The Causes of the Bud Eccentricity and the Large Divergence Angles Between Leaves in Cucurbitaceae

Robert Snow

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THE CAUSES OF THE BUD ECCENTRICITY AND THE LARGE DIVERGENCE ANGLES BETWEEN LEAVES IN CUCURBITACEAE

By ROBERT SNOW, F.R.S.
Hurston, Budleigh Salterton, Devon

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[Plate 4]

CONTENTS

In Cucurbitaceae the axillary buds are eccentric, being much displaced in the anodic or up-hill direction of the genetic leaf spiral: also the angles between successive leaves are unusually large. In the young seedling the leaf spiral and the bud eccentricity both start with the third leaf from the base. If a radial vertical cut of some little depth is made in the apex at the kathodic side of P_1, the youngest leaf primordium, or of I_1, the next primordium due to arise, the bud of the P_1 or I_1 is often median or nearly so. Also, if P_1 or I_1 is isolated from the apex with a vertical cut made in the tangential direction and is then extirpated, the bud of the next primordium, I_1 or I_2, is often subeccentric or nearly median. But shaving down the upstanding part of P_1 flush with the apex, or preventing with shallow cuts P_1 or its bud or both from developing does not diminish the eccentricity of any subsequent bud. It is concluded that an axillary bud is made anodic by some repelling influence, perhaps a hormone, which originates from an older leaf primordium and travels in the anodic direction of the leaf spiral. This influence is interrupted by a cut of moderate depth in the apex, but not by a very shallow cut.

The changes in the positions of certain younger leaves due to the formation of median or nearly median buds after these operations strongly support an explanation of the large angles between successive leaves based on a space-filling theory of phyllotaxis. This explanation is that the position in which any leaf n is determined is displaced in the anodic direction by the anodic axillary bud of leaf $n-3$, which encroaches from the kathodic side upon the space available for n between $n-3$ and $n-2$. When the bud of $n-3$ is median or nearly so, the position of n is much less far anodic. The repelling influence that makes the buds anodic is further discussed in relation to the course of the conducting strands and other relevant facts.

1. Introduction

The phyllotaxis of the Cucurbitaceae and the positions of their axillary buds present two special and very interesting peculiarities. The first is that in the spiral phyllotaxis the divergence angles between successive young leaves near the apex, though they differ in

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different genera and species, are always considerably greater than the usual Fibonacci divergence angle of approximately 137°. Thus in the apices of three species, which Hagerup (1930) illustrates with drawings of transections, *Ecballium elaterium*, *Luffa cylindrica*, and *Thladiantha dubia*, the mean divergence angles can be found from measurements to be respectively 146·6°, 157° and 164·7°. In *Cucurbita pepo*, the writer finds the mean angle to be approximately 153° (see figure 1). The second peculiarity is that the axillary buds are not median in their axils, but are much displaced to the anodic side—that is, in the uphill direction along the genetic spiral, the spiral along which the leaves originate (see figure 1). These buds are displaced to different distances in different genera and species, and their displacements appear to be positively correlated, at least roughly, with the divergence angles.

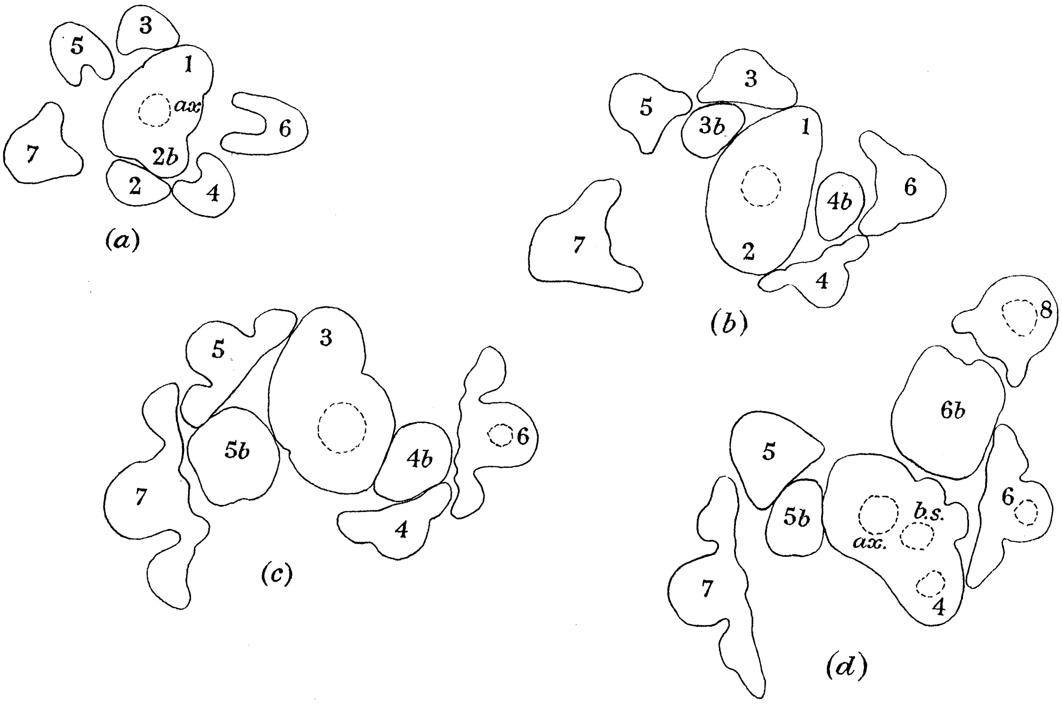


FIGURE 1, *a* to *d*. Transections in collodion through the shoot apex of a normal seedling of *Cucurbita pepo* from the level of the youngest two leaves downwards. The leaves are numbered downwards 1, 2, 3, etc., and their axillary buds, when present, 2*b*, 3*b*, etc. *b.s.*, bud stele; *ax.*, contour of xylem of axis. *a* to *c*, × 66; *d*, × 56.

It was suggested to me long ago by my wife that the abnormally large divergence angles of the Cucurbitaceae might be explained as being due to the eccentric axillary buds, on the basis of a 'space-filling' theory of phyllotaxis such as we have later supported with various evidences (Snow M. & Snow R. 1931, 1952, 1962). The explanation would be as follows.

In the Cucurbitaceae each new leaf '*n*' is determined and arises in accordance with space-filling theory as a roughly circular rudiment at a side of the apex in a depression between two older members, its contact members, below it. In *Cucurbita pepo* the contact member to the kathodic side of a new leaf '*n*' (the side that is 'down-hill' in respect to the genetic spiral) appears to be the leaf $n-3$, three plastochrons older, together with its axillary bud, and the contact member to the anodic side to be the leaf $n-2$, as will be explained later (see figure 1). But although leaves $n-3$ and $n-2$ were themselves similarly determined as roughly circular rudiments, by the time that leaf n is about to be determined, during the last plastochron before it arises, the axillary bud of $n-3$ has already been determined and is often already visible; and this bud, being displaced in the anodic direction, has encroached from one side upon the space that would otherwise have been available for leaf n in the depression between leaves $n-3$ and $n-2$. Leaf n, therefore, is determined in a more anodic position between its contact members than it would otherwise have occupied, and so with its centre at a larger divergence angle from the centre of the previous leaf.

The use of the term 'eccentric' to describe these axillary buds may be criticized, since it is also applied to some buds in another and more proper sense, meaning that their centre of growth does not coincide with their geometric centre. 'Non-median' is more accurate for the present buds, but this term leads to expressions that are too awkward when the distances of different buds from the median are compared.

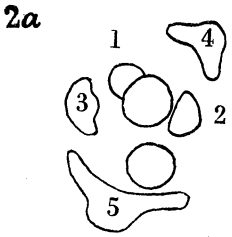
The writer set out both to test the above explanation of the phyllotaxis and also to investigate what causes the axillary buds to be eccentric, by means of experiments and observations on *Cucurbita pepo*. But in the course of the work the second problem came to predominate, and the evidence concerning it will be presented and discussed first.

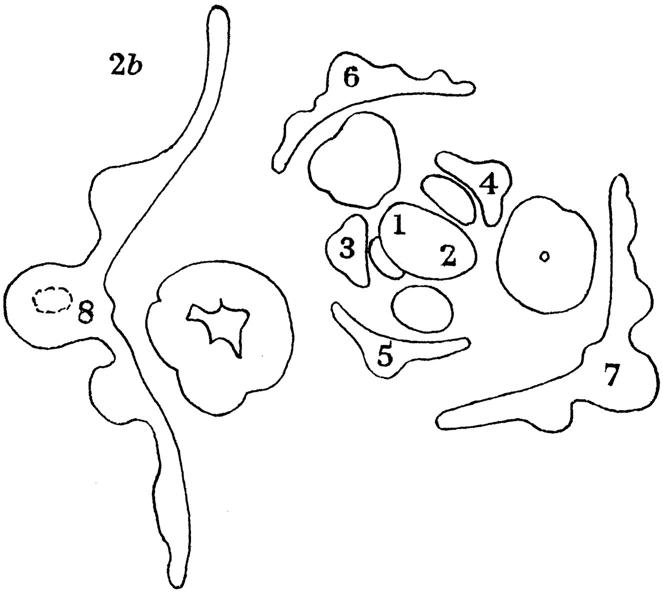
2. The morphology of the Cucurbitaceae

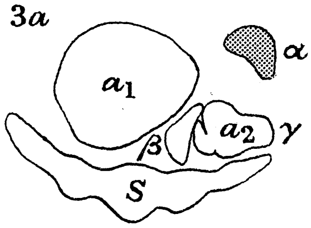
In order to make easily intelligible the results to be reported and the drawings illustrating them, it is necessary to run briefly through a few points in the morphology of the Cucurbitaceae. For the morphological interpretations and for the descriptions of genera other than *Cucurbita*, the writer will rely on an excellent paper by Hagerup (1930). There have indeed been many interpretations, especially of the tendrils, but the one given by Hagerup is convincing and based on very good illustrations and will in any case serve as a descriptive framework.

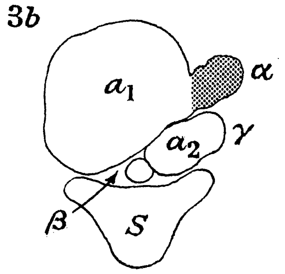
In the Cucurbitaceae the leaves have a bud in every axil, and as soon as the leaf spiral is established, which in *Cucurbita pepo* is at the third leaf of the seedling from the base, the axillary buds are all displaced in the anodic direction to distances that differ in different species. Hagerup states indeed that in *Ecballium elaterium*, a species without tendrils, the buds are roughly median in their axils. But his own drawings (1930, p. 37) of transections through five axillary buds on different shoots of this species show them all clearly anodic, though much less so than those of other genera, and transections of shoot apices made by my wife show the same even more clearly. One of these is shown in figure 2. The axillary buds of the leafy shoots all form terminal flowers, except, according to Hagerup, those of the cotyledons and of the first few leaves of the seedling. Below each terminal flower there may be three bracts, arranged in spiral sequence, which may most simply be called α, β and γ, though Hagerup calls them α, β and 0 (figure 3). Bract α is at the anodic side of the flower, β at the kathodic, and γ at the anodic side again, but closer than α to the subtending leaf. Bract α subtends and is carried up with the tendril, β subtends an inflorescence and γ a leafy shoot. But in a few genera, including *Cucurbita*, *Cucumis*, and

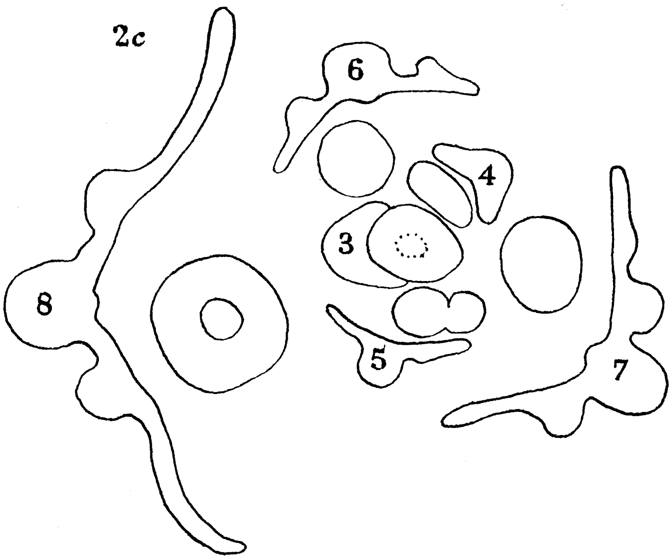
Bryonia, bract β with its inflorescence is aborted or quite rudimentary, so that in these genera there are two lateral members at the anodic side, and none at the kathodic. The rudiment of the tendril, including its subtending bract, is precocious and is inserted very low, and in some genera, including *Cucurbita*, it arises so far to the anodic side of the leaf axil that it is quite beyond it and appears to be arising not from the axillary bud at all,











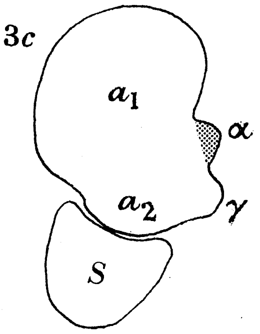


FIGURE 2, *a* to *c*. Transections of a normal shoot apex of *Ecballium elaterium*.

FIGURE 3, *a*, *b*. Main axis, young leaf and its axillary bud with rudiments of floral bracts α, β and γ in *Coccinia moghadd* and *Thladiantha dubia*. Bract α (stippled) is also the tendril rudiment. *c*, *Cucurbita pepo*, a younger stage, also with tendril rudiment stippled. a_1, main axis; a_2, axillary bud; *S*, subtending leaf. (After Hagerup 1930, figures 67, 68 and 70.)

but from the main axis. Thus, as Hagerup (1930) points out, the impression is given that some growth tendency is progressively shifted in the anodic direction, first in the leaf axil and then in the axillary bud itself, and that the tendril always comes at the farthest point of this shift. Perhaps it comes at the summit of some kind of gradient.

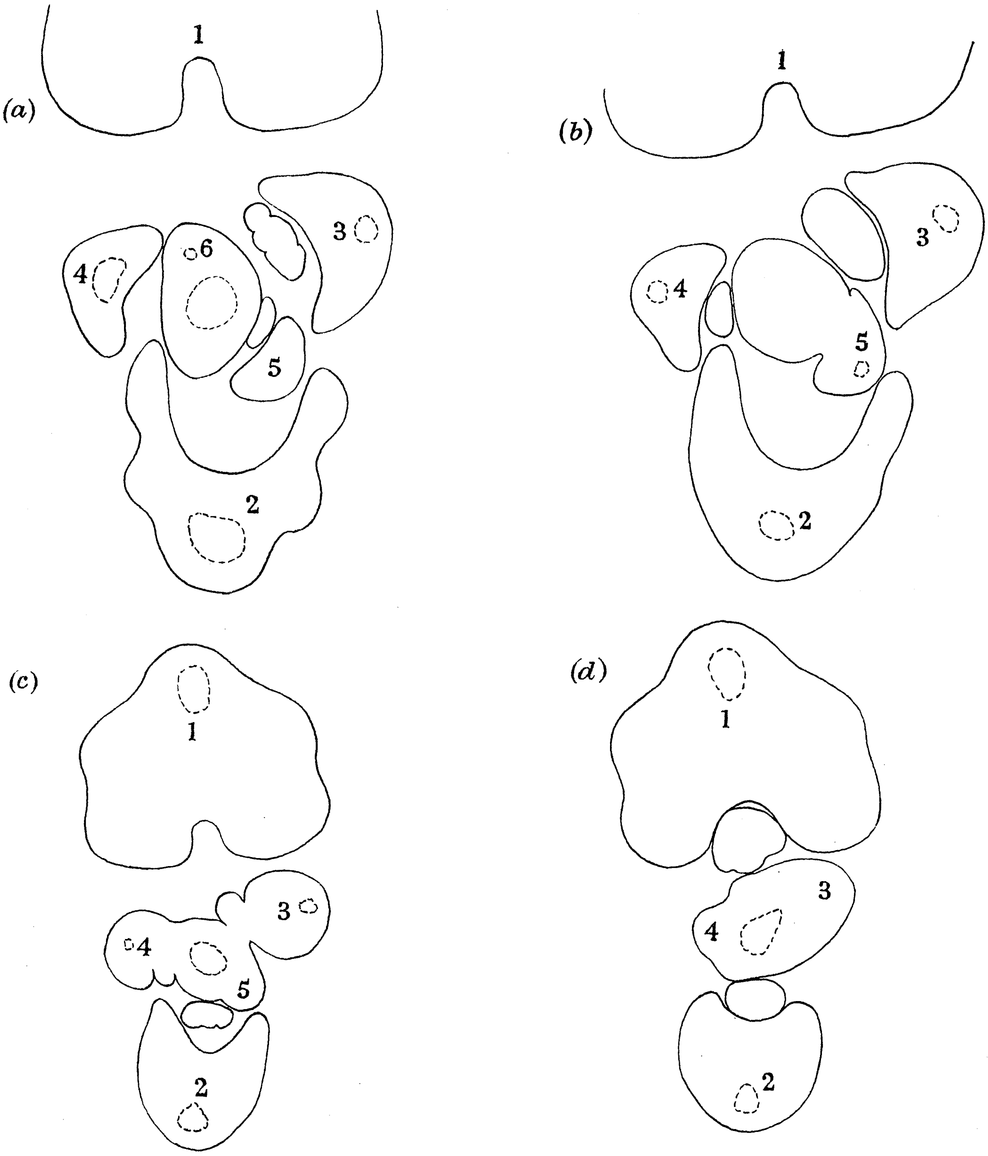


Figure 4, *a* to *d*. Transections in collodion of apex of a very young seedling of *Cucurbita pepo*. The leaves are numbered from the oldest upwards.

3. Preliminary observations on *Cucurbita* seedlings

The first two leaves of the seedling, which are the only leaves present in the seed, are opposite each other, but at different levels, and they have median axillary buds, as would be expected (figure 4). Leaf 3 arises at one side or the other of the plane through leaves 1 and 2, and thereby it determines the direction of the genetic spiral through itself and subsequent leaves. Its axillary bud is the first that is displaced in the anodic direction of the spiral, and this suggests that the bud may be displaced by some kind of repelling influence coming from the next older leaf and bud. The directions of genetic spiral and bud eccentricity are not inherited, since they may differ in different shoots of the same plant.

It can also be seen that the centre of leaf 3 is closer to the centre of the older two leaves between which it arises, as is the usual rule in spiral phyllotaxis, whereas the centres of leaf 6 and all later leaves are closer to that of the younger of the two leaves on each side (see figures 4 and 1). This difference can readily be explained as follows, in accordance with the interpretation of the phyllotaxis suggested in §1. Leaves 1 and 2, with which leaf 3 makes contact, have median buds, whereas from leaf 6 onwards at the latest each leaf n, as will be shown below, makes contact with an anodic axillary bud of $n-3$, which displaces its position away from $n-3$ and towards $n-2$.

The positions of leaves 4 and 5 of the young seedling are transitional, and their contacts have not been ascertained. Figure 3*d* suggests that leaf 4 may make contact with leaf 3 at its base. Further evidence concerning the phyllotaxis, based on results of experiments, will be offered in §11.

The contacts that are made, or most nearly approached, by each new leaf with older members below cannot easily be seen in transections, since the internodes begin to elongate very early. Nor can they be seen when the apex is observed in the solid from directly above, since the sides of the axis fall away steeply on all sides of the stem apex and youngest two leaves, so that even the third leaf from the apex is already some way down the side. The best method is to observe the apical region in the solid from one side with the axis tilted away, so that the insertions of the third and fourth leaves below the apex, *L.* 3 and *L.* 4, are clearly visible. It can then be seen that at late plastochron *L.* 1 makes contact, or very nearly, with *L.* 3, and with the axillary bud of *L.* 4 as already stated (see figure 5, plate 4). Thus the contact numbers of the system are 2 and 3, but contact 3 is peculiar in being made with an axillary bud.

The axillary buds develop early. The bud of P_2, the second youngest leaf primordium, can be seen rising up as a slight hummock at late or even middle plastochron; yet even the determination of the axillary bud of P_1 must be incomplete at early or sometimes even at middle plastochron, since this bud can be shifted by cuts made elsewhere in the apical region, as will be shown in §6.

4. ‘BUSH’ AND ‘LONG’ RACES OF *CUCURBITA PEPO* AND SPONTANEOUS REVERSALS IN THE FORMER

About the first half of the whole number of experiments, which have continued for many years, were made on apices of seedlings of bush marrows, chiefly Sutton’s ‘Tender and true’, but most of the second half on marrows of a ‘long green’ race. The purpose in using at first bush marrows was to get more leaves into each transection of the apical bud when the results were examined; for it was hoped that in the bush marrows the internodes would not begin to elongate so early. But actually very little difference was found in this respect.

On the other hand the ‘long green’ marrows had the big advantage that in them the directions of the genetic spirals and bud eccentricities when once established appeared to

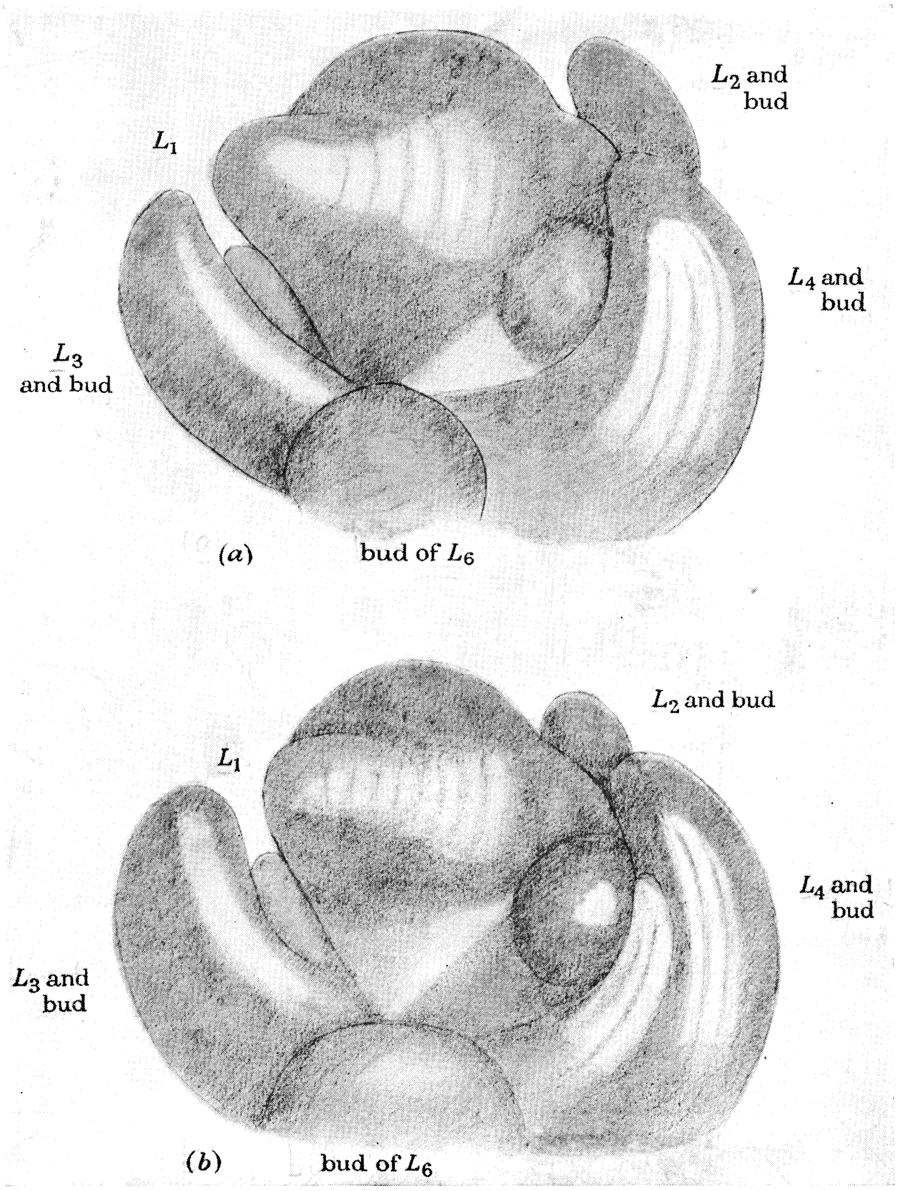
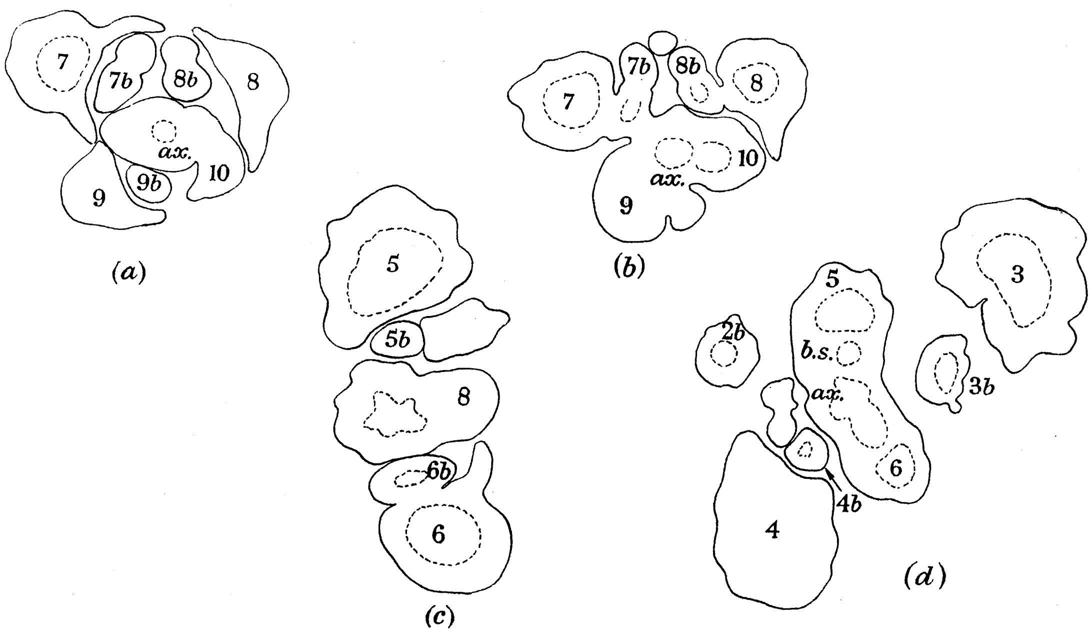


FIGURE 5, *a*, *b*. An apex of *Cucurbita pepo* seen in the solid from one side at late plastochron. *a*, seen from between leaves 1 and 2; *b*, seen more nearly from leaf 1, when one day older. The genetic spiral winds clock-wise and the leaves are numbered from the youngest downwards. The parallel lines across leaf 1 are boundaries between rows of cells.

be quite constant. Only two spontaneous reversals were ever found in the 'long green' race, and these were both in plants from one small packet of seed, which may have been hybrids due to accidental pollination from bush marrow plants. But the bush marrows reversed spontaneously quite often, as was observed when their apical buds were opened up for operation. Naturally those that were found to have reversed recently were not operated upon, but this involved a waste of labour. The spontaneous reversals were often spectacular, but it was difficult to learn much from them, since the immediate causes of the reversals were not known. After the operations also this possibility of spontaneous reversals in bush marrows had always to be considered, though they could usually be distinguished from reversals due to the operations. However for operations the 'long' races are much to be preferred.



(a) (b) (c) (d)

FIGURE 6, *a* to *d*. Apex no. 53, R. Transections of an apex of a bush marrow seedling reversing spontaneously. The leaves are numbered from the oldest of the seedling upwards, leaf 3 being the oldest in the sections shown. The original direction of the genetic spiral in this and subsequent figures is recorded after the apex number with R for right (clockwise) or L for left. Also the inner contour of the xylem is drawn at levels at which the pith has developed. *a*, *b* × 30, *c* × 22, *d* × 14.

In being liable to reverse their genetic spirals spontaneously the bush marrows differ sharply not only from the 'long' races of marrows, and apparently from the wild species of Cucurbitaceae, but also from species of other families with spiral phyllotaxis and with median axillary buds or with none. For in other families the rule is that in any one shoot apex the genetic spiral along which the leaves are formed does not reverse unless the apex suffers an experimental operation or an accidental injury, such as an insect bite. This difference suggests that in the bush marrows the spontaneous reversals of bud eccentricity may be the primary abnormality, and may be the cause of the reversals of the genetic spirals. In one bush marrow seedling, of which transections at different levels from above downwards are shown in figure 6, the bud eccentricity did indeed reverse spontaneously at leaf 8, one step before the reversal of the genetic spiral, and two steps before the first leaf of the reversed spiral (see figure 6*d*). A drawing of another similar apex showed that it had reversed in just the same way.

These two reversals of genetic spiral can readily be explained on space-filling principles as being due to the previous reversals of bud eccentricity. Thus in figure 6, leaf 8 is the first leaf having a bud with reversed eccentricity. The genetic spiral continues nevertheless to leaf 9, which is not in contact with 8, but 10 arises in a quite abnormal angular position, in the gap between 8 and 9 instead of that between 7 and 8, and thus on the wrong side of the apex, so that the direction of the spiral is reversed at (or from) leaf 9, as figure 6*a* shows. Clearly the abnormal position of leaf 10 can very well be explained as being due to the reversed eccentricity of the bud of 8; for this bud, together with the bud of 7, fills up the gap between leaves 7 and 8, in which leaf 10 would normally have arisen, so that 10 arises between 8 and 9 instead. However, in a few other 'bush' marrow apices the reversals of bud eccentricity and genetic spiral were reached gradually through transitional stages, so that the time relations of the changes were not clear. These apices would be more difficult to interpret.

It may be mentioned that neither in the spontaneous reversals of the bush marrows, nor in reversals caused by operations in apices of either race were the directions of genetic spiral and bud eccentricity ever in disagreement for more than two consecutive steps. The spontaneous reversals were found especially frequently in the earliest stages of seedling growth, from the fourth leaf of the seedling to about the tenth. The spontaneous irregularities in the bush marrows were not studied further, since for the purpose of solving the two main problems proposed the experiments seemed more promising.

5. Methods of experiment

The marrow seedlings, grown in pots in a greenhouse in summer, were operated upon as soon as the young shoots had grown up from between the cotyledons far enough to be easily accessible. At this stage the youngest leaf primordium is usually the 11th, 12th, or 13th of the seedling, and the oldest leaf about 2·5 cm. long. The stem apices were exposed in a cool room and under a dissecting binocular microscope, by cutting down, only just so far as was necessary, those young leaves that covered the apex or got in the way. This was usually done free-hand and then, usually after a delay of several hours or overnight with the apices lightly covered over, the operational cut was made with the knife held in a Saunders-Singer 'pantograph' micro-dissector at magnification × 50 or × 75. The delay served to allow the bleeding water which came from the earlier cuts to dry off or be re-absorbed. This makes the vision much clearer, and it was further improved by arranging as close as possible above the apex a small diffusing screen of ground glass which was illuminated from opposite the observer and steeply from above with a low-voltage projection lamp. Only just enough room was left between the diffusing screen and the apex for the nearly horizontal cataract knife used in operating. This arrangement is helpful in operating on any kind of stem apex since it prevents glittering reflexions, and it is especially needed with the *Cucurbita* apex which is very transparent and glittering without it.

After a day or two for recovery, the plants were returned to the greenhouse with their apices well covered with cotton-wool and were left to continue growing, usually for a period of 12 to 14 days from the operation, after which time P_1 could usually be seen just emerging from the apical bud. The petioles of the older leaves were left to facilitate counting up from the base in order to identify P_1, but their blades were removed to keep the plants from becoming pot-bound, since the large leafy cotyledons were found to be fully enough to keep the shoot apex growing and developing rapidly. Finally the shoot apices operated upon were fixed and stored in alcohol and were later embedded in collodion, sectioned free-hand, and drawn under a projecting microscope. The *Cucurbita* shoot apices often curve strongly towards the wound at about the P_1 level, which makes it difficult to keep the sections transverse.

The stem apices in *Cucurbita* are reached very quickly and seem less protected than in most families; yet they were seldom found to have been attacked by insects. The younger leaves of the bud, cut down part way, regenerated very quickly. Unfortunately no convenient way was found of comparing quantitatively the eccentricities of different axillary buds. The chief reason for this was that not only do the eccentric buds arise at the anodic sides of their subtending leaves, but they also grow out obliquely in the anodic direction, and not in a radial plane through the subtending leaf and the main axis. Consequently the higher the level of a section through an axillary bud, the greater appears to be its eccentricity or lateral displacement measured in degrees (see figure 1). To avoid this difficulty, the eccentricities of buds were estimated, whenever possible, in sections at a standard level, the lowest level at which the bud concerned was just about to separate from the main axis. But a suitable section at this level was not always available, and with various other complications after the operations it only seemed justifiable to judge the buds by eye and group them as fully or normally eccentric, subeccentric, nearly median and median, with a similar grouping for reversed buds. However when a bud is median or nearly median it leaps to the eye as being so, especially since such buds grow straight up instead of obliquely sideways. The position of the bud was naturally taken to be that of its morphological centre, marked by the stele.

6. KATHODIC CONFINEMENTS OF I_1 OR P_1 IN *CUCURBITA PEPO*

The simplest and best experiment for revealing something of the cause of bud eccentricity in *Cucurbita* was found to be to make a radial vertical cut in the apex at the kathodic side of I_1, the presumptive area of the next leaf rudiment due to arise, and extending a little way below it. A similar cut is also effective, though less so, if made at the kathodic side of P_1, the youngest visible leaf rudiment, at early plastochron. These operations will be called kathodic confinements of I_1 or P_1, and when operations upon I_1 or P_1 are being considered together the leaf area or leaf operated upon will be called 'n', and successively younger or older leaves $n+1$, or $n-1$, and so on. The cuts were usually continued downwards far enough to separate n from $n-3$ and its bud, as well as from $n-1$ (see figure 7*a*).

The results of thirteen kathodic confinements of I_1 and six of P_1 are given in tables 1 and 2. It can be seen that when I_1 was confined, in three apices its axillary bud was nearly median, in six it was very subeccentric or subeccentric, and in four it was only slightly subeccentric or normal. When P_1 was confined at early plastochron, the changes were similar but smaller. Figures 7 and 8 show transections of two of these apices from above downwards.

These results strongly support the suggestion made in §3, and based on observations of young seedlings, that normally the buds are made anodic by some repelling influence coming from the next older leaf and bud. Further, although the confining cuts usually separated the confined leaf *n* from $n-3$ as well as from $n-1$, in six apices marked by asterisks in the tables they did not separate *n* from $n-3$, or not completely, either because the cut was not continued downwards to the level of insertion of $n-3$ and its bud (apices 129, 132, 125) or because the cut was deliberately placed further kathodic than the kathodic margin of *n*, so that it passed down right through the insertion of $n-3$ (apices 101, 102, 107). Yet the changes caused in the eccentricity of the bud of *n* were about as big in these six apices as in the others. This indicates that little, if any, of the influence repelling the bud of *n* comes from $n-3$, the leaf closest to its kathodic side. Most of it must come from $n-1$, or at least from the direction of $n-1$.

Table 1. Thirteen kathodic confinements of I_1

protocol number of apex	stage of plastochron at operation	eccentricity of bud of *n* (I_1)
77	middle	nearly median
93	early	
102*	not noted	
101*	middle	very subeccentric
129*	middle	
132*	middle	subeccentric
96	late	
107*	early	
109	late	
123	early	slightly subeccentric
76	middle	normally eccentric
113	late	
122	early	

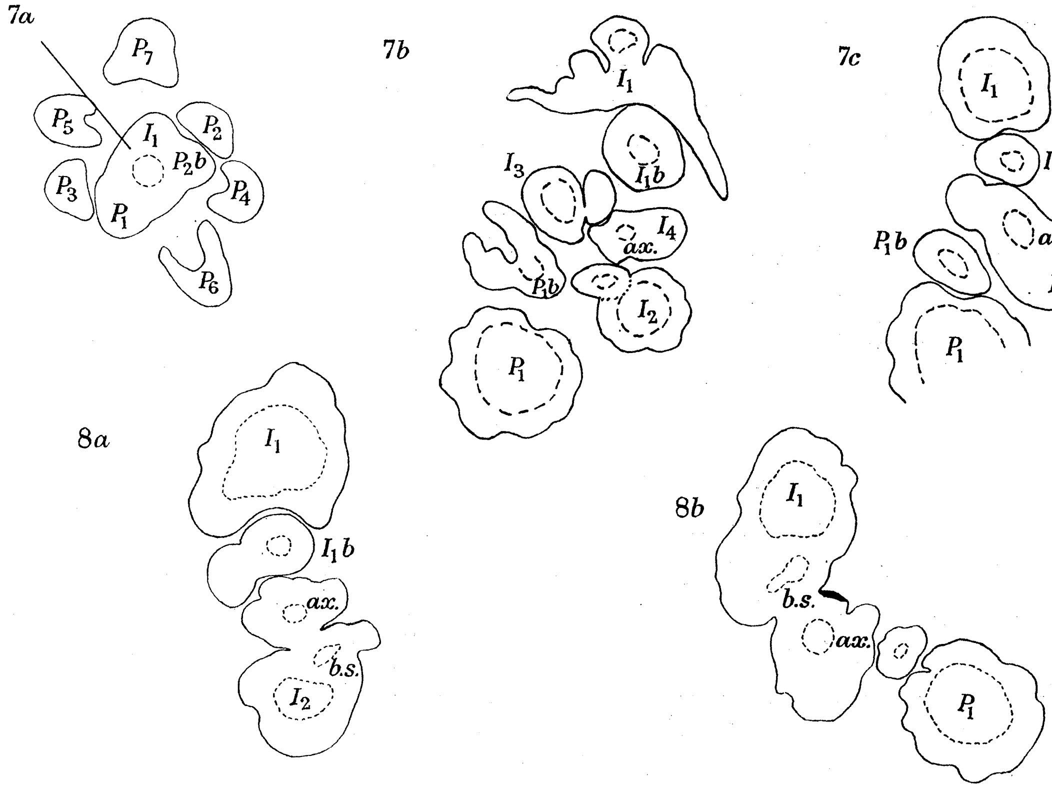
Note: in this table and the next an asterisk signifies that the cut separated *n* from $n-1$ only: see text.

Table 2. Six kathodic confinements of P_1

protocol number of apex	stage of plastochron at operation	eccentricity of bud of *n* (P_1)
114	early	very subeccentric
125*	early	subeccentric
126	early	subeccentric
95	early	slightly subeccentric
112	middle	slightly subeccentric
115	early	normally eccentric

It needs, however, to be considered how it was that after a few of the kathodic confinements, as the tables show, the eccentricity of the bud of the confined leaf was diminished only slightly, or not at all. The explanation is probably that in these apices the sides of the cut came into contact and grew together again soon after the operation, leaving not even a scar to show where the cut had been made. It is indeed a great difficulty with the *Cucurbita* apices that cuts made in them are very liable to close up again in just this way without leaving any trace, more so than in any other species on which the writer has operated. Consequently, after making a cut in a *Cucurbita* apex, it was always necessary to continue observing it for some minutes at least, and to press apart gently the sides of the cut when they tended to come together, besides making sure that the cut was not becoming filled with water of bleeding. Only then were the apices covered over with an arch of cotton-wool and left, and even so it quite often seemed later that the cuts had healed up, usually without leaving any scar.

A great advantage of the kathodic confinements was that apart from the changes in the eccentricity of the bud of the confined leaf *n*, they made, with one exception, no significant changes in the buds of any later leaves, and they did not upset the genetic spiral of the leaves, nor change their positions more than a little. For they consequently led to clear and simple pictures, which quickly drew attention to the main significant effect of the operation, the change in the eccentricity of the bud of leaf *n*. The one exception was found in apex no. 125, with P_1 confined, in which I_1 had a median bud, while the I_1 leaf itself failed not far above its insertion. This failure of the upstanding part of I_1, which of course was not present at operation, suggests a secondary and later injury of some kind, probably an insect bite, and the same injury may in some way unknown have made the bud of I_1 median. The bud of P_1 was subeccentric.



FIGURES 7, *b*, *c*, and 8, *a*, *b*. Transections of two apices, nos. 77, R, and 129, L, with I_1 confined kathodic. 7*a* is a transection of a normal apex showing the position of the cut in such an operation. In 8*a* a scar from the operation is shown black, and P_1 is small and its bud small and subeccentric, perhaps from an accidental injury.

These statements are based on examination of transections of the apices, which at the time of pickling had usually made from 8 to 10 leaves after the operation, or even more. But some of the younger leaves and buds were missed, either because the section at the necessary level was spoilt, or because a few of the apices made only a few leaves after the operation and then ceased growing, perhaps as a result of insect bites. Some small changes in the positions of leaf $n+3$, which are critical for the phyllotactic theory, will be discussed in §11.

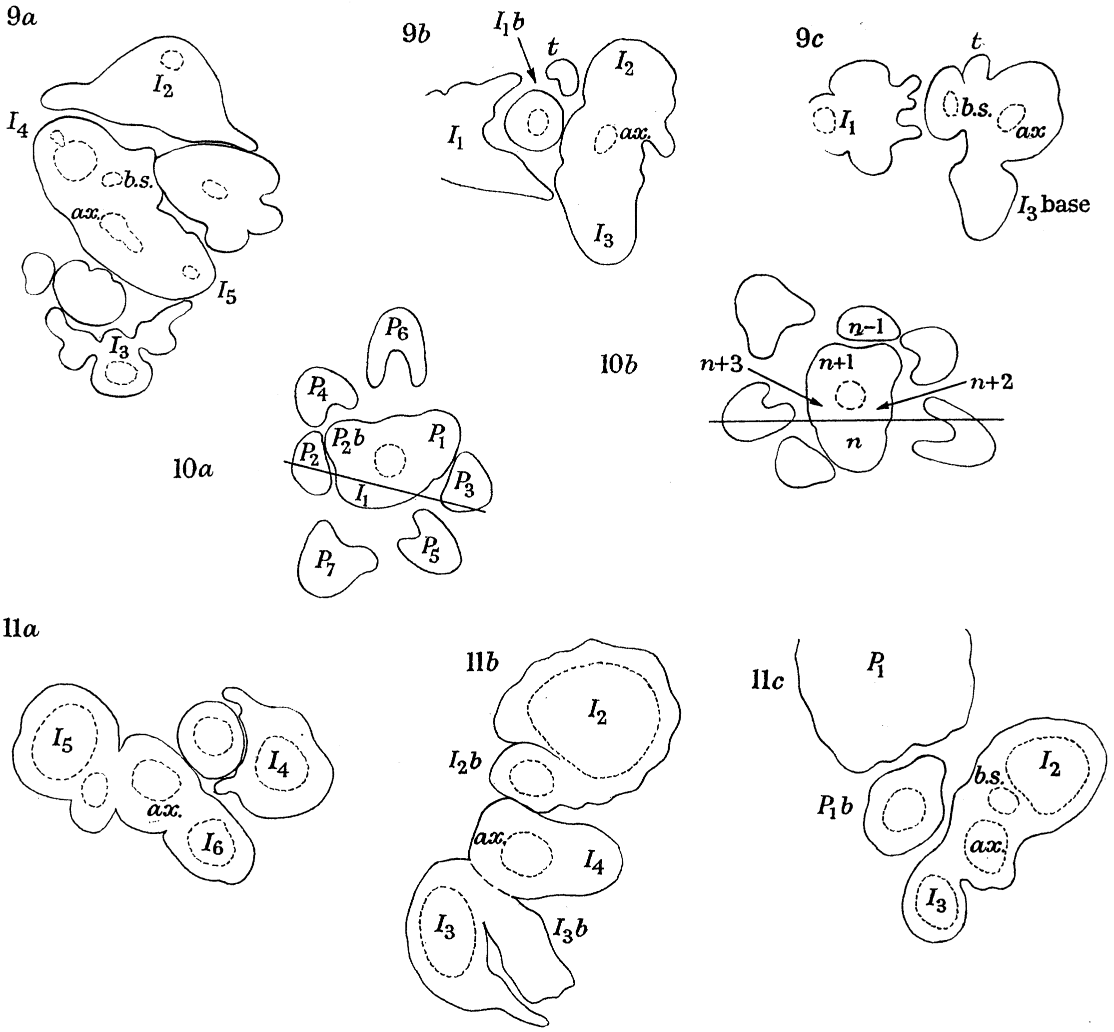
Anodic confinements of P_1 or I_1—that is, radial vertical cuts at the anodic side of P_1 or I_1, encounter the difficulty that they are likely, if at all close to the confined leaf, to enter and damage its presumptive bud area. When this happens the bud may arise in a more nearly median position instead, sometimes showing marks of injury on its anodic side; but not much can be concluded from this. When the cut was far enough away from the anodic side of the P_1 or I_1 to leave the bud area intact, it was not found to change the position in which the bud arose. Only a few operations, indeed, were made deliberately to test this point; but if anodic confining cuts at this distance could change the positions of the buds of P_1 or I_1, then some of the kathodic confinements of I_1 reported above would have been expected to act also as anodic confinements of P_1, and to change the position of its bud; and the same would have been expected of some of the isolations and extirpations of I_1 to be reported in the next section. Also several isolations and extirpations of I_2, to be reported in §10, would have been expected to displace the bud of I_1. But no such effects were found. It seems, therefore, that the eccentricity of an axillary bud in *Cucurbita* depends on an influence that travels in the anodic direction of the genetic spiral only.

7. ISOLATIONS AND EXTIRPATIONS OF I_1 OR P_1

In an earlier series of operations I_1 or P_1 was isolated from the apex with a vertical cut at right angles to the radius through the isolated area or leaf, which will again be called 'n'. The main purpose was to test whether the bud of $n+1$ would be less eccentric if separated from any repelling influence from n or its bud. But although the isolated pieces were well pressed back, they often managed to join up again with the main apex, and then the operation had no effect. So except in the earliest operations the isolated pieces were usually cut right off at the base so that the operation became an extirpation. This caused the loss of some information that might have been of interest, concerning the development of buds on the isolated leaves, but it seemed best to concentrate on the main question, how the bud of $n+1$ would be affected.

The results of five operations on P_1 and eight on I_1 are summarized in table 3, and sections of two of the apices are shown in figures 9 to 11. It can be seen that the bud of $n+1$ was indeed often made much less eccentric than the normal; but the pictures presented by transections of these apices were mostly less clear than those of the kathodic confinements, since the isolating cuts made the apices grow more irregularly and disturbed the phyllotaxis more. The genetic spirals of the leaves did, however, continue in all the apices of table 3, with the eccentricities of their buds, apart from those of $n+1$ and $n+3$, not far from normal, so far as could be made out. A few apices in which the spirals reversed or the phyllotaxis became irregular have been omitted from the table since the eccentricities of their buds are not comparable.

It can also be seen that the bud of $n+3$ was often made very subeccentric or even median. Indeed the eccentricity of this bud was diminished quite as much as that of the bud of $n+1$. This unexpected result was at first thought to be due to a persisting effect of the isolation and extirpation of n. But later when the lateral confinements of I_1 or P_1 were found so clearly and definitely to diminish the eccentricity of the axillary bud of that one leaf only, another explanation seemed more probable. This is that when I_1 or P_1 was isolated from the apex, which in *Cucurbita* is rather flat-topped and not provided with any easy guiding marks, the cut often approached rather too close to the centre of the apex, so that it acted as a kathodic confinement of $n+3$, as well as of n, in the manner indicated in figure 10*b*. In apices of other families the cuts would usually have left scars, and the positions of the scars might have settled the question. But in these *Cucurbita* apices the cuts, as usual, left no scars nor traces of any kind.



FIGURES 9 to 11. 9, *a* to *c*, an apex, no. 37, R, with P_1 isolated and extirpated. In *c* can be seen what is apparently a downward prolongation of I_3. *t*, tendril. 10*a*, section of a normal apex with the position of a cut isolating I_1 marked on it. 10*b*, section of a normal apex showing how a cut isolating a leaf or area n may act as a kathodic confinement of $n+3$. 11, *a* to *c*, an apex, no. 49, L, with I_1 isolated and extirpated.

It should be mentioned that the first effect of the isolating cuts on the centre or growing point of the stem apex appeared to have been to make it shift away from the cut, as in apices of other families, this being often indicated by changes in the divergence angles between the leaves arising just after the operation. But soon the young axis below curved strongly *towards* the cut, which much increased the difficulty of keeping the sections transverse.

Table 3. Thirteen isolations of P_1 or I_1

protocol number of of apex	identity of n, the leaf or area isolated	stage of plastochron at operation	eccentricity of bud of $n+1$	eccentricity of bud of $n+3$
11	P_1	early	very subeccentric	very subeccentric
13	P_1	early	subeccentric	median and double
37	P_1	middle	very subeccentric	normally eccentric
38	P_1	middle	very subeccentric	very subeccentric
35	P_1	middle	normally eccentric	median
43	I_1	late	very subeccentric	very subeccentric
48	I_1	late	very subeccentric	median
49	I_1	late	very subeccentric	slightly subeccentric
45	I_1	middle	subeccentric	normally eccentric
47	I_1	late	subeccentric	subeccentric
46	I_1	late	normally eccentric	subeccentric
41	I_1	middle	normally eccentric	slightly subeccentric
32	I_1	late	normally eccentric	normally eccentric

Note: in apices 11 and 13, n was only isolated. In the others it was isolated and then extirpated.

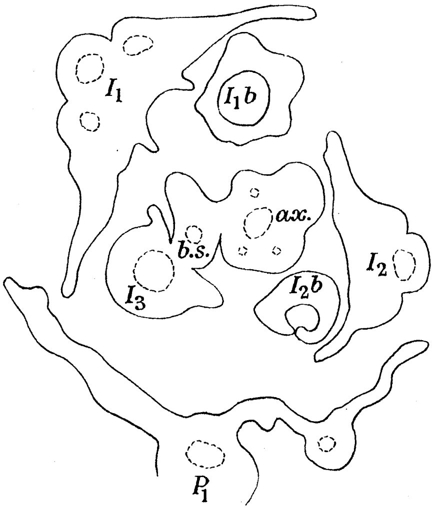
It can be understood that the kathodic confinements acted on the bud of leaf n only, whereas extirpations acted on the bud of $n+1$. For the confinements interrupted the influence repelling the buds which would have reached n from the kathodic direction, but left n itself and its bud intact, so that a similar influence may have started again from n, and may then have made the bud of $n+1$ normally eccentric. But the extirpating cuts, which also penetrate quite deeply into the apex and axis, not only interrupt the repelling influence but destroy n and its bud also. Consequently a repelling influence may not be able to start again until it can start from $n+1$: and then the bud of $n+2$ will be the first bud which it will make normally eccentric. The origin and course of the repelling influence will be further discussed in the next section.

8. Experiments revealing the importance of the depth of the cut

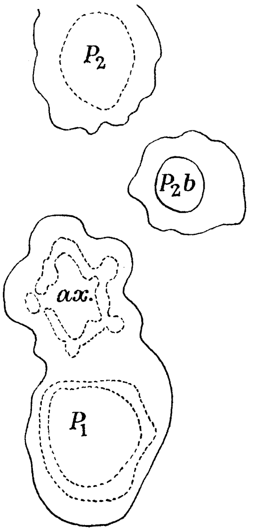
The results reported so far have supported strongly the suggestion that an influence repelling the bud of any leaf n and so making it anodic comes mainly from leaf $n-1$ or from the bud of that leaf, thus travelling along the genetic spiral. But a surprise was still in store, and it came to light when an attempt was made to decide whether the repelling influence comes from the bud of leaf $n-1$ or from the leaf itself. In four apices the area presumptive for the bud of P_1 was extirpated with two cuts inclined in a V, in two of the apices at mid-plastochron and in two at late-plastochron. The P_1's, which all developed well, had no buds, but yet the buds of the I_1's were all quite normally eccentric, and so also were those of the subsequent leaves. The genetic leaf spirals continued normally. Sections of one of these apices are shown in figure 12.

In six apices the upstanding part of P_1 was cut off with a cut or cuts which were inclined outwards flush with the surface of the apex, instead of descending vertically into the tissues of the apex and axis as in the previous isolations and extirpations. The presumptive area of the bud of P_1 was left intact, so far as possible. The results are summarized in table 4, and an example is shown in figure 13.

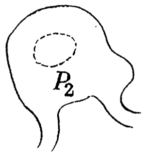
12*a*

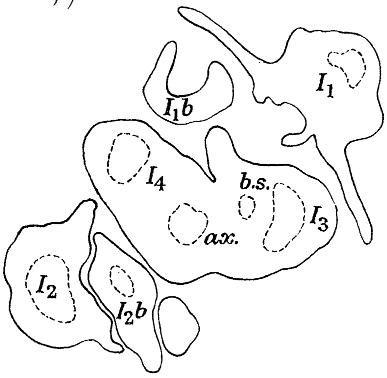


12*b*



13*a*





13*b*

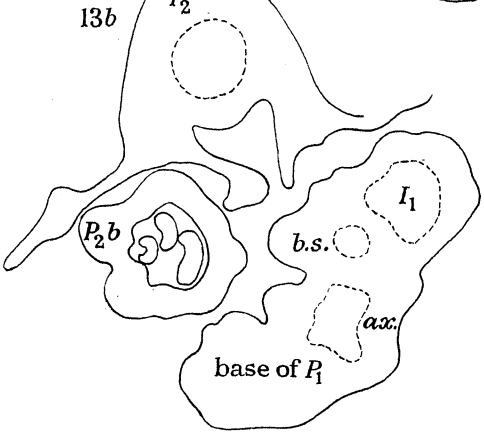


FIGURE 12, *a*, *b*. An apex, no. 142, R, from which the area presumptive for the bud of P_1 had been extirpated.

FIGURE 13, *a*, *b*. An apex, no. 133, L, in which P_1 had been cut down to its base.

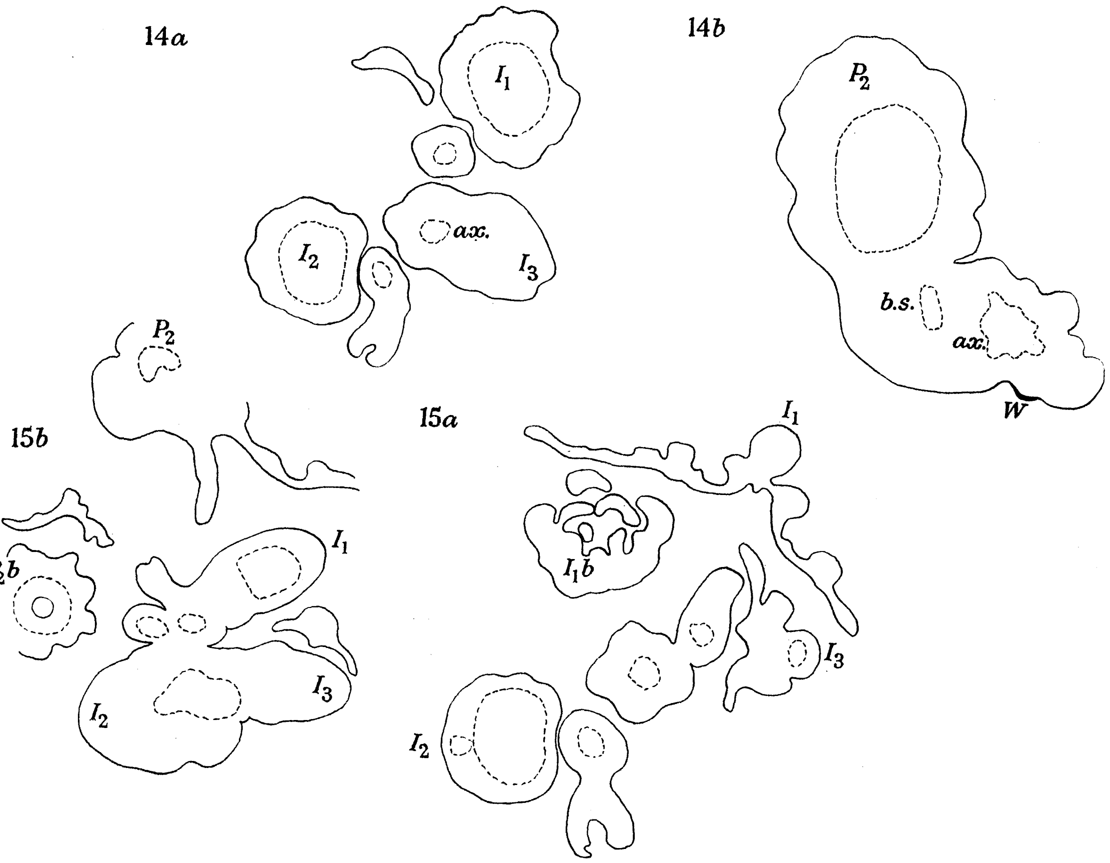
TABLE 4. SIX APICES IN WHICH P_1 WAS CUT DOWN TO THE BASE

protocol number	stage of plastochron at operation	development of P_1	development of bud of P_1	eccentricity of bud of I_1
136	middle	nil	normal	normally eccentric
137	middle	nil	normal	
134	late	disorganized base only	normal	
133	middle	disorganized base only	nil	
135	late	nil	nil	
143	early	disorganized base only	nil	

Table 4 shows that although nothing or almost nothing of P_1 developed, the I_1 buds were again all normally eccentric, both in three apices in which the bud of P_1 developed and in three others in which it did not. The later buds were also normally eccentric, and the genetic spirals continued normally, except that in one apex (no. 137) I_2 and its bud had apparently been eaten away by an insect, and in another (no. 135) the positions of I_3 and I_4 were abnormal.

TABLE 5. SIX APICES IN WHICH P_1 WAS SPLIT VERTICALLY AT EARLY PLASTOCHRON

protocol number	development of P_1	development of bud of P_1	eccentricity of bud of I_1
158	moribund base only	anodic bud	slightly subeccentric
165	basal swelling divided by wound		
170	base only		
164	small moribund base	nil	normally eccentric
166	nil		
190	nil		



FIGURES 14, *a*, *b*, and 15, *a*, *b*. Two apices, nos. 158, L, and 190, L, in which P_1 had been destroyed with a vertical split at early plastochron. A scar from the wound is labelled *W* in 14*b* and shown black.

Another method of preventing P_1 from developing, without making any deep cut into the apex or axis is to split it with a median vertical cut at early plastochron: for P_1 at this stage seems to be especially sensitive to injury. The results of six such operations are summarized in table 5, and two examples are illustrated in figures 14 and 15. Table 5 supports the results of table 4 by showing that although little or nothing of P_1 developed, the I_1 buds were normally eccentric or, in three apices, only slightly subeccentric. In these last three apices the buds of the P_1's had developed and not in the other three, probably because they lacked a determining influence from the missing leaf (see Pellegrini 1963; Snow, M. & Snow, R. 1942); but it is uncertain whether this has any significance or is only coincidence. The later buds were all normally eccentric and the genetic spirals continued normally.

The negative or almost negative results of these experiments differ markedly from those of the kathodic confinements and of the isolations and extirpations of P_1 or I_1; and there can be little doubt that the cause of this difference was that in the confinements and isolations the cuts penetrated fairly deeply into the apex and axis, whereas in the experiments of this section they penetrated only slightly or not at all. It may indeed seem that in the radial splits of young P_1's the cuts were not very different from those in the kathodic confinements; but they almost certainly were made less deep, partly because they started by penetrating a leaf rudiment that was already projecting slightly from the apex, and partly because in the splitting of the P_1's the idea was not to confine anything by making a lateral barrier in the apex, but only to inactivate the P_1 itself.

If this explanation is correct, it follows that the influence repelling the buds and so making them eccentric must travel at some little depth below the surface in the direction of the genetic spiral of the leaves, and must be able, when once started, to continue past a node at which the normal P_1 leaf rudiment and its bud area are missing or inactivated, as if it possessed a kind of physiological momentum. Yet there is evidence that the starting of the bud-repelling influence, or the re-starting of it when it has been blocked, does depend on a leaf or its bud. For, as pointed out in §3, the direction of the genetic spiral is decided as soon as the third leaf of the young seedling arises to right or left of the median plane through the first two leaves; and therewith the direction of travel of the bud-repelling influence is also decided, the bud of the third leaf being displaced right or left to the side away from the second leaf, which appears to repel it. Similarly, when P_1 or I_1, to be called here leaf n, is confined with a cut at its kathodic side, then, as pointed out already, n has a subeccentric or median bud, but $n+1$ has a normally eccentric bud, since the repelling influence can start again from leaf n or its bud after being interrupted by the cut.

As a further test of this interpretation based on differences in depth of cut, the following operation was performed. From the side of one apex between P_2 and P_1 there was cut out at early plastochron a piece of tissue which was made as nearly as possible equal in width and in radial depth to the pieces cut out in the isolations and extirpations of P_1 and I_1. The leaves on each side were, so far as possible, left uninjured. The idea was that if in the isolations and extirpations the radial depth of the cut was the factor effective in causing the bud of the next leaf, $n+1$, to be subeccentric or median, then the present operation might be expected to do the same to the bud of P_1, the leaf at the anodic side of the piece removed.

Actually in one apex operated on in this way, figure 16, the bud of P_1 was indeed median and the subsequent changes were also of interest. For the angle P_1 and I_1 was increased to nearly 180°, probably through the shift of the growing-point away from the wound, and I_2 arose on the opposite side of the apex to the normal, probably because the cut reached up rather high, as was noted at the time of operation, and so made the normal position of I_2 unavailable. Thereby the genetic spiral was reversed and consequently the buds from I_2 onwards had the reversed eccentricity appropriate to the new counter-clockwise direction of the spiral. The cut left exceptionally two small scars, as figure 16*d* shows. This apex forms an instructive contrast with the bush marrow apex, figure 6, no. 53, in which a spontaneous change of bud eccentricity appeared to have been primary, and to have led to a reversal of spiral by its space-filling effects.

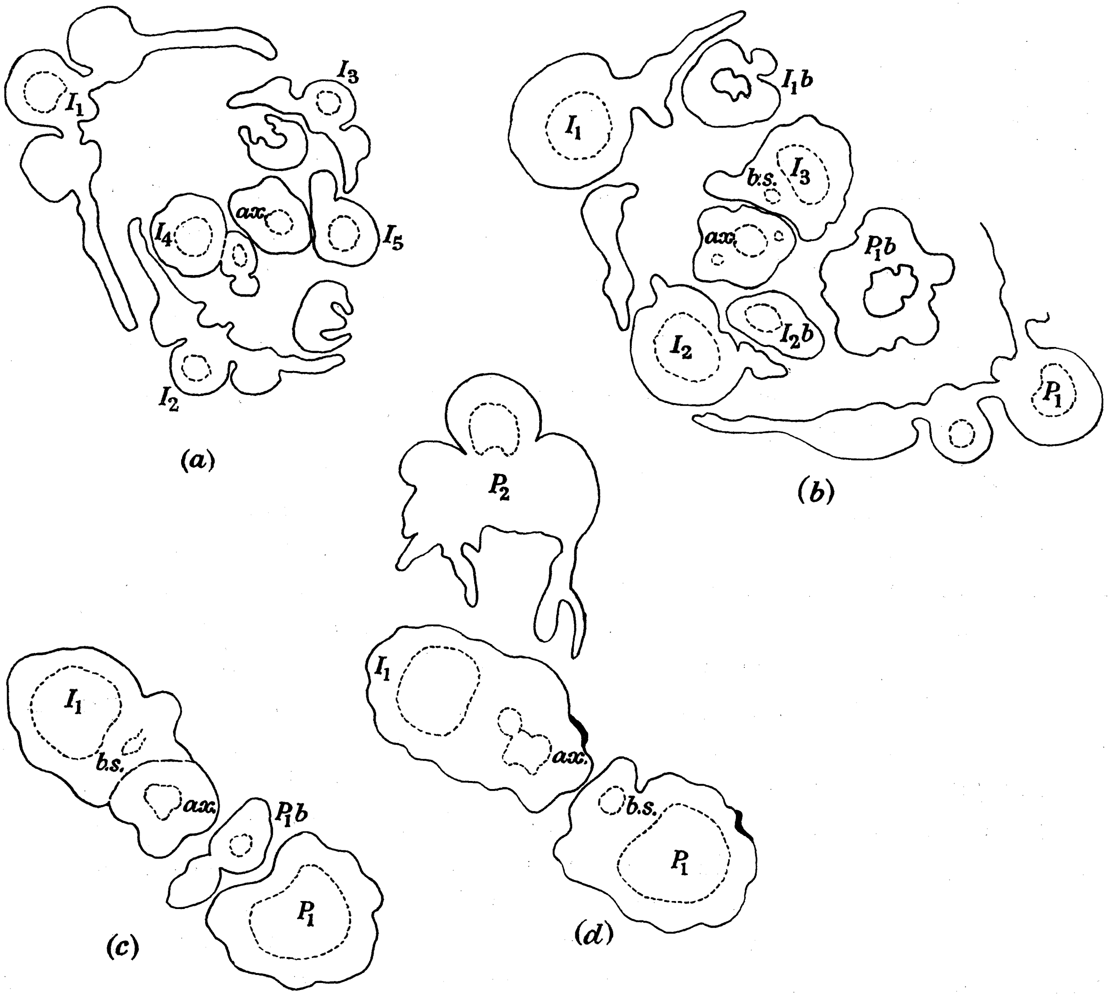


FIGURE 16, *a* to *d*. An apex, no. 153, R, from which a wide piece of tissue between P_2 and P_1 had been removed. Scar from the operation is shown black in *d*.

The last described operation is, however, very awkward since there is scarcely enough room for it between two successive leaves; and in three other apices similar operations attempted did not change the position of the bud of the leaf at the anodic side of the piece of tissue removed, probably because the cuts were not made radially deep enough for fear of injuring the neighbouring leaves.

9. The course of the conducting strands in comparison with that of the bud-repelling influence

It is difficult to decide how relevant may be the course of the conducting strands to the problem of the bud-repelling influence in *Cucurbita*. Since the bud of P_2 can sometimes be seen arising as a low hummock at late plastochron, or occasionally even at middle plastochron, it must have been determined in its anodic position by early plastochron at latest. At this early stage examination of the tissues in transections makes it seem very unlikely that there is yet any functional conducting strand leading to P_2 and its bud, even if a few cell divisions might be found indicating where such a strand was about to develop. However, it seemed desirable to discover whether actually the courses followed by the bud-repelling influence and later by the developing conducting strands do correspond in any way; and for this purpose the writer has made some simple observations on young zones of *Cucurbita* shoots cleared in chloral hydrate and examined at low magnification in the chloral hydrate over a black glass. The opaque white strands showed up very well, and when necessary the pieces of shoot were split down one side and opened out flat, to simplify the picture.

If one follows the three trace bundles from a leaf down into the stem, one finds that just below the leaf insertion they enter a partial node within which they are all laterally connected, and with which the axillary bud is also connected. Below the node the three leaf traces continue downwards, but the node also gives off lateral branch bundles, of which two wind very obliquely downwards round the axis in the kathodic direction and connect with the partial node of the next older leaf. A third branch starting from the anodic side of the node winds rather more steeply downwards in the opposite direction and connects with the partial node of $n-2$.

The course, therefore, of the conspicuous band formed by the median and kathodic traces of each successive leaf does correspond remarkably with the course followed upwards by the bud-repelling influence: but it hardly seems possible that this influence can move along the traces, since, as already pointed out, they differentiate too late. However there is another possibility worth bearing in mind. It may be that in the Cucurbitaceae the cells of the apical tissue acquire at an early stage a new polarity along the line of the genetic spiral of the leaves, and that this polarity guides both the bud-repelling influence and also, at a later stage, the formation of a continuous band by the joining up lengthways of two of the branch bundles from sucessive nodes. It would be of interest to find whether a similar band is formed in the Passifloraceae, in which family also the axillary buds are shown in Hagerup's illustrations (1930) to be regularly eccentric and anodic, though only very slightly so.

10. Isolations and extirpations of I_2, and the origin of stunted leaves

In a last series of operations the presumptive area of I_2, or in two apices of I_1 at early plastochron, was isolated with a vertical cut and extirpated in the manner described for P_1 and at later plastochron for I_1 in §7. The reporting of the results has been left to the present section since, although some incidental points of much interest appeared in them, these operations turned out to be not well suited to the purpose of studying the bud-repelling influence. This was because after most of them a new leaf which will be called $I_{2'}$ or $I_{1'}$, or sometimes n', arose near the normal angular position of the extirpated I_2 or I_1, though usually a little way kathodic from it and at a higher level, often as high as that of $n+1$ or even higher, and so after a delay of one plastochron or more. An example is shown in figure 17.

Rather similar results were obtained previously after 13 out of 21 isolations of I_2 in *Lupinus albus* (Snow, M. & Snow, R. 1931, pp. 27, 42), in which species it was much easier to make out afterwards just where the $I_{2'}$ had arisen in relation to the isolating cut. We concluded that the origin of an $I_{2'}$ leaf was due to the very big shift of the growing-point of the stem apex away from the wound, a shift which became progressively greater as the operation was made on progressively younger leaf areas, and so made progressively more space available for leaf-formation above the wound: and this explanation may well hold good for the *Cucurbita* apices also. In some of the lupin apices both I_2 and $I_{2'}$ developed, but in the *Cucurbita* apices I_2 had been extirpated after being isolated.

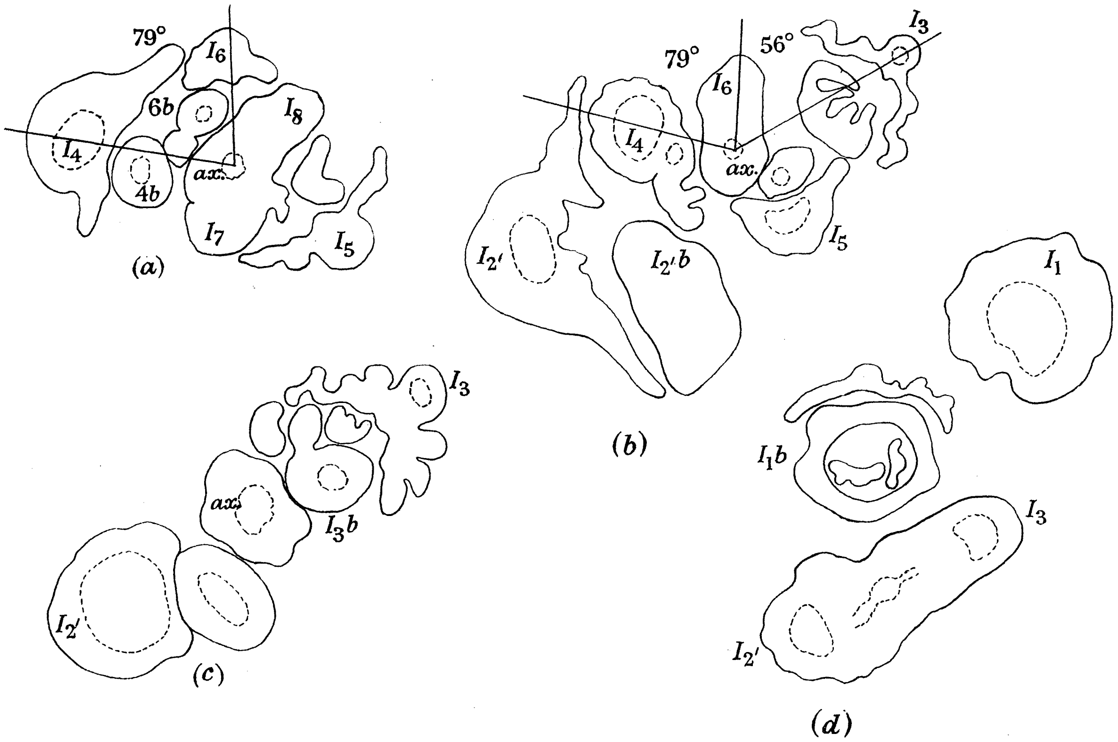
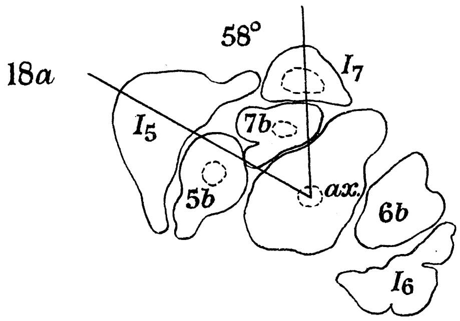
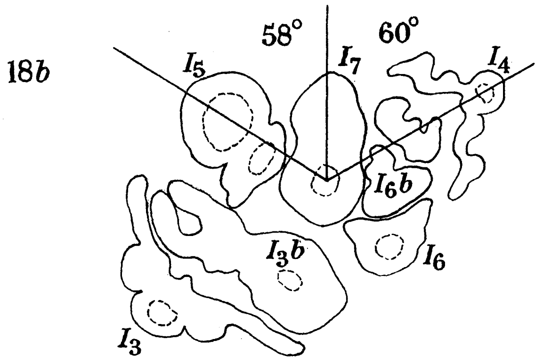


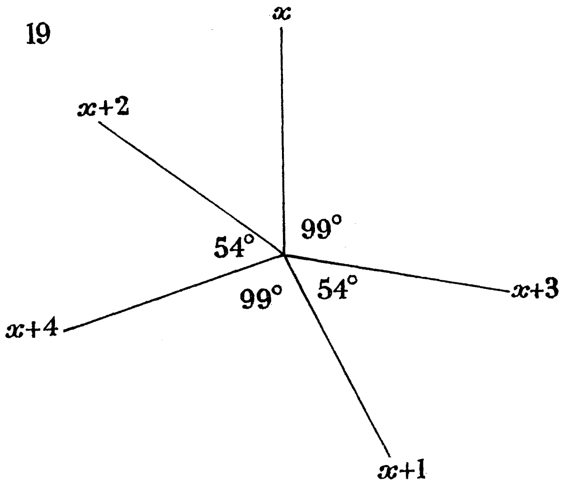
FIGURE 17, *a* to *d*. An apex, no. 172, L, in which I_2 had been isolated and extirpated, and an $I_{2'}$ leaf developed. The bud of I_3 was just reversed and nearly median, and I_3 itself was stunted.

The n' leaves rather spoilt the present operations as tests of the repelling influence of leaf n on the bud of $n+1$. For when the bud of $n+1$ was normally eccentric, then n' might have acted on it like a normal leaf n; and when the bud of $n+1$ was subeccentric or median, then n' might have developed too late to make it fully eccentric. Actually out of eight of the present apices in which the genetic spiral continued, in two the bud of $n+1$ was nearly median, in two it was subeccentric and in four normally eccentric. In the remaining six apices the leaf positions were completely changed, and this complicated the interpretations of the changes in bud eccentricity.

But in compensation these operations led to some other interesting results of which one seems to deserve mention, though not relevant to the two main problems of this paper. There often arose after these operations on I_2, or on I_1 at early plastochron, leaves of a kind which will be called 'stunted'. These leaves developed from bases of about the normal size, but their upper parts, though they formed petioles and blades of normal shape, were much reduced in their upward growth, so that they were commonly outgrown by one or more leaves that were younger and inserted higher. Examples are shown in figures 17 and 18. Leaves of this kind were not found after operations on P_1 or on I_1 at middle or late plastochron, but two of the thirteen $I_{2'}$ leaves formed after the previous isolations of I_2 in *Lupinus albus* were perhaps rather similar, since one of them formed no petiole and the other only an abnormal petiole after some delay (Snow, M. & Snow, R. 1931, figs. 15, 16 and p. 30). These two $I_{2'}$ leaves contrasted with the eight I_2 leaves which were formed on the isolated pieces in *Lupinus*, and which often developed from much smaller bases, but then made petioles and leaflets of nearly normal form.







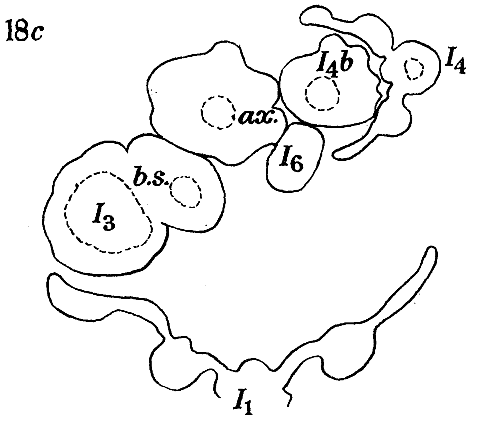


FIGURE 18, *a* to *c*. An apex, no. 179, L, in which I_2 had been isolated and extirpated, and no $I_{2'}$ developed. I_4 had a median bud and was stunted.

FIGURE 19. A diagram showing the angular positions of the centres of five successive leaves with divergence angles of 153° between them. The genetic spiral winds clockwise.

In the present series of thirteen isolations and extirpations of I_2 or of a young I_1, to which may be added one radial vertical split of a young I_1, seven stunted leaves were formed, four of them being n', one $n+1$ and two $n+2$. One of the stunted n' leaves arose above the radially split I_1 area, which did not itself develop. An interesting point is that the buds of five of these seven stunted leaves were median or nearly median, whilst those of the remaining two were very subeccentric and slightly subeccentric respectively. However there were also amongst these apices three median or very subeccentric buds that were subtended by normal leaves, one by an n', one by an $n+1$ and one by an $n+2$. It does not seem possible to interpret the stunted leaves and their nearly median or very subeccentric buds without further investigation.

11. THE LARGE DIVERGENCE ANGLES OF THE CUCURBITACEAE IN RELATION TO THE ECCENTRIC BUDS

There remains to be considered the problem of the abnormal spiral phyllotaxis of the Cucurbitaceae, with its unusually large divergence angles, which was briefly described in the introduction. An explanation suggested to the writer was also there set out, according to which the large divergence angles are due to the anodic axillary buds, which successively displace in the anodic direction the position in which each new leaf will be determined. Evidence in favour of this explanation from the development of the young seedlings was given in §3, and evidence from the experiments can now be added.

When after one of the various operations a median or nearly median bud is formed in the axil of any leaf, which will be called x since it is not always the same leaf as n, the leaf operated upon, then, if the suggested explanation of the phyllotaxis is correct, the centre of leaf $x+3$, which makes contact with the bud of x, will be expected to divide the angle between the centre of x and that of $x+1$, the other contact member, in a ratio different from that which is normal in *Cucurbita*: for the centre of $x+3$ should then be nearer to the centre of x than of $x+1$, as in other families.

To see whether this was so, a search was made through the drawings of transections of those apices operated upon which had formed nearly median buds, and the relevant angles between the leaf centres were measured in any apices that seemed suitable. Unfortunately the number of suitable apices is much less than the number with nearly median buds. Only those apices are suitable in which the genetic spiral has continued after the operation and has not been excessively disturbed, since after a big disturbance any change in the position of $x+3$ may be due to some cause other than the median bud of x. Moreover, it seemed necessary to reject several spirals in which the angle x, $x+1$ was very large, approaching 180°, since within so large an angle the situation might not be quite comparable. Also many apices could not be used because they had suffered some secondary injury, or because the internodes had elongated too far for measuring angles, or because the necessary sections were missing or were not accurately enough transverse. However, there remain nine instances which seem free from objection, and the measurements of the relevant angles in them are recorded, with other details, in table 6.

Table 6 shows the angles, in various apices operated upon, to the centre of leaf $x+3$ from the centres of x and $x+1$ between which $x+3$ arises, x being the leaf with a nearly median bud. It also shows the angle x, $x+3$ as a decimal of the angle x, $x+1$, since this provides perhaps the simplest comparison with the normal. If the normal divergence angle in *Cucurbita* may be taken as approximately 153° on the basis of the writer's drawing (figure 1), then it follows that normally $\angle x, x+3 = 99°$, and $\angle x+1, x+3 = 54°$, as figure 19 makes clear. Thus $x+3$ is normally much closer to the centre of $x+1$ than of x, as a drawing of a normal apex confirms (see figure 1). The normal ratio $(\angle n, n+3)/(\angle n, n+1)$ is 0·647. In all the nine instances of table 6, of which two, in apices 172 and 179, are illustrated in figures 17 and 18, the corresponding ratio was considerably less than this, in six of them very much less. In the other three the ratio did exceed 0·5, but only slightly. The mean ratio was 0·454.

Table 6. Angles in degrees between leaf centres after formation of a nearly median bud in axil of leaf x

apex number	operation	identity of leaf x and position of its bud	$\angle x, x+1$	$\angle x, x+3$	$\angle x+1, x+3$	ratio $\frac{\angle x, x+3}{\angle x, x+1}$
77	I_1 confined kathodic	I_1, nearly median	160	86	74	0·537
101	I_1 confined kathodic	I_1, very subeccentric	155	58	97	0·374
102	I_1 confined kathodic	I_1, median	129	55	74	0·426
37	P_1 extirpated	I_1, nearly median	114	62	52	0·543
171	I_2 extirpated	I_3, nearly median	166	65	101	0·392
171	I_2 extirpated	I_4, median	124	54	70	0·435
172	I_2 extirpated	I_3, median	135	56	79	0·415
179	I_2 extirpated	I_4, nearly median	118	60	58	0·508
187	I_2 extirpated	I_4, nearly median	110	50	60	0·454
						mean 0·454

The evidence, therefore, from these results supports strongly the proposed explanation of the large divergence angles of the Cucurbitaceae, and at the same time the agreement of the changed positions of the $n+3$ leaves with the positions of the available spaces forms one more piece of evidence supporting a space-filling theory of phyllotaxis, for which the evidence, in the writer's belief, is already convincing.

12. Discussion and conclusions

With regard to the non-median axillary buds of the Cucurbitaceae, the chief point of interest revealed by the present results is that in *Cucurbita* the anodic positions of the buds depend on a bud-repelling influence of some kind, which travels upwards in a low spiral, or more strictly in a helix, following, at least roughly, the genetic spiral along which the leaves are formed. Yet the genetic leaf spiral itself does not depend on any spirally travelling influence, but is a secondary consequence of the way in which the leaves are placed by a different process, which in the writer's belief is a space-filling process. The bud-repelling influence must travel at some little distance below the surface, since in order to interrupt it a longitudinal cut in the apex needs to have a certain depth (§8). The repelling influence must also originate from a leaf, or perhaps from a leaf with its bud, for reasons given in §§3 and 7; but yet, when once it has started, it can continue past a node at which the whole upstanding part of a leaf rudiment has been removed or destroyed in its first plastochron, and at which no bud is formed at any time (§8). This continued movement is not, indeed, itself so surprising, since a similarly continued movement is, for example, the downward movement of the auxin in a shoot, which originates mainly in the young leaves near the top, but may then continue to the base of the plant and into the roots.

The puzzling question is how to account for the course which the bud-repelling influence follows; but a possible explanation is suggested by the observation that in *Cucurbita* two strands from the node of each leaf develop in the stem along a very flat helix and become inserted on the node of the next older leaf (§9). Reasons were indeed given for thinking it very unlikely that these strands have yet begun to conduct at the time when the axillary bud of their leaf above them is being determined in its anodic position; but there must earlier be some kind of physiological disposition set up in the stem which causes the strands to follow the course which they do follow in developing, and this same disposition in the stem may also guide the bud-repelling influence in an upward helix along the same course. It would be valuable to know whether anodic axillary buds of this kind are always associated with leaf traces that follow the course of the genetic leaf spiral.

It may be noted that the bud-repelling influence is not associated with any inhibition of growth, but rather with promotion of it. For the axillary buds of the *Cucurbita* apices are very precocious and large and the tendril, which is regularly the most precocious lateral member of the axillary bud, is also always its most anodic member and so the furthest repelled. In a rather similar way the auxin formed by the young leaves and travelling down a shoot both promotes the formation and growth of lateral roots, if the shoot is taken as a cutting, and also repels them since it often causes them to be formed mainly or entirely at the extreme base. But that example is simpler, since the auxin is stopped at the base of the cutting and accumulates there, whereas in the Cucurbitaceae the repelling influence is not stopped in the region of each axillary bud, where it acts, but continues still further. An explanation based on some hormone is indeed rather suggested by the progressive shift of growth in the anodic direction, shown first by the axillary bud as a whole and then by its first formed lateral member, the tendril, but the question of the nature of the bud-repelling influence must remain open until further studied. It needs also to be remembered that the buds are caused to grow out obliquely in the anodic direction, besides being repelled in that direction (see § 5).

The axillary buds of *Rhoeo discolor* also, a species with spirodistichous phyllotaxis, were found by M. Snow (1955, p. 85), to be eccentric and anodic, though only slightly so, about 5°. The effects of various operations on the eccentricities of certain buds in that species were reported, and the effects of one kind of operation, a radial split of I_1, supported very well the idea that a bud is made anodic by a repulsion from the next older leaf. But the operations were designed for another purpose, and the effects of some of them of other kinds on the bud eccentricities were difficult to interpret, so that the question was left open. It should perhaps be pointed out that in all these examples the terms 'repulsion' or 'displacement' are not used in their literal senses, since it is not the actual buds or bud rudiments, but the positions in which they will be formed that are repelled or displaced.

The functional value of the anodic positions of the buds of the Cucurbitaceae is a difficult question. One possibility is suggested by the progressive shift in the anodic direction first of the axillary bud and then of its earliest and most anodic lateral member, the tendril, which actually arises out of the main axis just beyond and slightly below the axillary bud to which it belongs. It may be that in this family the process of formation of tendrils depends on the physiological change leading to this anodic shift, so that if a species were to lose the anodic shift and form median buds, it would lose its tendrils also. It is in favour of this suggestion that in *Ecballium elaterium*, in which species the tendrils are replaced by leafy shoots, the axillary buds are less far anodic than in species with tendrils, while in *Acanthosicyos horrida*, in which the tendrils are replaced by thorns, the axillary buds are almost median (see figure 2, and Hagerup (1930, pp. 35 to 42 and figures 10 to 19)). Against the suggestion, however, is the fact that after the present operations on *Cucurbita* apices, several median and nearly median buds did have tendrils, one such bud being shown in figure 9. But the idea still seems worth considering.

Another possible explanation of the functional value of the anodic shift is suggested by the association of median buds with stunted leaves, reported in §10. May it be that without this shift the primordia of the leaves, which are large in many Cucurbitaceae, would risk being stunted by their own large and precocious axillary buds confining them too closely from above?

Finally the phyllotactic consequences, reported in §11, of the formation of median or nearly median axillary buds after various operations have provided very strong support for an explanation of the large divergence angles of the Cucurbitaceae based on space-filling principles. For they show clearly that the position in which a new leaf n, is determined depends on the extent to which the axillary bud of $n-3$ encroaches from the kathodic side on the space available for n between $n-3$ and $n-2$. These results would be difficult to explain on any theory of phyllotaxis based on physiological inhibitions or repulsions between leaves or leaf centres; for one would need to extend such a theory by supposing that in Cucurbitaceae leaf n can be physiologically repelled by the bud also of $n-3$, which seems rather unlikely and has not been postulated before in any of these theories. Also in order to explain how it was that n was repelled by the bud of $n-3$ when that bud was anodic, but not when it was median, one would need to postulate an extremely rapid falling off of the repulsion with distance. But on a space-filling theory none of these difficulties arise, since anything whatever which occupies or inactivates space on the surface of the leaf-forming zone of the apex will change the positions into which new leaves will be fitted. This includes any lateral member, however young, and any area already determined to form a lateral member, even before that member arises. The special difficulty presented by the ferns has been discussed previously (Snow, M. & Snow, R. 1952, pp. 565, 566).

I am much indebted to my wife for suggesting the phyllotactic problem of the Cucurbitaceae and its explanation, for drawing figure 5, plate 4, for help in preparing the other figures and for performing more than half of the last fourteen operations reported in §10.

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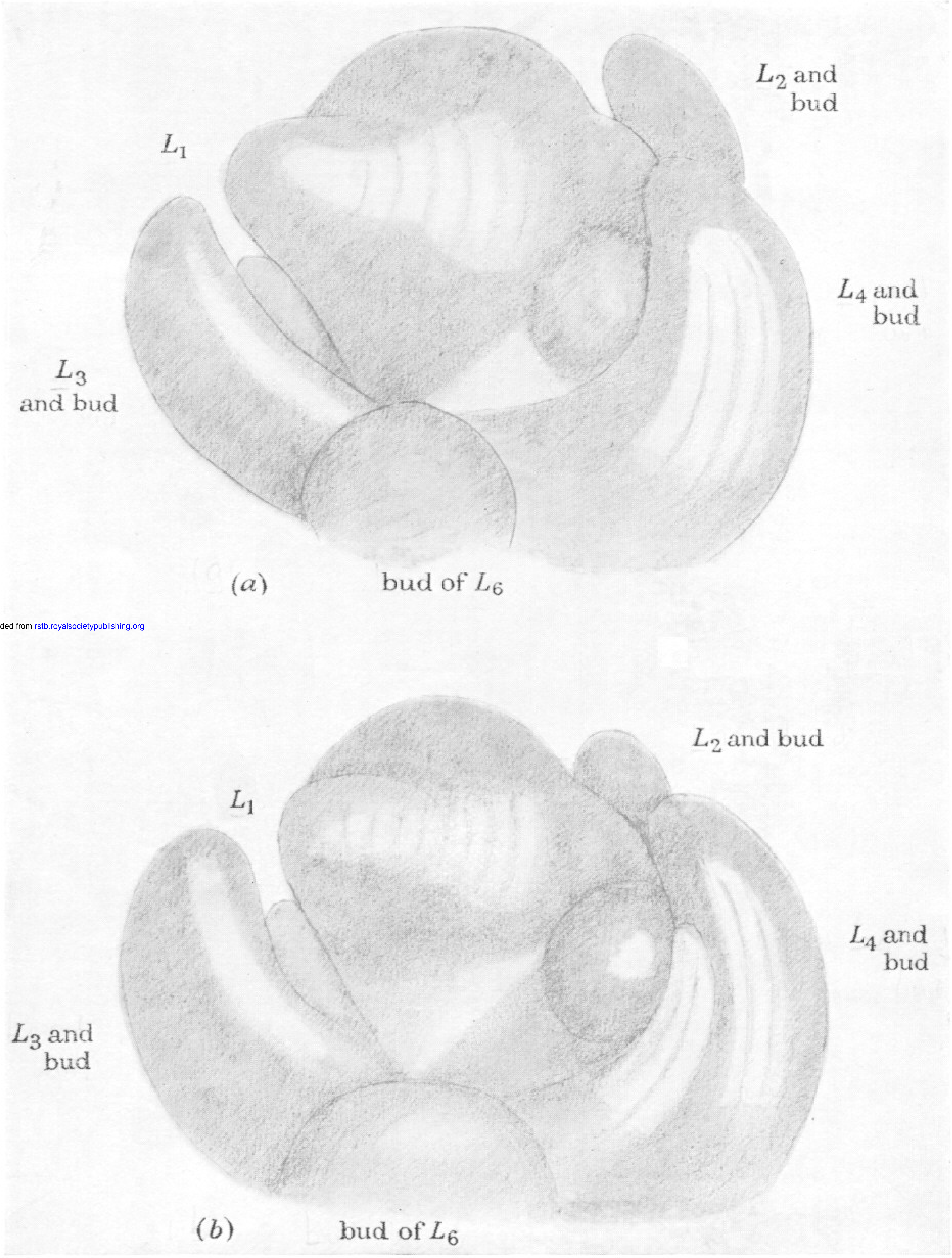

FIGURE 5, *a*, *b*. An apex of *Cucurbita pepo* seen in the solid from one side at late plastochron. *a*, seen from between leaves 1 and 2; *b*, seen more nearly from leaf 1, when one day older. The genetic spiral winds clock-wise and the leaves are numbered from the youngest downwards. The parallel lines across leaf 1 are boundaries between rows of cells.