers have begun to market refined seed lines. Refined seed is characterized as having increased germination percentage and more vigorous seedling growth. It is separated from thoroughly cleaned bulk seed lots according to weight, size and density (1,13,20).

The cleaning and separation procedures vary among seed species and seed companies (1). The separated fractions are more uniform physically than the original bulk lot. These fractions are tested for germination and vigor and those with the highest percentages are designated refined seed.

The objectives of this research were to study the effect of temperature and VPD on germination of refined and standard seed of Petunia x hybrida 'Blue Flash' and 'White Cascade' grown as plugs.

MATERIALS AND METHODS

Standard and refined seed (High Energy™ seed, Ball Seed Co., West Chicago, IL) of Petunia x hybrida 'Blue Flash' and 'White Cascade' were studied under 9 environments. The treatments were a factorial combination of 3 temperature levels and 3 VPD levels, controlled by relative humidity. Temperature levels were 20°, 25°, and 30°C with VPD levels of 0.80, 1.33, and 1.87 kPa (Table 1).

Square 406-cell plug flats were filled with medium composed of Sphagnum peat (35%), Hypnum peat (35%), and sand-finish perlite (30%) (Persolite Co., Florence, CO). Sphagnum and Hypnum peats were passed through a 0.64-cm mesh screen before use. The medium was amended with 2.97 kg/m^3 calcium carbonate, 1.78 kg/m^3 single superphosphate (0-8.7-0), and 0.59 kg/m^3 of a minor element mix containing 1.0% S, 0.02% B, 0.3% Cu, 2.0% Fe, 0.5% Mn, 0.0006% Mo, 1.0% Zn, and not more than 2.6% Cl (Esmigran, Mallinckrodt, Inc., St Louis, MO). Calcium carbonate and single superphosphate were ground in a mortar and pestle before incorporation into the medium. Flats were filled with dry medium, by hand, and filled flats were placed under intermittent mist overnight for moistening before sowing. Each flat, representing either 'Blue Flash' or 'White Cascade', was divided into 4 sections each containing 100 cells. Two sections were sown with refined seed and the remaining 2 sown with standard seed. A Blackmore automatic seeder (Blackmore Transplanter Co., Ypsilanti, MI) was used for all sowing, and the seeder manifold was checked for complete seed pick up on every row sown.

After sowing, flats were placed in growth chambers maintained at the constant temperature (+/- 1.5°C) and vapor pressure (+/- 5% relative humidity) of the environment to be tested (Table 1). All chambers were lighted with cool white fluorescent fixtures (135 μmol sec^{-1} m^{-2}) for a 24-hour day length. Flats were watered as needed to maintain a constantly moist medium. Deionized water (>18 megohm resistance) was kept in portable pressurized sprayers (Hudson Manufacturing Co., Chicago, IL) inside the chambers so water temperature equalled air temperature. In order to avoid changes in relative humidity within the chamber, watering was done outside the chamber.

Germination counts were taken daily for 15 days. Seed was considered germinated when the cotyledons were expanded completely. Germination rate was determined by Maguire's equation with a larger number signifying a faster rate (14). At 15 days after sowing, the number of no-sheds and abnormals were recorded. No-sheds were those seedlings unable to shed their seed coat and abnormals were translucent seedlings that grew horizontally on the soil surface. At 15 days, flats were moved from the chambers into a greenhouse maintained at 68° nights for the duration of the study. Flats were sown from April 22 through November 28, 1985.

Height, diameter of leaf span, whole plant fresh weight, shoot fresh weight, and shoot dry weight were recorded at 2, 4, and 6 weeks after sowing. Three seedlings were selected randomly from each quarter of a flat for growth data. Six plants from each quarter of each flat were transplanted into a marketable pack, 6 weeks after sowing. Days to flowering and dry shoot weight at first flowering were recorded.

The experiment was conducted as a split plot design where whole-plot treatments were a factorial combination of 3 levels of temperature and 3 levels of VPD and split plot treatments were the 2 seed types. Whole-plot treatments were conducted in a randomized block design with chambers as blocks. Data for cultivars were analyzed separately.

RESULTS AND DISCUSSION

The analysis of variance tables show that both temperature and VPD had significant effects on many of the parameters measured (Tables 2 and 3). Temperature consistently affected germination rate (Table 2), while VPD affected plant growth (Table 3). The interaction of temperature and VPD was significant for germination percentage and germination rate for both cultivars studied.

Temperature had a significant effect on germination rate, number of no-sheds, plant diameter, and shoot dry weight for seedlings produced from 'Blue Flash' seed (Tables 2 and 3). 'White Cascade' seed showed a significant response to temperature for germination percentage, germination rate, and plant diameter (Tables 2 and 3). Both cultivars showed a significant increase in germination rate with each increase in temperature (Table 4). This is consistent with the results of many germination studies where germination rate steadily increased with increasing temperatures up to an optimum temperature usually between 15^{o} and 30^{o}C (5,12,15).

The effect of temperature on plant diameter was similar for both cultivars. In each case, plants produced from seed germinated at 25^{o} and 30^{o}C were significantly larger in diameter than those produced from seed germinated at 20^{o} (Table 4). At 25^{o} and 30^{o} seed germination was faster and the seedlings produced were larger in diameter than seed germinated at 20^{o}. Research with soybean seed showed similar results with the first compound leaves of rapidly germinating plants being larger than those of slower germinating plants (16).

There were significantly more no-sheds, per 100 seed sown, observed at 20° (0.47) than at either 25° (0.19) or 30° (0.08) in the 'Blue Flash' cultivar (LSD = 0.24). Rate of seed germination at 20° was much slower, therefore the final germination count may have been taken before germination was complete and seed coats shed. 'Blue Flash' plants produced from seed germinated at 25° and 30° had significantly larger shoot dry weight measurements than plants produced at 20° (21.3 and 19.8 versus 11.8 mg). Temperatures of 25° and 30° resulted in larger, heavier seedlings in a shorter amount of time than seedlings produced at 20°. Seedlings of barley that emerged earlier showed advantages over later emerged ones, with larger shoots, increased photosynthetic rate, and a more developed root system (2).

'White Cascade' seed showed no significant differences in response to temperature for number of no-sheds or shoot dry weight (data not presented). A greater percentage of 'White Cascade' seed germinated at 30° than at 20° (Table 4). At 25°, germination was was not significantly different from either 20° or 30°. Similar results were found for tomato seed with low temperatures resulting in reduced germination percentage and rate (8). 'Blue Flash' showed no significant response to temperature for germination percentage (Table 4), which is consistent with results obtained with oats where percentage germination was statistically the same at the 5 temperatures studied (24).

VPD influenced germination rate, plant diameter, plant fresh weight, shoot fresh weight, and shoot dry weight for both cultivars (Tables 2 and

3). Rate of germination was significantly faster at VPDs of 0.80 and 1.33 kPa than at 1.87 kPa for 'Blue Flash' (Table 5). 'White Cascade' showed a faster germination rate at VPDs of 0.80 and 1.33 kPa but the difference between 0.80 and 1.87 kPa was not significant. Plant diameter and all weight measurements recorded were consistently larger at 0.80 and 1.33 kPa than at 1.87 kPa, but there were no differences between the 0.80 and 1.33 kPa treatments (Table 5). The weight differences were still apparent 6 weeks after sowing.

The interaction of temperature and VPD affected germination percentage and germination rate for both cultivars studied. The number of no-sheds in 'Blue Flash' also was affected by the interaction of temperature and VPD as was plant diameter in 'White Cascade'. Germination percentage was the highest at $25^{o}/1.87$ for both cultivars studied (Figures 1 and 2). Both cultivars showed the lowest germination percentage in the $20^{o}/1.87$ environment. Studies have shown that low temperatures and high substrate moisture tension decreased germination percentage and rate (18). At $30^{o}C$ the germination percentage was statistically the same at all 3 VPDs, at 25^{o} VPDs of 0.80 and 1.33 kPa were the same while 1.87 kPa gave significantly higher germination percentages. High moisture levels also have been found to inhibit germination by limiting aeration (4,9,17).

The interaction of temperature and VPD was significant for germination rate; this may have resulted from the overwhelming influence of temperature on germination rate. When the least significant differences for the interaction was computed VPDs within a temperature were not different from each other except at $20^{o}C$ where a VPD of 1.87 kPa was significantly lower than 0.80 or 1.33 kPa (Table 6). This is similar to lima bean research where it was thought that low temperatures and high water tension increased time to the beginning of water uptake and therefore decreased germination rate. Number of no-sheds for the 'Blue Flash' cultivar were significantly higher in the 20°/1.87 environment (Table 6). Germination rate was slower at 20°/1.87, and some seed may not have completed germination and shed their seed coat. 'White Cascade' seedlings showed a significantly smaller plant diameter in the 20°/1.87 environment than in any of the other environments studied (Table 6). Slow germination rate at 20°/1.87, decreased plant establishment and growth resulting in a smaller plant diameter.

Seed type, refined and standard, were not statistically different for any parameter measured in the 'White Cascade' cultivar (Tables 2 and 3). Refined seed of the 'Blue Flash' cultivar had increased germination percentage (82.6 versus 78.1), and increased germination rate (92.8 versus 86.1). Days to flowering and weight at first flowering were not statistically different between refined and standard seed (data not presented). These results show the characterization of refined seed as having increased germination percentage and rates held true for only 1 of the 2 cultivars studied. The claims of more vigorous seedling growth were not detectable for either cultivar. Because there were few differences in seed germination and seedling growth, individual growers will have to decide if the increased price of refined petunia seed would justify its use (1,20,13).

Germination environment is the most important factor to consider when germinating seed. Without the proper environment, seed (refined or standard) will fail to germinate or germinate more slowly. At 30°C, germination percentage and rate were the same for all 3 VPD levels studied. At 25°, germination percentage was significantly higher at 25°/1.87, while rate was the same at all 3 VPDs studied. Germination percentage and rate were significantly lower at 20° with a VPD of 1.87 kPa that at either 0.80 or 1.33 kPa. This would suggest at 30° a grower would have to worry less about moisture levels than at 20° or 25°. At 25° a somewhat drier environment, 41% RH should be maintained for maximum germination percentage, but at 20° the drier environment, VPD of 1.87, may result in poor seed germination and slow plant growth.

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Table 1. Temperature x vapor pressure deficit (VPD) treatment description

Treatment description Temp ^{o}C/VPD kPa	VP^{z}_{satd}	VP^{z}_{air}	% RH
20^{o}/0.80	2.34	1.54	65.8
20^{o}/1.33	2.34	1.01	43.0
20^{o}/1.87	2.34	0.47	20.2
25^{o}/0.80	3.17	2.37	74.8
25^{o}/1.33	3.17	1.84	57.9
25^{o}/1.87	3.17	1.30	41.1
30^{o}/0.80	4.24	3.44	81.1
30^{o}/1.33	4.24	2.91	68.6
30^{o}/1.87	4.24	2.37	56.0

^{z}Vapor pressure, in kPa.

Table 2. Analysis of variance for Petunia x hybrida 'Blue Flash' and 'White Cascade' germination data

Source	df	Blue Flash				Sums of squares White Cascade			
		D15[z]	Rate[y]	Abnormals	No-sheds	D15[z]	Rate[y]	Abnormals	No-sheds
Chamber	2	65.037	733.386*	0.028	0.111	29.370	323.205	0.009	0.287
Temperature	2	103.898	33625.128**	0.028	1.444**	448.398*	41021.882**	0.120	0.593
VPD[x]	2	61.676	1517.218**	1.333	1.083*	89.370	1444.530*	0.065	0.704
Temp x VPD	4	881.963**	3984.005**	0.222	2.556**	813.824*	4188.584**	0.185	1.296
Error a	16	365.630	1001.889	5.389	1.806	687.296	2711.896	1.074	4.629
Type	1	277.894**	599.256**	0.042	0.116	112.667	218.194	0.116	0.074
Temp x Type	2	10.731	10.717	0.028	0.037	13.361	151.796	0.120	0.037
VPD x Type	2	22.343	52.634	0.778*	0.065	21.778	45.444	0.176	0.148
Temp x VPD x Type	4	11.407	16.340	0.278	0.074	54.028	201.652	0.463	0.741
Error b	18	202.500	357.185	1.250	1.583	614.667	941.925	1.750	2.250

[z]Germination percentage, 15 days after sowing.

[y]Germination rate.

[x]Vapor pressure deficit.

*,**Indicates significance at the 0.05 and 0.01 levels, respectively.

Table 3. Analysis of variance for Petunia x hybrida 'Blue Flash' and 'White Cascade' growth data

Source	df	Blue Flash					White Cascade				
		Sums of squares									
		Height	Diameter	Fresh Plant Weight	Fresh Shoot Weight	Dry Shoot Weight	Height	Diameter	Fresh Plant Weight	Fresh Shoot Weight	Dry Shoot Weight
Chamber	2	167.930	104.531	0.032	0.032	0.001	175.003	72.501	0.047	0.043	0.000
Temp	2	1205.378	2076.461**	0.335	0.242	0.003*	723.492	1101.565*	0.215	0.149	0.001
VPD[z]	2	928.818	3773.055**	1.282*	0.656*	0.005**	962.070	2336.884**	1.092*	0.574*	0.003**
Temp x VPD	4	915.697	1408.573	0.453	0.315	0.000	863.839	1122.138**	0.424	0.325	0.001
Error a	16	3949.588	2316.706	1.652	0.940	0.005	4745.856	1424.064	1.846	1.164	0.003
Type	1	0.702	0.210	0.004	0.001	0.000	6.654	0.176	0.000	0.000	0.000
Temp x Type	2	2.607	4.173	0.003	0.001	0.000	9.735	6.093	0.000	0.000	0.000
VPD x Type	2	6.406	25.483	0.003	0.003	0.000	26.790*	21.188	0.002	0.001	0.000
Temp x VPD x Type	4	1.720	7.287	0.018	0.010	0.000	20.430	17.322	0.000	0.000	0.000
Error b	18	38.981	108.417	0.053	0.028	0.003	48.006	106.028	0.019	0.016	0.000

[z]Vapor pressure deficit.

*,**Indicates significance at the 0.05 and 0.01 levels, respectively.

Table 4. The effect of temperature on germination percentage, germination rate, and plant diameter of Petunia x hybrida 'Blue Flash' and 'White Cascade'

Temperature	Germination Percentage	Germination Rate	Diameter (mm)
		Blue Flash	
20°C	78.5	56.5	28.7
25°C	80.6	95.0	34.9
30°C	81.9	116.9	37.1
LSD (5%)	3.4	5.6	4.9
		White Cascade	
20°C	77.0	54.0	28.6
25°C	79.9	94.0	33.5
30°C	84.0	121.2	34.6
LSD (5%)	4.6	9.2	3.9

Table 5. The effect of vapor pressure deficit (VPD) on germination rate and growth data of <u>Petunia</u> x <u>hybrida</u> 'Blue Flash' and 'White Cascade'

VPD (kPa)	Rate	Diameter (mm)	Fresh Plant Weight (mg)	Fresh Shoot Weight (mg)	Dry Shoot Weight (mg)
Blue Flash					
0.80	93.0	37.4	433.90	328.32	20.72
1.33	93.4	36.6	407.24	311.89	22.23
1.87	82.0	26.8	225.64	180.41	9.95
LSD (5%)	5.6	4.9	133.86	100.97	7.25
White Cascade					
0.80	90.8	35.4	436.66	346.56	19.87
1.33	95.3	34.4	425.64	334.69	19.42
1.87	82.8	26.9	250.18	209.66	10.79
LSD (5%)	9.2	3.9	141.48	112.36	5.81

Table 6. The interaction of temperature and vapor pressure deficit (VPD) as it affects germination rate of _Petunia_ x _hybrida_ 'Blue Flash' and 'White Cascade', number on no-sheds in 'Blue Flash', and plant diameter of 'White Cascade'

	Temperature °C					
VPD	20	25	30	20	25	30
	Blue Flash			White Cascade		
	Germination Rate					
0.80	70.03	93.28	115.78	67.01	87.06	118.41
1.33	67.29	93.25	119.64	63.95	94.05	127.86
1.87	32.24	98.40	115.27	31.17	99.88	117.31
	Interaction LSD (5%) = 9.69			Interaction LSD (5%) = 15.94		
	No-sheds (%)			Diameter (mm)		
0.80	0.0	0.0	0.2	34.1	35.0	37.1
1.33	0.4	0.4	0.1	33.7	34.9	34.7
1.87	1.0	0.2	0.0	18.1	30.6	32.1
	Interaction LSD (5%) = .41			Interaction LSD (5%) = 6.67		

Figure 1. Effect of temperature and vapor pressure deficit on germination percentage of Petunia x hybrida 'Blue Flash'

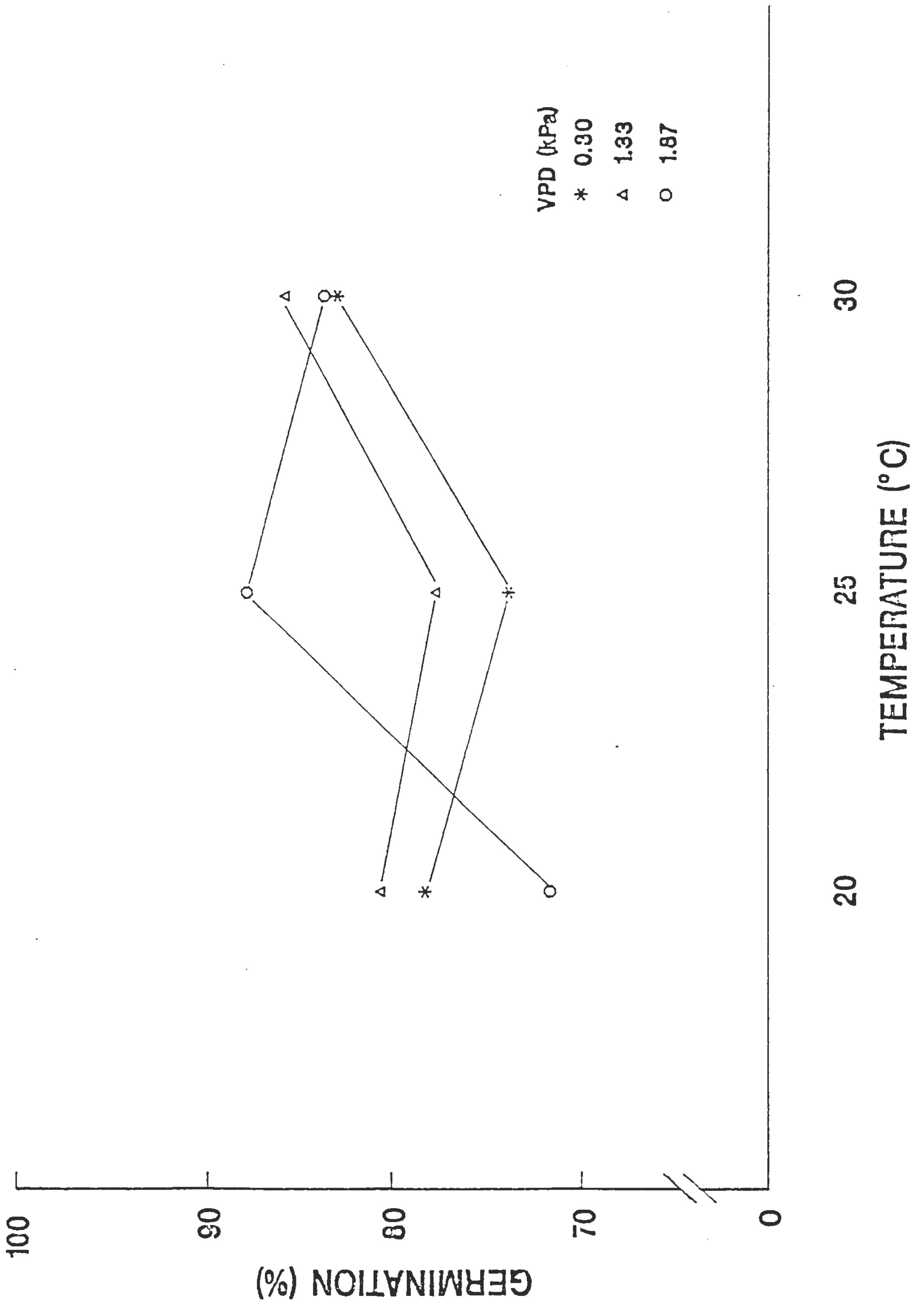
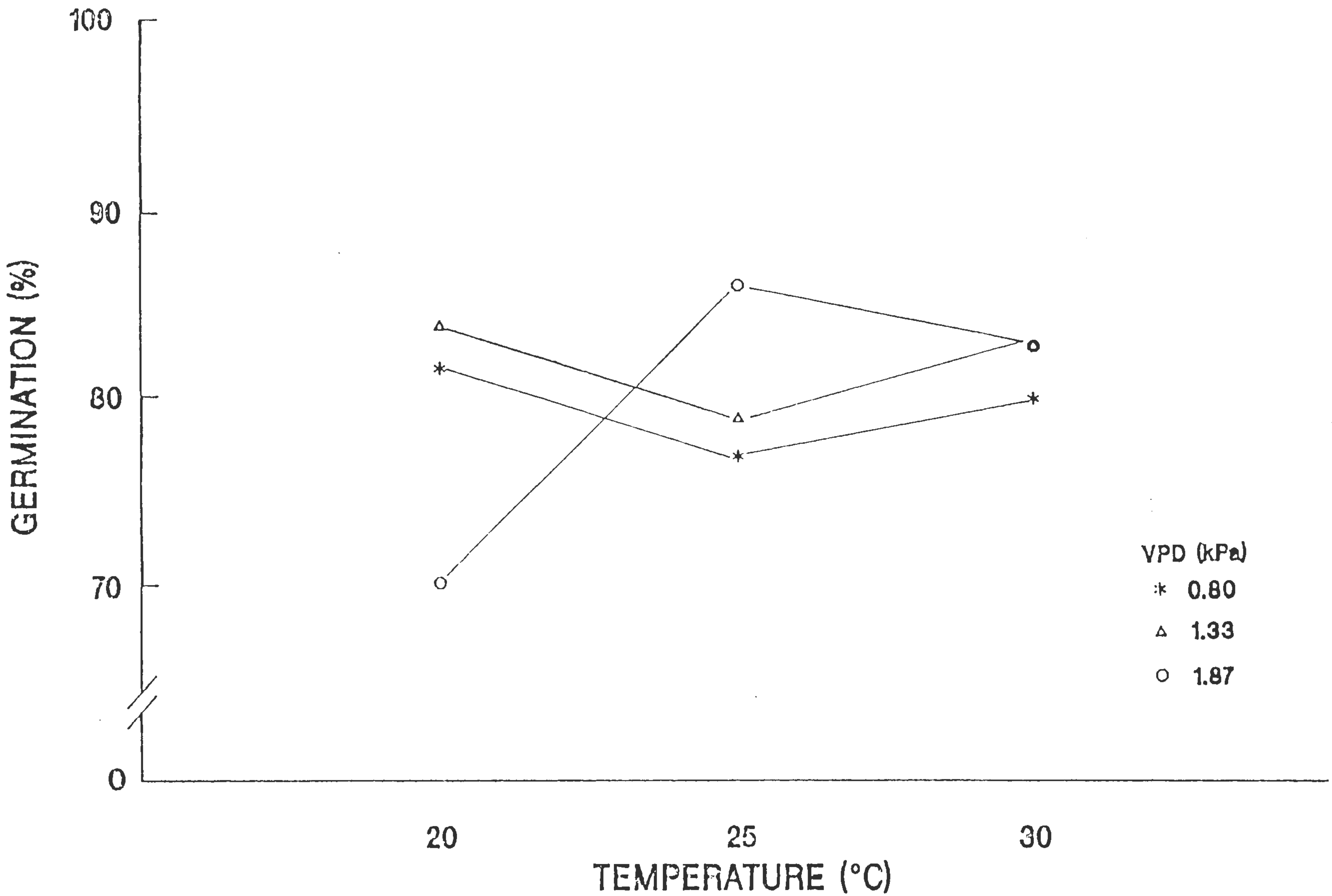

VPD (kPa)
* 0.30
Δ 1.33
○ 1.87
GERMINATION (%)
100
90
80
70
0
20
25
30
TEMPERATURE (°C)

Figure 2. Effect of temperature and vapor pressure deficit on germination percentage of _Petunia_ x _hybrida_ 'White Cascade'


100
90
80
70
0
GERMINATION (%)
20
25
30
TEMPERATURE (°C)
VPD (kPa)
* 0.80
△ 1.33
○ 1.87

SECTION III. GERMINATION AND EARLY SEEDLING GROWTH COMPARISON OF REFINED AND STANDARD SEED OF SEVERAL CULTIVARS OF PLUG-GROWN *PETUNIA* X *HYBRIDA* (VILM.)

Germination and early seedling growth comparison of refined and standard seed of several cultivars of plug-grown *Petunia* x *hybrida* (Vilm.)

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ABSTRACT

Refined (High Energy™ seed, Ball Seed Co.) and standard seed of 5 cultivars of *Petunia* x *hybrida* were germinated in plug flats at 25°C and 70% relative humidity. Germination counts, number of seed unable to shed their seed coat, number of abnormal seedlings, and number of runts were recorded. Percentage germination and germination rate were calculated from daily germination data. Height, diameter, and dry shoot weight were measured in order to compare early growth of seedlings. 'Happiness' refined seed germinated 20% more frequently, 40% faster, and with 81% fewer runts than standard seed. Refined seed of 'White Flash' also germinated at a higher percentage and at a faster rate than standard seed, 4.5% and 6.5%, respectively. The 3 other cultivars studied showed either no differences between refined and standard seed or differences for only 1 of the 8 parameters measured. Because the performance of refined seed was not consistent for all cultivars studied, the increased price of refined seed may not be justified because of the uncertainty of the results.

INTRODUCTION

Seed germination is a complex physiological event involving many metabolic processes resulting in seedling emergence and growth (5,9). The reactions involved in these processes are dependent on proper levels of temperature, moisture, aeration, and in some cases light. Varying the levels of any of these essential components for seed germination can reduce germination percentages and seedling growth. Due to the complexity of seed germination, difficulties in germination and seedling establishment are among the most troublesome in the plug industry (1). These problems may be due to improper germination environment, improper seed storage, or mechanical damage to the seed. Many seed companies have begun to market a new product, called refined seed, designed to help alleviate these problems in germination. This seed in characterized by increased germination and more vigorous seedling growth.

Refined seed is derived from bulk seed, by thoroughly cleaning the lot and by separation of the seed according to the physical characteristics of weight, size, and density (2,10,14). The separation procedures vary among seed species and seed companies (2). The separated seed fractions are more uniform physically than the original bulk lot. These fraction are tested for germination and vigor and those with the highest percentage viable seedlings are designated refined.

Studies with broccoli showed that size-graded seed performed more uniformly than bulk seed (16). Investigations into the effect of seed size on germination, growth, and development, have shown that a greater percentage of large seed germinate, produce larger more vigorous seedlings, and result in greater production and yield (3,6,7,8,13,16). Many times the difference is more pronounced during the early growth stages, while the seedling is dependent on stored food reserves of the seed (9). Studies also have shown that size has no effect on germination percentage, germination rate, or size of seedlings produced (4,12,13). This study was designed to compare germination and early seedling growth of standard versus refined seed for 5 cultivars of *Petunia* x *hybrida*.

MATERIALS AND METHODS

Standard and refined seed (High Energy™ seed, Ball Seed Co., West Chicago, IL) of _Petunia_ x _hybrida_ 'Happiness', 'Pink Flash', 'Salmon Flash', 'Super Cascade Pink', and 'White Flash' sown into square, 406-cell plug flats. Flats were filled with a medium composed of _Sphagnum_ peat (35%), _Hypnum_ peat (35%), and sand-finish perlite (30%) (Persolite Co., Florence, CO). _Sphagnum_ and _Hypnum_ peats were passed through a 0.64-cm mesh screen before use. The medium was amended with 2.97 kg/m^3 calcium carbonate, 1.78 kg/m^3 single superphosphate (0-8.7-0), and 0.59 kg/m^3 of a minor element mix containing 1.0% S, 0.02% B, 0.3% Cu, 2.0% Fe, 0.5% Mn, 0.0006% Mo, 1.0% Zn, and not more than 2.6% Cl (Esmigran, Mallinckrodt, Inc., St. Louis, MO). Calcium carbonate and single superphosphate were ground in a mortar and pestle before incorporation into the medium. Flats were filled with dry medium, by hand, and filled flats were placed under intermittent mist overnight for moistening before sowing. Each flat, representing a cultivar, was divided into 4 sections each containing 100 cells. Two sections were sown with refined seed and the remaining 2 sown with standard seed. A Blackmore automatic seeder (Blackmore Transplanter Co., Ypsilanti, MI) was used for all seed sowing and the seeder manifold was checked for complete seed pick up on every row sown.

After sowing, flats were placed in a growth chamber maintained at a constant temperature of 25°C (+/- 1.5°) with relative humidity of 70% (+/- 5%) and continuous fluorescent light at 135 mol s^{-1} m^{-2}. Flats were watered every 12 hours with a fine mist of tap water for the duration of the experiment. Germination counts were taken daily for 12 days. Seed was considered germinated when cotyledons expanded completely. Germination rate was determined by Maguire's equation with a larger number signifying a faster rate (11). At 12 days after sowing the number of no-sheds, abnormals, and runts were recorded. No-sheds were those seedlings unable to shed their seed coat; abnormals were translucent seedlings that grew horizontally on the soil surface; and runts were seedlings that were noticeably smaller. Height, plant diameter, and shoot dry weights were recorded for 3 seedlings (observations) harvested at the soil surface from each quarter of each flat. The study was repeated twice. The experiment was analyzed as a completely random design with treatments being a factorial combination of 2 runs and 2 seed types. Growth parameter observation within treatments were averaged before data analysis.

RESULTS AND DISCUSSION

'Pink Flash' showed no significant differences between seed type for any of the criteria measured. Refined seed had slightly larger means for all of the criteria measured except runts which were observed less frequently in refined seed (Tables 1 and 2). However, these differences were not statistically significant.

The 'Salmon Flash' cultivar showed no significant differences for type on day 12 for germination percentage, germination rate, number of abnormal seedlings, and number of no-sheds, but did show significantly fewer runts for refined seed (Table 1). The growth data, height, diameter, and dry weight, showed no significant differences between seed types (Table 2).

Type was significant in 'White Flash' for germination percentage and rate (Table 1). Refined seed had a 4.5% greater germination percentage, and 6.6% faster germination rate. While differences were not significant between seed types for number of abnormal seedlings and number of no-sheds, standard seed showed fewer seedlings for each of these parameters (Table 1). Standard seed also produced taller and heavier seedlings than refined seed, but the differences were not significant (Table 2).

'Happiness' showed the most striking differences in seed type with refined seed germinating over 20% more often than standard seed (Table 1). Refined seed showed a similarly impressive response for germination rate (90.1 versus 64.4). There were 81% more runts produced from standard seed and this difference was significant. 'Happiness' was the only cultivar to show a statistically significant difference in seed type for any of the measured growth parameters. Plant diameter for refined seed was significantly larger than standard seed (8.4 versus 8.0 mm).

'Super Cascade Pink' showed no significant difference for final germination at day 12, but did show a significant difference in germination rate with refined seed germinating faster than standard seed (Table 1). No significant differences were found for any of the other parameters measured (Tables 1 and 2).

Type was significant for germination rate in 'White Cascade', 'Happiness', and 'Super Cascade Pink' with refined seed germinating more rapidly than standard seed. If germination rate is considered a criterion for vigor, then refined seed could be considered a more vigorous seed type. A positive result from a single test for seed vigor is not a reliable measure, therefore, several tests should be employed to evaluate vigor (6). This would eliminate 'Super Cascade Pink' refined seed as a more vigorous type, because germination rate was the only parameter for which type was significant.

Germination percentage was significantly higher for refined seed in 2 of the 5 cultivars studied, with the difference highly significant for the cultivar 'Happiness'. In a study done on a wide variety of seed it was found that in almost all cases, there was a direct relationship between germinability as counted by primary root emergence or hypocotyl development, and subsequent viability of the seedling (15). An investment in refined seed of 'Happiness' may result in a 20% increase in saleable plants.

'Pink Flash' showed no difference for seed type for any criteria measured, and number of runts was the only significant parameter measured for 'Salmon Flash'.

These results show that the characterization of refined seed as having increased germination percentages and rates held true for only 'White Flash' and 'Happiness' (2 out of 5 cultivars). The claims of more vigorous seedling growth held true for only 1 out of the 5 cultivars studied ('Happiness').

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Table 1. Comparison of germination parameters between refined and standard seed for 5 *Petunia* x *hybrida* cultivars

Seed Type	Pink Flash	Salmon Flash	White Flash	Happiness	Super Cascade Pink
			Germination Percentage		
Refined	87.3	90.3	84.9	88.9	87.4
Standard	87.2	89.3	80.6	68.0	85.6
Pr > F[z]	.919	.517	.024	.0001	.328
			Germination Rate		
Refined	74.26	79.48	77.26	90.10	77.35
Standard	72.33	78.21	72.49	64.45	72.94
Pr > F	.285	.525	.018	.0001	.013
			Number of Abnormal Seedlings[y]		
Refined	1.0	0.8	2.4	0.5	0.1
Standard	0.6	1.3	2.1	0.7	0.3
Pr > F	.229	.167	.624	.526	.283
			Number of No-sheds[y]		
Refined	0.0	0.0	0.1	0.1	0.0
Standard	0.0	0.0	0.0	0.1	0.0
Pr > F	—	—	.329	1.000	—
			Number of Runts[y]		
Refined	0.9	0.8	1.4	0.3	0.68
Standard	1.6	1.5	1.6	1.8	1.18
Pr > F	.127	.040	.680	.001	.253

[z]Probability of a greater F statistic.

[y]Number per 100 seed counted.

Table 2. Comparison of growth parameters between refined and standard seed for 5 *Petunia* x *hybrida* cultivars

Seed Type	Cultivar				
	Pink Flash	Salmon Flash	White Flash	Happiness	Super Cascade Pink
			Height (mm)		
Refined	2.4	2.6	2.8	2.6	2.6
Standard	2.4	2.5	2.8	2.6	2.6
Pr > F[z]	1.00	.60	.83	.88	1.00
			Diameter (mm)		
Refined	8.4	8.8	8.7	8.4	8.9
Standard	8.1	8.4	8.6	8.0	8.7
Pr > F	.08	.09	.59	.04	.40
			Dry Weight (mg)		
Refined	.39	.55	.63	.70	.54
Standard	.37	.53	.64	.66	.55
Pr > F	.47	.38	.74	.23	.72

[z]Probability of a greater F statistic.

SUMMARY AND CONCLUSIONS

Rapid, uniform seed germination is essential for the production of high quality bedding plants in plug flats. Low germination counts and uneven seedling growth have been observed in single-sown plug flats of many bedding plant annuals. Seed companies have begun to market a refined seed lines to improve germination and vigor. The objectives of this research were to study the effects of the germination environment on refined and standard seed of petunia and impatiens.

Refined and standard seed within a species, petunia or impatiens, gave similar germination results in petri dishes at 20°, 25°, and 30°C except for number of no-sheds in impatiens, where refined seed had fewer no-sheds than standard (22% versus 33%) in 1 cultivar but more no-sheds for the other cultivar (48% versus 37%). Data for seed type were pooled so temperature effects could be studied. Temperature consistently affected germination rate. Petunia seed achieved the same germination percentage 15 days after sowing, but rate increased as temperature increased. A higher percentage of impatiens seed germinated at a faster rate at 25° than at 20° or 30°. The results of this research suggest that impatiens seed should be germinated at 25°, and that petunia seed should be germinated at 30°.

In plug grown petunia flats germination temperature had a profound effect of germination rate with each increase in temperature resulting in increased germination rate. Temperature also affected plant diameter with germination temperatures of 25° and 30°C resulting in larger plants than those germinated at 20°. VPD influenced many of the growth parameters measured. Generally plants produced at 0.80 and 1.33 kPa were larger and heavier than those produced at 1.87 kPa. The interaction of temperature and VPD affected germination percentage and germination rate. At 30° germination percentage and rate were the same for all 3 VPD levels studied. At 25° germination percentage was significantly higher at 1.87 kPa than at 0.80 or 1.33 kPa, while rate was the same at all 3 VPDs studied. Germination percentage and rate were significantly lower at 20°/1.87 that at 20°/0.80 or 20°/1.33.

When 5 cultivars of refined and standard seed of _Petunia_ x _hybrida_ were compared under a single environment germination percentage and rate were significant higher in refined seed for 2 cultivars. Plant diameter was larger for refined seed in 1 of the 5 cultivars studied.

This research has shown that germination environment is a more important factor to consider when germinating petunia seed than seed type. With the proper environment similar results, in most cases, can be obtained with either refined or standard seed. At 30°C growers without good humidity control should be able to produce and market plug flats with uniform germination and growth. At 25°, a drier environment (41% RH) should be maintained for maximum germination percentage and rate. At 20° a rate of germination will be slower and fewer seed will germinate at a high VPD.

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