

AN ABSTRACT OF THE THESIS OF

Hans-Joachim von Amsberg for the Ph.D. in Farm Crops  
(Name) (Degree) (Major)

Date thesis is presented February 26 1965

Title FLORAL AND VEGETATIVE DEVELOPMENT IN DACTYLIS GLOMERATA

AS INFLUENCED BY DIFFERENT TEMPERATURE LEVELS AND

DURATIONS

Abstract approved

(Major Professor)

Growth chamber and field experiments were conducted to investigate the effects of different temperature levels and durations on floral induction and vegetative growth in orchardgrass. Propagules of two clones, XLI-8 (clone R) and MID-5 (clone O), from the synthetic variety Pennlate, were used in all experiments.

Clone R plants were less dependent than clone O plants on extended low temperature exposure for floral induction. Temperature regimes in the growth chambers, where daily maxima were above 65°F for the durations of exposure used in these experiments, did not induce clone O propagules. However, clone R plants were induced to some degree with all temperature regimes except where the maximum temperature reached over 90°F for 10 hours each day.

The longer the exposure periods of inductive regimes, the higher the percentage of plants initiating an inflorescence and the

larger the number of tillers induced. Clone R plants were usually induced in a shorter time than was true for clone O. A positive linear relationship between floral induction and exposure time through the longest duration of 10 weeks suggests that the optimum duration of exposure under growth chamber conditions was not reached.

Constant 38°F and alternating 38°-60°F induced the highest percentage of plants of both clones. Cyclic daily temperatures of 15 hours at 38°F and 5 hours at 60°F temperatures gave the highest number of inflorescences per plant for clone O. This effect appeared to be due in part to the more favorable conditions for tillering with alternating temperatures, which resulted in more potentially inducible tillers. Alternating temperatures of 38°-60°F did not show a similar advantage over 38°F constant in number of inflorescence per plant for clone R, but did increase the number of tillers per plant.

Cyclic temperatures with high temperature maxima usually favored tiller development, both in the controlled environment chamber and after they had been transferred to the greenhouse. Constant 38°F temperatures were less promotive to tillering and reduced leaf development on the main tiller. Leaf development was accelerated by high temperatures with response related to the duration of high temperature received in each cyclic regime.

Field results indicated that the earlier the plants were transplanted to the field, the larger were the number of inflorescences. Differences in the number of floral primordia were more pronounced

for clone O than they were for clone R plants. Earlier planting dates for either clone resulted in earlier inflorescence exsertion. Inflorescence exsertion started about a week earlier for clone O than for clone R.

Clone O plants were superior to clone R plants in number of reproductive primordia, as well as in the total number of tillers developed in the field for early dates of planting. These differences were smaller or nonexistent with the later planting dates.

FLORAL AND VEGETATIVE DEVELOPMENT IN DACTYLIS GLOMERATA  
AS INFLUENCED BY DIFFERENT TEMPERATURE LEVELS  
AND DURATIONS

by

HANS JOACHIM VON AMSBERG

A THESIS

submitted to

OREGON STATE UNIVERSITY

in partial fulfillment of  
the requirements for the  
degree of

DOCTOR OF PHILOSOPHY

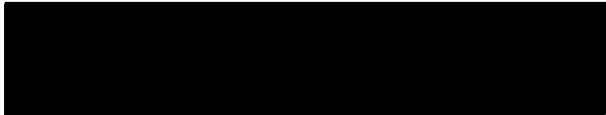
June 1965

APPROVED:



Assistant Professor of Farm Crops

In Charge of Major



Head of Department of Farm Crops



Dean of Graduate School

Date thesis is presented

February 16, 1965

Typed by Gloria M. Foster

## ACKNOWLEDGMENTS

The financial assistance provided by the Oregon Agricultural Experiment Station is gratefully acknowledged.

The writer wishes to thank Dr. J. R. Cowan, Head of the Farm Crops Department, and the other staff members, for their assistance during graduate study at Oregon State University. Thanks are also extended the advisory committee for their help in preparation of the thesis.

A special note of gratitude is extended to Dr. D. O. Chilcote for his help during the course of this study.

My wife Anne's constant help and encouragement during the course of studies is gratefully acknowledged.

## TABLE OF CONTENTS

	Page
INTRODUCTION	1
REVIEW OF LITERATURE	3
Floral Induction	4
Floral Initiation and Development	12
Modifying Effects in Floral Induction	15
Biochemical Changes in Vernalization	16
Genetic and Ecological Aspects of Vernalization	17
Vegetative Growth and Interrelationships with Floral Development	20
MATERIALS AND METHODS	24
Part I. Growth Chamber Experiments	24
Part II. Field Investigations	29
EXPERIMENTAL RESULTS	32
Part I. Growth Chamber Experiments	32
Part II. Field Experiment	61
DISCUSSION	73
SUMMARY AND CONCLUSIONS	80
BIBLIOGRAPHY	83

## LIST OF FIGURES

Figure		Page
1	Percent plants which exerted at least one inflorescence after exposure to various temperature levels and durations	34
2	Percent plants which exerted at least one inflorescence after exposure to various temperature levels and durations	35
3	Total number of tillers on 12 plants which initiated an inflorescence after exposure to various temperature treatments and durations	38
4	Total number of tillers on 12 plants which initiated an inflorescence after exposure to various temperature treatments and durations	39
5	Total number of tillers for 12 plants which exerted an inflorescence after exposure to various temperature treatments and durations	41
6	Total number of tillers for 12 plants which exerted an inflorescence after exposure to various temperature treatments and durations	42
7	Number of days required for first panicle exertion after removal from various temperature treatments and durations	43
8	Number of days required for first panicle exertion after removal from various temperature treatments and durations	44
9	Number of days required for first visible sign of anthesis after removal from various temperature treatments and durations	48
10	Number of days required for first visible sign of anthesis after removal from various temperature treatments and durations	49
11	Average number of tillers gained per plant during exposure to various temperature treatments and durations	51



LIST OF FIGURES (Continued)

Figure		Page
12	Average number of tillers gained per plant during exposure to various temperature treatments and durations	52
13	Average number of tillers gained per plant during and after exposure to various temperature treatments and durations	54
14	Average number of tillers gained per plant during and after exposure to various temperature treatments and durations	55
15	Average number of leaves gained per tiller during exposure to various temperature treatments and durations	57
16	Average number of leaves gained per tiller during exposure to various temperature treatments and durations	58
17	Average number of leaves gained per tiller during and after exposure to various temperature treatments and durations	59
18	Average number of leaves gained per tiller during and after exposure to various temperature treatments and durations	60
19	Total number of initiations for 6 plants including exerted and non-exserted floral primordia for various planting dates of clone R and O	64
20	Total number of initiated but non-exserted floral primordia for 6 plants for various planting dates of clone R and clone O	65
21	Total number of exerted inflorescences for 6 plants observed at weekly intervals for various planting dates	67

LIST OF FIGURES (Continued)

Figure		Page
22	Total number of exerted inflorescences for 6 plants observed at weekly intervals for various planting dates	68
23	Total number of tillers per plant developed for various planting dates of clone R and O	70
24	Percent tillers per plant which developed an inflorescence for various planting dates of clone R and O	71

## LIST OF TABLES

Table		Page
1	Temperature-time treatments for two clones of orchardgrass	28
2	Planting dates and hours of daylight	31
3	Percentage of plants, of a total of twelve, which initiated a floral primordia compared with the percentage which exerted an inflorescence following various temperature treatments	33
4	Total number of exerted and non-exerted inflorescences developed on 12 clone R and clone O plants after exposure to various temperature treatments	37
5	Average number of vegetative and reproductive tillers developed per plant under various temperature treatments	46
6	Percentage of total tillers per plant which became floral after exposure to various temperature treatments	47
7	Accumulated degree-hours in two degree categories calculated by averaging the temperatures over three-hour intervals for two-week field exposure periods measured at one-inch soil depth	63

FLORAL AND VEGETATIVE DEVELOPMENT IN DACTYLIS GLOMERATA  
AS INFLUENCED BY DIFFERENT TEMPERATURE LEVELS  
AND DURATIONS

INTRODUCTION

Orchardgrass (Dactylis glomerata) is a cool season perennial forage grass considered one of the leading pasture species in temperate regions of the world. In the United States this species is important, both as a seed and a forage crop. Improved varieties of this species are being utilized in several areas with seed often produced outside the region of usage. More complete knowledge of the environmental effects on growth and development of this forage grass could greatly facilitate forage and seed production.

Temperature has been found to be an important factor in the growth and particularly floral development of orchardgrass. Low temperature exposure, as well as short day length, appear to be involved in floral inductions of this species. Of the factors which influence growth and development, temperature is probably the least well understood. Under natural field conditions, temperature levels are seldom constant, but rather involve day-to-night variations. These diurnal temperature regimes may be expected to have considerable daily and seasonal variation in the temperature range and duration. Much remains to be learned about the influence of different temperature levels, alternation, and duration on growth and floral development of orchardgrass, especially since orchardgrass is a cross fertilized species and genotypes vary considerably in their

behavior under different temperature environments.

With the above factors in mind, a study was undertaken to evaluate the effect of different temperature regimes and exposure periods on the floral development and growth of two genotypes of orchardgrass. The genotypes selected were two of the four clones making up the synthetic variety Pennlate. These clones were known to behave differently under different temperature environments, thus providing a range for study of genotype-environment interaction. Field and growth chamber studies were conducted using vegetative propagules. Particular attention was given floral inductive requirements as measured by floral initiations and panicle exertion. Data on the vegetative development of the plant included tillering and leaf production.

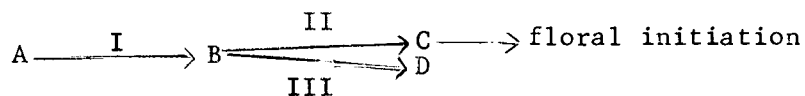
## REVIEW OF LITERATURE

Flowering and the conditions leading to the change from a vegetative to a reproductive primordia have been reviewed (42, 35) and discussed by many authors.

Gregory (32, p. 75-103) concluded from the existing evidence that flowering occurs in three phases:

1. Attainment of the ripe-to-flower condition
2. Floral initiation
3. Macroscopic bud development. Gardner and Loomis (30, p. 213) were more specific, giving the following floral stages for orchardgrass under natural conditions:
  - a. Induction in the fall
  - b. Initiation in early spring
  - c. Further development in late spring. The authors felt that the photoperiodic action was basic in floral induction and low temperature was needed to insure induction.

Early work done in the middle and at the end of the nineteenth century (14, p. 191) marked the beginning of vernalization and thermal induction research. Chouard (14, p. 228-238) cited the large amount of research devoted to this subject. Details for a suggested mechanism of photoperiod-vernalization interaction were described by Lang (42, p. 290-296). For the attainment of the induced state (vernalization), Lang and Melchers (41, p. 444) suggested the following scheme:



where C is the attainment of a definite (probably specific) chemical condition. B is an intermediary product leading either to C or D. High temperatures probably divert all of B to D. At lower temperatures, at least part of B goes to C.

Other graphic representations of the vernalization process by Purvis and Gregory and by Bakhuyzen, have been reviewed by Chouard (14, p. 216). Dooreaboos and Wellensiek (24, p. 172-174) suggested that vernalization and photoperiod are interrelated to flowering responses. The authors also pointed to the difficulty in separating the different physiological responses caused by temperature. Low temperature will bring about floral induction in different species, such as Triticum, Brassica and Streptocarpus, but the same process may not be involved. The terms floral induction, competence and vernalization have been used by several authors to describe the same phenomenon in the flowering sequence.

#### Floral Induction

A. Blühreife. The attainment of the ripe-to-flower condition or "Blühreife", has interested many investigators (12, 18, p. 521, 66). Wheat (1) and rye (64, p. 1-21) can usually be induced to flower by exposing the seed to low temperatures. Hamner (34, p. 108-116) found some evidence that the attainment of the ripe-to-flower condition is a variable factor. Treatment of the

developing zygote with low temperature while attached to the mother plant satisfied the floral induction requirements of some species.

Purvis and Gregory (61, p. 571-576) stated that about 7 leaf primordia were necessary before floral induction in winter rye could occur. Hansel (33, p. 417-425), working with the same crop, found that seedlings 20-25 mm long were just as well vernalized as were seeds where the radicle had just started to push through the pericarp.

An interaction of seed and plant vernalization was discovered by Wellensiek (73, p. 561-571) which led to a more rapid floral development. He theorized that in the juvenile plant phase, induction is not yet complete, and is fulfilled later on by the appropriate temperature and photoperiod. Cooper (19, p. 268-270) suggested that low temperatures advanced the point of "ripe-to-flowering" where the corresponding photoperiod could act. Once the plant achieved this "ripeness-to-flowering", then the environment dictated the further development of the plant.

The success or failure of seed vernalization in perennial grasses depends on the species and strain used. The attainment of floral competence in reaction to the cold stimulus has been demonstrated extensively (8, 18, p. 521, 60, 49) in the perennial grasses. Cold treatments given to water imbibed seed or seedlings of Lolium tamulentum grown to the third leaf stage were almost equally effective in their flowering response (60). McCown and Peterson (49) were able to vernalize seed of Lolium perenne. Bommer (8) exposed nine germinated perennial grass seed species to



five different cold periods and got no flowering response from Lolium perenne with less than 28 days at 2°C. Longer cold periods (up to 120 days) satisfied the low temperature requirements of the germinated seed, giving an average flowering response of 50%. Festuca rubra, Festuca pratensis, Poa pratensis, and Dactylis glomerata were shown to possess a juvenile phase and did not flower even after exposure to 120 days at 2°C.

Calder (12, p. 187-206) investigated five strains of Dactylis glomerata, placing young seedlings showing one leaf into the refrigerator at +2°C ± 2°C for three, four, five and six months. No floral induction was noted after the exposed plants were placed in continuous light for 18 weeks (p. 190). Calder concluded that orchardgrass had a juvenile stage at which inductive cold temperature treatments were without effect (p. 194).

B. Light and Temperature. The effect of light and temperature on floral induction has been reviewed extensively (14, p. 191-238; 24, p. 147-184; 35, p. 67-98; 42, p. 265-306; 48, p. 177-210). Doorenboos and Wellensiek (24, p. 147-180) stressed the importance of interacting environmental factors. They also described the short day-long day requirements of plants, as well as their vernalization needs.

Purvis (65) considered photoperiodism and vernalization so closely interrelated in winter rye that neither of the two factors should be considered separately. The author also emphasized the importance of the length of short-day for induction. In another paper, Purvis (63, p. 183-206) showed that a temperature of 1-7°C

was most effective in the vernalization of winter rye. Roberts et al. (67, p. 675-676) confirmed the modifying effect of temperature on photoperiodic responses and considered them to be systemic in the plant.

Using cereals and perennial grasses, Cooper (19, p. 268-270) found that summer annuals did not need previous low temperature exposure in order to flower. The flowering of winter annuals on the other hand, was linear to cold treatments. Many of our temperate perennial grass species need short days and/or low temperature to insure floral induction (59, p. 37, 49, 27; 23, p. 232; 30, p. 209). However, different light and/or temperature requirements, even within a species and for a particular genotype, make generalizations difficult (11; 12, p. 194-196; 21; 23, p. 232-234; 13, p. 88-95).

Peterson and Loomis (59, p. 37) found that Kentucky bluegrass required short days and cool temperatures for floral induction. For most species of Lolium perenne, a cold treatment is necessary to promote flowering (49; 27; 23, p. 232-246). Cooper (17, p. 353-355) established that the low temperature effect is stable in Lolium perenne and is maintained for about twelve months. In most strains, six weeks of cold temperature at 3°C was sufficient for the attainment of floral competence. Uniform induction of perennial ryegrass was achieved by Fejer (27) with short days and 40°F temperatures. Induction of Lolium perenne in another case (23, p. 232) was brought about by supplying 0-3°C or by an eight-hour day. Short days with low temperatures were ineffective in flower induction.

After orchardgrass has passed through the juvenile phase (12, p. 194-196), it becomes receptive to inductive environmental conditions. Under field conditions, Calder (11) noticed differential floral induction responses in different strains of orchardgrass depending on time of sowing in the spring. Gardner and Loomis (30, p. 209-210) found short days and low temperatures necessary for orchardgrass induction. These authors considered the short photoperiodic reaction basic and low temperatures necessary to insure induction. Primordia became induced when low temperatures and short days were given simultaneously, or when the low temperatures were preceded by short days. Chilcote (13, p. 29-87), comparing two different clones of orchardgrass, noted that they differed in their short-day, as well as low temperature requirements needed for floral induction. Alternating daily temperatures of 72°-45°F were superior in floral induction to continuous 72°F or 35°F temperatures.

Most recently, Calder (12, p. 187-194) studied the response of strains of orchardgrass under several natural and artificial environments. He found an absolute dark period requirement for induction. The author expressed doubts concerning the necessity of low temperatures to insure induction.

Not all temperate perennial grasses require cold treatments or short days for floral induction. None of the strains of timothy tested by Cooper (21) needed winter conditions or short days to become florally induced. All plants, however, were long-day types and high temperatures inhibited flowering. In three perennial grass species, Wycherley (76, p. 76-86) found low temperature

needed, but short days were not always essential for floral induction. Cynasurus cristatum and Lolium perenne were vernalized at 5°C. Dactylis glomerata, on the other hand, failed to become induced at that temperature. Wycherley's concurrent field experiments indicated that Dactylis glomerata required less than 5°C or needed alternating temperatures.

The time of flower induction under field conditions appears important in determining next years flowering. Knowles (40) tested the light and temperature responses of various grasses in Canada and noted that induction was fairly complete by October. Other authors (30, p. 213) concluded that orchardgrass was induced in the fall only, since day-length was a limiting factor in the spring. Winter temperatures in the Midwest were regarded as harmful or neutral to induction (13, p. 59-67). Calder (12, p. 194-196), on the other hand, feels that time of planting is less important than a long day, which is required by the maturing plant.

C. Stabilization of Induction and Devernalization. Data by Friend and Gregory (28) supported the idea of a thermostable vernalization product. Four weeks of vernalizing conditions in winter rye were followed by different periods of warm temperatures. Three to six weeks at 25°C, which usually devernalize, did not show any devernalizing effects. In another experiment (64, p. 1-21), where a 15°C period preceded warm temperatures (25°C), a thermostable vernalization product resulted. After twelve weeks of vernalizing treatment, subsequent high temperatures were not devernalizing. In winter rye, 10-15°C were found to be neutral and neither

vernalization nor devernalization occurred.

Purvis (65) also demonstrated that the vernalized stage may be reversed by high temperatures. Young rye plants could be devernalized and revernalized again, which indicated a reversible reaction. Additional work by the same author (63, p. 183-206) stressed the importance of daily alternating temperature cycles which may influence the speed of vernalization. Moisture also played an important role in achieving a thermostable product. With an unlimited moisture supply, the irreversible vernalized condition was reached much more rapidly than if a moisture stress existed. Devernalization was also much more rapid under high temperatures if moisture was limiting.

Temperatures of 15-17°C resulted in some devernalization which increased with a temperature rise to 40°C. Purvis and Gregory (64, p. 1-21) described the cyclic nature of vernalization showing that an increase in the ratio of

$$\frac{\text{time at high temperatures}}{\text{time at low temperatures}} = \text{increase in the time required for vernalization.}$$

Friend and Gregory (28), using Petkus winter rye, and Schwabe (68, p. 435-450) working with Chrysanthemum, found that high temperatures of 25° and 35°C respectively, would not devernalize plants after a prolonged period of those temperatures. Schwabe achieved complete devernalization only by long periods of low intensity light. It was rather surprising that partially vernalized rye seed was intensified in its vernalization by three to six weeks at 25°C, while six to twelve days at the same temperature were

devernalizing (28).

The importance of a neutral greenhouse temperature to prevent natural vernalization or devernalization was pointed out by Cooper (20, p. 380-382). On the basis of his experiments, he suggested that temperatures between 50<sup>o</sup> and 70<sup>o</sup>F were non-vernalizing.

Early vernalization experiments with Petkis winter rye (61, p. 569-590) led Purvis and Gregory to conclude that the whole plant was a vernalization receptor, because laterally emerging tillers flowered later on in the season. Gregory's (32, p. 75-103) subsequent experiments, however, indicated that leaves were not involved in the vernalization process. He explained lateral tiller flowering as being the result of an increase of the florally effective substance B during the autocatalytic period. Purvis and Gregory (66) demonstrated later on that substance B was also contained in rye seeds at the embryonic stage. The length of cold exposure appeared important in the determination of the total number of flowers in Lolium perenne (49). Heading could be increased from 50-90% by exposing the germinated seed for 4-20 weeks at 35<sup>o</sup>F. Four weeks of cold treatment to seedlings was more effective than 20 weeks of cold exposure to germinated seed. Chilcote (13, p. 59-67) considered the quantitative aspect of orchardgrass induction and found an increase in the number of fertile tillers with extended exposure to the inductive treatment.

### Floral Initiation and Development

Once induction has taken place, floral initiation is usually triggered by changes in photoperiod and/or temperature. Winter rye was found to be a long-day plant (65). Unvernalized rye would flower under continuous light, but would require twice as long as vernalized rye. Similar results with continuous light being superior for floral initiation and development were found by Gott *et al.* (31, p. 87-126) in winter rye and by Evans (25, p. 1-16) in subclover.

Floral initiation can take place in short days. However, stem elongation and flower exsertion will not occur (65). Winter wheat and rye, therefore, are to be considered as short-day long-day plants. Evans (26) tried to explain the photoperiodic response by suggesting that in long-day plants, the flower initiating substance is formed in the leaves and translocated to the apex. In short-day plants, on the other hand, inhibitors are formed.

Khalil (37, p. 28-32) studied the importance of night temperature on floral development of wheat. Spike development was earlier at 10°C night temperature than if the night temperature was kept at 27°C. If temperature was kept constant, spike development was delayed.

Initiation requirements for perennial grasses generally are similar to those of the cereals. Heading of perennial ryegrass (74, p. 915-932) was inhibited by day and night temperatures of 90° and 70°F respectively. The author suggested that inhibition

was probably due to a lack of floral initiation. Exposure to continuous light before cold treatment (60) of Lolium temulentum seedlings annulled the subsequent cold effect. Cooper (21) found all strains of Lolium perenne investigated to be long-day sensitive but high temperatures interfered with flower initiation. Stem elongation was not inhibited by these high temperatures, which showed the separate needs for floral initiation and stem elongation.

Several Southern grasses, such as Paspalum (38), were florally inhibited by low night temperature. A gradual delay (6) in flower initiation was observed by lowering temperatures to 55<sup>o</sup>F. Knight (39) found that high night temperatures and a 14-hour day were most favorable for flowering in Dallisgrass. An eight-hour photoperiod and low night temperatures inhibited seedhead formation.

Allard and Evans (4, p. 193-228) transferred orchardgrass plants from the field to the greenhouse in March, and with 12 hours of light, weak flowering resulted. Sixteen-hour photoperiods produced the largest number of inflorescences, longer and shorter photoperiods were less effective.

Bromegrass flowered most profusely under high light intensities and a 12-hour photoperiod. Gall (29, p. 59-71) thought long days were promotive to culm elongation. However, 13-hour photoperiods inhibited flowering in some brome grass clones. According to Cooper (15), ryegrass spikelet initiation does not take place until March or April under natural conditions.

Floral initiation in ryegrass is first distinguished by the formation of double ridges on the growing point. Cooper (17,



p. 368) showed that the initiation is a response to increased day-length. Ear development and emergence depend mainly upon temperature.

Development of the panicle and head exsertion in Lolium perenne depend on the duration of floral inductive low temperature exposure (49). For older seedlings, the shortest time between floral initiation and head exsertion was 18 days and the time required increased with successively shorter inductive low temperature treatments. The rate of floral development increased with age of plants and continuous light accelerated flowering.

Dactylis glomerata and Lolium perenne responded favorably to continuous photoperiods for initiation in Wycherley's (76, p. 87) experiments. Under field conditions (76, p. 87-88) none of the orchardgrass plants had received sufficient low temperatures for floral induction by the middle of December. Plants removed from the field after December showed more rapid head development with each succeeding transplanting to the greenhouse (30, p. 208).

In orchardgrass, the initiation of the flower primordia took place only after an exposure to long days and moderate temperatures according to Gardner and Loomis (30, p. 210-213). They considered a 24-hour photoperiod and warm temperatures as optimum for initiation. Contrary to Wycherley (76, p. 87-88), they considered fall induction only for orchardgrass, which could be maintained until initiation took place. Chilcote (13, p. 98) confirmed the long photoperiodic initiation requirements for orchardgrass and found eight hours of light to be inadequate.

Floral initiation, however, is still no guarantee of normal floral development. Orchardgrass plants might have formed reproductive primordia and later returned to the vegetative state under marginal inductive treatments (13, p. 307). Tincker (71, p. 727-728) observed that floral development of orchardgrass was abnormal under a 12-hour photoperiod. The inflorescence contained vegetative and reproductive structures which resembled viviparous fescues.

#### Modifying Effects in Floral Induction

Endogenous rhythms in plants have received limited attention concerning photoperiodic requirements. Bunning (10) proposed that plants may have a photo- and scotophile phase, where light is promotive in flowering in the photophile phase, but inhibiting in the scotophile phase. These photophases occur during a 24-hour day and are directly related to the diurnal endogenous rhythms of the plant.

Tukey (72) substantiated the endogenous rhythm of approximately 24 hours in plant growth processes. He added that the optimum cycle length probably depends on the prevailing temperatures. However, he did not consider that germination and early growth conditions were affected by it qualitatively.

Ahrens and Loomis (1) emphasized the intricacies of vernalization and the possible role of endogenous rhythms in vernalization. Vernalizing winter wheat at 1-3°C for less than three to four weeks had little effect on time of flowering, whereas periods of six to eight weeks at the same temperature produced the earliest flowering. However, when temperatures were raised to 7°C, six weeks of

exposure produced earlier anthesis than when plants were subjected to longer cold temperature treatments.

#### Biochemical Changes in Vernalization

Attempts to explain flowering on a biochemical basis have been reviewed by several authors (14, p. 191-238; 35, p. 67-98; 42, p. 265-306). Vernalization experiments by Purvis (63, p. 183-206) led to the conclusion that a substance was synthesized in the embryo, which then is transferred from the whole grain into the endosperm or aleurone layer. Low temperatures result in synthesis or accumulation of this substance, which cannot be replaced by auxins, dried yeast or ground endosperm. Purvis (62, p. 269-283) also concluded from other tests that small amounts of free sugar were necessary for successful vernalization.

Induced embryos of rye (66) which were vernalized for 10 weeks, were ground up and extracted. The extract, when applied to non-vernalized embryos, accelerated their flowering by three weeks. Nanda et al. (54, p. 5-16) postulated that during vernalization reserve carbohydrates are transformed to sugars which aid in the biosynthesis of ascorbic acid. Then if a critical level of ascorbic acid is reached, it reacts with IAA to bring about a faster transformation of the growing point.

Leopold and Guernsey (45) stressed the importance of an auxin-temperature interaction for barley vernalization. They found it essential that auxin (NAA) be accompanied by low temperature to increase flowering. A 3°C temperature and the presence of auxin

increased the number of barley flowers, while high temperature inhibited flowering.

A close relationship between a chemical component and vernalization treatments was discovered by Nitsan (57, 58). Petkus winter rye seeds kept at 2°C slowly accumulated a macromolecular compound which moved slowly when subjected to electrophoresis. This same compound was almost absent at temperatures of 26°C. At devernaling temperatures of 35°C, this component was reduced considerably. Seedlings grown at alternating high and low temperatures also lacked the macromolecular compound.

Mitchell (53) suggested that floral changes due to varied temperature periods may also involve changes in the chemical composition of the plant. Peterson and Loomis (59, p. 37) detected a close correlation between cold temperature and carbohydrate accumulation in Kentucky bluegrass. Cool temperatures and short days produced the largest carbohydrate storage. Tincker (71, p. 749) interpreted carbohydrate accumulation as being favored by long nights when condensation could take place.

#### Genetic and Ecological Aspects of Vernalization

Under natural conditions, the local environment may determine the survival of a species or ecotype. The ability to develop a normal flower and set seed are of foremost importance to plant persistence.

In Hawaii, Britten (9) observed a large genotypic variation in the flowering of Trifolium repens grown at low elevations.

Many of the "non-flowering" clones could be induced to flower at higher elevations where night temperatures were lower. Beatty (5) noticed some clover flowering modifications with different temperatures. Low temperature caused long-day plants to flower in short days and short-day plants to flower in a long photoperiod.

Cooper (20, p. 361-383) found a close correlation between temperatures needed for floral induction and the climatic origin of Lolium species. He indicated (20, p. 382) that the vernalization method could be used to assess the genetic variation within a population. Aitken (3, p. 245-257) confirmed the importance of winter temperatures for flowering in Trifolium subterraneum, showing that certain varieties of subclover were retarded in flower initiation by high mean monthly temperatures of 45-55<sup>o</sup>F.

In another article (2, p. 223-227), Aitkens showed the importance of temperatures and photoperiods in determining the length of growing season for subclover varieties. Later maturing varieties (3, p. 235-239) required longer cold temperature treatments than early varieties. The length of the cold treatment needed, however, could be shortened by long photoperiods. Kasperbauer and Loomis (36) tried to establish why sweet clover behaved like a biennial. They exposed plants to 40-45<sup>o</sup>F for four weeks, which then flowered with 13-hour photoperiods. Fifty degrees Fahrenheit were not sufficient for floral induction. The biennial varieties behaved like annuals with photoperiods from 18 to 20 hours, with 20 to 30 days needed to produce flowers. The authors concluded that the cold period requirement is the

explanation of the biennial growth habit of sweet clover.

Experiments with different strains of Trifolium subterraneum led Evans (25, p. 1-16) to the following conclusions. Flower initiation is under control of three interacting partial processes. Two of these processes are synergistic and promotive while the third one is inhibitory. The promotive processes could be independent of light and favored by low temperature, while the other one is accelerated by high temperatures. The inhibitory stage is limited to the diurnal dark period and is favored by high temperatures. Strain differences for these three partial processes revealed that two strains of subclover responded to dark inhibition which was due to night temperatures rather than dark period length. Two other strains, on the other hand, reacted just the opposite.

Generalizations concerning cold temperature requirements of perennial grasses are difficult, since great differences exist between and within species and varieties. Calder (11, 12, p. 190-192) detected population differences in orchardgrass strains derived from different geographic origins. With the same treatments, he found only an incomplete correlation between degree of heading and climatic origin. Cooper (21) related variations of May temperature inhibition in timothy. Cooper (18, p. 521-556, 22) also concluded that flowering response was closely adapted to local temperature conditions. Experimental results indicated that competence and response to photoperiods were polygenetically controlled. Chilcote (13, p. 24), who used two different clones of the synthetic variety Pennlate, noted considerable difference

between these two in the requirements for floral induction. Plants produced from seed harvested from the mother plant in a polycross nursery (13, p. 40), showed floral behavior characteristics of the parent clone.

#### Vegetative Growth and Interrelationships with Floral Development

Depending upon species, photoperiod and temperature acting as modifying agents, the vegetative development of grasses occurs usually in a rather orderly manner. Optimum growth temperatures for English grasses were considered to be between 65° and 75°F by Mitchell (53). At 65°F and 12 hours of light, orchardgrass produced the same dry matter weight as ryegrass while possessing only half the number of tillers.

Studies of leaf appearance in Lolium (16, p. 244-246) showed leaf increase to be closely related to temperature, but was not affected by photoperiods ranging from 9 to 24 hours. Leaf development was linear with time, and could be used to calculate when leaf formation ceased and floral induction took place (16, p. 244-246, 60, 70).

Cooper (19, p. 262-279) made the following points describing floral development in cereals and grasses:

1. The rate of leaf appearance is linear and is unaffected by the initiation of spikelet buds on the shoot apex.
2. The rate of apex elongation in advance of the expanding leaves is exponential. Elongation is gradual during the vegetative growth period, but spikelet initiation is

associated with an increased rate of elongation.

3. Once ear initiation has occurred in a suitable photoperiod, the elongation rate of the young inflorescence is mainly influenced by temperature.
4. Internode elongation does not occur until the late spikelet bud stage, when the apical meristem is transformed into an apical spikelet.

Purvis and Gregory (61, p. 571-576) followed the leaf development of initiated winter rye and found the final leaf number to be approximately 22. Templeton et al. (69) found that plant development was greatly influenced by the temperature and light conditions in which the plants grew. Morphological development was not a simple response to these environmental factors. Interactions among environmental factors and between environmental and genetic factors had important effects. The rate of leaf appearance on the primary shoot was increased by short days, but low temperature during the same period appeared to have the opposite effect. The interaction with age of plant, photoperiod and temperature were all significant.

Measuring number of tillers on Lolium, Cooper (16, p. 244-246) found no correlation between tillering and heading time. In young vegetative ryegrass plants, the main tiller did not inhibit lateral bud growth significantly (50, p. 433-436). Taylor (60) pointed out that new orchardgrass tillers arise from the crown of old tillers and that one fully expanded leaf would develop in 10-15 days. Gall (29, p. 68-70) found the greatest number of bromegrass tillers under natural daylength rather than with 18-hour photoperiods.



Wilson (74, p. 926) observed that under natural conditions, a high number of fertile orchardgrass tillers were formed before mid-winter.

Studying timothy tiller development, Langer (43, p. 537) labeled tillers at weekly intervals from June to September. Only those tillers which had appeared before the end of July were able to form inflorescences. Only 10 of the 88 tillers marked produced flowers. In a later study, Langer (44) showed that timothy and meadow fescue had their lowest tiller productions in July.

Tillering of tall fescue, Festuca arundinacea Schreb. (69) was increased by exposure to low temperature and by short photoperiods. At low light intensities, tillering was favored by long photoperiods. The tiller number was highest when low temperatures occurred during the light period only. Number of heads and size of perennial ryegrass increased with longer short-day periods and cool temperature, according to Fejer (27). Increasingly long days and warm temperature during floral initiation and development accelerated timothy flower exsertion (29, p. 68-70). Photoperiods above threshold daylengths had little effect on heading and anthesis time (4, p. 203-204). Peterson and Loomis (59, p. 39) obtained three times as many heads from bluegrass plants induced under low temperatures than from those grown at higher temperature levels. Detailed flowering studies by Wolfe (75, p. 605-618) indicated that flowering of a single orchardgrass clone took about 13.5 days to completion. The strains investigated did not flower at temperatures below 52°F.

Literature dealing with influence of moisture and nutrient levels on the reproductive development of the plant are scarce. For three Paspalum species, the order of anthesis time was independent of moisture levels (6). Calder (37, p. 58-60), using different orchardgrass populations, found superior flower production in two strains, with wide row spacing and high nitrogen levels compared to plants which were closely spaced at low nitrogen levels. A high C/N ratio in wheat was conducive to earlier anthesis.

Gardner and Loomis (30, p. 212-213) confirmed increased quantitative floral responses due to increased fertilization. In an experiment on orchardgrass by Wilson (74, p. 920-922), high nitrogen promoted earlier floral initiation, ear emergence, and gave a larger percentage of fertile tillers than did low nitrogen. The need of adequate fertility for floral induction experiments was supported by Sprague (69). Orchardgrass which had become pot-bound in the greenhouse, failed to flower under long photoperiods, while adequately fertilized plants flowered under the same treatment.

From the previous survey of the literature, it is apparent that floral induction and development is influenced by multiple environmental interactions. Despite the voluminous literature existing, much remains to be learned about the individual factors affecting floral induction and growth.

## MATERIALS AND METHODS

Studies were undertaken in 1962 and continued until 1964 to determine the influence of various temperature exposures on floral induction and development of orchardgrass propagules. Two orchardgrass clones, MID-5 (clone O) and XLI-8 (clone R), which are two of the four clones constituting the synthetic variety Pennlate, were used in the experiments. Henceforth, these two clones will be referred to as clone O and clone R. Both clones differ in morphological characteristics, growth habit and floral induction requirements.

Studies were conducted in the growth chamber and the field. Part I entails the controlled environment work with different daily temperature levels and durations. Photoperiods of eight hours were used in all experiments. Part II deals with the field investigations, studying the effects of duration of exposure in the field on floral induction and vegetative development of clone O and clone R.

### Part I. Growth Chamber Experiments

Growth chamber studies were started in 1962 and concluded in 1964. Two walk-in freezers obtained from U. S. Navy surplus were modified to be used as growth chambers. The chambers were equipped with automatic light, temperature and relative humidity controls.

Each chamber was lighted by 42 eight-foot fluorescent Grow-Lux Sylvania tubes, supplemented by five 100-watt incandescent

light bulbs to provide a more balanced light spectrum. Light intensity at the plant level varied from 1800 foot-candles at the start of the trials, to 1200 foot-candles during the last experiment, when the lamps had become less efficient.

A plastic, transparent barrier separated the light section from the main chamber. Heat was removed from the light section by forced air circulation. Fans forced conditioned air evenly throughout the chamber. Temperature levels were controlled by a 24-hour programmer-controller clock system, using a plastic cam cut to desired dimensions for the diurnal temperature variations. Deviations from the desired temperatures within the chambers did not exceed  $\pm 2^{\circ}$  Fahrenheit. Relative humidity was maintained at approximately 60%, regulated by an automatic control system.

Plant propagules were grown in the greenhouse. In order to maintain the orchardgrass clones vegetatively, 24 hours of continuous light were provided. Natural daylength was supplemented and extended by Sylvania Grow-Lux 40-watt fluorescent tubes. The artificial light intensity during the night at the plant level ranged from 450-600 foot-candles.

After the orchardgrass plants had been exposed to the various treatments in the growth chamber, they were transferred back into the same continuous light regime to insure rapid flowering.

Greenhouse temperatures were maintained at  $70^{\circ}\text{F}$  during the day and  $60^{\circ}\text{F}$  at night. Central steam heating was available for cold periods and a water cooled air flow system kept temperatures close to the desired limits during summer months. Continuous

temperatures were recorded in the greenhouse for both air and soil of the potted plant materials. The highest soil temperature recorded for the warmest time periods reached 85°F. The highest air temperature for the same time period was 88°F. During most of the year, temperatures ranged between the desired 60°F and 70°F temperatures.

Individual propagules of the two orchardgrass clones were planted into one-gallon metal cans. Propagule growth stage differences were reduced by separating clones periodically into individual propagules. The soil consisted of a composition of sand, loam and greenhouse peat mixed thoroughly. Each plant was watered regularly and every week a complete Hoagland nutrient solution was applied.

Plants with approximately the same visible leaf number were selected and the leaf number was recorded before they were subjected to the various growth chamber treatments. The leaf number of the main tiller was counted and the last fully emerged leaf blade was cut off about three inches from the end. This facilitated record-keeping of the number of leaves developed on the main tiller. The number of emerging tillers was also recorded for each plant.

After the vegetative development data had been taken, the plants were placed into the growth chambers. Plants were introduced into the growth chambers at two-week intervals and at the end of the test, all were removed simultaneously. This provided uniform floral development conditions for all time treatments and

facilitated record keeping.

Each temperature-time treatment was applied to twelve plants which were placed into the growth chamber according to a completely randomized design. Temperature cycles were designed to comply with a 24-hour day. The objective of the treatments chosen was to test the effect of different diurnal temperature cycles on floral induction and development of clone R and clone O. Table 1 summarizes the various temperature regime-time treatments. A constant two hours of change from one temperature level to another was chosen. This approached natural temperature changes and also was within the capacity of the growth chamber's cooling system. The time sequence of the experiments followed the order found in Table 1.

After experiments 3 and 4 had been completed, data indicated that neither of the two clones used were effectively induced with the two-week treatments. Experiments 5 and 6, therefore, had four weeks of temperature cycles as the minimum time treatment, and the longest time exposure was increased to ten weeks.

At the end of each experiment, all plants were moved to the greenhouse and set up on benches following a completely randomized design. The leaf number and number of tillers gained were counted and added to the previous figures. The last fully emerged leaf blade was clipped three inches from the end. Flower emergence was checked periodically and the date when the inflorescence became visible was recorded. The date when anthers first visibly protruded from a floret was recorded as the time of anthesis.

## Growth Chamber

Table 1. Temperature-time treatments for two clones of orchardgrass

<u>Treatment number</u>	<u>Temperature regimes</u>	<u>Time</u>
1	10 hours at 38°F	(2 weeks
	10 hours at 70°F	(4 weeks
	+ 4 hours change	(6 weeks
		(8 weeks
2	24 hours at 38°F	(2 weeks
		(4 weeks
		(6 weeks
		(8 weeks
3	15 hours at 38°F	(2 weeks
	5 hours at 70°F	(4 weeks
	+ 4 hours change	(6 weeks
		(8 weeks
4	10 hours at 38°F	(2 weeks
	10 hours at 90°F	(4 weeks
	+ 4 hours change	(6 weeks
		(8 weeks
5	15 hours at 38°F	(4 weeks
	5 hours at 60°F	(6 weeks
	+ 4 hours change	(8 weeks
		(10 weeks
6	20 hours at 38°F	(4 weeks
	to peak at 65°F	(6 weeks
	+ 4 hours change	(8 weeks
		(10 weeks

Each experiment was discontinued when no additional elongating inflorescences could be observed. The total number of initiated, but not fully exerted, primordia (as evidenced by partial internodal elongation or rudimentary floral development) and the total number of flowers per plant were recorded. The final number of leaves on the flowering stem and the total number of tillers per plant were tallied.

## Part II. Field Investigations

The outdoor experiment was conducted during the winter of 1963 and the first half of 1964. Plantings were located on the Oregon State University campus in the Farm Crops nursery area.

The soil of the experimental site, a Woodburn silt loam, was deep-rototilled twice before planting. Nitrogen and phosphorus at a rate of 80 pounds per acre were spread evenly over the whole experimental area with a hand-drawn fertilizer spreader.

Clone R and clone O propagules were used for the field experiment. Plants were periodically separated into individual propagules and plants of a similar growth stage were selected for each treatment. Altogether there were eight treatments, each involving six plants of clone R and O, which were set out into the field two weeks apart, beginning November 22, 1963 and ending February 28, 1964.

The plot design was a randomized block with six replications, each plant representing one replication. The propagules were planted three feet apart, a plant of clone O being adjacent to the



corresponding plant of clone R.

Temperature was measured during the course of the field experiment by a continuous chart temperature recorder. The sensitive element was inserted one inch below the soil surface under a selected plant. Twenty-four hour daily temperatures were divided into three-hour segments and averages were calculated for these intervals. Table 2 indicates planting times and hours of daylength received by the various treatments. Data for sunrise and sunset were obtained from the United States Naval Observatory.

During the fall and early winter months, annual bluegrass, wild onions and annual ryegrass began competing with the crop. In December 1963, these weeds were effectively controlled by directed spraying between the rows with paraquat at the rate of one pound of active material per acre. Soil moisture was adequate in the Willamette Valley from November to May. In June when soil moisture became limiting, two irrigations provided sufficient moisture until the experiment was discontinued in July.

Data were taken at weekly intervals on number of exerted inflorescences. At the end of the experiment, in July 1964, the total number of flowers and the initiated, but not fully exerted floral primordia per plant was recorded. The total number of tillers per plant was counted and the percentage reproductive tillers of the total tillers per plant was calculated.

## Field Experiment

Table 2. Planting dates and hours of daylight

<u>Treatment</u>	<u>Date of planting</u>	<u>Daylight</u>	
		<u>Hours</u>	<u>Minutes</u>
1	Nov. 22	9	24
2	Dec. 6	9	00
3	Dec. 20	8	53
4	Jan. 3	9	01
5	Jan. 17	9	22
6	Jan. 31	9	53
7	Feb. 14	10	32
8	Feb. 28	11	11

## EXPERIMENTAL RESULTS

Part I. Growth Chamber Experiments

## 1. Floral Induction

## A. Floral initiation and panicle exertion

In order to evaluate the effectiveness of various temperature treatments and durations for floral induction, subsequent floral initiation and panicle exertion were observed in the greenhouse under 24-hour photoperiods.

The percentage of plants, from a total of twelve, which had initiated at least one floral primordia compared with the percentage of plants with at least one exerted inflorescence, are presented in Table 3. The histograms in Figures 1 and 2 give a graphical concept of the percentage of plants exerting an inflorescence when given various temperature treatments. Those temperature levels and durations which failed to induce flower development are not included.

Inflorescence exertion was observed in clone R propagules under all temperature treatments, except where daily maxima were 90°F (Figure 1). However, only temperature levels with a maximum temperature of 65°F or below were able to promote panicle exertion in clone O. With day temperatures of 70°F or more, normal flower exertion and development failed.

Six weeks of treatment 1, 2 and 5 appeared sufficient for every plant of clone R to exert at least one florescence. Other temperature regimes (Figure 1) with from four to eight weeks exposure, satisfied the requirements for only part of the clone R

Table 3. Percentage of plants, of a total of twelve, which initiated<sup>1/</sup> a floral primordia compared with the percentage which exerted<sup>2/</sup> an inflorescence following various temperature treatments.

		Treatment Number											
		1		2		3		4		5		6	
		Temperature Regimes											
		10 hrs. 38°F		24 hrs. 38°F		15 hrs. 38°F		10 hrs. 38°F		15 hrs. 38°F		20 hrs. 38°F	
		10 hrs. 70°F		24 hrs. 38°F		5 hrs. 70°F		10 hrs. 90°F		5 hrs. 60°F		to peak 65°F	
		+4 hrs. change		38°F		+4 hrs. change		+4 hrs. change		+4 hrs. change		+4 hrs. change	
Type of floral development	Exposure time in weeks	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O
Exserted <sup>2/</sup>	10									100	92	100	58
Initiated <sup>1/</sup>										100	100	100	67
Exserted	8	67	0	92	75	83	0	0	0	100	75	50	33
Initiated		100	8	100	92	83	0	0	0	100	83	92	33
Exserted	6	100	0	100	67	83	0	0	0	100	67	50	0
Initiated		100	0	100	75	92	8	0	0	100	83	100	8
Exserted	4	33	0	58	8	0	0	0	0	42	0	8	0
Initiated		83	0	75	8	0	0	0	0	75	0	50	0
Exserted	2	0	0	0	0	0	0	0	0				
Initiated		17	0	0	0	8	0	0	0				

<sup>1/</sup>Initiated - as measured by a nodal stem or rudimentary inflorescence development

<sup>2/</sup>Exserted - an inflorescence which became visible

Figure 1. Percent plants which exerted at least one inflorescence after exposure to various temperature levels and durations

Treatments:

1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change

2=24 hrs. 38°F

3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change

5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change

6=20 hrs. 38°F to peak 65°F+4 hrs. change

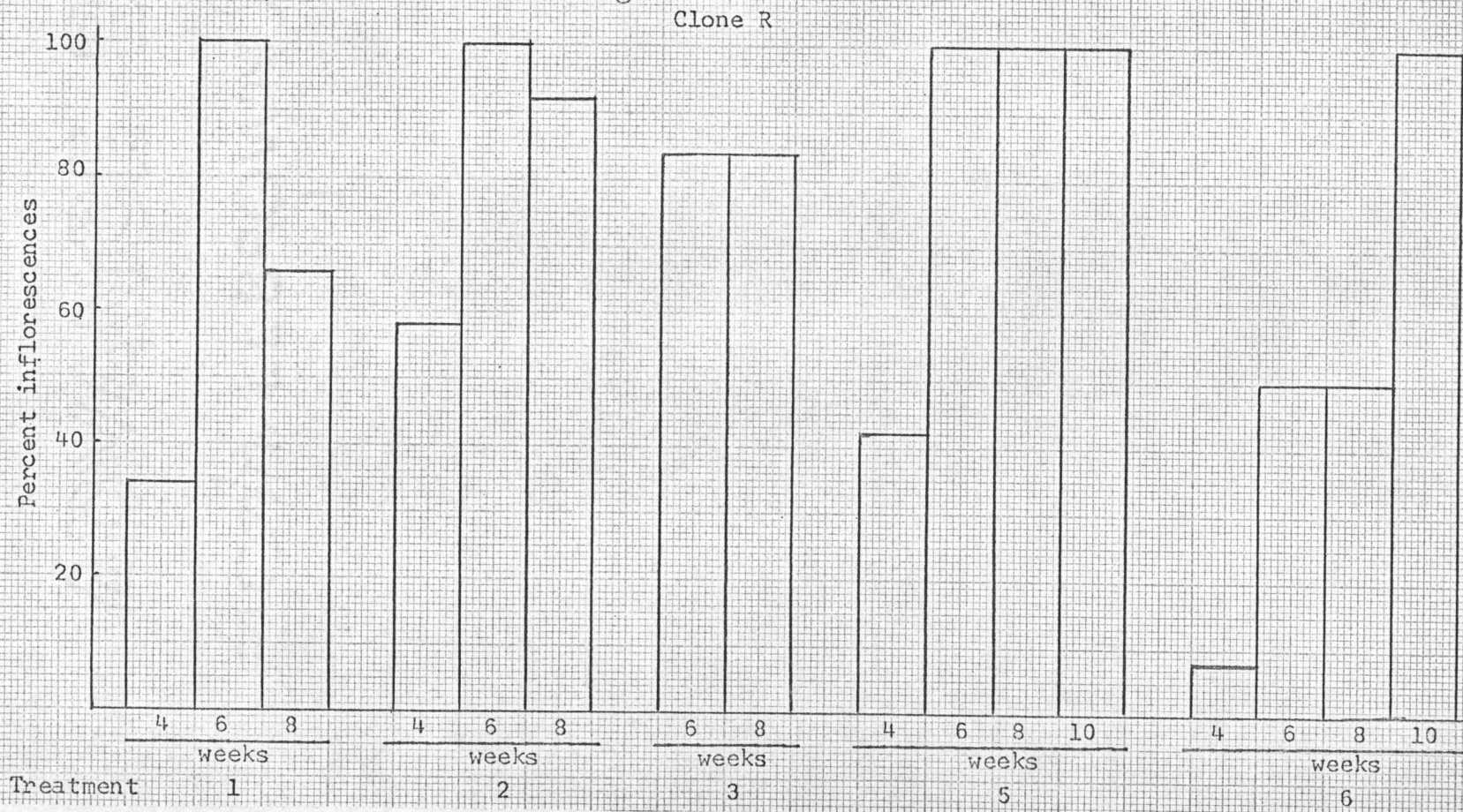
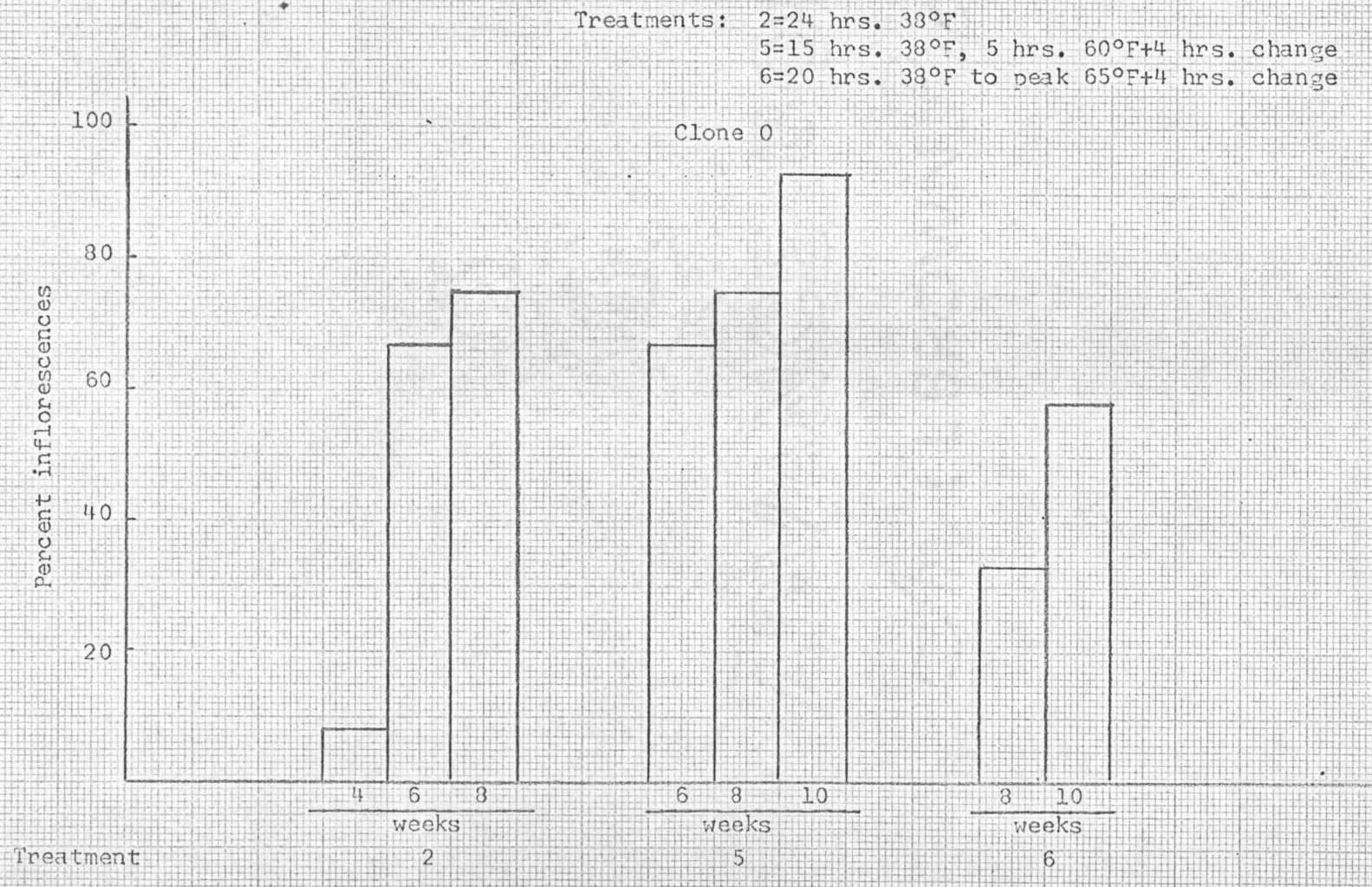


Figure 2. Percent plants which exerted at least one inflorescence after exposure to various temperature levels and durations



propagules. A comparison of treatment 2 and 5 in Figure 1 shows these two to be the most effective treatments, as far as percentage of plants exerting an inflorescence.

Clone 0 appears to require a minimum duration of six to eight weeks at temperature levels below 70°F to give a high percentage of florally induced plants with subsequent panicle exertion (Table 3 and Figure 2). Length of treatment exposure was especially important in clone 0 as seen in Figure 2. Exposure to short day temperature cycles less than six weeks in duration were largely ineffective in promoting inflorescence exertion in clone 0, whereas in clone R considerable inflorescence exertion resulted from four weeks treatment.

Data showing the effect of different daily temperature cycles of varying duration on the total number of both exerted and non-exserted inflorescences on all plants are summarized in Table 4. A graphic comparison between clone R and clone 0 (Figures 3 and 4) shows that a larger number of tillers developed an inflorescence for clone R than clone 0 under all effective temperature levels and durations.

As far as total number of exerted inflorescences are concerned, treatment 5 (15 hours at 38°F and 5 hours at 60°F with four hours change) proved to be the most effective one for clone 0. The longest time exposure of ten weeks also gave the largest number of inflorescences.

In clone R (Figure 3) treatments 3 and 6, with 5 hours at 70°F and a peak of 65°F respectively, were less effective in total

Table 4. Total number of exerted and non-exserted inflorescences developed on 12 clone R and clone 0 plants after exposure to various temperature treatments.

		Treatment Number											
		1		2		3		4		5		6	
		Temperature Regimes											
		10 hrs. 38°F		24 hrs. 38°F		15 hrs. 38°F		10 hrs. 38°F		15 hrs. 38°F		20 hrs. 38°F	
		10 hrs. 70°F		24 hrs. 38°F		5 hrs. 70°F		10 hrs. 90°F		5 hrs. 60°F		to peak 65°F	
		<u>+4 hrs.change</u>		<u>38°F</u>		<u>+4 hrs.change</u>		<u>+4 hrs.change</u>		<u>+4 hrs.change</u>		<u>+4 hrs.change</u>	
Type of floral development	Exposure time in weeks	Number of floral primordia											
		Clone R	Clone 0	Clone R	Clone 0	Clone R	Clone 0	Clone R	Clone 0	Clone R	Clone 0	Clone R	Clone 0
Exserted	10									90	44	57	14
Non-exserted										9	6	17	2
Exserted	8	18	0	50	12	28	0	0	0	52	24	10	5
Non-exserted		40	1	41	9	19	0	0	0	22	4	27	3
Exserted	6	35	0	53	15	25	0	0	0	65	26	16	0
Non-exserted		41	0	27	3	16	0	0	0	15	8	23	1
Exserted	4	4	0	8	1	0	1	0	0	7	0	1	0
Non-exserted		25	0	11	0	0	0	0	0	23	0	6	0
Exserted	2	0	0	0	0	0	0	0	0				
Non-exserted		4	0	0	0	1	0	0	0				



Figure 3. Total number of tillers on 12 plants which initiated an inflorescence after exposure to various temperature treatments and durations

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

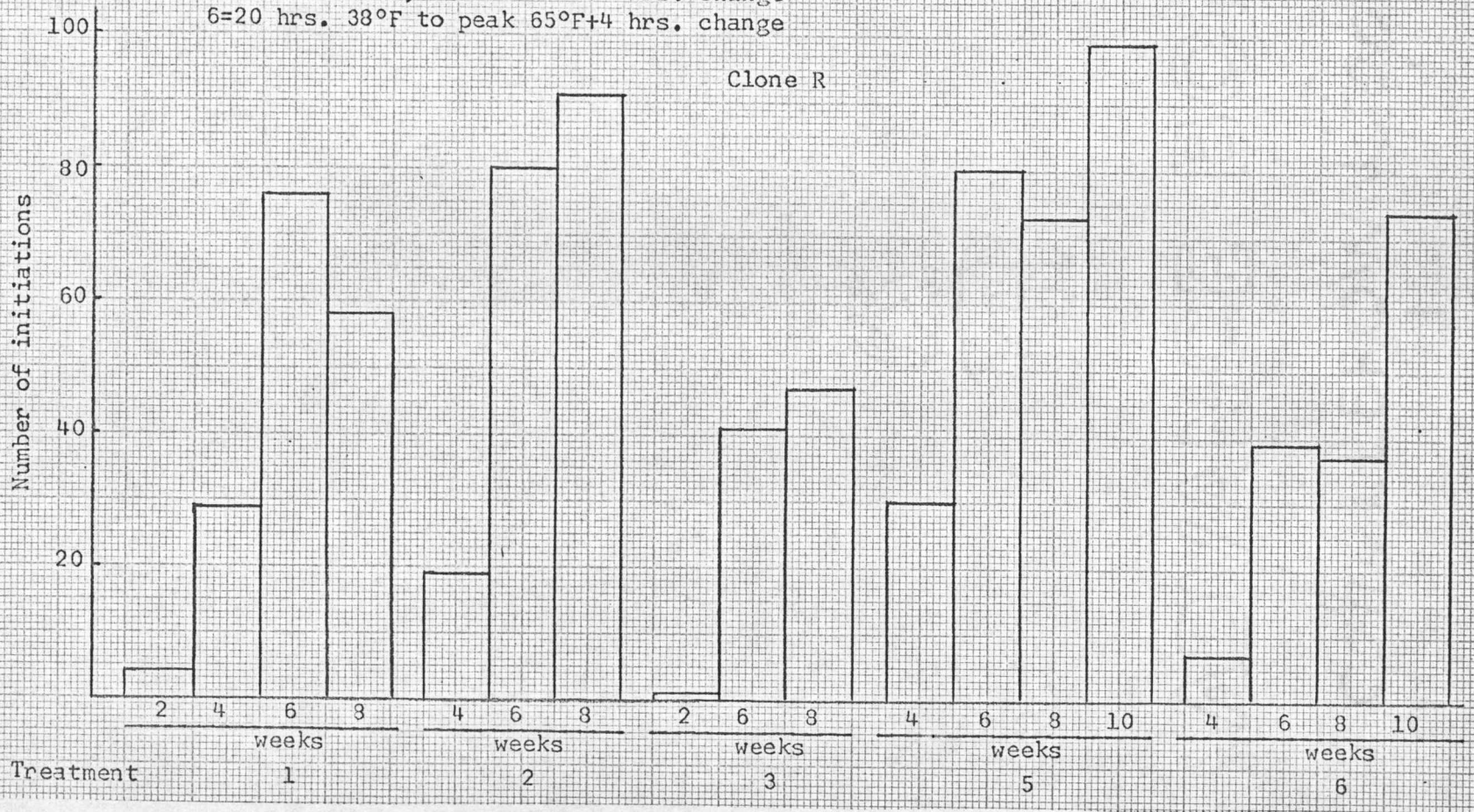
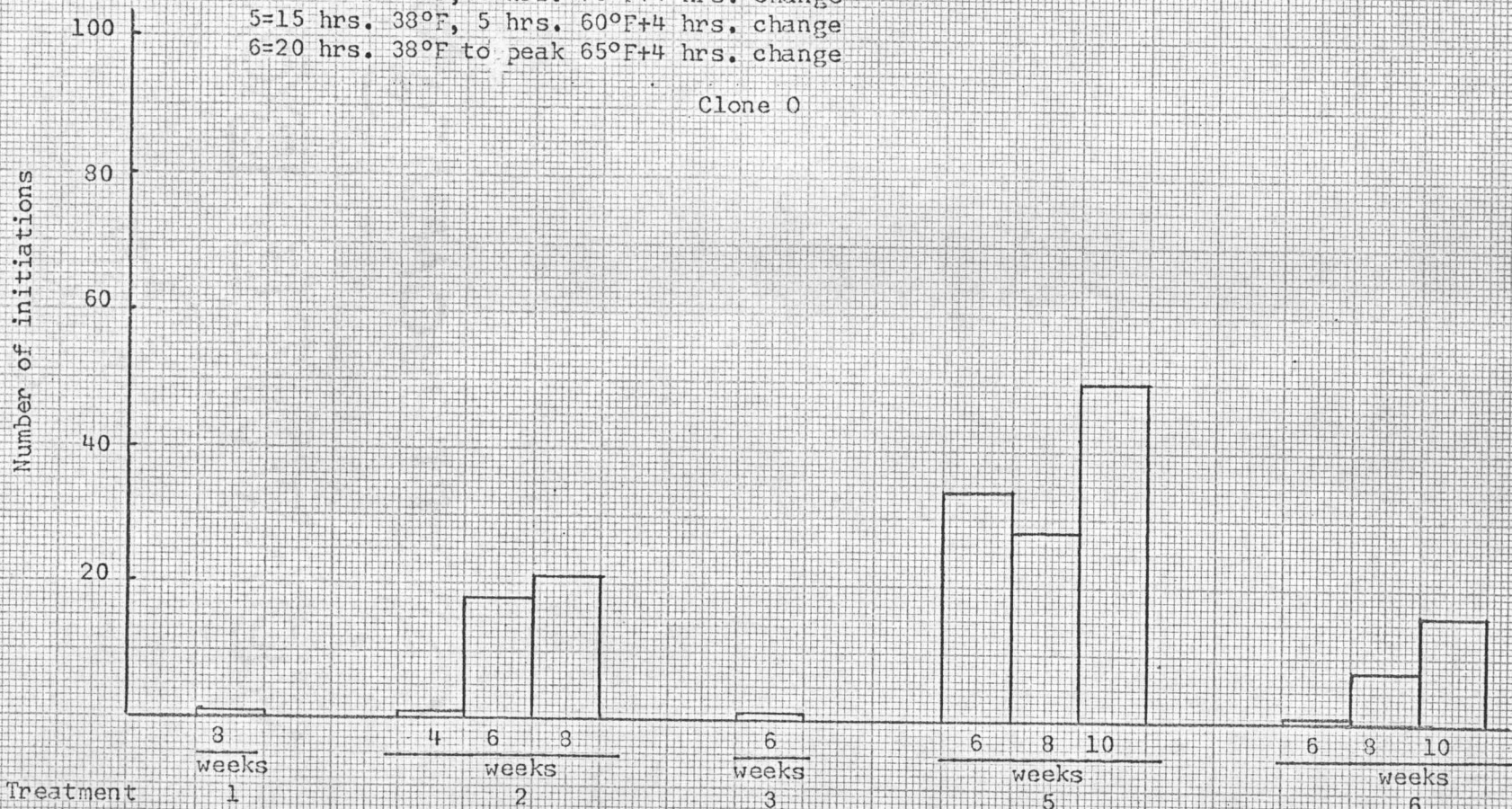


Figure 4. Total number of tillers on 12 plants which initiated an inflorescence after exposure to various temperature treatments and durations

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

Clone 0



number of floral inductions than were treatments 1, 2 and 5 at the same six-week exposure duration. The longer the time exposure to a given treatment, the higher the number of reproductive primordia developed. However, there was little apparent difference in total number of floral primordia developed between the six- and eight-week exposure periods.

The total number of exerted inflorescences for each temperature-exposure time treatment are presented as histograms in Figures 5 and 6. These figures show that initiation can occur with shorter duration of less favorable temperature regimes than are required for completion of the flowering process. The number of floral initiations were always considerably higher than exerted inflorescences. Effects of treatments on exertion followed the same general trend as for floral initiations. Of interest was the relatively lower number of non-exserting floral primordia with treatment 5 compared to treatment 2 in clone R.

#### B. Heading and anthesis

The number of days required for the first panicle per plant to exert are presented in form of histograms in Figures 7 and 8 for clone R and O respectively.

With the most favorable temperature-duration treatments, the minimum time period required for panicle appearance was 20 to 22 days for either clone (Figures 7 and 8). Time exposures within a treatment generally showed an inverse linear relationship with days needed for panicle exertion. The six-week exposure period

Figure 5. Total number of tillers for 12 plants which exerted an inflorescence after exposure to various temperature treatments and durations

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hours 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

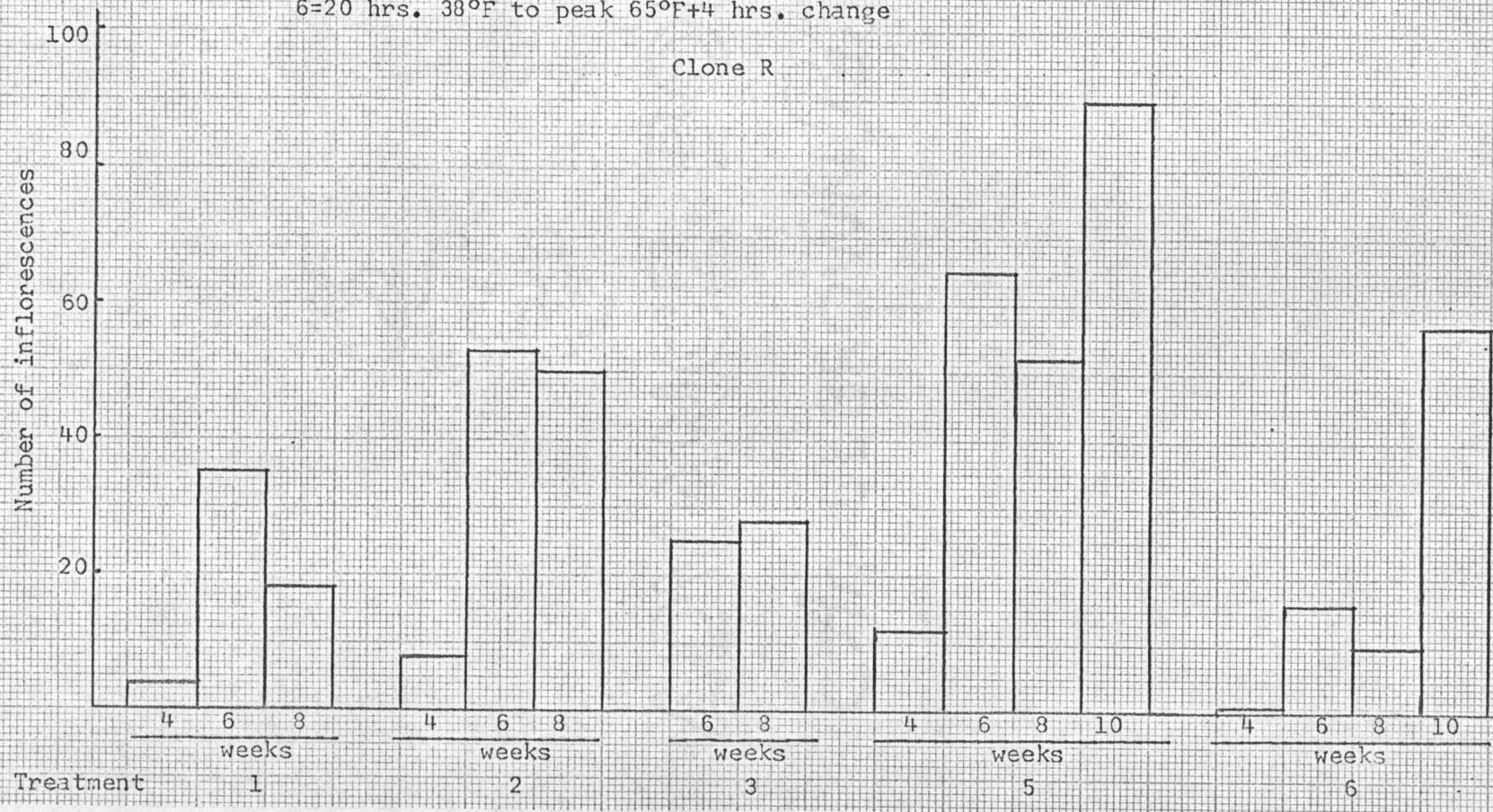


Figure 6. Total number of tillers for 12 plants which exerted an inflorescence after exposure to various temperature treatments and durations

Treatments: 2=24 hrs. 33°F  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

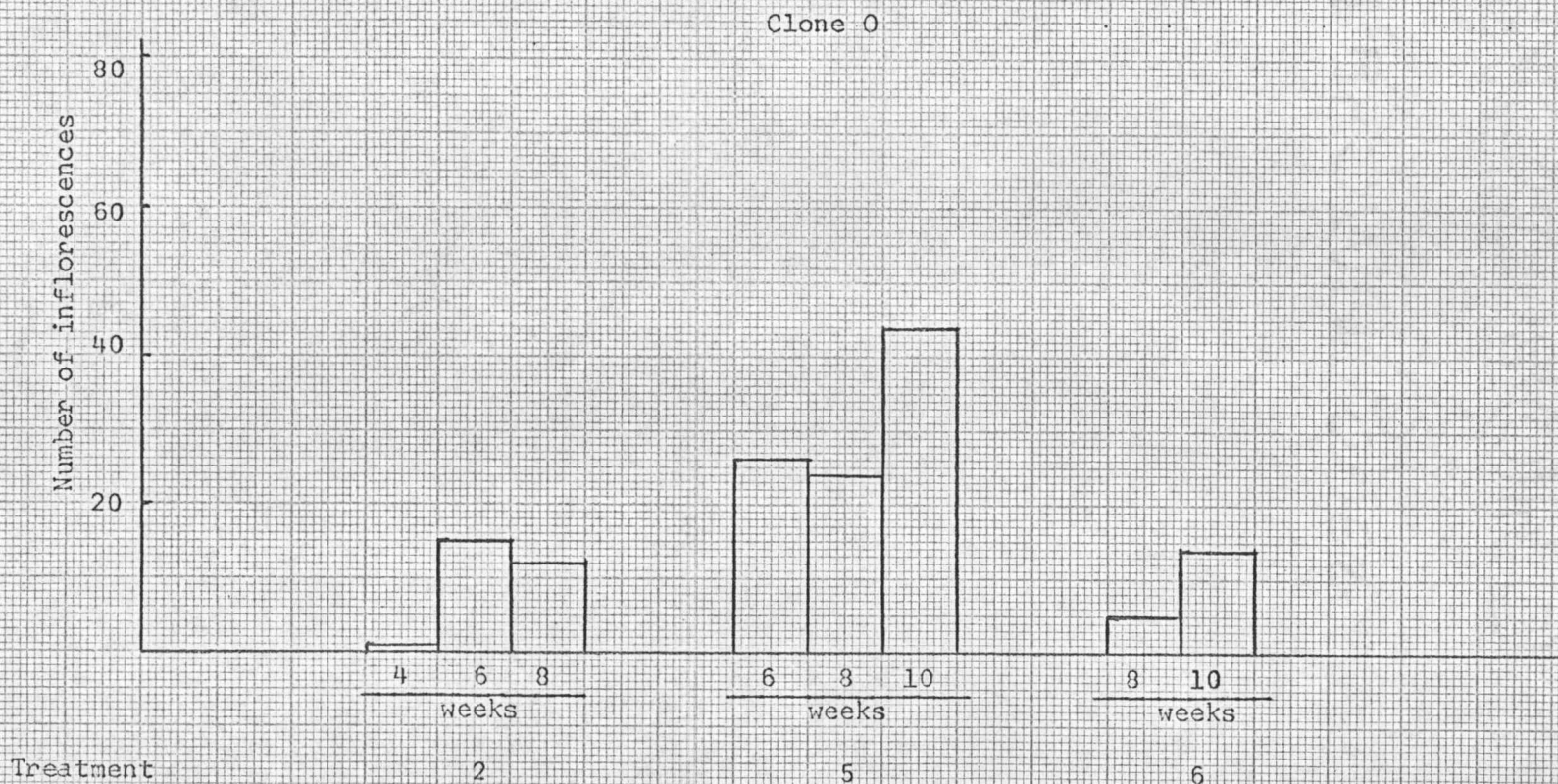


Figure 7. Number of days required for first panicle exertion after removal from various temperature treatments and durations

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change      5=15 hrs. 33°F, 5 hrs. 60°F+4 hrs. change  
 2=24 hrs. 38°F      6=20 hrs. 33°F to peak 65°F+4 hrs. change  
 3=15 hrs. 33°F, 5 hrs. 70°F+4 hrs. change

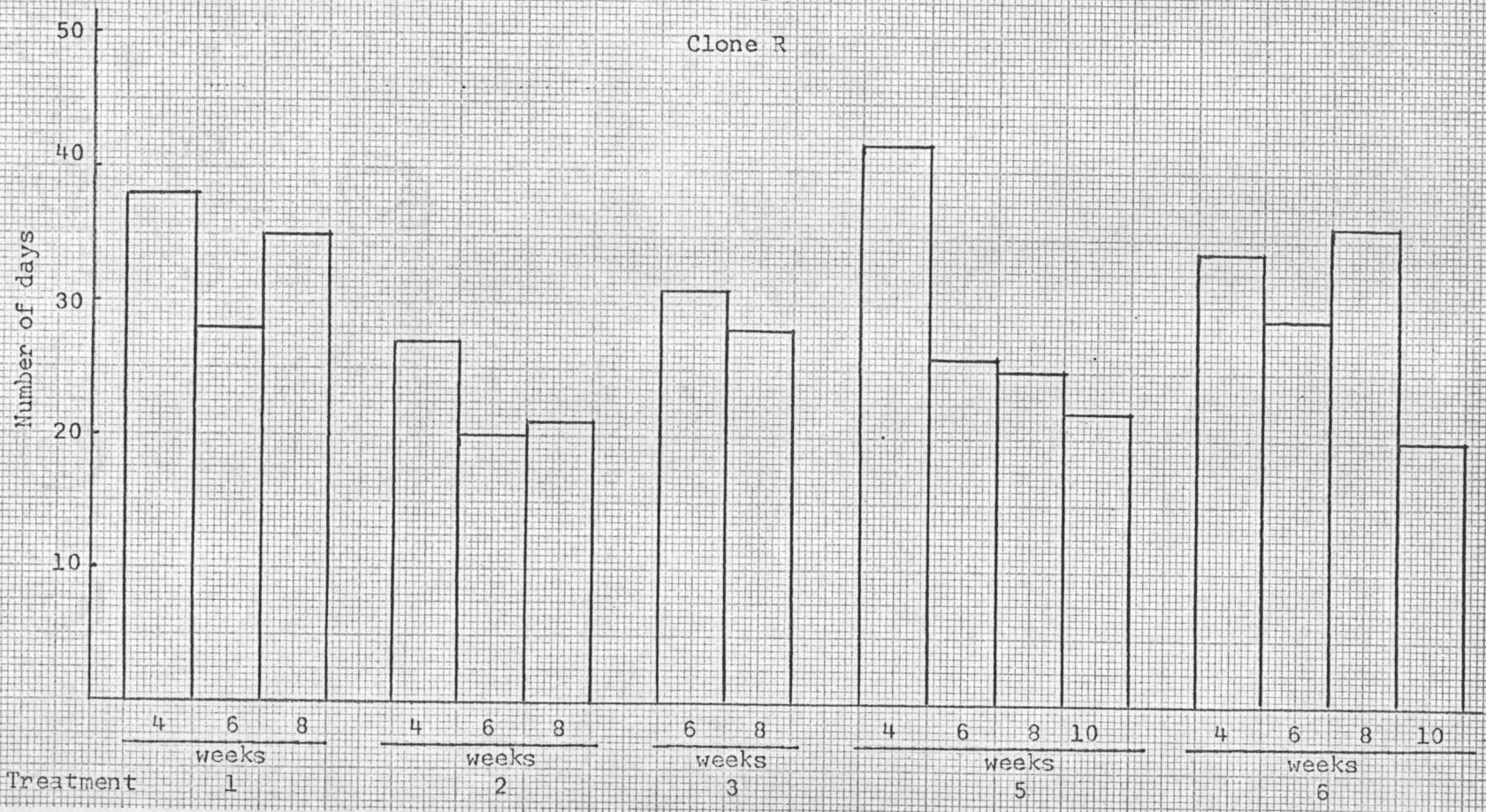
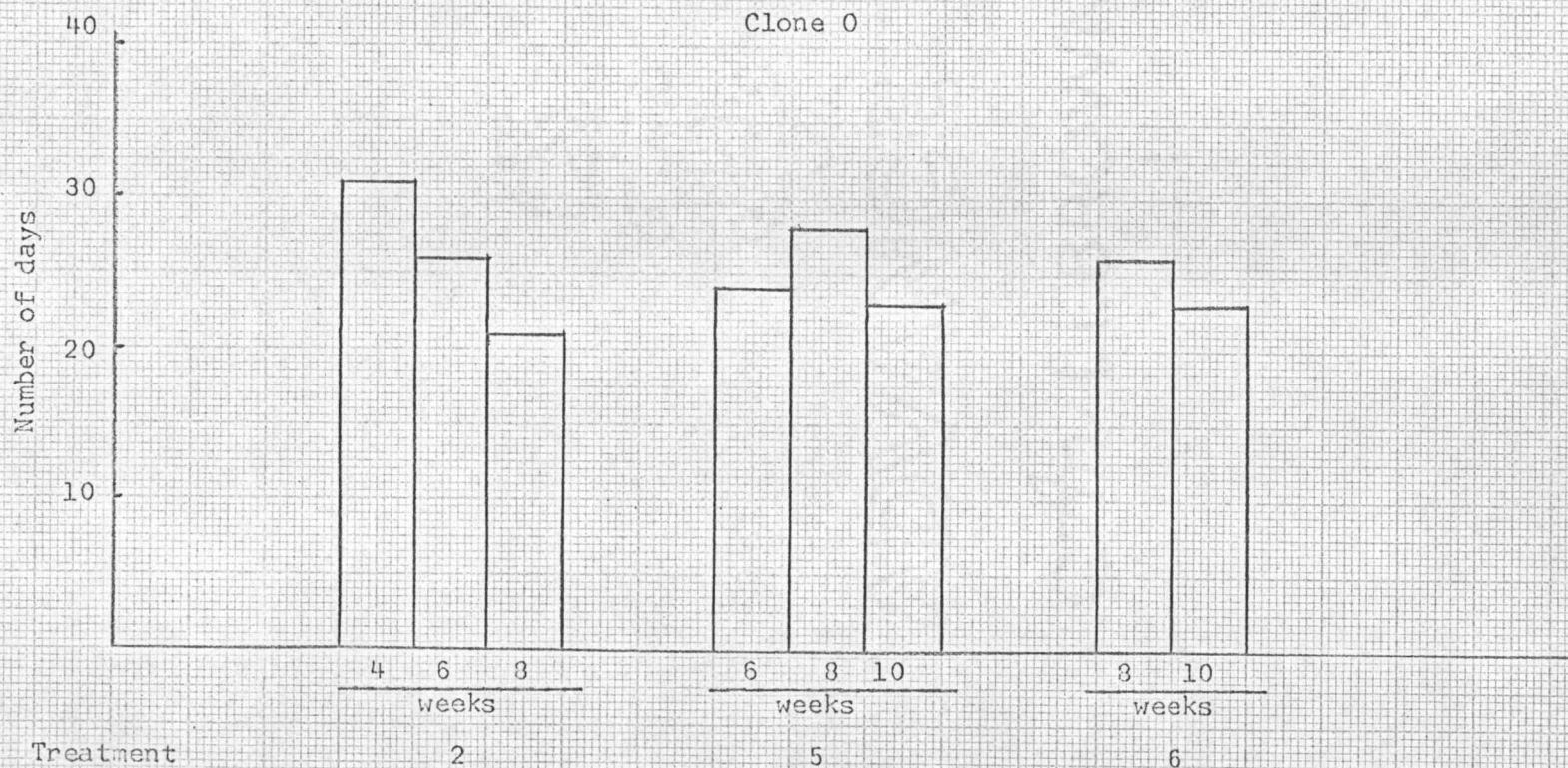


Figure 8. Number of days required for first panicle exertion after removal from various temperature treatments and durations

Treatments: 2=24 hrs. 38°F  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change



exhibited earlier floral exsertion in certain treatments (1 and 6 for clone R, 5 for clone O) than did the eight-week exposure treatment (Figures 7 and 8).

The first visible sign of anthesis generally followed panicle exsertion in a relatively similar amount of time for all treatments (see Figures 9 and 10). Like panicle exsertion, anthesis time was generally inversely related to cold temperature exposure durations. Small deviations were noted in treatments 1 and 6 for clone R, where six weeks of cyclic temperature levels showed earlier signs of anthesis than did eight weeks at the same temperature levels.

#### C. Vegetative and reproductive tiller development

Data presented in Tables 5 and 6 compare the reproductive tillers developed per plant with those that remained vegetative under the various temperature treatments and durations. Clone R always had a larger number of fertile tillers per plant than did clone O under the same temperature treatment conditions. The number of fertile tillers per plant was generally directly related to an increase in duration of exposure (Table 5). The longer exposure periods in treatments 2, 5 and 6 were particularly effective in increasing the number of fertile tillers of clone O. A similar positive linear trend held true for the length of treatment exposure and the number of fertile tillers developed in clone R. However, some six-week temperature treatments (treatment 1 and 5) were higher in fertile tiller number per plant than were the comparable eight-week treatments. Overall, treatments 2 and 5 appeared to be



Table 5. Average number of vegetative and reproductive tillers developed per plant under various temperature treatments

		Treatment Number											
		1	2	3		4		5		6			
		Temperature Regimes											
		10 hrs. 38°F	24 hrs. 38°F	15 hrs. 38°F	5 hrs. 70°F	10 hrs. 38°F	10 hrs. 90°F	15 hrs. 38°F	5 hrs. 60°F	20 hrs. 38°F	to peak 65°F		
		+4 hrs.change	38°F	+4 hrs.change	+4 hrs.change	+4 hrs.change	+4 hrs.change	+4 hrs.change	+4 hrs.change	+4 hrs.change	+4 hrs.change	+4 hrs.change	+4 hrs.change
Tillers per plant	Exposure time in weeks	Fertile tillers and total tillers per plant											
		Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O
Total Fertile	10							21.7	18.7	23.0	13.3		
								8.3	4.2	6.2	1.3		
Total Fertile	3	15.6	13.7	10.8	8.9	20.1	18.7	22.0	11.0	21.0	12.3	15.3	14.7
		4.8	.1	7.6	1.8	3.9	0	0	0	6.1	2.3	3.1	.7
Total Fertile	6	13.8	11.1	11.8	12.7	17.7	12.3	11.0	17.0	20.7	11.3	17.7	14.3
		6.3	0	6.7	1.5	3.4	.1	0	0	6.7	2.8	3.2	.1
Total Fertile	4	12.2	11.3	8.8	8.5	15.3	9.0	11.3	9.3	11.7	17.0	12.3	8.7
		2.4	0	1.6	.1	0	0	0	0	2.5	0	.6	0
Total Fertile	2	10.9	9.3	8.0	6.8	7.0	9.3	9.2	7.3				
		.2	0	0	0	.1	0	0	0				

Table 6. Percentage of total tillers per plant which became floral after exposure to various temperature treatments.

	Treatment Number											
	1		2		3		4		5		6	
	Temperature Regimes											
	10 hrs. 38°F		24 hrs. 38°F		15 hrs. 38°F		10 hrs. 38°F		15 hrs. 38°F		20 hrs. 38°F	
	10 hrs. 70°F				5 hrs. 70°F		10 hrs. 90°F		5 hrs. 60°F		to peak 65°F	
	<u>+4 hrs.change</u>		<u>38°F</u>		<u>+4 hrs.change</u>		<u>+4 hrs.change</u>		<u>+4 hrs.change</u>		<u>+4 hrs.change</u>	
Exposure time in weeks	Percent tillers induced											
	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O	Clone R	Clone O
10									36	22	27	10
8	31	.7	70	20	19	0	0	0	29	19	20	5
6	46	0	57	12	19	.8	0	0	32	25	18	.7
4	20	0	18	1	0	0	0	0	21	0	5	0
2	.9	0	0	0	1	0	0	0				

Figure 9. Number of days required for first visible sign of anthesis after removal from various temperature treatments and durations

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change      5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 2=24 hrs. 38°F      6=20 hrs. 38°F to peak 65°F+4 hrs. change  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change

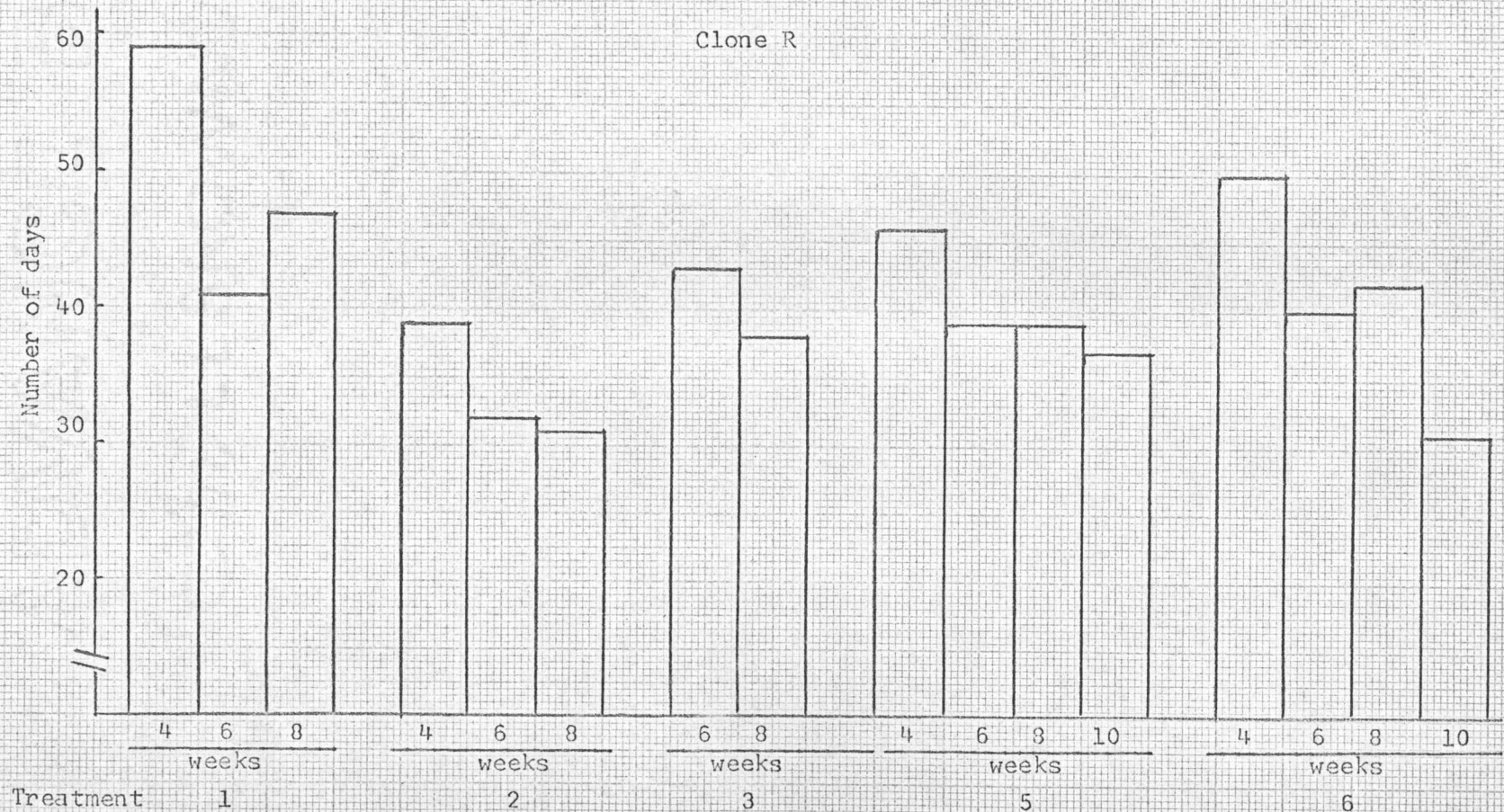
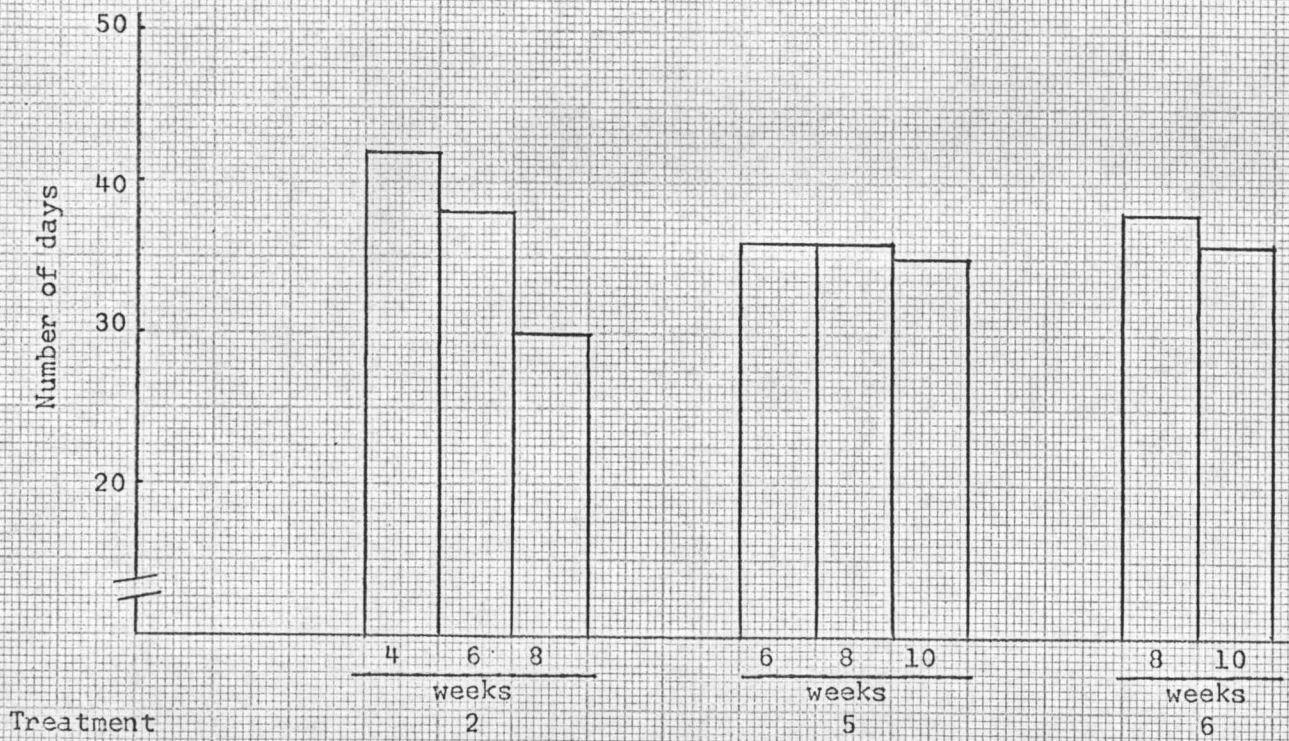


Figure 10. Number of days required for first visible sign of anthesis after removal from various temperature treatments and durations

Treatments: 2=24 hrs. 38°F  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

Clone 0



most promotive in the number of fertile tillers developed per plant (Table 5).

The percent fertile tillers developed of the total tillers per plant was calculated and the results summarized in Table 6.

With eight weeks at continuous 38<sup>o</sup>F temperatures, clone R produced the highest percentage of floral tillers of any temperature treatment period. However, this may have been due to the small total number of tillers produced per plant (Table 6). The percentage of reproductive tillers per plant usually increased with each two-week increase in exposure to a certain treatment. The six-week exposure of treatment 1 and 5 for clone R and the 10-week exposure to treatment 5 for clone 0 was superior in percentage of reproductive tillers. Treatments 3 and 6 were less effective than treatments 1, 2 and 5 in the ratio of vegetative to reproductive apices.

## 2. Vegetative Development

### A. Tiller development

Tillering during the various treatments generally showed a positive linear trend with duration of treatment exposure. Treatment 3, with 15 hours at 38<sup>o</sup>F and 5 hours at 70<sup>o</sup>F temperature (Figures 11 and 12), appeared to be most conducive to tiller development for both clone R and clone 0. The slowest tiller development for both clones was found with a continuous 38<sup>o</sup>F temperature regime.

Figure 11. Average number of tillers gained per plant during exposure to various temperature treatments and durations

Clone R  
 Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 4=10 hrs. 38°F, 10 hrs. 90°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

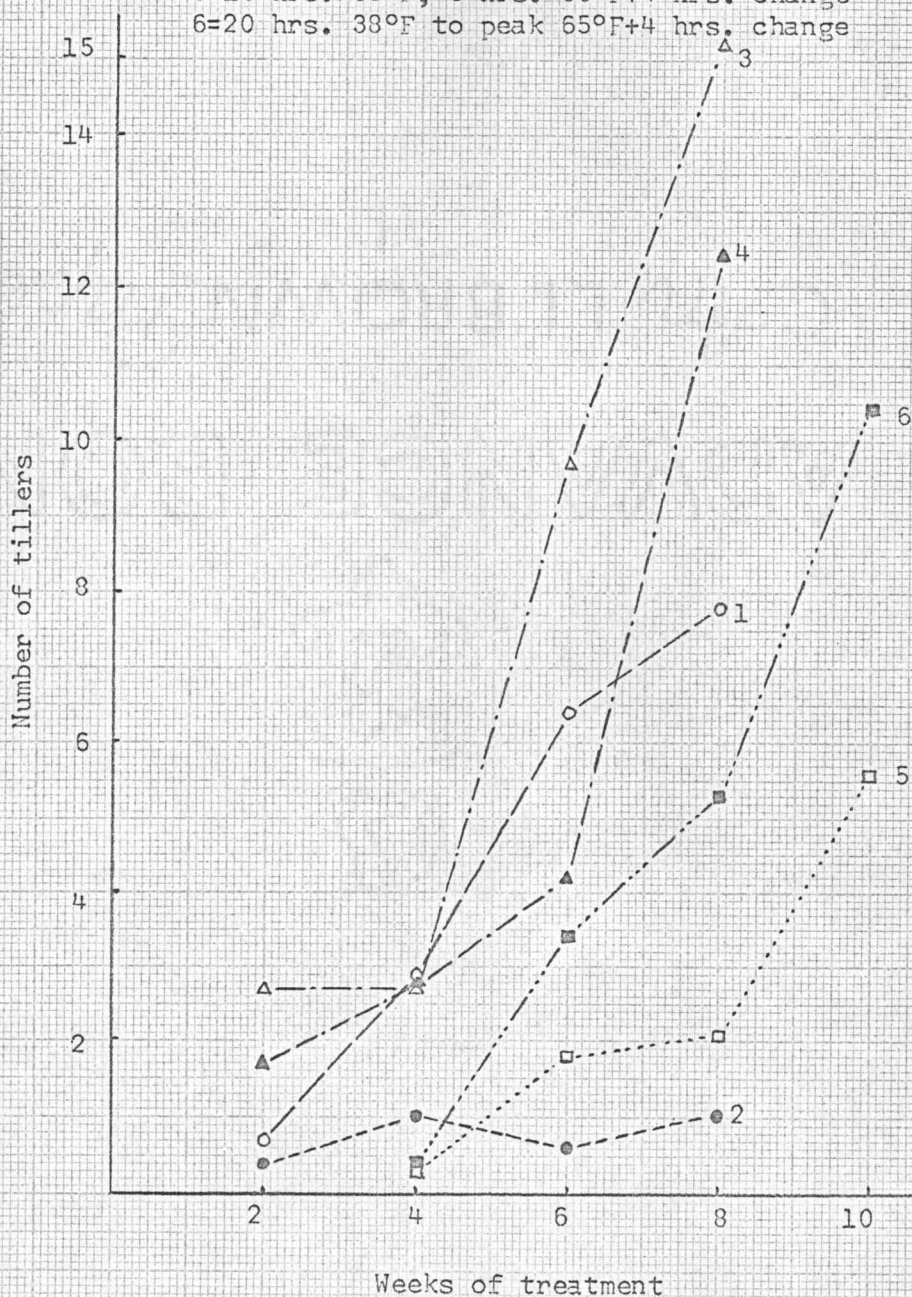
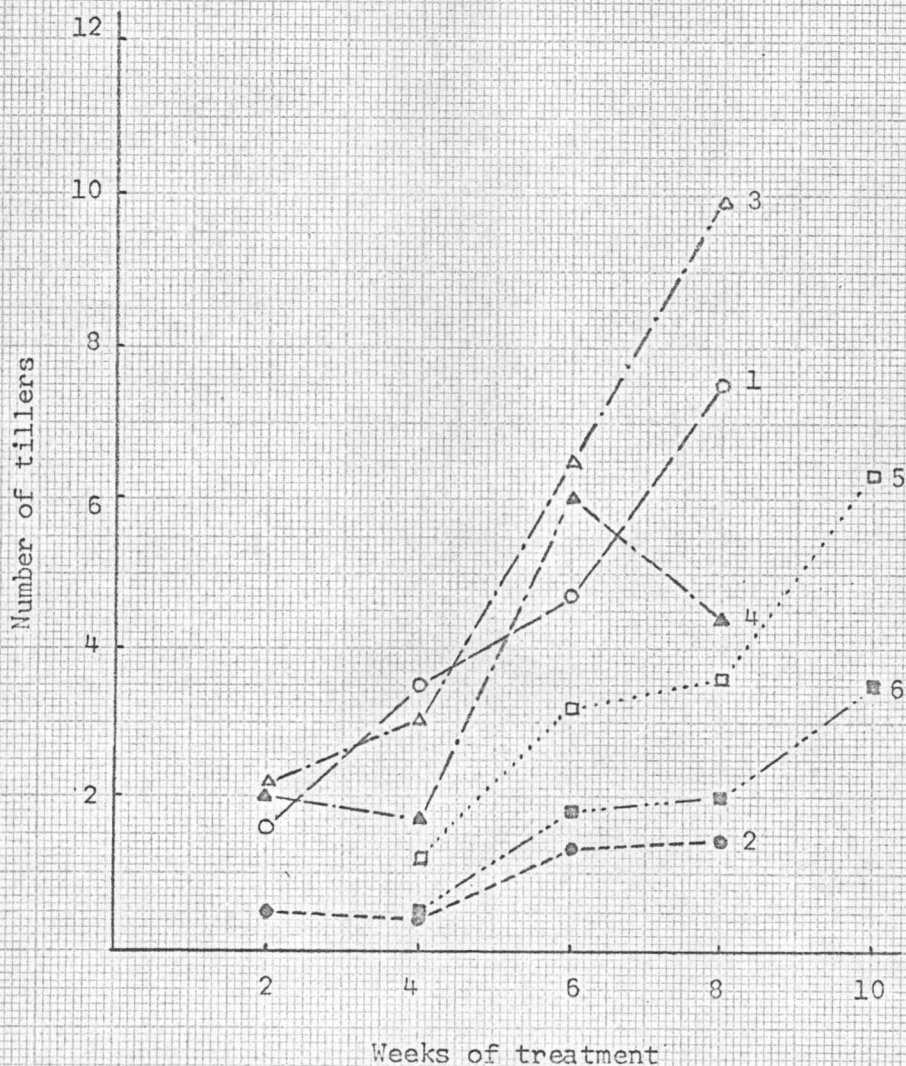


Figure 12. Average number of tillers gained per plant during exposure to various temperature treatments and durations

Clone 0

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 4=10 hrs. 38°F, 10 hrs. 90°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change



A comparison between treatments 2 and 6 showed that a peak of 65°F with 20 hours of 38°F temperatures encouraged tiller development above that of a continuous 38°F temperature regime (Figures 11 and 12). Alternating temperatures of 10 hours at 38°F and 10 hours at 70°F or 90°F were more favorable for tillering than were those temperature regimes with lower than 70°F maximum temperatures.

In clone O, the difference between treatment 2 (continuous 38°F) and treatment 6 (20 hours at 38°F and a peak of 65°F) were less pronounced than it was in clone R.

The total number of tillers gained by clone R and clone O during the treatment and during the floral initiation and development period are presented in Figures 13 and 14 respectively. The overall trend was generally a direct linear relationship between number of tillers gained and exposure time to various temperature treatments. Interesting exceptions to this linear trend are treatments 2, 4 and 5 for clone O, which showed irregular tiller development, particularly for the longest temperature durations.

In some instances, the six-week exposure period produced a larger number of tillers overall than did the eight-week treatment, as evidenced by treatments 2 and 6 for clone R (Figure 13) and treatments 2 and 4 for clone O (Figure 14).

#### B. Leaf development

The number of leaves gained during different temperature treatments and durations are shown in Figure 15 and 16 for clone R



Figure 13. Average number of tillers gained per plant during and after exposure to various temperature treatments and durations

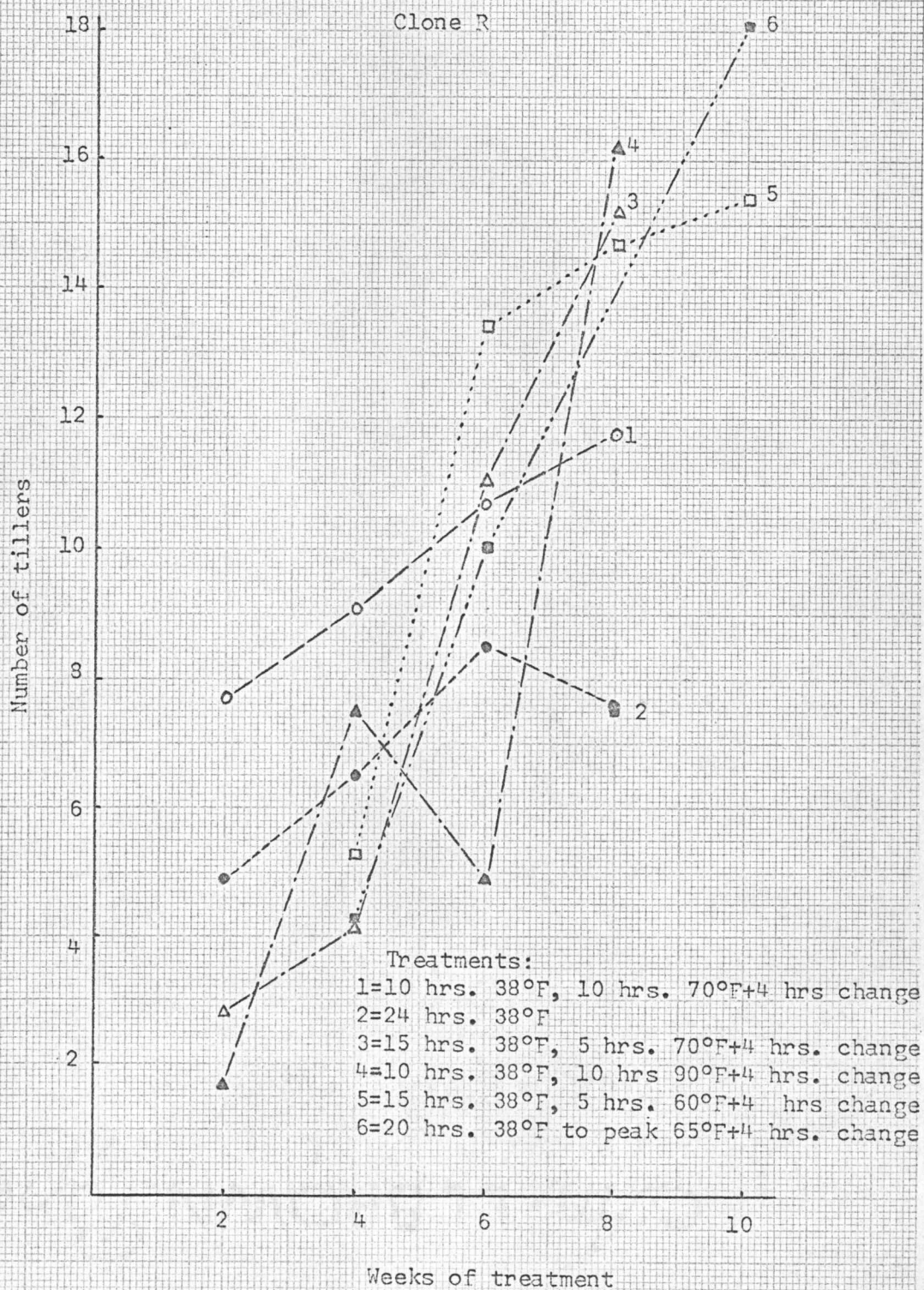
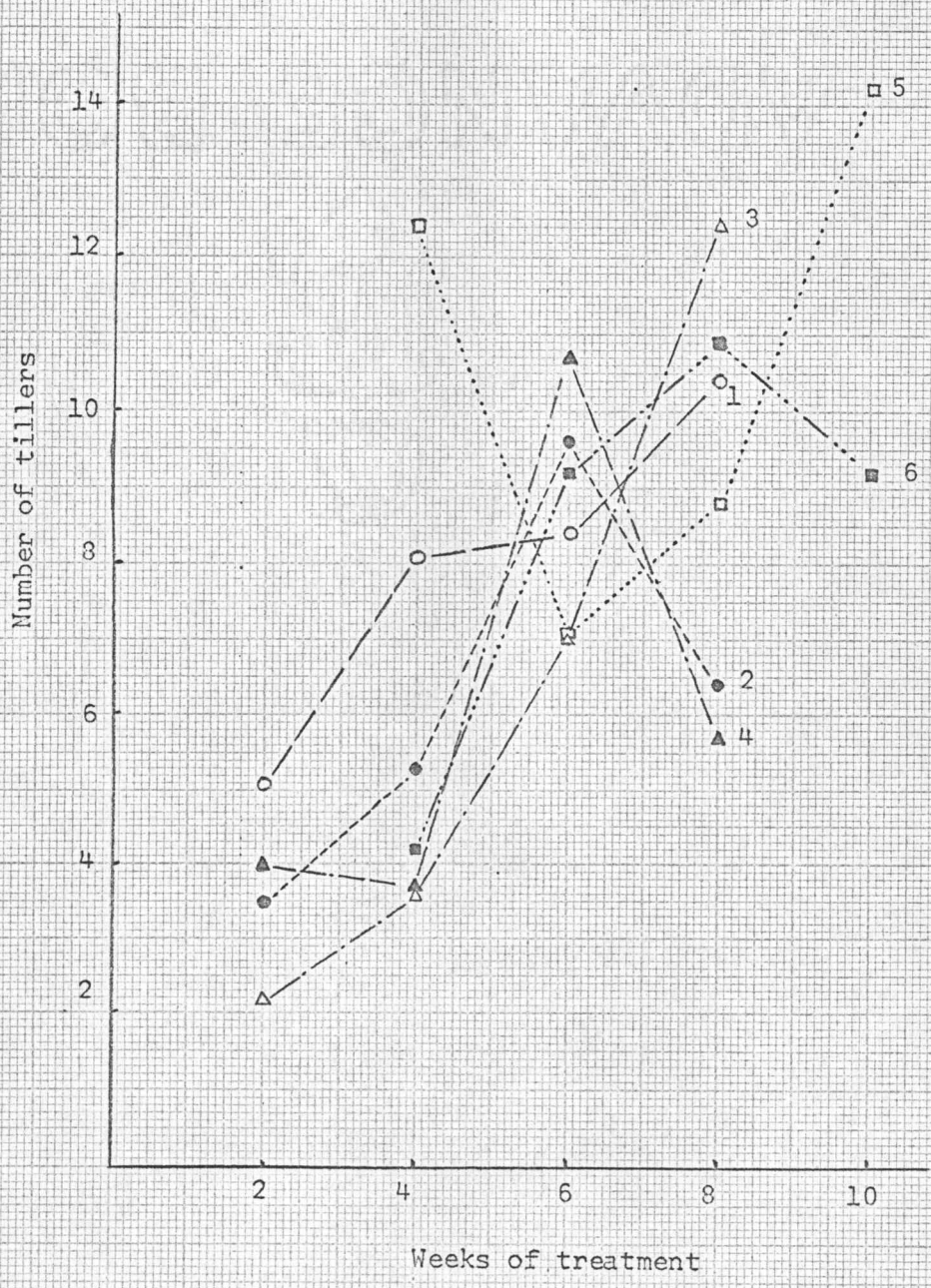


Figure 14. Average number of tillers gained per plant during and after exposure to various temperature treatments and durations

Clone 0

- Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change
- 2=24 hrs. 38°F
- 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change
- 4=10 hrs. 38°F, 10 hrs. 90°F+4 hrs. change
- 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change
- 6=20 hrs. 38°F to peak 65°F+4 hrs. change



and 0 respectively.

Treatment 4, with 10 hours of 38°F alternating with 10 hours of 90°F temperatures, was one of the most favorable treatments for leaf development in either clone, particularly for the eight weeks treatment. Next to treatment 4, treatments 1 and 3 with 10 and 5 hours of 70°F temperatures respectively, showed the best leaf development for both clone R and 0 exposure periods.

Leaf development was slowest in continuous 38°F temperatures. However, it was rather surprising that tillers with six weeks of treatment gained more leaves than those tillers subjected to eight weeks of treatment. The peak of 65°F and 4 hours change to and from 38°F temperatures in treatment 6 was apparently enough to encourage leaf development, after eight weeks exposure, to levels beyond those found in treatment 2 (continuous 38°F temperatures).

Figures 17 and 18 show the relationship between weeks of treatment exposure and the total number of leaves gained on the main tiller, beginning at the time when plants were subjected to various treatments up to the time when all leaves on the reproductive culm were visible. In certain instances, where the main tiller was still vegetative, leaf development of the non-reproductive primordia was recorded, thus indicating the number of leaves gained by the vegetative primordia. The two-week exposure periods of treatments 1 to 4 involved strictly vegetative tillers. Four-week exposure periods of treatments 3, 5 and 6 were often non-inductive for clone R with only some tillers developing an inflorescence (Table 4).

Figure 15. Average number of leaves gained per tiller during exposure to various temperature treatments and durations

Clone R

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 4=10 hrs. 38°F, 10 hrs. 90°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

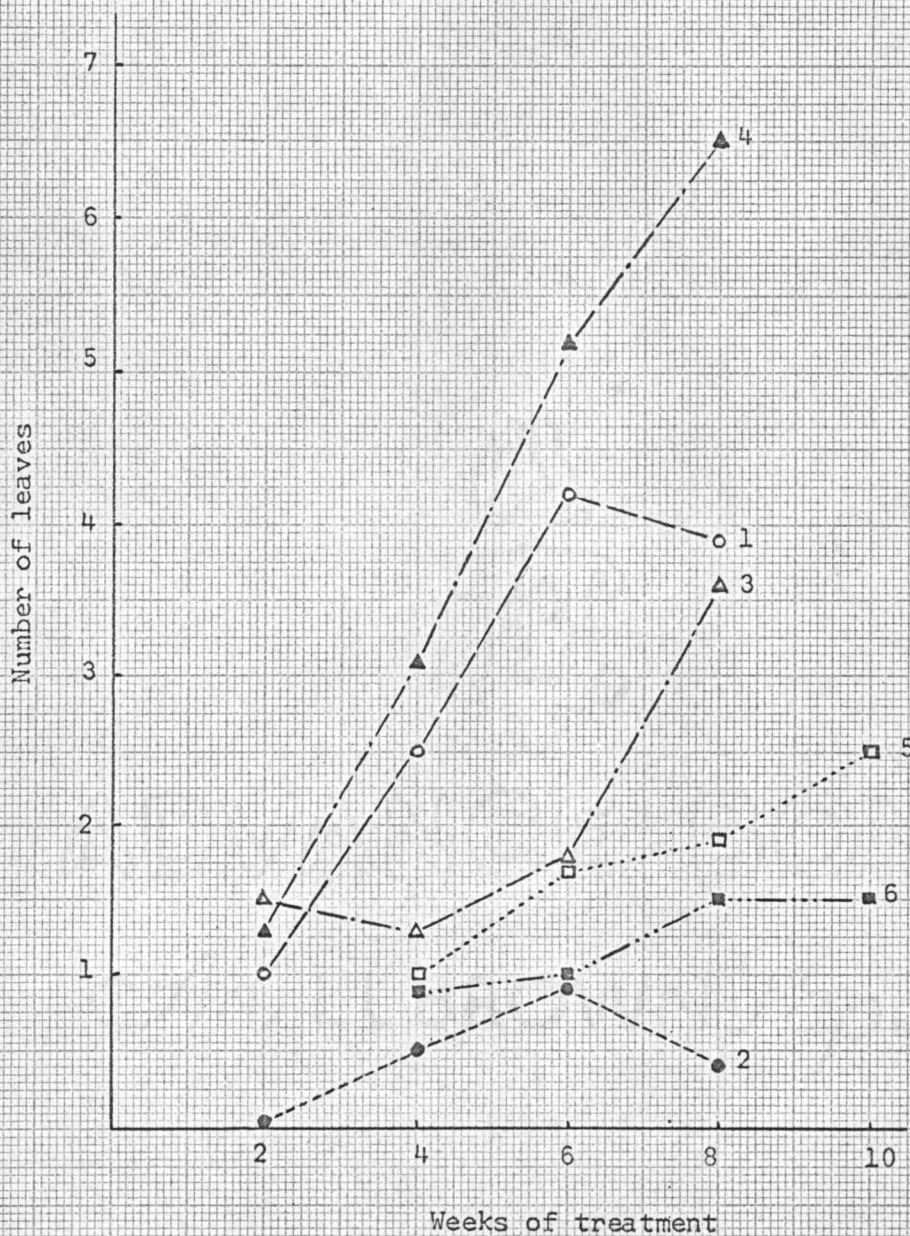


Figure 16. Average number of leaves gained per tiller during exposure to various temperature treatments and durations

Clone 0

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 33°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 4=10 hrs. 38°F, 10 hrs. 90°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

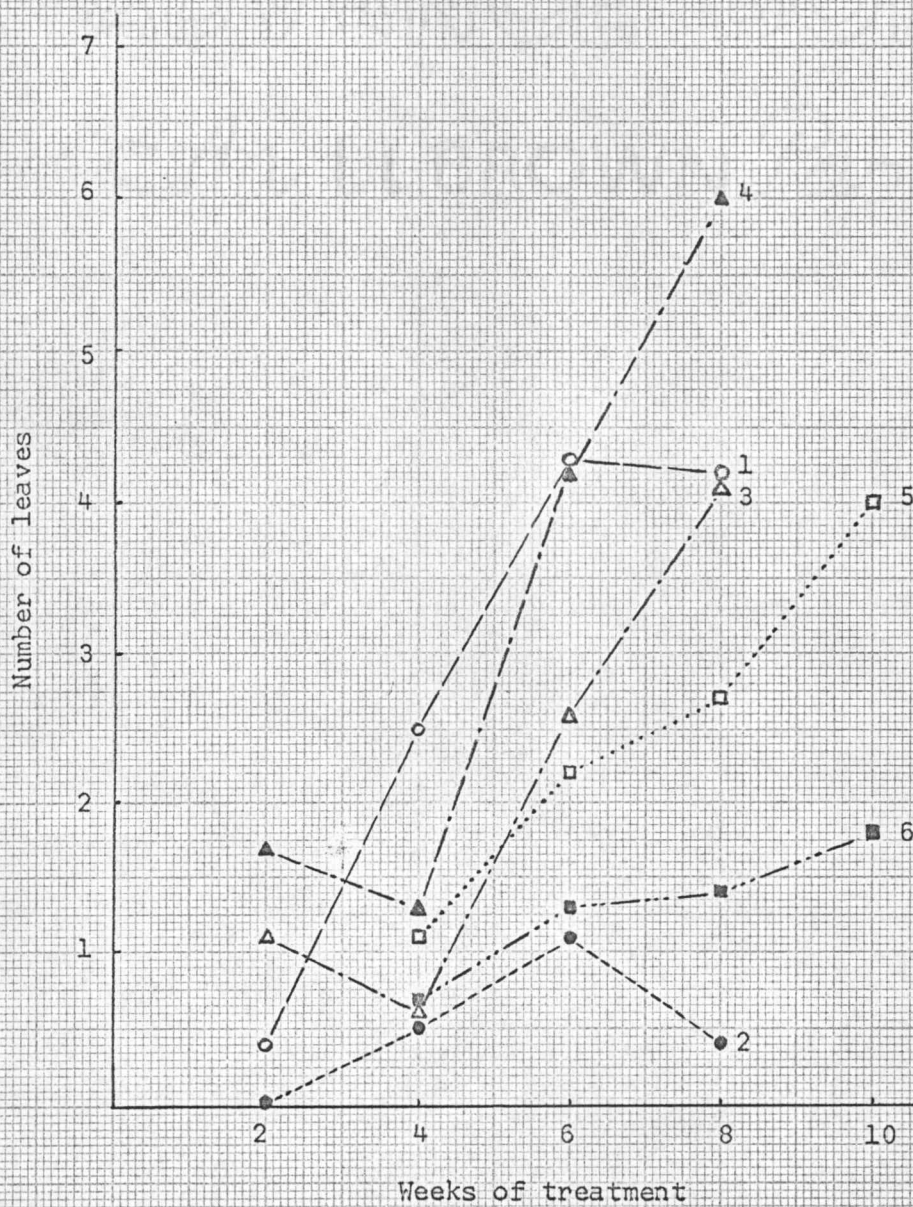


Figure 17. Average number of leaves gained per tiller during and after exposure to various temperature treatments and durations

Clone R

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 4=10 hrs. 38°F, 10 hrs. 90°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change

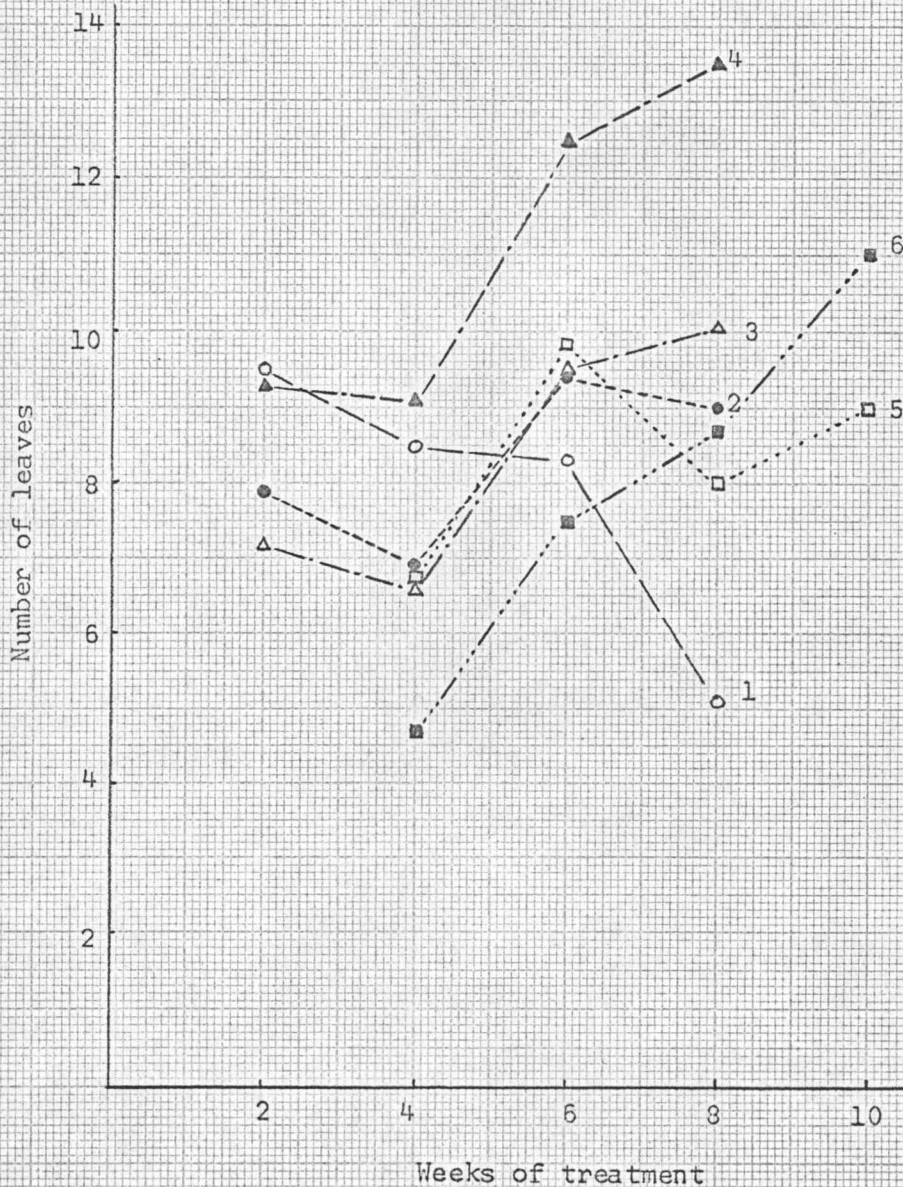
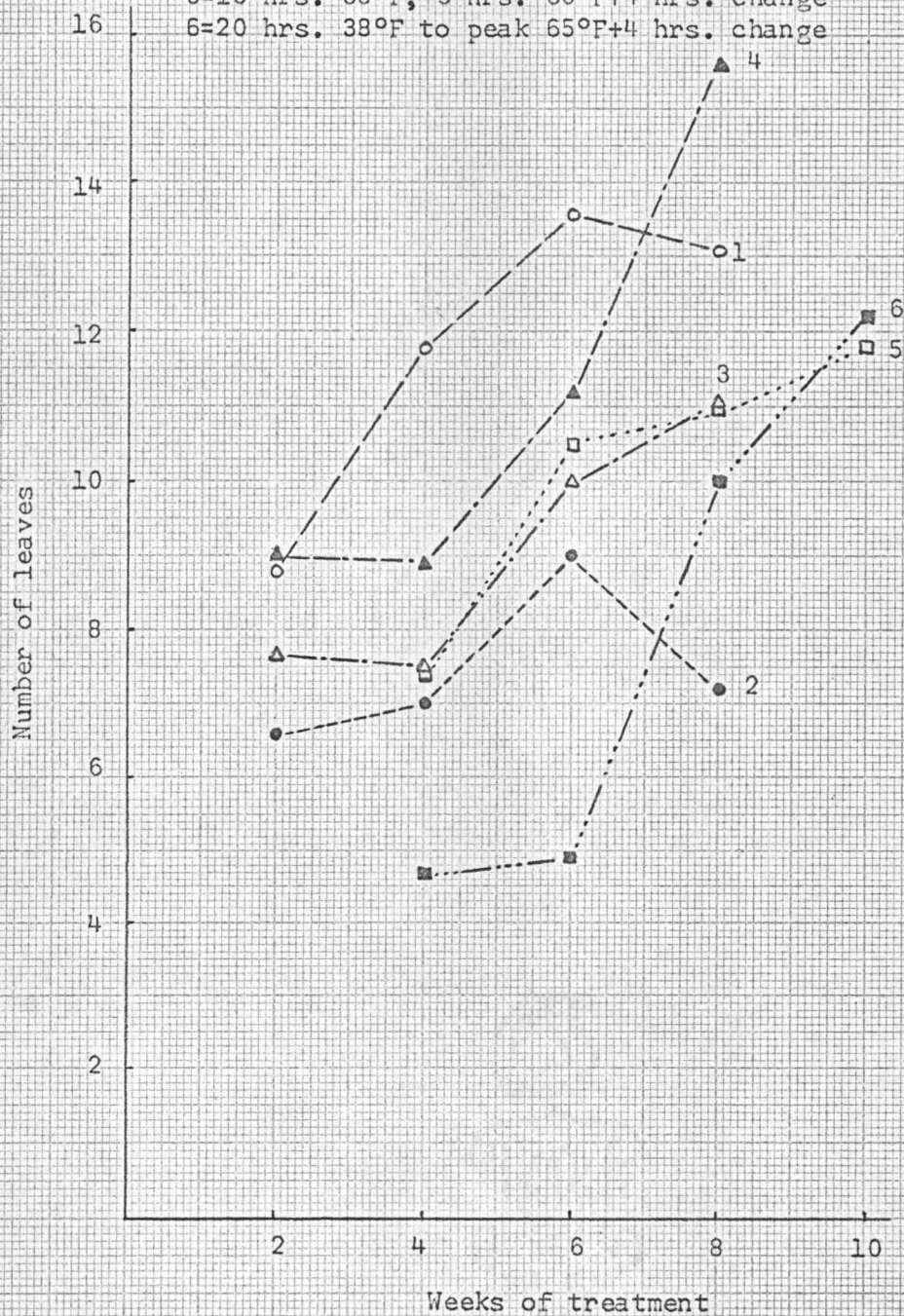


Figure 18. Average number of leaves gained per tiller during and after exposure to various temperature treatments and durations

Clone 0

Treatments: 1=10 hrs. 38°F, 10 hrs. 70°F+4 hrs. change  
 2=24 hrs. 38°F  
 3=15 hrs. 38°F, 5 hrs. 70°F+4 hrs. change  
 4=10 hrs. 38°F, 10 hrs. 90°F+4 hrs. change  
 5=15 hrs. 38°F, 5 hrs. 60°F+4 hrs. change  
 6=20 hrs. 38°F to peak 65°F+4 hrs. change



Treatment 4, which was 10 hours of 38°F and 10 hours of 90°F temperatures on a daily basis, was generally superior to the other regimes in terms of number of leaves formed. Treatment 1, with 10 hours at 38°F and 10 hours at 70°F temperature, was promotive to leaf formation in clone O but not in clone R. Most of the treatments showed a trend for increased leaf number with an increase in duration of the various temperature treatments.

The number of leaves developed on clone O generally showed a direct relation to the duration of exposure (Figure 18) with the exceptions of treatments 1 and 2, where a slight decline in the number of leaves was noted between the six- and eight-week exposure durations. In general, the warmer the cycle temperatures (treatment 4, 1 and 3), the faster the leaf development during and after treatment exposure.

## Part II. Field Experiment

### 1. Floral Induction

#### A. Floral initiation and panicle exertion

In order to obtain a better understanding of floral induction under field conditions, propagules were planted at various dates during the fall and winter. A graphical illustration of the total number of initiations, including exerted and non-exserted inflorescences, is shown in Figure 19 for each planting date and each clone. A comparison between clone R and clone O points out the different floral induction responses of these two clones. Clone O responded



to an increased period of exposure to winter conditions with a larger number of reproductive primordia. A breaking point was found for clone 0 (Figure 19) between planting treatments 3 and 5 where there was a rapid decline in the total number of initiations.

To evaluate the temperature regime in the field, daily temperatures were averaged over three-hour periods and summed within two-degree categories for the two-week intervals, beginning on November 22, 1963. The degree-hour temperatures for each treatment period, as well as five additional weeks after the first planting date, are found in Table 7.

The decrease in total number of floral primordia in clone 0 for the second planting date (Figure 19) could perhaps be explained by the extended period of sub-freezing temperatures (Table 7), which may have been injurious to the clone 0 apices.

Clone R (Figure 19) also showed an increased number of total floral initiations with an increase in field exposure time. These differences, however, were less pronounced than the differences found in clone 0.

Figure 20 shows the relationship between date of planting and the total number of floral primordia which failed to exert from the boot in plants of clone R and 0. Clone 0 generally had larger numbers of non-exserted inflorescences than did clone R. The earlier the planting date of clone 0, the larger the number of non-exserted inflorescences (Figure 20). An interesting deviation from the linear trend of initiated but not exerted primordia is found in treatments 4 and 7 for clone 0 and treatment 7 for clone R.

Table 7. Accumulated degree-hours in two degree categories calculated by averaging the temperatures over three-hour intervals for two-week field exposure periods measured at one-inch soil depth.

Treat ment no.	Dates in two-week intervals	Temperatures in degree Fahrenheit																						
		68	66	64	62	60	58	56	54	52	50	48	46	44	42	40	38	36	34	32	30	28		
		<u>Hours</u>																						
1	Nov. 22- Dec. 5									3	3	6	39	69	18	27	30	24	63	51				
2	Dec. 6- Dec. 19											3	3	6	30	18	51	48	30	54	60	21	12	
3	Dec. 20- Jan. 2										3	12	24	72	63	48	45	24	9	36				
4	Jan. 3- Jan. 16											6	3	18	48	99	42	42	63	15				
5	Jan. 17- Jan. 30										9	6	15	42	66	30	66	30	42	30				
6	Jan. 31- Feb. 13												18	27	27	12	48	42	48	60	54			
7	Feb. 14- Feb. 27									9	15	6	18	12	33	39	18	42	51	27	30	30	6	
8	Feb. 28- Mar. 12												9	9	21	12	36	45	24	63	57	18	27	15
	Mar. 13- Mar. 26						3	6	9	27	6	27	24	39	30	72	36	33	21	6				
	Mar. 27- Apr. 9																							
	Apr. 10- Apr. 15		6	18	12	6	9	24	39	21	21	27	33	60	27	24	3	3	3					
		3	9	0	3	3	6	12	15	30	3	3	18	12	27	9	9	6						

Figure 19. Total number of initiations for 6 plants including exerted and non-exserted floral primordia for various planting dates of Clone R and O

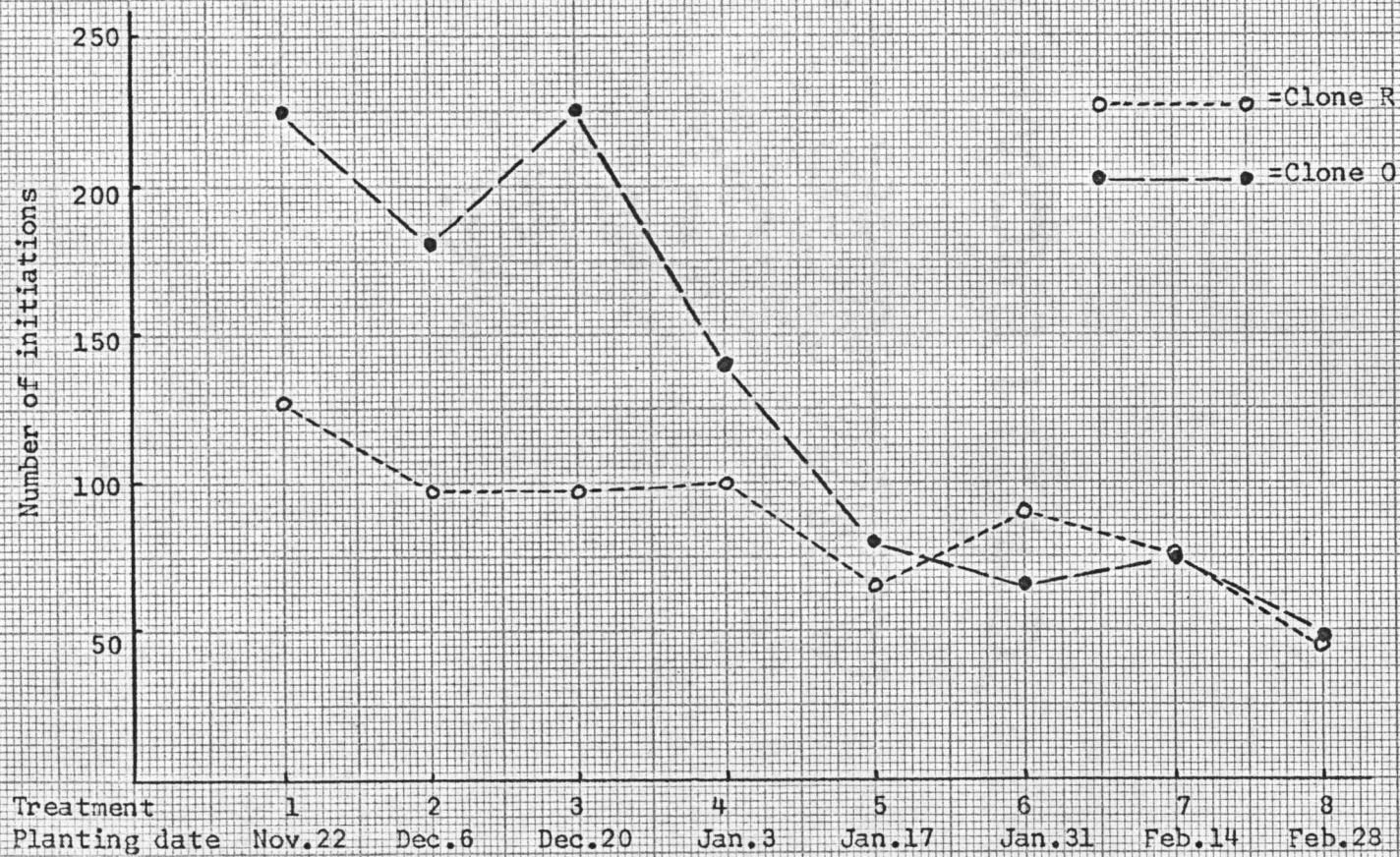
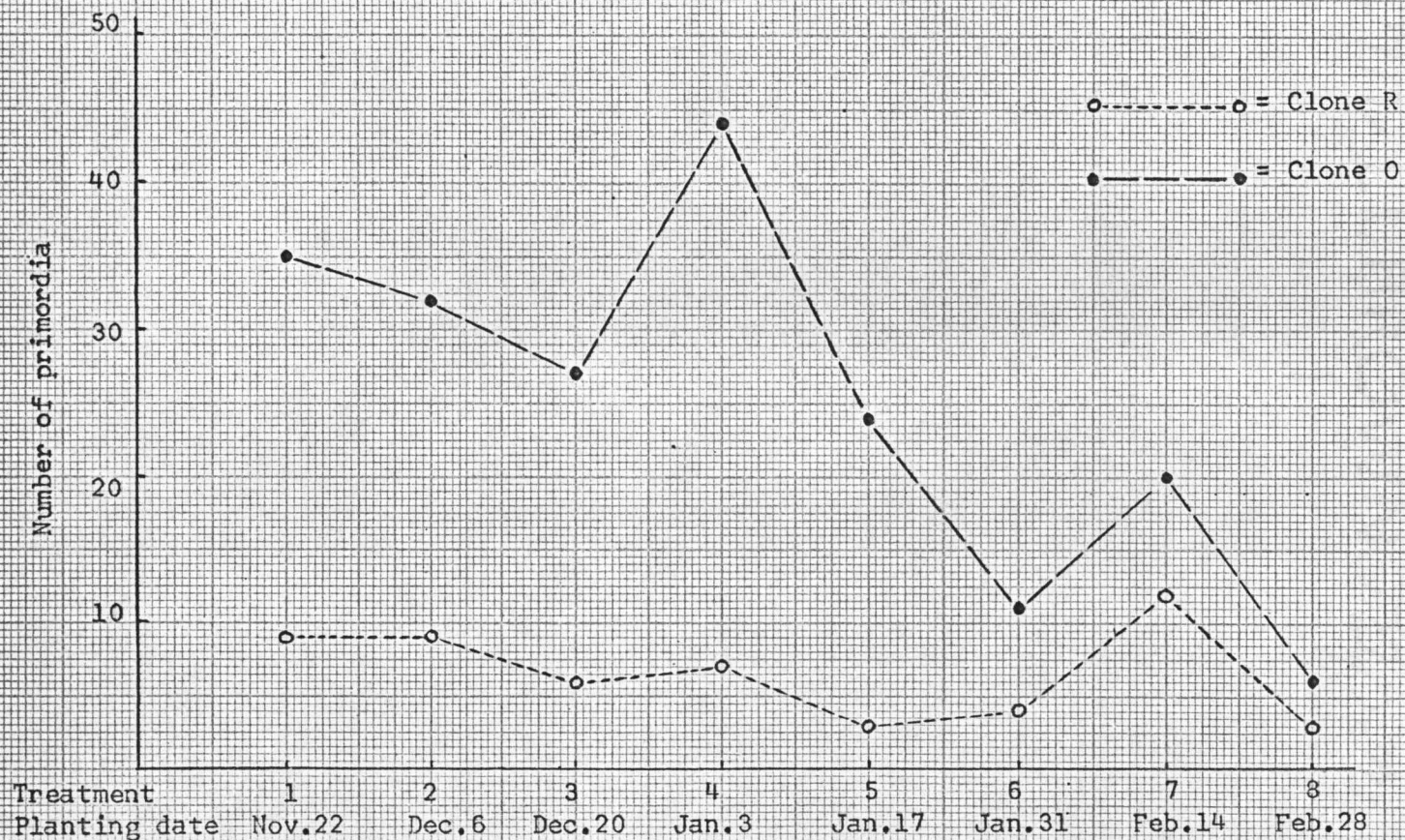


Figure 20. Total number of initiated but non-exserted floral primordia for 6 plants for various planting dates of Clone R and Clone O



A possible explanation for the higher than expected number of non-exserted inflorescences is that the apices concerned suffered some injury, therefore failing to exert inflorescence. Such injury could have been brought about by freezing temperatures in treatments 4 and 7 with 15 and 36 hours of 30° to 32°F temperatures respectively (Table 7).

#### B. Rate of inflorescence development

The number of exerted inflorescences were counted at weekly intervals for each clone and for each planting date (Figure 21 and 22). Plants transferred to the field on November 22, December 6 and December 20 were the first to exert an inflorescence. Clone O started to show inflorescences one week earlier than did clone R. Panicle exertion was most rapid during the first three weeks after appearance of the first panicles, and then started to decline.

Three of the eight plantings of clone R had exerted an inflorescence on May 25. On June 1, seven of the eight plantings exhibited panicles, and by June 8 the last planting showed exerted panicles. The most rapid inflorescence exertion for clone R was by plants transferred to the field on the first date of planting. Also, the total number of inflorescences was markedly higher for clone R propagules planted on the first date. In general, the numbers of exerted panicles decreased with later planting dates.

The floral exertion pattern for clone O was somewhat similar to that of clone R, except that panicle exertion started earlier and a higher number of visible inflorescences developed. Plants

Figure 21. Total number of exerted inflorescences for 6 plants observed at weekly intervals for various planting dates

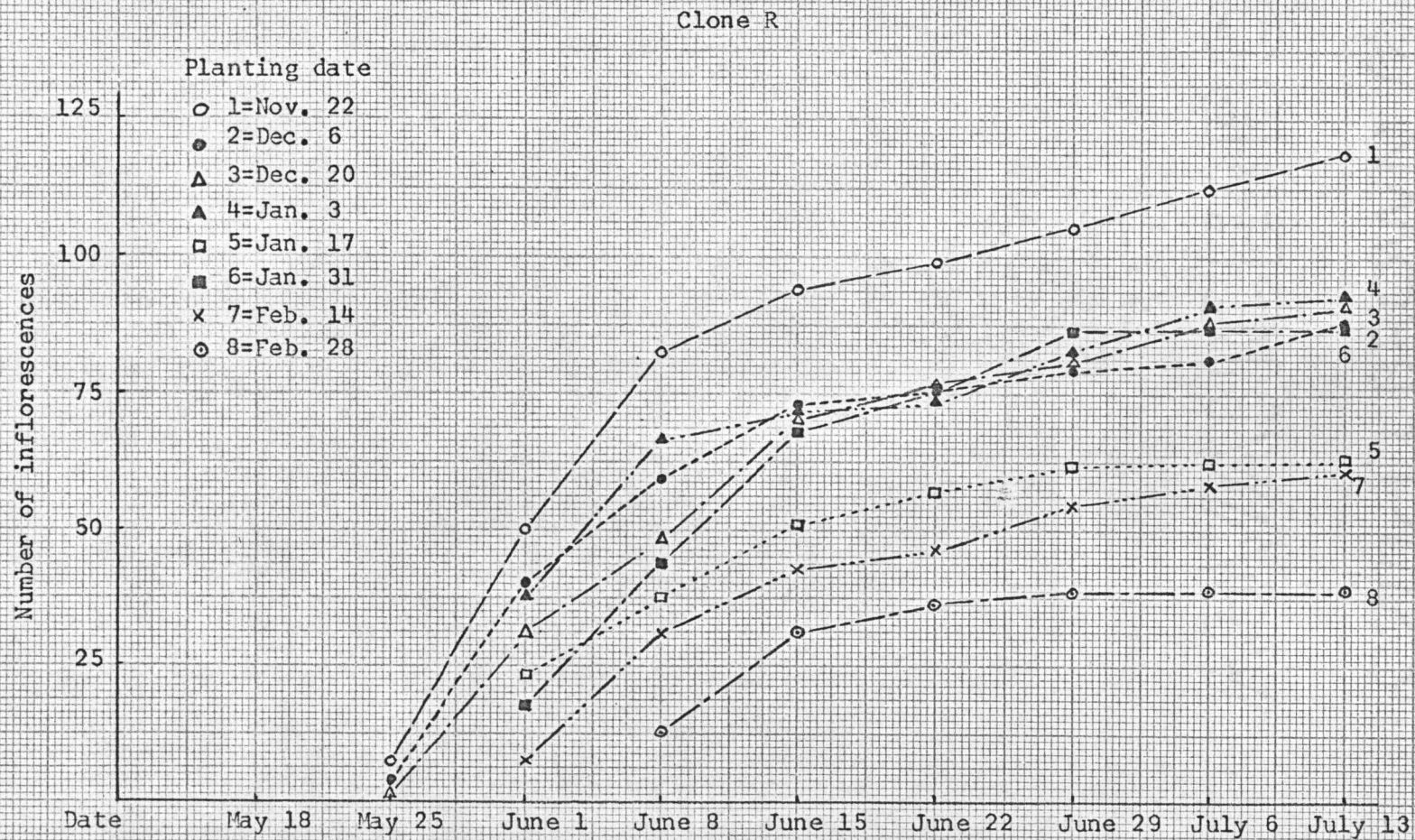
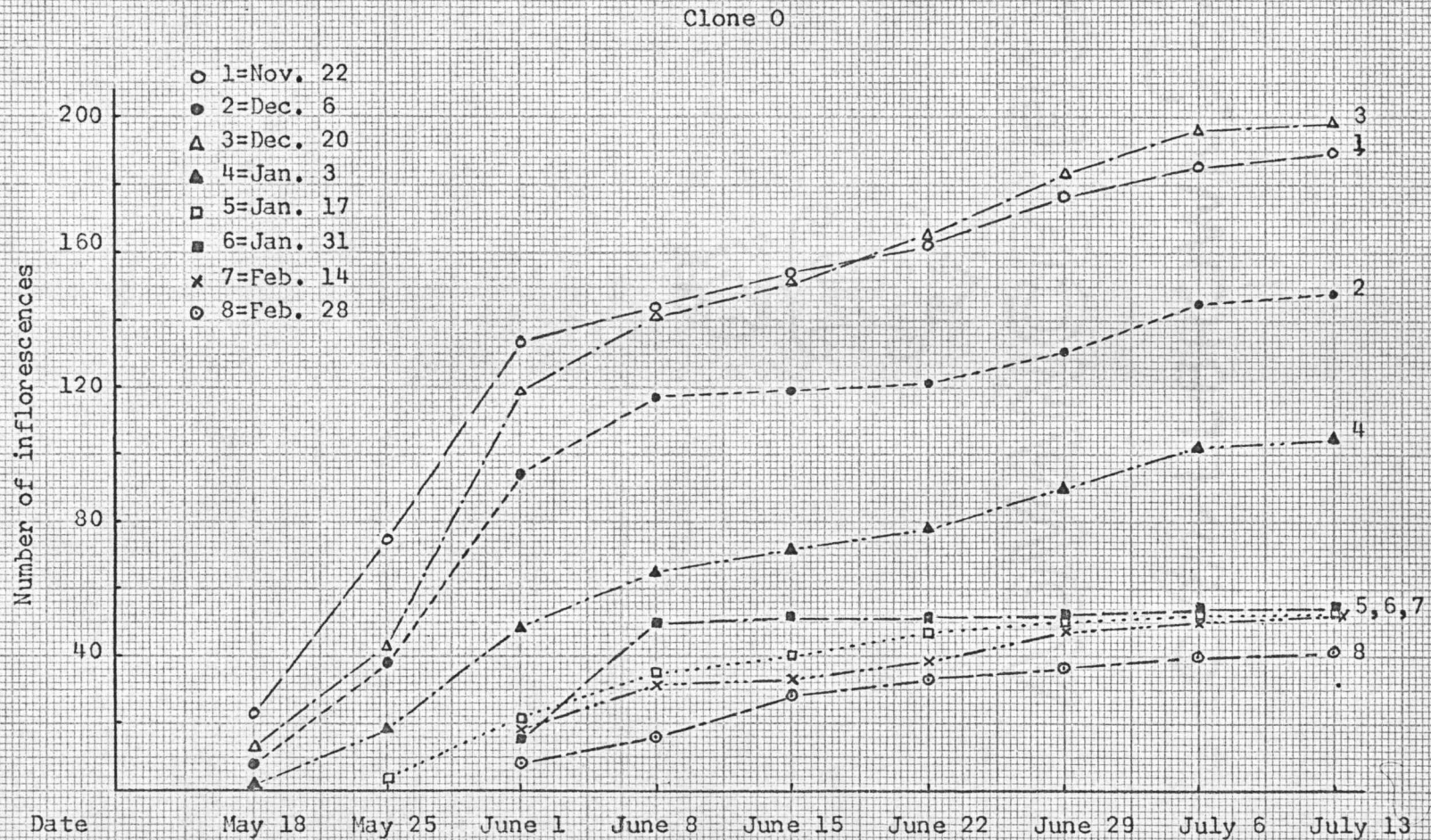


Figure 22. Total number of exerted inflorescences for 6 plants observed at weekly intervals for various planting dates



transferred to the field on dates 1 to 4 had exerted inflorescences on May 18, plants of treatments 5 and 7 on May 25, and treatments 6 and 8 on June 1. A rather noticeable difference in floral exertion time was found between the first four and the second four planting dates. Inflorescences developed most rapidly for plants transferred to the field prior to January 17.

## 2. Tiller Development

Figure 23 represents the tiller development per plant for various planting dates of clone R and clone O. Both clones showed a positive linear trend between earliness of planting time and number of tillers developed per plant. Tiller number differences between the earliest and latest planting date were more pronounced in clone O than they were in clone R (Figure 23).

The trend in total number of tillers per plant at various transplanting dates shown in Figure 23 follows a pattern similar to that for number of floral initiations.

The total number of tillers developed on plants placed in the field on the first three dates showed that clone O had about 20 more tillers per plant than did clone R for the same planting dates. These tiller number differences became smaller in the later planting dates until finally, with the last planting date, both clones produced an equal number of tillers.

The percentage of tillers per plant which showed floral development in the various planting treatments are found in Figure 24. The percentage of reproductive tillers for the various planting



Figure 23. Total number of tillers per plant developed for various planting dates of Clone R and O

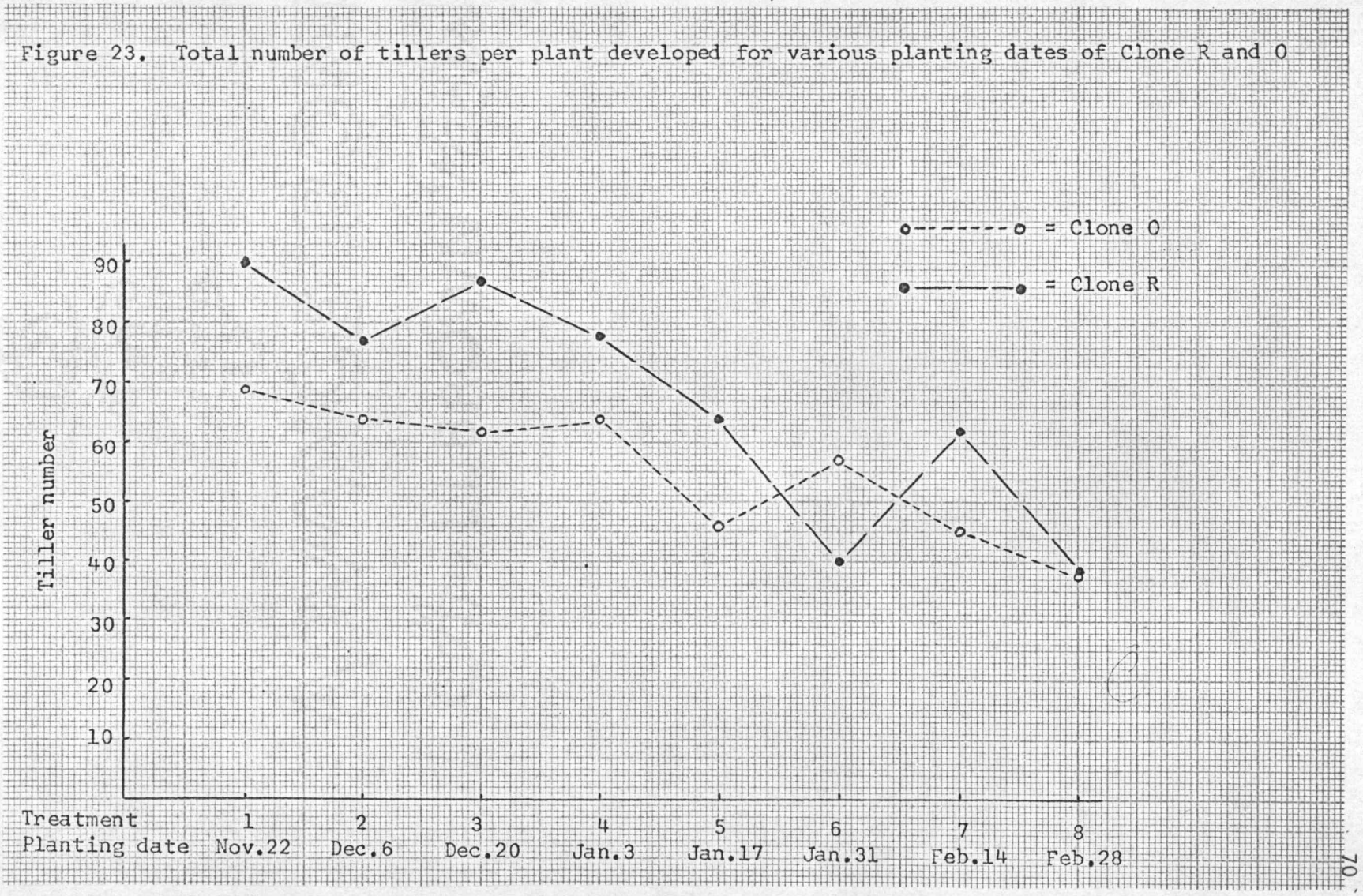
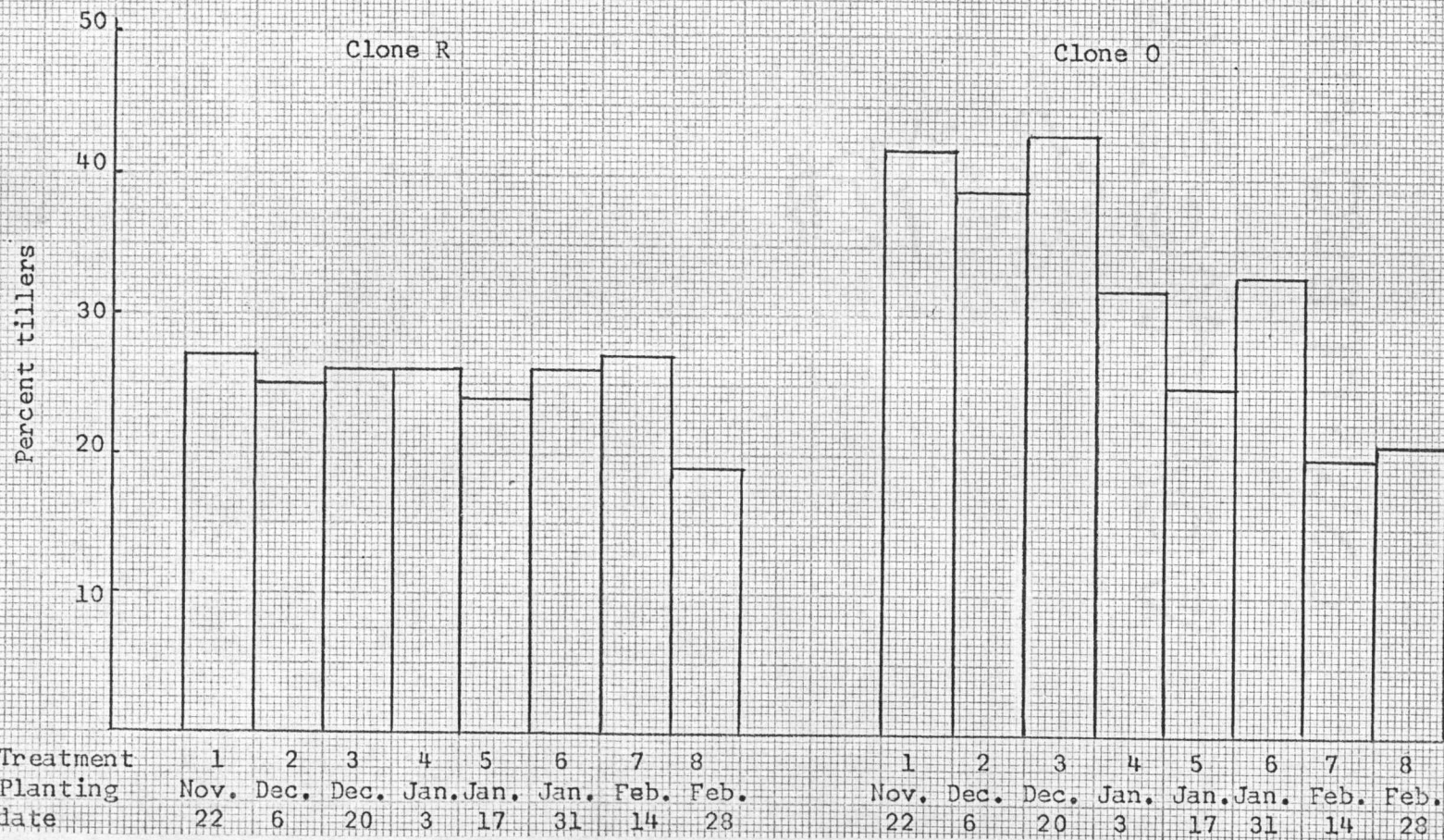


Figure 24. Percent tillers per plant which developed an inflorescence for various planting dates of Clone R and O



dates in clone R showed considerably less variability than those calculated for clone O (Figure 24). Reproductive tiller percentages per plant for clone R varied from 19 to 29 percent, whereas percentage figures in clone O ranged from 20 to 43 percent.

The percentage reproductive tillers per plant were similar for clone R plants of all planting dates, with the possible exception of the last planting date, when only 19 percent of the total number of tillers per plant became reproductive.

Clone O, on the other hand, appeared to produce higher percentages of fertile tillers with earlier planting dates (Figure 24). The unusually low total number of tillers developed per plant (Figure 23) may account for the high percent fertile tillers found in treatment 6 for clone O.

## DISCUSSION

Although clonal propagules of the selected genotype were used in all experiments and material was selected for each growth chamber and field treatment, differences in response of propagules resulted within treatments. The reasons for this variability are not entirely clear. Differences in response within a treatment may have been caused by gradients in soil fertility or moisture in individual lots. More important may have been the differential age or development of propagules following subdivision. One of the genotypes used in the trials was also affected to some degree by a disease which could have influenced floral induction and inflorescence development to some extent.

Duration of exposure to the various temperature regimes was an important factor in floral induction. Exposure periods of four to six weeks appeared to be the minimum duration to insure inflorescence exertion of clone R and clone O plants under favorable temperature treatments. Shorter time periods resulted in non-exserted panicles or tillers remained vegetative.

Temperatures of 70°F or higher failed to induce clone O tillers and reduced the number of inflorescences developed in clone R plants. Temperature regimes with highs of 60°F and 65°F were slightly more and slightly less promotive, respectively, than were continuous 38°F temperatures. The response at higher temperatures, especially 90°F temperatures, seemed to support the concept of a reversible reaction occurring similar to that during

vernalization (14, p. 216)

Cyclic diurnal temperature exposures with 5 hours of 60°F and 15 hours of 38°F with 4 hours of change, resulted in a higher number of floral tillers for clone 0 than where temperatures were maintained at constant 38°F. However, both the cyclic exposure treatment and 38°F constant temperatures were similar in the percentage of induced plants. Cyclic temperatures not exceeding 60°F appear to be important in increasing the number of fertile tillers per plant. The increased percentage of tillers found induced to flowering with increasing durations of low treatment agrees with the results published by Purvis (63, p. 186-206) for vernalization in winter rye.

The fact that clone R and 0 were florally induced in the field, even at the last field planting date (Figure 19, Table 7), suggests that interaction effects in the field may be important, since there were limited hours of 38°F temperatures and photoperiods exceeded 11 hours. Wycherley (76, p. 76-86) showed that earlier temperatures below 5°C (41°F) or alternating temperature cycles, were necessary for floral induction of orchardgrass. The increased range of alternating temperatures, particularly during the last planting dates, may provide a possible explanation for the relative success in floral induction despite increasing photoperiods in the field. However, interaction effects of photoperiod, light intensity, temperature levels and soil fertility in the field, should also be considered.

Longer treatment exposure periods to favorable temperature regimes in the growth chamber and field increased the number of inflorescences per plant for both clones. This is in agreement with results found by Chilcote (13, p. 59-67), who worked with two genotypes of orchardgrass. A larger number of tillers was induced under continuous 38°F temperatures in the controlled environment chamber than there were with 20 hours of 38°F and a short peak of 65°F temperatures. On the other hand, 15 hours at 38°F and 5 hours at 60°F temperatures were equal to (clone R) or more promotive (clone O), than 24 hours at 38°F temperatures in the number of induced floral tillers. The above data suggest that alternating temperatures may fulfill the temperature requirements for floral induction, as long as a critical maximum temperature level is not surpassed. This maximum temperature level may vary with genotype.

With all growth chamber treatments studied, clone R plants showed a higher number of induced apices than did clone O plants under comparable temperature levels and durations. However, field results indicated generally a higher number of floral apices for clone O than for clone R plants. The last three planting dates, however, were almost equal in fertile tiller number. The inconsistency between growth chamber and field trials concerning inflorescence number suggests that growth chamber trials were either too short in duration to produce comparable flowering data to field results, or other facts, such as photoperiod and soil fertility levels, were important for floral induction.

The results obtained in the present study seem to support Gardner and Loomis (30, p. 209-210), who found low temperatures essential for floral induction of orchardgrass. The hypothesis of Calder (12, p. 187-194) concerning light-dark alternation effects in floral inductions would not appear to apply to the genotypes used in this study.

All plants transferred to the field showed varying degrees of floral development. Since planting started as late in the year as November 22 and the last planting date was not until February 23, it could not be ascertained if a larger number of inflorescences per plant would have developed in earlier planting. Conversely, since even with the last plant date, February 28, plants still showed floral development, it was impossible to determine when environmental conditions may cease to be promotive for floral induction.

The field results showed that floral induction in these genotypes may take place in mid or late winter, as well as under fall conditions. However, fall planting seemed to favor fertile tiller production, at least in clone 0. The present findings appear slightly different from those reported by Gardner and Loomis (30, p. 210-213), who considered fall induction for orchardgrass under local conditions. However, these authors conducted their research in the Midwest where winter temperatures are usually below freezing which possibly are non-promotive to floral induction. Wycherley (71, p. 87-88), on the other hand, found that none of the orchardgrass strains investigated were florally induced by December under

the warmer climatic conditions of the Netherlands.

Inflorescence exertion time showed an inverse linear relationship with temperature exposure. The longer the exposure to a particularly effectious treatment, the shorter the time required for the first tiller to show a panicle. This relationship held true with only a few exceptions for the field, as well as the greenhouse treatments. These results seem to be in accordance with the results obtained by Mitchell (49) with Lolium and Wycherley (76, p. 87-88) with orchardgrass. The latter author reported more rapid head development from transplants to the greenhouse after December.

Tiller development under growth chamber conditions generally showed a positive linear relationship to increasing exposure durations. With the temperature regimes studied, clone R plants appeared to develop tillers more rapidly than did clone O plants. The total number of tillers developed per plant to the time when head exertion was completed, was also higher for clone R than for clone O plants. Cyclic temperatures were superior in tiller development than were continuous 38°F temperatures, suggesting that the increase in number of flowers after cyclic cold treatments may be partially due to the potentially larger number of apices present, which may then be induced to flower. Temperatures higher than 65°F during the diurnal cycle were promotive to tiller development. However, these same temperature levels were little or non-promoting for floral induction.

Tiller development in the field followed a positive linear trend with plants of earlier planting dates showing the higher



number of tillers. Clone O plants generally developed a larger number of tillers than did clone R plants in the field, while in the growth chamber the reverse was true. It should be pointed out that the maximum growth chamber exposure periods of eight to ten weeks were roughly comparable with the last two field planting dates, where neither clone showed clear superiority in number of tillers developed. Brevity of exposure duration might explain the diverse results obtained between field and growth chamber studies.

The highest percent floral tillers was obtained from clone R plants which had been subjected to continuous 38°F temperatures for eight weeks in the growth chamber. However, this treatment also had a relatively low number of total tillers developed. The high percentage floral tillers was probably due to the low number of total tillers developed. Overall, the longer the treatment exposure, the higher the percentage floral tillers, although the longer treatment exposures were also higher in the total number of tillers developed per plant.

Field results showed clone O plants of most planting dates producing higher percent fertile tillers than the respective clone R plants. Differences in percent floral tillers among the various planting dates for clone O plants were more pronounced than they were for the corresponding clone R plants. The data furthermore suggested that percent floral tillers in clone O plants was directly related to the length of field exposure. The field exposure period showed a lesser effect on percent fertile tillers in clone R plants. The longer exposure to winter conditions appeared

to exert a positive influence on the number of fertile tillers developed. Quantitative floral responses due to temperature durations had been previously substantiated by Fejer (27) for ryegrass and Peterson and Loomis (59, p. 39) for bluegrass.

Leaf development due to the various temperatures treatments and durations followed generally a linear trend, the leaf number increasing proportionally to exposure periods. This substantiates earlier published results (19, p. 262-279). The higher the temperature and the longer the period of the same temperature during the diurnal cycle, the more rapid the leaf development. Cold temperatures during the cycle reduced leaf development. This also had been found previously by Templeton et al. (69), who also considered important the interaction between photoperiod and temperature in subsequent leaf development. Cooper (19, p. 262-279) found leaf development independent of apical condition. The results found for leaf development in the main tiller after growth chamber treatment substantiates Cooper's findings.

## SUMMARY AND CONCLUSIONS

Two genotypes of the synthetic orchardgrass variety Pennlate, clone R and clone O, were used to investigate the influence of various temperature levels and durations on floral induction, floral development and vegetative growth. Temperature levels selected for the growth chamber trials were as follows:

1. 10 hours at 38°F, 10 hours at 70°F and 4 hours change
2. 24 hours at 28°F
3. 15 hours at 38°F, 5 hours at 70°F and 4 hours change
4. 10 hours at 38°F, 10 hours at 90°F and 4 hours change
5. 15 hours at 38°F, 5 hours at 60°F and 4 hours change
6. 20 hours at 38°F to a peak of 65°F and 4 hours change

All growth chamber treatments had an eight-hour photoperiod and exposure durations varied from two to ten weeks.

The field experiment consisted of eight dates during late fall and winter with time intervals of two weeks between plantings.

Growth chamber results indicated that 90°F temperatures for 10 hours offset 10 hours of 38°F temperatures and inhibited floral induction in the clones used. Generally, the longer the exposure to any of the other five treatments, the higher the percentage of plants induced to flowering, and the larger the number of visible inflorescences. Twenty-four hours at 38°F and 15 hours at 38°F with 5 hours at 60°F and 4 hours change, were the most effective temperature treatments for increasing the number of inflorescences produced per plant.

Clone R plants appeared to be less dependent on extended periods of inductive temperature levels than were clone O plants. Six weeks appeared to be the minimum duration for effective floral induction under favorable temperature regimes. Clone R plants, on the other hand, were florally induced with only four weeks exposure to favorable temperatures. Flowering of 100 percent for clone R plants was obtained with several temperature levels and durations. In clone O, cyclic diurnal temperatures of 15 hours at 38°F and 5 hours at 60°F, as well as continuous 38°F temperatures, appeared best in percent of floral apices and number of exerted inflorescences recorded. Temperatures above 60°F reduced floral induction and number of inflorescences developed for clone O plants.

Tiller development during the different temperature treatments was apparently favored by cyclic temperatures rather than by continuous 38°F temperatures. Leaf development during the different treatments was related to the amount of higher temperatures received. Tillering, even after removal from the growth chamber, was generally proportional to treatment durations and accelerated by those cycles consisting of temperatures of 70°F and 90°F during the day.

Field trial results showed an increased number of panicles and non-exserted inflorescences for the earlier planting dates. Clone O plants in particular responded positively in the number of inflorescences developed for the first three planting dates. Clone O plants showed a rapid decline in the number of inflorescences developed with later planting dates. Clone R plants also showed a

positive linear relationship between flower number and exposure time to winter conditions. Differences in the number of inflorescences developed at various planting dates, however, were less pronounced in clone R plants than in clone O plants.

Clone O plants generally produced a larger number of reproductive tillers, as well as a higher percentage of floral primordia than did clone R plants at the same planting dates. Clone R plants furthermore showed little difference in percentage of reproductive tillers for the various planting dates. Clone O plants, on the other hand, had a range from 43 to 20 percent reproductive tillers for the first and last planting date respectively.

Vegetative tiller development in the field was positively related with earliness of planting time for both clones. Tiller number differences between planting dates, however, were more pronounced in clone O than they were in clone R plants.

Leaf development appeared to be related to the amount of high temperature received.

## BIBLIOGRAPHY

1. Ahrens, J. F. and W. E. Loomis. Floral induction and development in winter wheat. *Crop Science* 3:463-466. 1963.
2. Aitken, Yvonne. Flower initiation in pasture legumes. I. Factors affecting flower initiation in Trifolium subterraneum L. *Australian Journal of Agricultural Research* 6:212-244. 1955.
3. Aitken, Yvonne. Flower initiation in pasture legumes. II. Geographical implications of cold temperature requirements of varieties of Trifolium subterraneum L. *Australian Journal of Agricultural Research* 6:245-257. 1955.
4. Allard, H. A. and M. W. Evans. Growth and flowering of some tame and wild grasses in response to different photoperiods. *Journal of Agricultural Research* 62:193-228. 1941.
5. Beatty, D. W. Effect of photoperiod and temperature on flowering of white clover (Trifolium repens). *Dissertation Abstracts* 20:3460. 1960.
6. Bennett, H. W. The effect of temperature upon flowering in Paspalum. *Agronomy Journal* 51:169-171. 1959.
7. Bennett, H. W. The effect of temperature upon flowering in Paspalum. *Agronomy Journal* 51:191-193. 1959.
8. Bommer, D. Samenvernalisation perennierender Gräserarten. *Zeitschrift für Pflanzenzuchtung* 46:105-111. 1961.
9. Britten, E. J. Genetic and environmental control of flowering in Trifolium repens in the tropics. *Science* 131:100-101. 1960.
10. Bunning, Erwin. Die entwicklungsphysiologische Bedeutung der endogenen Tagesrythmik bei den Pflanzen. In: A. E. Murneek et al.'s Vernalization and photoperiodism. Waltham, Mass., *Chronica Botanica*, 1948. p. 167-174.
11. Calder, D. M. Flowering responses in Dactylis. *Herbage Abstracts* 32:1525. 1962.
12. Calder, D. M. Stage development and flowering in Dactylis glomerata L. *Annals of Botany* 28:187-206. 1964.

13. Chilcote, D. O. Environmental control of flowering in selected genotypes of orchardgrass (Dactylis glomerata). Ph.D. thesis. Lafayette, Purdue University, 1961. 107 numb. leaves.
14. Chouard, P. Vernalization and its relation to dormancy. Annual Review of Plant Physiology 11:191-238. 1960.
15. Cooper, J. P. Day-length and head formation in the ryegrasses. Journal of the British Grassland Society 5:105-112. 1950.
16. Cooper, J. P. Studies of growth and development in Lolium. II. Pattern of bud development of the shoot apex and its ecological significance. Journal of Ecology 38-39:228-270. 1950-51.
17. Cooper, J. P. Studies on growth and development in Lolium. III. Influence of season and latitude on ear emergence. Journal of Ecology 40:352-379. 1952.
18. Cooper, J. P. Studies on growth and development in Lolium. IV. Genetic control of heading responses in local populations. Journal of Ecology 42:521-556. 1954.
19. Cooper, J. P. Developmental analysis of population in the cereals and herbage grasses. I. Methods and techniques. Journal of Agricultural Science 47:262-279. 1956.
20. Cooper, J. P. Developmental analysis of populations in the cereals and herbage grasses. II. Response to low temperature vernalization. Journal of Agricultural Science 49:361-383. 1957.
21. Cooper, J. P. The effect of temperature and photoperiod on inflorescence development in strains of timothy. Journal of the British Grassland Society 13:81-91. 1958.
22. Cooper, J. P. The use of controlled life cycles in the forage grasses and legumes. Herbage Abstracts 30:71-79. 1960.
23. Cooper, J. P. Short-day and low temperature induction in Lolium. Annals of Botany 24:232-246. 1960.
24. Doorenboos, J. and S. J. Wellensiek. Photoperiodic control of floral induction. Annual Review of Plant Physiology 10:147-184. 1959.
25. Evans, L. T. Flower initiation in Trifolium subterraneum L. I. Analysis of the partial processes involved. Australian Journal of Agricultural Research 10:1-16. 1959.

26. Evans, L. T. Clue to photoperiodic effect. *Australian Journal of Biological Science* 13:429-440. 1960.
27. Fejer, S. O. Ear emergence in perennial ryegrass as affected by differences in light and temperature before ear initiation. *New Zealand Journal of Agricultural Research* 3:558-564. 1960.
28. Friend, D. J. C. and F. G. Gregory. Acceleration of flowering in partially vernalized grain of Petkus winter rye by subsequent treatment at high temperature. *Nature* 172(4380):667-668. 1953.
29. Gall, H. J. F. Flowering of smooth brome grass under certain environmental conditions. *Botanical Gazette* 109:59-71. 1947.
30. Gardner, F. P. and W. E. Loomis. Floral induction and development in orchardgrass. *Plant Physiology* 28:201-217. 1953.
31. Gott, M. B., F. G. Gregory and O. N. Purvis. Studies in vernalization of cereals. XIII. Photoperiodic control of stages of flowering between initiation and ear formation in vernalized and unvernallized Petkus winter rye. *Annals of Botany n.s.*, 19:87-126. 1955.
32. Gregory, F. G. The control of flowering plants. *Symposia of the Society for Experimental Biology* 2:75-103. 1948.
33. Hansel, H. Vernalization of winter rye by vegetative temperatures and the influence of vernalization upon the laminar length of the first and second leaf in winter rye, spring barley, and winter barley. *Annals of Botany n.s.*, 17:417-432. 1953.
34. Hammer, K. C. Factors governing the induction and development of reproductive structures in plants. *Symposia of the Society for Experimental Biology* 2:104-116. 1948.
35. Hillmann, William S. *The physiology of flowering*. New York, Holt Rinehart and Winston, 1962. 164 p.
36. Kasperbauer, M. J. and W. E. Loomis. Interaction of vernalization and photoperiod in flowering of sweet clover. *Plant Physiology* 36 (supplement):51. 1961.
37. Khalil, M. S. H. The interrelation between growth and development of wheat as influenced by temperature, light and nitrogen. *Mededelingen Landbouwhogeschool Wageningen* 56(7):1-73. 1956.



38. Knight, W. E. and H. W. Bennett. Preliminary report on effect of photoperiod and temperature on flowering and growth of several Southern grasses. *Agronomy Journal* 45:268-269. 1953.
39. Knight, W. E. The influence of photoperiod and temperature on growth, flowering and seed production of Dallisgrass (Paspalum dilatatum). *Agronomy Journal* 47:555-559. 1955.
40. Knowles, R. P. Seed production of perennial grasses in the greenhouse. *Canadian Journal of Plant Science* 41:1-9. 1941.
41. Lang, A. and G. Melchers. Vernalisation und Devernalisation bei einer zweijährigen Pflanze. *Zeitschrift für Naturforschung*, ser B, 2:444-449. 1947.
42. Lang, A. The physiology of flowering. *Annual Review of Plant Physiology* 3:265-306. 1952.
43. Langer, R. H. M. Growth and nutrition of timothy (Phleum pratense). II. Growth of the plant in relation to tiller development. *Annals of Applied Biology* 45:528-541. 1957.
44. Langer, R. H. M. A study of growth in swards of timothy and meadow fescue. I. Uninterrupted growth. *Journal of Agricultural Science* 51:347-351. 1958.
45. Leopold, A. C. and F. S. Guernsey. Modification of floral initiation with auxins and temperatures. *American Journal of Botany* 40:603-607. 1953.
46. Liverman, J. L. The physiology of flowering. *Annual Review of Plant Physiology* 6:177-210. 1955.
47. McCown, R. L. and M. L. Peterson. Effects of low temperature and age of plant on flowering in Lolium perenne. *Crop Science* 4:338-391. 1964.
48. Mitchell, K. J. Influence of light and temperature on the growth of ryegrass. I. The control of lateral bud development. *Physiologia Plantarum* 6:425-443. 1953.
49. Mitchell, K. J. Influence of light and temperature on the growth of ryegrass, Lolium species. I. Pattern of vegetative development. *Physiologia Plantarum* 6:21-46. 1953.
50. Mitchell, K. J. Growth of past species. II. Perennial ryegrass (Lolium perenne), cocksfoot (Dactylis glomerata), and paspalum (Paspalum dilatatum). *New Zealand Journal of Scientific Technology* 37:8-26. 1955.

51. Mitchell, K. J. The influence of light and temperature on the growth of pasture species. Seventh International Grassland Congress, Proceedings, 1956, p. 58-69.
52. Nanda, K. K., J. J. Chinoy and G. S. Sirohi. Energy relations of crop plants. I. The quantitative relationship between the duration of vernalization treatment and photothermic quanta in wheat. Indian Journal of Plant Physiology 1:5-16. 1958.
53. Nitsan, J. Accumulation of a distinct macro-molecular compound in rye embryos as a result of vernalization. Nature 194:400-401. 1962.
54. Nitsan, J. Possible role of polysaccharides in floral induction. Nature 198:100. 1963.
55. Peterson, M. L. and W. E. Loomis. Effects of photoperiod and temperature on growth and flowering of Kentucky bluegrass. Plant Physiology 24:31-43. 1949.
56. Peterson, M. L., J. P. Cooper and L. Bendixen. Thermal and photoperiodic induction of flowering in Darnel (Lolium temulentum). Crop Science 1:17-20. 1962.
57. Purvis, O. N. and F. G. Gregory. Studies in vernalization of cereals. I. A comparative study of vernalizations of winter rye by low temperature and by short days. Annals of Botany n.s., 1:569-590. 1937.
58. Purvis, O. N. Studies in the vernalization of cereals. X. The effect of depletion of carbohydrates on the growth and vernalization response of excised embryos. Annals of Botany n.s., 11:269-283. 1947.
59. Purvis, O. N. Studies in vernalization. XI. The effect of date of sowing and of excising the embryo upon the responses of Petkus winter rye to different periods of vernalization treatment. Annals of Botany n.s., 12:183-206. 1948.
60. Purvis, O. N. and F. G. Gregory. Studies in vernalization. XII. The reversibility by high temperatures of the vernalized condition in Petkus winter rye. Annals of Botany n.s., 16:1-21. 1952.
61. Purvis, O. N. The physiology of flower differentiation in grasses. Sixth International Grassland Congress, Proceedings 1:661-666. 1952.
62. Purvis, O. N. and F. G. Gregory. Acceleration effect of an extract of vernalized embryos of winter rye on flower initiation in unvernallized embryos. Nature 171(4355):687-688. 1953.

63. Roberts, R. H. and Esther B. Struckmeyer. The effects of temperature and other environmental factors upon the photoperiodic responses of some of the higher plants. *Journal of Agricultural Research* 56:633-677. 1938.
64. Schwabe, W. W. Factors controlling flowering in the *Chrysanthemum*. V. Devernalization in relation to high temperature and low light intensity treatments. *Journal of Experimental Botany* 6:435-450. 1955.
65. Sprague, V. G. The relation of supplementary light and soil fertility to heading in the greenhouse of several perennial forage grasses. *Journal of the American Society of Agronomy* 40:144-154. 1948.
66. Taylor, T. H. Some morphological and physiological aspects of the orchardgrass tiller. Report of the Joint Meeting of the Western Grass Breeders Work Planning Conference and Nineteenth Southern Pasture and Forage Crop Improvement Conference, 1962. p. 30-31.
67. Templeton, W. C. Jr., G. O. Mott and R. J. Bula. Some effects of temperature and light on growth and flowering of tall fescue, *Festuca arundinacea* Schreb. I. Vegetative development. *Crop Science* 1:216-219. 1961.
68. Tincker, M. A. H. The effect of length of day upon the growth and reproduction of some economic plants. *Annals of Botany* 39:721-754. 1925.
69. Tukey, H. B. Jr. and H. J. Ketellapper. The effect of temperature and length of the light-dark cycle on plant growth. *Plant Physiology* 34 (supplement):iv. 1959.
70. Wellensiek, S. J. Vernalization and age in *Lunaris biennis*. Koninklijke Nederlandse Akademie van Wetenschappen Proceedings, series C 61(5):561-471. 1958.
71. Wilson, J. R. The influence of time of tiller origin and nitrogen level on the floral initiation and ear emergence of four pasture grasses. *New Zealand Journal of Agricultural Research* 2:915-932. 1959.
72. Wolfe, T. H. Observations on the blooming of orchardgrass flowers. *Agronomy Journal* 17:605-618. 1925.
73. Wycherley, P. R. Temperature and photoperiod in relation to flowering in three perennial grass species. *Mededelingen Landbouwhogeschool Wageningen* 52(2):75-92. 1952.