Amer. J. Bot. 75(9): 1253–1261. 1988.

REGENERATION OF THE SUNFLOWER CAPITULUM AFTER CYLINDRICAL WOUNDING OF THE RECEPTACLE[1]

LUIS F. HERNANDEZ[2] AND JOHN H. PALMER

School of Botany, University of New South Wales, Sydney 2033, Australia

ABSTRACT

Using the young capitulum of *Helianthus annuus* L., a cylindrical plug of undifferentiated receptacle tissue, 1 mm in diameter, was isolated from lateral communication with the rest of the receptacle surface by a vertical circular wound cut, while retaining continuity with the subapical meristem. Within 24 hr, active cell division was induced at the inner and outer surfaces of the wound and in the receptacle epidermis bordering the wound edges, creating a rounded rim at the top of the wound. Within 3–6 days, floral initials, spaced 133–166 μm apart appeared on the flanks of both rims and later on the top of the plug and surrounding receptacle surface. The first formed initials developed into involucral bracts or ray florets and the later ones into disc florets which were organized into contact parastichies, the number of which did not conform with the Fibonacci series. The base of the plug developed into a stem-like structure completing the regeneration of a fully formed functional capitulum. This operation was demonstrated for two sunflower cultivars and occurred in both long and short daylengths.

WOUNDING MATURE SHOOT TISSUE can initiate cell division, redifferentiation and tissue development (Küster, 1916; Hardham and McCully, 1982; Warren Wilson and Grange, 1984; Warren Wilson and Warren Wilson, 1984). Apical wounding has commonly involved bisection of the apex. In the vegetative sunflower apex this results in the regeneration of two identical apices (Davis and Steeves, 1977) and the vegetative apices of other species respond similarly (Pilkington, 1929). Bisection of flowering apices of *Primula bulleyana* (Cusick, 1956), *Portulaca grandiflora* (Soetiarto and Ball, 1969) and *Aquilegia formosa* (Jensen, 1971) demonstrated a capacity for regeneration of floral organs in the wound vicinity, although organs already formed, or about to be formed, were not regenerated by wounding, indicating a progressive diminution in developmental potential in the determined flowering apex (Hicks and Sussex, 1971). Palmer and Marc (1982) induced the regeneration of all the floral organs of the sunflower capitulum, by pinprick wounding the uncommitted central region of the young receptacle.

Wound-induced organogenesis and tissue development are similar in that cell division and dedifferentiation are the first responses shown by the cells around the wound and appear to be prerequisites in the redifferentiation process (Warren Wilson and Grange, 1984). While the physiological basis for wound-induced cell division is unknown, applied IAA can induce cell division and tissue differentiation (Jacobs, 1952; Sussex, Clutter, and Goldsmith, 1972; McArthur and Steeves, 1972), although there is no evidence that increased hormone production is the initial biochemical step in the wounding response. The way in which mature cells can be induced to divide so that the new walls in adjacent cells are often in the same plane and precisely oriented parallel to the wound surface (Sinnott and Bloch, 1941), has led to the proposal that wound-induced changes in tissue stress provide the stimulus for initiating cell division and orienting the division plane (Kny, 1902; Lintilhac and Vesecky, 1981).

In the sunflower capitulum, the receptacle which bears the floral organs appears in floral stage 4 as a saucer-shaped structure with a prominent rim (Marc and Palmer, 1981). Wounding the receptacle surface in floral stages 4, 5, or 6 induced regeneration of all the floral organs, namely, involucral bracts, ray and disc florets, from the cells around the wound rim, without affecting the natural growth or development of the parent capitulum (Palmer and Marc, 1982). However the positions of the induced involucral bracts and ray florets were irregular. The wounding procedure has now been refined and used to induce the generation of a complete functional capitulum with normal morphology. The implications of this for

[1] Received for publication 20 July 1987; revision accepted 4 November 1987.

This research was completed while LFH was supported by a scholarship from the Consejo Nacional de Investigaciones Científicas y Técnicas, Argentina.

[2] Permanent address: Departamento de Agronomía, Universidad Nacional del Sur, 8000, Bahia Blanca, Argentina.

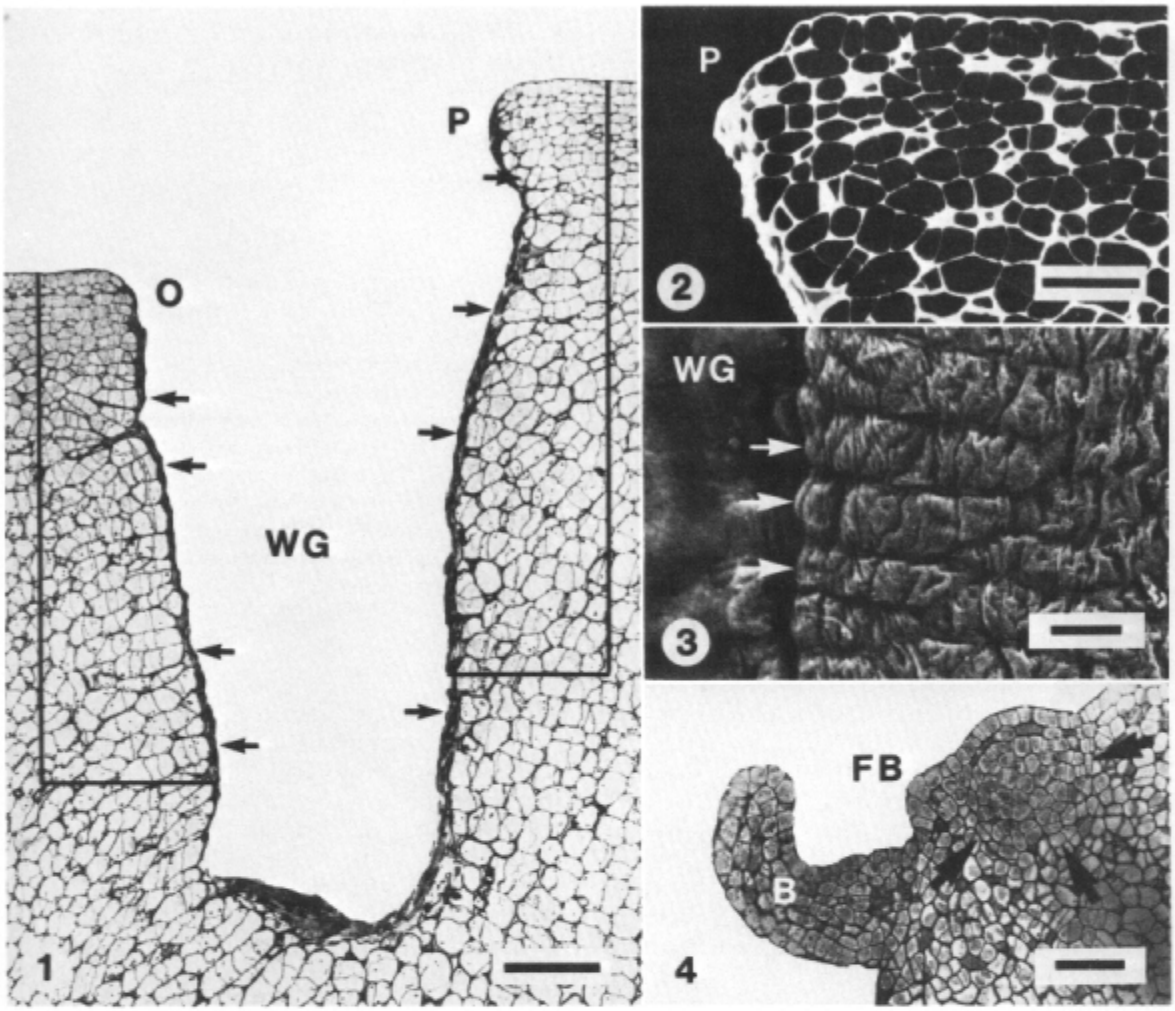

Fig. 1–4. Induced cell division and organ development following wounding of the receptacle in cv. Hysun 30 reared in an 18-hr photoperiod. **1.** Vertical radial section of the wound after 24 hr. Arrows show induced cell division with formation of new cell walls parallel to the wound surfaces. Outlined areas used for determination of cell division frequency (Table 1). Bar = 100 μm. **2.** Fluorescence micrograph of vertical section of the junction of the inner wall of the wound and the plug surface 36 hr after wounding showing formation of the plug rim by cell divisions parallel to the wound. Note absence of organ initials. Bar = 50 μm. **3.** Scanning electron micrograph of the edge of the plug 47 hr after wounding. Arrows showing radial files of 4–6 cells at the plug rim formed by cell division in the epidermis parallel to the wound. Bar = 10 μm. **4.** Vertical section of the plug 5 days after wounding to show an involucral bract initial on the flank of the wound rim and a floret initial (outlined by arrows) forming beneath the plug rim. Bar = 50 μm. **B** = involucral bract initial; **FB** = floret bract initial; **O** = outer rim; **P** = plug rim; **WG** = wound gap.

the control of organogenesis in the sunflower capitulum are discussed.

MATERIALS AND METHODS—*Plant culture*—Sunflower (*Helianthus annuus* L.) seedlings, cvs. Sunfola 68-2 and Hysun 30 were reared in 1-liter pots containing a sand/peat/vermiculite mixture (ratio 1:1:1) and grown in a controlled artificial environment at a constant air temperature of 28 C. Cool white fluorescent tubes and supplementary incandescent lighting provided a photon flux density of 550 μmol s^{-1} m^{-2} (400–700 nm) and an 18-hr long-day photoperiod (LD), for the first 13 days after sowing. On day 14, half of the plants of each cultivar were transferred to an 11-hr short-day photoperiod (SD) while retaining the same temperature and irradiation conditions. The remaining plants were kept in LD. The daily irradiation flux in SD was 61% of the LD value. The plants were regularly supplied with inorganic nutrient solution to maintain optimal growth. Capitulum growth and development was followed by sampling 4–6 plants in each treatment at 2–3 day intervals, dissecting the apical bud and measuring the diameter of the apex. A mean value for the stage of capitulum development for each sample was obtained by

grading each plant against the ten stage classification scale of Marc and Palmer (1981).

Wounding procedure—The saucer-shaped receptacle which bears the floral organs of the capitulum appears in floral stage 4 and persists until the end of floral stage 7. Apices at late floral stage 4 or floral stage 5 were used, since in these stages the receptacle surface was either undifferentiated (FS 4) or had produced only a few rows of floret initials (FS 5). It was also large enough to accommodate the wounding treatment and had just begun active expansion. The receptacle surface was exposed by partial removal of involucral bracts developing on the receptacle flanks. A cylindrical wound 50 μm wide and about 200 μm deep was then made at the receptacle center with a truncated, resharpened, flame-sterilized hypodermic needle.

This operation produced a cylindrical plug of receptacle tissue 1 mm in diameter which was isolated from lateral contact with the rest of the receptacle (Fig. 6) while retaining continuity with the subapical meristem. In some treatments the surrounding receptacle tissue, together with all the involucral bracts on the flanks of the capitulum, were cut away leaving only the plug. For the first 20 hr after treatment, the wound was protected from desiccation by covering the receptacle surface with moist tissue paper. Although no precautions were taken to maintain asepsis, infection of the wound was not observed. Growth of the treated receptacle and plug was followed by measuring their diameters at regular intervals, using a micrometer scale and dissecting microscope. Controls comprised capitula in which the receptacle was exposed by partial removal of the involucral bracts, but was otherwise untreated.

SEM and light microscopy—Some capitula were allowed to develop to maturity, while the remainder were harvested at regular intervals and prepared for light or scanning electron microscopy (SEM). After dissection, capitula were fixed under vacuum in ice-cold 8% glutaraldehyde in 0.025 M phosphate buffer (pH 6.8). For SEM, capitula were dehydrated in methoxyethanol followed by a graded acetone/ethanol series, then critical point dried in liquid carbon dioxide and coated with gold-palladium in a Polaron 5000 sputter-coater. Specimens were photographed using a Cambridge S4-10 scanning electron microscope at 20 kV. The SEM prints were used to calculate firstly, the distances between the centers of symmetry of the floret primordia arising naturally at the generative front (Palmer and Steer, 1985) and at the wound rims, and secondly the diameters

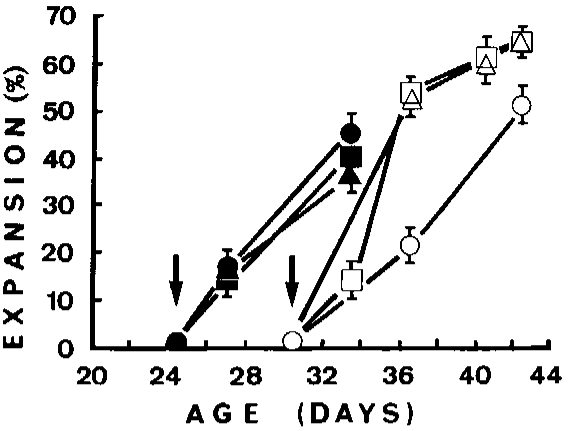

Fig. 5. Expansion growth of the wounded receptacle (■, □), plug (●, ○) and intact receptacle (▲, △) commencing either 24 days after sowing in SD or 30 days after sowing in LD. Data for cv. Hysun 30. Arrows indicate time of wounding. Closed symbols = 11-hr short day; open symbols = 18-hr long day; vertical bars = ±SD.

of the emerging floret primordia. Allowance was made for specimen shrinkage during critical point drying. Mean values for each capitulum were based on 15–25 values for floret spacing and 40–50 values for floret diameter. There were four replicates for each treatment.

For light microscopy, capitula were fixed as above and dehydrated in graded ethanol/water series, then embedded in LR White acrylic resin (The London Resin Co., London, UK). Vertical sections, 1–2 μm thick, were cut using a glass knife on a Reichert-Jung ultra microtome and stained for 2 min with 0.05% Toluidine Blue O in acetate buffer (pH 5.4). Median sections were photographed using a Leitz photomicroscope. The prints were used to assess cell division activity in the wounded receptacle by recording the frequency of newly formed cells with a thin dividing wall. Measurements were made 15 min, 10 and 24 hr after wounding.

Fluorescence microscopy—Sections were stained in 0.001% aq. solution of Fluorescent Brightener 28 (Sigma, USA) for 2 min, washed, dried, and mounted in immersion oil (n^d = 1.515) for observation in a Leitz Ortholux epifluorescence microscope.

RESULTS—Ten hr after wounding, some induced cell division activity was evident in undamaged cells around the plug wall and outer wall of the wound. Within 24 hr new cell divisions had become frequent in 4 or 5 layers of cells immediately beneath the wound surfaces (Fig. 1). The majority of division planes were oriented to give new cell walls parallel to the wound edges (Table 1). Similar cell division

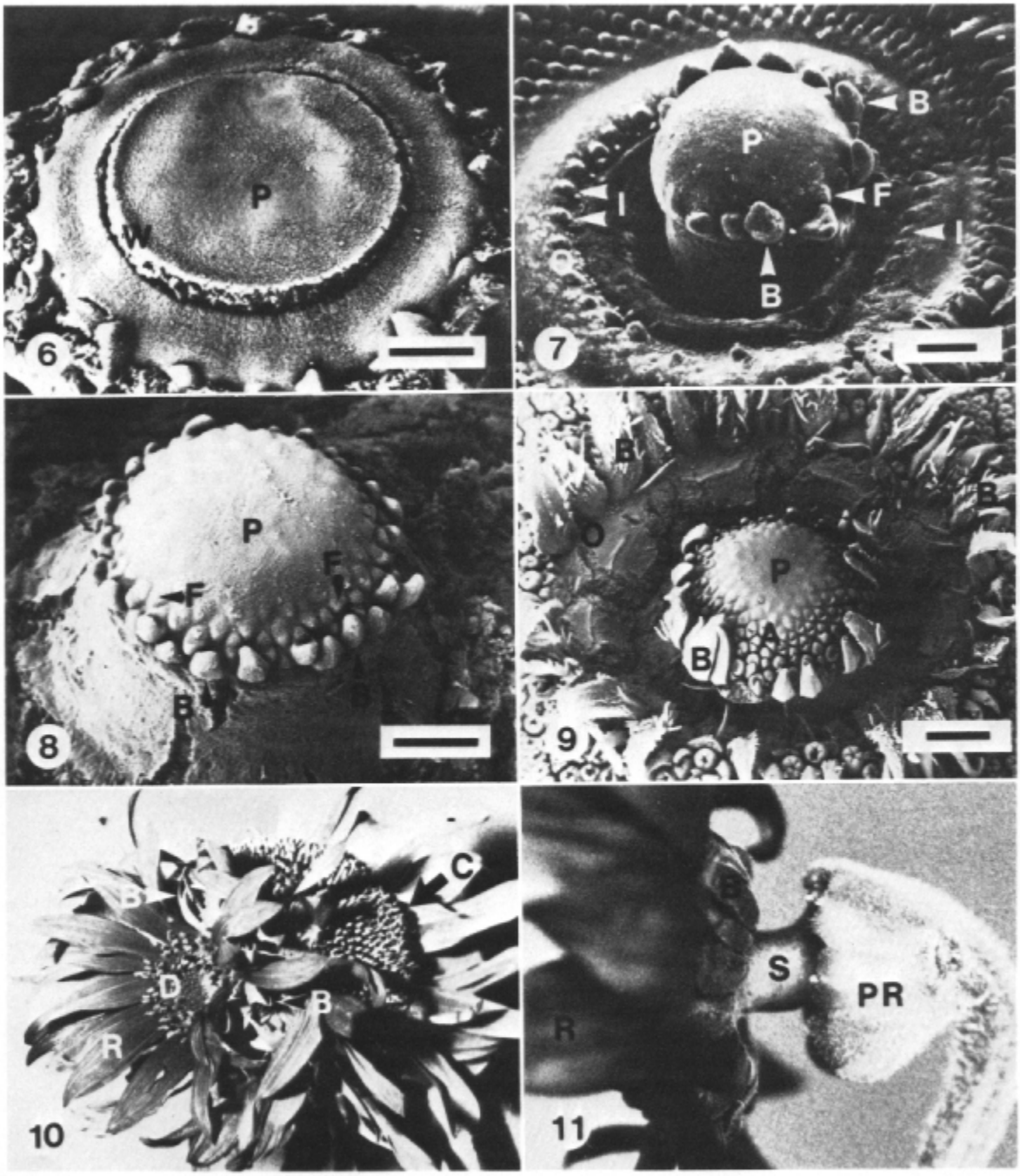

Fig. 6–11. 6–9. Scanning electron micrographs showing development of the central plug. **6.** Circular incision in the receptacle surface to form a plug of undifferentiated tissue; 15 min after wounding. Bar = 300 μm. **7.** Five days after wounding. Involucral bracts and ray floret initials have appeared around the rim of the plug and unidentified initials are regularly spaced around the outer rim of the wound. The plug has elongated above the receptacle surface and has developed a prominent dome. Bar = 300 μm. **8.** Six days after wounding and removal of parent receptacle. Plug with developing involucral bracts and ray and disc floret initials. Bar = 300 μm. **9.** Eight days after wounding. Involucral bracts at the plug rim and outer wound rim and emerging disc floret initials on the plug dome forming contact parastichies. Bar = 500 μm. 10–11. Wound-induced capitulum at anthesis. **10.** Involucral bracts, functional ray florets and disc florets at maturity. **11.** Side view of capitulum to show stem-like support formed from the plug base. **A** = parastichies of disc floret initials; **B** = involucral bract; **C** = parent capitulum; **D** = disc florets; **F** = floret initial; **I** = unidentified initials; **O** = outer rim; **P** = plug; **PR** = parent receptacle; **R** = ray floret; **S** = stem-like support; **W** = circular wound.

TABLE 1. *Frequency of newly formed cells with thin dividing wall parallel to plug wall or outer wound wall and located at the upper part of the wound. Data for Hysun 30 grown under long days*

Time after wounding (hr)	Number of cells mm^{-2} with new cell walls parallel (P) or not parallel (NP) to the wound surface			
	Plug wall		Outer wall	
	P	NP	P	NP
0.15	472	326	342	380
10.00	439	162	578	315
24.00*	1,372	256	2,173	246

* Measured areas are shown in Fig. 1.

activity also occurred in and beneath the receptacle epidermis adjacent to the junction with the wound surfaces, forming parallel files of cells (Fig. 2, 3) and creating an overhanging rim around the top of the plug and the outer wound wall about 48 hr after wounding. These rims provided the first sites for organ initials which were seen as regularly spaced mounds that suddenly appeared on the flank of the plug rim 3 to 6 days after wounding and on the flank of the outer rim, about one day later (Fig. 7). They were not distributed uniformly, but formed first in one region of each rim from where they spread in a left or right direction to encompass the entire circumference of the rim in one or two days, and then they rapidly developed into involucral bracts, while those that formed later gave rise to ray or disc florets (Fig. 9). Both types of initial appeared to originate from cells in subepidermal sites. These cells located in the 4th, 5th or 6th cell layers, divide to give daughter cells with periclinal cell walls, or walls radially organized around a focal point located in the 3rd or 4th layer of cells (Fig. 4). Subsequent cell divisions create an axis for the new initial which then appears as a bulge in the receptacle epidermis. In *Pisum*, leaf primordia are initiated on the vegetative apex in a similar way (Cunninghame and Lyndon, 1986). Since the appearance and subsequent development of the initials on the outer rim of the wound was similar to that previously described for pinprick wounding (Palmer and Marc, 1982), only the development of the plug initials are reported here.

On first sighting 5 to 7 days after wounding, the mean diameter of the initials on the plug rim ranged from 88.8 μm in SD to 110.3 μm in LD for Sunfola 68-2 and from 119.0 μm in SD to 103.3 μm in LD for Hysun 30. These values were comparable with those for initials arising naturally at the same time on the parent receptacle (Table 2). In both cultivars and photoperiods the mean spacing between florets forming around the plug rim was greater than the natural floret spacing; the increase ranged from 14 to 45% (Table 2). In both cultivars the development of the capitulum was accelerated in SD (Fig. 5), confirming the finding of Palmer and Steer (1985). Consequently, floral stages 4 and 5 occurred about 6 days sooner in SD than in LD, permitting the earlier wounding of SD plants. The expansion growth of the circular plug under SD and LD is shown in Fig. 5, for Hysun 30. Identical results were obtained for Sunfola 68-2. In SD the relative growth of the plug was the same as that for both the parent receptacle and the intact control receptacle, while in LD, relative expansion of the plug was consistently less than that of the parent and the control receptacle.

Plug development was essentially the same in both cultivars and was unaffected by daylength either directly or by the lower rate of expansion growth in LD. Floral organs appeared in the natural sequence. The first rows of initials arising on the flank and top of the rim, developed into involucral bracts. These were followed by a row of ray floret initials located at the plug perimeter. All the subsequent initials, arising on the top of the plug developed into disc florets. As previously re-

TABLE 2. *Spacing and diameter for initials formed naturally by the capitulum or generated at the wound rim of the plug*

Cultivar		Sunfola 68-2				Hysun 30			
Position	Day-length	Spacing (μm)	Variation (%)	Diameter (μm)	Variation (%)	Spacing (μm)	Variation (%)	Diameter (μm)	Variation (%)
Natural	SD[v]	116.4 a[x]		109.7 ac		113.6 a		100.6 a	
Plug rim	SD	132.8 b	+14	88.8 b	−23	165.6 b	+45	119.0 b	+18
Natural	LD[w]	112.6 a		100.2 bc		110.3 a		98.7 a	
Plug rim	LD	156.7 b	+39	110.3 a	+10	152.9 b	+38	103.3 ab	+4.7

[v] 11-hr short day. [w] 18-hr long day.

[x] Values with the same letter in the same column do not differ significantly ($P < 0.05$), as determined by Duncan's multiple range test.

Measurements were made 5 days after wounding in SD treatment and 7 days after wounding in LD treatment in both cultivars.

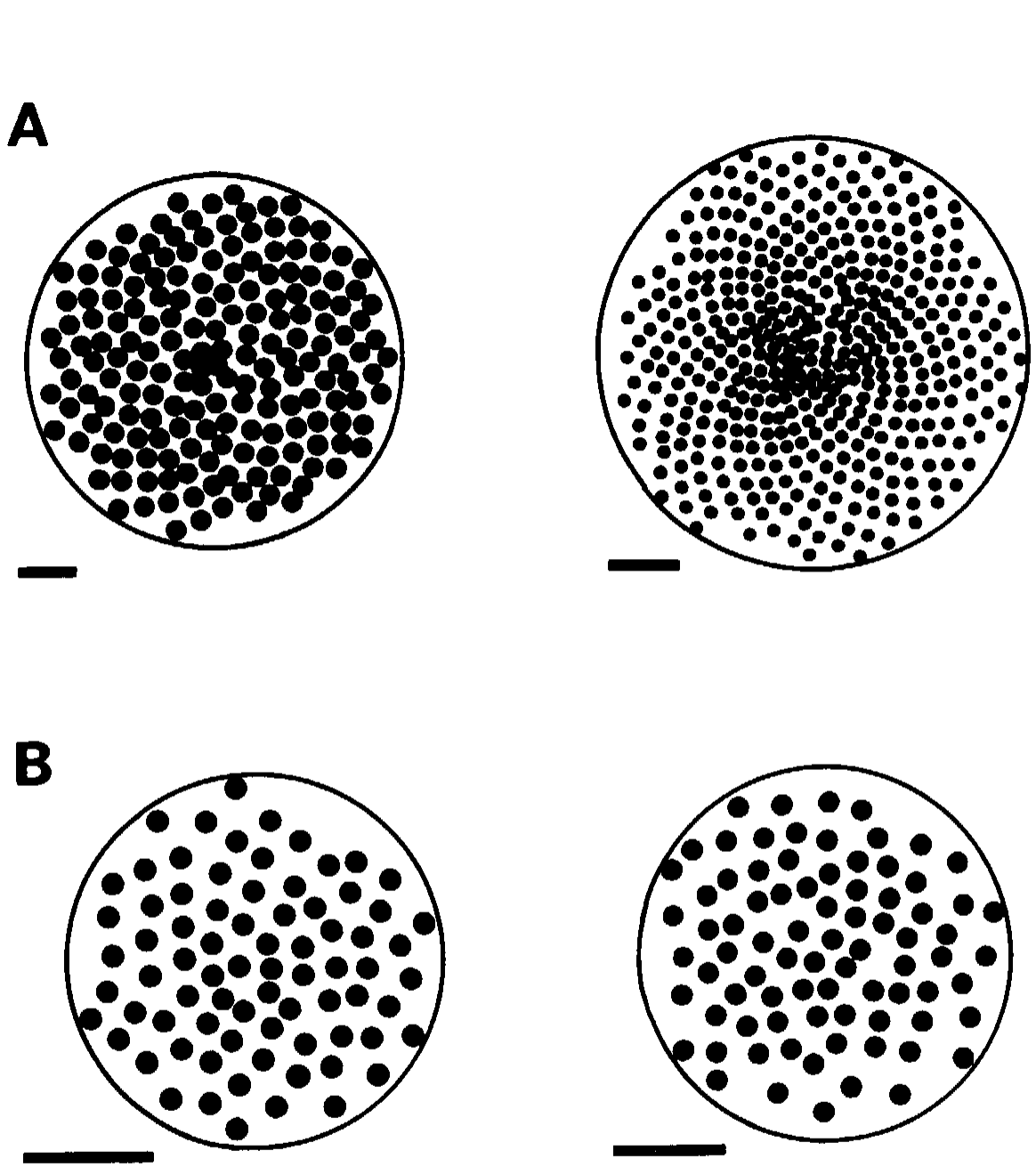

Fig. 12. Camera lucida drawings to show the pattern of disc floret parastichies at maturity, formed either naturally on the receptacle surface (**A**), or on the top of the plug (**B**), in long or short days. Note numbers of radial parastichies on the receptacle conform to the Fibonacci series (SD: 21/34; LD: 34/55), whereas the plug parastichies form nonradial irregular patterns not assignable to the Fibonacci series. Data for cv. Sunfola 68-2. Bar = 10 mm.

ported (Palmer and Marc, 1982), the function of the wound-induced floral organs was normal in every respect. The plug surface became domed as expansion growth continued and at maturity it appeared to be identical to a naturally formed receptacle (Fig. 10), except that it possessed fewer rows of involucral bracts and while the disc florets were organized into contact parastichies, these were irregular in length and shape and their number did not conform with the Fibonacci series (Fig. 12). The cylindrical base of the plug developed into a green, stem-like structure (Fig. 11), but the natural tissues of the stem were absent, there being no

epidermis, collenchyma or regularly arranged vascular tissue. Removing the surrounding receptacle and involucral bracts at the time of wounding, to leave only the plug had no effect upon either the development of the plug or the rate or magnitude of its subsequent growth (Fig. 8).

DISCUSSION—Circular wounding created a new flowering axis within the developing sunflower capitulum, leading to the regeneration of a secondary inflorescence. Reversion of flowers and inflorescences to vegetative growth is a well documented phenomenon and organized vegetative structures, known as "proliferations," have been induced in floral apices by varying daylength (Brulfert, 1965). By comparison the duplication of flowering apices in vivo is comparatively rare and confined to woody perennials, for example *Banksia* spp. (Carr, 1984).

Wounding characteristically induces cell responses in the immediate vicinity, so that the development of wound-induced floral organs from the tissues of the uncommitted receptacle can be regarded as a local event controlled by the plug and tissues adjacent to the outer wound rim. Wounding causes gene activation and an increase in messenger, ribosomal and transfer-RNA (Kahl and Wielgat, 1976), which may explain the rapid cell division and dedifferentiation around the wound during the first 1–2 days. These events appeared to be prerequisites for the developmental stages that followed in the next 2–3 days; namely the formation of the wound rims and the generation of spaced floral initials by the rims. The ability of the plug to produce the three types of floral organ, involucral bract, ray and disc floret, under the different conditions of short and long days and even after removal of much of the parent receptacle (Fig. 8), can be accounted for if the generation of floral initials is an innate function of receptacle tissue. The natural receptacle rim may be able to release this initial generating potential and in so doing give a centripetal direction to organ initiation. On this view the wound rims duplicate the action of the natural rim. The subsequent utilization of the entire receptacle and all of the plug for the production of disc floret initials may be explained if the disc floret generating process becomes self-perpetuating once it has commenced. The development of initials into either involucral bracts, ray or disc florets may reflect their relative positions on either the natural or wound rims (Palmer and Marc, 1982). Alternatively, the first-formed initials, may control the fate of later initials (Heslop-Harrison, 1964).

The sunflower capitulum is the culmination of determined shoot development and the organization of the disc florets in the capitulum is an example of high order Fibonacci phyllotaxis since the numbers of contact parastichies formed by the disc florets invariably conforms to the Fibonacci series; 34/55/89 being the most common numbers. In this context the plug disc florets are of interest because they are regularly spaced and form contact parastichies, apparently without external phyllotactic influence, either from the leaves on the stem, or from existing initials at the natural rim of the parent capitulum (Fig. 9). This is based on the assumption that the width and depth of the circular wound effectively isolates it from chemical diffusion fields (Roberts, 1987) which in theories of phyllotaxy are assumed to control the formation of leaves or floral initials and their positions on the shoot (Schwabe, 1984). However, while the plug disc florets formed parastichies (Fig. 9), the number of these did not conform to the Fibonacci series (Fig. 12), indicating that there needs to be surface continuity or close contact between developed and developing initials for Fibonacci phyllotaxis to influence the floret initial generating sites.

It is generally assumed that the developmental process which precisely positions the florets in a capitulum so that contact parastichies are created, is identical with spiral phyllotaxis which controls the positions of leaf primordia at the shoot apex. In the case of the sunflower capitulum this kind of extrapolation overlooks major morphological and developmental differences between the capitulum and the vegetative apical bud. Thus, the "bare" apex with its assumed role in leaf phyllotaxis is absent. The rate at which new primordia arise in the capitulum, as many as 6 hr^{-1} (Palmer and Steer, 1985), is much greater than in leaf phyllotaxis, requiring a control mechanism with a fast relocation time. For much of the floret generation period the width of the bare center of the receptacle, 1.5–2.0 mm, is the distance over which existing primordia would need to interact, if future primordia are to be positioned around the receptacle rim in the "first available space," in accordance with the rules of spiral phyllotaxis. Because this distance is relatively great in developmental terms and new initials may be appearing at intervals of only 10 min, chemical diffusion would seem to be too slow to qualify as the medium for communication between the developing disc floret initials. For these reasons chemical gradients appear to be inadequate as a basis for the operation of Fibonacci phyllotaxis in the sunflower capitulum. Green (1985, 1987) has

proposed that the interaction of physical reinforcement fields, generated by two adjacent leaves can result in the sectioning of the angle between them in the Fibonacci ratio 1.618:1 to locate a new leaf site and thereby maintain Fibonacci leaf phyllotaxis.

This "local" theory could explain the prevalence of Fibonacci phyllotaxis in the natural positioning of the floret initials in the sunflower capitulum as well as its absence from the plug, where the first initials arising at the wound rim are isolated from the influence of existing initials.

Green's theory depends upon the interaction of surface forces, but the floret initials in the sunflower capitulum appear to be propagated beneath the receptacle epidermis without disturbing the surface geometry until their development is relatively advanced (Palmer, unpublished data). If physical forces determine the positions of involucral bracts and floret sites at the natural rim of the receptacle, the outer wound rim and in the plug then these may be expressed as fields of pressure or stress within the tissue. Lintilhac and Vesecky (1984) have shown that an applied compressive stress of 0.075 bar can induce spatially organized coplanar cell divisions within pith derived tissue in vitro. However, it seems unlikely that naturally generated forces in expanding receptacle tissue could have similar magnitudes or foci in the range of experimental sites at which floral initials were produced, for example, the convex plug rim, the concave outer rim of the wound (Fig. 6), and the plug rim after removal of the residual receptacle tissue (Fig. 7). This leads to the conclusion that while the ability of the capitulum receptacle to spontaneously generate floral initials in a range of sites and conditions has been demonstrated, the mechanism controlling their initiation and location remains elusive.

LITERATURE CITED

BRULFERT, J. 1965. Physiologie de la mise a fleurs d'*Anagallis arvensis* L., ssp. *phoenicea* Scop., et du développement végétatif en retour des méristèmes floraux. Bull. Soc. Franç. Physiol. Vég. 11: 247–251.

CARR, D. J. 1984. Positional information and plant morphology. *In* P. Barlow and D. J. Carr [eds.], Positional controls in plant development, 349–374. Cambridge University Press, Cambridge.

CUNNINGHAME, M. E., AND R. F. LYNDON. 1986. The relationship between the distribution of periclinal cell divisions in the shoot apex and leaf initiation. Ann. Bot. 57: 737–746.

CUSICK, F. 1956. Studies of floral morphogenesis. I. Median bisections of flower primordia of *Primula bulleyana* Forrest. Trans. Roy. Soc. Edinburgh 63: 153–166.

DAVIS, E. L., AND T. A. STEEVES. 1977. Experimental studies on the shoot apex of *Helianthus annuus*: the effect of surgical bisection on quiescent cells in the apex. Canad. J. Bot. 55: 606–614.

GREEN, P. B. 1985. Surface of the shoot apex: a reinforcement-field theory for phyllotaxis. J. Cell Sci. Suppl. 2: 181–201.

———. 1987. Inheritance of pattern: analysis from phenotype to gene. Amer. Zool. 27: 657–673.

HARDHAM, A. R., AND M. E. MCCULLY. 1982. Reprogramming of cells following wounding in pea (*Pisum sativum* L.) roots. I. Cell division and differentiation of new vascular elements. Protoplasma 112: 143–151.

HESLOP-HARRISON, J. 1964. Sex expression in flowering plants. *In* Meristems and differentiation. Brookhaven Symposia in Biology. No. 16, 109–125.

HICKS, G. S., AND I. M. SUSSEX. 1971. Organ regeneration in sterile culture after median bisection of the flower primordia of *Nicotiana tabacum*. Bot. Gaz. 132: 350–363.

JACOBS, W. P. 1952. The role of auxin in the differentiation of xylem around a wound. Amer. J. Bot. 39: 301–309.

JENSEN, L. C. W. 1971. Experimental bisection of *Aquilegia* floral buds cultured in vitro. I. The effect on growth, primordia initiation, and apical regeneration. Canad. J. Bot. 49: 487–493.

KAHL, G., AND B. WIELGAT. 1976. Regulation of transcriptional activity in wounded potato tuber tissues. Physiol. Vég. 14: 725–738.

KNY, L. 1902. Ueber den Einfluss von Zug und Druck auf die Richtung der Scheidenwändein sich theilenden Pflanzenzellen. Jahrb. Wiss. Bot. 37: 55–98.

KÜSTER, E. 1916. Pathologische Pflanzenanatomie. Gustav Fischer, Jena.

LINTILHAC, P. M., AND T. B. VESECKY. 1981. Mechanical stress and cell wall orientation in plants. II. The application of controlled directional stress to growing plants; with a discussion on the nature of the wound reaction. Amer. J. Bot. 68: 1222–1230.

———, AND ———. 1984. Stress-induced alignment of division plane in plant tissues grown in vitro. Nature 307: 363–364.

MARC, J., AND J. H. PALMER. 1981. Photoperiodic sensitivity of inflorescence initiation and development in sunflower. Field Crops Res. 4: 155–164.

MCARTHUR, I. C. S., AND T. A. STEEVES. 1972. An experimental study of vascular differentiation in *Geum chiloense* Balbis. Bot. Gaz. 133: 276–287.

PALMER, J. H., AND J. MARC. 1982. Wound-induced initiation of involucral bracts and florets in the developing sunflower inflorescence. Pl. Cell Physiol. 23: 1401–1409.

———, AND B. T. STEER. 1985. The generative area as the site of floret initiation in the sunflower capitulum and its integration to predict floret number. Field Crops Res. 11: 1–12.

PILKINGTON, M. 1929. The regeneration of the stem apex. New Phytol. 28: 221–232.

ROBERTS, D. W. 1987. The chemical contact pressure model for phyllotaxis—application to phyllotaxis changes in seedlings and to anomalous phyllotaxis systems. J. Theor. Biol. 125: 141–161.

SCHWABE, W. W. 1984. Phyllotaxis. *In* P. Barlow and D. J. Carr [eds.], Positional controls in plant development, 403–440. Cambridge University Press, Cambridge.

SINNOTT, E. W., AND R. BLOCH. 1941. The relative position of cell walls in developing plant tissues. Amer. J. Bot. 28: 607–617.

SOETIARTO, S., AND E. BALL. 1969. Ontogenetical and experimental studies of the floral apex of *Portulaca grandiflora*. 2. Bisection of the meristem in successive stages. Canad. J. Bot. 47: 1067–1076.

SUSSEX, I. M., M. E. CLUTTER, AND M. H. M. GOLDSMITH. 1972. Wound recovery by pith cell redifferentiation: structural changes. Amer. J. Bot. 59: 797–804.

WARREN WILSON, J., AND R. I. GRANGE. 1984. Regeneration of tissues in wounded stems: a quantitative study. Ann. Bot. 53: 515–525.

———, AND P. M. WARREN WILSON. 1984. Control of tissue patterns in normal development and in regeneration. *In* P. Barlow and D. J. Carr [eds.], Positional controls in plant development, 225–280. Cambridge University Press, Cambridge.