THE PHYSIOLOGY OF SPROUTING AND FLOWERING IN ONION BULBS: PHOTOPERIOD AND TEMPERATURE EFFECTS

D. S. Bertaud

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Abstract

Onion bulb growth and development are affected by photoperiod, light quality and light interception. In short-days no bulbs are formed; in long-days large bulbs are formed, but take some time to mature; in very long-days small bulbs are formed and mature quickly. The effect of photoperiod depends on light quality; bulb growth is stimulated by far-red light. In conditions favourable to bulbing, sucrose and oligosaccharides accumulate in the bulb. Bulb growth and carbohydrate accumulation depend on current environmental conditions and cease if conditions are no longer favourable. High light intensity and leaf area enhance bulb growth.

Photoperiod and temperature effects on sprouting and floral development, and the effect of duration of chilling at 8°C on inflorescence development, were investigated in three cultivars of onion. The cultivars used were early maturing ('Sakigake Yellow'), midseason ('Gladalan Brown') and late maturing ('Early Long Keeper'). These cultivars were subjected to treatments comprising all combinations of two photoperiods (8 h and 14 h) and two constant temperatures (15°C and 20°C), except that 'Sakigake Yellow' was subjected to constant 8°C in 8 h days instead of 15°C and 14 h days. 'Sakigake Yellow' sprouted readily under all conditions except 8°C; 'Gladalan Brown' sprouted much faster in short-days than in long-days; and 'Early Long Keeper' sprouted poorly in all conditions. In long-days, some 'Gladalan Brown' plants showed evidence of remobilisation of assimilates, leading to growth of new bulbs within the original one, without growth of leaf blades.

'Early Long Keeper' plants were slow to develop roots as well as to sprout. Leaf appearance rates after sprouting were affected by photoperiod in 'Sakigake Yellow', and in 'Gladalan Brown' they were affected by both photoperiod and temperature. Rates were lower in long-days, and photoperiod had more effect at 15°C than at 20°C. It appears that bulbs of 'Sakigake Yellow' and 'Gladalan Brown' were ecodormant when planted, requiring only sufficient water to begin growth; however, leaf appearance in 'Sakigake Yellow', and both sprouting and leaf appearance in 'Gladalan Brown', showed a photomorphogenetic response to photoperiod. 'Early Long Keeper' bulbs were very slow to root and to sprout, indicating that they required more than the availability of water to begin growth, and were therefore not simply ecodormant when planted. The effects of duration of chilling at 8°C and subsequent photoperiod and temperature on floral initiation and development in three onion cultivars were investigated. Four weeks of chilling at 8°C followed by four weeks of growth at 15°C was sufficient for floral initiation in 'Gladalan Brown' and 'Early Long Keeper', but the bolting-resistant cultivar 'Sakigake Yellow' required eight weeks of chilling at 8°C followed by four weeks of growth at 15°C. With all cultivars, subsequent emergence of inflorescences was more rapid in 14 h days than in 8 h days, but at 20°C there was abortion of some inflorescences in 14 h days. Development of the emerged inflorescence to anthesis was also rapid in 14 h days, and faster at 20°C than at 15°C. There was apparent competition between leaf growth and inflorescence development in 8 h days, with resultant abortion of some inflorescences. At 15°C in 8 h days many of the inflorescences had bulbils or malformed florets in the inflorescence. Mature onion bulbs may be brought into flower in 35 weeks by sprouting them for 8 weeks in 15°C and 8 h days, followed sequentially by 8 weeks chilling at 8°C, 10 weeks at 15°C and 8 h days (until most plants have emerged inflorescences), and 8-10 weeks at 20°C in 14 h days. Knowledge of the responses of specific cultivars would allow the time to flowering from planting to be reduced further. Malformations occurring during floral development in unfavourable

conditions are described; these are compared with floral development in *Allium cepa* var. *proliferum*. A conceptual model for dry weight partitioning in onion bulbs is described.

Keywords: photomorphogenesis, *Allium cepa* L., dormancy, bulb sprouting, inflorescence initiation, floral initiation, inflorescence development, floral development, light interception, phytochrome, red/far-red ratio, plant growth models.

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Chapter 1

Introduction

The time taken to breed new cultivars of onion is largely determined by the length of a generation. Onions sown as seed in winter develop a bulb the following spring which matures in summer. This bulb remains dormant for a period ranging from a few weeks to several months, depending on cultivar. In late summer, autumn or winter the bulb sprouts, and during cool weather in winter inflorescences are initiated which mature, and set seed over the following spring and summer. Seed cannot be planted too early in autumn or winter under New Zealand conditions, or seedlings may bolt to flower instead of bulbing. Bulbing is required for the evaluation of progeny in breeding programmes. If the time from bulbing to seed production could be shortened to give a one year seed to seed generation time, breeding programmes could be accelerated.

Studies which have evaluated the effects of light on bulbing (Chapter 2) have suggested that the photomorphogenetic effects of photoperiod might have some effect on the leaf elongation required for sprouting, and possibly also on control of dormancy of bulbs. In this review, the hypothesis that bulbing is a consequence of changes in the partitioning of photosynthate in growing cells in response to current daylength is developed. The results reviewed by Brewster (1977), together with more recent work on carbohydrate metabolism (Lercari, 1982a, b, 1983), light

interception (Brewster, 1982b), light quality and planting density (Mondal *et al.*, 1986a, b, c; Brewster *et al.*, 1986) and growth rates (de Ruiter, 1986), are used in developing the hypothesis.

Onion flowering physiology has been reviewed by Brewster (1977) and more recently by Rabinowitch (1985). Studies have shown that a period of low temperature is required for inflorescence initiation, and abortion can occur if the newly formed inflorescence is subjected to high (20-30°C) temperatures. However, development of the emerged inflorescence and seeds was advanced by temperatures in this range.

This work was carried out to investigate the effect of temperature and photoperiod on sprouting of newly harvested bulbs, and of duration of chilling and subsequent photoperiod and temperature treatments on floral development in prematurely sprouted plants. The objective was to define a set of environmental conditions which would allow production of onion seed from seed in one year, and would also allow for synchronous flowering of cultivars.

Sections 1 and 2 of Chapter 2 have appeared as a paper in Proceedings of the New Zealand Agronomy Society (Bertaud, 1986); Chapter 4 is written as a paper to be submitted to the New Zealand Journal of Experimental Agriculture; and Chapter 5 will be submitted as a paper for the Fourth Eucarpia *Allium* Symposium.

1.1 REFERENCE

Bertaud, D. S. 1986: The effect of light on bulbing in onions. *Proceedings of the New Zealand Agronomy Society 16* : 79-86.

Chapter 2

Onion physiology

2.1 THE STRUCTURE OF AN ONION PLANT

Onions (*Allium cepa* L.) are monocotyledonous plants, and may be placed in either the Liliaceae or Amaryllidiaceae families (Rabinowitch, 1985). The genus *Allium* has also been placed in a separate family, the Alliaceae (Willis, 1966). Leaves arise from a very short, flat stem (Fig. 2.1). The outermost leaves are the oldest, and new leaves arise from the apical meristem at the tip (in the middle) of the stem.

Onions produce two kinds of leaves; green foliage leaves with a long, cylindrical sheath and a hollow blade that grows from one side of the sheath, and non-green scale leaves, where the blade does not develop and the hollow sheath is thickened as storage tissue (Fig. 2.1). Younger foliage leaves emerge through the hole at the top of the sheath (where the ligule would be on a grass leaf). There is a high turnover of leaves on a growing onion (de Ruiter, 1986). Older leaves die and are sloughed off as the diameter of the stem increases with successive leaves. Some of the scales on a mature onion bulb are thickened leaf bases (the lower parts of the sheaths) and some are scale-leaves. These organs store carbohydrate in the form of sucrose and more complex oligosaccharides (mainly fructans with various degrees of polymerisation). Growth of the bulb is a result of thickening of leaf-bases and

scale-leaves. There are generally 4 - 8 scales on a mature bulb, plus a number of immature leaves and primordia. The proportion of the bulb made up of scale leaves varies between cultivars and between plants (de Ruiter, pers. comm.). Generally, most of the volume of the bulb is made up of leaf-bases; however, scale leaves may comprise up to 74% of the fresh weight (Bertaud, unpublished work).

When conditions are suitable for bulbing to occur, the growing bases of leaf sheaths thicken as cells expand and fill with complex sugars (Abdalla and Mann, 1963). The stem apex begins to produce scale leaves, which do not extend above the top of the bulb, so that the last green leaves, when fully grown, are not supported by emerging new leaves. Eventually the remaining green leaves fall over, and the onion is said to be 'top down', or mature.

Onion leaves mature first at the tip (Bertaud, unpublished work); we assume that mature leaf tissue is continuously pushed up by growth at the leaf base until the leaf is mature, as in grass leaves. This assumption is supported by the work of Denne (1960) on *Narcissus*, a bulb-forming plant which also produces foliage leaves and scale leaves. Patterns of leaf growth and cell elongation in *Narcissus* were the same for both foliage and scale leaves that were 0.5-1 mm long. In foliage leaves, the intercalary meristem present in the primordium was maintained and enlarged, though it never extended more than 50 mm up the blade, even when the blade was 250 mm long (Denne, 1960). When the blade was nearly mature, cell division became restricted to the sheath. In scale leaves, however, leaf growth and cell division were restricted to the sheath very early in development, without the intervening stage of blade growth (Denne, 1960). This difference in growth suggests that partitioning of photosynthates between sheath and blade is the process that is affected by environmental conditions causing bulbing.

Inflorescence initiation occurs during a period of low (7-15°C) temperature (Heath and Mathur, 1944; Holdsworth and Heath, 1950; Brewster, 1982a, 1983; Currah, 1981). The stem apex broadens and flattens, and the last foliar primordium develops into a sheathing structure called a *spathe* which will envelop the florets (Fig. 2.2).

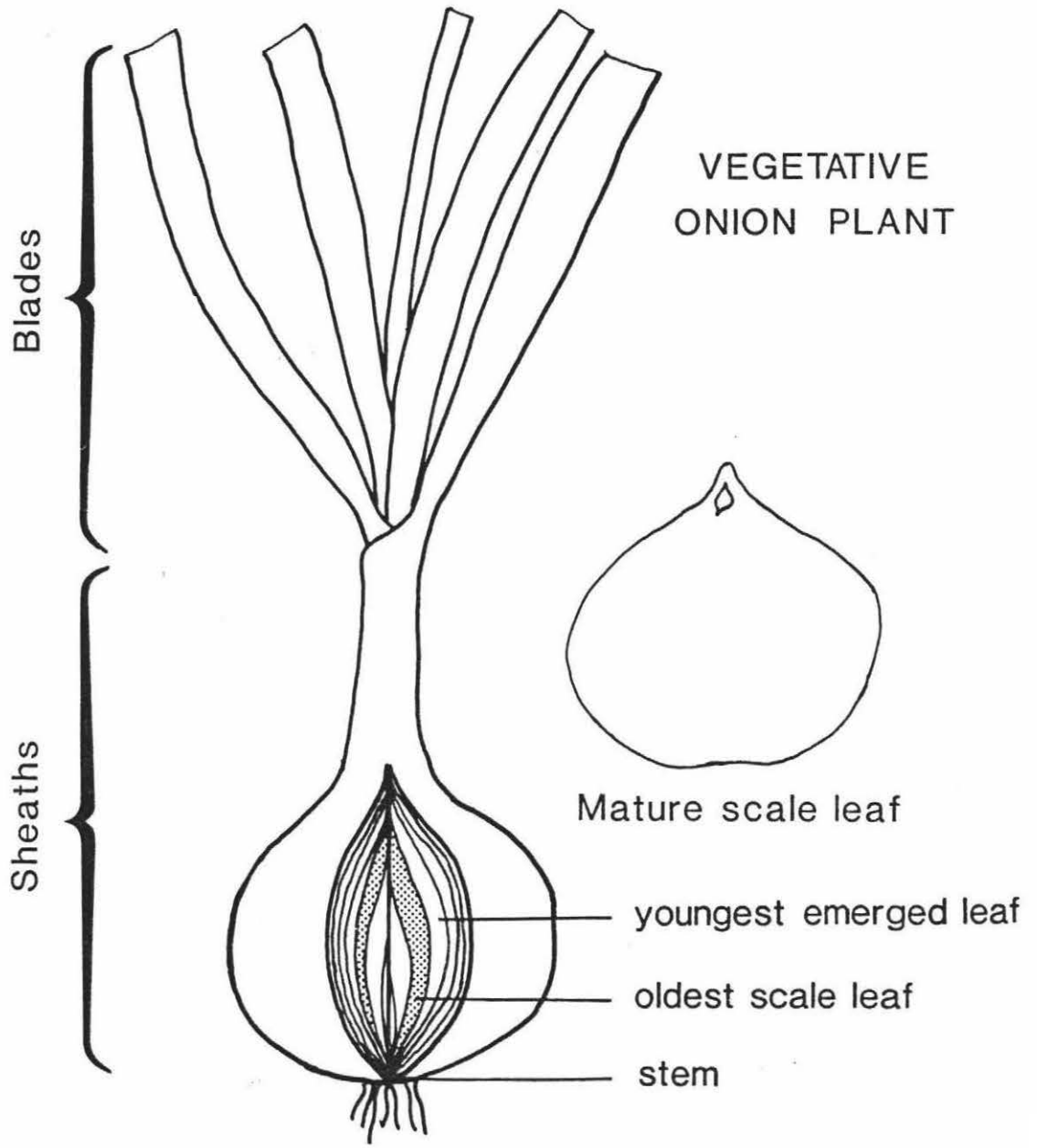

Figure 2.1: Schematic drawing of an onion plant, showing growth habit and (inset) a scale leaf. The two outermost leaves on the bulb appear as a single line on this scale.

The internode between the spathe and the last leaf elongates, forming an unbranched stem, the *scape*. The apical dome within the spathe expands to form a meristematic *mantle* which produces florets (Jones and Emsweller, 1936).

Elongation of the scape pushes the spathe up through the sheaths of the subtending leaves, and continues until the head of the inflorescence is well above the leaf canopy. When the spathe emerges from the sheaths of older leaves floral primordia are visible on the mantle, but florets mature after emergence. As florets mature, the spathe dries and eventually splits to reveal the florets in bud. A single inflorescence may contain 50-2000 florets (usually about 600), which open over a period of 3 - 5 weeks (Currah, 1981).

2.2 THE EFFECTS OF LIGHT ON BULBING

2.2.1 Photoperiod effects

Bulbing in onions has been known to depend on photoperiod since the work of Garner and Allard (1920). Magruder and Allard (1937) studied the effects on a range of varieties of photoperiods ranging from 10 h to 14.25 h, along with naturally increasing photoperiods. Only one variety produced bulbs that eventually dried down normally in 10 h days; most produced good bulbs in 14 h days. A typical pattern of response was that exhibited by the variety 'Early Yellow Globe' (Fig. 2.3), which showed very little evidence of bulbing in 10 h days. In 12 h days, bulbs formed, but the plants continued producing green leaves and the tops did not fall. In 13 h and 13.5 h days, the plants produced large bulbs; in 14 h and 14.25 h days, maturity was earlier and bulbs were progressively smaller. Bulb size decreased with increasing photoperiod, as plants reached topdown earlier. There was a considerable range in response to photoperiod within each variety, as well as between varieties. Austin, (1972) using a number of varieties, found that bulbs formed in continuous light (14h days extended by low intensity incandescent light, in a glasshouse) had weights of 1-5 g (about the size of a large bean).

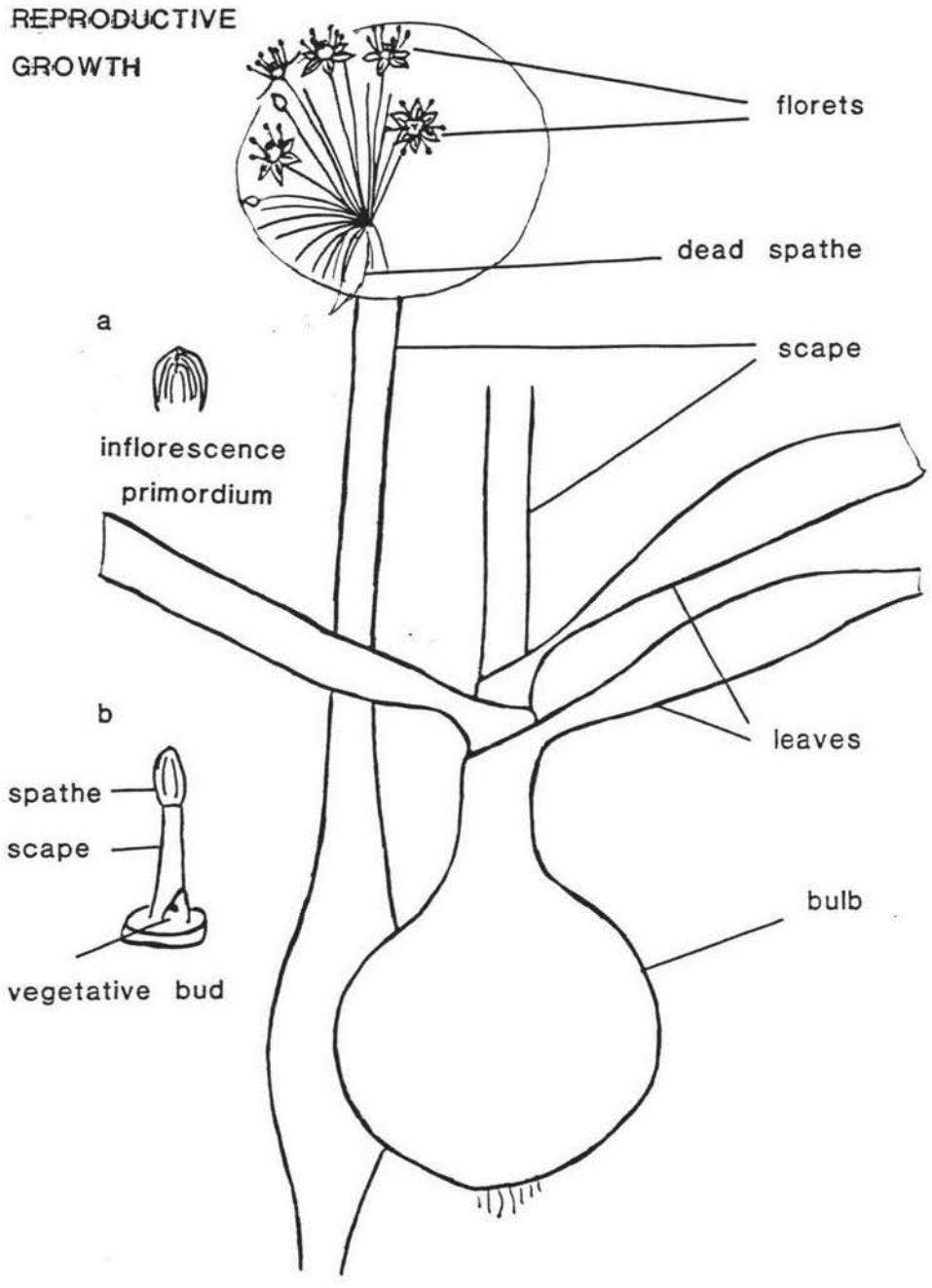

Figure 2.2: Schematic drawing of the inflorescence on an onion plant, showing (a) the inflorescence primordium, (b) a young inflorescence with scape elongating, (c) a mature inflorescence.

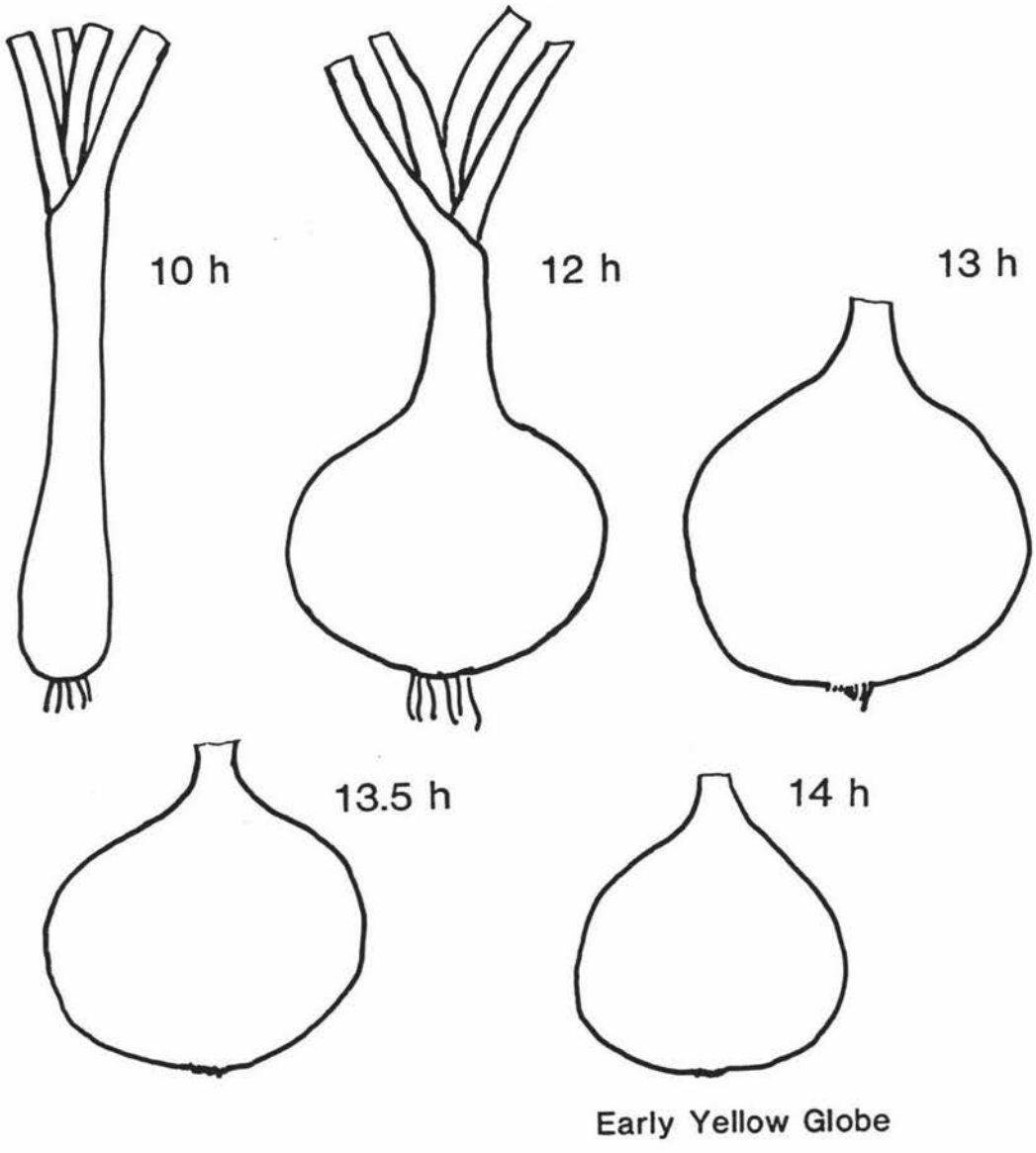

Figure 2.3: Effect of photoperiod on shape and size of onion bulbs. Redrawn from Magruder and Allard, 1937.

This pattern could be explained by a gradient in partitioning of photosynthates in response to photoperiod. In short-days, most photosynthates go to leaf extension; a higher proportion goes to bulb growth as days lengthen. Bulbs will form under some photoperiod conditions as a result of thickening of the leaf bases, but scale leaves are not produced, so bulb tops never fall. By contrast, in very long-days, all photosynthates go to the formation and development of scale tissue, leaving none for blade growth; leaf production stops early and bulbs mature early, so the period for bulb growth is short, and bulbs are small. However, although bulbs will form under changing photoperiods, bulbing does not require increasing photoperiods. Bulbs can be grown in decreasing photoperiods providing the photoperiod does not fall below a certain critical minimum (Kedar *et al.*, 1975).

The possibility that bulbing depends on total photosynthate rather than on photoperiod *per se* was investigated by Wright and Sobeih (1986). Plants exposed to a photosynthetic photon flux (PPF) of 67.5 μmol m^{-2} s^{-1} during 16 h days formed bulbs while those given 135 μmol m^{-2} s^{-1} during 8 h days did not, though the total photosynthetically active radiation was the same in the two treatments. Plants exposed to the high PPF for 16 h days, however, bulbed faster than those exposed to the low PPF. Extending an 8 h photoperiod with low light (11 W m^{-2}) was also effective in inducing bulbing (Wright and Sobeih, 1986).

Bulb growth can be stopped or reversed by transferring bulbing plants to short days. Bulb diameter growth slows and stops, green leaves are produced, and bulb diameter decreases as scales lose reserves and senesce. Renewed production of green leaves and reversal of bulbing can occur even after the tops have fallen (Kedar *et al.*, 1975). Bertaud (unpublished results) transferred immature, bulbing onions with mature, or near mature, green leaves from long days to short-days. Bulb development stopped, and instead new green leaves of atypical form were produced (Fig. 2.4). Since onion leaves mature first at the tip, the shapes of these leaves provide further information. Blades on scale leaves are reduced to residual points (Fig. 2.1) because growth becomes confined to the sheath at a very early stage of development (Denne, 1966). Evidently leaves which started maturing as scales have changed partway through

their development, while some tissue at the base of the blade was still capable of growing, and produced short green blades with normal sheaths. Under some conditions (e.g., low temperatures) mature scale-leaves were observed between the last leaf emerged before transfer and the first leaf emerged after transfer.

A similar phenomenon is observed when plants are transferred from short (8 h) days to long (14 h) days. The sheaths of young leaves with immature blades swelled to form thick, white scales, while the growing tissue at the base of the blades senesced, apparently because of a reduction in photosynthate supply (Fig. 2.5). Older leaves, which had mature blades and upper parts of the sheaths, developed white scale tissue in the lower parts of the sheaths.

This plasticity of the leaf tissue is probably responsible for the phenomenon observed during bulb growth, where long leaves with green blades finish development at the sheath base with a region of white, thickened scale tissue. Thus it seems that green or scale leaves are not initiated as such, but that a more general 'leaf' is initiated (c.f. Denne, 1960) which has the potential to develop as a green leaf, as a scale leaf, or as an intermediate form. The path of development taken is determined by the environmental conditions which prevail at the time cells within the leaf blade and sheath are growing and maturing.

These results indicate that bulbing is a continuous response to current photoperiod; it is not triggered by a short exposure to long-days. This is a different phenomenon from the classic flowering response to photoperiod, where flowering may often be induced irreversibly by a short exposure to inductive conditions (Salisbury, 1982). It also suggests that a term like 'onset of bulbing' (Mondal *et al.*, 1986a) or 'start of bulb growth' would be preferable to 'bulb initiation', since a bulb is not an organ to be initiated in the same way as, for instance, a flower.

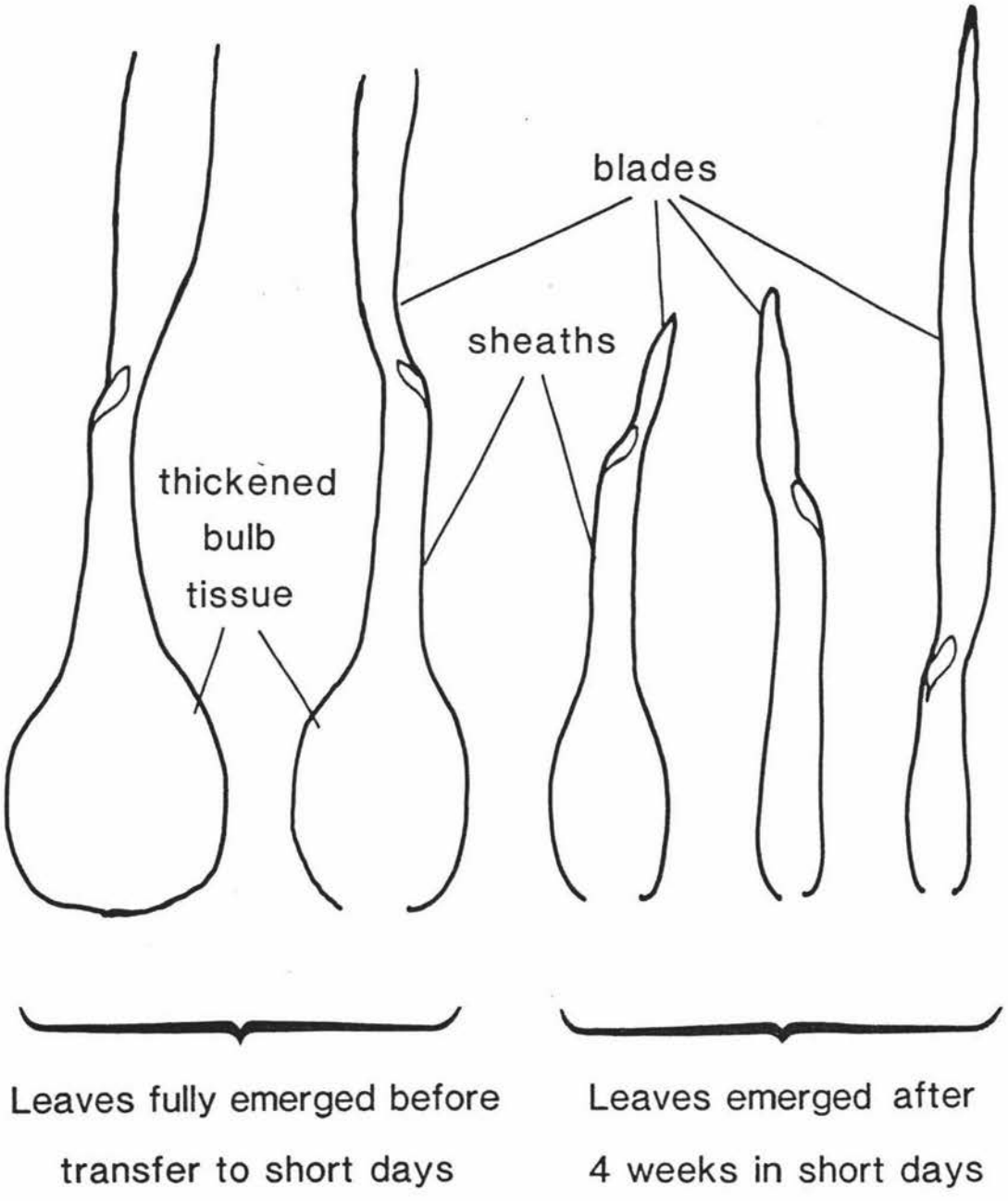

Figure 2.4: Schematic drawing of successive leaves from an immature, bulbing onion transferred from long to short-days (Bertaud, unpublished work).

Figure 2.5: Leaf from an onion transferred from short to long-days; growing tissue has senesced at the base of the blade.

2.2.2 Light quality effects

A number of authors (e.g. Austin, 1972; Lercari, 1982a, b; Mondal *et al.*, 1986b, c) have reported that bulbing in onions requires far-red light (FR). A higher proportion of far-red in the available light will increase the rate of bulbing. Red:far-red ratios of about 3.5 required 18 h days to produce bulbs comparable with those produced in sunlight (R:FR = 1.1) in natural photoperiods of 14 h (Austin, 1972).

Lercari (1982a) also found that the rate of bulbing increased with the proportion of far-red light (Fig. 2.6). There is a circadian rhythm in this response; far-red light applied alone for 3 h in the middle of the day was more effective than that applied at the beginning or end of the day; red light (R) and blue light were ineffective at all times. A 3 h dark break in the middle of an inductive photoperiod of 18 h also prevented bulbing. Since the results for red and blue light were similar to those for darkness, while that for far-red light was different, this is probably a photoperiodic effect rather than a total photosynthate effect. As such it may reflect a circadian rhythm in the response to light, or disruption of the phytochrome photoequilibrium.

Bulbing may be reversed by changing light quality as well as by a change from long to short (below the critical daylength required for bulbing) photoperiods (Lercari, 1982a). The accumulation of reducing sugars in the leaf blades and bulb observed under conditions of long-days with a high level of far-red light is also reversible, and there appears to be no lag when conditions are changed (Fig. 2.6) (Lercari, 1982b). Levels of oligosaccharides and sucrose also increase in the bulb under these conditions - this is reversible, but there is a lag between the change of conditions and a drop in oligosaccharide levels. The action spectrum of the response suggests that phytochrome may be involved, possibly because phytochrome photoequilibrium position affects carbohydrate metabolism in some way (Lercari, 1982b). The responses are observed very soon after conditions are changed.

Mondal *et al.* (1986b, c) have demonstrated an effect of light quality on bulb growth and maturity. Light transmitted through leaves is known to be enhanced in far-red

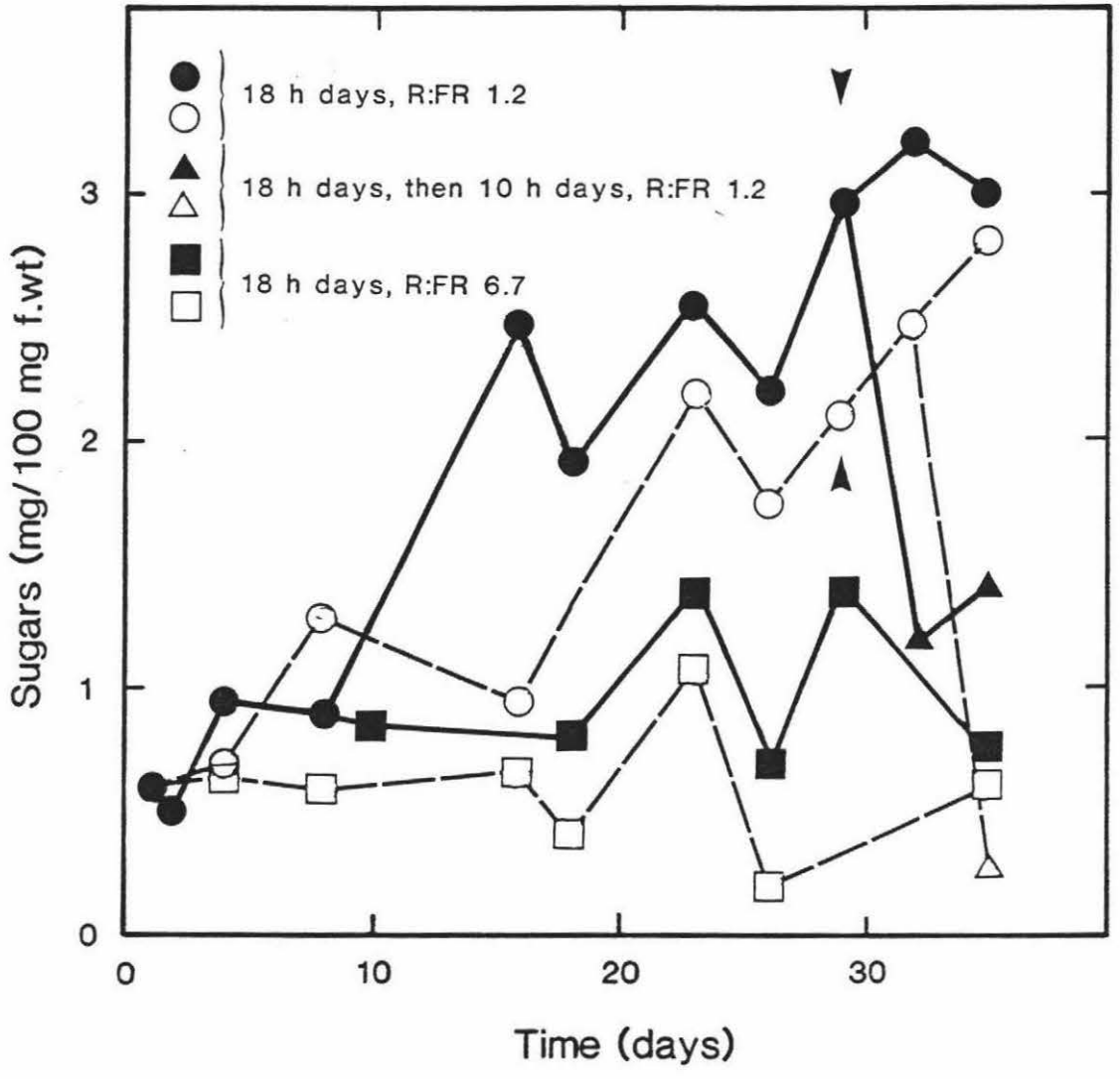

Figure 2.6: Effect of light quality on levels of oligosaccharides and reducing sugars in onion bulbs (redrawn from Lercari, 1982). Solid symbols: reducing sugars. Open symbols: oligosaccharides and sucrose.

light relative to red light. Onions grown under a canopy of leaves (climbing peas and beans) matured about 10 days earlier than those grown under neutral shade or in sunlight. Similar effects were observed for plants in very high density plantings (400 plants m^{-2}). In both cases, leaf appearance ceased earlier and fewer leaves were produced. Unfortunately the sizes of the resulting bulbs were not recorded. Some evidence of fluence rate effects has been reported (Lercari, 1983; Mondal *et al.*, 1986b). Wright and Sobeih (1986) report that bulbing is delayed by low light intensity for a given photoperiod.

Bulbing appears to be a classic, fluence-rate dependent, 'high irradiance response' (e.g. Lercari 1982a, 1983). This suggests it is analogous to the photoperiod responses reported for internode elongation, which have been demonstrated to involve phytochrome.

2.2.3 Light interception effects

Brewster (1982) showed that total shoot dry matter yields of onion crops were linearly related to total radiation intercepted during bulb growth. On average the onion crop, planted at a density of 100 plants m^{-2}, intercepted 49% of total radiation. The maximum observed was 65% for early sowings of late maturing cultivars which had time to establish a larger leaf area.

Maturity date decreases linearly with fraction of light intercepted. Brewster *et al.*, (1986) showed that crops which established a high leaf area index (LAI) early in growth matured early. They achieved this by very high density (400 plants m^{-2}) plantings, irrigated and fertilised to minimise competition for water and nutrients. However, duration of bulb growth was reduced by high light interception, at least in high density plantings. The difficulty with this method of achieving high light interception is that the results could also be affected by changes in light quality under the canopy.

The longer duration of bulbing in autumn-sown crops, which led to bigger bulbs, may

have occurred as a result of lower LAIs established over winter. Spring-sown crops, earlier sowings within a season, and later-maturing cultivars, which intercepted a higher proportion of the incident radiation, had a shorter duration of bulbing. A cold winter, which resulted in smaller plants in spring, delayed maturity of normally early-maturing cultivars (Brewster 1982b).

It appears that bulb scale development may be delayed by low LAI in some cultivars (Brewster *et al.*, 1986). Early maturing cultivars appear to respond more to photoperiod or temperature and begin bulbing at a low LAI. This tends to reduce yield, probably because the bulb scales compete more strongly with the new blades as sinks for photosynthate, reducing the later development of leaf area. As the photoperiod increases, the leaf bases become stronger sinks, until eventually no blades are formed, and scale leaves are produced. Since yield is increased both by high LAI and by long duration of bulbing, but duration of bulbing is decreased by high LAI, these results suggest that final yield will always be limited by physiological responses.

Sobeih and Wright (1986) found that bulbing was delayed by reducing the leaf area of plants. The ages of the remaining leaves were also important; later formed leaves appeared to be more sensitive to photoperiod, regardless of leaf area.

2.2.4 Implications for bulb sprouting

Onion bulbs appear to go through two phases of dormancy; a phase where the bulb will not sprout even if supplied with water and nutrients, and a phase where only water (and possibly nutrients) is needed to produce visible growth (Abdalla and Mann, 1963). Work of Isenberg *et al.* (1974), indicating that growth inhibitors are produced in the leaves and translocated to the bulb during bulbing, suggests that the first phase may be called paradormancy (Lang *et al.*, 1987), since it appears that the stimulus for dormancy is perceived by the leaves rather than by the stem apex. The second phase appears to be a form of ecodormancy, where the bulb requires

only provision of normal conditions, such as availability of water and nutrients, for growth.

The period of time after harvest that elapses before the bulb becomes ecodormant varies with cultivar (Abdalla and Mann, 1963). Data of these authors suggest that the transition to ecodormancy is temperature-dependent, proceeding fastest at 10-15°C. The effect of photoperiod on dormancy and sprouting has not been studied explicitly.

Sprouting of bulbs involves growth of bladed leaves rather than scale leaves. It seems likely, therefore, that photoperiod may play a part in promoting sprouting, resulting from the photomorphogenetic response of growing leaves, whether emerged or not. It is also possible that photoperiod responses contribute to the maintenance of dormancy. If this is so, an effect of photoperiod on sprouting in freshly harvested onions should be observed. Short days would be expected to be more conducive to sprouting than long-days.

2.3 FLOWERING

2.3.1 Initiation and emergence

Onion plants require a period of low temperature to initiate inflorescences (Holdsworth and Heath, 1950, Brewster, 1977). For temperate cultivars, inflorescence initiation is most rapid in the range 7-13°C; cultivars grown in Northern Russia require 4-6°C (Brewster, 1986), while varieties grown in tropical Africa are reported to initiate inflorescences in the 15-20°C temperature range (Sinnadurai, 1970). Storing dormant bulbs at 10°C from harvest in early summer until planting in autumn has no effect on subsequent floral development (Brewster, 1982a). Bulbs stored at 15-20°C for four weeks after harvest, then transferred to 7-10°C for one to three months (Behairy and El-Habbasha, 1979) or six months (DeMille and Vest, 1976), showed an effect of temperature on floral development. Behairy and El-Habbasha

(1979) found an an increase in flowering with chilling compared to 25°C, and with duration of chilling. DeMille and Vest (1976) found that a temperature of 7°C was more favourable to floral induction than 2°C, and that the effect of 2°C temperatures varied with time of application. These bulbs would be expected to have lost dormancy rapidly when stored at at 15-20°C (Abdalla and Mann, 1963), and the effect of chilling could be different from that reported by Brewster (1982a) as a result. Holdsworth and Heath (1950) reported that when bulbed and unbulbed plants of the same age (grown in long-days and short-days respectively) were placed in 11 h days at a mean temperature of 12.6°C, bulbed plants were significantly slower to produce inflorescences. Since Abdalla and Mann (1963) report that the stem apex of an onion plant ceases to produce leaf primordia up to two weeks before bulb maturity, this could also be an effect of dormancy of the apical meristem.

Long photoperiods during chilling increase the rate of initiation; low nitrogen status increases the rate if temperature and photoperiod are marginal (Brewster, 1983). Emergence of inflorescences is inhibited by temperatures of 0°C or above 20°C (Heath and Mathur, 1944; Holdsworth and Heath, 1950); temperatures of 20°C and long-days can lead to death of developing inflorescences. Long-days and cool temperatures tended to accelerate emergence but also increased abortion of inflorescences (Brewster, 1982). Plants revert to vegetative growth in unfavourable conditions, or develop abnormal structures, such as bulbils or long leafy spathes, in the inflorescence (Roberts and Struckmeyer, 1951).

2.3.2 Development after emergence.

Brewster (1982a), using temperature-controlled glasshouses, found that development of the inflorescence after initiation was strongly dependent on temperature, and less strongly dependent on mean photoperiod. However, his experiment used only the natural progression of photoperiods experienced by the plants, and may not have given an accurate indication of the effects of photoperiod on all stages of development. His analysis is unusual in that each inflorescence is treated as an independent

estimator of the time to flowering. A more common approach to this type of problem would be to treat inflorescences on a given plant as correlated and use the mean for each plant as an estimator.

Incomplete development of flowers (Scully *et al.*, 1945) and development of topsets, or bulbils, in the inflorescence in short-days (Roberts and Struckmeyer, 1951) have been reported.

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Chapter 3

Effect of light quality and daylength

3.1 INTRODUCTION

Conditions for loss of dormancy in onion bulbs were characterised by Abdalla and Mann (1963), but conditions favouring sprouting in planted bulbs are not well known. Since sprouting involves growth of bladed leaves, it might be expected that the photomorphogenetic effect of photoperiod observed during bulbing might play a role (Section 2.2.4). If this is the case, sprouting and subsequent leaf growth would be expected to proceed faster in short-days (Section 2.2.1) and in light of a low red:far-red ratio (Section 2.2.2).

Floral initiation in onions requires a period of cold temperatures and normally takes place in winter (Holdsworth and Heath, 1950). If bulbs can be sprouted prematurely shortly after harvest in summer, it might also be possible to induce early floral initiation by subjecting sprouting plants to a cold treatment in autumn. This would enable plants to flower in winter, and, if conditions were favourable and pollinators available, to produce seed within a year of planting seed.

High temperatures (20-30°C), and daylengths which favour bulbing (generally days longer than 13 h), early in inflorescence development, suppress emergence and lead to abortion of the developing inflorescence (Holdsworth and Heath, 1950). Later in development, however, these conditions favour development of flowers (Brewster, 1982).

This experiment was undertaken to establish the kinds of variation likely to be encountered in different daylength environments; whether or not there was an effect of light quality; and to establish a time scale for the main experiment.

Bulbs were planted in autumn, when they would be expected to be breaking dormancy. After a six week sprouting period, a prolonged (14 weeks) chilling treatment was imposed, to maximise floral initiation. Temperatures were raised slowly, in 4°C steps, to minimise abortion of developing inflorescences. Once a set of favourable conditions had been established, a range of less favourable conditions could be tested in later experiments to determine how much the time from planting bulbs to flowering could be reduced, and whether the same responses were obtained from freshly harvested bulbs.

3.2 MATERIALS AND METHODS

Onion bulbs of four cultivars, 'Pukekohe Long Keeper', 'Early Long Keeper', 'Porter's Early Globe' and 'Yozui Yellow', previously grown in the field at Palmerston North, were used.

On 7 April 1986, bulbs were planted in six-inch pots by making a shallow depression in the potting medium. The potting medium used comprised Opiki loam, coarse sand and coarse pumice (4 : 3 : 1 v/v) with incorporated Nitrophoska fertiliser and dolomite lime (31.3 g l^{-1}). Pots were placed in reach-in controlled environment cabinets at Plant Physiology Division, DSIR. Plants were watered by hand twice weekly until sprouting, three times weekly during chilling, then daily until the experiment

Table 3.1: Numbers of bulbs of each cultivar per treatment.

Cultivar	Short-day/ low R:FR	Long-day/ low R:FR	Long-day/ high R:FR
'Pukekohe Long Keeper'	4	10	12
'Early Long Keeper'	4	5	3
'Porter's Early Globe'	4	2	4
'Yozui Yellow'	4	4	6

was terminated.

Three treatments, short-day/low R:FR (8 h days, z = 0.95), long-day/low R:FR (14 h days, z = 1.1), and long-day/high R:FR (14 h days, z = 6.4) were used. Red:far-red ratios were obtained as follows: z = 0.95, 4 x 700 W mercury vapour, 2 x 750 W tungsten halogen, and 8 x 65 W blue fluorescent lamps; z = 1.1, 6 x 400 W high pressure iodide and 2 x 1000 W tungsten halogen lamps; z = 6.4, 17 x 80 W cool white fluorescent tubes. Spectra were measured using a spectroradiometer (Model 740A, Optronic Laboratories Inc., Silver Spring, U.S.A.) with a cosine-corrected head and a 5 nm slit width. The red:far-red ratio, z, is the ratio of the photon flux density at 660 nm to that at 730 nm. Mean photosynthetic photon flux was 350 μmol m^{-2} s^{-1} except for the first 12 weeks in the long-day/low R:FR treatment, which had 760 μmol m^{-2} s^{-1}. All treatments were carried out at 20°C +/- 1°C, and humidity was uncontrolled.

Bulbs were allocated in varying numbers to each treatment, depending on the number of bulbs of that cultivar available (Table 3.1).

Additional bulbs of each cultivar, chosen at random, were dissected at intervals to check for signs of sprouting (in unsprouted bulbs) and of floral initiation.

Plants were kept in these conditions at 20°C for six weeks to determine the effect on sprouting, then the temperature was lowered to 8°C for floral initiation. After 14 weeks of chilling, the temperature was raised to 12°C for three weeks, then to 16°C for six weeks, then to 20°C for three weeks before the experiment was terminated.

Numbers of plants sprouted, numbers of plants with emerged inflorescences and with open flowers were determined weekly. Results were analysed using a χ^2 test of homogeneity (see Appendix).

3.3 RESULTS

Unsprouted bulbs of all cultivars in the short-day/low R:FR and long-day/high R:FR treatments showed green-tipped leaves inside the bulb 4 weeks after planting, as did 'Early Long Keeper' in the long-day/low R:FR treatment. However, 'Pukekohe Long Keeper' showed no sign of greening in the long-day/low R:FR treatment. Eight weeks after planting, green-tipped leaves were found inside unsprouted bulbs of all cultivars. Bulbs of 'Porter's Early Globe' and 'Yozui Yellow' were well rooted in all treatments. Two bulbs of 'Pukekohe Long Keeper', and one of 'Early Long Keeper', were only sparsely rooted, but others had many roots. The first sign of inflorescence initiation was found 12 weeks after planting (after 6 weeks chilling) in a bulb of 'Porter's Early Globe' under short-day/low R:FR conditions. At this time rooting in 'Pukekohe Long Keeper' and 'Early Long Keeper' was still variable; one of the bulbs selected for dissection had only two roots. After 13 weeks of chilling, spathes were formed in all of the bulbs that were dissected, and in many the scape had started to elongate. Where the scape was longer than 2 cm, the surface of the mantle was warty in appearance as floral primordia began to develop. Three plants without emerged inflorescences, one of 'Pukekohe Long Keeper' and two of 'Yozui Yellow', from each of the long-day treatments were dissected when the experiment was terminated. All three from the high R:FR treatment contained aborted inflorescences; one plant of 'Yozui Yellow' from the low R:FR treatment did not.

Table 3.2: Effect of light quality and photoperiod on sprouting of four onion cultivars

Cultivar	Fraction of plants sprouted in 6 weeks		
	Short-day/ low R:FR	Long-day/ low R:FR	Long-day/ high R:FR
'Pukekohe Long Keeper'	1.0	0.1	0.2
'Early Long Keeper'	0.5	0.3	0.8
'Porter's Early Globe'	0.6	1.0	0.7
'Yozui Yellow'	1.0	1.0	1.0

Overall there was a strong effect of treatment on number of plants sprouted in 6 weeks ($\alpha < 0.01$). More bulbs sprouted in the short-day treatment than in either of the long-day treatments. The small number of bulbs of each cultivar in each treatment means that the results for cultivars must be treated with caution. The only cultivar which showed differences between treatments was 'Pukekohe Long Keeper' (Table 3.2). No differences between cultivars were apparent in short-day conditions, but in long-days fewer bulbs of 'Pukekohe Long Keeper' and more bulbs of 'Yozui Yellow' sprouted than expected ($\alpha < 0.01$ for long-days/low R:FR, $\alpha < 0.025$ for long-days/high R:FR). All 'Yozui Yellow' bulbs sprouted in two weeks, but in long-day/low R:FR conditions three out of four plants were top down four weeks later. Plants in the other two treatments continued to produce green leaves. In the initial dissections at the start of the treatments, bulbs of 'Yozui Yellow' showed development of green shoots, whereas all other cultivars appeared completely dormant.

Inflorescences appeared earliest in the short-day/low R:FR treatment (Table 3.3), with only about two weeks separating the fastest and slowest cultivars. In this treatment all plants of all cultivars produced inflorescences. Both long-day treatments took longer, with greatest differences between cultivars in the long-day/low R:FR

Table 3.3: Effect of light quality and photoperiod on time to emergence of inflorescences, measured from the end of chilling at 8°C

Cultivar	Mean time to emergence (weeks)		
	Short-day low R:FR	Long-day low R:FR	Long-day high R:FR
'Pukekohe Long Keeper'	6.0	7.5	10.8
'Early Long Keeper'	6.0	7.0	10.0
'Porter's Early Globe'	5.8	11.0	10.0
'Yozui Yellow'	4.5	-	8.3

treatment (Table 3.3), where plants of 'Yozui Yellow' produced no emerged inflorescences, and 'Pukekohe Long Keeper' was the only cultivar where more than half the plants flowered. In the long-day/high R:FR treatment, half the plants of 'Yozui Yellow' and 'Porter's Early Globe', and one third of the plants of 'Pukekohe Long Keeper' and 'Early Long Keeper' produced emerged inflorescences.

Plants of 'Yozui Yellow' showed thickened leaf bases on flowering plants in both short- and long-days in low R:FR conditions, but not in high R:FR conditions.

3.4 DISCUSSION

The longer keeping cultivars of onion did show a response to daylength when sprouting, and 'Early Long Keeper' and 'Porter's Early Globe' also responded to light quality. This suggests a continuation of the photomorphogenetic response observed during bulbing. However, since 3-5 months elapsed between harvest and planting, the bulbs would have been expected to be breaking dormancy anyway. The responses

may have been different in bulbs planted soon after harvest.

Time to floral emergence of inflorescences was shortest in the short-day treatment (Table 3). Fewer plants produced inflorescences in the long-day treatments, where plants bulbed rapidly.

Light quality affected the degree of thickening of leaf bases, but had little effect on time to sprouting or on inflorescence emergence compared to the effect of photoperiod.

Both the numbers of inflorescences emerged and the mean times to emergence were affected by photoperiod, suggesting that the highest number of emerged inflorescences could be achieved most rapidly by keeping photoperiod short until the inflorescence was well developed.

A time scale for the main experiment was selected as follows. Bulbs would be maintained for 8 weeks in sprouting treatments of 15°C and 20°C in 8 and 14 h days, to allow extra time for freshly harvested bulbs, which may be more dormant, to respond. Since the 14 weeks of chilling at 8°C used in this experiment was much longer and more intense than would be experienced by plants in the field, three shorter periods of 4, 8 and 12 weeks would be evaluated, followed in each case by a period of 4 weeks in 15°C. Plants would then be transferred to post-chilling treatments of 15°C and 20°C in 8 and 14 h days.

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Chapter 4

Sprouting

4.1 INTRODUCTION

Bulbing in onions is controlled by photoperiod and temperature. In warm long-day conditions, assimilates are allocated to storage tissue formed in the growing parts of leaf sheaths, so that part or all of a sheath develops as a bulb scale. Growth of bladed leaves involves allocation of assimilates to structural tissue, forming leaf blades and long sheaths (Bertaud, 1986). Bulbs produce leaf primordia throughout storage at rates which vary with cultivar and storage conditions (Abdalla and Mann, 1963). In sprouting bulbs, the leaves which develop from these primordia develop the blades of green leaves rather than the storage tissue of bulb scales (Bertaud, 1986); blades and sheaths of such leaves elongate to protrude from the top of the bulb.

Evidence for the production and translocation to the apex of a growth inhibitor in mature onion bulbs during long-days (Isenberg *et al.*, 1974) indicates that onion bulbs may initially be in a state of *paradormancy* as defined by Lang *et al.* (1987). Eventually bulbs reach a state of *ecodormancy* (Lang *et al.*, 1987) where all they require for growth is the basic needs of water and nutrients. Bertaud (1986) hypothesised that the transition from a state of paradormancy and ecodormancy to

one of ecodormancy alone, as well as sprouting of ecodormant and non-dormant bulbs, is also photoperiod-dependent. A continuation of the photomorphogenetic response observed during bulbing ought to result in increased sprouting and subsequent leaf appearance under short-day conditions, since under short-days allocation of photosynthates to structural tissue rather than storage tissue would be expected. A preliminary experiment with small numbers of bulbs planted three to four months after harvest suggested that there was a response to photoperiod and light quality in some longer keeping cultivars at constant 20°C (Bertaud, 1988).

The objective of this experiment was to test the hypothesis that sprouting and leaf growth of freshly harvested bulbs should be affected by photoperiod and temperature. The results may also help to define the nature of dormancy and its relationship to the keeping quality of onions.

4.2 MATERIALS AND METHODS

Onion bulbs of three cultivars, 'Sakigake Yellow', 'Gladalan Brown' and 'Early Long Keeper', grown in the field at Crop Research Division, DSIR, Pukekohe, were used. Agronomic details for these plants are shown in Table 1.

Bulbs were planted in 15 cm pots by making a shallow depression in the potting medium. The potting medium used comprised Opiki loam, coarse sand and coarse pumice (4 : 3 : 1 v/v) with incorporated Nitrophoska fertiliser and dolomite lime (31.3 g l^{-1}).

Bulbs were placed in controlled environments (CE) after planting. Two walk-in CE rooms at the Climate Laboratory (Anon, 1981) and three reach-in CE cabinets were used to obtain the range of temperature and photoperiod treatments required. The Climate Laboratory rooms were run at 8°C and 15°C, both for 8 h days (chilling short-days and cool short-days), while the cabinets were run at 15°C for 14 h days (cool long-days), and at 20°C for 8 h and 14 h days (warm short-days and warm

Table 4.1: Development details for onion bulbs used in this study. All cultivars were sown on 9 May, 1986.

Cultivar	50% Top down	Lifting	Harvest	Planting
'Sakigake Yellow'	9 Nov 1986	19 Nov 1986	20 Nov 1986	2 Dec 1986
'Gladalan Brown'	27 Nov 1986	3 Dec 1986	22 Dec 1986	31 Dec 1986
'Early Long Keeper'	17 Dec 1986	19 Dec 1986	15 Jan 1987	30 Jan 1987

long-days). Temperature was controlled to +/- 0.5°C in CE rooms, and +/- 1.0°C in cabinets. Mean photosynthetic photon flux (PPF) was 558 μmol m^{-2} s^{-1} in the cabinets, 677 μmol m^{-2} s^{-1} in the 15°C room, and 474 μmol m^{-2} s^{-1} in the 8°C room, for the entire photoperiod. Relative humidity was 82% $\pm$ 5% at 15°C, and 87% $\pm$ 5% at 20°C, in the CE rooms, but uncontrolled in the cabinets. Plants were watered twice weekly until the bulbs sprouted, when the rate was increased as necessary.

Bulbs were divided into 2 size classes, assigned randomly to each treatment, and planted within a month of harvest (Table 4.1). Six bulbs of each cultivar were dissected at the start of the treatments to inspect for greening of leaf primordia, which appears to be a sign of growth resumption following dormancy (Bertaud, 1988).

Twenty bulbs of each cultivar were placed in each of the photoperiod x temperature treatments, except the cool short-day combination where 60 plants were used. Forty bulbs of 'Sakigake Yellow' were placed in the chilling short-day treatment, but no bulbs of 'Sakigake Yellow' were grown under cool long-days. Plants were scored weekly for number of leaves on each shoot. Plants of 'Early Long Keeper' were also scored for rooting at intervals during the experiment, and time to 50% plants rooted was calculated.

Data for sprouting were analysed by analysis of variance for categorical data (Grizzle et al., 1969), and cumulative normal distributions were fitted to the data for time to sprouting. Mean leaf numbers per shoot were regressed against time from sprouting for each treatment, and the regressions compared.

4.3 RESULTS

4.3.1 Sprouting

In initial dissections, no bulbs of any cultivar showed any greening of leaf primordia or other sign of sprouting. However, differences between cultivars were significant and increased with time ($\alpha < 0.001$ after 8 weeks).

There was no significant effect of photoperiod on sprouting of 'Sakigake Yellow' in warm temperatures. The first plants sprouted after approximately 4 weeks (Fig. 4.1). After 8 weeks in short-days, 83% of bulbs sprouted in cool and 85% in warm temperatures; in long-days, 75% of bulbs sprouted in 20°C (Fig. 4.1). Sprouting was, however, very slow in chilling (8°C) short-days where only 2 bulbs had sprouted after 8 weeks.

'Gladalan Brown' plants also started sprouting after approximately 4 weeks (Fig. 4.2). A significant effect of treatment was observed after 8 weeks (Fig. 4.2). In cool temperatures, 97% of bulbs had sprouted in short-days and 45% in long-days, while in warm temperatures the numbers were 70% and 40%, respectively. Analysis of variance showed a significant effect of photoperiod from the sixth week ($\alpha < 0.01$) that increased over time (Fig. 4.2). After 8 weeks an effect of temperature was almost significant ($\alpha < 0.08$), as a result of separation of the short-day treatments (Fig. 4.2); this may indicate a trend that would be apparent if plants had been observed for longer. Some bulbs in the warm long-day treatment swelled and split into two or three smaller bulbs without sprouting (Fig. 4.3).

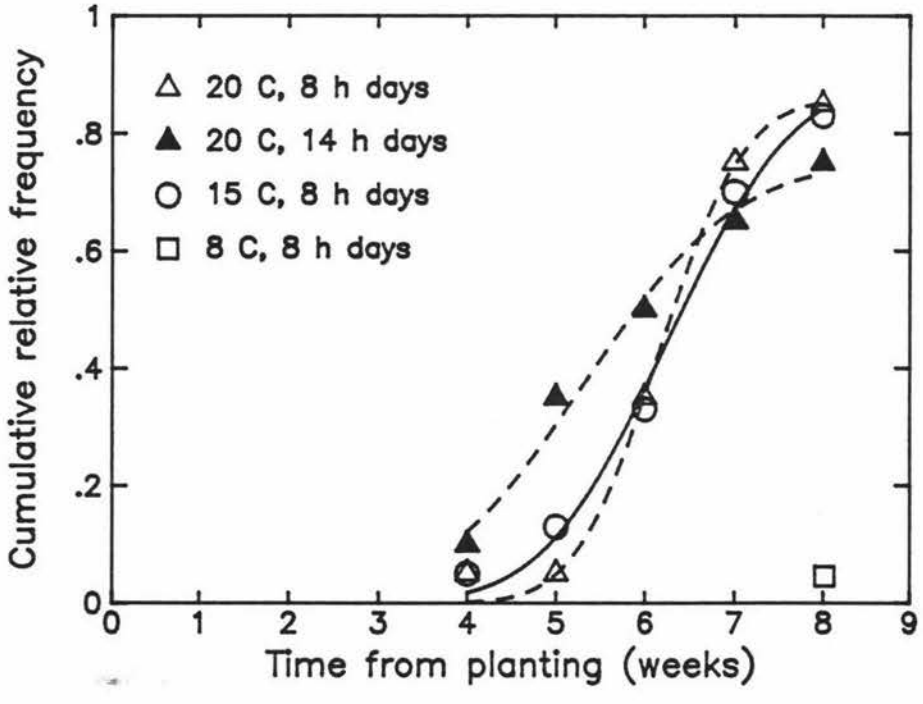

Figure 4.1: Cumulative relative frequency of times to sprouting for 'Sakigake Yellow'. Lines are fitted cumulative normal distributions.

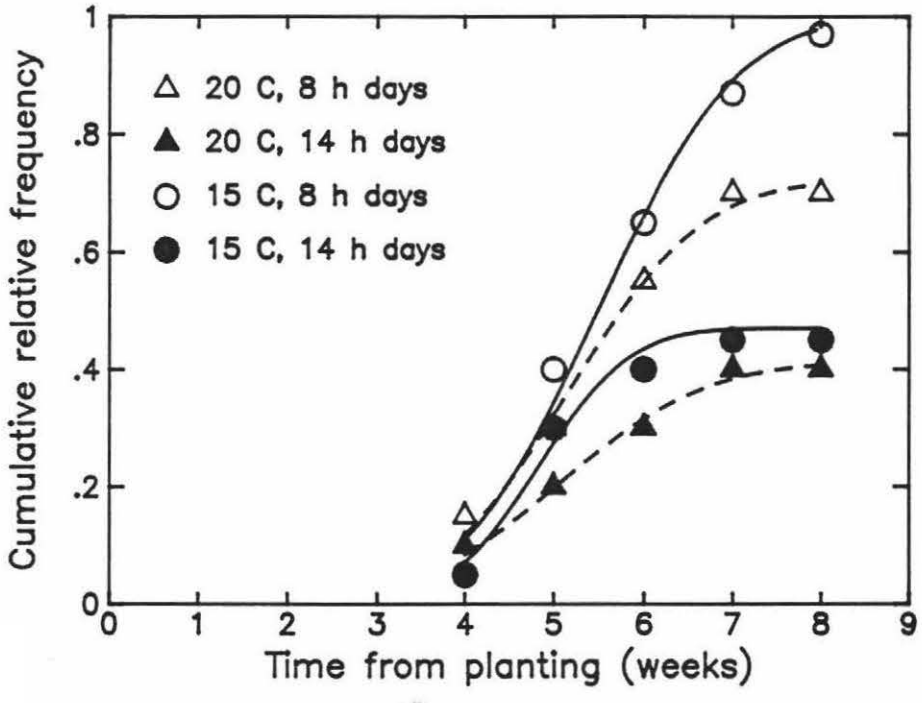

Figure 4.2: Cumulative relative frequency of times to sprouting for 'Gladalan Brown'. Lines are fitted cumulative normal distributions.

Figure 4.3: 'Gladalan Brown' bulbs in 20°C and 14 h days. Note swelling and splitting of unsprouted bulbs as 'daughter' bulbs are produced.

No significant treatment effects were observed on 'Early Long Keeper'. Bulbs were slow to root; time to 50% rooting was 5 weeks in cool short-days, and 8 weeks in both warm treatments. The cool long-day treatment had not reached 50% rooting after 12 weeks. Sprouting was also delayed. The first sprouts appeared after 5 weeks in cool short-days, warm short-days and warm long-days; none had appeared after 12 weeks in cool long-days. In 8 weeks, at cool temperatures, 13% of bulbs sprouted in short-days but none in long-days; at warm temperatures, the numbers were 5% and 17%, respectively.

4.3.2 Leaf appearance

Sprouted plants showed no obvious apical dominance. In most plants a number of shoots of similar size emerged from the bulb in a short time. These shoots generally remained of similar size throughout the experiment, and the total number of leaves on a plant was directly proportional to the number of shoots. Mean leaf numbers per shoot were calculated by dividing the total number of leaves by the number of shoots visible the following week, since shoots could not be separately identified until the leaf sheaths emerged.

Sprouted plants showed differences in rate of leaf appearance with treatment. Quadratic equations were fitted to data for 'Sakigake Yellow' because there was evidence of a non-linear response in 14 h days (Fig. 4.4). Plants in both cool and warm short-days were not significantly different ($\alpha < 0.05$), and were described by the regression equation

$$L = 2.15 + 1.01t - 0.016t^2,$$

where L is the number of leaves and t is time in weeks. The coefficient of t^2 was not significantly different from zero. In warm long-day conditions the regression equation was

$$L = 1.72 + 1.41t - 0.17t^2,$$

The coefficient of t^2 was more significant ($\alpha < 0.075$), and much larger, under

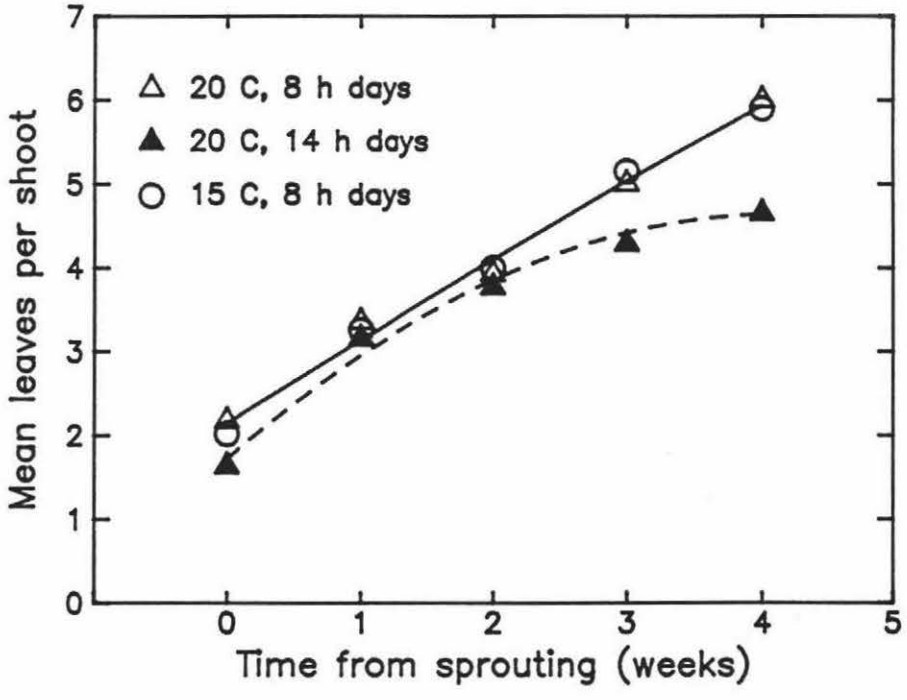

Figure 4.4: Mean leaf number per shoot with time for 'Sakigake Yellow'. Lines are fitted quadratic regressions.

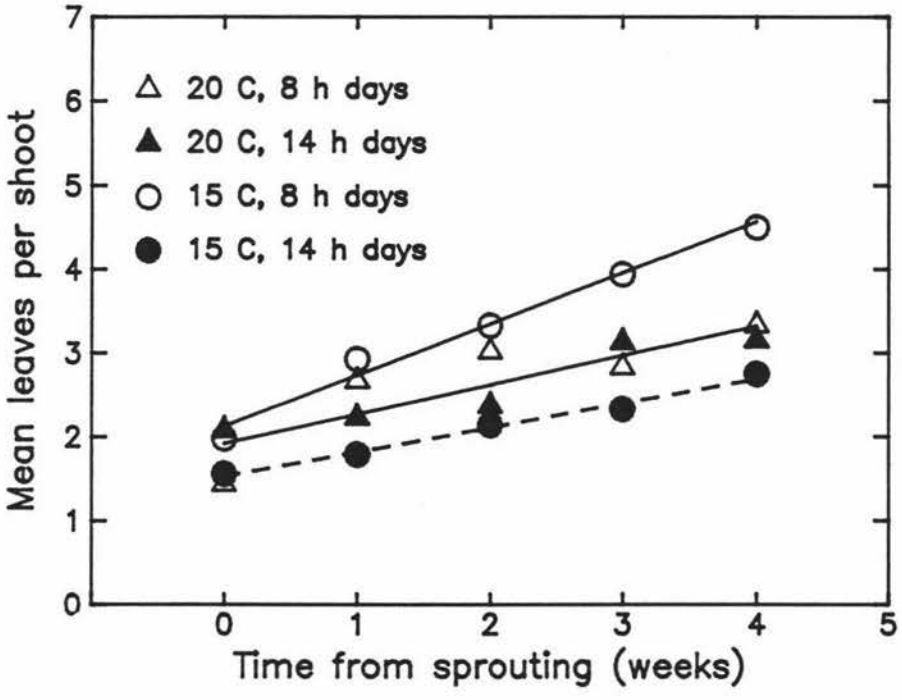

Figure 4.5: Mean leaf number per shoot with time for 'Gladalan Brown'. Lines are fitted linear regressions.

these conditions, reflecting the plant response; at 5 weeks from sprouting many of the plants had stopped producing bladed leaves altogether, formed new bulbs, and in some cases the tops had fallen over. 'Gladalan Brown' showed no such behaviour; linear regressions accounted for the leaf appearance observed. Rates were significantly different ($\alpha < 0.05$) for both cool temperature treatments; the rates were 0.61 leaves week^{-1} in short-days and 0.29 leaves week^{-1} in long-days (Fig. 4.5). The warm treatments had the same rate, 0.35 leaves week^{-1}.

So few plants of 'Early Long Keeper' sprouted that it was not possible to test for significant differences.

4.4 DISCUSSION

4.4.1 Sprouting

Dormancy is defined as a state of 'no visible growth' by Lang *et al.* (1987). Salisbury and Ross (1985) and more recently Lang *et al.*, (1987) differentiate between dormancy caused by lack of basic necessities for growth such as water or light, which may be referred to as *ecodormancy* (Lang *et al.*, 1987), and dormancy caused by specific physiological responses to environmental factors such as photoperiod or chilling, which Lang *et al.*, (1987) refer to as *endodormancy* when the stimulus is perceived by the dormant meristem itself, and as *paradormancy* when it is perceived by other organs such as leaves. This terminology may be applied to seeds or buds.

An onion bulb is a whole plant and therefore has both shoot meristems and root meristems. Since roots formed by a bulb are adventitious, it seems likely that their emergence is a sign that growth is possible. Lack of sprouting in rooted plants therefore suggests a specific response to environmental conditions. If shoot and root meristems responded in the same way, sprouting would be expected to become visible later than rooting because although both sprouting and rooting involve the elongation of preformed initials, the sprout has to grow upward through the bulb before

emerging. Because the shoot meristem(s) of a bulb are hidden by a number of bulb scales, which may total some centimetres in thickness and in height, growth may proceed for some time before becoming visible. Bulbs that grow without sprouting, such as those observed in long-days in 'Gladalan Brown', are not dormant. Classification of plants that are rooted, but not producing either bladed leaves or bulb, is more difficult, and raises the question of whether the bulb should be treated as a whole, or dormancy of shoot and root meristems should be considered separately. Some of the evidence produced here suggests that separate treatment of root and shoot meristems would be appropriate under some conditions.

Onion bulbs of cultivars 'Excel' and 'Australian Brown 5' have been reported to go through a period when they will not sprout, followed by a period when bulbs can sprout in favourable conditions (Abdalla and Mann, 1963). Evidence for translocation of a growth inhibitor from leaves into the bulb (Isenberg *et al.*, 1974) suggests that in the initial period the bulb is in a state of paradormancy. At some stage this dormancy disappears and the bulb becomes simply ecodormant. An ecodormant bulb will root if planted in moist conditions, but may or may not sprout subsequently. If left long enough, it will sprout whether planted or not. Two points remain to be resolved here; what controls the transition to a state of ecodormancy alone, and what controls sprouting (i.e., leaf elongation) of the formerly ecodormant bulb, once it has produced roots.

Sprouting is a result of at least two major changes in the onion apex and associated leaf primordia. The first is mobilisation of assimilates for leaf growth and for root growth (where the base plate is kept moist). Leaves and roots may not become active sinks simultaneously; Abdalla and Mann (1963) observed a decrease in the interval between rooting and sprouting of bulbs of cultivars 'Excel' and 'Australian Brown 5' with increasing time of storage, and bulbs of 'White Creole' did not sprout if their roots were removed as they appeared. Abdalla and Mann (1963) suggest that sprouting is stimulated by a growth hormone produced in the roots. The second change is partitioning of assimilates to structural carbohydrate, forming bladed leaves, rather than storage carbohydrate of scale leaves. Development of leaf

primordia initiated before harvest as bladed leaves instead of scale leaves leads to bulb sprouting (Abdalla and Mann, 1963). Partitioning in some cultivars responds strongly to photoperiod during bulbing (de Ruiter, 1986). In warm long-day conditions, some plants showing evidence of metabolic activity were continuing to allocate assimilates to storage tissue in new leaves (Fig. 2), suggesting that partitioning in rooted, growing bulbs also responds to photoperiod.

Heath and Holdsworth (1948) reported that onions stored for a year in the dark produced leaves with shortened blades, and suggested that this might be due to secondary bulbing. Such leaves were observed in all treatments of both experiments in the present work, where no bulbs were stored for longer than a month. Bertaud (1986) suggests that this is a result of leaf tips maturing under conditions conducive to bulbing, so that subsequent growth of the blade under non-bulbing conditions is limited.

The longer bulbs are stored, the less time they take to sprout if planted in moist conditions, though the actual times to sprouting are longer for long-keeping than for poor-keeping cultivars (Abdalla and Mann, 1963). Temperature of storage also affects time to sprouting; longer times to sprouting at 0°C and 30°C occurred in parallel with a decrease in mitosis and in rate of leaf initiation at the apex at those temperatures (Abdalla and Mann, 1963). These results suggest a temperature-sensitive dormancy period after harvest.

Sensitivity to photoperiod during bulbing appears to vary between cultivars (de Ruiter, 1986). 'Yozui Yellow', which like 'Sakigake Yellow' is an early maturing, poor-keeping cultivar of Japanese origin, appears to be relatively insensitive to photoperiod during bulbing (de Ruiter, 1986). This insensitivity could result in partitioning of assimilates to structural carbohydrates in long days, forming bladed leaves, and thus accounting for the sprouting response of these cultivars. 'Yozui Yellow' bulbs used in a preliminary experiment sprouted in 2 weeks (Bertaud, 1988), but the difference between 'Sakigake Yellow' and 'Yozui Yellow' in initial sprouting time can probably be accounted for by the fact that in the preliminary experiment,

'Yozui Yellow' bulbs dissected before planting were already forming green leaves.

Sprouting in 'Gladalan Brown' was clearly influenced by photoperiod. Long-day treatments resulted in rooted bulbs either showing no visible signs of growth, or forming new bulbs within the old one without leaf growth. Bulbs of both 'Sakigake Yellow' and 'Gladalan Brown' rooted and sprouted rapidly in short-days at both cool and warm temperatures, indicating that they were ecodormant when planted, and this dormancy was broken by contact with moist soil.

Rooting was very slow in 'Early Long Keeper', although bulbs were planted in moist potting medium, suggesting that most bulbs had not yet become ecodormant. Even long keeping cultivars generally root if the base plate is kept moist (Abdalla and Mann, 1963), and bulbs of *Allium moly* planted in moist soil will produce roots in April in Palmerston North, though leaves do not appear until September. The slowness of rooting in 'Early Long Keeper', and the different times to 50% plants rooted, suggest that the inability of this cultivar to grow is a more specific form of dormancy. Since sprouting is so slow, a longer experiment with larger numbers of plants would be needed to characterise the response. In a preliminary experiment, where bulbs were planted two months later, faster sprouting was observed, implying faster rooting (Bertaud, 1988).

Rooting was studied by Abdalla and Mann (1963) using cultivar 'Excel', a poor-keeping cultivar. Fifty percent of bulbs planted immediately after harvest rooted in 9 days, while 50% of bulbs stored for 16 weeks were rooted in 2 days. These results are comparable with the observation that cultivars 'Sakigake Yellow' and 'Gladalan Brown' rooted in 10 days.

A number of attempts have been made to influence the length of the dormant period in onions (Brewster, 1977b). Sprays of synthetic auxins or shading, applied before harvest, have been reported to reduce the dormant period (Brewster, 1977b). Injections of NAA, GA3 or GA4/7 failed to stimulate sprouting (it is in fact very difficult to inject anything into an onion bulb, which is a solid structure with few airspaces).

However, wounding of bulbs, and removal of outer scales, have been reported to stimulate sprouting (Brewster, 1977b).

4.4.2 Leaf appearance

Rates of leaf appearance were affected by photoperiod; in 'Sakigake Yellow' at 20°C and 'Gladalan Brown' at 15°C. In both cases leaf appearance was faster in short-days. Slower rates observed in long-day conditions, and the cessation of leaf appearance observed in 'Sakigake Yellow', demonstrate the photomorphogenetic response observed in bulb development. The intercepts of the fitted regression lines are not zero; this reflects the fact that when a bulb sprouts, a number of leaves appear at the same time.

4.5 CONCLUSIONS

The hypothesis that sprouting of onion bulbs should be affected by photoperiod appears to be correct for cultivar 'Gladalan Brown'. However, in some cultivars there is clearly another mechanism involved in addition to the photoperiod response. It appears that two factors stop an onion bulb sprouting after the initial formation of scale leaves: whether or not the bulb has become ecodormant, indicated by the ability of bulbs to develop roots; and a photomorphogenetic response to long-days. 'Sakigake Yellow' and 'Gladalan Brown' were both ecodormant when planted, but sprouting is sensitive to photoperiod only in 'Gladalan Brown'. However, subsequent leaf appearance is affected by photoperiod in both cultivars. In 'Early Long Keeper', bulbs are not simply ecodormant a month after harvest, and there may be a photoperiod response involved in subsequent sprouting. A two-stage response of this sort also explains the inverse relationship of keeping quality to maturity. If dormancy in onions was entirely a response to photoperiod, it might be expected that late maturing cultivars such as 'Early Long Keeper', which require very long-days for bulbing, would be the poorest keepers, since days long enough (about 14 h) to

maintain the state of paradormancy of the bulb only last for a short period in the summer.

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Chapter 5

Flowering

5.1 INTRODUCTION

Onion bulbs planted in the field for seed flower in late spring and produce seed in summer. Under New Zealand conditions this seed cannot be sown immediately as the cool temperatures of winter increase the risk of bolting in the crop. This results in a generation time (seed to seed) of 2 years. Cultivars also flower at different times, which means some cultivars are difficult to incorporate into breeding programmes. Present methods for reducing the generation time to one year involve growing the plants continuously but without the formation of a bulb which can be harvested and assessed (Brewster, 1982). If it were possible to induce onions to flower earlier, using controlled environments, it might be practicable to produce bulbs in the field, assess them for characteristics such as colour, hardness or flavour (but not storage quality), plant them in modified environments in the summer, and produce seed in time for planting late the following winter. Bulb storage quality could be assessed concurrently with seed production.

A study of sprouting onion bulbs in controlled conditions was described (Bertaud, 1988). In this chapter the effects of duration of chilling, and of subsequent temperature and photoperiod treatments on inflorescence initiation and development are

described.

Onion flowering physiology was reviewed by Brewster (1977) and more recently by Rabinowitch (1985). Onion plants require a period of low temperature to initiate inflorescences (Heath and Holdsworth, 1950; Brewster, 1977). For temperate cultivars, inflorescence initiation is most rapid in the 7-13°C range; cultivars grown in Northern Russia require 4-6°C (Brewster, 1986), while varieties grown in tropical Africa are reported to initiate inflorescences in the 15-20°C temperature range (Sinnadurai, 1970). Temperatures of 25-30°C and long-days can suppress emergence and cause death of a developing inflorescence (Holdsworth and Heath, 1950). However, development of the emerged inflorescence and seeds is advanced by temperatures of 25-30°C and long-days (Brewster, 1982).

Brewster (1982), using temperature-controlled glasshouses, found that development of the inflorescence after initiation was strongly dependent on temperature, and less strongly dependent on mean photoperiod. However, his experiment used only the natural progression of photoperiods experienced by the plants, and may not have given an accurate indication of the effects of more extreme photoperiods or the effect of photoperiod on different stages of development.

The objectives of this work were (a) to assess whether plants with green leaves and mature bulbs responded to chilling in the same way as unsprouted and sprouted bulbs; and (b) to explore variation between cultivars in sensitivity to duration of chilling, and to subsequent photoperiod and temperature treatments, during inflorescence initiation, emergence and flowering. From these results too it was expected that a sequence of controlled environment treatments could be defined that would allow (a) a reduced generation time, and (b) a range of cultivars to be brought into flower at the same time. The three cultivars used were 'Gladalan Brown', which is susceptible to bolting; 'Sakigake Yellow', which is resistant to bolting in the first year but flowers readily thereafter; and 'Early Long Keeper', which is intermediate in behaviour in its first year, but often flowers late or not at all in the second year, when it is very slow to sprout (D. Grant, pers. comm.).

5.2 MATERIALS AND METHODS

Bulbs were planted in 15 cm pots by making a shallow depression in the soil. The potting medium used comprised Opiki loam, coarse sand and coarse pumice (4 : 3 : 1 v/v) with incorporated Nitrophoska fertiliser and dolomite lime (31.3 g l^{-1}).

Bulbs were placed in controlled environments (CE) after planting. Two walk-in CE rooms at the Climate Laboratory (Anon, 1981) and three reach-in CE cabinets were used to obtain the range of temperature and photoperiod treatments required. The Climate Laboratory rooms were run at 8°C, and 15°C, both for 8 h days, while the cabinets were run at 15°C for 14 h days, and at 20°C for 8 h and 14 h days. Temperature was controlled to +/- 0.5°C, in CE rooms, and +/- 1.0°C, in cabinets. Mean photosynthetic photon flux (PPF) was 558 μmol m^{-2} s^{-1} in the cabinets, 600 μmol m^{-2} s^{-1} in the 15°C room, and 474 μmol m^{-2} s^{-1} in the 8°C room, for the entire photoperiod. Mean red to far-red ratio was 1.03, except for the cool long-day treatment which had a ratio of 2.14. Relative humidity was 82% $\pm$ 5% at 15°C, and 87% $\pm$ 5% at 20°C, in the CE rooms, but uncontrolled in the cabinets. Plants were watered twice weekly until the bulbs sprouted, when the rate was increased as necessary to avoid stress. Four months after the start of the experiment, a modified Hoaglands-type nutrient solution was substituted for water, to supplement the fertiliser incorporated in the growing medium.

Sixty bulbs each of cultivars 'Sakigake Yellow', 'Gladalan Brown', and 'Early Long Keeper' were sprouted for 8 weeks in 8 h days at 15°C (Bertaud, 1988) before transfer to chilling conditions. Bulbs were divided into 2 size classes, assigned randomly to each treatment, and planted within a month of harvest. In addition, 60 unsprouted bulbs of 'Sakigake Yellow' were planted in pots and placed directly into 8°C chilling conditions, along with 58 plants of 6 cultivars that had formed bulbs, but still had green leaves, which in some cases were still upright. Before planting, bulbs were divided into three groups of 20, which were to be chilled at 8°C in 8 h days for 4, 8, and 12 weeks (Fig. 5.1); these groups were divided between 4 post-chilling

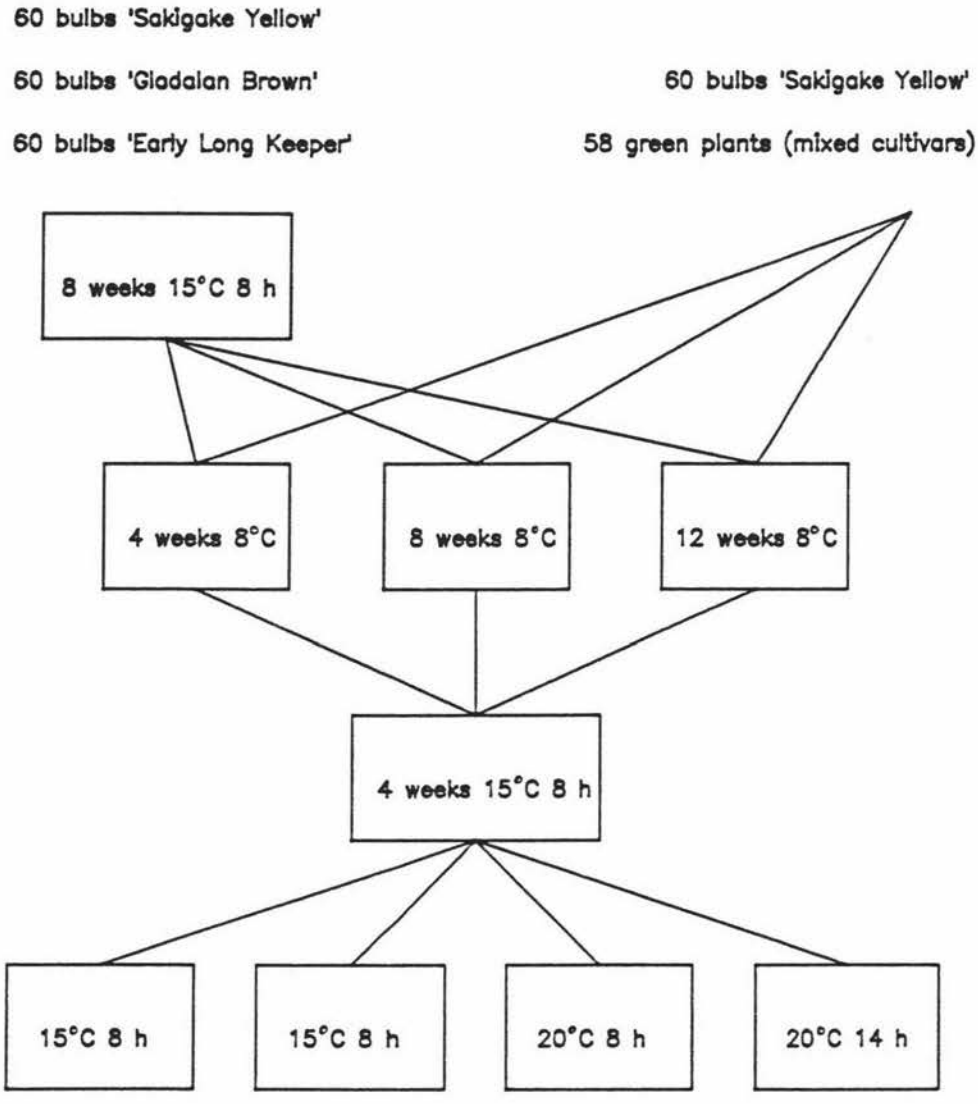

Figure 5.1: Flow chart of protocol for flowering experiment

treatments of 15°C/8 h (cool short-days), 15°C/14 h (cool long-days), 20°C/8 h (warm short-days) and 20°C/14 h (warm long-days) (Fig. 5.1) to give five plants per treatment in a 3 x 4 factorial design. The green plants were also divided between these 12 treatments, 5 plants going into each of 10 treatments and 4 each into the remaining 2, so that any treatment had one plant of each of 4 or 5 cultivars. After chilling, all plants were maintained for 4 weeks in cool short-day conditions, before being transferred to the appropriate post-chilling treatment (Fig. 5.1).

Numbers of leaves and stage of inflorescence development were recorded for each tiller on each plant at weekly intervals. Plants in the post-chilling treatments that did not produce inflorescences were dissected after three months (in the short-day treatments) or after all shoots reached top-down (in the long-day treatments) to check for inflorescence initiation. Plants were classified by the most advanced growth stage reached: 0 (no initiation), 1 (at least one inflorescence initiated), 2 (at least one inflorescence emerged) or 3 (at least one inflorescence flowered). Times of emergence of the inflorescence from the sheath of the subtending leaf, of opening of inflorescences (spathe rupture) and of first floret anthesis were recorded. The number of florets per inflorescence was determined for cultivar 'Early Long Keeper'.

Sample sizes were restricted by the number of plants that could be fitted into a CE room (144) or cabinet (32). The sample size of 5 was chosen because work by Brewster (1982a, b) with sample sizes as small as 2 plants produced satisfactory SEDs (see Section 5.4, Discussion).

Effects of treatments on stage of floral development were analysed using analysis of variance for categorical data and χ^2 tests (see Appendix). To analyse the effect of treatment on initiation, growth stages 1, 2 and 3 were pooled and compared to growth stage 0. For emergence, growth stages 0 and 1 were pooled and compared with pooled data for 2 and 3, while for flowering stages 0, 1 and 2 were pooled and compared to stage 3. When tables were drawn up for individual cultivars, a number of cells had fewer than five values. When combined with the small sample size, this meant that non-significant results were often obtained from the analysis of variance.

In cases where this happened, χ^2 tests were used to analyse for main effects.

Effects of photoperiod, temperature and chilling duration on times to floral emergence and opening were analysed using generalised linear models (see Appendix). Times were measured from the transfer from chilling to cool short-day conditions. Plants which initiated inflorescences that did not emerge, or that failed to flower, were defined as having infinite times to emergence or from emergence to flowering. The analysis was carried out on the reciprocals of times to emergence and from emergence to flowering, which are referred to as rate to emergence and rate to flowering.

Floret counts were analysed using generalised linear models.

5.3 RESULTS

Plants in the warm short-day treatment were infected with *Fusarium* species (M. Christensen, pers. comm.) and became infested with thrips. Both these organisms are recognised pathogens of onions in the field (Jones and Mann, 1963), although *Fusarium* species do not seem to be regarded as serious pests under New Zealand conditions (Fullerton *et al.*, 1986). Most plants in this treatment had to be discarded before flowering. As a result, it is not possible to test for interactions between photoperiod and temperature for number of plants that reached flowering or for time to flowering. However, effects of photoperiod in cool temperatures, and of temperature in long-days, can be assessed.

5.3.1 Floral initiation

Analysis of variance produced very large standard errors and indicated neither temperature, photoperiod, chilling duration nor cultivar had a significant effect on initiation. Analysis of main effects using the χ^2 test indicated that when results for all

cultivars were combined, chilling and temperature could have significant effects on the results ($\alpha < 0.05$). Cultivars were also significantly different ($\alpha < 0.05$), and as differences between these cultivars are significant in the field, this result should be reliable.

All plants of 'Gladalan Brown' initiated inflorescences, regardless of chilling duration (Table 5.1). A small number of plants of 'Early Long Keeper' that experienced only 4 weeks chilling did not initiate inflorescences, but the effect of duration of chilling was not significant ($\alpha < 0.05$). 'Sakigake Yellow', however, showed some effect of duration of chilling (Table 5.1); 8 out of 19 plants given 4 weeks chilling did not initiate inflorescences.

Large standard errors meant that the analysis of variance showed no significant effects on unsprouted bulbs of 'Sakigake Yellow' or on green plants. Tests of main effects showed that temperature, photoperiod, and chilling duration were all effective when results for sprouted and unsprouted plants of 'Sakigake Yellow' and for green plants were combined ($\alpha < 0.05$). Unsprouted bulbs and green plants were significantly different from sprouted plants of 'Sakigake Yellow' ($\alpha < 0.05$); most bulbs and green plants did not initiate inflorescences. Only after 12 weeks chilling did 50% of unsprouted bulbs of 'Sakigake Yellow' initiate inflorescences (Table 5.1), and only when subsequent treatments involved cool temperatures. Green plants showed no consistent trend in effect of chilling (Table 5.1), although over 80% of plants initiated inflorescences after 12 weeks. Overall, the effect of chilling duration was only apparent when subsequent floral development took place at warm temperatures, when inflorescences failed to initiate (Table 5.2), or, when initiated, failed to emerge. Plants exposed to cool short-days showed no effect of duration of chilling. Chilling duration had a significant effect on subsequent floral development in both short- and long-days.

Table 5.1: Effect of chilling on stage of floral development. Percentages of the total are given for each treatment.

Cultivar	Duration of chilling (weeks)	Stage of floral development 0[1]	1[2]	2[3]+ 3[4]	Number of plants
'Early Long Keeper'	4	16.7	16.7	66.7	18
	8	0.0	17.7	82.3	17
	12	0.0	12.5	87.5	16
'Gladalan Brown'	4	0.0	27.8	72.2	18
	8	0.0	6.3	93.8	16
	12	0.0	0.0	100.0	18
'Sakigake Yellow'	4	42.1	31.6	26.3	19
	8	0.0	10.5	89.5	19
	12	0.0	5.3	94.7	19
Unsprouted bulbs of 'Sakigake Yellow'	4	72.0	8.0	20.0	25
	8	69.6	8.7	21.7	23
	12	52.6	21.1	25.3	19
Green plants (mixed cultivars)	4	47.4	26.3	25.3	19
	8	65.0	5.0	30.0	20
	12	17.7	23.5	58.8	17

[0]0: no floral initiation

[1]1: at least one inflorescence initiated

[2]2: at least one inflorescence emerged

[3]3: at least one inflorescence with open flowers

Table 5.2: Effect of subsequent temperature on manifestation of effect of chilling duration (both photoperiods and all cultivars included). Percentages of the total are given for each treatment.

Temperature (°C)	Duration of chilling (weeks)	Stage of floral development 0[1]	1[2]	2[3]+ 3[4]	Number of plants
15	4	3.5	20.7	75.9	29
	8	0.0	13.8	86.2	29
	12	0.0	11.5	88.5	26
20	4	38.5	30.8	30.8	26
	8	0.0	8.7	91.3	23
	12	0.0	0.0	100.0	27

5.3.2 Inflorescence emergence

Differences between cultivars were not significant, but are described here because of their field importance.

Temperature (Table 5.4) and chilling duration (Table 5.1) appeared as significant factors in both the analysis of variance ($\alpha < 0.05$) and the χ^2 tests. Chilling duration interacted significantly with temperature, photoperiod, and cultivar ($\alpha < 0.05$). Inflorescences were more likely to emerge in cool temperatures and after a longer period of chilling, but the effect of chilling duration only manifested itself in warm temperatures (Table 5.2), and to a lesser extent in long-days. Chilling duration had much more of an effect on emergence in 'Sakigake Yellow' and 'Gladalan Brown' than in 'Early Long Keeper' (Table 5.1).

[0] 0: no floral initiation

[1] 1: at least one inflorescence initiated

[2] 2: at least one inflorescence emerged

[3] 3: at least one inflorescence with open flowers

Unsprouted bulbs and green plants showed no significant effects in analysis of variance, but χ^2 tests of main effects showed bulbs and green plants differed from sprouted plants of 'Sakigake Yellow', responding to temperature, photoperiod and chilling duration.

Table 5.3: Effect of photoperiod on stage of floral development at 15°C. Percentages of the total are given for each treatment.

Cultivar	Photoperiod	Stage of floral development				Number
		0[1]	1[2]	2[3]	3[4]	of plants
'Early Long	8	0.0	0.0	64.3	35.7	14
Keeper'	14	0.0	46.7	6.7	46.7	15
'Gladalan	8	0.0	0.0	45.5	54.6	11
Brown'	14	0.0	0.0	13.3	86.7	15
'Sakigake	8	0.0	0.0	13.3	86.7	15
Yellow'	14	7.1	42.9	0.0	50.0	14

[0] 0: no floral initiation

[1] 1: at least one inflorescence initiated

[2] 2: at least one inflorescence emerged

[3] 3: at least one inflorescence with open flowers

5.3.3 Flowering

Analysis of variance indicated that none of the main effects were significant, but that there was a significant interaction between photoperiod and cultivar ($\alpha < 0.05$). Inspection of Table 5.3 shows that, at 15°C, photoperiod had little effect on flowering in 'Early Long Keeper', but that flowering was improved by long-days in 'Gladalan Brown', and by short-days in 'Sakigake Yellow'. Effects of chilling duration on cultivar approached significance ($\alpha < 0.05$).

Tests of main effects using χ^2 showed that there could also be effects of chilling duration and cultivar.

Table 5.4: Effect of temperature on stage of floral development in 14 h days. Percentages of the total are given for each treatment.

Cultivar	Temperature	Stage of floral development				Number
	(°C)	0[1]	1[2]	2[3]	3[4]	of plants
'Early Long	15	0.0	46.7	6.6	46.7	15
Keeper'	20	14.3	7.1	14.3	64.3	22
'Gladalan	15	0.0	0.0	13.3	86.7	15
Brown'	20	0.0	30.8	7.7	61.5	13
'Sakigake	15	0.0	0.0	13.3	86.7	15
Yellow'	20	7.1	42.9	0.0	50.0	14

Analysis of variance showed no significant effects on bulbs or green plants. Tests of main effects showed that sprouted plants of 'Sakigake Yellow' differed from bulbs and green plants, most of which produced no flowers. An effect of photoperiod

[0] 0: no floral initiation

[1] 1: at least one inflorescence initiated

[2] 2: at least one inflorescence emerged

[3] 3: at least one inflorescence with open flowers

was apparent in cool temperatures. Unsprouted bulbs of 'Sakigake Yellow' only produced emerged inflorescences when subsequent growth took place in cool short-days (Table 5.1). Of 23 plants that initiated inflorescences, only 15 plants produced emerged inflorescences.

5.3.4 Rate to emergence and from emergence to flowering

Treating plants that did not produce emerged inflorescences as having infinite times to emergence, or zero rates to emergence, means that mean rates to emergence for some treatments are very low. These low rates do not reflect the actual behaviour of the inflorescences that did emerge, but rather the unfavourable effect of the treatment on emergence.

Cultivar differences were significant for rate to emergence, but not for rate from emergence to flowering. Inflorescences in all varieties emerged faster after a longer period of chilling ($\alpha < 0.05$). In 'Early Long Keeper', rate to emergence was slightly faster in warm than in cool temperatures (Fig. 5.2a; $\alpha < 0.05$). Photoperiod had no effect on rate to emergence. Rate to emergence in 'Gladalan Brown' was affected by chilling duration ($\alpha < 0.001$), but not by photoperiod. Temperature interacted with chilling duration; when subsequent growth took place in cool temparatures there was little response to duration of chilling, while in warm temperatures rate to emergence was significantly faster after longer periods of chilling (Fig. 5.3a). Rate to emergence in 'Sakigake Yellow' responded to chilling duration ($\alpha < 0.05$), but not to photoperiod or temperature (Fig. 5.4a). However, there were significant interactions between chilling and photoperiod, and between chilling and temperature ($\alpha < 0.001$).

Rate from emergence to flowering showed strong effects of chilling duration (reflecting higher numbers of aborted inflorescences after short chilling durations) and of photoperiod (in both cases $\alpha < 0.001$). The interaction between these two factors was also highly significant ($\alpha < 0.001$) reflecting higher rates of abortion in

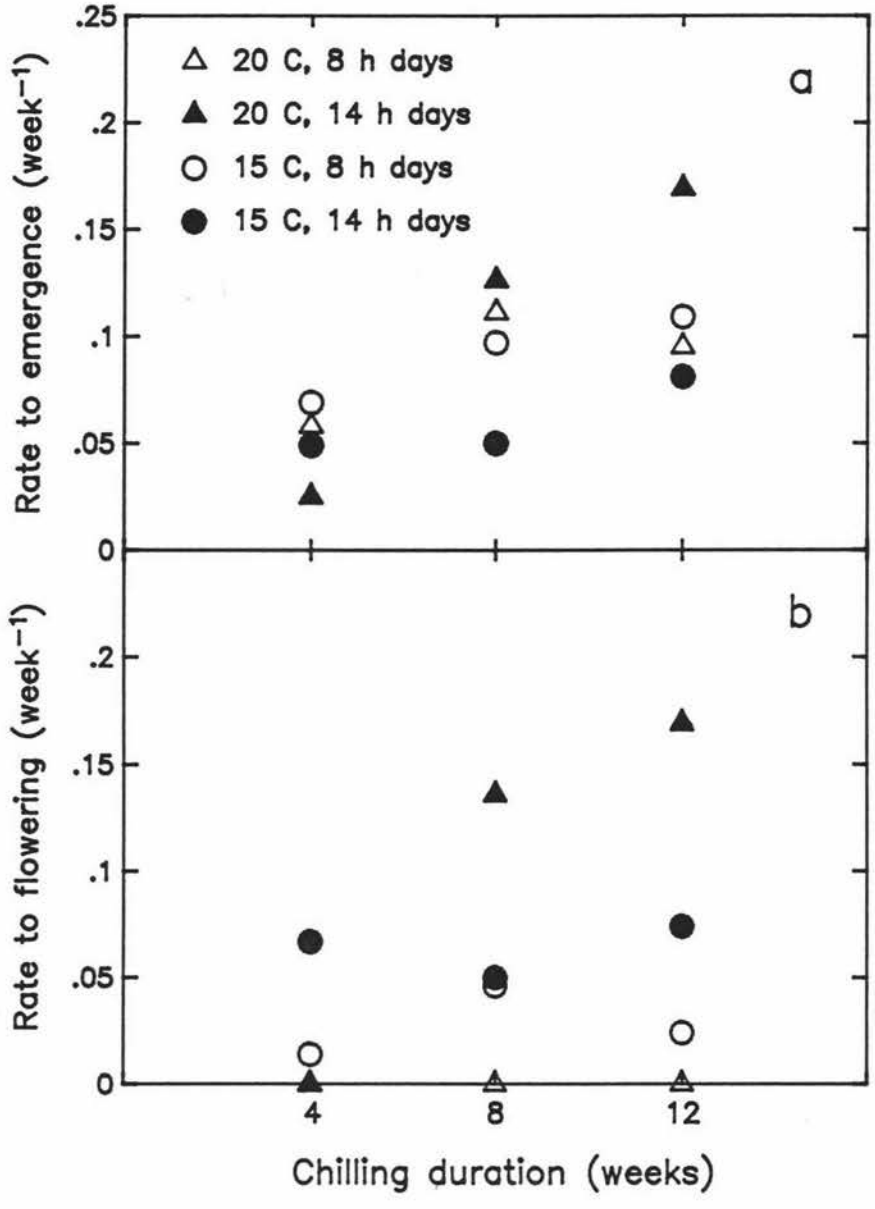

Figure 5.2: Effect of duration of chilling on (a) rate to inflorescence emergence and (b) rate to flowering for cultivar 'Early Long Keeper'.

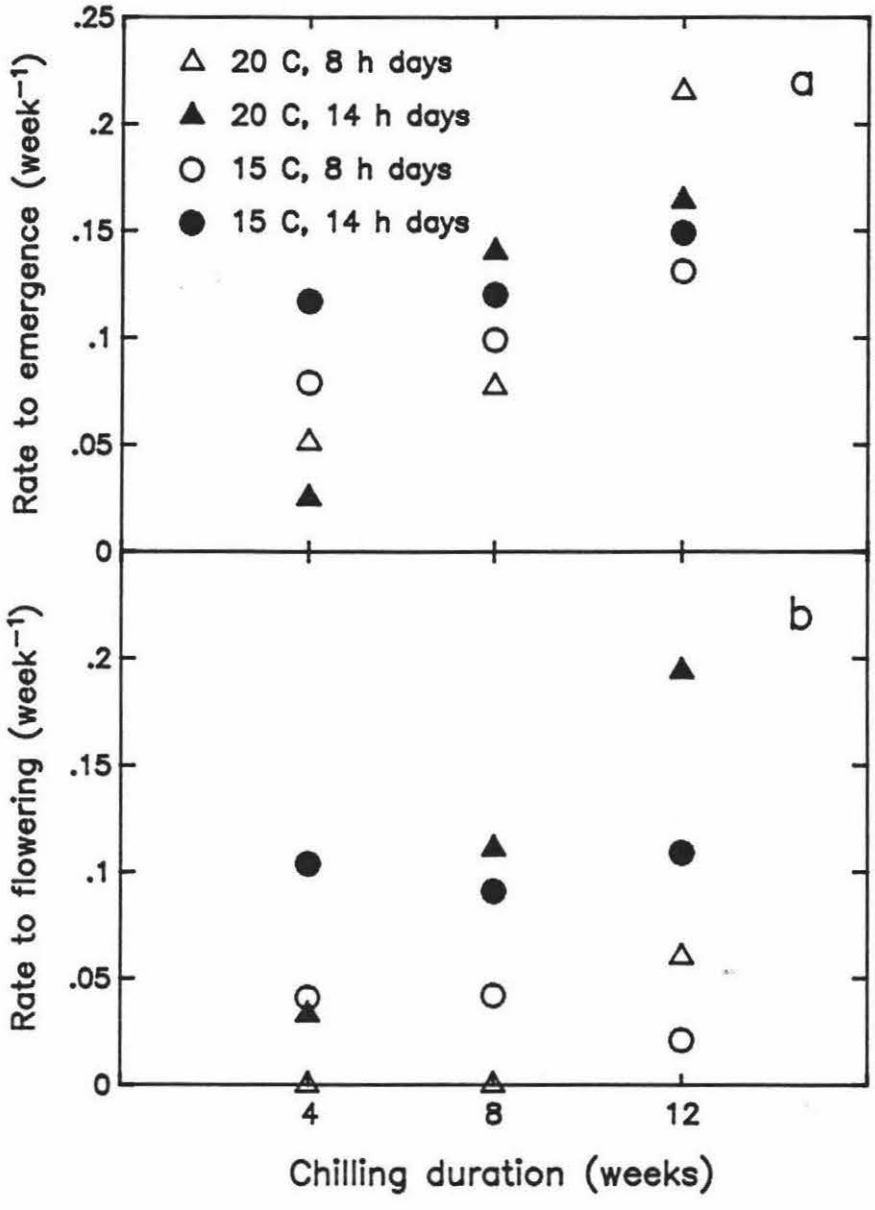

Figure 5.3: Effect of duration of chilling on (a) rate to inflorescence emergence and (b) rate to flowering for cultivar 'Gladalan Brown'.

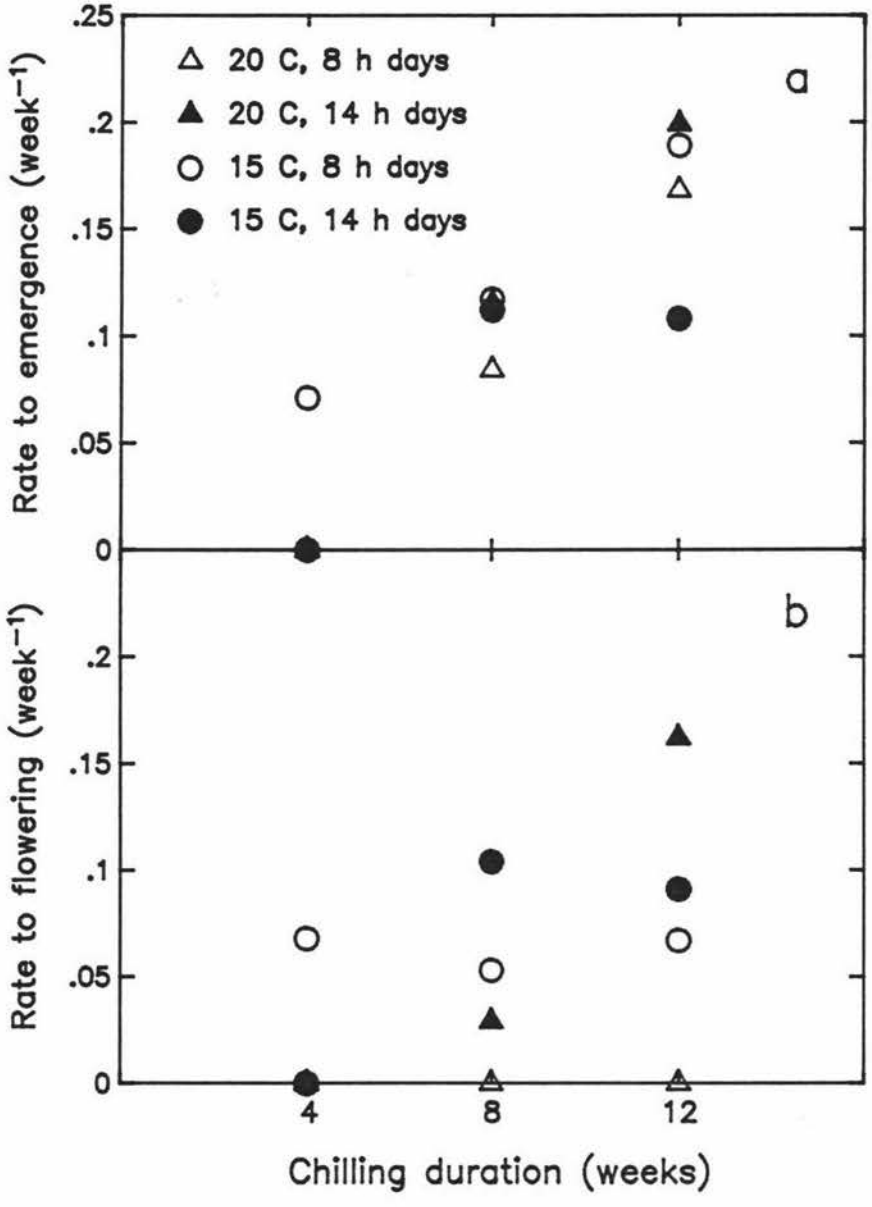

Figure 5.4: Effect of duration of chilling on (a) rate to inflorescence emergence and (b) rate to flowering for cultivar 'Sakigake Yellow'.

long-days. The effect of temperature was not significant overall, though 'Sakigake Yellow' did show faster development under 15°C conditions ($\alpha < 0.001$)), reflecting the higher rates of abortion under 20°C, except after 12 weeks chilling (Fig. 5.4b).

All cultivars showed some malformations of florets and inflorescences in cool short-days, though very few appeared in 'Early Long Keeper'. 'Sakigake Yellow' and 'Gladalan Brown' also showed some abnormalities in warm long-days. These observations are discussed more fully elsewhere (Chapter 6).

5.3.5 Seed production

Floret numbers in 'Early Long Keeper' showed no effect of chilling duration, but did respond to photoperiod ($\alpha < 0.01$), and less strongly to temperature ($\alpha < 0.05$). Mean floret numbers produced per inflorescence were 162 in cool short-days, 596 in cool long-days, and 696 in warm long-days.

Six inflorescences on four plants of 'Sakigake Yellow' were enclosed in a pollination cage (Fig. 5.5) in warm long-days when all plants had reached anthesis after receiving 8 weeks chilling, 4 weeks in cool short-days, and 12 weeks in cool long-days. Twenty blowfly pupae (Currah, 1981) were also put into the cage, which was sealed around the scapes of the inflorescences. After six weeks, when the first signs of seed shedding were seen, the inflorescences were harvested and the seeds from each inflorescence were counted.

The inflorescences in the cage differed in height by over 60 cm, and the blowflies tended to congregate at the top of the cage. The results show a trend from top to bottom of the cage; in order from the top, the inflorescences had 213, 525, 178, 273, 5 and 14 seeds.

The tallest inflorescence had been flowering for 4 weeks before transfer to the cage, so many of its florets would not have been fertilised, and no additional blowflies were added to the cage after the initial group. Both these factors would have tended to

Figure 5.5: Cage for pollinating onion flowers with blowflies

reduce yield. However, clearly the florets were fertile and it is possible to produce seed in a controlled environment by this method.

5.4 DISCUSSION

It is clear from the results of the analyses of variance that the sample size chosen was too small to allow firm conclusions to be drawn about many factors. Lack of significance of chilling duration on floral initiation in 'Sakigake Yellow', for instance, appears to be a result solely of the small sample size. Brewster (1982a) does not state how the SEDs obtained were calculated. However, in the second paper (Brewster 1982b), it appears that the regression he obtained was fitted to data for individual inflorescences rather than for plants. If the same method was followed by Brewster (1982a), the apparent sample sizes would have been 2-5 times bigger than the number of plants. This is an unusual method of analysis; a more common approach is to treat inflorescences on a plant as correlated and take a single value for each plant, such as time to first anthesis or mean time to anthesis.

In this experiment, very few plants flowered in short-days. However, it is possible to develop inflorescences normally in short-days. In a preliminary experiment (Chapter 3), plants were chilled for 14 weeks and warmed more gradually, in 4°C steps, from 8°C to 20°C, and produced healthy inflorescences with few aborted florets or other malformations. This suggests that the low floret numbers and malformations observed in cool short-days are a physiological response to temperatures which are too low for floral development. The incidence of disease in the warm short-day treatment accounts for the fact that most inflorescences did not open before the plants were discarded. Warm temperatures early in inflorescence development may have favoured fungal growth.

5.4.1 Stage of floral development

Early development of inflorescences in the onion cultivars studied was favoured by low temperature conditions where growth was slow, while development after inflorescence emergence was favoured by warm temperatures and long-days. This is in general agreement with the results of Holdsworth and Heath (1950). However, there was considerable variation between cultivars in specific responses.

Brewster (1986) describes a temperature-dependent model for predicting floral initiation in onion seedlings. Substituting the environmental conditions in the present work into his equation gives a prediction that initiation should occur in 2 weeks, and that development of the inflorescence to 'growth stage 4' (a well-defined spathe completely covers the apical dome) should take a further 7 weeks at 8°C. Since plants were only dissected when they failed to flower, it is not possible to test this accurately, but it would seem likely that initiation in sprouting bulbs of 'Gladalan Brown' and 'Early Long Keeper' could be predicted using this model. 'Sakigake Yellow', however, appears to respond to chilling much more slowly.

Abortion of inflorescences in long-days occurred most often before emergence, apparently in plants where the inflorescence was still small at the time of transfer out of chilling temperatures. In cool short-days abortion of inflorescences and florets tended to occur after emergence, when conditions did not favour development of the inflorescence and withering of the spathe. In these cases, as Roberts and Struckmeyer (1951) observed, the axillary bud produced at the base of the scape grew vigorously and either continued to produce bladed leaves (in short-days) or produced scale-leaves and formed a bulb (in long-days). This suggests that competition between leaf growth and the reproductive apex continues throughout, but that reproductive development is favoured by different environmental conditions at different stages.

Plants with mature bulbs, but with the leaves still green, appeared to initiate inflorescences somewhat more readily than unsprouted bulbs, but both were very much

slower than sprouted plants. Abdalla and Mann (1963) found that leaf initiation by the apex in bulbed plants stops while the leaves are still green, before the tops have fallen, indicating that apices of green plants and unsprouted bulbs may be in similar states. This suggests that a dormant apex may be less receptive to chilling than one that is actively growing.

Sensitivity to duration of chilling confers bolting resistance on 'Sakigake Yellow', which also showed a response to warm temperatures for floral development. This probably reflects an origin in Japan with cold winters and hot summers. 'Gladalan Brown', which bolts easily in the field, required only a short period of chilling to initiate inflorescences. 'Early Long Keeper', while not as quick to respond to chilling as 'Gladalan Brown', initiated inflorescences readily, suggesting that difficulties experienced by breeders in the the field (D. Grant, pers. comm.) result more from the prolonged dormancy of the bulb than from a slow response to chilling.

5.4.2 Rate to emergence and from emergence to flowering

The effects of both temperature and photoperiod observed here were much stronger than observed by Brewster (1982). Brewster (1982) derived two equations for predicting time from inflorescence appearance to spathe rupture. Substituting into his equations, and adding 20% to account for the difference between spathe rupture and first floret anthesis (instead of the 25% for the difference between spathe rupture and peak flowering; Brewster, 1982) gives predictions of mean time to flowering. The equation based on temperature alone is a poor predictor of behaviour observed in this experiment; predicted values are 5.9 weeks at 20°C and 7.8 weeks at 15°C, while observed values were 6.4 weeks at 20°C and 11.2 weeks at 15°C. (The observed means used in this comparison are based only on plants that did flower, to agree with Brewster's (1982) work.) The equation based on photoperiod and temperature gives good agreement with experiment in cool long-days, where predicted and observed values are both 8.2 weeks. In cool short-days the predicted value is an underestimate (10.4 weeks) of the observed value (14.6 weeks). The predicted value

for warm long-days is a slight overestimate (6.5 weeks) of the observed value (5.9 weeks).

These results probably reflect the difference between using the mean of increasing temperatures and photoperiods and having two fixed and widely differing values as in the present work. The cool short-day prediction probably involves extrapolating Brewster's equation outside the range of conditions for which it was derived.

5.4.3 Development of protocols for use in breeding programmes

Some plants of all cultivars were sprouted after 8 weeks in cool short-day conditions, and all plants given 8 weeks chilling at 8°C in short-days initiated inflorescences. Emergence of the inflorescences occurred in 8-10 weeks in cool short-days in all cultivars used, with very little abortion. The daylength could be increased to increase the rate of floral initiation, but this carries an increased risk of inflorescence abortion. Further development of the inflorescence, however, occurred best in warm long-days. Such a protocol would be expected to bring bulbs into flower in 30-35 weeks from planting. The 8 weeks allowed for sprouting could be shortened to the time required to achieve 20% sprouting in the slowest cultivar. (In this experiment less than 20% of 'Early Long Keeper' bulbs had sprouted when the plants were transferred to chilling conditions, but no effect of this on flowering was detectable.)

The red to far-red ratio used in the cool long-day treatment would be expected to inhibit bulbing somewhat; if a lower ratio were to be used, it would be necessary to shorten the photoperiod (e.g. to 12 h) to achieve comparable results. Increasing the photoperiod to an intermediate value such as 10 h immediately after chilling might speed the emergence of inflorescences.

A method such as this would allow the breeder both to synchronise the flowering of a range of varieties and, if bulbs were planted in January, to obtain seed in time for planting in late winter.

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Chapter 6

Organ development

6.1 INTRODUCTION

Development of abnormal structures in or in place of flowers or inflorescences in marginal or unfavourable conditions for flowering has been reported in a number of species. Most of these reports concern responses to marginally inductive, or too few fully-inductive, photoperiods. In *Chrysanthemum morifolium* (Popham and Chan, 1952; Schwabe, 1951; Cockshull, 1976), plants kept in continuous long-days developed a capitulum, but few or no florets, and did not reach anthesis. Transfer of plants to short-days resulted in normal flowers, though with more bracts than usual. Plants forced to flower in long-days by removal of all lateral shoots produced proliferous inflorescences, where a number of secondary inflorescences grew from the primary one. Other flowers forced to open in long-days by re-rooting as cuttings produced leafy shoots from the corolla tube (Schwabe, 1951). Plants of *Impatiens balsamina* L. cv. 'Buisson Fleuri' developed a double flower if given 22 or more short-days (8 h), but produced abnormal structures intermediate between floral and vegetative growth if given 3-14 short-days (Simon, 1974). Galinat and Naylor (1951) described production of proliferous tassels, containing leafy shoots, in maize plants in photoperiods of 15 h or longer.

Reversion of inflorescences to vegetative growth in unfavourable temperature conditions has been reported in wallflower (*Cheiranthus cheiri* L.) by Diomaiuto-Bonnand (1972). Plants were induced to flower at low temperatures (2 or 5°C); floral development was completed at 5 or 12°C, but plants transferred to 22 or 24°C reverted to vegetative growth. These reverted plants could be induced to flower again by returning them to 2°C. Development of bulbils in the inflorescence of *Poa bulbosa* in short-days at temperatures below 20°C was reported by Youngner (1960). *P. bulbosa* normally requires vernalization at 10°C before normal florets are developed at 21-27°C.

Normal development of leaves and inflorescences of onion (*Allium cepa* L.) has been reviewed by Jones and Mann (1963) and is outlined in Chapter 2. Experiments that involved growing onions in cool short-days (15°C and 8 h days), cool long-days (20°C and 14 h days), warm short-days (20°C and 14 h days), and warm long-days (15°C and 8 h days), and some transfers between conditions (Chapters 4, 5) during bulb sprouting and inflorescence development, resulted in some abnormal morphology of leaves and inflorescences. These malformations are described, and compared with inflorescence development in *Allium cepa* var. *proliferum*. Implications for the course of organ development in onions are discussed.

6.2 LEAF DEVELOPMENT

In short-days, leaves grow long blades and long unthickened sheaths. In plants transferred from short-days to long-days, partly mature leaves with emerged green blades, which were still growing at the base, developed thickened, scale-like sheaths or sheath bases (where some sheath tissue had already matured). Where the leaf blade was still growing at the time of transfer to long-days, the immature tissue senesced and died (Fig. 6.1), leading to the death of any mature tissue at the leaf tip; again presumably as a result of diversion of assimilates.

Plants transferred from long-days to short-days often produced a number of leaves

with shortened blades (Fig. 6.2). Since onion leaves mature first at the tip, this is likely to be a result of leaves starting to mature in long days when leaf blades do not develop; elongation of the blade on transfer to short-days is limited to the remaining immature tissue.

6.3 DEVELOPMENT OF THE INFLORESCENCE

6.3.1 Long-days and warm temperatures

Many inflorescences do not develop fully in plants transferred to warm long-days, especially after short periods (e.g. 4 weeks) of chilling. Most abortion occurred before the inflorescence emerged; the plants matured as apparently normal bulbs, and inflorescences could only be detected by dissection (Fig. 6.3). In a small number of plants, inflorescences were aborted later in development, after emergence, when the leafy top fell over.

Holdsworth and Heath (1948) reported that inflorescences were crushed by the development of the axillary bud at the base of the scape as bulb scales. No evidence for this was observed in the present work; rather it appeared that inflorescences had aborted, perhaps as a result of vigorous growth of bulb scales depriving them of photosynthates. The subtending scale was generally large, and often showed a fold that accommodated the inflorescence (Fig. 6.3).

Plants of 'Sakigake Yellow' produced small numbers of bulbils in the inflorescence in warm long-days (Fig. 6.4).

Some plants of 'Early Long Keeper' developed abnormal spathes, with one or two hollow, blade-like structures arising from the top of the sheath (Fig. 6.5).

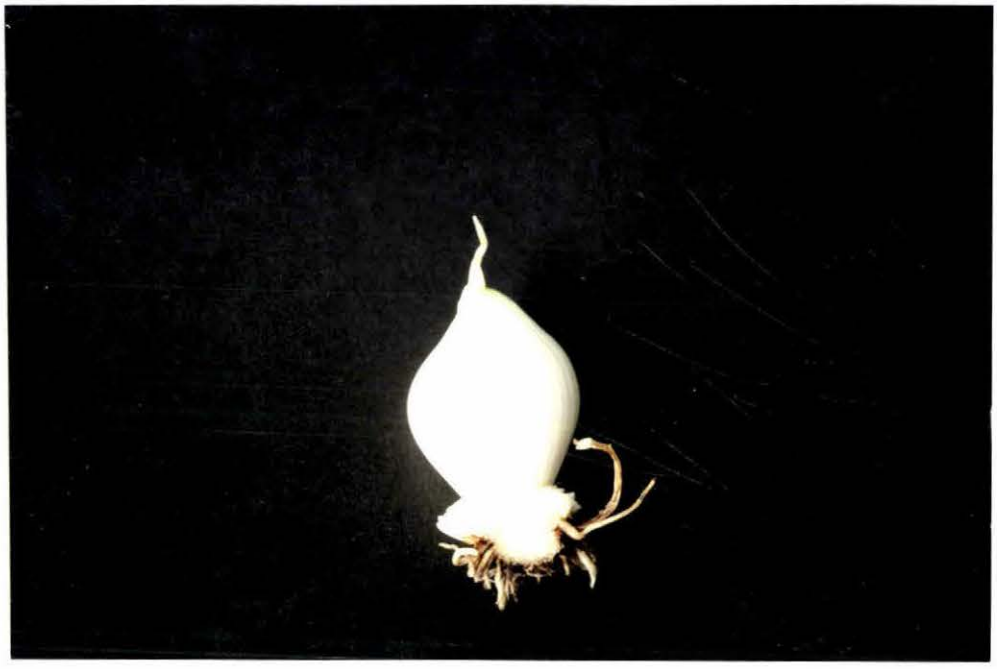

Figure 6.1: Leaf of plant transferred from short-days to long-days. Note senescence of immature leaf blade tissue.

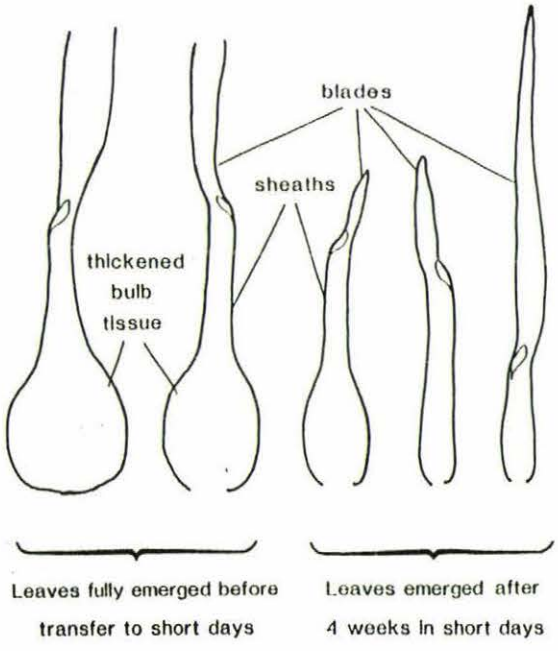

Figure 6.2: Schematic diagram of leaves from a plant transferred from long-days to short-days.

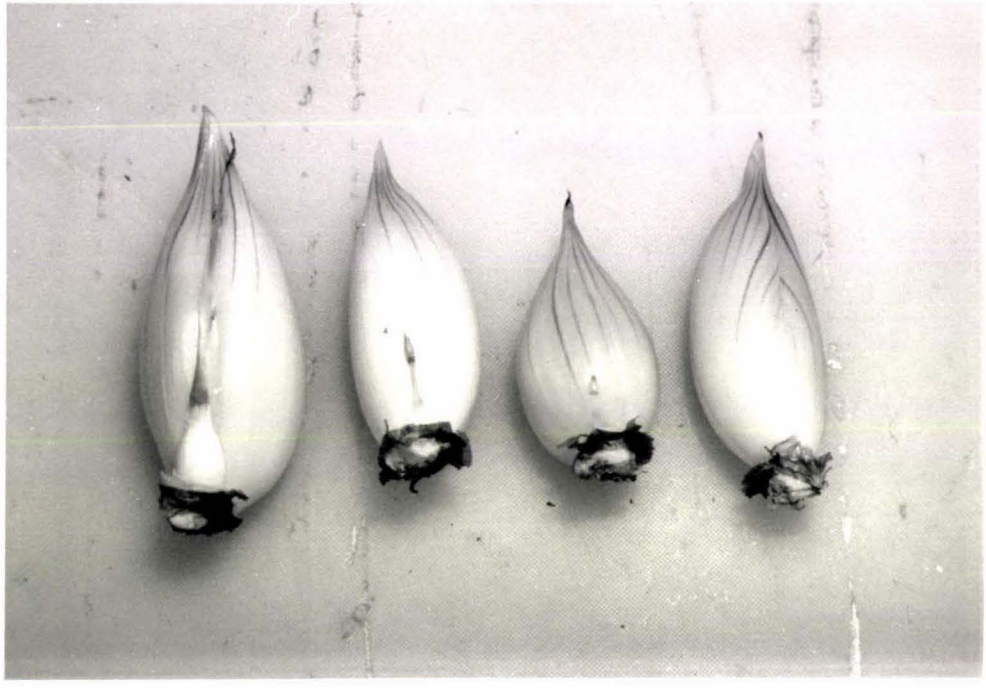

Figure 6.3: Abortion of inflorescences in long-days. The plant at right shows a fold in the scale subtending the inflorescence and vegetative bud.

Figure 6.4: Bulbil produced in otherwise normal inflorescence in long-days.

Figure 6.5: Spathe of plant transferred to long-days. Note the two blade-like structures (arrowed) and the sheath-like appearance of the spathe itself.

6.3.2 Short-days and cool temperatures

Inflorescence emergence and early development was slow but otherwise normal in most plants in cool short-days. However, in some plants of 'Early Long Keeper', spathes appeared abnormally thick and green (Fig. 6.6). Late in development, however, it became apparent that inflorescences were not maturing normally. The spathes did not dry out to become papery and brittle, but turned brown and stayed moist. Some spathes were burst by growth of floret pedicels in the umbel, while in others all florets aborted. In one case florets opened inside the spathe. Surviving florets in inflorescences that were split by hand opened normally. Florets that opened under cool short-days did not appear to shed pollen.

All inflorescences produced under cool short-day conditions showed high levels of floret abortion (Chapter 5), and deformed pedicels were common (Fig. 6.7). 'Sakigake Yellow' plants produced many inflorescences on which the florets were replaced by bulbils, some forming between sepals on a pedicel (Fig. 6.8). Plants of 'Yozui Yellow', 'Sakigake Yellow', 'Early Long Keeper' and 'Gladalan Brown' produced florets where the floral parts were partly or completely replaced by florets or bulbils (Fig. 6.9). Some of these florets matured normally and some aborted. Incomplete development of flowers in short-days (Scully *et al.*, 1945) and development of topsets, or bulbils, in the inflorescence have been reported (Roberts and Struckmeyer, 1951).

'Early Long Keeper' florets had short pedicels.

The axillary bud produced at the base of the scape in a reproductive plant became a leafy shoot in cool short-days. In 'Sakigake Yellow' and 'Gladalan Brown' many of these shoots produced 6-8 leaves followed by a new inflorescence (Fig. 6.10).

Some plants of 'Gladalan Brown' produced shoots with multi-bladed leaves (Fig. 6.11). Normal phyllotaxis was not retained in the subsequent growth of a shoot; many multibladed leaves were produced around several growth centres, some of which produced fasciated inflorescences (Fig. 6.12), and several of which produced small inflorescences which subsequently aborted. Similar structures composed of

thickened scale tissue raised on a short stem were discovered in a plant of 'Early Long Keeper' in cool long-days.

6.3.3 Long-days and cool temperatures

Cool long-days were the most favourable conditions for inflorescence emergence. One plant of 'Early Long Keeper' that had bulbed, and was dissected to check for inflorescence initiation, contained two abnormal structures which looked like inflorescences composed of thickened white bulb scale tissue. These inflorescences were raised on short stems (Fig. 6.13). Since the only internode that elongates in an onion plant is the scape bearing the inflorescence, it appears that these structures were inflorescences that had reverted to vegetative growth. When the swollen outer spathes/scales were removed, the axes bore what appeared to be fasciated scale leaves grouped around four apices, three of which bore recognisable inflorescences.

6.3.4 'Tree onions'

Inflorescences of *Allium cepa* var. *proliferum* grown in temperate climates normally contain a number of bulbils (Fig. 6.14), and the florets that are produced are generally not fertile (Jones and Mann, 1963). Plants grown in Palmerston North in 1987 produced bulbils exclusively in inflorescences that opened in early spring; later opening inflorescences produced a higher proportion of florets and fewer bulbils (Fig. 6.14). This was the case whether the inflorescences were primary (produced from the base of the plant) or secondary (produced from a bulbil in a primary inflorescence) (Fig. 6.14). Florets did not appear to produce pollen.

Spathes surrounding the inflorescences had leaf-like blades up to 30 cm long (Fig. 6.14).

Figure 6.6: Spathe of plant developing in short-days. Note heavy texture and dark colour.

Figure 6.7: Deformed pedicels produced in cool short-days.

Figure 6.8: Bulbils formed in cool short-days.

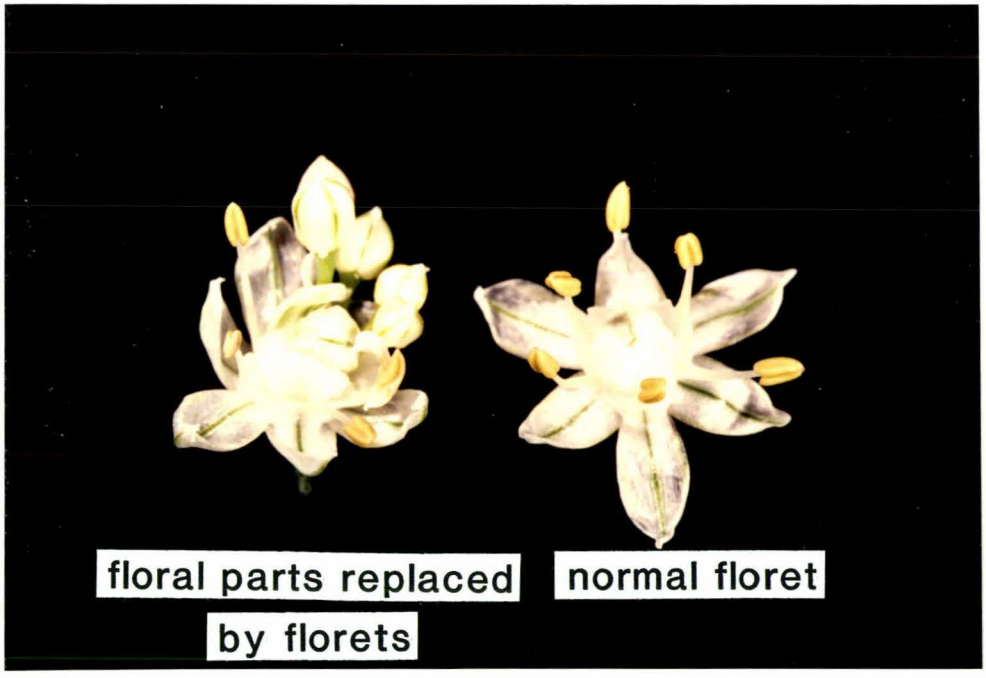

Figure 6.9: Abnormal florets produced in cool short-days. Close-up of abnormal floret (left) and normal floret (right).

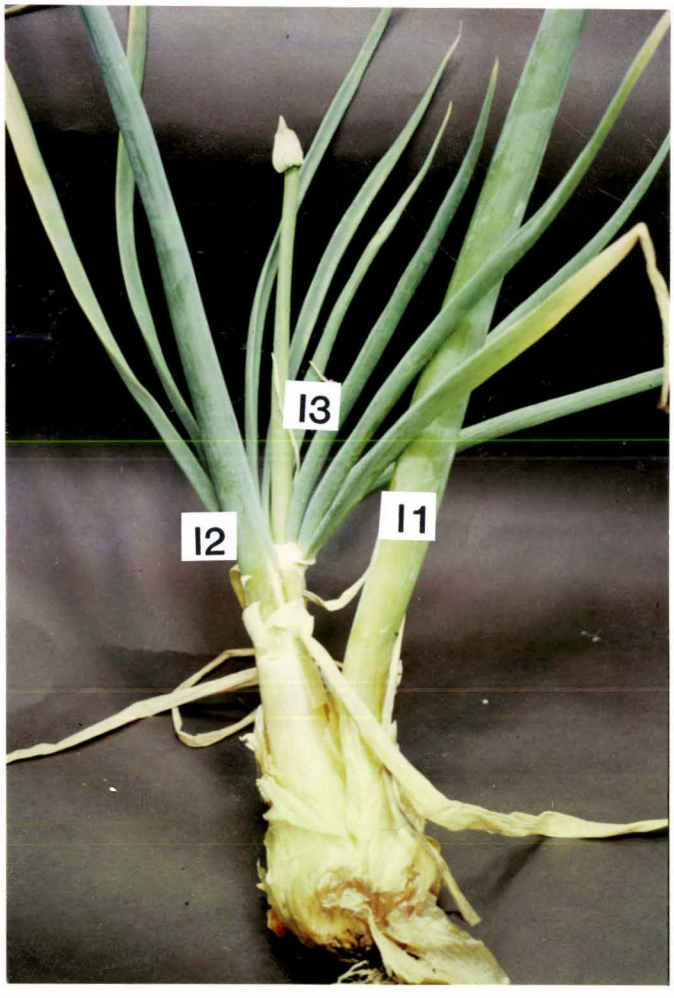

Figure 6.10: Three inflorescences (I1, I2, I3) on one shoot produced in cool short-days. Two axillary buds in succession have become reproductive after producing 6-8 leaves.

Figure 6.11: Leaf with multiple blades produced in cool short-days.

Figure 6.12: Fasciated inflorescence produced in cool short-days.

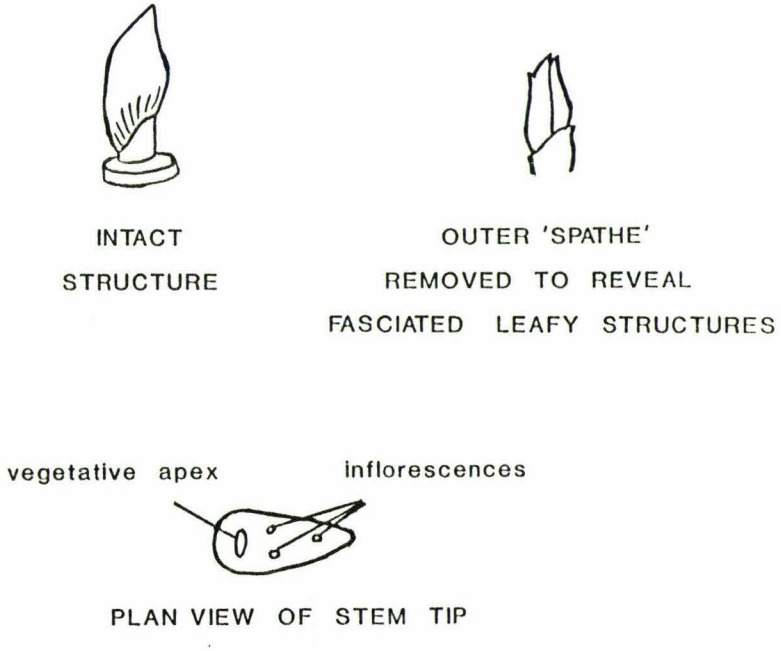

Figure 6.13: Bulb-inflorescence formed in cool long-days.

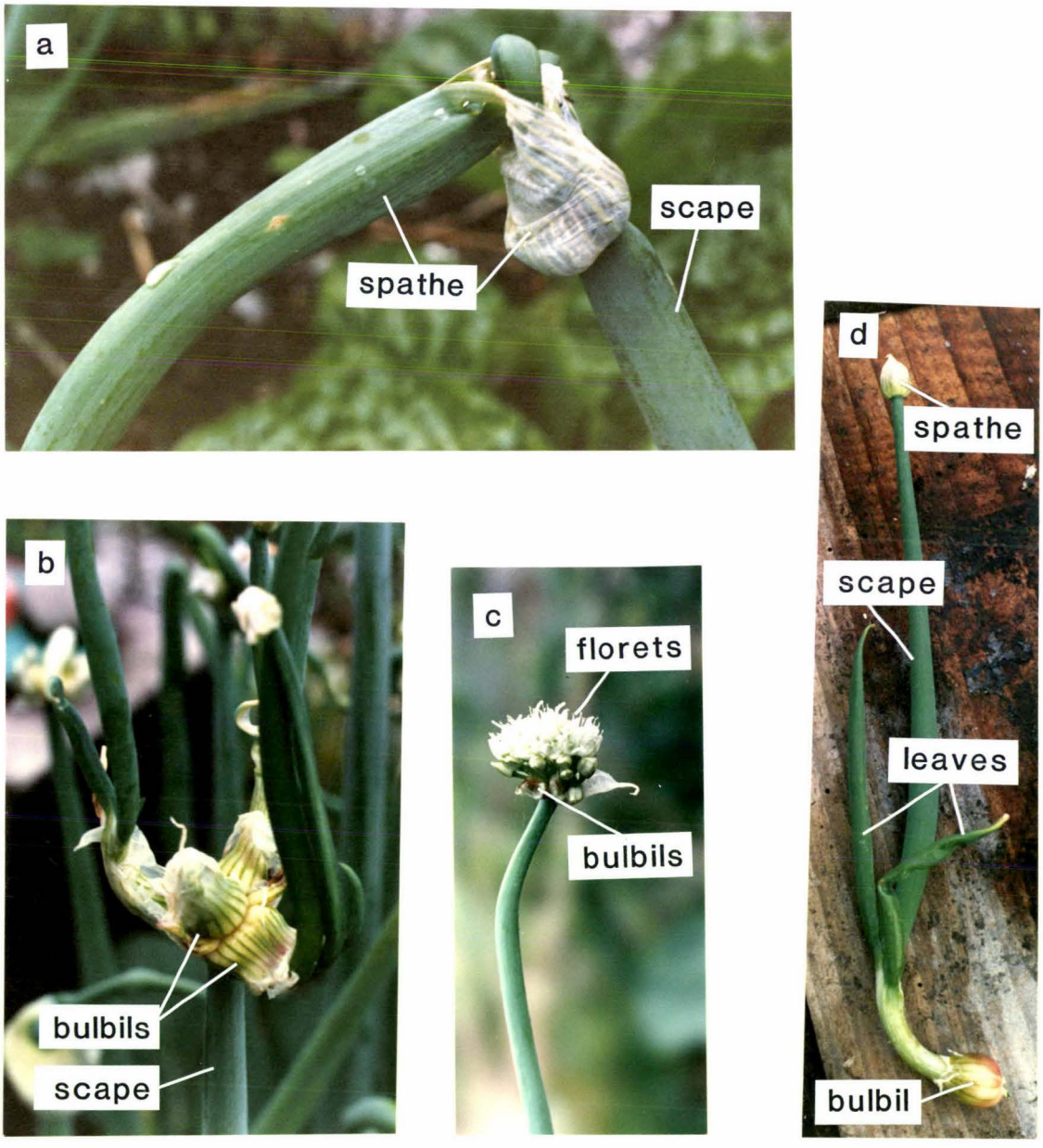

Figure 6.14: Inflorescence of *Allium cepa* var. *proliferum*, (a) leafy spathe; (b) bulbils in inflorescence; (c) florets produced in late spring, apparently without pollen production; (d) secondary inflorescence growing from bulbil.

6.4 DISCUSSION

6.4.1 Leaf development

The phenomena observed when growing leaves are subjected to abrupt changes in conditions indicate that all leaf tissue remains plastic until it is mature. The plant does not initiate scale leaves or bladed leaves as such, but a more general foliar structure which can mature either as a scale leaf or as a bladed leaf or as a combination of the two, depending on environmental conditions. This is in agreement with the results of Denne (1960), who found that bladed leaves and scale leaves of *Narcissus* were indistinguishable until they were 1 mm long. Subsequently, leaf growth and cell division became restricted to the sheath in scale leaves, while in foliage leaves the intercalary meristem in the blade was maintained and enlarged, though it never extended more than 50 mm up the blade (Denne, 1960).

6.4.2 Floral development

Flowering in onions appears to be a multistep process, and development of different parts of an onion inflorescence becomes irreversible at different times. Reproductive development can be divided into three stages. First, spathe initiation occurs, phyllotaxis changes, and the scape starts to elongate. Spathe and scape differentiation becomes irreversible very early - once the inflorescence reaches this stage it will generally either develop or die. Only four plants showed evidence of floral structures reverting to vegetative growth before significant scape elongation had occurred. These showed changes in phyllotaxis and leaf structure analogous to those observed by Simon (1974) in *Impatiens balsamina* cv. 'Buisson Fleuri', when plants were given only 3-8 inductive cycles.

The second stage of reproductive growth involves maturity of the spathe, development of the inflorescence and scape elongation. Optimum temperature and photoperiod increase during this process. Unfavourable conditions (high temperature

and long-days) before emergence tend to result in abortion of the inflorescence, while cool temperatures and short-days later in development slowed maturity of the spathe and resulted in the production of leafy shoots in the inflorescence. Some parallels are seen in the development of leaves in inflorescences of *Cheiranthus cheiri* when plants are transferred to high (24°C) temperatures.

Floret development and the development of floral parts are completed last. A floral primordium may still behave as an apical meristem and produce a vegetative axis (a bulbil), or new florets rather than floral parts, late in development. The apex of the axillary bud can also become reproductive if conditions are conducive to floral initiation and the plant is growing vigorously rather than bulbing.

These observations suggest that reproductive growth in onions is subject to alteration by environmental conditions at most stages of development. In situations where the environment is modified to accelerate or enhance floral development in breeding and selection programmes, account must be taken of the changes that can occur in floral development to avoid the production of abnormal inflorescences and infertile florets.

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Chapter 7

Modelling onion growth

7.1 INTRODUCTION

Models are representations of reality. Plant growth models are not intended to be complete replicas of plants or plant systems; instead they serve to summarise salient knowledge about the system, delineate areas of the system which are not adequately understood, and make predictions about the behaviour of the system. By comparing predictions with reality, a model can be tested (Thornley, 1976). A plant growth model could be described as a formalised hypothesis about the plant system in question. The value of a model depends on how well it fulfils the functions listed above. It does not necessarily depend on the accuracy of the predictions derived from the model. More information can often be gained from a model which, by inaccurate predictions, eliminates possible mechanisms for the system of interest (Fukshansky and Schäfer, 1983). Accurate predictions are necessary if a model is to be used as a management tool.

The choice of salient knowledge and the structure of the model depend both on the system under study and on the purpose of the modeller. A model of crop dry weight production may be purely descriptive, intended to summarise knowledge and to delineate a problem more clearly; or it may be predictive, intended to use knowl-

edge and hypotheses about the crop's responses to environmental factors such as temperature, to predict crop dry weight at harvest, or the consequences of management decisions. Such models may be speculative, intended to test ideas, or may be intended as a tool in making management decisions.

In this chapter, the environmental physiology of onion growth and development is outlined as a problem system where a model, or hypothesis, is needed as a synthesis of available knowledge and as a predictor of behaviour for the system. A range of mathematical approaches to plant growth models are outlined, and two models for onion bulb growth, a descriptive dry weight model (Lancaster and Gandar, 1986) and a model for the effects of daylength and temperature on bulbing (Bertaud, 1986) are outlined. Extensions of the models to deal with bulb sprouting, inflorescence initiation and inflorescence development are discussed.

7.2 ONIONS: THE PHYSIOLOGICAL PROBLEM

Bulbing in onion plants is controlled by daylength and temperature (see review by Bertaud, 1986). In winter, in short-days and cool temperatures, onion plants produce a succession of leaves but do not bulb. In the lengthening days and warmer temperatures of spring, some photosynthate is stored as fructans in swollen leaf bases, rather than used as structural carbohydrate to produce more leaves. Eventually all photosynthate produced is stored in these swollen leaf bases and modified leaf sheaths to form bulb scales, and leaves cease to elongate and produce blades. Old leaves die and are not replaced; the remaining leaves are not supported by new blades growing up through their sheaths, and the leafy top of the plant falls over, when the bulb is said to be mature.

Mature onion bulbs keep for varying lengths of time (depending on cultivar) without sprouting or rooting. Bulbs that will root readily when planted in moist soil can be said to be ecodormant, while bulbs that will not root even when provided with optimum conditions such as adequate water, light and nutrients can be said to

be paradormant (Chapter 4). Long keeping varieties such as 'Early Long Keeper' display paradormancy for a period after harvest. Later in storage paradormancy disappears to leave only ecodormancy in stored bulbs (Chapter 4). The apex appears to become dormant before the leaves die (Abdalla and Mann, 1963).

Sprouting of bulbs involves a resumption of leaf blade growth; it is not clear whether the stem apex in an unsprouted, ecodormant bulb is actively growing or not. Leaf production at the rate of one primordium every two weeks has been reported in the poor-keeping cultivar 'Excel' (Abdalla and Mann, 1963). Sprouting in ecodormant bulbs involves a similar photomorphogenetic response to that observed in bulbing (Chapters 3, 4), where bladed leaves are produced in short-days and thickened bulb scale tissue is produced in long-days.

Floral initiation requires a period of cool (7-15°C) temperatures. Early development of the inflorescence also proceeds more rapidly at temperatures of 15°C or below (Heath and Holdsworth, 1950; Chapter 5). Once the inflorescence has emerged, temperatures of 20°C, or 15°C in 14 h days, ensure normal development of the spathe and florets (Chapter 5).

A model for onion growth must account for partitioning of photosynthate between structural tissue and storage tissue in response to photoperiod and temperature, as well as for increase in dry weight by photosynthesis.

The phenomena of beginning and ending dormancy, or of transition from one kind of dormancy to another, are not well understood, and any attempt to model them at present would be largely speculative. Instead it is useful to look at bulbing, at sprouting in ecodormant bulbs, and at subsequent floral initiation and development. The latter phenomena are particularly important to breeders.

7.3 RELATIONSHIPS BETWEEN MATHEMATICAL MODELS

7.3.1 Stochastic models

Stochastic models are generally used where numbers are too small or events too random to approximate the system's behaviour with continuous functions. A stochastic model expresses the state of the system in terms of probability distributions, rather than producing a single number, as in the case of deterministic models (e.g. Chalabi *et al.*, 1986).

Various kinds of models are available, and choice of the most appropriate model may not be a trivial problem. A population model for cells, for instance, might be formulated as a stochastic branching process, where an entity gives 'birth' to a number of entities of the same type as itself. The behaviour of the population is expressed as a probability distribution for the number of offspring. A population of leaves on a plant would be better formulated as a Markov chain, with the state of the plant defined as the number of leaves. In this case the plant has conditional probabilities of having various numbers of leaves at time $t+1$, given that at time t it has a number of leaves x. Where the system is not observed continuously, a model must account for the observation process, which is also discrete.

Only certain types of stochastic models are appropriate to plant growth and development processes. Stationary processes and some types of Markov chains, for instance, are only appropriate where the temporal average over a process,

$$\bar{f} = \frac{\int f(t)dt}{\int dt}, \tag{7.1}$$

is the same as the arithmetic mean,

$$< f >= \frac{\sum f(t^*)}{n}, \tag{7.2}$$

where t^* is the time at which a number of individuals are measured. Such a process is decribed as *ergodic*.

For most plant processes, this is not so. The changing seasonal pattern of leaf appearance and maturity, reproductive development, and leaf senescence and death means that plant processes change both qualitatively and quantitatively throughout the life of a plant. Ephemeral plants in high stress environments such as deserts and arctic conditions, and annual plants in temperate climates, which live their entire life cycles from seed to seed in a matter of a few weeks or months, depart furthest from ergodic behaviour; perennial plants in tropical areas, where seasonal changes are less marked, depart least.

Since stochastic models are mathematically complex and can be hard to formulate, continuous deterministic approximations are generally used where possible (Thornley, 1976).

7.3.2 Size density distributions

Density distributions are continuous functions which describe properties which vary between individuals. They can be estimated from discrete data of the sort one might obtain from observations of a very large population. The data are normalised by dividing each size class by the total number of observations (the number in the population, or sample), and added to obtain cumulative relative frequency data, which sum to one. A curve fitted to this is an estimate of the cumulative probability density function. Differentiating this function gives a probability density distribution, where the area under the curve equals one. The mean, $\int w.p(w)dw$, is a useful measure of the value of the property for a normally distributed population.

Distributions of properties imply underlying stochastic behaviour in the form of random variation between individuals in a population. This may be a result of random events in a process. Systematic variations occuring between groups of individuals mean that one is really dealing with more than one population. A deterministic process is one where the *process* is fixed regardless of the states of the individuals being processed, and a deterministic process carried out on a population containing

random variations can result in distributions of properties.

7.3.3 Deterministic models

Deterministic models produce a single number as output for a given initial condition. Such models may be discrete, in which case the system's behaviour is described by difference equations, or continuous, when differential equations are used.

It is common in biological systems to model stochastic processes using deterministic models for the mean value of a property of the system. Differential equations are generally used (e.g. Thornley, 1976), as they may be solved analytically using calculus; difference equations apply in the discrete case, but may give less information. The use of deterministic models involves assumptions about how the mean and variance of the system behave under the transformation modelled. There is some evidence to show that individuals of mean value in a population may not retain this position if, for example, the growth of individuals is non-linear (Gandar, pers. comm.). Use of the mean also implies an assumption that the population is normally, or at least symmetrically, distributed.

7.3.4 Compartment models

It is often possible to treat an experimental system, at plant or crop level, as a number of linked compartments. Each of these compartments represents an aggregate of similar parts, e.g. leaves; structure within the compartment, such as a hierarchy of leaf age, is ignored. One or more state variables may be associated with each compartment. Relations between variables are generally expressed as differential equations for simplicity, with the assumption of continuous, deterministic processes, although there is no *a priori* reason why stochastic or difference equations should not be used if appropriate.

Equations used are of two types; conservation equations such as number balances

or mass balances, and constitutive equations which account for the constitution of individual variables. Properties such as number and mass are conserved, while age is not.

In the compartment model proposed by Bertaud (1986) for an onion leaf (Fig. 7.1), a mass balance may be written for the blade compartment thus:

$$W' = P' + G' - I' - R' \tag{7.3}$$

where W' is the rate of change of dry weight, P' is rate of photosynthesis, G' is rate of growth of new leaf (Bertaud uses L'), I' is rate of import to the sheath, and R' is rate of respiration. A constitutive equation for P' might be

$$P' = \frac{\alpha F P'_{max}}{\alpha F + P'_{max}}.L \tag{7.4}$$

where α is a constant, F is light flux density at the leaf surface, P'_{max} is rate of photosynthesis at saturating light levels, and L is leaf area (Thornley, 1976). Development of relationships like these allows the behaviour of the model to be related to environmental factors or physiological characteristics of the system.

7.3.5 Models involving environmental forcing functions

It is something of a truism that plants respond to environmental conditions. Defining these responses can be very difficult, however. Suppose we have observed a distribution of times to flowering, and we want to relate this distribution to environmental conditions. The usual way to do this involves assuming the population to be normally distributed, calculating the mean, and using differential equations to describe a model.

This is equivalent to stating that a notional rate of development, r, is a function of the environment:

$$r = r(environment). \tag{7.5}$$

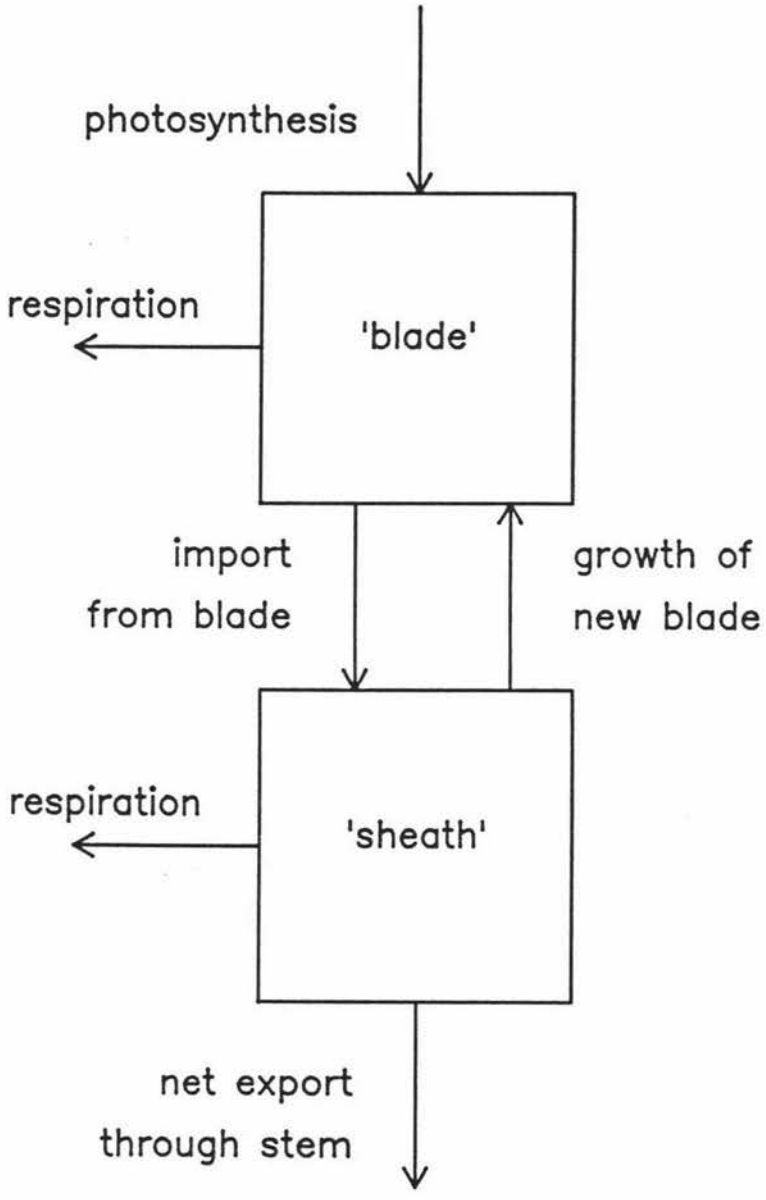

Figure 7.1: Compartment model for onion leaf showing inflows and outflows of dry matter.

(McNaughton, Gandar and McPherson, 1985). In fact, it is probably more accurate to write

$$r = r(state\ of\ plant, environment), \tag{7.6}$$

but in experimental work, plants are sorted for uniform size, age and other characteristics, to ensure that they are as nearly of the same state as possible, and the variable is ignored.

The state of the plant is defined as the integral of the rate :

$$State = \int_0^{flowering} r dt; \tag{7.7}$$

at flowering, the state of the plant is equal to 1. This definition involves the assumption that the plant responds only to current conditions, with no pre-conditioning effects of previous conditions.

The total environment is impractical as a variable because it is really a large number of variables, most of which cannot be specified. Many of these variables are correlated with seasonal changes in the natural environment, and most of them are probably of minor importance; certainly a number of models that successfully account for most of the variation in experimental data have been written which depend only on factors such as temperature or solar radiation (see for example Hughes, *et al.*, 1984). Variables such as planting density, soil moisture, fertiliser levels, shelter, and grazing can generally be controlled, and are usually treated as management variables, while soil type, temperature and daylength are uncontrollable except in controlled environment experiments. Soil type is a categorized variable, and thus difficult to include in equations; however, its effect on plant growth is usually a result of availability of moisture or nutrients, so specific properties such as bulk density may be measured and used instead. In practice, soil is often ignored; partly due to the tendency of plant scientists to concentrate on what happens above ground, and partly because, on most farmable land, soil type has little effect relative to temperature (T) and photoperiod (P):

$$r = r(T, P). \tag{7.8}$$

In many cases we can ignore photoperiod (which tends to be correlated with temperature anyway), in which case we get

$$r = r(T). \tag{7.9}$$

This is an empirical equation involving no assumptions about the form of the temperature dependence other than that the plant responses do not change with time, and that the plant responds the same way by night as by day to any given temperature. These are both testable hypotheses. There are good physiological reasons for suspecting the latter hypothesis to be inaccurate, since plants gain dry weight by photosynthesis only during the day, but for the sake of the argument we will accept it. The importance of the effect depends on the timescale of interest; for events measured in hours, such effects could be significant.

In attempting to define the effects of night temperature, some authors have applied treatments involving two different night temperatures with the same day temperature (e.g. Steer, 1980). In this work the effect of night temperature is complicated by the fact that the overall mean temperature is different in two such treatments. To elucidate the effect of night temperature, the plants must be exposed to the same temperatures for the same lengths of time, the only variable being the time period in the temperature cycle when the plants are exposed to light. Friend and Helson (1976) adopted this approach, but their data is analysed in terms of treatments rather than day and night rates for each temperature. It appears from their figures that plants did respond differently to temperature by night and by day.

There are times when it is useful to make some assumptions about the nature of the plant's response. This is particularly true in field situations, where temperatures may vary continuously over a considerable range, and models for r cannot easily be fitted because of the lack of independent treatments. Also continuous logging of temperature would be required, which can result in large and unwieldy data sets. The assumed plant response is called an environmental forcing function, because an outside factor is forcing the system to behave in certain ways (Patten, 1971). If an

environmental forcing function is used, the equation for the state of the plant is

$$S(t) = \int_0^t r(T(t))dt, \tag{7.10}$$

where T(t) is the environmental forcing function.

One of the commonest temperature response functions used is the degree-day or heat-sum model, where it is assumed the plant does not grow below a minimum temperature, and (usually) that the response to temperature above the minimum is linear. The heat sum, H, can be calculated from the equation:

$$H = \sum_{j=1}^{T} f(T_j \Delta t), \tag{7.11}$$

where T_j is the jth temperature class, and Δt is the time increment of interest. The values for each day are added to the sum accumulated since sowing. Assuming a linear response could lead to predictions of crop requirements that were significantly in error. It is a technique used most for arable and horticultural crop plants, whose temperature responses are often incompletely understood. Investigation of these responses could lead to better predictions of crop requirements, but is not a trivial problem. Data on growth rates must be collected over a sufficient range of temperatures to determine the form of f, and each temperature must appear in at least two independent treatments. Such work is most effectively carried out in controlled environment facilities.

7.4 A DESCRIPTIVE DRY WEIGHT MODEL.

A compartment model for an onion crop, comprising a framework which is a population balance equation, and a descriptive dry weight submodel for individual bulbs, was proposed by Lancaster and Gandar (1986). The compartments of the model are size classes. The bulbs are distributed over size classes by weight and the number of bulbs in each class can be divided by the total number to give a number density. For an arbitrary size class:

$$\begin{pmatrix} \text{rate of change} \\ \text{of number} \\ \text{in class} \end{pmatrix} = \begin{pmatrix} \text{rate of inflow} \\ \text{into class by growth} \\ \text{of smaller onions} \end{pmatrix} - \begin{pmatrix} \text{rate of outflow} \\ \text{from class by growth} \\ \text{of onions in class} \end{pmatrix} + \begin{pmatrix} \text{birth of} \\ \text{new onions} \\ \text{in class} \end{pmatrix} - \begin{pmatrix} \text{death of} \\ \text{onions} \\ \text{in class} \end{pmatrix} \tag{7.12}$$

In this equation, 'birth' occurs when swelling lateral buds cause bulbs to split, forming doubles, while 'death' is death and disappearance of plants. Lancaster and Gandar do not mention the contribution made to this term by splitting of large bulbs to form smaller ones (the 'birth' process mentioned above) resulting in a loss from the parent bulb's size class. However, it is not usually necessary to consider the birth and death terms, since mortality and splitting are likely to be low in commercial crops.

7.4.1 Growth of individual bulbs

The model for bulb growth is formulated in terms of the numbers and sizes of leaf blades and scales. The above-ground dry weight at time t is

$$w(t) = w_p(t) + \sum_{j=1}^{N_t} (b_j + s_j) \tag{7.13}$$

where N_t is the number of leaves produced at time t, $w(t)$ is the total dry weight, $w_p(t)$ is the dry weight of the base plate, b_j is the weight of the jth blade and s_j the weight of the jth scale (Lancaster and Gandar use ${}^{b}w_j$ and ${}^{s}w_j$).

Before differentiating to obtain growth rates, this equation must be modified to take

account of the change in leaf numbers over time resulting from leaf birth and death. N_t is changed to N_{max}, and unit step functions are introduced:

$$w(t) = w_p(t) + \sum_{j=1}^{N_{max}} U(t - t_j).(b_j + s_j) \tag{7.14}$$

Differentiating this product, we obtain

$$w'(t) = w_p'(t) + \sum_{j=1}^{N_{max}} U(t - t_j).(b_j\prime + s_j') + \sum_{j=1}^{N_{max}} \delta(t - t_j).(b_j + s_j) \tag{7.15}$$

where $\delta(t - t_j)$ is an 'impulse' function, the derivative of a unit step function. This third term only has a value at the time of initiation t_j, when its value is the initial dry weight of the primordium.

7.4.2 Discussion

The model is a deterministic description of a single plant. The population model described in the first paragraph would contain this as a submodel, and individual onions would be distributed over the weight classes according to some number density. This number density could be modelled in turn, or it could be measured. A measured density has the disadvantage that it only applies to the measured population; however, if measurements were made of a population over the period of its growth, and a model relating this to some environmental forcing function were fitted, then this could be tested against other populations after measuring some initial condition such as an initial size density.

To have explanatory value, the model must relate the plant to the environment. A dry-weight sum such as this is purely descriptive. However, it does serve to point up factors that must be taken into account when discussing the growth of onions.

The definitions of separate parts of a topographically continuous organism (one where the surface has no gaps or discontinuities) are inevitably somewhat arbitrary. This is particularly the case with leaf initiation in plants, where a few cells on the apical meristem form a slight bump which grows into a protrusion, and eventually becomes recognisable as a leaf primordium (Fig. 7.2). The point at which the new leaf ceases to be a part of the apex and becomes a separate organ is a matter of definition, and might well vary between individual observers, depending on their familiarity with the species in question. The matter is further complicated by the fact that very few observations have ever been made in growing tissue, since the apical meristem is generally inaccessible without destroying the plant. Poethig (1985) reports that tobacco leaves arise from a group of about 100 cells of the apical meristem. Onion leaves encicle the apex, so it seems likely that leaf primordia arise from a row of cells around the apex.

7.4.3 Leaf initiation - a term for the apex

The apical meristem is continuously growing, producing new tissue which differentiates into various organs - leaves and stem while the plant is vegetative, inflorescence and florets when it is in the reproductive phase. Here we are only concerned with vegetative growth. Although leaves appear sequentially, many leaf primordia may be growing, at different stages of development, at any one time. The growth function for the apical meristem is therefore made up of the growth functions of a number of leaves. The definition of these as separate structures at some point means that the apical meristem, as defined, shrinks at intervals by the size of a leaf primordium (assuming for the moment that all leaf primordia are defined as existing when they reach a certain size).

An apex term should cancel out the impulse function term in eqn 7.15. (7.15) says that each leaf springs into existence at t_j with an initial weight $(b_j + s_j)$. The apex term should grow each leaf until t_j, when it is defined as a separate organ and no

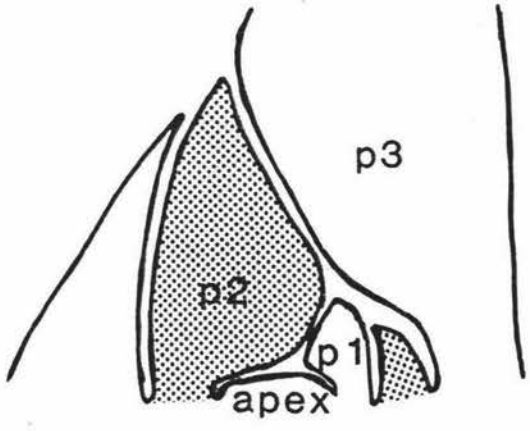

Figure 7.2: Section through apex of onion showing developing leaves. Primordia encircle the apex; shaded area represents one primordium (p1, p2, p3 are successive leaf primordia).

longer part of the apex. A possible expression might be:

$$w_a(t) = w_d(t) + \sum_{j=1}^{N_{max}} (1 - U(t - t_j)).(b_j + s_j). \tag{7.16}$$

apex dome developing primordia

The dome is the apical dome, a small area in the centre of the apex that continues to exist independent of the leaf primordia. Differentiating:

$$w'_a(t) = w'_d(t) + \sum_{j=1}^{N_{max}} (1 - U(t - t_j)).(b'_j + s'_j) - \sum_{j=1}^{N_{max}} \delta(t - t_j).(b_j + s_j) \tag{7.17}$$

7.4.4 Leaf appearance and death

Leaf appearance only applies to bladed leaves. It is potentially important as a turning point between autotrophic (self-feeding; i.e. able to support its own growth by photosynthesis) and heterotrophic (dependant for nutrient on other leaves) phases of growth. Although only a small part of the blade emerges at first (Fig. 7.3), I shall treat the leaf as though the time of appearance, t_a, marks the changeover between phases. Recent research on the light-trapping properties of plant tissue (Vogelmann and Björn, 1986) suggest that this is not an unreasonable assumption.

There are two aspects of death of leaf tissue. Leaves may suffer biological death, where tissue stops functioning but some material remains; alternatively, leaves may disappear or be removed. Ultimately, tissue that is biologically dead is likely to disintegrate and blow away as well. We treat death as disappearance of tissue, which involves a loss of dry weight. After bulbing, blades alone, or blades and unthickened sheaths of leaves may die, leaving scale tissue only. When appearance and death are included, the leaf blade term becomes:

$$\sum_{j=1}^{N_{max}} (U(t - t_j) - U(t - t_a))b_j^n + \sum_{j=1}^{N_{max}} (U(t - t_a) - U(t - t_d))b_j^e \tag{7.18}$$

where b_j^n is the weight of the unemerged blade and b_j^e is the weight of the emerged blade. A similar term could be written for leaf sheaths; however, problems could

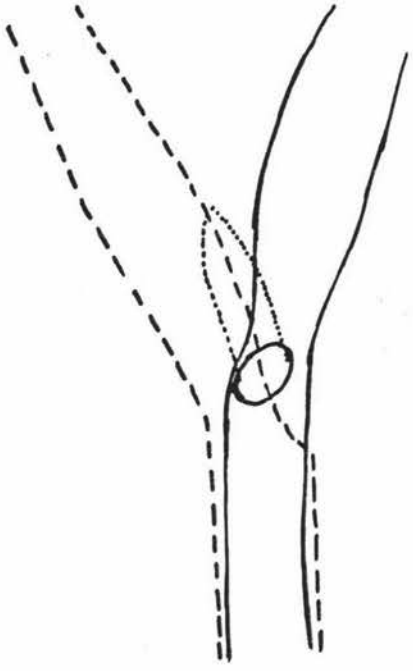

Figure 7.3: Leaves appear through a hole at the top of the sheath of the previous leaf.

arise in the case of sheaths that were made up of thickened scale tissue at the base, and unthickened sheath tissue above. After bulbing, the unthickened tissue will die off, leaving the thickened tissue as part of the bulb.

7.4.5 Flowering

Onion plants generally have a biennial habit, flowering in the second year of growth. (Plants are generally harvested for seed and discarded at this stage. If they were left in the ground they would continue to flower every year thereafter, so they are perennials rather than true biennials.) Susceptible varieties, planted early enough to have their chilling requirements met in the first winter, may bolt to seed in the first year.

The actual order in which events occur and the time scale involved are not relevant to the formulation of the dry weight model.

When the apex changes from vegetative to reproductive development, new structures are produced. Onion plants produce an inflorescence made up of a stem-like structure called a scape, a sheathing spathe (Fig. 7.4), and later, within the spathe, a large number of florets. Parts of florets (petals, stamens etc.) are ignored. The time at which the apex changes to the reproductive state is defined as t_f. At t_f, $w_a(t)$ becomes

$$w_a(t) = \underset{\texttt{spathe}}{U(t-t_f)p(t)} + \underset{\texttt{scape}}{U(t-t_f)c(t)} + \underset{\texttt{florets}}{\sum_{i=1}^{N_{max}} U(t-t_i)f_i(t)} \qquad (7.19)$$

If one was interested in obtaining seed from the plants, one would also have to account for abortion of inflorescences or florets, whether or not florets are fertilised, and growth and maturity of seeds.

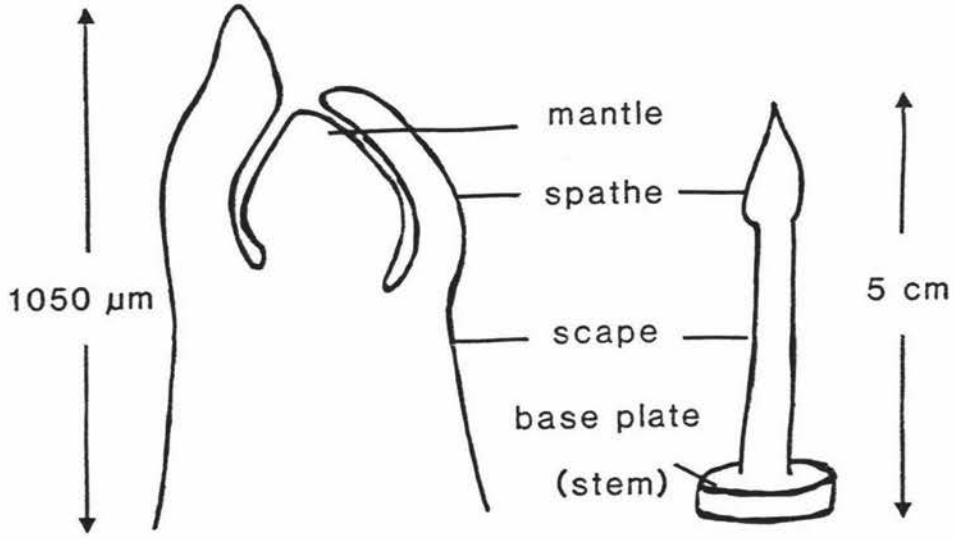

Figure 7.4: The reproductive apex produces an inflorescence consisting of stem-like scape, sheathing spathe, and a meristematic mantle which produces individual florets.

7.5 A MODEL USING FORCING FUNCTIONS

7.5.1 Introduction

A model for onion growth using environmental forcing functions allows the user to relate growth to environmental conditions, and potentially to account for or predict effects of different environments. In this section a conceptual model is outlined which relates changes in partitioning of assimilates, and indirectly carbohydrate metabolism (Chapter 2) to environment. Further work based on this conceptual model should enable development of a predictive model based on some understanding of the underlying physiology. In principle it would be possible to build the model up from the biochemical level, including the light sensing pigment phytochrome and treating carbohydrate metabolism explicitly. However, since much of the mode of action of phytochrome is still unknown (Quail, 1984), and and the problems of integrating from the cellular, much less the biochemical, level are notoriously complex (Passioura, 1973; Thornley, 1976), this would make the model much more involved to little advantage. However, basing it on what is known of gross physiology should make it possible to incorporate such information, if it becomes available, and if it improves the usefulness of the model.

7.5.2 A compartment model for an onion plant

It is useful to picture the leaf as having two notional compartments (Fig. 7.1). The 'blade' compartment, comprising emerged, photosynthesising blade, is a net source of photosynthate; non-structural carbohydrates produced in the blade by photosynthesis are exported to the the 'sheath' compartment, consisting of sheath and unemerged blade. The boundary between the two compartments is the point in space at which the blade emerges from the uppermost sheath. Most cell division and expansion takes place in the 'sheath' compartment; in monocotyledonous plants leaf tissue is largely mature by the time it emerges.

An onion plant can be considered as a series of such leaves (Fig. 7.5), at various stages of development, interconnected through the stem. The relative importance of the different inputs and outputs changes as the leaf matures (Fig. 7.5). In a growing leaf, the 'sheath' is the major sink for photosynthates. If the plant is not bulbing, they are used as structural carbohydrates for leaf extension; if the plant is bulbing, they are used as storage carbohydrates in scale tissue. Leaves that are no longer growing export carbohydrates through the stem to supply the youngest leaves (Mann, 1983), while in very young leaves, scale leaves with no photosynthetic blade, and inflorescences, all carbohydrates are imported through the stem from older, photosynthesising leaves (Fig. 7.5).

7.5.3 Bulbing

Bulbing in onions involves preferential growth of sheaths at the expense of blades, rather than the initiation of a discrete organ. Formation, growth and maturity of bulbs must therefore depend on the production of translocatable photosynthate, and the partitioning of this photosynthate between leaf blades and sheaths. In this section partitioning of photosynthate is discussed, and a model for onion growth involving light-controlled partitioning of photosynthate is presented.

All growth in an onion leaf is assumed to take place in the sheath compartment, and mature blade tissue is pushed up into the light, where the blade fixes carbon by photosynthesis. Some of this carbohydrate is lost by respiration, and the rest is assumed to be imported by the sheath rather than incorporated by the blade itself. Since bulbing is the process of interest, a mass balance for the blade is expressed in terms of the surplus available for import into the sheath, and hence for growth:

$$\begin{pmatrix} \text{rate of import} \\ \text{into sheath } I' \end{pmatrix} = \begin{pmatrix} \text{photosynthetic} \\ \text{rate } P' \end{pmatrix} - \begin{pmatrix} \text{respiration} \\ \text{rate } R' \end{pmatrix}$$

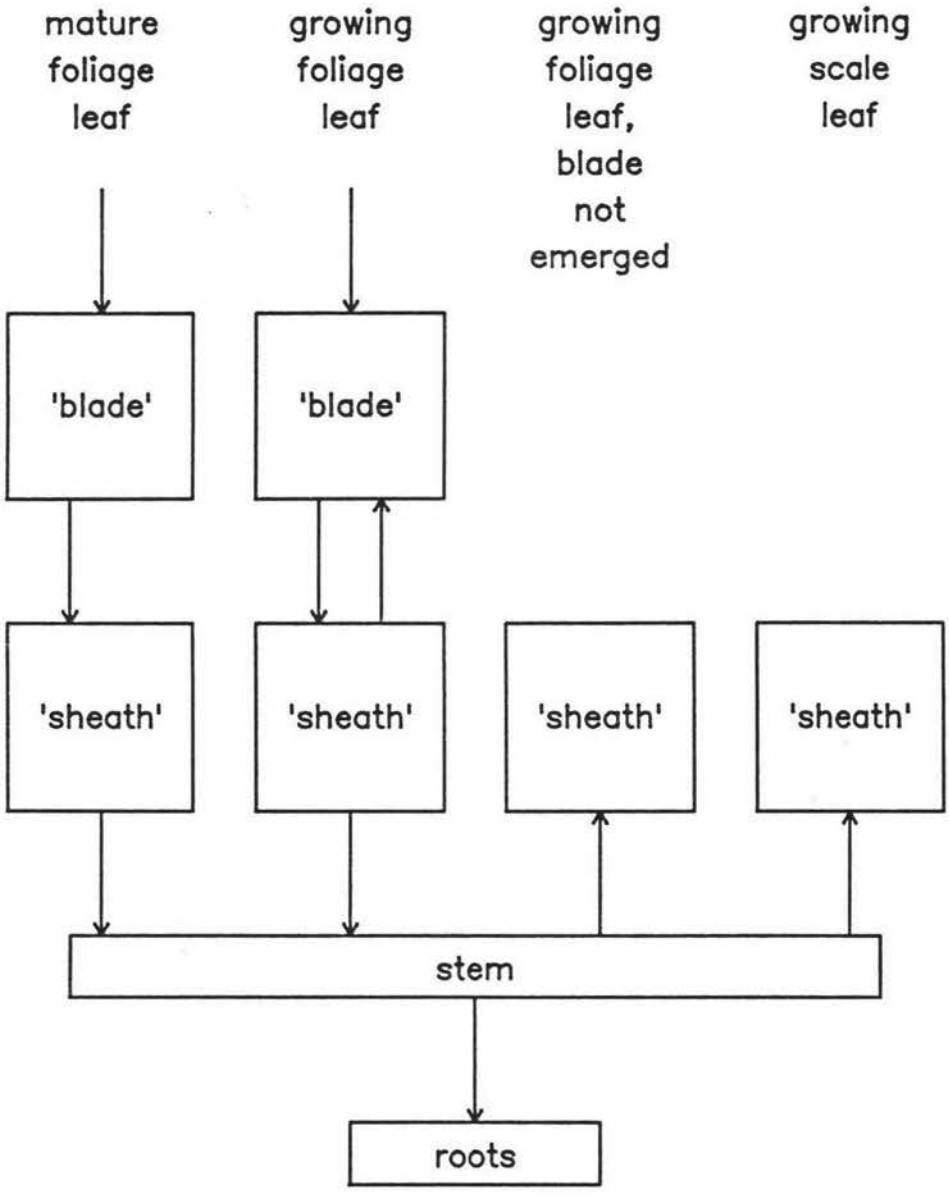

Figure 7.5: Compartment model for onion plant

$$+\left(\begin{array}{l}\text{increase in } P' \text{ from}\\ \text{growth in leaf area}\end{array}\right) \tag{7.20}$$

where the symbols are defined as for equation (11). P' can be calculated using equation (12). Respiration as a function of temperature, T, or light flux density, F, could be determined by experiment.

A mass balance for the sheath might be written in terms of G'.

$$G' = E' - R' - S' \tag{7.21}$$

for a growing leaf with with blade not emerged, which obtains its nutrition from older leaves; and

$$G' = I' - R' - S' \tag{7.22}$$

for a growing leaf with emerged blade, which is self-sufficient in photosynthate (Fig. 7.5). The rate of export of photosynthate to other tissues, E', is negative for an unemerged leaf which imports photosynthates from older leaves; and S' is rate of storage of carbohydrate in scale tissue. For a mature, non-growing, bladed leaf

$$E' = I' - R' \tag{7.23}$$

and all photosynthate not respired is exported.

G' and S' vary with daylength and temperature; in short days all the photosynthate available for growth goes to leaf extension, G, while in lengthening days an increasing amount is stored as fructans in bulb-scale tissue, S. Allocation of the net photosynthate available for growth, $E - R$ (eqn 7.21) or $I - R$ (eqn 7.22), to leaf growth and to storage could be calculated using two fitted forcing functions, $D(t)$ and $T(t)$. Then let

$$A = A(D(t), T(t)) \tag{7.24}$$

where $A(D(t), T(t))$ is the fraction of available photosynthate used for leaf extension. Then

$$G' = A.(I' - R'), \tag{7.25}$$

or, alternatively,

$$S' = (1 - A).(I' - R'). \tag{7.26}$$

Solution of a model of this sort would require initial conditions in the form of initial plant size, specifically the size of its first leaf; also the forcing functions for temperature, daylength, and light intensity would need to be known or estimated, along with factors such as the amount of growth a leaf undergoes before its blade emerges. The transitions between the three growth phases represented by eqns. (7.21), (7.22) and (7.23) are assumed to be continuous. Integration of eqn (7.25) gives an approximation to the dry weight of the scale, ignoring structural tissue (this approximation improves as the bulbs mature):

$$S = \int_0^t (1 - A(D(t), T(t))).(I' - R')dt. \tag{7.27}$$

Bertaud (1986) defined a rate of growth in dry weight for the bulb

$$W' = \sum_{all\ sheaths} I' - E' - G' - R' \tag{7.28}$$

In our notation,

$$W' = \sum_{all\ sheaths} S' \tag{7.29}$$

$$= \sum_{all\ sheaths} (1 - A).(I' - R'). \tag{7.30}$$

This ignores the net loss of dry matter to the roots.

The model discussed here is a dry weight model like that of Lancaster and Gandar (1986) discussed earlier, but because it incorporates environmental forcing functions it could, in principle, be used predictively. However, a lot of computation would be involved in using a model with so many compartments. It would be much simpler, and probably not unreasonable, to treat a whole plant (or even a crop) as two compartments, blade and scale. This would involve determining A for a plant instead of for individual leaves.

7.5.4 Sprouting and floral initiation

Sprouting in ecodormant bulbs shows a photomorphogenetic response similar to that observed for bulb growth. At first growth depends on remobilised assimilate from old bulb scales; after sprouting, photosynthesis in leaf blades supports growth. An equation similar to (7.25) could be derived to account for time to sprouting for a population and subsequent growth of plants.

The nutrient diversion hypothesis of Sachs and Hackett (1977, 1982) postulates that floral initiation is controlled by the availability of assimilates to the apex of the plant. There is also strong evidence that the maintenance of reproductive growth in onions depends on maintenance of conditions which favour reproductive over vegetative growth (Roberts and Struckmeyer, 1951; Chapter 5). The requirement of the developing inflorescence or flower for assimilates has been documented in *Gladiolus* (Robinson *et al.*, 1980), *Tulipa* (Ho and Rees, 1975, 1976) and *Rosa* (Mor and Halevy, 1979). The effect of temperature on carbohydrate metabolism of *Tulipa* bulbs was related to inflorescence development by Hobson and Davies (1978). These results suggest it is reasonable, as a first approximation, to treat inflorescence initiation and development in onions as a process dependent on intra-plant competition for assimilates.

A temperature-dependent allocation function for flowering can be derived based on a model with three compartments; a reproductive apex and a vegetative blade and sheath (Fig. 7.6). The vegetative compartments include both previously formed leaves and the bud that forms at the base of the inflorescence in the axil of the last leaf, which may grow and compete with the inflorescence if conditions are unfavourable for flowering.

An inflorescence may have one of three fates: it may develop normally and flower; it may abort at some stage of its development before flowering (generally before emergence in long-days, after emergence in short-days; Chapter 5); or it may revert to a vegetative state and continue growing (in cool short-days; Chapter 5). The

third possibility is much less common than the other two, and will be assumed to represent a case where competition for assimilates is very evenly balanced between the reproductive and vegetative compartments.

Since the inflorescence is always dependent on imported assimilates, the available photosynthate can be represented by $(E' - R')$. This could be calculated from the vegetative model described in the previous section. Rate of floral development is given by

$$K' = B(T(t))(E' - R') \tag{7.31}$$

where K' is the rate of growth in dry weight of the inflorescence and $B(T(t))$ is the temperature-dependent allocation function. Integrating,

$$K = \int_0^t B(T(t))(E' - R')dt. \tag{7.32}$$

After emergence of the inflorescence, its growth becomes photoperiod-dependent as well (Chapter 5):

$$K' = B(T(t), D(t))(E' - R') \tag{7.33}$$

In favourable conditions, the inflorescence would be expected to remain a strong sink as it grew, so $B(T(t))$ or $B(T(t), D(t))$ would tend to one. In unfavourable conditions, B would tend to zero. Conditions resulting in a value of B close to 0.5 might be expected to result in the malformed leaves and abnormal phyllotaxis observed on some plants in 15°C and 8 h days (Chapters 5, 6). Flowering of an inflorescence tends to be associated with death of the subtending leaves, so dry weight would not be expected to increase after anthesis.

7.5.5 Discussion

In this model, knowledge about the responses of onion plants to environment is related to knowledge about carbohydrate metabolism by a hypothesis about assimilate partitioning. This hypothesis could be tested by following patterns of growth

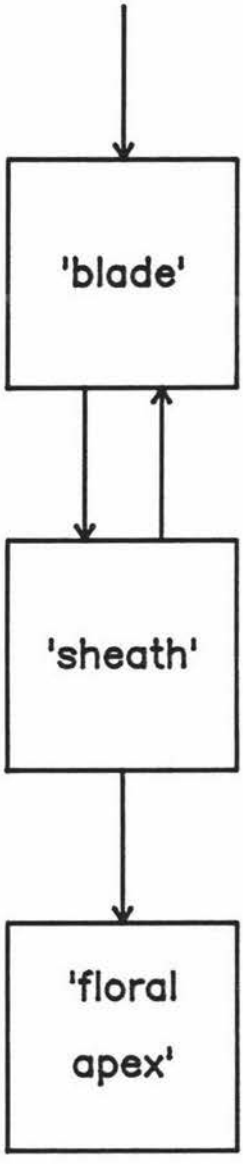

Figure 7.6: Compartment model for onion plant undergoing reproductive development

in dry weight under a range of environmental conditions, or by measurements of the redistribution of radioactively labelled carbon supplied as CO_2. Such measurements would allow estimates of partitioning functions and available photosynthate, the parameters of the model. The model could then be tested on a plant or crop where measurements of temperature and photoperiod as a function of time were available.

The model as presented is described by differential equations and is therefore deterministic. Such a model could be related to a real, variable crop by experiments to establish how the mean and distribution of the crop are affected by growth. It would then need to be tested against a different crop by measuring an initial distribution, evaluating A or B for the particular season, and predicting (say) the mean and variance of dry weight for the crop. Since onion cultivars differ in their response to photoperiod and temperature, the forms of A and B should differ as well.

The model is a summary of knowledge about the responses of onion plants to photoperiod and temperature. Specific assumptions about carbohydrate metabolism and partitioning have had to be made in its development, indicating that this area is inadequately understood. Using the model, it should be possible to make predictions about the behaviour of the plant under various temperature and photoperiod regimes. Whether the model will be of more value to physiologists, as a means of eliminating possible mechanisms of response, or to managers, as a predictive tool, depends on the results of further research.

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7.5.6 References

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Chapter 8

Conclusions

8.1 Onion physiology

This work was designed to explore the physiology of sprouting and flowering in freshly harvested onion bulbs. The experiments described in Chapter 3 were intended to test the hypothesis that sprouting and leaf growth of freshly harvested bulbs should be affected by photoperiod and temperature. The results also helped to define the nature of dormancy and its relationship to the keeping quality of onions.

The hypothesis that sprouting of onion bulbs should be affected by photoperiod appears to be correct for cultivar 'Gladalan Brown'. However, in some cultivars there is clearly another mechanism involved besides the photoperiod response. It appears that two factors stop an onion bulb sprouting after the initial formation of scale leaves: whether or not the bulb is simply ecodormant, indicated by the ability of bulbs to develop roots; and a photomorphogenetic response to long-days. 'Sakigake Yellow' and 'Gladalan Brown' were both ecodormant when planted, but sprouting is sensitive to photoperiod only in 'Gladalan Brown'. However, subsequent leaf appearance is affected by photoperiod in both cultivars. In 'Early Long Keeper', bulbs are not simply ecodormant a month after harvest, and there may be a photoperiod response involved in subsequent sprouting.

The experiment designed in Chapter 5 was designed to (a) to assess whether plants with green leaves and mature bulbs responded to chilling in the same way as unsprouted and sprouted bulbs; and (b) to explore variation between cultivars in sensitivity to duration of chilling, and to subsequent photoperiod and temperature treatments, during inflorescence initiation, emergence and flowering. From these results too it was expected that a sequence of controlled environment treatments could be defined that would allow (a) a reduced generation time, and (b) a range of cultivars to be brought into flower at the same time. The three cultivars used were 'Gladalan Brown', which is susceptible to bolting; 'Sakigake Yellow', which is resistant to bolting in the first year but flowers readily thereafter; and 'Early Long Keeper', which is intermediate in behaviour in its first year, but often flowers late or not at all in the second year, when it is very slow to sprout.

Mature bulbs, with or without green leaves, were much slower to initiate inflorescences in chilling conditions than sprouted bulbs. Bulbs of 'Early Long Keeper', most of which were rooted but not sprouted, also initiated inflorescences readily. Bulbs of 'Sakigake Yellow' were slower to initiate inflorescences, requiring a longer period of chillng than 'Early Long Keeper' and 'Gladalan Brown'. Plants exposed to warm long-days before emergence of inflorescences had a higher rate of abortion, especially in 'Sakigake Yellow'. Cool short days inhibit normal development of the inflorescence after initiation, resulting in abnormal florets, little pollen production, and abortion of inflorescences.

8.2 Development of protocols for use in breeding programmes.

Some plants of all cultivars were sprouted after 8 weeks in cool short-day conditions, and all plants given 8 weeks chilling at 8°C in short-days initiated inflorescences. Emergence of the inflorescences occurred in 8-10 weeks in cool short-days in all cultivars used, with very little abortion. The daylength could be increased to increase

the rate of floral initiation, but this carries an increased risk of inflorescence abortion. Further development of the inflorescence, however, occurred best in warm long-days. Such a protocol would be expected to bring bulbs into flower in 30-35 weeks from planting. The 8 weeks allowed for sprouting could be shortened to the time required to achieve 20% sprouting in the slowest cultivar. (In this experiment less than 20% of 'Early Long Keeper' bulbs had sprouted when the plants were transferred to chilling conditions, but no effect of this on flowering was detectable.)

The red to far-red ratio used in the cool long-day treatment would be expected to inhibit bulbing somewhat; if a lower ratio were to be used, it would be necessary to shorten the photoperiod (e.g. to 12 h) to achieve comparable results. Increasing the photoperiod to an intermediate value such as 10 h immediately after chilling might speed the emergence of inflorescences.

A method such as this would allow the breeder both to synchronise the flowering of a range of varieties, and, if bulbs were planted in January, to obtain seed in time for planting in late winter.

Appendix A

Statistical methods used in data analysis

All analyses were carried out using the SAS package (SAS Institute Inc., 1985).

A.1 ANALYSIS OF UNBALANCED DATA

A.1.1 Generalised linear models

Generalised linear models are the general class of which analysis of variance and linear regression are examples. It is particularly useful for carrying out analysis of variance on unbalanced data, such were obtained (Chapter 5) for time to flowering (Searle, 1971). The SAS procedure used was GLM, which is capable of carrying out regression, multiple regession, analysis of covariance, and analysis of variance (SAS Institute Inc, 1985).

A.2 ANALYSIS OF CATEGORISED DATA

Data where results involve classifying experimental material into categories are referred to as catagorised, or categorical, data. Since this data consists of counts of numbers in each category, the errors are not normally distributed. Classical analysis of variance techniques are therefore inappropriate. Data for sprouting (Chapters 3, 4) and stage of floral development (Chapter 5) are examples of categorised data.

A.2.1 χ^2 tests for homogeneity

This test is carried out on contingency tables with one margin fixed. In this case, the fixed margin is the number of plants in each treatment, or of each cultivar. The null hypothesis of homogeneity is that the number of plants sprouted or unsprouted is the same for each treatment (Bhattacharya and Johnson, 1977). The SAS procedure used was FREQ, which can compute χ^2 tests on multi-way tables (SAS Institute Inc, 1985). An example of the output is shown in Fig. A.1.

A.2.2 Generalised linear models

Grizzle *et al.* (1969) presented a method of analysing categorical data by linear models. One form of this is the analysis of variance used to analyse the data for sprouting and stage of floral development (Chapters 4, 5). The model may be fitted using the methods of least squares or maximum likelihood. The latter method is used to analyse the data for stage of floral development because it is more robust when a number of cells have a value of zero. The SAS procedure used was CATMOD, which can be used for linear or log-linear modelling, logistic regression and repeated measurement analysis.

SAS

TABLE OF TREAT BY TSPROUT

TREAT	TSPROUT		
FREQUENCY / PERCENT / ROW PCT / COL PCT	0	1	TOTAL
1	6	20	26
	6.90	22.99	29.89
	23.08	76.92	
	15.38	41.67	
2	19	10	29
	21.84	11.49	33.33
	65.52	34.48	
	48.72	20.83	
3	14	18	32
	16.09	20.69	36.78
	43.75	56.25	
	35.90	37.50	
TOTAL	39	48	87
	44.83	55.17	100.00

STATISTICS FOR TABLE OF TREAT BY TSPROUT

STATISTIC	DF	VALUE	PROB
CHI-SQUARE	2	10.008	0.007
LIKELIHOOD RATIO CHI-SQUARE	2	10.361	0.006
MANTEL-HAENSZEL CHI-SQUARE	1	1.957	0.162
PHI		0.339	
CONTINGENCY COEFFICIENT		0.321	
CRAMER'S V		0.339	

SAMPLE SIZE = 87

Figure A.1: Contingency table for sprouting data in Chapter 3.

A.3 References

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