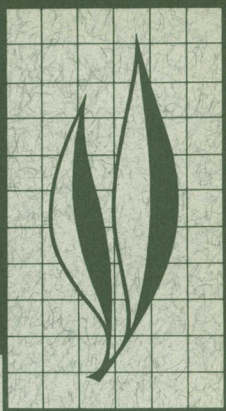


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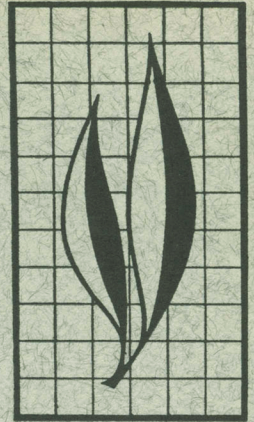
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Bulb Development in the Onion (*Allium cepa* L.) and the Effect of Storage Temperature on Bulb Rest

Abdalla A. Abdalla and Louis K. Mann



Wherever bulb onions are grown, part of the crop is usually stored. Since the bulbs can be kept for weeks or months, successful storage helps determine their availability as food.

It has long been known that onions do not sprout readily soon after harvest, even if the bulbs are planted in moist soil. Such bulbs are said to be in rest. As storage is prolonged, the resting condition disappears and the bulbs sprout easily. Storage conditions, however, may prolong the period of rest and, more important, may retard sprouting and rooting long after rest is past.

In this study data are presented on the morphological changes related to rest as the latter is measured by sprout and root production. Bulb growth and development are described for field-grown plants, and changes in bulbs leading to rooting and sprouting are followed during curing and during storage at several temperatures. The capacity of bulbs to root and sprout was measured by removing samples from storage to conditions favorable to bulb growth. Cell divisions in the stem apex were observed during curing and under several storage conditions, to relate stem growth to sprouting. The findings are presented in relation to the practice and problems of onion bulb storage.

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Bulb Development in the Onion (*Allium cepa* L.) and the Effect of Storage Temperature on Bulb Rest^{1, 2}

INTRODUCTION

MATURE ONION BULBS can be stored for several months or transported for long distances before use. The suitability of onions for storage or transportation depends on the variety (cultivar) and on cultural and storage methods. Apart from economic considerations, the maximum storage life of bulbs is limited by weight loss, storage diseases, or regrowth (appearance of sprouts and roots).

Stored onion bulbs that are neither sprouting nor rooting are described as dormant. Dormant bulbs may root and sprout quickly if placed in favorable conditions. Fully mature bulbs planted immediately or shortly after harvest may not sprout, however, and are said

to be in rest. For most cultivars stored under favorable conditions, rest ends long before the useful storage period is over. (Rest in onions may be prolonged by application of chemicals, an aspect of storage not considered in this paper.) Although rest in bulbs is of interest to physiologists, onion breeders, and seed producers, it has received surprisingly little attention.

The present study describes the development of the onion plant in the field, the morphological changes that occur during storage, the duration of rest, especially as affected by storage temperature, and changes in mitotic activity in the shoot tip from before harvest through storage at three temperatures.

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² A portion of a dissertation presented to the Graduate School of the University of California in partial fulfillment of the requirement for the degree of Doctor of Philosophy.

BULB DEVELOPMENT IN THE FIELD AND BULB GROWTH AT THREE STORAGE TEMPERATURES

Numerous workers have studied the development of the onion bulb, among them: Sachs (1863); Boswell (1924); Hoffman (1933); Hayward (1938); Heath and Mathur (1944); Heath (1945); Heath and Holdsworth (1948); Rauh (1950); and Aoba (1954). The following discussion is based on their findings.

The primary root and the lower portion of the cotyledon emerge from the onion seed during germination. The cotyledon soon folds to form the "knee," elongates rapidly upward, becomes green, and finally unfolds to an erect position. At this time the first adventitious roots emerge from the stem, and the first foliage leaf, elongating within the tubular sheath of the cotyledon, exserts its blade from the pore at the top of the cotyledon sheath. The young plant continues to grow by initiation of new leaves at the shoot apex and new adventitious roots on the stem. The new roots always arise from the younger (upper) part of the shoot.

A leaf primordium, first visible as a lateral protuberance on the shoot apex, soon extends itself around both sides of the apex to form a ring, somewhat higher on one side than on the other. The upward growth of this ring-like primordium produces the tubular leaf sheath and the higher side elongates to form the leaf blade. Meanwhile, the apical meristem, in like manner, produces a second foliage leaf, the blade of which emerges on the opposite side of the apex. Leaves are thus distichous and, because the internodes fail to elongate, appear to be arranged in two compact rows. The youngest leaf and the shoot apex are always enclosed in the tubular bases of the preceding leaves. Continued lateral expansion of the upper surface of the stem, and of the sheath bases attached to it, provides

space for newly formed leaves. The lack of internode elongation and the presence of the tubular leaf sheaths, open only at the top and forming an apparent or false stem, account for the characteristic habit of the vegetative onion plant.

Before changes in day length and temperature initiate bulbing, leaves bearing blades are continuously produced. With the onset of bulbing, the formation of leaf blades is inhibited, so that bladeless sheaths are produced. These, along with the sheaths of previously formed, bladed leaves, increase in thickness by cell enlargement and cause the swelling of the bulb. Commonly associated with bulbing is the production of lateral buds (within the main bulb) which may also function as storage structures. Following the formation of several bladeless leaves associated with bulbing, several small, unelongated, bladed leaves again arise from the shoot apex of the mature bulb. The bulb thus consists of three distinct types of leaves produced in sequence. Finally, the old leaf blades and some of the outer leaf sheaths collapse and die, root emergence ceases, and the bulb becomes "mature." Later, when dormancy is past, the primordial bladed leaves at the bulb center may emerge as sprouts.

MATERIALS AND METHODS

The onion cultivar 'Excel,' a 'Yellow Bermuda' type first selected in Texas (Jones, Perry, and Davis, 1947), was used. Grown mostly in Texas and Southern California, 'Excel' was chosen because of its early maturity, remarkable uniformity of size and color, and presumably short rest.

Data on 'Excel' were obtained during the 1960 and 1961 seasons. The 1960 crop was direct-seeded at Davis on March 3, 1960, and harvested June 22, 111 days later, when half the tops were

down. The mature bulbs averaged about 5 cm in diameter. Samples of 10 plants were dissected on March 30 and at two-week intervals thereafter until harvest.

The 1961 crop was direct-seeded at Davis on December 31, 1960, and harvested June 30, 1961, 181 days later, when the tops had fallen over. Samples of 10 plants were first dissected on April 6 and then at weekly intervals until harvest. In both plantings, leaf number (cotyledon = leaf No. 0) was determined by the use of tags placed on young leaves in the field. For the second planting, in which dissections were continued on bulbs in storage, the base of leaf No. 8, an outer scale, was marked with India ink when the bulbs were harvested.

The harvested bulbs were first cured in crates for one week in an uninsulated warehouse. The tops and roots were then removed, and the bulbs were stored in well-ventilated crates at 0°, 15°, and 30° C. The relative humidity and the temperature controls of these storages are described on page 93. Ten bulbs were dissected weekly by removing successive leaves until the shoot apex was exposed. Changes in total leaf number during storage were recorded. The mean length of leaf No. 12, one of the inner, bladed leaves which was part of the potential bulb sprout, was measured to assess the effect of storage temperature on sprout elongation.

RESULTS OF FIELD TESTS

1960 Season. When the 1960 plants were first examined on March 30, the cotyledons were erect and green and the first foliage leaf was exerted on about half of the seedlings. The second foliage leaf had been differentiated in all plants, and the third in about half of the plants. From this time until harvest, leaves were initiated at a rather uniform rate of about one per week, so that by the time the bulbs were mature, 15 or 16 leaves had been differentiated.

Each seedling had about three roots on March 30, and new roots were added at a rate of nearly four every week until

a maximum number of approximately 35 was reached on May 26, well after bulbing had started. Roots then died more rapidly than they were produced, so that each plant averaged about 19 roots at harvest. The primary root, being central and thickest, can be distinguished for one or two months after seedling emergence.

Bulb enlargement was first associated with the thickening of the foliage leaf bases (roughly the lower third of the sheath), which ultimately made up the bulk of the mature bulb. As bulbing progressed, however, the bladeless sheaths, formed soon after bulbing began, also thickened. By June 22, when the bulbs were harvested, the first three or four leaves produced either had disappeared, or persisted as dry sheathing scales. The bulbs had an average of seven thickened scales; the outer four were bases of foliage leaves and the inner three, sheaths of leaves with aborted blades. Finally, within these storage leaves were five or six small, bladed leaves, the potential sprouts.

The leaves described above arose from the main axis. Each 'Excel' bulb had at least one lateral bud, however, and some as many as three. These buds were axillary to the innermost foliage leaves or to the outer bladeless leaves, and were initiated when the bulbs began to enlarge. They consisted either of thickened, bladeless sheaths surrounding small, bladed leaves or of small, bladed leaves only. In some bulbs the lateral buds made up as much as one-third of the bulb's weight. The small, innermost leaves of the lateral buds, like those of the main axis, elongate when the bulb sprouts.

1961 Season. When first dissected on April 6, the plants were in about the same condition as those dissected on March 30 of the previous year. Since development during the two years was remarkably similar, and since observations in 1961 were made weekly, data on root and leaf production are given only for 1961 (figs. 1 and 2).

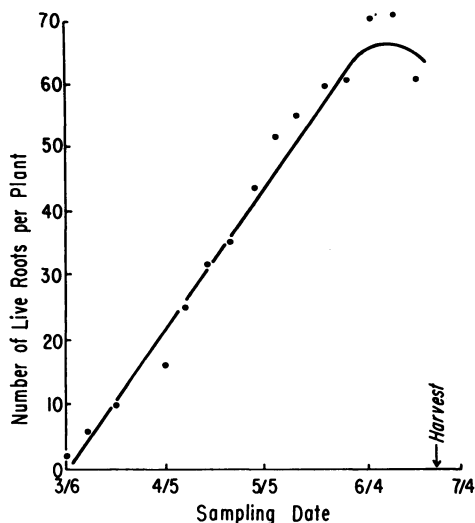


Fig. 1. Number of live roots in 'Excel' onion plants during growing season. Each point is based on 10 plants. Plants direct-seeded in field at Davis, December 31, 1960.

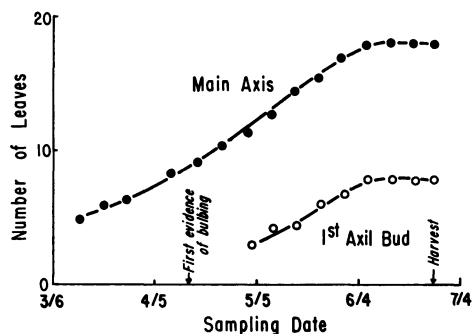


Fig. 2. Pattern, in time, of leaf initiation on main axis and first axillary bud of 'Excel' onion plants from early seedling stage to harvest. In order of initiation, the first 8 to 9 leaves were bladed, the next 3 to 4 bladeless, and the last 5 to 6 bladed primordia. Each point is a mean from 10 plants. Plants direct-seeded in field at Davis, December 31, 1960.

Adventitious roots emerged at a rate of four to five per week until about three weeks before harvest, when each plant had about 70 live roots. Death of older roots decreased this number to about 60 per plant at harvest (fig. 1).

The pattern in time of leaf initiation on the main shoot and on the first axillary bud during the growing season is shown in figure 2. Leaves were initiated at the rate of about one per week. Leaf initiation ceased 20 days before harvest, by which time each plant had produced 17 to 18 leaves on its main axis. Aoba (1954) reported that leaf initiation ceased at the bulbing period. Although he did not specify the bulbing date, his finding substantiates the observation made in this study that leaf initiation ceases while the plant tops are still erect and green in the field. Heath and Holdsworth (1948) observed that leaves were initiated about once every two weeks in the first part of the growing season and at an enhanced rate after bulbing began. They did not state when leaf initiation ceased.

In 1961 the number of visible green leaves reached a maximum of seven per plant and then decreased because of shedding of the dry, older leaves and the cessation of the production of leaf blades from the younger, inner leaves. Aoba (1954) reported that the number of visible green leaves in three Japanese onion cultivars increased up to about six and then decreased through the season.

Bulbing was first evident about four months after planting. The neck-to-bulb ratio (diameters) was then 0.31. At that time leaf-base thickening was confined to the three or four outermost, nondry, bladed leaves. A total of eight or nine bladed leaves was produced, followed by three or four bladeless leaves, and finally by five or six unelongated, bladed leaves (the potential sprout). The bladeless sheaths and the bladed leaf bases exterior to them (except for three or four which withered by the time the bulb was mature) thickened to form the bulk of the bulb.

The lower portion of the sheath thickens during bulbing by cell enlargement. In longisection, the number of cell layers in the neck and in the lower sheath were equal, but cell size was greater in the latter. No cell divisions

were evident in either portion of the sheath. Heath and Holdsworth (1948) reported that sheath cells enlarge *after* maturity, but gave no precise meaning to the term maturity. They noted that, for onion sets, all foliage leaf bases collapse and all storage is in the scales, i.e., bladeless leaves. In the plants we studied, foliage leaf bases were always involved in bulb formation. Aoba (1954) also concluded that the thickness of the bulbs is caused by cell enlargement in both the foliage leaves and the scales.

A lateral bud, usually in the axil of leaf 7, 8, 9, or 10, was first evident on April 25, when bulb enlargement began. In a number of plants, a second lateral bud was found in a later-formed leaf, and occasionally this bud also contained an additional lateral bud. New leaves in the first lateral bud were initiated at the same rate as in the main shoot (fig. 2). At maturity, the first lateral buds had seven to eight leaves, the outer ones bladeless and thickened, the inner bladed but very small.

We attempted to follow root initiation on the 1961 crop. A longitudinal half of the stem of each of 10 plants was cut crosswise into slices 250 μ thick, and these were examined under a binocular microscope. Samples were first taken on April 20 and at 10-day intervals until the end of storage (15° C).

The number of unemerged root primordia per plant varied widely during the growing season, possibly in response to field irrigation. For individual bulbs, it was not known whether water contacted the stem plates during irrigation. Because of this, the data are difficult to interpret, and further data should be taken under rigorously controlled moisture conditions.

Although the number of primordia appeared to increase between a sampling just before harvest (June 29) and the first sample from storage (July 9), bulbs always have many primordia, and root emergence at any time appears to be independent of root initiation. Thus, in the sampling on July 9, and in three

subsequent samplings in storage before the bulbs rooted, bulbs averaged between 42 and 43 primordia each.

Storage Observations. Elongation of the sprout-forming leaves (as exemplified by leaf No. 12) in the 1961 'Excel' bulbs stored at 0°, 15°, and 30° C is shown in figure 3. Leaf elongation was not evident at any temperature for the first two weeks in storage and was slight or absent for the entire 10 weeks of storage at 0° and 30° C. Inner leaves of bulbs stored at 15° C started to elongate after two weeks in storage, but visible sprouts were not produced until two months after harvest (fig. 4). The inner leaves of lateral buds elongated at the same rate as those of the main shoot.

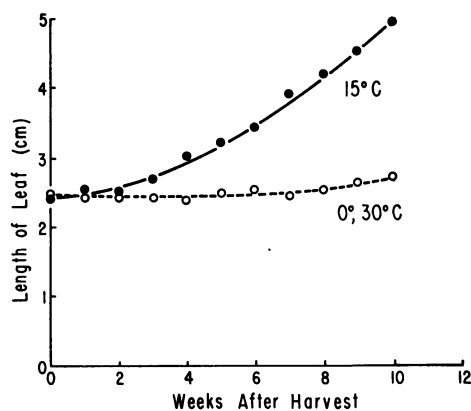


Fig. 3. Storage temperature and elongation of an inner bladed leaf (number 12) of 'Excel' bulbs stored at 0°, 15°, and 30° C immediately after harvest. The 30° and 0° C storages gave similar results; each circle is the mean of 20 bulbs. Each point on the 15° C curve is a mean of 10 bulbs.

Leaf initiation was resumed in bulbs stored at 15° C at about the same time as or somewhat earlier than leaf elongation, and four to five leaves had been initiated by the time the sprouts were visible (fig. 5). Leaf initiation was much slower in bulbs stored at 0° or 30° C. Heath and Mathur (1944) found that under ordinary storage temperatures (not specified) only one or two leaf

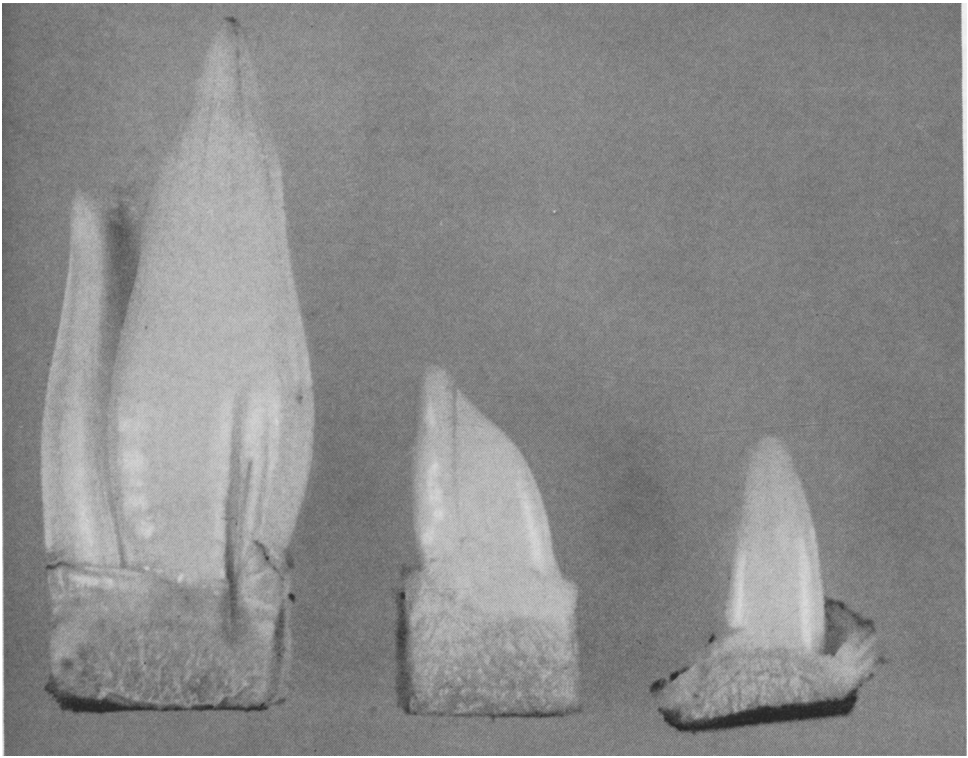


Fig. 4. Elongation of inner leaves of 'Excel' onion bulbs stored for five weeks at 15° C (left), 0° and 30° C (right). Exterior leaf on each bud is leaf number 10.

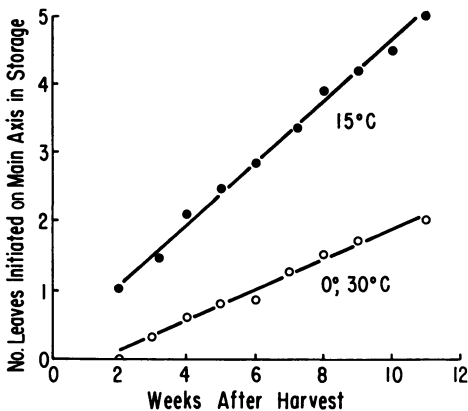


Fig. 5. Pattern, in time, of leaf initiation on main axis of 'Excel' onion bulbs stored at 0°, 15°, and 30° C. The 30° and 0° C storages gave similar results; each circle is the mean of 20 bulbs. Each point on the 15° C curve is a mean of 10 bulbs.

initials were produced in storage, mainly during the latter part of winter. Aoba (1954), dissecting stored bulbs of the Japanese cultivars 'Imai-wase,' 'Sapporo-ki,' and 'Aichi-shiro,' observed that new leaf initials were formed when the inner scales began to elongate, long before the sprouts were visible. Since sprouts are formed by the elongation of leaves initiated before harvest, sprouting appears to be independent of leaf initiation during storage.

In our bulbs stored at 15° C, root emergence was evident six weeks after the beginning of storage. Bulbs stored at 0° or 30° C remained without roots for the entire storage period.

EFFECT OF TEMPERATURE ON REST AND DORMANCY

Emilsson (1949), in studies on the potato, defines rest and dormancy as follows: "The term 'rest period' will refer to the period immediately following harvest during which the potato tuber will not sprout even under optimal sprouting conditions—the term 'dormant period' will signify that period during which the tuber may be stored at some temperature suboptimal for sprouting without beginning to sprout or break down physiologically." He also notes that dormancy includes and may coincide with the rest period. The same definition may be applied to rest and dormancy in the onion, with the added point that other factors, as well as temperature, may control dormancy. Boswell (1924) used similar terms, but considered that the dormant period followed and did not include the rest period. Some workers have used the terms interchangeably (Howard, 1910; Miller, 1938) or have added terms, such as quiescence (Meyer and Anderson, 1952; Bonner and Galston, 1952). Whatever the terminology, the growth processes indicating the end of rest or dormancy have almost always been conspicuous macroscopic changes, such as the unfolding and elongation of buds.

Rest in Onion Bulbs. In one of the earliest papers on rest and dormancy in onions, Jones (1921) concluded that onion bulbs were in a state of rest for some time following harvest and that the length of this rest period varied among cultivars. By periodically removing and planting bulbs from storage, Jones showed that rest disappears with time. He was also able to stimulate both rooting and sprouting by wounding the bulbs in various ways. Boswell (1924) concluded that the cultivar 'Yellow Globe Danvers' has a rest period of eight weeks when planted in warm, moist soil. Loomis and Evans (1929) stored onion sets at 40°, 86°, and 102° F and made weekly plantings for four weeks. They found that bulbs from the

102° F storage sprouted most quickly. Lots stored at 86° and 40° F, and the check, which was planted immediately after harvest, sprouted less rapidly, in the order listed. Sprouting temperature was not indicated, and some results were conflicting because of partial depletion of rest before the storage tests began.

While few studies have been made on storage of onion bulbs under controlled conditions followed by periodic plantings to conditions favorable to sprouting and rooting, an extensive study of this type was made on garlic bulbs by Mann and Lewis (1956). They stored garlic bulbs at a series of temperatures from 0° to 20° C and made periodic plantings to flats of moist sand kept at controlled temperatures. The mean days to 50 per cent sprout and root emergence were determined. Sprouts and roots emerged most quickly after pre-planting storages of 5°, 10°, or 15° C. Bulbs stored at 20° C sprouted and rooted more slowly; bulbs stored at 0° C were intermediate. For all storage temperatures, the time required for sprout and root emergence decreased as the time in storage increased. Mann and Lewis concluded that both roots and sprouts were in rest when garlic reached maturity. Bulb storage temperature also affected growth subsequent to sprouting and rooting, but this effect was shown to be separate from that of temperature on rest.

Most investigators to date have used externally visible sprouting or rooting as the first indication of resumption of growth and consequently the termination of the rest period. That this may be a rather arbitrary criterion is indicated by observations of J. P. Bennett (unpublished), who found that leaves within peach buds increased in number over a long period, although externally visible changes in the buds were slight. He maintained the resting condition of peach tree buds by bringing plants into a warm greenhouse after they had gone into the resting state in the autumn.

Periodic dissections showed that new leaves were being continually produced within the unexpanded buds. Davidson (1958) noted that the appearance of a sprout on a potato tuber is not actually the first sign of rest break, but only the first conspicuous indication of growth which had been progressing from the time of harvest. He placed freshly dug potato tubers in storage at 39° and 86° F. Samples were periodically removed, and median longitudinal sections of the sprout initials of the apical eyes were measured. Growth, as evidenced by elongation, was continuous at both temperatures, but most rapid at 86° F.

Storage Life of Onion Bulbs. While surprisingly little research has been done on rest in onions, extensive data are available on the storage life of mature bulbs, i.e., on factors affecting dormancy (not distinguished from rest). Magruder *et al.* (1941) studied the principal onion cultivars grown in the United States and classified them as poor, intermediate, fair, etc., with reference to their storageability. These workers concluded that the storage quality is consistent within each cultivar, despite considerable variability in the production methods, time of maturity, and storage conditions. Normal maturity and freedom from field diseases and insects are necessary for any cultivar to store well.

Mann and Lewis (1956) reviewed the studies on the responses of onions to the storage environment, primarily to temperature and humidity. Their findings may be summarized as follows: In most of the studies on onion storage, bulbs were held at a series of temperatures, occasionally with humidity control, and inspected periodically for storage losses. Most of the studies showed that bulbs produce roots and sprouts more quickly at intermediate temperatures than at low or high storage temperatures. A constant temperature of 32° to 35° F usually results in the highest yield of sound bulbs. Storage at 86° F will keep

bulbs unsprouted for as long as a year, but weight loss may be high. Root elongation, in contrast to sprouting, is closely associated with high humidities.

Recent findings by Yamaguchi *et al.* (1957) and Pratt and Morris (1958) are fairly typical. These authors stored 'South Port White Globe' onion bulbs for four months at seven constant temperatures ranging from 0° to 40° C. Relative humidities at the different temperatures were moderate but uncontrolled. Storage at 0° C gave the highest yield of sound bulbs at the end of the four-month storage period. The highest storage temperature, 40° C, was next best; the intermediate temperatures of 10°, 15°, and 20° C gave the lowest returns after four months of storage. Bulbs from 0° and 7.5° C storage appeared fresher than bulbs from 25° to 35° C. Yellow centers, "indicating sprout initiation," were found after storage at 10° and 15° C.

While these studies on continuous storage of onions show the influence of storage temperature and humidity on sprouting and rooting, they provide little evidence on either the presence or duration of rest.

Similar studies on potato tubers and gladiolus corms were reviewed by Mann and Lewis (1956). The effect of temperature on rest in the potato tuber is still not clear, but gladiolus corms lose rest more quickly at 5° to 10° C than at higher temperatures, a reaction similar to that observed in garlic (Mann and Lewis, 1956).

MATERIALS AND METHODS

The onion cultivars used were 'Excel' and 'Australian Brown 5,' an onion released in 1935 (Porter, Jones, and Davis, 1941). The latter, a uniform strain of good keeping quality, was selected because it was known to have a long dormancy.

Both cultivars were direct-seeded at Davis on December 31, 1960. 'Excel' was harvested six months later when the tops had fallen over. 'Australian

Brown 5' was harvested about a month after 'Excel.' During the week following harvest, the bulbs of each cultivar were cured in slatted crates in a well-ventilated onion warehouse where day temperature was approximately 25° to 30° C and night temperature, 15° to 20° C.

The dried tops and roots were removed immediately after curing and a control lot of 30 uniform, sound bulbs of each cultivar was planted in a flat of moist sphagnum. The flat was held in a 15° C storage room, but the temperature of the sphagnum remained about 1.5° C below that of the room, because of evaporative cooling from the sphagnum.

The remaining bulbs were transferred to controlled-temperature storage rooms held at 0°, 5°, 10°, 15°, 20°, 30°, and 40° C. The temperature in the storage rooms usually remained within $\pm 0.5^\circ$ C of the indicated temperature. The humidity in the rooms was not controlled, but was periodically checked. In

all the storage rooms except those at 30° and 40° C, the relative humidity varied between 95 and 98 per cent. In the 30° room, it varied between 80 and 85 per cent, and the 40° room ran as low as 20 per cent.

Random samples of 30 uniform, sound bulbs were removed from each of the storage temperatures after 2, 4, 8, and 16 weeks. These were planted in flats of moist sphagnum and held in the same manner as the control.

Daily counts were made of the number of bulbs from which at least one root had emerged to a length of 5 mm or more. After a bulb was recorded as rooted, it was not disturbed again. Counts of visible sprouts were made every other day. The time required for 50 per cent of the bulbs to root or to sprout was estimated from a graph of the number of rooted or sprouted bulbs vs. observation dates (fig. 6). The mean days for sprout emergence were also calculated by multiplying the number

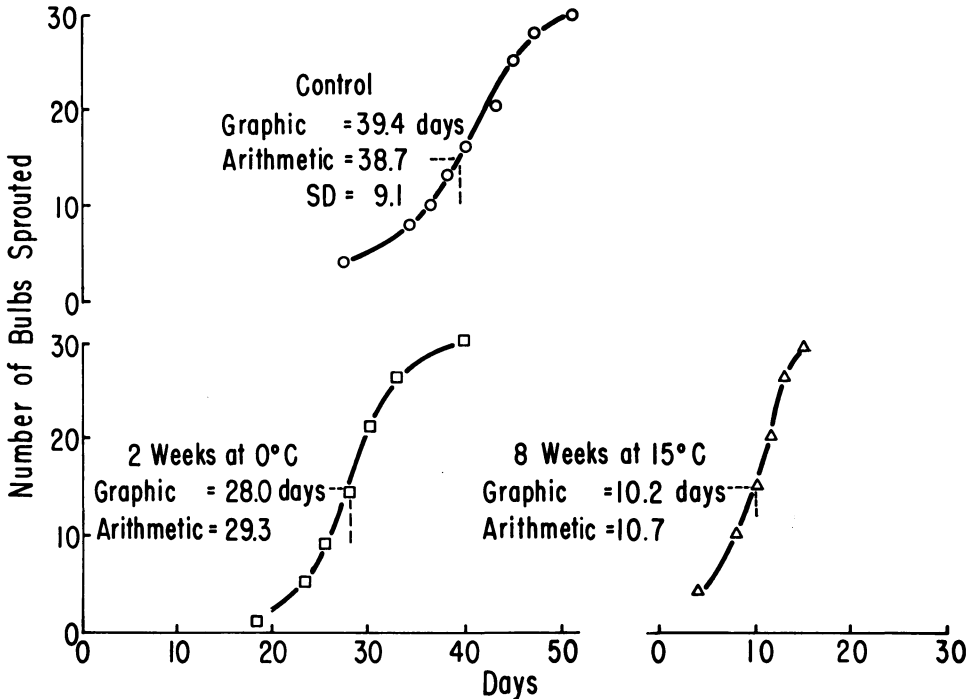


Fig. 6. Graphic and arithmetic mean estimates of days to 50% sprouting for 'Excel' bulbs at two storage temperatures and three storage periods. SD=standard deviation in days from planting to mean sprout emergence.

of the new sprouts on any observation date by the number of days since planting, summing all such products, and dividing by the total number of bulbs sprouted. This arithmetic-mean method, used by Kotowski (1927) for estimating seedling emergence, should be equivalent to days until 50 per cent sprout emergence estimated graphically if emergence is normally distributed with time. Values for the two methods (fig. 6) were sufficiently close so that the simpler graphic method of estimation was used.

RESULTS OF DORMANCY STUDIES

Sprout Emergence

'Excel.' Total sprout emergence was nearly 100 per cent in all the tests, with 93 per cent the lowest level observed. The range in days for total sprout emergence was relatively narrow, as indicated by the estimates of the standard

deviations of the populations (fig. 6). In the check treatment, for example, all sprouts emerged within 24 days after the first sprout had appeared, and the time to 50 per cent sprout emergence was 39.4 ± 9.1 days.

Figure 7 shows the number of days to 50 per cent sprout emergence of onion bulbs stored at 0° , 5° , 10° , 15° , 20° , 30° , and 40° C for 0 (control), 2, 4, 8, and 16 weeks and sprouted at 15° C. Pre-planting storage temperature clearly had a marked effect on the rapidity of sprouting. For any storage period, the most rapid emergence occurred after storage at 5° , 10° , and 15° C. Bulbs from 30° C storage sprouted most slowly; those from 0° C storage sprouted almost as slowly. Storage at 40° C for 2 or 4 weeks was more effective in decreasing the time for sprout emergence than was storage for similar periods at either 0° or 30° C. Bulbs stored at 40° C for more than 4 weeks were desiccated; therefore no sprout emergence data are

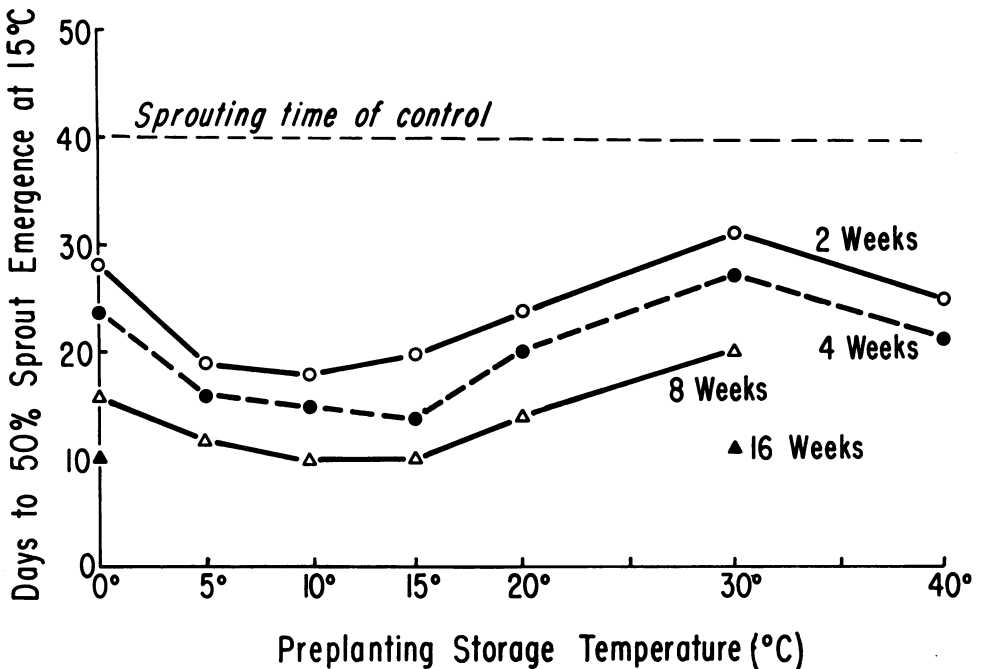


Fig. 7. Days to 50% sprout emergence for 'Excel' bulbs planted in moist sphagnum at 15° C after storage at the indicated temperatures for 2, 4, 8, and 16 weeks. Control was placed in sphagnum without storage. The 16-week storage graph is incomplete because bulbs held 16 weeks sprouted in storage except at 0° and 30° C. Each point is based on a 30-bulb sample.

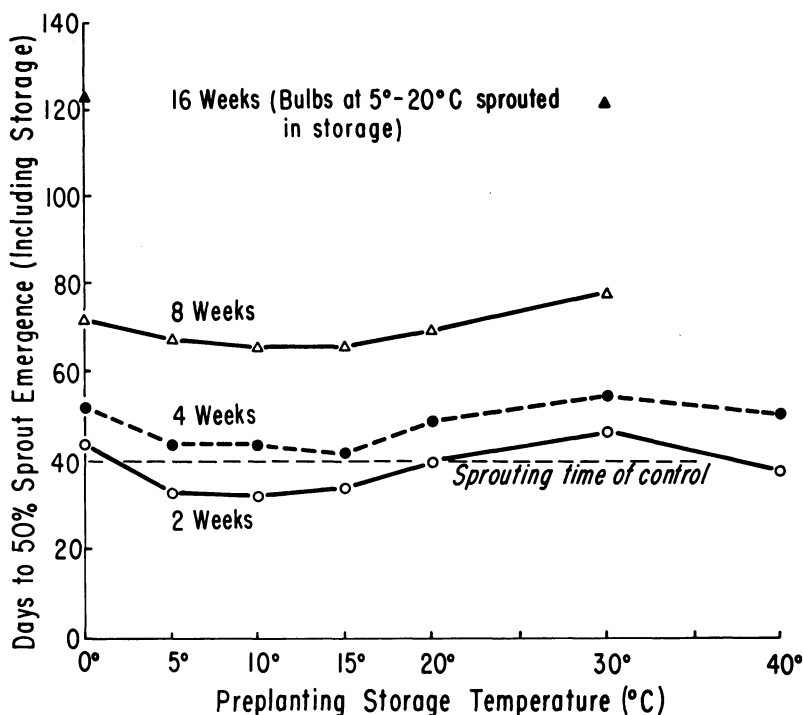


Fig. 8. Total time from beginning of storage to 50% sprout emergence for 'Excel' bulbs stored at indicated temperatures for 0, 2, 4, 8, and 16 weeks and then placed in moist sphagnum at 15° C (compare with fig. 7).

shown. The longer the bulbs were held, regardless of storage temperature, the more quickly they sprouted. This indicates that rest disappeared with time at all the storage temperatures used and that at none of the temperatures was the level of rest intensified.

Figure 8 shows the total number of days from the beginning of storage to the day of 50 per cent sprout emergence for the bulb lots shown in figure 7. This graph is interesting because it shows that bulbs stored for two weeks before planting sprouted so much more quickly than the control bulbs that the total time from the end of the cure to 50 per cent sprouting was roughly equal for bulbs planted immediately and for bulbs stored two weeks before planting. Actually, the bulbs stored for two weeks at 5°, 10°, 15°, and 40° C sprouted before the control. We have no explanation for this earlier sprouting, since the control was in a flat of moist sphag-

num at 15° C, a temperature which appears to be near the optimum for the depletion of rest. That the observation is real, however, is indicated by figure 11, a similar graph for 'Australian Brown 5,' in which sprouting in both the two- and four-week storage lots overlaps that of the control.

'Australian Brown 5.' The total sprout emergence of 'Australian Brown 5' was generally lower than for 'Excel.' Figure 9 shows the variability among the bulbs in days to sprout emergence. 'Australian Brown 5' sprouts slowly, and the range in days over which sprouts emerged was much wider than for comparable treatments on 'Excel.' For example, the time between the emergence of the first and last sprout in the check treatment of the 'Australian Brown 5' was 62 days, while that of 'Excel' was 24 days. The time to 50 per cent sprout emergence for the check treatment was 60 ± 11.7 days for 'Aus-

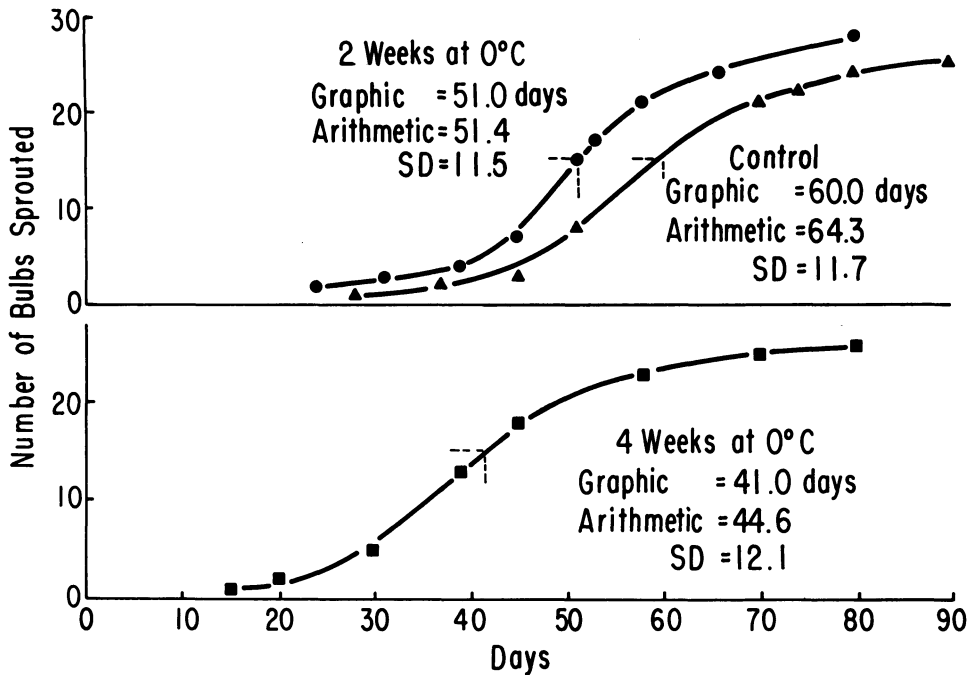


Fig. 9. Graphic and arithmetic mean estimates of days to 50% sprouting for 'Australian Brown 5' bulbs at two temperatures and three storage periods. SD = standard deviation in days from planting to sprout emergence.

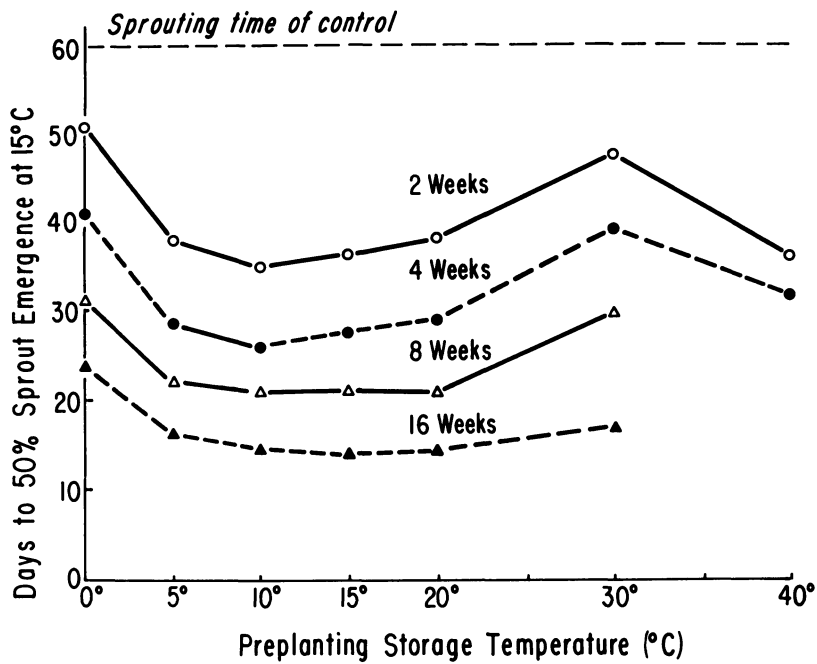


Fig. 10. Days to 50% sprout emergence for 'Australian Brown 5' bulbs planted in moist sphagnum at 15° C after storage at the indicated temperatures for 2, 4, 8, and 16 weeks. Control was placed in sphagnum without storage. Each point is based on a 30-bulb sample.

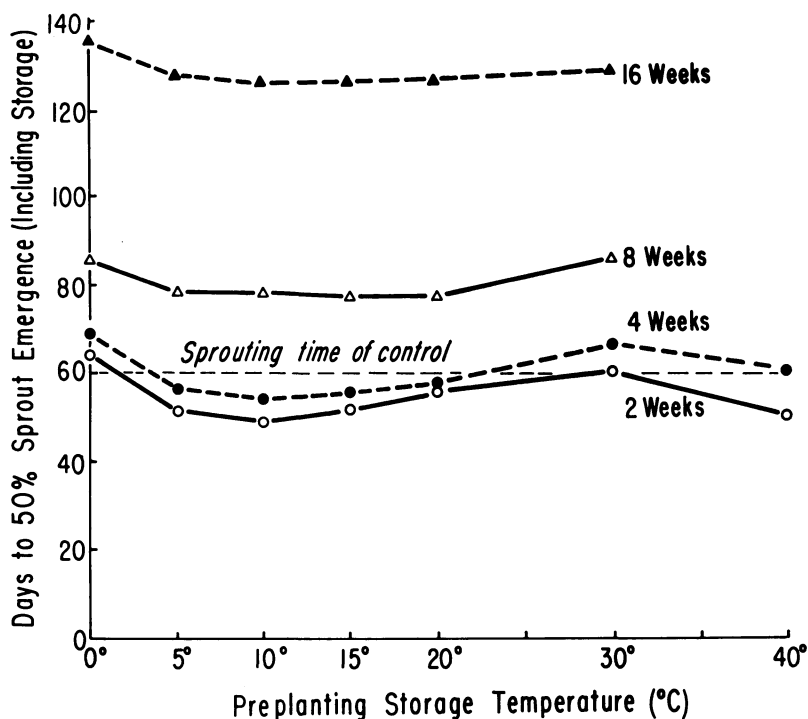


Fig. 11. Total time from beginning of storage to 50% sprout emergence for 'Australian Brown 5' bulbs stored at indicated temperatures for 0, 2, 4, 8, and 16 weeks and then placed in moist sphagnum at 15° C (compare with fig. 10).

tralian Brown 5' and 39.4 ± 9.1 for 'Excel'; the coefficient of variation for both was nearly 20 per cent. Figure 10 shows that, despite slower sprouting and rooting, the sprouting response to storage temperature of 'Australian Brown 5' was remarkably similar to that of 'Excel.' However, neither rooting nor sprouting, as seen in 'Excel,' was observed during the 16-week storage treatment. Also, in contrast to 'Excel,' storage at 20° C was nearly as effective in depleting rest as was storage at 5°, 10°, and 15° C. In common with 'Excel,' bulbs stored at either 0° or 30° C sprouted most slowly. The rather uniform period required for 50 per cent sprouting for all storage temperatures after 16 weeks suggests that rest had essentially disappeared by that time (except at 0° C). Figure 11 shows the total time from the beginning of storage to the day of 50 per cent sprout emer-

gence. Storage at temperatures between 5° and 20° C enhanced sprout emergence. This confirms the results obtained for 'Excel' and, in the sense that sprouting occurs more quickly as storage is prolonged, substantiates the evidence for a rest period in onion bulbs. Storage at either 0° or 30° C depletes rather than intensifies rest. As with the 'Excel' bulbs stored for two weeks at 5°, 10°, 15°, and 40° C, the 'Australian Brown 5' bulbs stored at 5°, 10°, 15°, and 20° C for two and four weeks before planting sprouted sooner than the control bulbs, which had been placed in moist sphagnum immediately after curing.

Root Emergence

The influence of preplanting storage temperatures on root emergence is shown for 'Excel' in figure 12 and for 'Australian Brown 5' in figure 13.

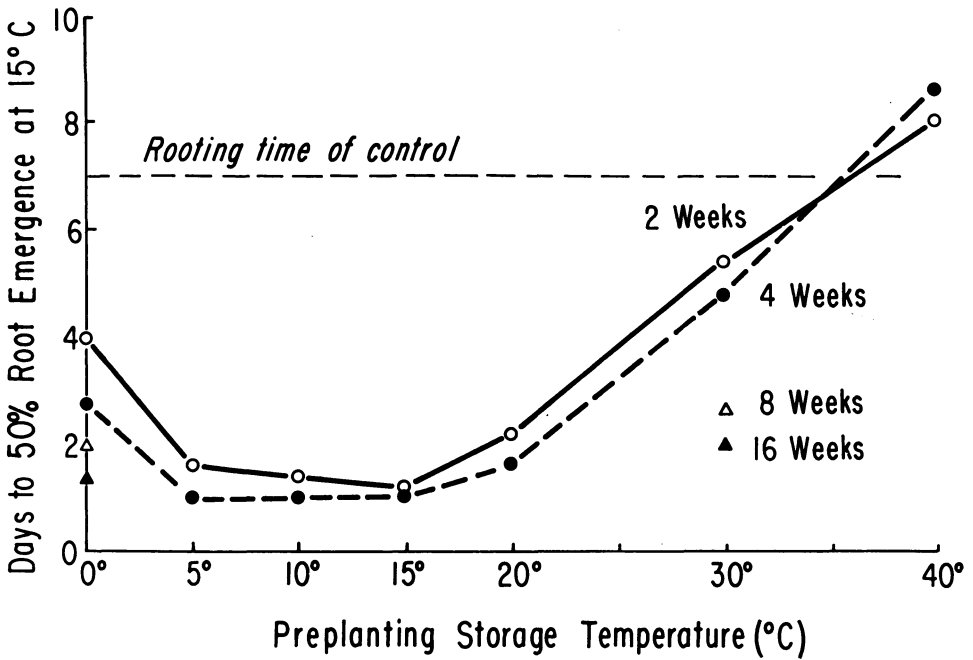


Fig. 12. Days to 50% root emergence for 'Excel' bulbs planted in moist sphagnum at 15° C after storage at indicated temperatures for 2, 4, 8, and 16 weeks. Control was placed in sphagnum without storage. The 8- and 16-week storage graphs are incomplete because lots held for those periods rooted in storage except at 0° and 30° C. Each point is based on a 30-bulb sample.

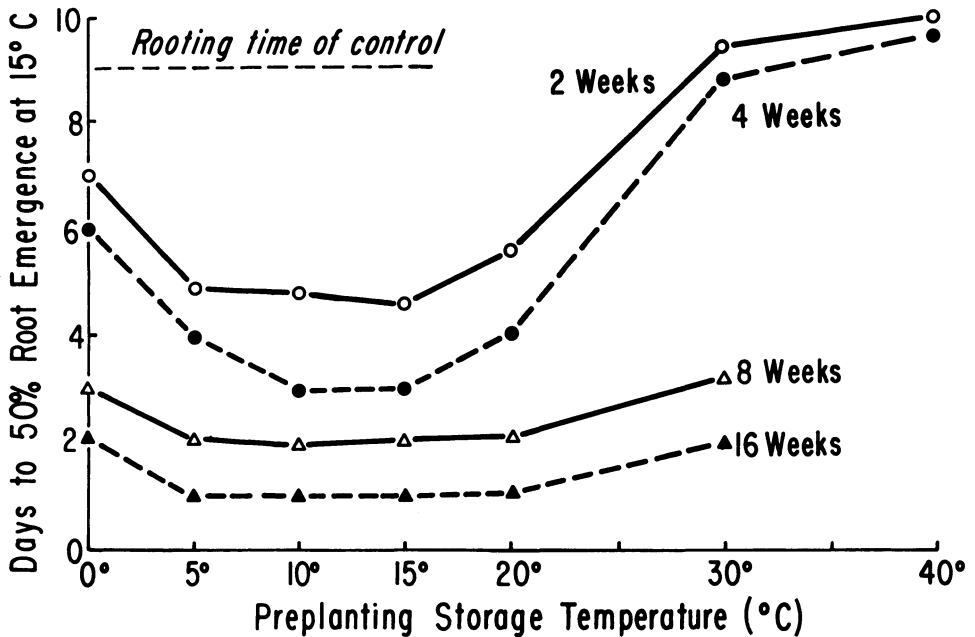


Fig. 13. Days to 50% root emergence for 'Australian Brown 5' bulbs planted in moist sphagnum at 15° C after storage at indicated temperatures for 2, 4, 8, and 16 weeks. Control was placed in sphagnum without storage. Each point is based on a 30-bulb sample.

Both cultivars show the same pattern of root emergence except that, as in its sprouting behavior, 'Australian Brown 5' is slower to root. For rooting in sphagnum at 15° C and for all the storage periods, the storage temperatures may be arranged in an order from least to greatest acceleration of root emergence as follows: 40°, 30°, 0°, 20°, 5°, 10°, and 15° C. Time to 50 per cent root emergence, like time for sprout emergence, decreases with increasing length of storage prior to planting, at all temperatures. (The reversal at 40° C in figure 12 is of doubtful significance.) For both cultivars, the highest storage temperature (40° C) increased the time required for root emergence beyond that of the check treatment. In other words, the temperature appeared to have increased the level of rest. It may be recalled that none of the storage treatments increased the time required for sprout emergence. The delayed rooting of bulbs held at 40° C may reflect the bulb desiccation observed in this storage.

Although rooting was usually more rapid in stored bulbs than in the con-

trols, the total time from the beginning of storage to 50 per cent root emergence was always increased by storage (which was not always true for sprouting, as shown by figs. 8 and 11). This difference between sprout and root emergence results from the much greater rapidity of root emergence and the fact that the shortest storage period chosen—two weeks—was much longer than the time required for the control bulbs to reach 50 per cent rooting. Otherwise, sprout emergence and root emergence appear to be remarkably similar in their response to storage time and temperature.

The data indicate that, if roots have a rest period, it is very brief and, further, that the availability of moisture is a dominant factor controlling root elongation. These ideas were investigated further by placing 10 freshly harvested, mature bulbs of each of the cultivars 'Sweet Spanish,' 'Yellow Bermuda,' 'Australian Brown 5,' and 'White Creole' (all grown at Davis) with their root plates immersed in water, at 15° C. Other lots of 10 bulbs of the same cultivars were placed in flats of moist sphagnum at 15° C. All bulbs

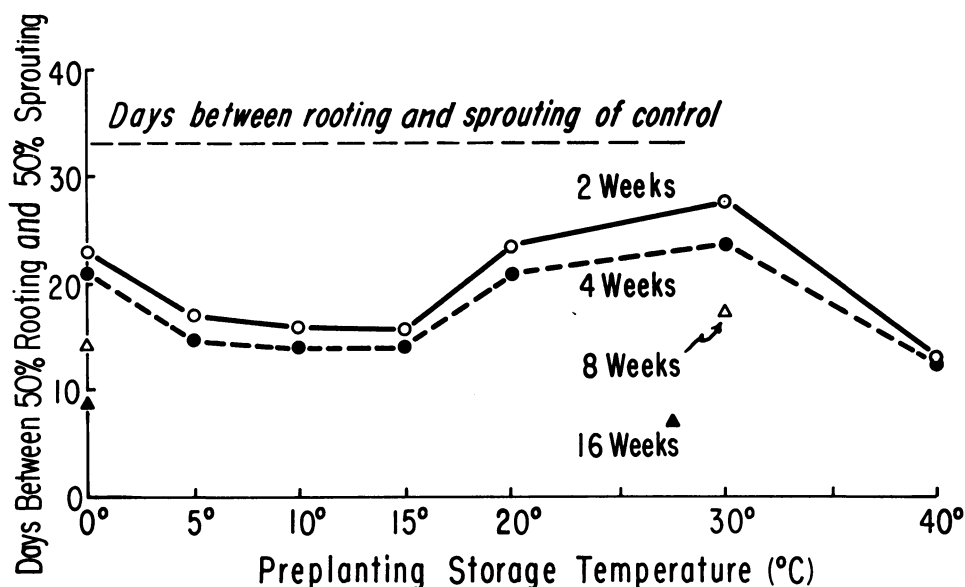


Fig. 14. Days from time of 50% rooting to time of 50% sprouting for 'Excel' bulbs stored for 0, 2, 4, 8, and 16 weeks at indicated temperatures and then placed in moist sphagnum at 15° C (compare with figs. 7 and 12).

placed in contact with water rooted three days after the beginning of the test. Roots first emerged from the bulbs placed in sphagnum two to three days later.

Apparently the time required for root emergence depends in part on the ease with which water has access to the stem plate or root primordia. The number of days required for roots to emerge from bulbs placed in wet sphagnum would undoubtedly have been reduced had the bulbs been placed with their stem plates in water.

Effect of Emerged Roots on Sprout Emergence

For all storage temperatures and all storage periods, root emergence invariably preceded sprout emergence. Certain sound bulbs which were noticeably much slower to sprout than others usually lacked roots. This observation suggested that sprout emergence may be

associated with prior root emergence. To determine if the data already at hand supported this idea, the time intervals between the days to 50 per cent rooting and 50 per cent sprouting were determined for 'Excel' (fig. 14) and 'Australian Brown 5' (fig. 15). These graphs have the same shape as those for sprout emergence because the periods to sprout emergence are little affected by subtracting the relatively short intervals between planting and root emergence. Clearly, rooting preceded sprouting in all treatments and, as rest was depleted, sprouting followed rooting more and more quickly.

The relationship between root emergence and sprout emergence was further tested by an experiment on root amputation. 'White Creole,' grown in the Imperial Valley, California, and shipped to Vacaville five days after harvest, was used. Sixty bulbs of uniform size were selected; the old, dry

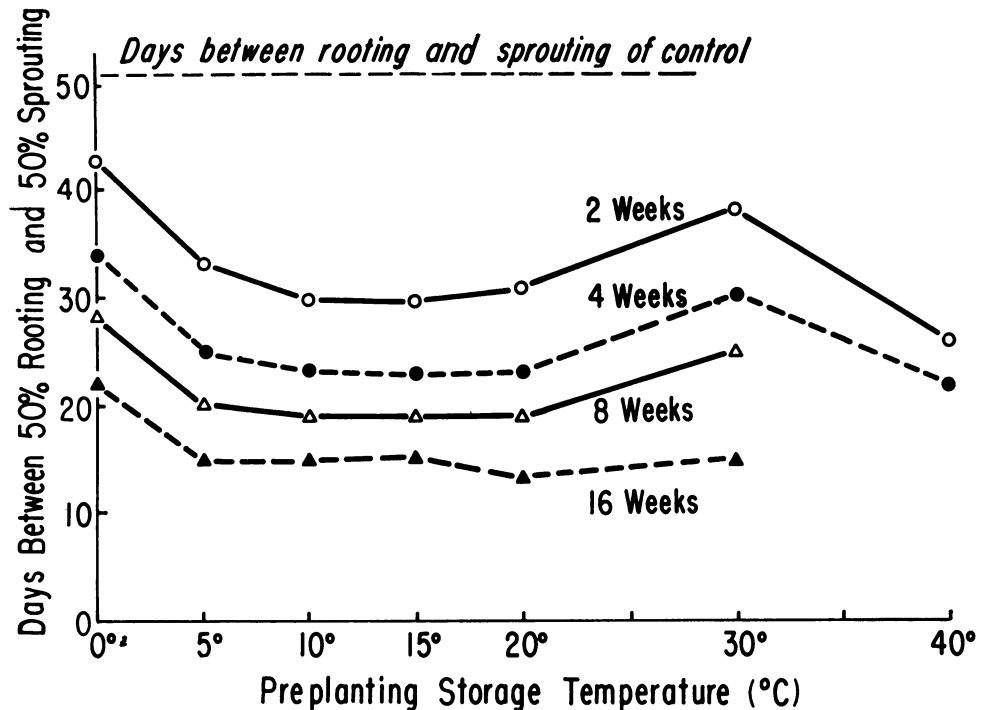


Fig. 15. Days from time of 50% rooting to time of 50% sprouting for 'Australian Brown 5' bulbs stored for 0, 2, 4, 8, and 16 weeks at indicated temperatures and then placed in moist sphagnum at 15° C (compare with figs. 10 and 13).

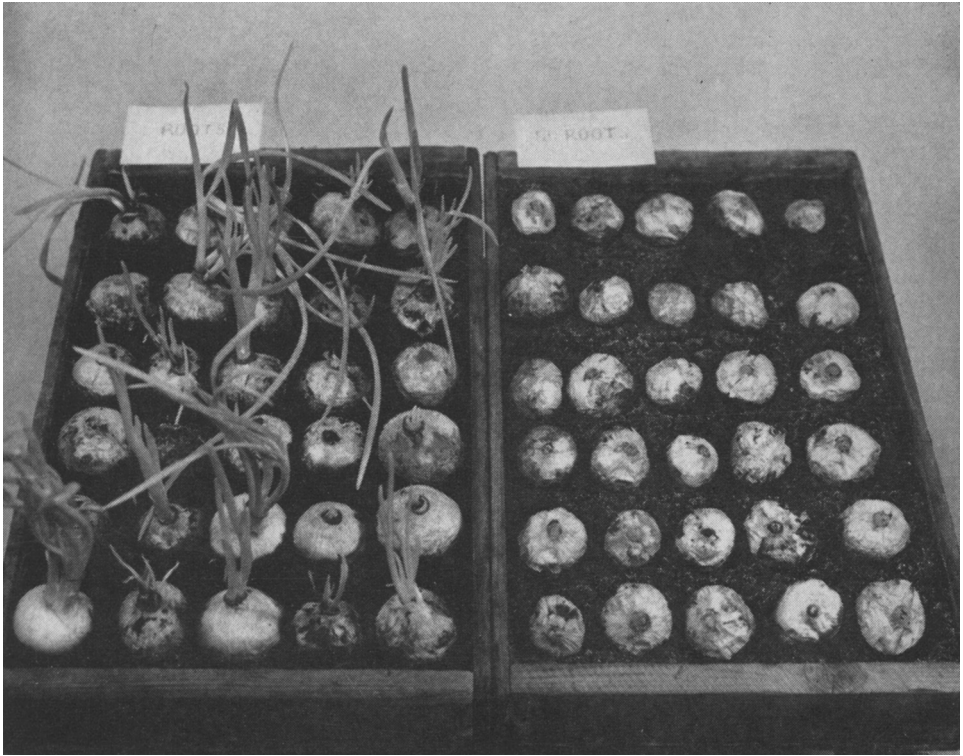


Fig. 16. 'White Creole' bulbs 6 weeks after being planted in flats of moist sphagnum held at 15° C. Roots were removed daily from bulbs in flat on right; they were allowed to develop on bulbs at left.

roots were trimmed off and 30 bulbs were planted in each of two flats of moist sphagnum held in the 15° C storage room. Roots emerging from the bulbs in one of the flats were removed daily as they appeared. Days to 50 per cent sprout emergence were determined as in previous tests. None of the bulbs from which the roots were removed sprouted during the six-week period of the test, while those bulbs that were allowed to develop roots were 50 per cent sprouted in 31 days (fig. 16). Root production in the former bulbs decreased with time, and ceased completely after about six weeks.

Storage Temperature and Total Root Production

Mann and Lewis (1956) observed that the temperature at which garlic bulbs were held before planting had a

marked effect on the volume of roots produced as well as on the time of root emergence. Because observations on roots in the above experiments indicated that onions may show a similar response, the following experiment was run. Thirty uniform-sized 'Excel' bulbs were stored at each of the seven storage temperatures for two weeks and then placed in flats of moist sphagnum in the 15° C storage room. One week after all the bulbs from the different treatments had at least one root 5 mm long, the bulbs were carefully dug and the roots were measured, trimmed, and weighed. Figure 17 shows the weights and lengths of roots developed at 15° C on bulbs from the seven preplanting storage temperatures. Representative bulbs are shown in figure 18. Bulbs stored for two weeks at 15° C had the most extensive root systems; bulbs stored at 30°

and 40° C had the least extensive root systems; bulbs from 5°, 10°, and 20° C had root systems of intermediate size.

Temperature and Sprout Elongation

Ten bulbs of 'White Creole' planted in each of three flats of moist sphagnum were held at 15° C until all bulbs had sprouted. The flats were then transferred to 5°, 15°, and 30° C, and leaf elongation of the five most vigorous plants was recorded for three successive weeks. The results are summarized in the table below.

TEMPERATURE °C	AVERAGE INCREASE IN TOP HEIGHT		
	1ST WEEK cm	2ND WEEK cm	3D WEEK cm
5	2	3	5
15	6	9	15
30	10	22	35

This test clearly shows that the elongation of leaves of already-sprouted

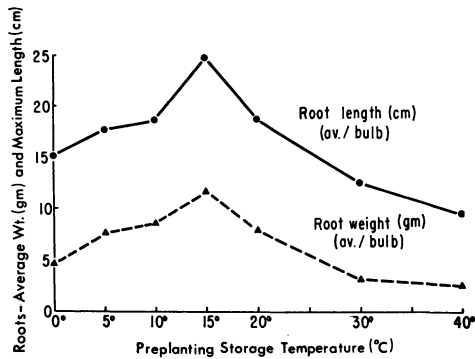


Fig. 17. Weight and length of roots of 'Excel' bulbs grown in moist sphagnum at 15° C and stored for two weeks at indicated temperatures. Each point is based on a 30-bulb sample (compare with fig. 18).

bulbs increases with temperature over the range 5° to 30° C. The effect of temperature on leaf elongation at this stage of growth is completely distinct from the effect of storage temperature on the initiation of sprouts.

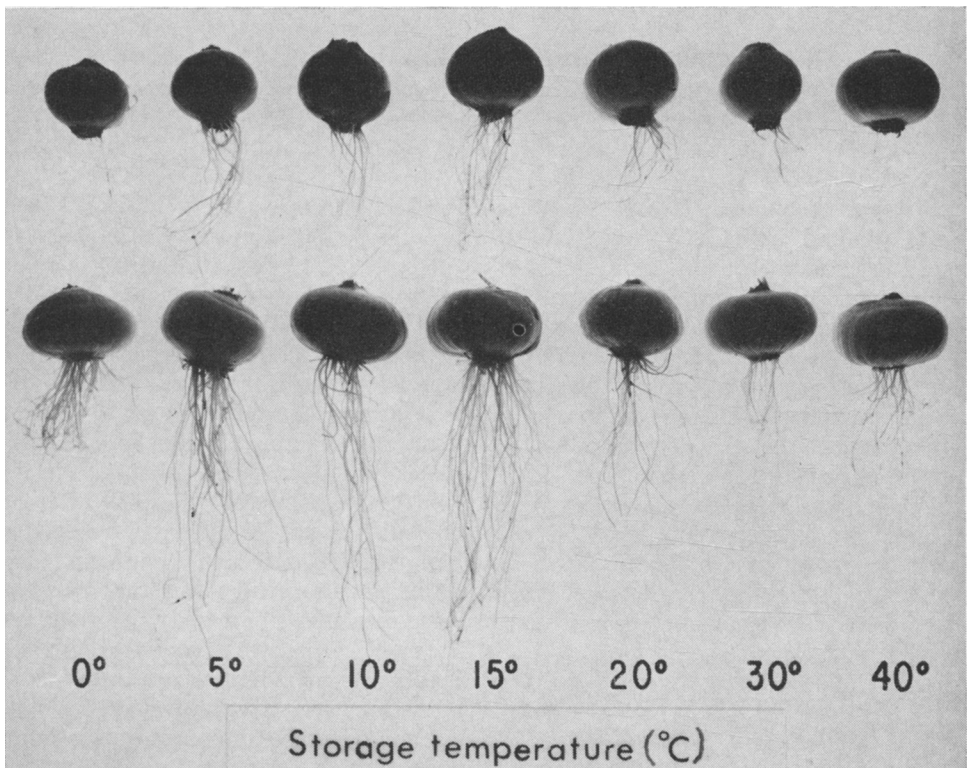


Fig. 18. Root production in 'Australian Brown 5' (top row) and 'Excel' (bottom row) bulbs stored for two weeks at indicated temperatures before being planted in moist sphagnum at 15° C. A representative bulb from each temperature is shown (compare with fig. 17).

MITOTIC ACTIVITY IN THE SHOOT APEX AT THREE STORAGE TEMPERATURES

In vegetative organs such as bulbs and tubers, we do not know whether cell division at the apices stops or merely slows down during the rest period. Clearly, cell enlargement is greatly restricted. Because of the marked effect of storage temperature on the rest of onion bulbs, data on the mitotic activity in shoot apices under different storage conditions are of special interest. Limited data on the processes associated with the resumption of growth are available from several studies on germinating seeds and from studies on shoot apices.

Pollock and Olney (1959) found that growth processes in cherry seeds after-ripened at 5° C are first detected after four weeks. This late-developing capacity for growth, evidenced by an increase in translocation and in dry weight, was preceded by cell division. In seeds after-ripened at 25° C, no comparable growth capacity was found, but cell divisions were evident within the first four weeks and then declined. Ledbetter (1960) concluded that the failure of the pith cells to divide is the basic cause of dwarf growth in *Rhodotypos tetrapetala*. He also noted that the inhibition of internode elongation by maleic hydrazide caused morphological changes analogous to physiological dwarfing. While the previous findings indicate cell division as a critical factor for growth, Haber and Luippold (1960) showed that the visible expansion of the embryonic axis of germinating lettuce seed is not necessarily preceded by cell division. They also showed that the embryo axis of nongerminating, soaked seed exhibits mitosis in addition to other processes usually associated with growth. They suggested that the initiation of cell expansion is the critical factor for growth in resting seeds and possibly for other plant organs.

The structure and development of the shoot apex—that portion of the

apical meristem distal to the latest-formed leaf primordium—have received considerable attention in detailed reviews by Foster (1939, 1941), Philipson (1949), and Gifford (1954).

Savelkoul (1957), on the basis of the distribution of mitotic activity in the shoot apex of *Elodea densa*, divided the shoot apex into distal and proximal zones. She concluded that zonation based on the frequency of mitotic divisions corresponds with the zonation based on other histological criteria, such as planes of wall formation as found by Stant (1952). Buvat (1952), realizing the inadequacy of describing the shoot apex only in terms of planes of wall formation, introduced the idea of the distribution of mitotic activity within the shoot apex as an additional criterion for identifying zonal patterns. The studies of Jacobs and Marrow (1961) on the apical buds of *Coleus blumei* Benth. showed that the distal 10 to 20 μ of the meristem consistently had fewer mitotic figures than did the proximal region.

Wildman and Ballard (unpublished) recorded a low frequency of mitosis in smears of resting buds of *Helianthus annuus* L. Scoring of mitotic divisions was based only on clearly evident chromosomes. These workers noted that sucrose induces resting nuclei to divide, indicating that cell division may be an energy-requiring process and not one governed only by specific hormonal agents.

The primary purpose of this section of our study was to determine the extent of mitosis in the onion shoot tip during the rest period and to observe how mitosis is affected by storage temperature.

MATERIALS AND METHODS

Bulbs from the 'Excel' planting of December 31, 1960, were used. Sam-

pling began in the field when leaf initiation ceased, continued through harvesting and curing, and during storage at 0°, 15°, and 30° C. Samples of 10 bulbs were taken at 10-day intervals over a period of 70 days. For each bulb, all leaves except the last well-defined primordium were removed from the stem. The shoot apex with some underlying stem tissue was then removed and fixed in Craf III (Sass, 1951). To assure that all apices were sectioned in a uniform manner, the blocks of tissue were trimmed so that the longest dimensions on the flanks of the apex were parallel to the plane of leaf divergence. The tissue was dehydrated with ethyl and tertiary butyl alcohols and embedded in paraffin (Johansen, 1940). Serial longitudinal sections were cut at 10 μ on a rotary microtome, stained with crystal violet with orange G as a counter stain (Johansen, 1940), and mounted in Permunt. The most nearly median sec-

tion was located by means of a binocular microscope, and marked with India ink. Mitotic figures were counted in this median section and in the five preceding and the five following sections. Any cells that showed well-defined stages of mitosis, from prophase through telophase, were counted as dividing cells. The parts studied were the shoot apex, the immediate subapical region to a depth of 100 μ from the center summit of the apex, and the youngest leaf primordium. The numbers of mitoses observed were used for relative comparisons among treatments.

RESULTS OF MITOSIS STUDY

In longitudinal section the shoot apex of the 'Excel' onion is broad, slightly convex, and closely overarched by the first leaf. The layer underlying the single protoderm layer is composed of mostly anticlinally dividing cells, but

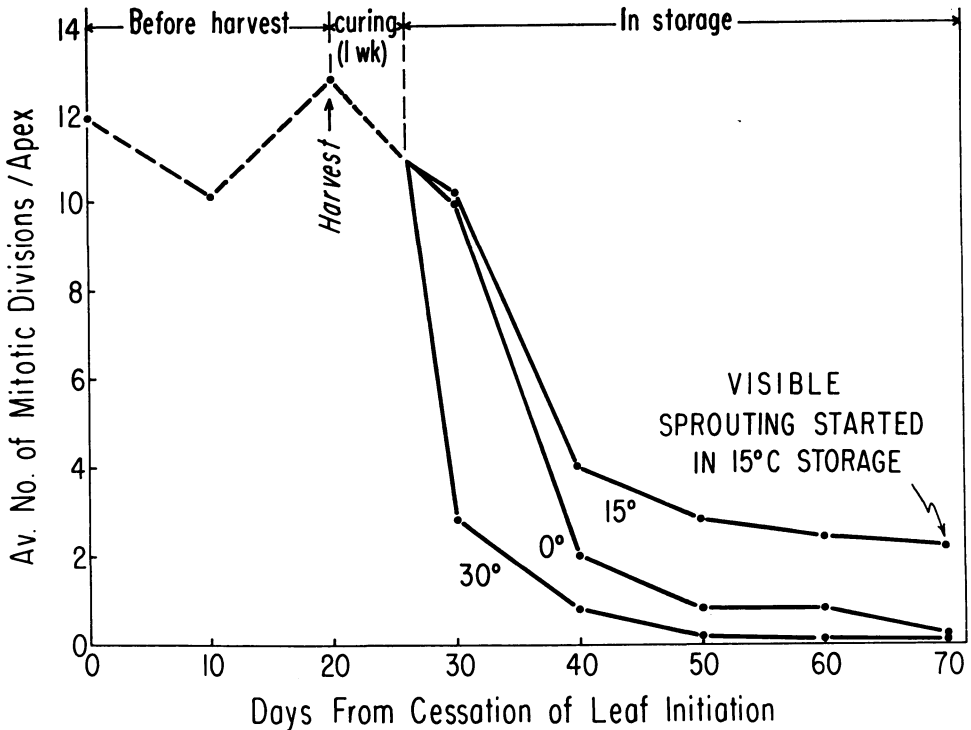


Fig. 19. Number of mitotic figures observed in shoot apices of 'Excel' onions during maturity, curing, and storage. Each point is based on 11 sections from each of 5 to 8 apices.

some periclinally dividing cells are present. These two peripheral layers of cells of the apex enclose a mass of thin-walled, homogeneous cells in the central region. The cells of the two peripheral layers are smaller and have denser cytoplasm than the cells of the central region.

Our counts showed that the proximal region of the apical meristem (25 to 100 μ) had more mitotic divisions than the distal region (0 to 25 μ). Mitoses were more common in the youngest leaf than in the apical meristem. In the young leaf, the mitotic divisions were usually at the base or near the margins.

Figure 19 shows the number of mitotic figures observed in the shoot apices of 'Excel' onion bulbs from the time of the cessation of leaf initiation (in the field) through storage at the three indicated temperatures. It is evident that during storage at all three temperatures the frequency of mitosis in the bud was relatively low. The frequency of mitotic divisions was greatest at 15° C, however, where divisions were observed throughout the storage period. After about three weeks in storage at 0° or

30° C, only a few buds were found to contain mitotic divisions and these were often in the early stages of mitosis, i.e., prophase and early metaphases. In shoots of bulbs stored at 15° C, later stages of mitosis (anaphase and telophase figures) were frequent. The later stages of mitosis were very common in plants sampled from the field at the time leaf initiation ceased, and also at harvest time.

In comparing data on mitotic activity in the shoot apex with data on leaf initiation and sprout elongation, we note that leaf initiation apparently stops shortly before harvest and begins again, in bulbs stored at 15° C after harvest, at a rate of about one new leaf every two weeks (figs. 2 and 5). In storage at 0° and 30° C, leaves are initiated at about half this rate. Sprouting depends on the elongation of leaves initiated well before harvest, and since these leaves are attached below the apex, sprouting is, at least morphologically, unrelated to cell divisions at the shoot apex, although both processes proceed more rapidly at 15° C than at 0° or 30° C.

DISCUSSION

Available data on rest and dormancy in onion bulbs, garlic bulbs, and gladiolus corms indicate that the effect of storage temperature is remarkably similar in the three species. For onion bulbs, exposure to intermediate temperatures of from 5° to 15° C before planting was most favorable for sprout emergence; i.e., at those temperatures rest appeared to be rapidly depleted. Higher and lower temperatures—around 30° and 0° C—always resulted in less rapid sprout emergence. In garlic bulbs and gladiolus corms rest also disappears most rapidly after storage at 5° to 10° C. In the potato tuber, however, high temperatures—25° to 30° C—are the most effective in depleting rest, although temperature responses here are less marked than they are in the onion,

and have been less clearly demonstrated.

The gradual depletion of rest in onion bulbs can be demonstrated by periodically transferring stored onions to favorable sprouting conditions. Measuring the depletion of rest by mean days to sprout emergence or by days to 50 per cent sprout emergence indicates the quantitative nature of rest. The gradual disappearance of rest in stored onion bulbs indicates that a "rest period" is not "broken" at any specific time, and thus that the term "rest period" is rather misleading. Mann and Lewis (1956) came to the same conclusion with regard to the rest condition in garlic bulbs. "Breaking" of rest may, of course, be arbitrarily defined. Wright and Peacock (1934) considered rest as "broken" if a sample of 10 potato tubers

all sprouted in two weeks at 70° F and high humidity.

The level of rest for sprouts and roots was measured by the time taken for sprouts or roots to elongate a specified amount. For roots, this was 5 mm beyond the base of the bulb; for sprouts, rest was considered ended when the sprout first became visible at the bulb surface. This externally visible sprouting has been regarded by most investigators as the first indication of the resumption of growth and, consequently, the termination of rest. Sprouting leaves have, of course, grown a certain distance inside the bulb before they become externally evident. Dissection of 'Excel' bulbs cured for one week after harvest and then stored at 15° C showed that the elongation of the sprout was first evident about two weeks after the start of storage. Such bulbs maintained in storage at 15° C did not sprout—that is, they did not produce an externally visible sprout—until over eight weeks of the storage period had passed (fig. 7). Bulbs placed in moist sphagnum at 15° C immediately after curing sprouted in about 40 days (fig. 7, control). If we assume that leaf elongation began in the planted bulbs and in the stored bulbs at approximately the same time (two weeks after storing or planting), then about 26 days at 15° C were required for the leaves to elongate until they emerged from the planted bulbs. Since 15° C is favorable for sprouting, this period must be close to the minimum required for sprouts to elongate until they become visible.

The greater effectiveness of storage at 40° C, as compared with 30° C, in decreasing rest of sprouts (figs. 7 and 8) is interesting. While this may possibly be attributable to heat injury, the bulbs transferred to moist sphagnum at 15° C after two or four weeks at 40° C showed no evidence of injury. Curing studies by Vaughan and Cropsey (1958, 1959) indicate that artificial drying of onion bulbs for more than 24 hours at 125° F (near 50° C) caused injury, and that

curing for more than 48 hours at 115° F (40.6° C) increased decay. Yamaguchi *et al.* (1957) stored 'Southport White Globe' onions at a wide range of temperatures for four months. They believed storage at 40° C to be injurious for that long a period. However, Mann and Lewis (1956) observed an effect of high temperature on sprouting of garlic similar to that reported here for onions: bulbs stored for 16 and 20 weeks at 20° C sprouted more quickly than did bulbs stored 16 and 20 weeks at 15° C. This suggests that the observed effect on onion may not have resulted from heat injury, because the similar response in garlic occurred at considerably lower temperatures. The possibility that storage at 40° C was injurious cannot, however, be eliminated.

Our evidence for rest in buds of freshly harvested onion bulbs supports the early experiments of Jones (1921) and Boswell (1924) and the observations of Heath and Holdsworth (1948) and of Jones, Perry, and Edmundson (1949).

The enhancement of sprout formation by emerged roots and the sprouting of rotted bulbs (usually in absence of roots) suggest that the availability of moisture may be important for sprout emergence. Thus, for brief storages at least, root emergence always preceded sprout emergence, and trimming the emerging roots as fast as they appeared prevented sprouting as long as the bulbs remained fairly sound. Nevertheless, a rather long time elapses between rooting and sprouting. This difference is due in part to the distance that the sprouts must grow to be evident, but a real time difference associated with rest may also be involved. This time interval can be reduced by injury to the bulb or by preplanting storage at temperatures between 5° and 20° C. The reactions to both of these treatments provide good evidence for rest in the onion bud.

While sprout emergence was slowest after preplanting storage at 30° C, the growth rate of well-emerged sprouts in-

creased with increasing temperatures when sprouted bulbs were placed at 5°, 15°, and 30° C. The quite different effect of temperatures between 5° and 30° C on the initial stages of leaf elongation and on the growth rate of sprouts after they had emerged from the bulbs furnishes additional evidence for rest in onion buds.

At present the evidence for rest in onion roots is less decisive than that for onion buds. Part of this uncertainty stems from the fact that access of water to the basal part of even freshly harvested bulbs causes root initials to emerge within a remarkably short time—several cultivars tested in this study rooted in three days, and some of these, e.g., 'Sweet Spanish,' are known to keep very well in storage.

Preplanting storage of bulbs at the intermediate temperatures decreased the time required for roots to emerge. Root emergence was slowest after the lowest (0° C) and highest (40° C) storage temperatures used, and the remarkable similarity between root and sprout emergence in response to storage temperature suggests a similar internal control of growth in both.

Other evidence, however, which at first suggests that roots have a rest period, is not convincing. Storage temperature not only affects root emergence, but the subsequent growth of the root system as well. Thus bulbs produced larger root systems if they had been stored at 15° C rather than at 0° or 30° C before planting (figs. 17 and 18). This ability of the root system to grow vigorously may be as satisfactory a measure of rest as is root emergence. If growth merely slows down during rest instead of stopping completely, as may be the case with some vegetative organs (Davidson, 1958), rest may be ended by an increase in growth rate.

Mann and Lewis (1956) observed large differences in the development of root systems of garlic cloves stored at different temperatures before planting. Those observations differed from the

present ones in that the root system of garlic developed most rapidly after storage at 10° C rather than after 15° C, as in onion. Garlic is more tolerant of low temperature than is onion, and the storage temperatures most favorable to subsequent sprouting or rooting have lower optimums than those for onion.

Storage temperature also affects the vigor of top growth of field-planted garlic (Mann and Minges, 1958). Although the greatest vigor is observed after storage at 0° C, sprouting is most rapid after storage at 5° or 10° C. Boswell (1924) noted that onion bulbs planted in the field after eight months of storage at 32° F and at 50° F showed the greatest vigor of top growth from the 32° F storage. The profound effect of storage temperature on flowering has, however, diverted attention from the effects of storage temperature on the vegetative growth. More information should be gathered on this interesting response.

In all the above examples of the stimulating effect of low storage temperatures on subsequent growth—either of roots or tops, in garlic or in onion—the data show that the storage temperatures most favorable to rapid sprout or root emergence are distinct from those most favorable to later growth. Thus the effectiveness of storage at 15° C for stimulation of the growth of onion roots after they emerge (figs. 17 and 18) is quite distinct from the broad 5° to 15° C range which enhances root emergence (figs. 12 and 13). Likewise, Mann and Lewis (1956) noted a similar distinction in garlic: "...the greatest weight of roots was produced by cloves previously held at 10° C, in sharp contrast to...root emergence (5 mm) in which a 5° C preplanting storage had the most stimulating effect." Thus, while the growth of roots subsequent to their initiation resembles other rest reactions in that it is a response to storage temperature, it apparently is not a rest reaction, since the temperatures controlling it are distinguishable from those controlling root elongation.

With reference to rest in buds, our results suggest some of the reasons why storage temperatures near 0° C have been found (except for economic reasons) to be the most satisfactory for holding onion bulbs for long periods. Near that temperature several factors favor bulb storage: the internal changes (depletion of rest), which appear necessary for leaf elongation, take place at a slow rate—i.e., the start of sprouting is delayed; leaf elongation is slow at 0° C, even when rest is no longer in effect; and loss of bulb weight from drying is usually reduced. Loss of weight from drying depends, however, on factors in addition to temperature, and warm storage does not necessarily imply rapid loss of moisture. Yamaguchi *et al.* (1957) and Pratt and Morris (1958) recovered about as many 'Southport White Globe' onions from storage at 25° to 35° C as from 0° to 15° C after four months of storage. Most serious losses were from the 10° to 20° C storage temperatures. These workers observed that, while the best storage for dehydration purposes was 30° C (high flake quality), bulbs from the low temperatures were fresher in appearance. While bulb storage at high temperatures may be disadvantageous if the environment favors rapid moisture loss, the maintenance of the resting condition within the bulbs appears to make prolonged storage at temperatures of 25° to 30° C entirely feasible and certainly more desirable than storage at the intermediate 10° to 20° C range.

It is interesting to note that a relatively large number of mitotic figures were observed during the 20-day period before harvest—the one time during the life of the vegetative plant when leaf initiation appears virtually to cease. Leaf initiation started again during

curing or at the beginning of storage and, in storage at 15° C, continued at about half the rate observed during the period of active growth in the field. This rate was again reduced by half in storage at 0° or 30° C, and continued to drop as these storages were prolonged.

Most of the mitotic figures observed in bulbs stored at 0° or 30° C were pro-phases or early metaphases. The later stages of mitosis were more common in bulbs stored at 15° C and in bulbs sampled from the field prior to harvest. The lack of the later stages of mitosis in bulbs stored at 0° or 30° C may indicate that these temperatures block the later stages, but without data on the effect of temperature on the duration of the separate mitotic phases, no analysis of the observed frequencies of certain phases is possible.

Sprouting, as measured by the elongation of leaf No. 12, began about two weeks after the start of storage at 15° C. At that time the number of mitotic figures in the apex was still decreasing, so that mitotic activity at the shoot apex appears not to be associated with sprouting, which involves the elongation of older leaves.

While the larger number of mitotic figures observed in storage at 15° C, as compared with storage at 0° or 30° C, may have been associated with the higher rate of leaf initiation at 15° C, the still larger number during the period just prior to harvest does not seem to have been associated with leaf initiation. Perhaps the most interesting observation is that, during the short pre-harvest period when leaf initiation ceases or reaches its lowest rate, mitotic figures are relatively abundant, so that at no time during rest or dormancy does the shoot apex appear to be morphologically inactive.

SUMMARY AND CONCLUSIONS

Bulb Development

For the field tests, the cultivar 'Excel' was direct-seeded in the field at Davis, California, December 31, 1960. The bulbs were harvested on June 30, when 80 per cent of the tops had fallen over, placed in crates in a bulb house for a week to dry thoroughly (cure), and then transferred to storage at 0°, 15°, and 30° C. Samples of plants were dissected weekly from shortly after seedling emergence until the time of storage.

Adventitious roots emerged at the rate of four or five per week, reached a maximum of 70 live roots per plant 20 days before harvest, and then declined in number, as old roots died more quickly than new roots were produced.

Leaves were initiated at intervals of about one week until about three weeks before the plants were harvested, when leaf initiation ceased.

As bulbing begins, the newly forming leaves develop without blades; i.e., they consist of sheaths only. Lateral expansion of these bladeless sheaths, and of the lower portions of the sheaths of bladed leaves exterior to them, results in bulb enlargement. Bladeless sheaths of lateral buds, initiated at the time of bulbing, also swell and contribute to bulb expansion.

The innermost (last formed and bladed) four to five leaves of the main and lateral buds remain small, but will later elongate as sprouts if the bulbs sprout in storage or are planted.

Bulb Growth in Storage

In dry storage at 15° C, leaf initiation begins near the beginning of storage and continues at about half the rate observed during the period of active growth in the field. In bulbs stored at 0° or 30° C, the rate of leaf initiation was about half that observed in storage at 15° C.

Elongation of the inner leaves during dry storage at 15° C started after about

two weeks of storage. Six weeks later sprouts were not yet externally visible on bulbs remaining at 15° C. No elongation of the inner leaves was observed in the 0° or 30° C storages during a period of 10 weeks.

Storage Temperature and Rest and Dormancy

'Excel' and 'Australian Brown 5' onion bulbs field-grown at Davis, harvested when mature, and cured for a week were placed in storages controlled at 0°, 5°, 10°, 15°, 20°, 30°, and 40° C. Bulbs removed from storage after 2, 4, 8, and 16 weeks were placed in moist sphagnum at 15° C, and checked for sprouting and rooting. A control lot was planted in sphagnum immediately after curing.

Preplanting storage temperature had a marked effect on the rapidity of sprouting. For any storage period, the most rapid emergence occurred after storage at 5°, 10°, or 15° C. Sprouts emerged least rapidly after storage at 0° or 30° C. The longer the bulbs were stored before planting, the more quickly they sprouted, indicating that at no storage temperature was rest intensified.

The effect of temperature on leaf elongation of sprouted bulbs is distinct from its effect on the initiation of sprouting. While the latter is most rapid after bulb storage at temperatures of 5°, 10°, and 15° C, growth subsequent to sprout emergence increases with increasing temperatures between 5° and 30° C.

Root emergence always preceded sprouting in our sprouting tests. For bulbs planted in sphagnum at 15° C, daily removal of roots as they emerged kept the bulbs from sprouting for six weeks, at which time the test was discontinued; control bulbs with intact root systems sprouted after about four weeks. That onion bulbs held in dry

storage for long periods sprout without rooting is, of course, well known.

Root emergence was most rapid after storage at the same temperatures that hastened sprout emergence. Roots emerge more quickly than sprouts, however, and placing the stem in contact with water induced root emergence in two or three days in freshly harvested bulbs.

While the response of onion buds to temperature indicates a definite rest in buds, evidence for rest in roots is less clear.

Storage temperature of bulbs has an effect on subsequent root growth similar to, but probably separate from, its effect on root emergence. Root lengths and weights after a period of growth were greater from bulbs stored at 15° C than from bulbs stored at 5° or 10° C; but root emergence was equally rapid after storage at 5°, 10°, or 15° C.

Mitotic Figures in the Shoot Apex

Mitotic figures in the shoot apex and in the first leaf of the 'Excel' onion bulb

were counted. Samples were taken at 10-day intervals over a period of 70 days, starting at the time of cessation of leaf formation (in the field) and continuing through curing (7 days) and through storage (42 days) at 0°, 15°, and 30° C.

Mitotic figures were present from the cessation of leaf initiation through curing and storage. Their frequency decreased with continued storage, especially at 0° and 30° C.

Sprouting in storage resulted from the elongation of well-formed leaf initials and, at least in its early stages, was not correlated with a change in the number of mitotic figures in the apical region of the stem nor with a change in the rate of leaf initiation.

Even during the period of about 20 days near the time of bulb maturity, when leaf initiation ceased, mitotic figures were evident in the shoot apex. From this standpoint, the shoot apex apparently remains morphologically active during rest and dormancy.

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