8. All animals received water and food *ad libitum*. Rats were fed with a commercial stock diet (Purina Laboratory Chow); cats, with ground horsemeat or beef replaced once weekly with ground beef, pork, or sheep liver and once weekly with fish; and dogs with three parts Purina Dog Kibble to one part of ground meat, replaced once weekly with one part of liver.
9. Drs. Abner Wolf and David Cowen concurred in the pathological findings of these preliminary studies.
10. This report has dealt only with the findings related to the central nervous system. A description of the course of intoxication, the hematological changes, and the visceral lesions is in preparation. After this study was completed, our attention was drawn to a description of the pathological effects of 6-AN in young rats and mice (*11*).
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Influence of Gibberellin on Xanthium Flowering as Related to Number of Photoinductive Cycles

In 1956 Lang (*1*) reported that biennial *Hyoscyamus niger*, when treated with gibberellin, bolted and bloomed the first season without cold or long-day treatments and that the long-day, annual form of this species bloomed under short days when treated with gibberellin. As early as 1928 Kurosawa (*2*) had noted that rice plants infected with *Gibberella fujikuroi*, the fungus which produces gibberellin, flowered earlier than uninfected plants. In 1951 Mitchell, Skaggs and Anderson (*3*) reported that bean plants, when treated with gibberellin, bloomed earlier than the controls. Lang, however, was the first to demonstrate that gibberellin could substitute for long-day and low-temperature treatments in the induction of bolting and initiation of flowering.

Soon after his first papers appeared, Lang (*4*) and other investigators, including Bünsow and Harder (*5*), Lona (*6*), Lona and Bocchi (*7*), Marth, Audia, and Mitchell (*8*), and Wittwer and associates (*9*), reported on the initiation or promotion of reproductive development by gibberellin in various other species of plants. Of the 38 species studied, 17 have been long-day annuals, 9 biennials, 9 day-neutral annuals, and 3 short-day annuals. Gibberellin has proved to be a substitute for long-day and cold treatments for all species in the first two groups, except rye and *Perilla*. While induction of reproductive development was not involved in the day-neutral species, gibberellin hastened flowering in all species studied except pepper and geranium. No success has been reported, however, with the few short-day species studied. Lang (*4*) found that Biloxi soybeans and *Xanthium*, when treated with gibberellin, remained strictly vegetative under long photoperiods. Marth, Audia, and Mitchell (*8*) observed earlier blooming in gibberellin-treated saliva, but the plants were apparently already induced. Lang (*4*) reached the following conclusion: "It thus appears that application of gibberellin allows numerous plants to overcome cold and long day requirements in flower formation but that it does not substitute for any short day requirement."

The present study, which was initiated before the publication of Lang's results with soybeans and *Xanthium*, was designed to secure information on possible effects of gibberellin on the reproductive development of *Xanthium pennsylvanicum*, which is one of the best known short-day species. A preliminary experiment, in which both gibberellin-treated plants and controls were kept continuously under both long and short photoperiods, confirmed Lang's observations that gibberellin would not induce reproductive development in *Xanthium* under long photoperiods.

In the experiment reported here all plants were kept under long photoperiods (18 hour minimum) for 28 days following the date of planting (15 July 1957). At that time 30 plants were set aside as controls and 30 were treated with gibberellin in the form of a $10^{-3}M$ solution of gibberellic acid (*10*) in 0.25 percent Dreft, each plant receiving 1 ml of the solution as drops applied to the leaves with a pipette. The controls were similarly treated with the 0.25 percent Dreft solution alone. Five controls and five treated plants were continued under long days, and 25 of each were placed under 8-hour photoperiods in the greenhouse by means of an automatic short-day device (*11*). After two photoinductive cycles, five plants of each group were returned to long photoperiods. This process was repeated on alternate days, providing groups of plants which had received two, four, six, eight, ten, and twelve photoinductive cycles. The data reported here was taken 21 days after the beginning of the short-day treatments.

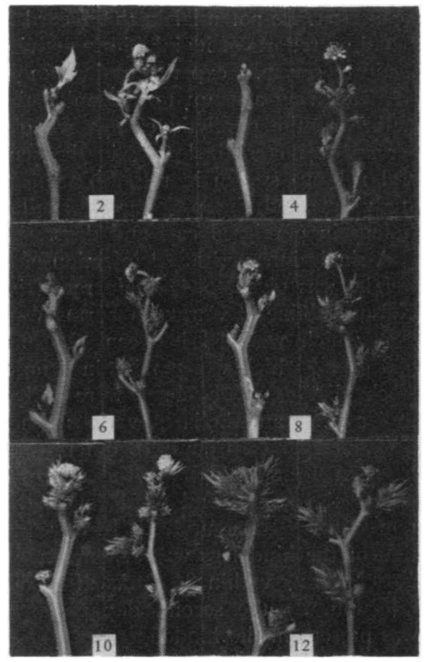

Fig. 1. Stem tips of representative *Xanthium* plants given 2 to 12 photoinductive cycles (figures between each pair of tips). The right-hand tip of each pair was from a plant treated with gibberellin, the left-hand tip from a control plant (× ½).

Table 1. Influence of gibberellin and photoperiod on the growth and reproductive development of *Xanthium pennsylvanicum* during a 3-week period after treatment.

Number of photo-inductive cycles	Mean stem growth (cm)		Number of nodes visible		Inflorescence buds formed	
	Controls	Gibberellin	Controls	Gibberellin	Controls	Gibberellin
0	27.0	54.7	5	8	–	–
2	25.6	62.3	5	8	–*	+†
4	17.0	55.1	4	8	–*	+
6	20.6	66.7	5	8	+	+
8	15.6	57.2	5	7	+	+
10	18.9	57.4	4	7	+	+
12	15.7	50.1	5	6	+	+

* Inflorescence primordia present in all five plants.
† Four plants with macroscopic inflorescence buds, one with inflorescence primordia.

The stem growth of the gibberellin-treated plants was much greater than that of the controls in all photoperiodic treatments (Table 1), the gibberellin overcoming the reduction in stem growth produced in the controls by four or more photoinductive cycles. All gibberellin-treated plants had more nodes than the controls, though this may have been due to accelerated internode elongation rather than to the formation of additional nodes. Stem diameter was not measured, but the gibberellin-treated plants all had stems which were obviously more slender than those of the controls (Fig. 1). At the end of the experiment all plants that had been retained continuously under long days were still strictly vegetative, while all plants that had been given six or more photoinductive cycles had well-developed macroscopic flower buds. The control plants that had been given two and four photoinductive cycles had developed only microscopic inflorescence primordia—stages 2 and 3 of Salisbury (*12*)—while all but one of the gibberellin-treated plants given two or four cycles had well-developed macroscopic flower buds, which were farther along than the controls which had received eight inductive cycles (Table 1; Fig. 1). Gibberellin also accelerated reproductive development in the six-, eight-, and ten-cycle groups, particularly with regard to pistillate inflorescences in the latter group.

These results substantiate Lang's conclusion that gibberellin does not substitute for any short-day requirement of *Xanthium* as far as *initiation* of reproductive development is concerned. Various investigators, including Mann (*13*), Naylor (*15*), and Salisbury (*12*), have shown that, while one photoinductive cycle is sufficient for the induction of *Xanthium,* additional cycles increase the rate of reproductive development. Gibberellin *can* substitute for such additional photoinductive cycles in promoting the reproductive development of induced *Xanthium* plants.

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References and Notes

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8 November 1957

Note on Absorption Spectra of Hill Reaction Oxidants

The selection of oxidants for the Hill reaction has been somewhat empirical heretofore. Mainly, complex inorganic ions and quinones have been found to be good Hill reaction oxidants. Hill and Whittingham (*1*) have listed the oxidation potentials of common oxidants. These range from – 0.44 for ferricyanide to 0 for ferrioxalate.

Yet surely the oxidation potential alone does not determine the suitability of an electron acceptor for the Hill reaction. For one thing, some oxidants of really negative potentials, which therefore should be easily reducible, are not reduced by illuminated chloroplasts. Examples of this kind are permanganate and periodate, neither of which participates in the Hill reaction at all (*2*).

It became of interest, therefore, to investigate some of the other characteristics which might influence the activity of various oxidants in the Hill reaction. For this reason, we have investigated the absorption spectra of various Hill reaction oxidants (*3*). The pertinent data are collected in Table 1. It is clear that Hill reaction oxidants absorb light near the blue peak of chlorophyll *a*, whereas the light absorption of those compounds which are inactive as Hill reaction oxidants is negligible around 420 to 430 mμ. This appears to be true despite the fact that the latter group includes compounds which are good oxidizing agents, such as permanganate and periodate. One test of this prediction was the finding that cobaltioxalate, which possesses the required absorption band at 420 mμ, acts as an electron acceptor in the Hill reaction (*4*).

Unfortunately, a real physical explanation of these facts cannot be given now. Neither the term levels of chlorophyll nor the term levels of complex inorganic ions are well enough characterized that an elucidation of their absorption spectra is possible at this time. The following explanation of the correlation which appears in Table 1 may, however, provide a reasonable approach to the problem.

It is well known that the Franck-Condon principle requires an overlap of energy levels of the reactants, because the actual time taken by electron transfer and by electronic excitation energy transfer is short compared with the time for ordinary atomic motion. If we consider that electronic excitation energy transfer from chlorophyll to the oxidant occurs in the Hill reaction, then the Franck-Condon principle requires chlorophyll and the Hill reaction oxidant to have overlapping energy levels. In the absence of precise knowledge of energy levels of chlorophyll and ordinary Hill reaction oxidants, which has already been referred to, the additional assumption has to be made that overlapping absorption bands in the absorption spectra of two substances also indicate overlapping energy levels. The data of Table 1 indicate that good Hill reac-

Table 1. Absorption peaks of chlorophyll *a* and of various hill reaction oxidants.

Hill reaction		Absorption		
Oxidant	Ref.	mμ	Log ε	Ref.
		Chlorophyll a		
		250	~ 4.43	*10*
		325*	4.41	*11*
		375*	4.69	*11*
		427.5*	5.08	*12*
		660*	4.96	*12*
		Ferricyanide		
Yes	*13*	260	3.28	*14*
		303	3.36	*14, 15*
		420	3.04	*15*
		p-*Benzoquinone*		
Yes	*16*	240	4.7	*17*
		300	2.5	*17*
		425	1.4	*17*
		Cobaltioxalate		
Yes	*4*	245	4.33	*18*
		420	2.34	*18*
		596	2.22	*18*
		1,2-*Naphthoquinone*		
Yes	*19*	250	4.35	*20*
		340	3.40	*20*
		400–500	3.4–2.0	*20*
		420	~ 3.0†	*20*
		1,4-*Naphthoquinone*		
Yes	*19*	250	4.4	*21*
		335	3.5	*21*
		400	1.7	*21*
		425	1.6	*21*
		Phenol-indophenol		
Yes	*2*	420	3.0†	*22*
		Ferrioxalate		
Yes	*23*	420	1.4†	*24*
		Chromate		
Yes	*2, 25*	280	3.58	*26*
		380	3.66	*26*
		420	2.7†	*4*
		Vanadate		
No	*2*	310–370	cutoff	*27*
		Thionine		
No	*2*	240	4.08	*28*
		285	4.64	*28*
		585	4.60	*29*
		Permanganate		
No	*2*	310	3.23	*30*
		525	3.35	*30*
		545	3.33	*30*
		Nitrate		
No	*2*	200	4.1	*31*
		300	2.53	*27*
		Cobalticyanide		
No	*4*	200	> 4	*32*
		259	2.15	*32*
		311	2.2	*32*
		Tetrathionate		
No	*2*	216	4.0	*33*
		Hypochlorite		
No.	*2*	290	2.54	*34*
		Periodate		
No	*2*	222.5	4.0	*35*
		Methylene blue		
Yes	*36*	250	4.30	*28*
No	*2*	290	4.65	*28*
		615	4.57	*29*
		662	4.83	*29*

* In ethyl ether. † Continuous absorption, not a maximum.