THE PREDICTION OF FLOWERING DATE OF PETUNIA HYBRIDA HORT., CV. WHITE CASCADE

Thesis for the Degree of M. S.

MICHIGAN STATE UNIVERSITY
STANLEY J. KAYS

1969

ABSTRACT

THE PREDICTION OF FLOWERING DATE OF
PETUNIA HYBRIDA HORT.,
CV. WHITE CASCADE

By

Stanley J. Kays

Accurate prediction of the flowering date in *Petunia hybrida* Hort. would facilitate the scheduling and growing of this plant by commercial growers. With petunia, many environmental factors exert considerable influence on the date of flowering. Correlation of several environmental factors to provide a prediction equation for first and fifty percent flower was undertaken.

Experiments were conducted with White Cascade between February and June of 1968 to determine the relative effect of growing temperature, date of planting and level of supplemental mineral nutrition on flowering and their value as factors in a working prediction equation.

Results indicated that flowering date for *Petunia hybrida* Hort., cv. White Cascade can be accurately predicted utilizing these factors under the given environmental and cultural conditions. Prediction equations for first and

50% flower were derived:

$$D = 28.55 + 7.08T + 13.41t + 4.00F \quad (1)$$

$$D_{50} = 29.04 + 7.75T + 13.42t + 5.67F \quad (2)$$

In the equation, D represents the number of days from seeding to first flower (D_{50} = days to 50% flower), T = the growing temperature, t = the time of planting and F = the level of supplemental mineral nutrients applied. The treatment levels are coded as either the number 1 or 2. The merits and demerits of the equations are discussed along with the relative effects of each factor.

THE PREDICTION OF FLOWERING DATE OF

PETUNIA HYBRIDA HORT.,

CV. WHITE CASCADE

By

Stanley J. Kays

A THESIS

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

MASTER OF SCIENCE

Department of Horticulture

1969

354056
3-18-69

ACKNOWLEDGMENTS

Appreciation is expressed to Drs. W. H. Carlson, Charles Cress, A. A. De Hertogh and A. L. Kenworthy for their advice and guidance during the course of this investigation.

Acknowledgment is made to Dr. Louis Aung for his constructive criticism and suggestions in preparation of the manuscript.

TABLE OF CONTENTS

Page

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

Hudson (21) once stated, "the condition of a plant at any time is the summation of the effects of all the environmental conditions it has experienced up to that time." Although this over simplifies the situation, it suggests that with a greater understanding and control of the environmental factors, the more closely one can manipulate and control the plant.

The prediction of plant response in relation to various environmental factors has been investigated by many workers. One of the early methods was to establish a correlation between temperature and date of harvest. Many of these approaches analyzed only what was considered the major factor affecting the variability in date of flowering or harvest. With bedding plants such as petunia, it is known that a number of environmental factors exert considerable influence on the plant's growth and subsequent flowering (34). Petunia growers use a wide range of environmental combinations for controlled production of plants according to a predetermined schedule.

Since many of the presently used methods of predicting the time of flowering correlate only those environmental factors contributing the greatest influence on this response (eg. temperature), the wide range in other factors utilized by the grower often renders them inadequate. Because of this, there is need for the correlation of several environmental factors into an equation to aid in predicting the time of flowering of petunia. An attempt to utilize this relationship for bedding plants presents a complex of problems in that many factors relate both directly or indirectly to flowering.

This study is primarily concerned with determining the effect of date of planting, temperature and mineral nutrients on the flowering of petunias. The specific question which has been raised is; can sufficient variability in flowering response be accounted for by these parameters?

REVIEW OF LITERATURE

I. Light and Flowering

(A). Some General Aspects of Light and Flowering: Energy derived from sunlight ultimately supplies all the energy for biological processes. Of the total light which strikes the earth's surface, only a small portion, 5 to 6%, is utilized in photosynthesis (1). The quality, duration and intensity of light can influence the behavior of plants in many ways: for instance, the red / far red interaction in seed germination (30).

The term photoperiodism was first used by Garner and Allard (14). It implies the ability of the plant to respond to the duration of the light period. This response has been the subject of extensive investigation since the work in 1920. Information relating to flowering has been both extensive and complex. As a result, a number of generalizations have been formulated from basic facts on flowering (40). Two of these relate to long day plants and may be expressed as:

(i). Long day plants flower in response to day lengths with light hours exceeding a certain minimum critical value. At the same time, it should be noted that the response to dark periods of less than a maximum value appears to be more critical than the response to the light period (45).

(ii). A combination of responses is commonly observed within a single plant. Many plants are day neutral in one particular temperature range and highly sensitive to photoperiod within another range (45).

The discovery (18) of the effect of intermittent light during the dark period led to the study of critical day length requirement. The critical day length concept for long day plants asserts a minimum required duration of the light period (on a 24 hour time cycle) for flowering to take place. Bonner and Liverman (7) suggested that length of the light period rather than shortness of the dark period determines the critical day length for long day plants. At present, it is generally accepted that the dark period when of a magnitude less than a maximum critical value appears to be the governing factor (40). With many long day plants (e.g. dill), a longer photoperiod, up to the critical value, results in earlier flowering (33). Near this critical length, very small changes in day length have had relatively great effects on the flowering response (24).

While it has been established that the total light intensity is only of secondary importance (14), there are several pertinent factors that merit review. Plants are very sensitive to low light intensities especially at the end of their photoperiod. The extreme daytime fluctuations in light intensity, due to clouds, for the most part do not affect the photoperiodic response of the plant (49). On the other hand, clouds may, during twilight, exert considerable influence on the measurement of day length (15, 45). Craig (12), in his study, concluded that cummulative solar energy, rather than the number of days from transplant to flowering was the major factor influencing the flowering of Pelargonium hortum class.

(B). Light and Flowering of Petunia: It was reported in 1930 that long days hastened flowering of petunia (3). Early work on the photoperiodic requirements of this plant suggested that night temperature could affect this response to the point of preventing flowering. It was concluded that petunia was day-length sensitive only between 63 and 76° F and under this regime it was a long day plant (36). This was generally accepted due perhaps in part to the fact that many plants were classed as day neutral at one temperature and day length sensitive at another (25).

Contrary to earlier reports or opinions, van der Vean and Meijer (48) found petunia to be a non-obligate long day plant. Under short day conditions and low temperature, flowering was materially delayed but eventually occurred. These effects were substantiated by Piringer and Cathey (34). Their experiments with *Petunia hybrida* Hort. showed the direct influence of photoperiod on growth and flowering. Using both early and late varieties, the plants were exposed to either an 8 or 16 hour photoperiod. In each case, earlier flowering occurred with the longer photoperiod. Many workers have since derived similar conclusions which lend additional support (4, 5, 28, 47).

II. Temperature and Flowering

(A). Some General Aspects of Temperature and Flowering: Sachs (39) in 1860 was one of the earlier workers to note and record the ability of temperature to alter the earliness of flowering in plants. The specific effect of temperature on flower initiation was studied with *Xanthium* (18). The results indicated that the temperature, during the dark period, greatly influenced initiation of flower buds. Conversely, varying the temperature during the photoperiod exerted little effect on the initiation of floral primoridia. Later research (27) showed that high and low temperatures could modify both light and dark processes in photoperiodism. The degree of variation depended upon the temperature, plant species, specific cycle, as well as the portion of light or dark chosen.

Temperature interacting with photoperiod causing a shift in phase and amplitude of the photoperiod has been shown in a number of plants (8, 9, 16, 17, 32). With

Hyoscyamus, a 3 hour cold treatment during the dark period resulted in a significant delay in flowering (41). This is also seen with Rudbeckia bicolor Nutt. The plant will flower at relatively high temperatures under photoperiods too short to permit flowering under cool conditions. However, Rudbeckia speciosa Wenderoth, remains a long day plant under either high or low temperature (31). Searle (42) suggested that low temperatures may substitute in part for darkness and high temperatures for light.

(B). Temperature and Flowering of Petunia: Roberts and Struckmeyer (36, 37) showed the effect of the interaction of temperature and photoperiod on the flowering of many plants. With Petunia hybrida Hort., they concluded that it was a temperature dependent long day plant. Later work (34), however failed to support this view. The flowering response, chronologically, may be shifted by temperature but the photoperiodic requirement is not completely eliminated. The plant will flower under low temperatures and short day conditions with an appreciable delay in flowering. Piringer and Cathey (34) demonstrated the effect of temperature on the flowering of petunia. Flowering was earliest at 80° F and slightly later at 70° F while at 60° and 50° F there was a significant delay. These results have been supported by research of a number of other workers (4, 8, 28, 43).

III. Nutrition and Flowering

(A). Some General Aspects of Nutrition and Flowering: There are numerous theories and opinions concerning the role of inorganic nutrition in the flowering of plants (6). Leopold (25) suggested that the rate of flower development was affected by inorganic nutrients; but that they had relatively little effect on floral initiation. This view has been supported by other workers (25), however, exceptions have been recorded. In tests with *Sinapis alba* Linn., nitrogen nutrition was found to be directly related to floral initiation (13). In this instance the photoperiodic reaction of the plant was not principally altered but date of flowering was appreciably modified. Very low levels of available nitrogen favored flower initiation. Calcium and phosphorous were also noted to exert an influence on flowering behavior (19). It was suggested that the effects of nitrogen on flowering were due to the production of "building materials" necessary for flower formation (13, 19).

A number of authors have reported that low nitrogen levels delayed the initiation and development of flower buds (6, 51). While prevention or restriction of vegetative growth may induce earlier anthesis in a number of plants (eg. cotton, grape) there are a number of cases where flower promoting treatments also may stimulate vegetative growth, especially for those long day plants that generally bolt before flowering (26). An instance where promotion of growth induced earlier flowering has been seen with gibberellin A_3 treatment of Lactuca sativa Linn. and Petunia hybrida Hort. (50). With Phleum pratense Linn., flowering was delayed by a low level of nutrition, especially in the instance of plants induced to flower soon after germination.

With Lycopersicon esculentum Mill., Wittwer and Teubner (51) found that high nitrogen levels (440 ppm) favored both earlier and increased flower formation. This was contrary to the earlier generally accepted idea that reproductive development in plants was favored by a high carbon to nitrogen ratio (40).

In general, the results show that low levels of available nitrogen may promote, while in other cases it may delay flowering in plants. The response will be species dependent. The strong promotive effects were almost without exception associated with long day plants while inhibition or delayed flowering was exhibited most by short day plants (19, 21).

(B). Nutrition and Flowering of Petunia: As is characteristic of most long day plants, flowering of Petunia hybrida Hort. is delayed by low levels of nutrition. A number of workers (4, 5, 44, 47) have reported data which supports this conclusion. An adequate nutritional level results in earlier anthesis without unfavorable morphological effects (4). When comparing the effect of frequency of fertilizer application, it was noted that earlier flowering occurred on those plants which had received the more frequent application even though the total amount of fertilizer applied was in all cases equal (44).

IV. Prediction of Plant Response

The prediction of plant response in relation to environmental factors has been studied by a number of workers (10, 11). Many different methods have been utilized in attempts to accurately characterize the plant and its response. One of the early methods was to establish a correlation between temperature and date of harvest. The length of temperature exposure was quantitatively represented in "heat units" or "degree days" (10). This procedure which was used extensively by de Candolle in 1854, has been useful in helping to predict flowering and harvest dates of both vegetable and fruit crops (2). This correlation, however presupposes a linear relationship between average temperature and growth and/or flowering. It is the general consensus that this relationship definitely does not exist. Over a narrow range of temperatures such a relationship may hold by approximation, and thus in relatively uniform climates the heat summary expresses the growing conditions over an entire growing season for certain plants. With many plants, such as some of the Solanaceae family where night temperatures predominately control many plant responses, heat sums, as used currently, appear to be of little value (40).

Since the advent of the digital computer, the multiple regression analysis has become more practical. This allows for the simultaneous analysis of a number of variables affecting a specific parameter. It has been used to determine a number of different relationships. Carlson (11) used plant and soil analysis data in multiple regression equations to predict the yield of roses. This, under the specified environmental and cultural conditions utilized, provided a relatively accurate method for the predetermining of yield (in this case the number of flowers produced). Hodgson (20), with this technique, investigated the seasonal changes in light radiation and temperature on the vegetative growth of *Helianthus annus* Linn. and *Vicia faba* Linn. The results demonstrated the positive dependency of net assimilation rate and relative growth rate on light and temperature. Equations to predict the net assimilation rate and relative growth rate were developed.

METHODS AND MATERIALS

I. Test Plant: *Petunia hybrida* Hort., cv. White Cascade was selected as the test plant because of uniform growth and flowering characteristics. Seed for these tests was supplied by Geo. J. Ball, Inc. of West Chicago, Illinois.

II. Seedling Growth: Seeds were germinated in a 12 x 4 x 5 ft. polyethylene film plastic intermittent mist chamber, located in a greenhouse. The ambient air temperature was maintained constant at 65° F during the first 16 days following seeding. Bottom heat was supplied by means of General Electric heating cables imbedded in a 3 in. layer of perlite. The flats were held in the chamber at 70° F for the duration of the seedling growth period. Intermittent overhead mist was utilized to maintain optimum humidity conditions. Natural photoperiod and intensity was augmented by 16 hours of light from two 40 watt cool white fluorescent lamps placed 60 cm. above the seedlings. The interval of supplemental illumination began at daylight in order to coincide with the natural light source. This provided from three to six hours of additional illumination to the natural day length depending on the time of the year.

The germination medium consisted of steam sterilized sandy loam soil, "Turface" and peat moss (1:1:1). Seeds were planted in 1/4 in. trenches in flats of moist medium and were not covered. The seeded flats were covered with panes of greenhouse glass and remained in place until after germination. The 16 day old seedlings at the two leaf stage, were then transplanted into 12 x 12 x 5 cm. undivided thin plastic containers holding approximately 600 ml. of the soil mix. Nine plants were grown per container. After transplanting, the plants were moved from the mist chamber to greenhouse benches.

III. Treatments

(A). Date of Planting: Commercial petunia growers plant seed and produce seedlings throughout an extended time period (22). A preliminary test was established in an effort to study the combined effects of a number of ecological factors (eg. longer natural photoperiod, higher daily solar radiation peaks and totals, a gradual increase in day temperature, etc. (See Figures 1 and 2) associated with differing planting dates.

Seed of the cultivar White Cascade were planted on February 1, 1968 and March 26, 1968. Seedling production methods were the same for both planting dates and were identical with those previously described. It should be noted that although the seedlings were started under a 16

Figure 1.--The increase in daylength from January 1 to May 30. The daylength at each planting date is denoted as well as at first and 50 percent flower. Data provided by Nautical Almanac Office, United States Naval Observatory, Washington, D. C.

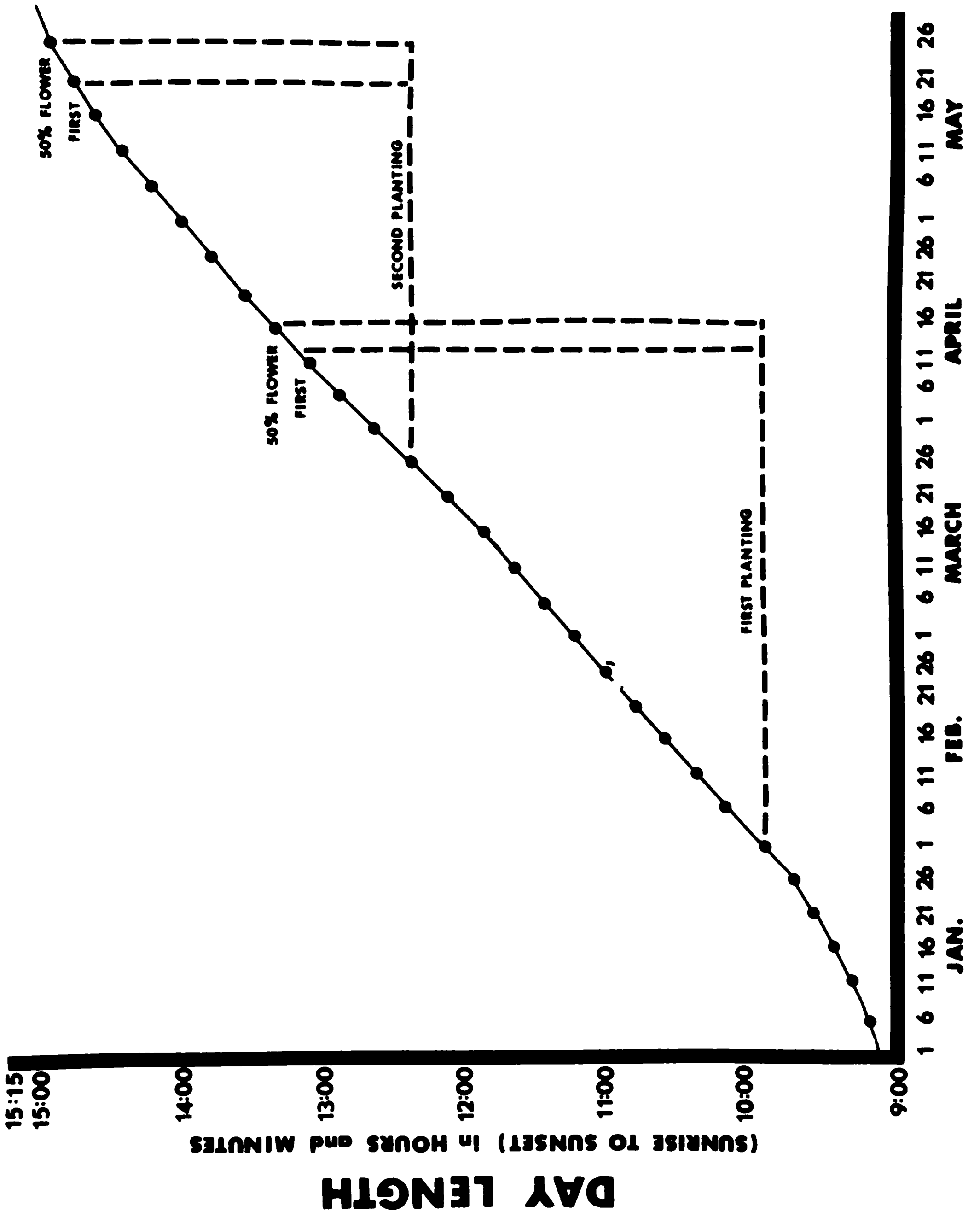
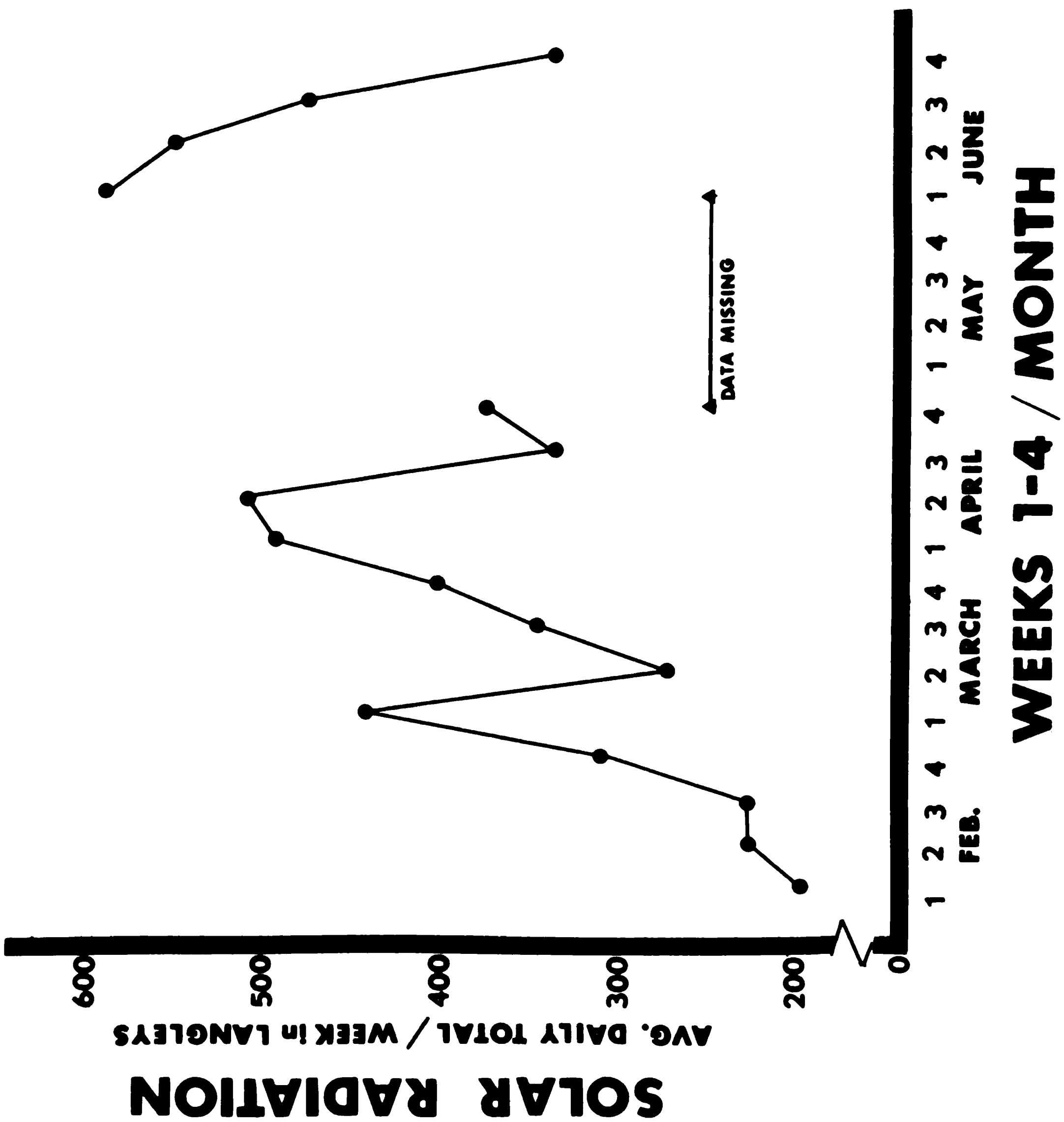

DAY LENGTH
(SUNRISE TO SUNSET) in HOURS and MINUTES
15:15
15:00
14:00
13:00
12:00
11:00
10:00
9:00
MONTH
JAN.
FEB.
MARCH
APRIL
MAY
1 6 11 16 21 26 1 6 11 16 21 26 1 6 11 16 21 26 1 6 11 16 21 26 1 6 11 16 21 26
FIRST PLANTING
SECOND PLANTING
FIRST
50% FLOWER
FIRST
50% FLOWER

Figure 2.--Cumulative (weekly) solar radiation totals in langleys are graphed in relation to date, between the first week of February and the last week of June, 1968. (Data from the U. S. Weather Bureau at East Lansing, Michigan.) Radiation readings for May, 1968 were not available.


SOLAR RADIATION
AVG. DAILY TOTAL / WEEK in LANGLEYS
600
500
400
300
200
0
DATA MISSING
1 2 3 4 1 2 3 4 1 2 3 4 1 2 3 4 1 2 3 4
FEB. MARCH APRIL MAY JUNE
WEEKS 1-4 / MONTH

hour photoperiod at both seeding dates, the natural photoperiod did increase along with a corresponding increase in solar radiation. Figure 1 shows the natural photoperiod at seeding and at 50% flower for both time treatments.

(B). Temperature: Thermostatically controlled night temperatures of 60° and 70° F were evaluated with respect to their effect on flowering date. Day temperatures were maintained as closely as possible to corresponding night temperatures. The temperature range selected for these tests was based upon recommended growing temperatures for petunia (34) and are those used by a majority of Michigan flower growers (22). The temperature treatments were initiated immediately following transplanting.

(C). Soil Fertility: The test plants were watered twice weekly with a 20-20-20 fertilizer solution at one of two rates.

(1) One pound of 20-20-20 per 100 gallons of water (240 ppm each of NO_3, P_2O_5, and K_2O).

(2) Three pounds of 20-20-20 per 100 gallons of water (720 ppm each of NO_3, P_2O_5, and K_2O).

The first application was made seven days following transplanting and continued bi-weekly to the termination of the experiment. The containers were watered sufficiently to provide for some leaching thus preventing a salt build-up in the medium.

IV. Flowering Data: The number of days to anthesis was used as a specific growth stage index. Two phases in the development of the plant were recorded.

(A). The date of first plant in each unit of nine plants to come into flower.

(B). The date when at least 50% of the plants per unit were in flower.

This information was converted to number of days from seeding until initial and 50% flowering and analyzed as such.

V. Statistical Design and Analysis: The experimental design was a split-split plot with splits for temperature and time. Variance was analyzed utilizing STAT Series program 14 of the Michigan State University Computer Laboratory (Analysis of Variance With Equal Frequency in Each Cell). In the statistical analysis, since the main concern was to determine if the factors accounted for sufficient variance to allow their use in prediction of flowering date, a multiple regression program was utilized. The least squares of variables program STAT Series (no. 8) of Michigan State University Computer Laboratory was selected. This provided among other statistics, the degree of variation accounted for by each variable and the total variation for all of the variables collectively. Using the regression coefficients for the variables, prediction equations for first and 50% flowering were derived.

RESULTS

I. Effect of Environmental Treatments on Date of First Flower

At first flower, only the main effects were found to be significant. Graphically, the response of the main effects in days to first flower is illustrated in Figure 3.

Temperature

Plants grown at 70° F flowered an average of 7 days earlier than similar plants grown at 60° F (Table 4). The temperature treatments did not interact appreciably in this test with either date of planting or level of supplemental mineral nutrients applied. Because of this, the same response in days to first flower was realized for the temperature effect at varying levels of the other environmental factors.

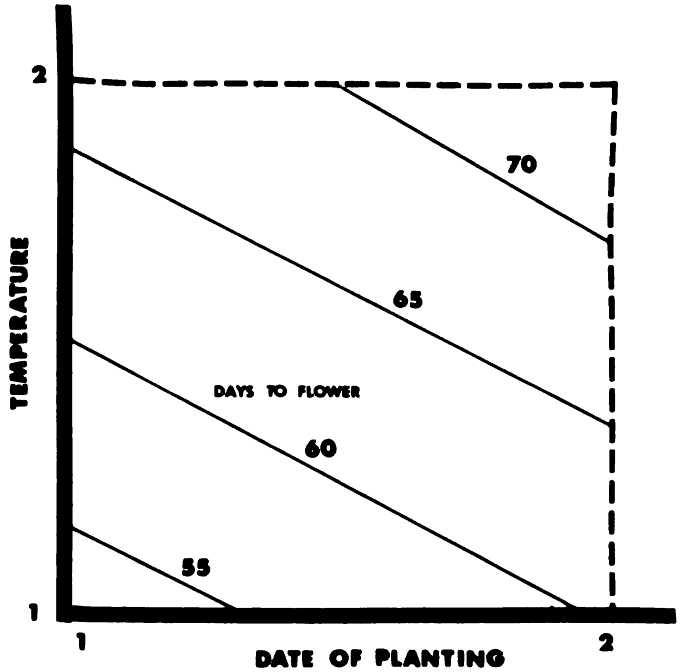
Planting Date

Plants from the later of the two planting dates (March 26) flowered an average of 13.5 days faster than those planted at the earlier time (February 1), Table 4. It may be noted that considerable change in the environment occurred between and during the two growing times. The natural daylength (as recorded by the U. S. Weather Bureau

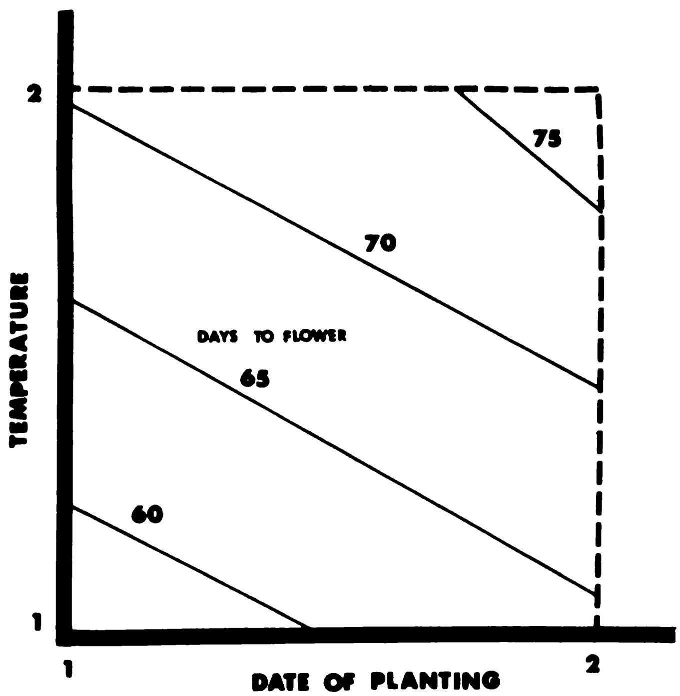
Figure 3.--The relationship of temperature and date of planting at two levels of supplemental mineral nutrients to the number of days to first flower.

HIGH LEVEL

OF SUPPLEMENTAL NUTRIENTS



LOW LEVEL



for East Lansing, Michigan) at the first planting was 9 hours and 58 minutes (Figure 1); whereas at the second planting the photoperiod was 12 hours and 26 minutes. By the time the plants in the first planting had reached first flower, the photoperiod had increased to 13 hours and 11 minutes. With the second planting at the same morphological stage of development, the photoperiod was 14 hours and 49 minutes.

Likewise, during the same time period, the level of solar radiation also increased (Figure 2). The general trend of increase was much more random.

The date of planting did not interact significantly in this test with the other factors under study.

Supplemental Nutrient Level

With the higher level of supplemental fertilizer (3# of 20-20-20 per 100 gallons of water), the number of days from planting to flower was reduced by 4 over the lower level (1# of 20-20-20 per 100 gallons of water).

Statistical Results

Only main effects were found to be significant in this study (Table 1). Consequently, the lack of significant interaction allowed the utilization of only linear terms in the multiple regression equation for characterization of the response of the plants to the environmental factors studied. Treatment means were used in the multiple

Table 1.--Analysis of variance table for first flower of *Petunia hybrida* Hort. cv. White Cascade. Split - split plot design.

Source of Variance	Degrees of Freedom	Sum of Squares	Mean Square	F Statistic	Approximate Significance
				0.6448	0.679
Temp.	1	602.0833	602.0833	46.4333	0.005**
Error (A)	10	64.8332	12.9666		
Time	1	2160.0833	2160.0833	422.1661	0.0005**
Temp. X. Time	1	0.7500	0.7500	0.1466	0.710
Error (B)	10	51.1667	5.1167		
Fert.	1	192.0000	192.0000	36.4557	0.0005**
Temp. X. Fert.	1	0.0000	0.0000	0.0000	1.000
Time X. Fert.	1	1.3333	1.3333	0.2532	0.620
Temp. X. Time X Fert.	1	8.3333	8.3333	1.5823	0.223
Error (C)	20	105.3333	5.2667		
Total	47	3185.9167			

(** Highly Significant)

regression program (Table 2). The least squares equation obtained for the prediction of first flower was:

$$D = 28.55 + 7.08T + 13.41t + 4.00F \qquad (1)$$

In the equation, D represents the number of days from seeding to first flower, T = the growing temperature, t = the time of planting and F = the level of supplemental mineral nutrients applied. Since two levels of each environmental factor were studied, the treatment levels are coded as either the number 1 or 2, i.e. the values for T, t and F ranged from 1 to 2. Using this equation, predicted values for the days to first flower were calculated and compared with the observed values (Table 3).

The multiple regression coefficients for this equation accounted for 99.65 percent of the variability in days to flower for the treatment means. The standard error of estimate for the equation was 2.29. The residuals for the means (the difference between the predicted and actual) are illustrated in Table 4. Although adequate data were not available to critically test the accuracy of the equation, it was tested against the individual measurements in this experiment (Table 3). Only two of the predicted flowering dates resulted in a residual (the difference between the predicted and the observed) that fell outside of one standard deviation from the mean.

Table 2.--Analysis of variance for overall regression (first flower of *Petunia hybrida* Hort., cv. White Cascade), regression coefficients, multiple correlation coefficient and standard error of estimate.

Analysis of Variance for Overall Regression

Source of Variance	Degrees of Freedom	Sum of Squares	Mean Square	F Statistic	Approximate Significance
Total	7	493.8837			
Regression	3	492.1539	164.0513	186.8891	0.005**
Residual + Error	20		.8778		

(** Highly Significant)

Regression Coefficients

	Constant	Temperature	Time	Fertility
Coefficients	28.55	7.08	13.41	4.00

Multiple Correlation Coefficient - $R^2 = 0.9965$

Standard Error of Estimate - $S = 2.2948$

Table 3.--Prediction equation for first flower of _Petunia hybrida_ Hort., cv. White Cascade and residuals (difference between the predicted and actual number of days to first flower).

$$D = 28.55 + 7.08T + 13.41t + 4.00F \quad (1)$$

Temperature	(High)	=1
	(Low)	=2
Time	(Second Planting)	=1
	(First Planting	=2
Fertility	(High)	=1
	(Low)	=2

Residuals Per Unit of Nine Plants

T	t	F	Actual	Pred.	Residual	T	t	F	Actual	Pred.	Residual
2	2	2	76.00	77.54	+1.54	1	2	2	66.00	64.13	-1.87
2	2	2	82.00	77.54	-4.46	1	2	2	62.00	64.13	+2.13
2	2	2	77.00	77.54	+0.54	1	2	2	62.00	64.13	+2.13
2	2	2	81.00	77.54	-3.46	1	2	2	66.00	64.13	-1.87
2	2	2	77.00	77.54	+0.54	1	2	2	65.00	64.13	-0.87
2	2	2	75.00	77.54	+2.54	1	2	2	61.00	64.13	+3.13
2	2	1	75.00	73.54	-1.46	1	2	1	60.00	60.13	+0.13
2	2	1	74.00	73.54	-0.46	1	2	1	61.00	60.13	-0.87
2	2	1	71.00	73.54	+2.54	1	2	1	62.00	60.13	-1.87
2	2	1	71.00	73.54	+2.54	1	2	1	61.00	60.13	-0.87
2	2	1	75.00	73.54	-1.46	1	2	1	60.00	60.13	+0.13
2	2	1	71.00	73.54	+2.54	1	2	1	61.00	60.13	-0.87
2	1	2	69.00	70.46	+1.46	1	1	2	54.00	57.05	+3.05
2	1	2	69.00	70.46	+1.46	1	1	2	58.00	57.05	-0.95
2	1	2	73.00	70.46	-2.54	1	1	2	65.00	57.05	-7.95
2	1	2	69.00	70.46	+1.46	1	1	2	59.00	57.05	-1.95
2	1	2	70.00	70.46	+0.46	1	1	2	53.00	57.05	+4.05
2	1	2	72.00	70.46	-1.54	1	1	2	54.00	57.05	+3.05
2	1	1	66.00	66.46	+0.46	1	1	1	52.00	53.04	+1.04
2	1	1	67.00	66.46	-0.54	1	1	1	55.00	53.04	-1.96
2	1	1	66.00	66.46	+0.46	1	1	1	52.00	53.04	+1.04
2	1	1	67.00	66.46	-0.54	1	1	1	54.00	53.04	-0.96
2	1	1	67.00	66.46	-0.54	1	1	1	51.00	53.04	+2.04
2	1	1	68.00	66.46	-1.54	1	1	1	52.00	53.04	+1.04

Table 4.--Residuals for means, first and 50% flower of Petunia hybrida Hort., cv. White Cascade.

Obs.	Temp.	Time	Fert.	Actual Y Value	Estimated Y	Residuals
FIRST FLOWER						
1	1	1	1	52.6700	53.0425	-0.3725
2	1	1	2	57.1700	57.0450	0.1250
3	2	1	1	66.8300	66.4550	0.3750
4	2	1	2	70.3300	70.4575	-0.1275
5	1	2	1	60.8300	60.1250	0.7050
6	1	2	2	63.6700	64.1275	-0.4575
7	2	2	1	72.8300	73.5375	-0.7075
8	2	2	2	78.0000	77.5400	0.4600
FIFTY PERCENT FLOWER						
1	1	1	1	56.0000	55.8725	0.1275
2	1	1	2	63.3300	61.5400	0.7900
3	2	1	1	69.8300	69.2900	0.5400
4	2	1	2	73.5000	74.9575	-1.4575
5	1	2	1	64.1600	63.6200	0.5400
6	1	2	2	67.8300	69.2875	-1.4575
7	2	2	1	75.8300	77.0375	-1.2075
8	2	2	2	84.8300	82.7050	2.1250

II. Effect of Environmental Treatments on Date of 50% Flower

The response recorded at 50% flower for the environmental parameters was very similar to that obtained at first flower. The index level of 50% flowering was considered to be attained when at least one half of the plants in a unit of nine had reached anthesis. The number of days from seeding to this stage was recorded. The data are graphically illustrated (Figure 4).

Temperature

Petunia plants that were grown at 70° F reached the 50% level of flowering an average of 7.8 days sooner than similar plants grown at 60° F. No two-factor interactions were significant, however, the three factor interaction was judged highly significant. This latter interaction will be discussed in a separate section.

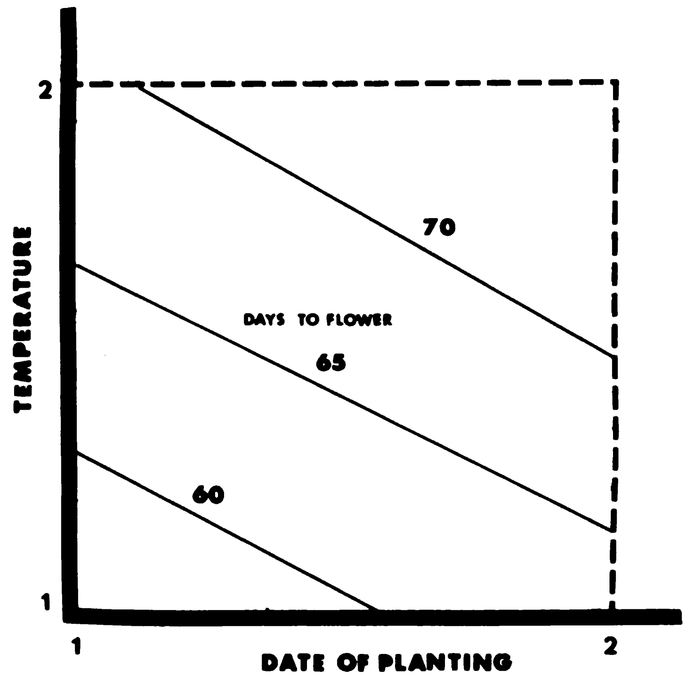
Date of Planting

As at first flower, petunias planted at the later date (March 26) flowered in a shorter period than those planted the first day of February (Table 4). The March planting reached flowering an average of 13.4 days earlier than the earlier planting. There was also a considerable change in the environment in which the plants were grown following first flower. The photoperiod (Figure 1) increased from 9 hours and 58 minutes on February 1, to 13 hours and 26 minutes (50% flowering). The photoperiod of

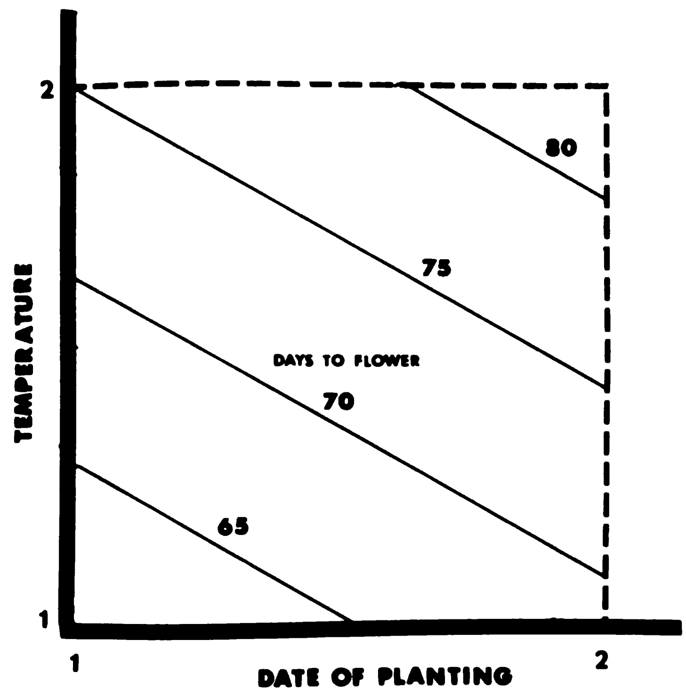
Figure 4.--The relationship of temperature and date of planting at two levels of supplemental mineral nutrients to the number of days to 50% flower.

HIGH LEVEL

OF SUPPLEMENTAL NUTRIENTS



LOW LEVEL



the later planting date (March 26) approached the critical day length for petunia when the 50% level of flowering was attained. The light period in this case increased from 12 hours and 26 minutes at seeding to 14 hours and 58 minutes at 50% flower (Figure 1).

Supplemental Mineral Nutrient Level

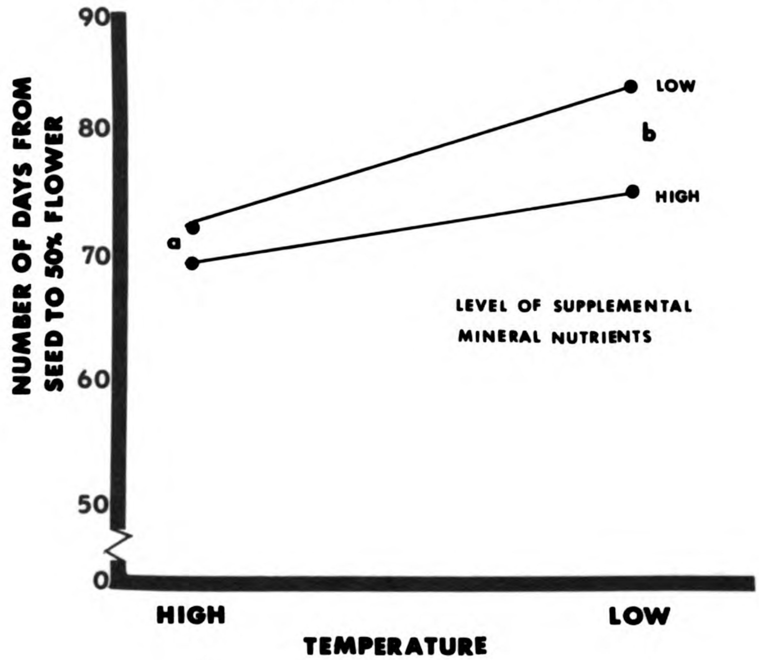
With the higher application of supplemental mineral nutrients, the petunia plants reached the 50% level of flowering an average of 5.7 days earlier than the low level. The concentrations here were 3 pounds and 1 pound of 20-20-20 fertilizer per 100 gallons of water, respectively.

Interaction

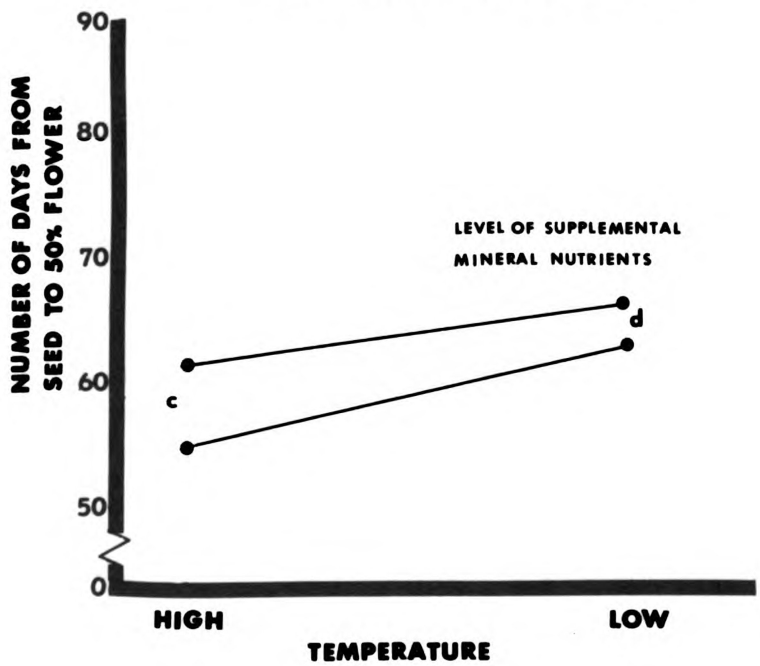
As noted, there was a significant three factor interaction between temperature, supplemental nutrient level and date of planting. Graphs of this interaction are presented in Figure 5. The magnitude of the interaction is exemplified by the failure of the two graphs to be alike. One may note that the interaction, although highly significant, is a matter of the degree of response and not difference in kind. It is possible to examine this from several aspects. For example, the delay in 50% flower at the low nutrient level compared to the high level at the early planting is (a) and (b) for the high and low temperature respectively (intervals (a) and (b) in Figure 5). The same intervals for the late planting date are (c) and

Figure 5.--Graphic illustration of the three factor interaction between temperature, date of planting and level of supplemental mineral nutrients.

PLANTING DATE 1



PLANTING DATE 2



(d) for the high and low temperatures (intervals (c) and (d) in Figure 5). The significant three factor interaction indicates that the lack of joint equality of intervals (a) and (b) with (c) and (d) is not likely to be a chance event.

The main effects were found to be statistically significant in this study (Table 5). Significance was also noted for the interaction of all three environmental factors. The presence of a significant interaction would normally require the use of cross-product terms in the multiple regression equation to characterize the response with the highest degree of accuracy. However, the overpowering size of the main effects resulted in a relatively minor improvement when cross-product terms were added to the equation (2). For this reason the simpler model with only linear terms was chosen for prediction. The treatment means were used in the regression program. From the analysis of variance, the variability in days to 50% flower accounted for by the regression model with linear terms was found to be highly significant (Table 6). The regression coefficients this determined give rise to the following prediction equation for predicting the number of days from seeding to 50% flower.

$$D_{50} = 29.04 + 7.75T + 13.42t + 5.67F \qquad (2)$$

In the equation, D represents the number of days from seeding to 50% flower, T = the growing temperature, t = the time of planting and F = the level of supplemental

Table 5.--Analysis of variation table for 50% flower of *Petunia hybrida* Hort., cv. White Cascade. Split-split plot design.

Source of Variance	Degrees of Freedom	Sum of Squares	Mean Square	F Statistic	Approximate Significance
Temp.	1	720.7500	720.7500	62.4929	0.005**
Error (A)	10	57.6667	11.5333		
Time	1	2160.0833	2160.0833	535.5579	0.0005**
Temp. X Time	1	10.0833	10.0833	2.5000	0.145
Error (B)	10	40.3333	4.0333		
Fert.	1	385.3333	385.3333	88.5824	0.0005**
Temp. X Fert.	1	5.3333	5.3333	1.2261	0.281
Time X Temp. X Fert.	1	48.0000	48.0000	11.0345	0.003**
Error (C)	20	87.0000	4.3500		
Total	47	3519.9167			

(** Highly Significant)

Table 6.--Analysis of variance for overall regression (50% flower of *Petunia hybrida* Hort., cv. White Cascade), regression coefficients, multiple correlation coefficient and standard error of estimate.

Analysis of Variance for Overall Regression

Source of Variance	Degrees of Freedom	Sum of Squares	Mean Square	F Statistic	Approximate Significance
Total	7	555.7931			
Regression	3	544.3472	181.4491	250.2746	0.005**
Residual + Error	20		.7250		

(** Highly Significant)

Regression Coefficients

	Constant	Temperatures	Time	Fertility
Coefficients	29.04	7.75	13.42	5.67

Multiple Correlation Coefficient - $R^2 = 0.9794$

Standard Error of Estimate - $S = 2.0856$

mineral nutrients applied. Since there were two levels of each factor studied, the levels were coded as either the number 1 or 2. These values were then substituted into the equation for the appropriate treatment combination to obtain the predictions in Table 7. Graphically, the predicted values are illustrated in Figure 6.

The multiple regression coefficients for this equation accounted for 97.94 percent of the variability. The residuals for the means (the difference between the actual and the predicted) are quite low in magnitude (Table 4). The standard error of estimate for the equation was 2.09. As with the equation for first flower, the equation for 50% flower was tested against individual measurements in the experiment (Table 7). The residuals indicate that the equation on the whole fairly accurately estimates the required number of days from seeding to 50% flower.

Table 7.--Prediction equation for first flower of *Petunia hybrida* Hort., cv. White Cascade and residuals (difference between the predicted and actual number of days to 50% flowering).

$$D_{50} = 29.04 + 7.75T + 13.42t + 5.67F \qquad (2)$$

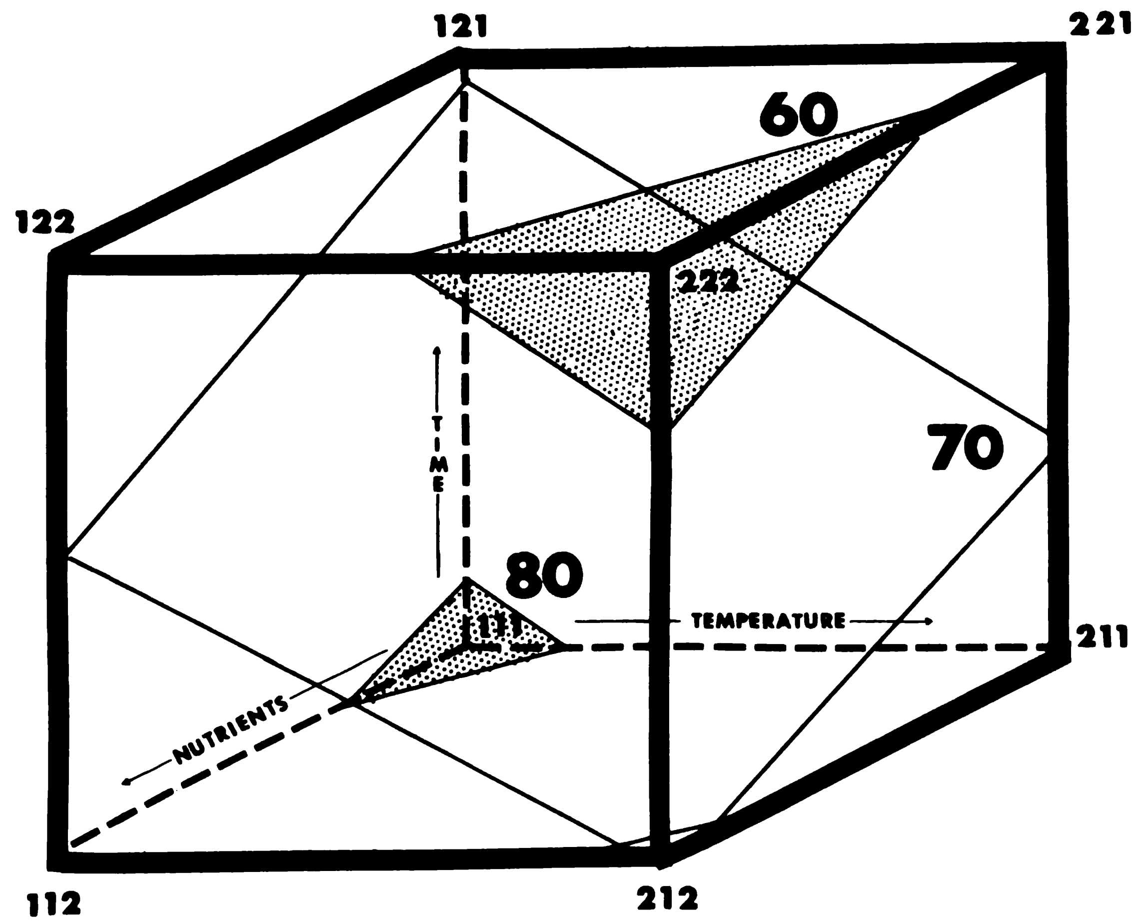
Temperature	(High)	=1
	(Low)	=2
Time	(Second Planting)	=1
	(First Planting)	=2
Fertility	(High)	=1
	(Low)	=2

Residuals Per Unit of Nine Plants

T	t	F	Actual	Pred.	Residual	T	t	F	Actual	Pred.	Residual
2	2	2	85.00	82.70	-2.30	1	2	2	69.00	69.29	+0.29
2	2	2	83.00	82.70	-0.30	1	2	2	69.00	69.29	+0.29
2	2	2	87.00	82.70	-4.30	1	2	2	67.00	69.29	+2.29
2	2	2	86.00	82.70	-3.30	1	2	2	66.00	69.29	+3.29
2	2	2	85.00	82.70	-2.30	1	2	2	68.00	69.29	+1.29
2	2	2	83.00	82.70	-0.30	1	2	2	68.00	69.29	+1.29
2	2	1	78.00	77.03	-1.97	1	2	1	63.00	63.62	+0.62
2	2	1	78.00	77.03	-1.97	1	2	1	63.00	63.62	+0.62
2	2	1	73.00	77.03	+4.03	1	2	1	63.00	63.62	+0.62
2	2	1	75.00	77.03	+2.03	1	2	1	67.00	63.62	-3.38
2	2	1	76.00	77.03	+1.03	1	2	1	66.00	63.62	-2.38
2	2	1	75.00	77.03	+2.03	1	2	1	63.00	63.62	+0.62
2	1	2	73.00	74.96	+1.96	1	1	2	58.00	61.54	+2.54
2	1	2	73.00	74.96	+1.96	1	1	2	67.00	61.54	-5.46
2	1	2	75.00	74.96	-0.04	1	1	2	69.00	61.54	-7.46
2	1	2	73.00	74.96	+1.96	1	1	2	62.00	61.54	-0.46
2	1	2	74.00	74.96	+0.96	1	1	2	58.00	61.54	+2.54
2	1	2	73.00	74.96	+1.96	1	1	2	60.00	61.54	+1.54
2	1	1	70.00	69.29	-0.71	1	1	1	57.00	55.87	-1.03
2	1	1	69.00	69.29	+0.29	1	1	1	57.00	55.87	-1.03
2	1	1	71.00	69.29	-1.71	1	1	1	57.00	55.87	-1.03
2	1	1	69.00	69.29	+0.29	1	1	1	57.00	55.87	-1.03
2	1	1	69.00	69.29	+0.29	1	1	1	54.00	55.87	+1.94
2	1	1	71.00	69.29	-1.71	1	1	1	54.00	55.87	+1.94

Figure 6.--A graph of the combination of temperature, date of planting and level of supplemental mineral nutrients required for selected days to 50% flower.

NUMBER OF DAYS FROM SEED TO 50% ANTHESIS



DISCUSSION

The response trends for the environmental factors tested support the conclusions reached by earlier workers in this area (3, 30, 35). The results indicated a definite relationship between the test factors concerning the amounts of the total variability accounted for by each. Therefore, within the test ranges for these factors, quantitative comparisons can be made. Because of the nature of the statistical design used, this presupposes a linear relationship between the parameters established for each environmental factor. Interpolation is valid if this condition is factual. The general quantitative trends are accurate estimates of variation accounted for by the particular chosen increment of each environmental factor. Extrapolation of these relationships outside the test conditions would probably lead to error.

From the regression coefficients, it is noted that the date of planting contributed the highest degree in variation in days to first and fifty percent flower. The variation between the first and second date amounted to 13.5 days for first flower and 13.4 days for fifty percent flowering. Although the conclusion that planting date per se contributes the greatest effect on the flowering response seems logical, it is, however, not substantiated. More correctly, the increment allocated for planting date (the degree of difference between the two dates) accounted for more of the variation in this test than either the range for temperature or supplemental mineral nutrient level.

The increment of ten degrees (Fahrenheit) resulted in a difference of 7 days to the first flower and 7.8 days for fifty percent flowering. As the temperature is increased up to a maximum (28), fewer days are required to obtain the flowering response. Although the linearity of this increase has not been substantiated, the general trend is valid.

The range in levels of supplemental mineral nutrients supplied to the plants accounted for the lowest degree of variation in flowering response of the factors tested. Both levels chosen were within a relatively optimal range for petunia, consequently if these levels were extended to more critical levels (e.g. nearing deficiency and/or excess) a higher variation would undoubtedly be realized. It would not be advisable to extrapolate linearly outside the experimental factor space. The variation due to level of supplemental mineral nutrients was 4 days at first flower and 5.4 days at fifty percent flower.

The high degree of correlation between these environmental factors and flowering (first and 50%) is no doubt partially due to several reasons.

One of the factors leading to the relatively high degree of accuracy of the regression equations has been the somewhat compound parameter, date of planting. Although with temperature and supplemental nutrients it was possible to construct a fairly critical test of their effects, date of planting undoubtedly entails many individual factors working collectively. It has been noted that considerable change in day length as well as daily solar radiation totals occurred during the experiment. Particularly notable is the extreme random variation in the solar radiation. Because of the potential seasonal variability, it was felt that this parameter (planting date) should be broken down into more precise components and tested with a higher degree of precision.

The use of mean values in the regression program also eliminated some degree of the inherent variation that would have otherwise been acquired. Had mean values not been utilized, the percent correlation would have been lower. In this study, it is felt that little would be gained with this procedure.

The non-universality of the present equation also merits mention. While under identical environmental and cultural conditions at Michigan State University in following years, the equations will probably yield reasonable estimates of the number of days to first flower and fifty percent flowering, few commercial growers have similar conditions. In fact, as previously stated, there is a diverse array of growing conditions used in the Southern part of Michigan alone. Because of this, factors such as structural cover, altitude, latitude, carbon dioxide level, etcetera, must be analyzed before an accurate working equation can be produced.

SUMMARY

(1) Flowering date can be predicted for *Petunia hybrida* Hort., cv. White Cascade using growing temperature, date of planting and level of supplemental mineral nutrition under given environmental and cultural conditions.

(2) Further analysis should be made of these environmental parameters, expanding the present ranges. Specific attention should be given to planting date. A more complete breakdown and analysis of contributing factors is suggested.

(3) Coefficients for other environmental factors and varieties should be formulated to increase the universality of the equations.

LITERATURE CITED

LITERATURE CITED

1. Anderson, M. C. 1964. Light relations of terrestrial plant communities and their measurement. Biol. Rev. 39:425.

2. Anstey, T. H. 1966. Prediction of full bloom date from air temperature date. Amer. Soc. Hort. Sci. 88:57.

3. Author, J. M., J. D. Guthrie and J. M. Newell. 1930. Some effects of artificial climates on the growth and chemical composition of plants. Amer. J. Bot. 17:416.

4. Ball, G. V. (ed). 1965. Bedding plants - how to grow 'em. Grower Talks. Nov. Issue.

5. ________. 1966. How to grow bedding plants. Grower Talks. Jan. Issue.

6. Blake, J. and G. P. Harris. 1960. Effects of nitrogen nutrition on flowering in carnation. Ann. Bot. 24(94):247.

7. Bonner, J. and J. L. Liverman. 1953. Hormonal control of flower initiation. p. 283 in W. E. Loomis (ed.), Growth and Differentiation in Plants. Iowa State University Press. Ames, Iowa.

8. Bunning, E. 1960. Cold Spring Harbor Symp. Quant. Biol. 25:249-56.

9. ________. 1964. The Physiological Clock. Academic Press, N. Y.

10. Candolle, A. de. 1855. Geographic Botanique Raisonnee. p. 606. Masson, Paris, France.

11. Carlson, W. H. 1966. Yield predictions of "Better Times" roses (*Rosa hybrida*) based on plant and soil analysis. Ph.D. Thesis, Pennsylvania State University.

12. Craig, R. and D. E. Walker. 1963. The flowering of Pelargonium hortorum Bailey seedlings as affected by cumulative solar energy. Amer. Soc. Hort. Sci. 83:772.

13. Doorenbos, J. and S. J. Wellensiek. 1959. Photoperiodic control of flower induction. Ann. Rev. Plant Physiol. 10:147.

14. Garner, W. W. and H. A. Allard. 1920. Effect of the relative length of day and night and other factors of the environment on growth and reproduction in plants. J. Agri. Res. 18(11):553.

15. Greuach, V. A. 1942. The length of the photoperiodically effective twilight period. Ohio J. Sci. 42:71.

16. Hammer, K. 1960. Cold Spring Harbor Symp. Quant. Biol. 25:269.

17. ________. 1963. Environmental control of plant growth. p. 215. L. T. Evans (ed.). Academic Press, New York.

18. ________, and J. Bonner. 1938. Photoperiodism in relation to hormones as factors in floral initiation and development. Bot. Gaz. 100:388.

19. Hinnawy, E. I. 1956. Mededel. Landbouwhogeschool Wageningen. 56:1.

20. Hodgson, G. L. 1967. Physiological and ecological studies in the analysis of plant environment. XIII. A comparison of the effects of seasonal variations in light energy and temperature on the growth of Helianthus annuus and Vicia faba in the vegetative phase. Ann. Bot. 31(122):291.

21. Hudson, J. P. (ed.). 1957. Control of the Plant Environment. p. 240. Butterworths Scientific Publications, London.

22. Kays, S. J. and W. H. Carlson. 1967. Unpublished data. Department of Horticulture, Michigan State University.

23. Kraus, E. J. and Krayhill. 1918. Vegetation and reproduction with special reference to the tomato. Oregon Agri. Expt. Sta. Bull. No. 149. As cited in E. J. Kraus. 1925. Soil nutrients in relation to vegetation and reproduction. Amer. J. Bot. 12:510.

24. Lang, A. and G. Melechers. 1943. Die photoperiodischi reakton von Hyoscyamus niger. Planta 33:653.

25. Leopold, A. C. 1951. Photoperiod in plants. Quart. Biol. Rev. 26:247.

26. ________. (ed.). 1964. Plant Growth and Development. p. 266. McGraw-Hill. New York.

27. Long, E. M. 1939. Photoperiodic induction as influenced by environmental factors. Bot. Gaz. 101:168.

28. Mastalerz, J. W. 1963. Controlling growth of petunias. Penn. Fl. Gr. Bull. 145:4.

29. McCree, K. J. and J. H. Thoughton. 1966. Prediction of growth rate at different light levels from measured photosynthesis and respiration rates. Plant Physiol. 41:559.

30. Mhor, H. 1964. The control of plant growth and development by light. Biol. Rev. 30:87.

31. Murneek, A. C. 1940. Length of day and temperature effects in Rudbeckia. Bot. Gaz. 102:269.

32. Nanda, K. K. and K. C. Hamner. 1959. Planta 53:45.

33. Naylor, A. W. 1941. Effects of some environmental factors on photoperiodic induction of beet and dill. Bot. Gaz. 102:557.

34. Piringer, A. A. and H. M. Cathey. 1960. Effect of photoperiod, kind of supplemental light and temperature on the growth and flowering of petunia plants. Amer. Soc. Hort. Sci. 76:649.

35. Rafter, M. E. and W. L. Ruble. 1967. Stepwise deletion of variables from a least square equation. STAT Series Description No. 8. Michigan State University Computer Laboratory (mimeo).

36. Roberts, R. H. and B. E. Struckmeyer. 1938. The effects of temperature and other environmental factors upon the photoperiodic responses of some of the higher plants. J. Agri. Res. 56:633.

37. ________. 1939. Further studies of the effects of temperature and other environmental factors upon the photoperiodic responses of plants. J. Agri. Res. 59:699.

38. Ruble, W. L., D. F. Kiel and M. E. Rafter. 1968. Analysis of variance with equal frequency in each cell. STAT Series Description No. 14. Michigan State University Computer Laboratory (mimeo).

39. Sachs, J. 1860. Physiologische untersuchungen iiber die abhangigkeit der keimung von der temperatur. Jahrb., Wiss. Bot. 1:49.

40. Salisburg, F. B. 1961. Photoperiodism and the flowering process. Ann. Rev. Plant Physiol. 12:293.

41. Schwemmle, B. 1960. Cold Spring Harbor Symp. Quant. Biol. 25:239.

42. Searle, N. E. 1965. Physiology of flowering. Ann. Rev. Plant Physiol., 16:97. (L. Machlis and W. R. Briggs, eds.) Ann. Rev., Inc., Palo Alto, California.

43. Seeley, J. G. 1955. Petunias for sale 8 weeks after potting. Penn. Fl. Gr. Bull. 51:1.

44. ________. 1965. As cited in Grower Talks, J. G. Ball (ed.). Nov. Issue.

45. Spector, W. S. (ed.). 1956. Handbook of Biological Data. p. 584. W. B. Saunders Co., Philadelphia, Pa.

46. Takimoto, A. and K. Ikeda. 1959. Studies on the light controlling flower initiation of Pharbitis nil, V. On the light following the inductive dark period. Bot. Mag. (Tokyo) 73:9o.

47. Tayama, H. K. and D. C. Kiplinger. 1966. Tips on growing bedding plants. Flor. Rev. Dec. 12.

48. van der Veen, R. and G. Meijer (eds.). 1959. Light and Plant Growth. p. 162. MacMillan Co., New York.

49. Withrow, R. B. (ed.). 1949. Photoperiodism and Related Phenomena in Plants and Animals. p. 439. Amer. Assoc. Advance. Sci., Washington, D. C.

50. Wittwer, S. H. and M. J. Bukovac. 1957. Gibberellins: new chemicals for crop production. Mich. State Agri. Expt. Sta. Quart. Bull. 39:469.

51. ________, and F. G. Teubner. 1957. The effect of temperature and nitrogen nutrition on flower formation in the tomato. Amer. J. Bot. 44:125.

APPENDIX

Appendix Table 1.--Number of days from planting to first and 50% flower for each observation.

Treatment			Number of Days to		Treatment			Number of Days to	
T	t	F	First Flower	50% Flower				First Flower	50% Flower
1	1	1	52	57	2	1	1	66	70
			55	57				67	69
			52	57				66	71
			54	57				67	69
			51	54				67	69
			52	54				68	71
1	1	2	54	58	2	1	2	69	73
			58	67				69	73
			65	69				73	75
			59	62				69	73
			53	58				70	74
			54	60				72	73
1	2	1	60	63	2	2	1	75	78
			61	63				74	78
			62	63				71	73
			61	67				71	75
			60	66				75	76
			61	63				71	75
1	2	2	66	69	2	2	2	76	85
			62	69				82	83
			62	67				77	87
			66	66				81	86
			65	68				77	85
			61	68				75	83

MICHIGAN STATE UNIV. LIBRARIES
31293105284297