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17. Under HPLC conditions, which separate BP *cis*- and *trans*-7,8-diols, we found that the metabolically formed 7,8-diol is a *trans* isomer. Mass spectral analysis indicated that the *trans*-7,8-diol formed enzymatically from BP under $^{18}O_2$ contained ^{18}O. Acid treatment of this ^{18}O-containing *trans*-7,8-diol yielded 7-^{18}OH-BP (97 percent) and 8-OH-BP (3 percent), which were separated by HPLC. An ^{18}O-containing *trans*-7,8-diol was also obtained by incubating racemic BP 7,8-epoxide in $^{18}OH_2$ with partially purified epoxide hydratase. On acid treatment of this ^{18}O-containing *trans*-7,8-diol, the ^{18}O was found exclusively in 8-OH-BP by mass spectral analysis. (S. K. Yang, P. P. Roller, H. V. Gelboin, in preparation).
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Phyllotaxis in *Xanthium* Shoots Altered by Gibberellic Acid

Abstract. *Gibberellic acid treatment of vegetative* Xanthium *shoots induced a change in phyllotaxis and almost doubled the rate of leaf production. Phyllotaxis in control plants displayed a 2,3 contact parastichy pattern; that of the treated plants could be approximated with a 3,5 pattern. Thus, the* Xanthium *apex switched to a new mode of growth and a higher order of phyllotactic leaf arrangement not seen in untreated plants. It may be inferred from these experiments that gibberellic acid plays a role in determining the site of leaf initiation.*

Single application of gibberellic acid (GA) produced a striking change in the shape of leaves and accelerated the rate of leaf initiation in vegetatively grown plants of *Xanthium pennsylvanicum* by a factor of 1.8 (*1*). The stimulation of leaf production coincided with enhancement of cell division and enlargement of the apical dome (*2*). We provide evidence here that the hormone treatment also leads to a striking change in phyllotaxis, from what might be designated as a 2,3 pattern of leaf arrangement to a 3,5 pattern, where the index numbers 2, 3, and 5 designate sets of contact parastichies (*3*). To our knowledge, this is a first report and data presentation indicating that prolonged treatment with GA causes marked changes in phyllotaxis.

Plants of *X. pennsylvanicum* were grown in a walk-in type growth chamber where the incandescent light of about 700 foot-candles (~ 7500 lu/m^2) cycled to give 18 hours of illumination per day, to prevent flowering. Other plants were grown in a greenhouse, supplemented with incandescent light to provide 16 hours of illumination. A lanolin paste containing GA (0.7 mg, 75 percent K salt, in 100 mg of paste per plant) was applied once to each of a number of randomly chosen plants of nearly uniform plastochron age, and comparable plants of similar plastochron age, not treated with the paste, were designated as controls. Leaves on each shoot were numbered in order of their appearance distally from the cotyledons, and records were kept of leaf numbers for each shoot. The morphological ages of plants and leaves were estimated by the plastochron index (*PI*) and the leaf plastochron index (*LPI*) (*4*). Apical buds were fixed overnight in a 3:1 mixture of ethanol and acetic acid at various times after treatment. After embedding in Tissue-Prep, 8-μm serial sections were cut transversely to the shoot axis, through the apical region, and stained with Feulgen reagent and fast green, or Delafield hematoxylin and fast green. Camera lucida drawings were made, at a magnification of 63×, of sections just below the apical meristem, chosen to include the youngest visible primordium.

Determination of leaf arrangement was carried out by methods of Richards (*3*) and Maksymowych and Erickson (*5*). The latter method is based on measurements of divergence angle and determination of the plastochron ratio (a), which involves measurements of the chord lengths between three successive leaf primordia in cross sections of apical shoots (Fig. 1). The relative plastochron rate of radial displacement is ln a. Values of the parameters α and ln a characterize various patterns with orthogonal and contact parastichies (Table 1). Details of the analysis are presented elsewhere (*5*). Richards's method (*3*) is based on measurements of radial distances of successive leaf primordia from the stem apex and estimation of the divergence angle. To designate a pattern of leaf arrangement, he proposed the use of the phyllotaxis index. This index is designated to have approximately the value of 1 for a 1,2 pattern of orthogonal parastichies, 2 for 2,3, 3 for 3,5, and so on, for an assumed divergence angle $\alpha = 137.51°$. Both methods are extensions and modifications of Van Iterson's (*6*) models of phyllotaxis.

The effect of the GA treatment on the organization of the shoot apex was similar in all of the treated plants. This is illustrated in Fig. 1. There is an increase in the number of leaf primordia on the treated shoot, and they appear to be more closely spaced than those of the control. The apparent larger size of the apical meristem in Fig. 1A is in part due to the level at which the section was cut,

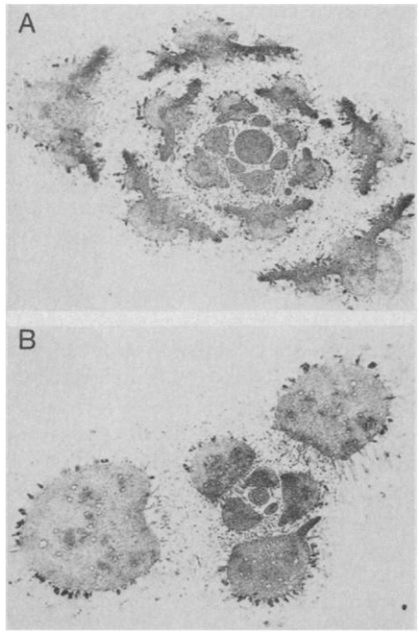

Fig. 1. (A) Cross section of a *Xanthium* shoot fixed 28 days after GA treatment. The phyllotaxis of this shoot fits a 3,5 parastichy pattern. (B) Cross section of a control apical shoot in which the arrangement of leaf primordia can be approximated to a 2,3 contact parastichy pattern. Note the increased number of leaf primordia in the treated shoot.

but it has about twice the volume of the control meristem (*2*).

Three series of plants have been analyzed. Eight plants grown in the growth chamber in 1975 were fixed 12, 20, 28, and 34 days after the treatment. These plants were of nearly uniform plastochron age at the time of treatment. Eight plants grown in the greenhouse in the summer of 1975 were processed 12, 20, 27, and 33 days after treatment. Twelve plants grown in the growth chamber in 1974 were collected 12, 16, 26, 31, 32, and 57 days after treatment. Each of these 28 plants was analyzed. The results for the 1975 experiments are summarized in Table 1. They allow a comparison of the effect of greenhouse versus growth chamber culture and of time of collection as well as the effect of GA treatment. There appears to be a clear effect of the treatment in reducing the plastochron ratio, but no effect on the divergence angle. The result of the 1974 experiment was the same. It is worth noting that plants grown for 57 days after treatment were essentially similar to others, appearing not to have reverted to their normal growth pattern. The variance analysis for data on ln a, the relative rate of radial displacement, revealed a highly significant difference between GA-treated and untreated plants, with $F_{1,3} = 34.2$. In summary, the striking change in phyllotaxis of the *Xanthium* shoot apex caused by the GA treatment is shown to consist of a highly significant reduction of the plastochron ratio, a, from a mean value of 1.35 in control plants to 1.19 in GA-treated plants, with no significant change in the divergence angle, α. There is no significant effect of time of collection on the phyllotactic parameters. This means (i) that the phyllotactic pattern is stable in the control plants and (ii) that the GA treatment has caused a change in this growth pattern and that the new pattern is also relatively stable.

For a direct comparison of the phyllotactic parameters estimated from the shoot apices with the parameters of the ideal models, mean values of ln a have been plotted against mean divergence angle in Fig. 2 for each of the apices of the 1975 experiment. The dashed lines in Fig. 2 represent the parameters of ideal diagrams for contiguous folioids, which

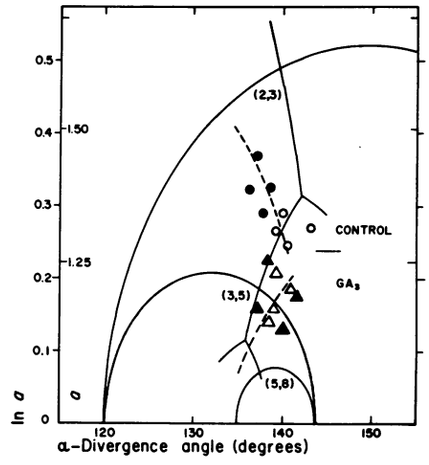

Fig. 2. Parameters of theoretical phyllotactic patterns of orthogonally intersecting parastichies (solid arcs), contiguous circles (branching solid lines), and contiguous folioids (dashed lines). In each case the parameters α and ln a are shown for 2,3 and 3,5 parastichy systems. Experimental points are for apices from control plants (circles) and treated plants (triangles) cultured in the growth chamber (solid symbols) and in the greenhouse (open symbols). Gibberellic acid treatment induced a change in phyllotaxis from a 2,3 kto 3,5 parastichy system.

are vertical projections into the plane of circles drawn on the surface of a circular cone (*6*). The data points are located in proximity to the dashed lines. We might therefore characterize the control apices as exhibiting 2,3 phyllotaxis and the GA apices as showing 3,5, recognizing that patterns of folioids best approximate these phyllotactic parameters. In a statistical sense, the differences between these parameters, as measured by the plastochron ratios, are highly significant. The phyllotaxis index based on Richards's method was 2.39 for all control shoots and 3.10 for GA-treated shoots. The first value approximates a 2,3 and the second an almost orthogonal 3,5 parastichy system. We conclude that the GA treatment induced a change in phyllotaxis from a 2,3 to a 3,5 pattern, not observed in untreated *Xanthium* plants.

Hormonal regulation of leaf arrangement may open new avenues of investigation and contribute to a better understanding of processes in the apical meristem, especially those which determine the loci of leaf primordium inception.

Table 1. Divergence angles (α), plastochron ratios (a), and relative rates of radical displacement (ln a) of control and GA-treated shoot apices of *Xanthium*. Mean values for each apex are given. The plastochron index of the plant at the time of treatment is PI_0 and at the time of excision of the apex is PI_f.

Days	Control					GA-treated				
	PI_0	PI_f	α (deg)	ln a	a	PI_0	PI_f	α (deg)	ln a	a
					Greenhouse					
12	16.5	20.9	135.5	0.324	1.383	16.6	21.1	141.7	0.178	1.195
20	16.8	23.6	137.7	0.293	1.340	16.9	29.2	138.4	0.226	1.254
28	15.9	23.9	137.2	0.371	1.449	17.0	34.9	137.2	0.159	1.173
34	16.0	27.2	138.7	0.327	1.387	16.2	36.1	139.6	0.127	1.135
					Growth chamber					
12	13.0	18.9	139.9	0.289	1.335	12.8	22.5	141.4	0.187	1.206
20	13.9	23.5	139.3	0.264	1.302	12.3	29.0	139.2	0.150	1.161
27	12.7	25.9	140.8	0.245	1.278	12.5	37.0	139.5	0.153	1.166
33	13.5	28.4	143.4	0.269	1.309	12.7	37.6	139.4	0.209	1.233
Mean			139.1	0.298	1.347			139.6	0.174	1.190
Standard error			±0.9	±0.014				±0.5	±0.012	

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17 December 1976

Nucleation on Photoexcited Molecules

Abstract. *On irradiation with light of suitable wavelength and intensity, certain organic compounds, even at very low concentrations, cause very efficient nucleation of supersaturated vapors. A mechanism is suggested to account for this phenomenon. Nuclei containing only a few photoexcited molecules are responsible for the nucleation.*

In a paper presenting results on the homogeneous nucleation of *n*-alkyl benzenes (*1*), Katz *et al.* reported that light of wavelength shorter than 420 nm caused marked increases in the observed rates of nucleation. We have now determined that the light-induced nucleation was probably due to the contamination of the alkyl benzenes by very small amounts of *o*-alkylbenzaldehydes (*2*). This report describes studies on the nucleation of liquid droplets from a supersaturated vapor phase caused by irradiation with light of suitable wavelength when this vapor phase contains trace amounts of certain organic substances. On the basis of careful observations, we have determined that (i) this nucleation is not a homogeneous nucleation process, (ii) the nucleation center is not charged, (iii) this nucleation is not a heterogeneous nucleation process in the ordinary sense, and (iv) the nucleation is not caused by dust particles or other artifacts. In short, this phenomenon cannot be explained by existing nucleation theories.

The thermal diffusion cloud chamber employed in this nucleation study has been developed and used in this laboratory for many years (*1*, *3*, *4*) for the study of the condensation of liquid drops from the vapor phase. The chamber is designed so that one-dimensional diffusion takes place through a gas of low molecular weight, usually helium, from a hotter to a cooler plate. The bottom plate is covered by a shallow liquid pool of a nucleating substance doped with a low concentration [typically 0.1 to 100 parts per million (ppm)] of the light-sensitive organic substance under investigation. The vapors evaporate from the surface of the liquid pool, diffuse upward, condense on the upper plate (which is slightly conical), flow to its edge, and return to the liquid pool by way of the chamber wall. The temperature, partial pressure, and thus the supersaturation of nucleating vapor inside the chamber are well defined and are obtained by solving the heat and mass flux equations (*1*, *3*). At sufficiently large supersaturations, homogeneous nucleation occurs; small droplets can be readily observed to form and fall to the lower plate. At slightly lower supersaturations, no droplet formation whatever is observed (*5*). However, upon irradiation with light of suitable wavelength and intensity, after a brief delay (1 to 30 seconds), nucleation commences and rapidly rises to a constant rate (*6*) even at supersaturations much too low to cause homogeneous nucleation (or even nucleation on ions). Once the light has been turned off, the rate of nucleation decreases to zero over a period of 20 to 300 seconds.

We have investigated the effects of supersaturation, the concentration of the light-sensitive organic substance, the wavelength and the intensity of the light, and the irradiation time on the rate of nucleation. A quartz-ringed thermal diffusion cloud chamber with a 1000-watt xenon light source and a 0.25-m monochromator was used (yielding a maximum light intensity for 10-nm resolution of about 10 watt m^{-2} at 400 nm). The cloud chamber was set to a steady state, with a light-insensitive nucleating vapor which is supersaturated. Carefully purified samples of heptane and nonane were used. For each of these substances we verified that, in the absence of the nucleating dopant, no light effect at any supersaturation, light wavelength, or light intensity was observable. Initial studies of the dopants *o*-tolualdehye, *p*-tolualdehyde, crotonaldehyde, benzoic acid, and *o*-nitrotoluene have shown that they all induce nucleation upon irradiation, with a relative nucleation efficiency which varies strongly from compound to compound and very strongly with wavelength.

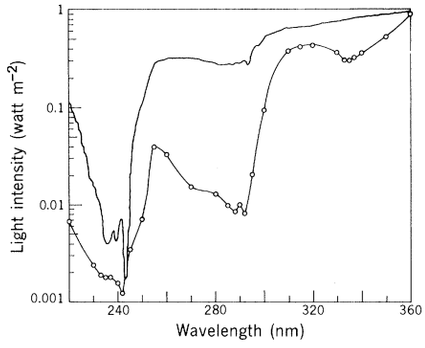

Fig. 1. Comparison of the ultraviolet absorption spectrum of pure *o*-tolualdehyde vapor (solid line) with the light intensity required for 100 ppm of *o*-tolualdehyde to cause nonane to exhibit a nucleation rate of two drops per square centimeter per second at a supersaturation $S = 7.249$ at 290.99 K (solid line with open circles).

Systematic studies have been carried out on *o*-tolualdehyde in nonane at one concentration (100 ppm by weight) (*7*). No light effect was observed at wavelengths longer than 400 nm. From 400 nm to about 300 nm there is a definite nucleating effect with a peak at 335 nm and a minimum at 320 nm. The light effect then increases very strongly, reaching two peaks in the range 285 to 295 nm. It decreases somewhat and then reaches another strong peak at about 242 nm (*8*). In principle, one would like to determine the rates of nucleation as a function of wavelength at constant intensity; however, these rates can be reliably determined over only a limited range (about 0.2 to 100 drops crossing 1 cm^{-2} sec^{-1}) because of fluctuation effects at low rates and depletion of the nucleating material (for example, the nonane) at high rates. Since, at constant light intensity, the change in the rate of nucleation as a function of wavelength is much wider than this range, we were forced to change the light intensity. Studies at several wavelengths showed approximately the same dependence of the rate of nucleation on intensity. Thus, a study of the intensity required to obtain the same rate of nucleation as a function of wavelength is roughly reciprocally equivalent to a study of the rate as a function of wavelength at constant intensity. Superposing (see Fig. 1) a vapor-phase ultraviolet absorption spectrum of pure *o*-tolualdehyde on a plot of wavelength as a function of the light intensity required to cause a given rate of nucleation, one sees significant correlation. The peaks in the region 285 to 295 nm correspond to a weak vapor-phase absorption in the pure aldehyde which is due to a n-π^* transition. The 242-nm peak corresponds to a much stronger π-π^* transition.

We have studied the steady-state nu-