Physiological Observation of the Gibberellin Effects on the Development and Growth of Plants

by

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I. Introduction

Various reports have accumulated with regard to the physiological and morphological effects of gibberellin on plants, as reviewed by STOWE and YAMAKI (60), STODOLA (59), CHOUARD (15), PHINNEY and WEST (49), STUART and CATHEY (61), and others. BRIAN and many other workers (1, 3, 21, 27, 29, 58, 62, 67) reported that the extraordinary elongation of stem as well as leaf sheath by gibberellin treatment is due to the increase of cell length. While, according to many other workers the elongation of stem is induced by activated cell divisions of internodal cells (31, 37, 52, 56). FEUCHT and WATSON (17), and GREULACH and HAESLOOP (18) found that not only cell length but also cell number is increased by gibberellin. Later, SACHS and LANG (52, 53), and SACHS *et al.* (54) noted that the cell number in the tissue below the apical meristem is increased by gibberellin. It has also been found by many workers that the vernalization requirements were replaced and the flowering was induced by gibberellin treatment in some biennials or perennials (12, 13, 32, 37, 69). There are many reports which revealed that gibberellin could replace the chilling but not the short day requirement of long day plants (19, 47, 77).

In his previous papers (45, 46) the present author has reported that the flowering in *Erigeron* is stimulated by gibberellin, and that the extraordinary stem elongation in *Pharbitis* by gibberellin is mainly caused by the increase in cell number and is partly caused by the increase of cell length. The present paper deals chiefly with various effects of gibberellin on plants of different life forms.

II. Materials and Methods

"Kidachi", a dwarf strain, and "Violet", a tall strain of *Pharbitis nil* CHOIS.[1], "Tamanishiki", a dwarf strain of *Oryza sativa* L.[2], and a wild growing biennial plant, *Erigeron annuus* L. have been used in the present studies.

The methods of gibberellin[3] treatment will be described in respective paragraphs. The continuous illumination were made by natural light supplemented by additional illumunation from incandescent lamps at night, and the short day

1) The *Pharbitis* strains are the same which were used by HIRONO *et al.* (25) for the gibberellin test, and also by IMAMURA and TAKIMOTO and others (26, 63, 64) for the photoperiodic experiments.

2) A strain of *Oryza* used by OGAWA for the studies of gibberellin effects (44).

3) The used gibberellin was the product of the Kyowa Fermentation Industry Co. Ltd. Tokyo and contained gibberellin A_3 in 96.6 per cent.

conditions were made by covering the material plants by wooden shades from 5:00 pm to 9:00 am, thus making the 8 hour day and 16 hour night lengths.

For the histological observations in *Pharbitis*, the stem of a plant fixed in 70 per cent alcohol was handsectioned, then the sections were stained with safranin or treated with phloroglucin and HCl. In *Oryza*, the leaves of a plant fixed in alcohol were made clear with "*eau de Javelle*".

III. Experimental results

1. *Pharbitis nil* Chois.

The seeds germinated in 24 hours when they were treated with conc. H_2SO_4 for 45 minutes, and washed with tap water, while otherwise they took several days for germination. The germinating seeds were planted in soil filled in a wooden flat at intervals of 5 cm. The cotyledons expanded to their full size after two days. Then 0.02 cc of a 1000 ppm gibberellin solution was given to the plumule of the seedling two times at 24 hour intervals. The same volume of distilled water was similarly given for the control.

A. Elongation of stem

The lengths of hypocotyl, epicotyl and cotyledonary petiole in both dwarf and tall strains of *Pharbitis* were measured after seven days from the first treatment. In this experiment the plants were treated two times with a 1, 10, 100 or 1000 ppm solution. The results are given in Figure 1. The response to gibberellin differed strikingly between two strains ; in "Kidachi", the lengths of hypocotyl, cotyledonary petiole and epicotyl of the treated plants with a 1000 ppm solution were found to be 2.4, 1.6 and 9.0 times as long as those of the control plants. In "Violet", the lengths of those parts were 1.1, 1.2 and 2.4 times as long as those of the controls. The most sensitive organ was the epicotyl, which showed increases in length proportional to the gibberellin concentrations applied. The epicotyl of the dwarf plant treated with a 1000 ppm gibberellin solution became nearly as long as the normal tall one on the seventh day. In Figure 2, epicotyl length is plotted logarithmically against time from four to twelve days after the first treatment. The lengths of hypocotyl, the first and second internodes obtained from another experiment are also given in Table 4.

B. Elongation of internode

In Figure 3, the lengths of respective internodes are plotted logarithmically

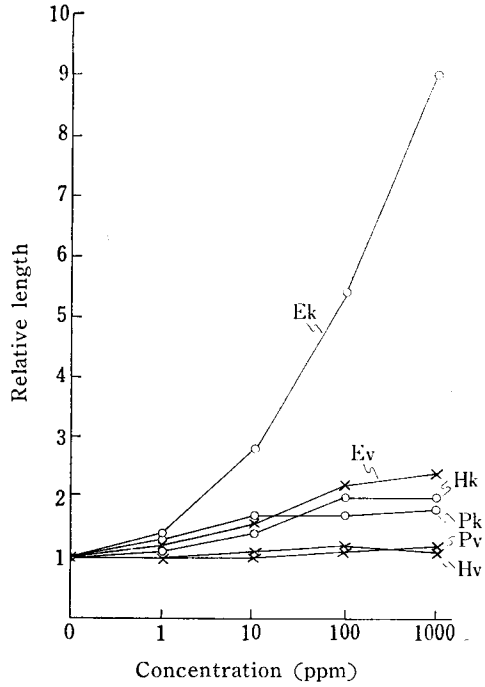


Fig. 1. Relative length of hypocotyl, epicotyl and cotyledonary petiole of the *Pharbitis* plants treated with gibberellin as compared with those of the untreated controls. Hk, Ek and Pk are hypocotyl, epicotyl and cotyledonary petiole of "Kidachi", a dwarf strain, respectively. Hv, Ev and Pv are the same of "Violet", a tall strain. Sown on 12 July, treated on 14 and 16 July, and measured on 21 July. Lengths of the control plants of "Kidachi", hypocotyl: 46. 1 mm, epicotyl : 19. 8 mm and cotyledonary petiole: 40. 5mm. Lengths of the control plants of "Violet", hypocotyl: 111. 5mm, epicotyl: 102. 0 mm, and cotyledonary petiole: 50. 8 mm.

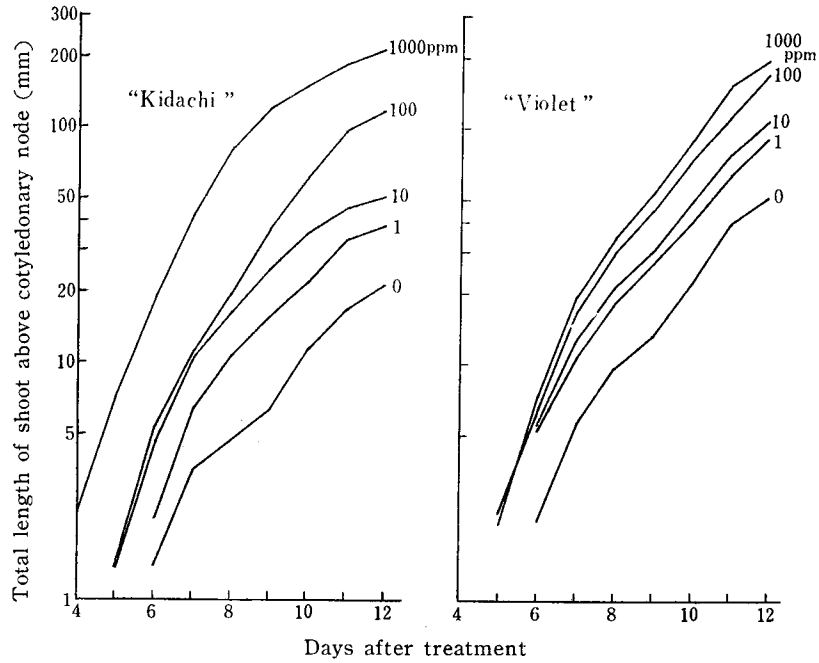


Fig. 2. Shoot lengths of both dwarf ("Kidachi") and tall ("Violet") strains of *Pharbitis nil* plotted logarithmically against time. The plants were given 0. 02 cc of respective gibberellin solutions.

against time from four to twelve days after the first treatment. The results show that in "Kidachi", the increase in stem length caused by gibberellin is mainly due to the pronounced elongation of respective internodes and somewhat to the increase in the number of internodes. In "Violet", any clear tendency as such was not observed. The acceleration of stem elongation by gibberellin treatment was more pronounced in the dwarf strain than in the tall one. The second internode elongated most pronouncedly.

In another experiment, the first, second and third internodes were marked at a distance of 5 mm with Indian ink on the fourth, seventh and tenth day, respectively, after gibberellin treatment. Stem septums were measured two days after marking. Table 1 shows the results. The growth ratio of the final to the initial length was greater in upper portions than in lower ones in all internodes. These results indicate that the elongation in the upper portions in every internode was more striking than in the lower. Observations in the untreated tall plant are given

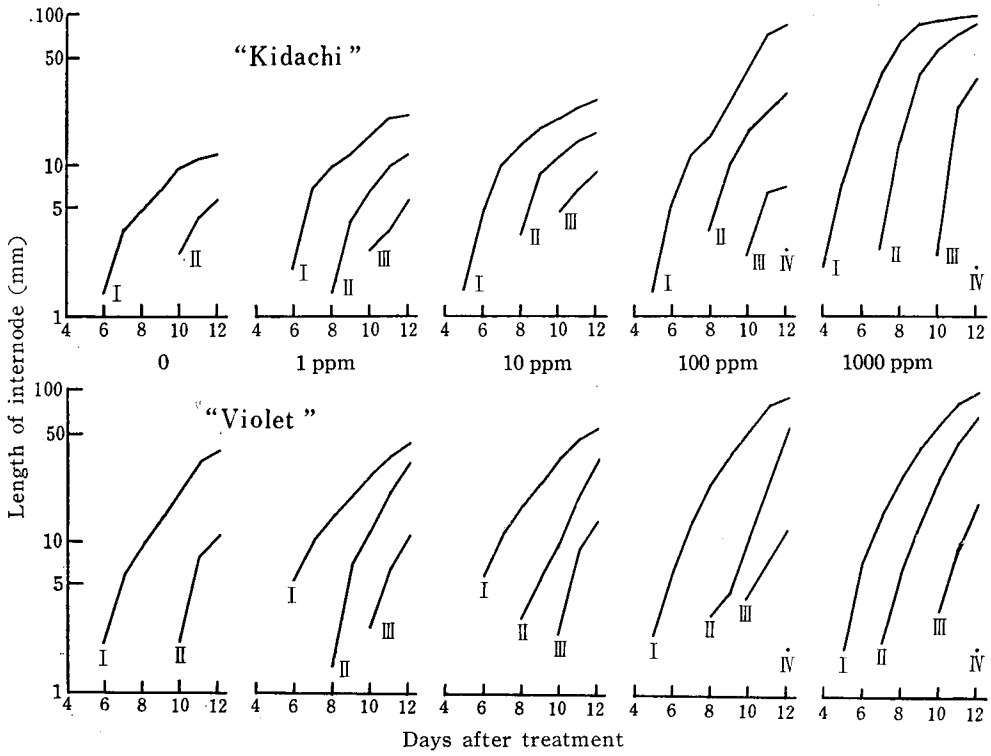


Fig. 3. Lengths of internodes of *Pharbitis nil*, dwarf ("Kidachi) and tall ("Violet") strain, plotted logarithmically against time. Serial roman figures show the order of internodes numbered from base to top.

Table 1.
Elongation of septums in internodes of the treated dwarf plant* of *Pharbitis nil* and cell length in respective septums.

	No. of Septum**	1	2	3	4	5	6	7	8
1st internode	Initial length (mm)	5.0	5.0	5.0	5.0	5.0	4.0		
	Final length (mm)	8.4	10.1	12.8	15.7	17.8	19.3		
	Ratio of final to initial length	1.7	2.0	2.6	3.1	3.6	4.8		
	Cell length (μ)	143.3	190.4	194.5	199.2	188.2	125.2		
2nd internode	Initial length (mm)	5.0	5.0	5.0	5.0	5.0	5.0	3.0	
	Final length (mm)	7.2	6.4	7.6	7.3	8.5	9.1	10.6	
	Ratio of final to initial length	1.4	1.3	1.5	1.5	1.7	1.8	3.5	
	Cell length (μ)	103.1	106.2	113.3	118.1	96.4	73.2	60.3	
3rd internode	Initial length (mm)	5.0	5.0	5.0	5.0	5.0	5.0	5.0	4.0
	Final length (mm)	6.5	7.4	7.6	9.3	11.2	12.7	17.4	22.3
	Ratio of final to initial length	1.3	1.5	1.5	1.9	2.2	2.5	3.5	5.6
	Cell length (μ)	78.6	94.1	108.9	124.8	112.2	101.6	89.7	59.9

* "Kidachi".

** Septums were numbered serially from base to top. Plants were treated with 0.02cc of a 1000 ppm gibberellin solution. The first, second and third internodes were marked on the fourth, seventh day after treatment, respectively, and measured after two days.

Table 2.
Elongation of septums in internodes of tall plant of *Pharbitis nil.* *

	Septum**	1	2	3	4	5	6	7	8
1st internode	Initial length (mm)	5.0	5.0	5.0	5.0	5.0	3.0		
	Final length (mm)	5.5	5.5	6.1	6.3	8.1	4.6		
	Ratio of final to initial length	1.1	1.1	1.2	1.3	1.6	1.5		
2nd internode	Initial length (mm)	5.0	5.0	5.0	5.0	5.0	5.0	3.0	
	Final length (mm)	5.0	5.4	6.3	7.8	8.3	8.9	5.2	
	Ratio of final to initial length	1.0	1.1	1.3	1.6	1.6	1.8	1.0	
3rd internode	Initial length (mm)	5.0	5.0	5.0	5.0	5.0	5.0	5.0	4.0
	Final length (mm)	5.7	6.1	6.9	7.3	7.9	9.0	10.8	8.4
	Ratio of final to initial length	1.1	1.2	1.4	1.5	1.6	1.8	2.2	2.1

* "Violet".

** Septums were numbered serially from base to top. The first, second and third internodes were marked on the fifteenth, twentyth and twenty-fifth day after sowing, respectively, and measured after two days.

in Table 2. The distribution of elongation in an internode of the dwarf plant were the same as those of the treated tall plant.

C. Differentiation of leaf primordia

A question arose as to whether the differentiation of leaf primordia at the terminal bud was accelerated by application of gibberellin. Consequently, 0.02 cc of a 100 ppm solution was applied to a two-day old seedling and on various days after the treatment, the leaf primordia in the terminal buds were counted under a dissection microscope. On the day of the treatment, there were three leaf primordia in all individuals ; on the second day, the treated and control plants initiated five and four leaf primordia, respectively. The differences in the number of the leaf primordia between the treated and control plants increased with the lapse of time. After eight days 10.6 leaf primordia in the treated and 7.5 in the control plants were observed, as shown in Figure 4.

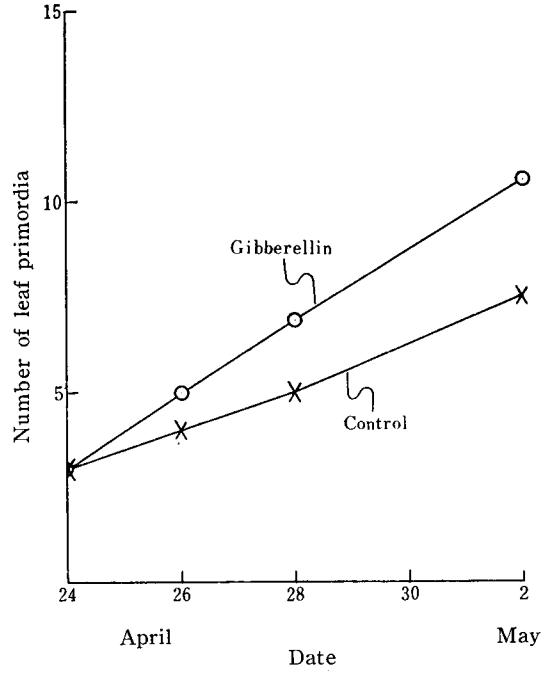


Fig. 4. Effect of gibberellin on the differentiation of leaf primordia in the shoot apex of *Pharbitis* seedling ("Kidachi").

D. Leaf development

When the young seedlings of *Pharbitis* were treated, the leaves became some-

what pale in color and expanded more rapidly than those of the control plants.

The lengths of lamina of the treated and control plants are plotted logarithmically against time in Figure 5. It is clearly shown that the growth of the leaves of the treated dwarf plants was very rapid, and even exceeded that of the treated tall plants.

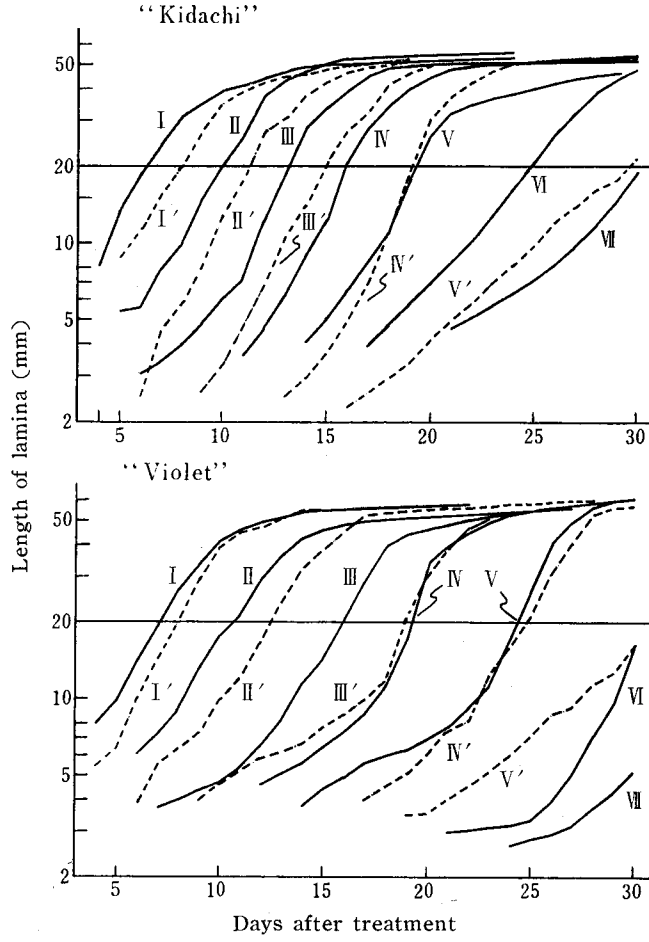


Fig. 5. Lengths of successive leaves (lamina only) in *Pharbitis nil* plotted logarithmically against time. Each growth curve applied to a single leaf. Leaves of the plants were numbered progressively from base to top and the respective positions were designated by roman numerals.
-------- : Control —— : Gibberellin

E. Plastochron age

The physiological age of a plant is indicated rationally by the plastochron

index (P. I.), proposed by ERICKSON and MICHELINI (12) as follows :

$$P.I. = n + \frac{\log L_n - \log k}{\log L_n - \log L_{n+1}} \qquad \cdots\cdots\cdots\cdots(1)$$

where n represents the serial number of a lamina longer than the arbitrary reference length, k, and $\log L_n$ or $\log L_{n+1}$ is the logarithm of the length of the nth or (n+1) th lamina which are respectively longer or shorter than the length, k, at a given time. In the present experiment, 20 mm was used for the reference length of a lamina, because the lamina was unfolding and growing exponentially when it attained this length. The plastochron indexes are given in Table 3.

Table 3.
Plastochron index in *Pharbitis nil.*

Days after 1st treatment	"Kidachi"		"Violet"	
	Control	Gibberellin	Control	Gibberellin
9	1. 26	1. 69	1. 28	1. 56
12	2. 22	2. 55	1. 83*	2. 26
15	3. 03	3. 54	2. 41*	2. 67
18	3. 60	4. 49	2. 69*	3. 54
21	4. 26	5. 42	3. 53*	4. 40*
24	—	5. 83*	—	4. 82*
27	—	6. 95*	—	5. 42*

* Formula 1 is based on the assumption that the curves are parallel. Since these curves in Figure 5 are not parallel, the values indicated are only approximate.

In both dwarf and tall strains the indexes of the treated plants were greater than those of the untreated plants. The indexes of the treated dwarf plants were greater than those of the treated tall ones, indicating that the effects of gibberellin on the aging of the dwarf plant were more stimulative than on that of the tall one.

F. Cell division in the shoot apex

The plants were treated once with gibberellin and after certain periods the apical region of the plants were fixed with CARNOY's solution, dehydrated by normal butyl alcohol, and imbedded in paraffin. Serial longitudinal sections, 8μ thick, were stained with DELAFIELD's haematoxylin. The meta-, and- and telophase figures were classified according to the orientation of the cell plate by the following two categories : (1) longitudinal division, when the cell plate inclined between 0 and 45° of the plant axis, and (2) transverse division, when the cell plate inclined between 45° and 90° of the same. The data from five replications of the observations in

Table 4.

Number of cell divisions in shoot apex of *Pharbitis nil.* *

A : Transverse division**

Distance below shoot apex (mm)	No treatment					Treatment***					
	Hours after treatment					Hours after treatment					
	0	24	48	72	96	12	18	24	48	72	96
0-0.1	0	2.6	3.4	0	0	22.3	17.1	1.2	1.0	4.3	0
0.1-0.2	0	6.8	4.1	1.3	3.7	12.5	24.8	6.7	2.4	4.7	1.7
0.2-0.3	1.4	3.3	7.4	2.9	10.4	13.9	18.2	11.7	2.8	1.6	0
0.3-0.4	0	1.4	7.1	7.0	5.7	14.7	16.5	12.5	17.4	6.3	1.3
0.4-0.5	0	0	3.7	7.9	9.6	1.7	18.3	8.2	10.6	7.7	2.3
0.5-0.6			1.5	9.2	4.9	0	7.8	5.2	8.4	4.3	4.1
0.5-0.7			3.0	6.4	5.5		3.7	5.7	9.2	3.7	6.2
0.7-0.8			2.8	5.3	4.2		0	7.2	1.6	5.7	2.5
0.8-0.9			0	1.4	3.8			5.3	4.8	11.2	5.3
0.9-1.0				0	1.6			1.6	3.3	6.8	3.2
1.0-1.1					3.6			0	0	13.6	7.4
1.1-1.2					2.3					10.8	7.4
1.2-1.3										1.2	5.7
1.3-1.4										0	3.8
1.4-1.5											3.2
1.5-2.0											14.8
Total	1.4	14.1	33.0	41.4	55.3	65.1	106.4	65.3	61.5	81.9	68.9

B : Longitudinal division**

0-0.1	0	1.3	1.6	2.7	2.3	22.7	11.6	1.3	2.6	4.3	1.2
0.1-0.2	0	1.8	0	1.2	0	6.3	4.7	5.2	1.6	4.8	0
0.2-0.3	1.5	2.7	8.3	5.6	0	3.0	7.4	3.5	4.5	4.1	0
0.3-0.4	2.0	0	3.7	2.8	0	3.2	4.7	6.3	2.5	2.7	0
0.4-0.5	0	0	5.8	6.6	1.4	1.6	0	7.4	5.2	1.1	1.0
0.5-0.6			2.2	5.3	3.6	0	0	5.1	3.9	1.3	1.6
0.6-0.7			1.9	1.6	0		1.5	6.4	3.1	0	2.4
0.7-0.8			1.3	1.1	3.2		0	4.5	2.2	1.6	2.0
0.8-0.9			0	1.1	3.3			1.2	4.6	1.1	2.3
0.9-1.0				0	2.6			1.2	0	1.3	6.2
1.0-1.1					3.7			0	0	2.6	1.2
1.1-1.2					1.4					1.3	2.4
1.2-1.3										0	2.5
1.3-1.4											2.2
1.4-1.5											6.3
1.5-2.0											11.7
Total	3.5	5.8	24.8	28.0	21.5	36.8	29.9	42.1	30.2	26.2	43.0

* "Kidachi", a dwarf strain.

** See the text on the methods of the classification of cell divisions.

*** Plants were treated with 0.02 cc of a 1000 ppm solution of gibberellin.

the middle eight sections are shown in Table 4. At the twelfth hour after the treatment a greater number of cell divisions were found in 500 μ of the shoot apex with the maximum in the uppermost 100 μ. During the period from 18 to 72 hours, the later the observation was made, the wider was the active meristematic zone. Many of the cell divisions were observed at ninety-sixth hour after treatment in the zone between 500 and 2000 μ below the shoot apex. In the untreated plants the

Table 5.
Stem length, cell length and the ratios of respective lengths of the treated to those of the control plants of *Pharbitis nil.*

Part of plant			"Kidachi"			"Violet"		
			Cont.	Gib. *	Gib. as % of Cont.	Cont.	Gib. *	Gib. as % of Cont.
Total length of epicotyl in mm			21	233	887	44	197	448
Length × Diameter** of hypocotyl in mm			45×2.4	74×3.0	164×125	109×2.3	120×2.6	111×113
Length × Diameter** of the first internode in mm			12×2.3	89×1.5	733×65	33×2.2	80×1.6	246×73
Length × Diameter of the second internode in mm			6×2.0	69×1.3	1150×65	9×2.1	62×1.5	689×71
Cell dimension in μ***	Hypocotyl	Cortex	longitudinal	156	276	176	279	370	133
			radial	52	49	95	41	50	122
			tangential	53	41	78	45	51	113
		Pith	longitudinal	157	267	170	218	278	128
			radial	74	73	99	66	83	126
			tangential	73	75	104	67	70	104
	1st internode	Cortex	longitudinal	134	178	132	149	153	103
			radial	50	18	36	30	15	50
			tangential	43	26	61	32	22	69
		Pith	longitudinal	86	165	192	92	103	112
			radial	88	57	65	84	55	65
			tangential	69	54	78	73	51	70
	2nd internode	Cortex	longitudinal	113	101	89	76	85	112
			radial	43	15	35	25	18	72
			tangential	37	22	59	22	20	91
		Pith	longitudinal	76	97	128	72	83	115
			radial	69	41	60	49	38	78
			tangential	63	39	62	45	36	80

* The plants were treated with a 1000 ppm solution of gibberellin, and measured after ten days.

** Length of major axis.

*** Mean of 30 cells in the middle portion in respective tissues.

cell divisions were found in the distance between 200 to 1200μ. The total number of transverse and that of longitudinal cell divisions in the treated plants were 3 and 2.5 times as many as those in the control, respectively.

G. Cell length

The middle protions of the first, second and third internodes were hand-sectioned on the twelfth day after the first treatment, and the longitudinal, radial and tangential dimensions of 30 cells of the cortical and pith parenchyma were measured with an ocluar micrometer. The results are given in Table 5. The data from the third internode were excluded because it was still growing. In "Kidachi", the longitudinal cell lengths of all organs of the treated plants with an exception in the cortical cells of the second internode were longer than those of the untreated ones. Especially the cells in hypocotyl were conspicuously elongated. On the contrary, the radial and tangential dimensions of the cells were smaller than the untreated plant in many organs. The tendency was the same in "Violet", but the increase in cell length was not so much as in "Kidachi".

Table 6.

Cell length in shoot apex of *Pharbitis nil.* (μ)

Distance below shoot apex (mm)	Control					Gibberellin					
	Hours after treatment					Hours after treatment					
	0	24	48	72	96	12	18	24	48	72	96
0–0.1	9.5	9.8	10.9	9.7	9.7	9.9	10.2	9.8	9.8	10.4	9.4
0.1–0.2	13.0	15.0	17.1	20.2	20.4	16.4	17.7	15.0	13.8	14.4	12.8
0.2–0.3	13.2	17.8	19.8	20.2	23.5	19.4	21.2	16.8	16.1	19.7	18.8
0.3–0.4	14.6	20.1	20.8	21.0	20.2	18.2	21.9	22.3	19.6	19.5	21.1
0.4–0.5	15.9	23.2	20.7	14.5	21.4	17.6	18.8	19.0	20.7	20.0	20.3
0.5–0.6	23.3	25.4	21.7	19.3	20.8	22.1	18.9	22.9	23.0	21.2	21.2
0.6–0.7	49.2	32.1	21.6	20.5	21.4	37.2	28.2	31.4	20.7	26.4	22.5
0.7–0.8	61.3	43.5	23.3	20.0	20.8	52.7	30.4	33.6	20.7	24.4	25.4
0.8–0.9	83.2	57.8	25.6	23.5	22.1	64.7	41.6	35.7	21.0	21.9	23.8
0.9–1.0	97.8	67.6	28.2	24.9	23.6	74.5	63.5	38.3	23.6	27.2	27.8
1.0–1.1						84.6	64.8	43.4	24.3	27.9	28.3
1.1–1.2						93.6	65.9	47.3	25.2	28.3	26.4
1.2–1.3						101.8	68.8	50.7	26.4	29.1	28.5
1.3–1.4										28.7	28.2
1.4–1.5										30.6	29.3
1.5–1.6											30.2
1.6–1.7											29.6
1.7–1.8											30.2
1.8–1.9											29.8
1.9–2.0											28.9

Parts below the line is hypocotyl.

The cell lengths in the shoot apex are given in Table 6. In the uppermost 1 mm of the growing point the cell lengths of the treated plants were not greatly different from the control until 96 hours after gibberellin treatment.

H. Ratio of stem length to cell length

The ratios of internodal length to cell length are given in Table 7. In the treated plant of "Kidachi", the ratios in hypocotyls were not vastly different from the control, but those in the treated epicotyl were higher than in the control. In the second internode the ratios were 11.4 times in cortex and 9.5 times in pith as high as those of the control. The tendency was the same in "Violet", but the ratios were lower than in "Kidachi". These facts indicate that the internodal cells of the treated plant continued to divide at least three times as more than those of the control before they attained to a permanent state, but in the hypocotyl the cell division was not affected by gibberellin.

Table 7.
The ratios of stem length to cell length in *Pharbitis nil.*

	Part of plant		Ratio of stem length to cell length		Treated as compared to Control
			Control	Gibberellin	
"Kidachi"	Hypocotyl	Cortex	290	270	1.0
		Pith	290	280	1.0
	First internode	Cortex	90	500	5.6
		Pith	140	540	3.8
	Second internode	Cortex	50	570	11.0
		Pith	80	710	9.0
"Violet"	Hypocotyl	Cortex	390	320	0.8
		Pith	500	430	0.9
	First internode	Cortex	230	520	2.3
		Pith	360	780	2.2
	Second internode	Cortex	120	730	6.0
		Pith	130	750	5.8

The ratios were calculated from the data in Table 5.

As shown in Table 1, the most striking elongation was found in the upper portions of an internode. The cells in the upper portions were not so elongated, however, indicating that these cells continued to divide after division had ceased in the middle and lower portions. From these facts it can be said that the elongation of the stem by gibberellin was mainly caused by the elongation of the upper portion of the internodes as a result of increase in cell number.

I. Plant weight

The *Pharbitis* plants were grown under two conditions, *i. e.* (a) in soil under long day conditions in the field and (b) in water culture under cntinuous artificial illumination. Both fresh and dry weights of shoot and root were measured every day after the gibberellin treatment. The results were as follows :

(a) The data from ten plants grown in soil are shown in Figures 6 and 7. In "Kidachi", the fresh weights of the shoot and root of the treated plants on the

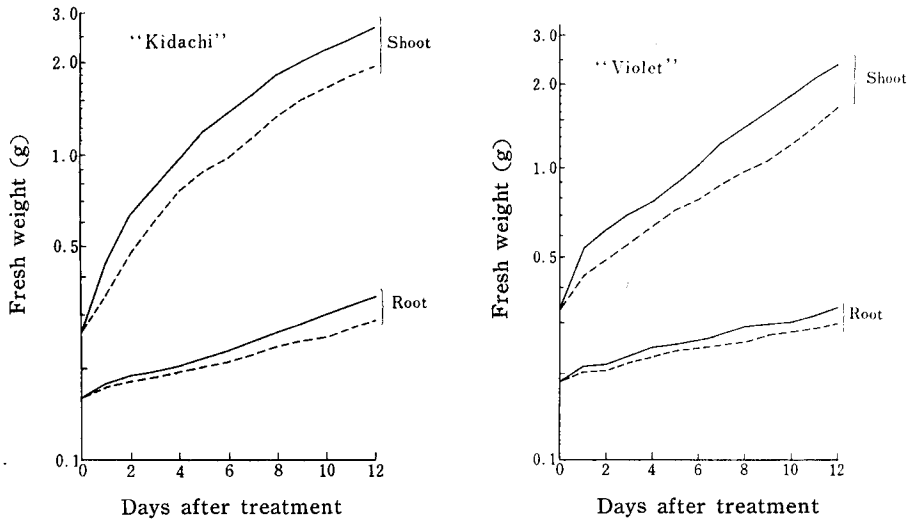


Fig. 6. Fresh weight of *Pharbitis nil* plotted logarithmically against time.
-------- Control ———— Gibberellin

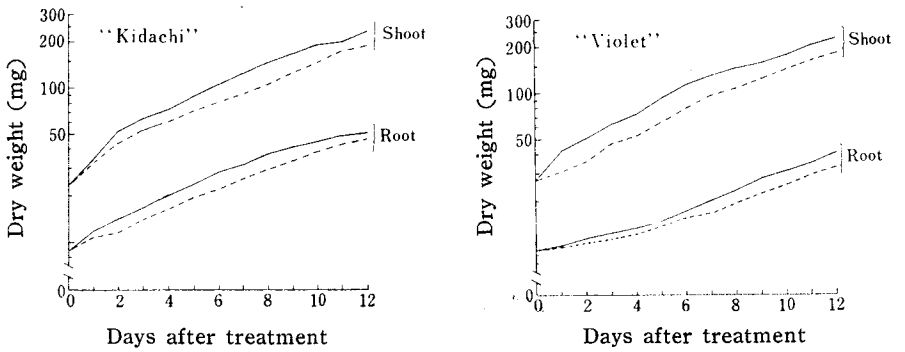


Fig. 7. Dry weight of *Pharbitis nil* plotted logarithmically against time.
-------- Control ———— Gibberellin

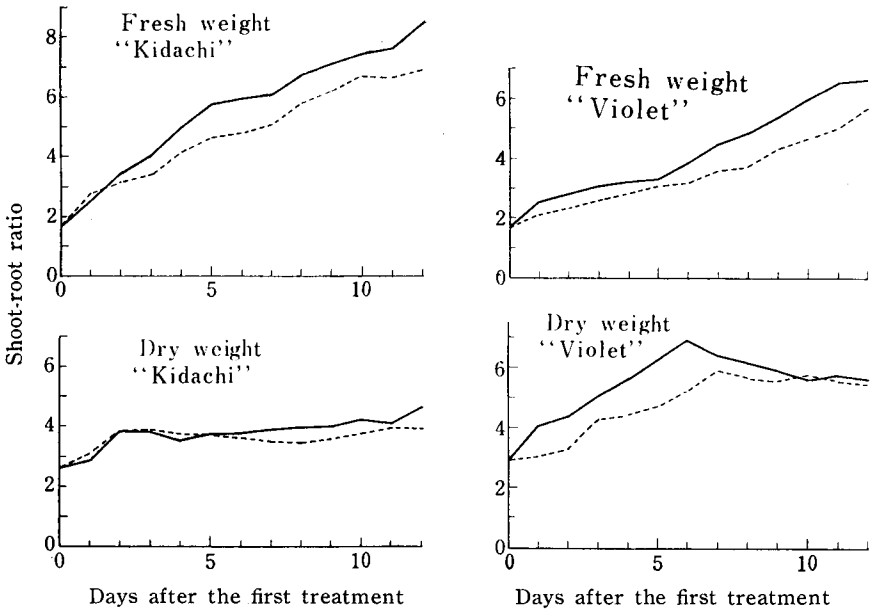


Fig. 8. Shoot-root ratio of fresh and dry weight of *Pharbitis nil.*
-------- Control ———— Gibberellin

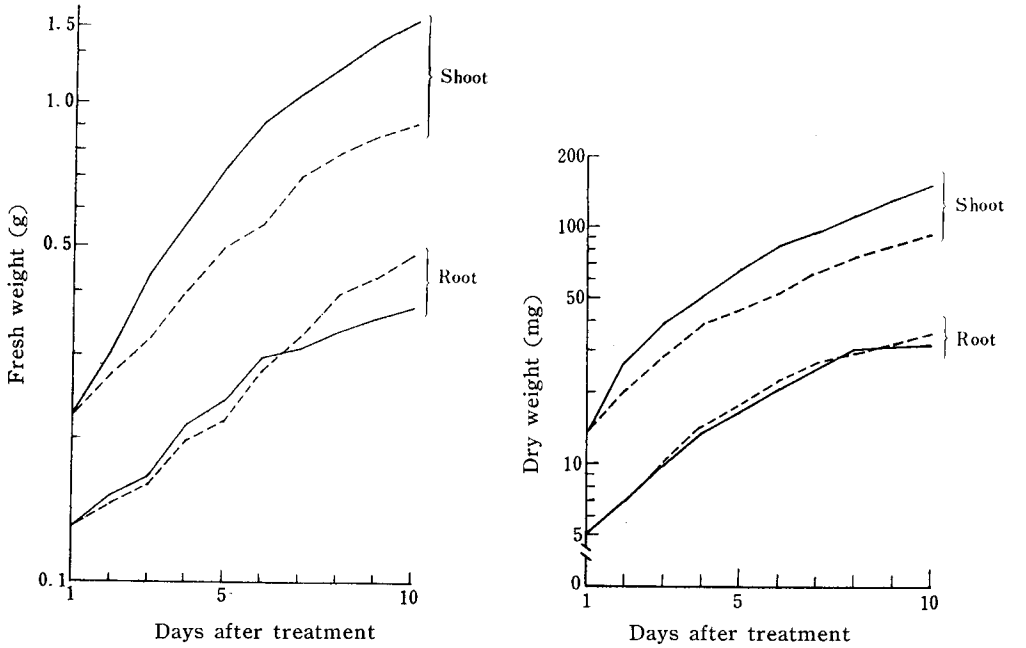


Fig. 9. Effect of gibberellin on plant weight of "Kidachi", a dwarf strain of *Pharbitis nil,* cultured with WHITE's mineral mixtures.
-------- : Control ———— : Gibberellin

twelfth day after the first treatment were 1.4 and 1.2 times as large as the control. The dry weights of these portions of the treated plants were 1.2 and 1.1 times as large as the control. The ratios of the treated to the untreated plant were higher in the shoot than in the root. The tendency was the same in "Violet".

Shoot-root ratios are shown in Figure 8. The ratios, with a few exceptions, were higher in the treated plants than in the control.

(b) The results of dwarf strain in the case of the water culture with WHITE's mineral mixtures are given in Figure 9 in which each dot represents the mean of ten plants. The plant weight increased less than in the case of the soil culture. The shoots of the treated plants were heavier than those of the control. The root growth of the treated plant was promoted in the earlier period but inhibited in the later period. The same tendency was shown in dry weight analysis, but the tendency was not so pronounced. Shoot-root ratios (Fig. 10.) of the treated plants were higher than the control in both fresh and dry weights. The reasons for the differences between water and soil culture, in the root weight especially, are not apparent, but the lack of oxygen in the culture medium could be a factor.

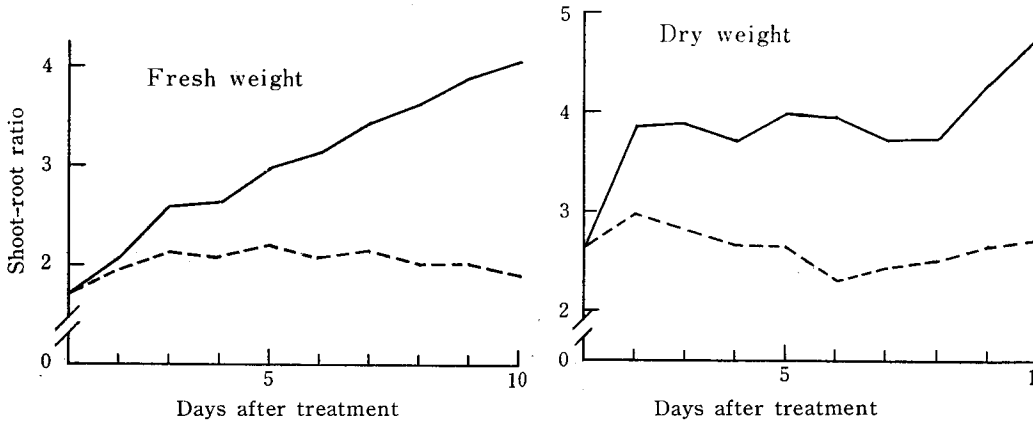


Fig. 10. Shoot-root ratio of "Kidachi", a dwarf strain of *Pharbitis nil*, cultured with WHITE's medium.

- - - - Control ——— Gibberellin

J. Histological changes in the stem

Gibberellin induced also the following histological changes in the stem in *Pharbitis* : (a) reduction of the activity of cambial growth, and (b) acceleration of cell-wall thickening accompanied with the lignification in xylem and pericyle. The processes of these changes are illustrated in Plate I.

Cambium in the first internode was found to consist of one or two cell layers

on the third day after the treatment. With the lapse of time, however, the number of cell layers decreased, so that the cambial zone could hardly be observed on the tenth day, while the cambial zone in the control consisted of three or more cell layers on the same day.

In the first internode on the fifth day after treatment, slight cell-wall thickening accompanied with lignification was observed in the zone from one to three cell layers in the secondary xylem, and the width of the sclerenchyma was almost the same in and between the bundles. On the tenth day, the cell-wall thickening was also observed in the second and third internodes.

In the control, slight cell-wall thickening was first observed on the seventeenth day after sowing. The sclerenchyma on the thirty-second day consisted of many cell layers and was wider in the fascicular portion than in the interfascicular one.

If one side of the middle portion in the first internode of ten-day old seedlings was pasted with lanolin containing one per cent gibberellin, cell-wall thickening was observed all around the internode. When the middle portion in the first internode was pasted with the gibberellin entirely around the stem, the cell-wall thickening was accelerated in both upper and lower directions from the treated portion. In the majority of pericycle cells, cell-wall thickening began a little later than in the xylem. Thin-walled cells were found scattered randomly among sclerenchymatous pericycle cells.

K. Comparison in the effects between gibberellin and IAA

About 20 mg of lanolin paste containing 1.0 per cent gibberellin or 0.1 per cent IAA was applied to one side of the hypocotyl of six-day old seedlings. Applications were made 1 cm below the cotyledonary node at right angles to the plane formed by cotyledonary petioles, as shown in Figure 11. The results after two

Table 8.
Effects of gibberellin or IAA on hypocotyl of *Pharbitis nil**

Treatment	Length of hypocotyl	Degree of curvature	Cell-wall thickening in xylem	Activity in cambium
Control	41.3	0	+	++
Gibberellin	57.8	0	+	+
IAA	43.8	-37.2	+	++

* "Kidachi", a dwarf strain.
See the text and Figure 11 on the methods of the treatment.
Observation on the second day after the treatment.

Table 9.
Cell length in hypocotyl of *Pharbitis nil.** (μ)

Treatment	Part of plant	Radial	Tangential	Longitudinal
Control	Treated side	45.4	44.9	175.5
	Opposite side	—	—	—
Gibberellin	Treated side	47.8	48.1	278.2
	Opposite side	45.7	45.8	276.1
IAA	Treated side	49.4	50.6	265.8
	Opposite side	44.5	42.2	219.3

* "Kidachi", a dwarf strain.
See the text and Figure 11 on the methods of the treatment.
Observations on the second day after the treatment.

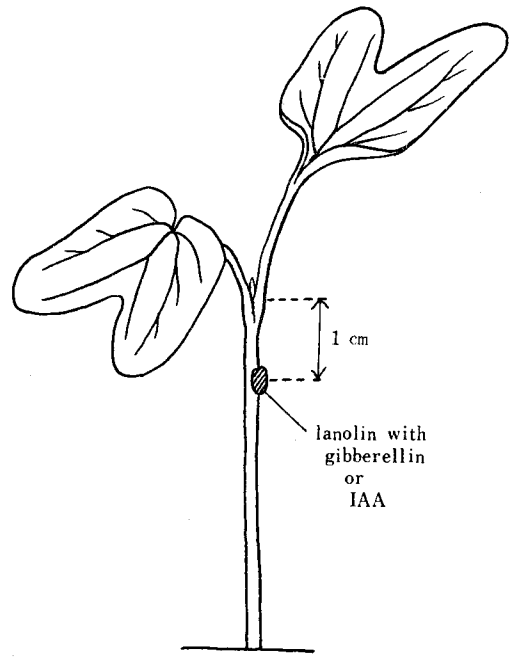


Fig. 11.

days from the treatment are given in Tables 8 and 9 and shown in Plate II. The hypocotyls treated with gibberellin grew more pronouncedly than either the control or IAA treated ones. By IAA treatment, the hypocotyls became a little longer than the control, showing a curvature caused by growth promotion of the treated side.

It was found by anatomical observations that the dimensions in both tangential and radial directions of the cells in the treated portion were not affected, but those in longitudinal direction became longer than the control if treated with gibberellin or IAA. The cambial zone became thinner only in the case of gibberellin treatment.

2. *Oryza sativa* L.

The seeds of rice were (1) soaked in absolute alcohol for five minutes, (2) sterilized with ten per cent calcium hypochlorite solution for 30 minutes, (3) washed thoroughly with tap water, and then (4) sown in deionized water in a Petri dish, which was kept in an incubator regulated at 30°C.

When the coleoptiles grew about 1 mm in length, the seedlings were planted in six linear cuts on a piece of foamed polyurethane (5cm×5cm×1cm); the seedlings were planted so as to place the embryos into six small holes made at equal distances along a linear cut. The polyurethane piece was kept in a vessel (6cm×6cm×2cm) containing 40 cc of an aqueous gibberellin solution without renewal until the end of the experiment. For the control, the box was filled with the same volume of deionized water. The plants were grown in an incubator regulated at 30°C under continuous illumination by a fluorescent lamp.

A. Elongation of leaf

The seedlings were treated with a 1, 10 or 100 ppm solution of gibberellin, and

Table 10.
Effects of gibberellin on leaf elongation of rice seedlings*. (cm)

No. of Leaf	Conc. of Gibberellin (ppm)	Days after treatment 2	4	6	8	10
1st leaf	0	0.97	1.32	1.38	1.41	1.47
	1	1.46	2.82	2.92	3.01	3.24
	10	1.54	3.42	3.54	3.65	3.68
	100	1.58	3.54	3.68	3.90	4.04
2nd leaf	0	—	3.52	4.32	4.88	5.06
	1	—	8.52	12.53	13.74	13.89
	10	—	7.78	13.61	15.60	15.73
	100	—	9.26	15.31	16.47	16.63

* "Tamanishiki", a dwarf strain of *Oryza sativa*.

their first and second leaves, sheaths and blades altogether, were measured after 2, 4, 6, 8 and 10 days. The results are given in Table 10. It is apparent from the data that the effects on the elongation of the first leaf were noticed on the second day, and the rate of the leaf elongation was highest on the fourth day. The first leaf of the plant treated with a 1 ppm solution was 2.2 times as long as the control on the tenth day. The second leaf was 2.7 times as long as the control on the same day. The leaf lengths increased in proportion to the concentration of gibberellin.

The lengths of the leaf sheath and leaf blade measured separately are given in Table 11. The second leaf sheath was 4 times as long as the control and seemed to be most sensitive to gibberellin. While the leaf blade was 2.5 times as long as the control.

When grown at 33°C, the second leaf sheath of the treated plants increased in

Table 11.
Leaf length, lower epidermal cell length and the ratio of leaf length to cell length of rice seedling.

	No. of leaf	Control	Gibberellin	Gibberellin to Control
Leaf length in cm	1st leaf sheath	0.99	2.21	2.2
	2nd leaf sheath	2.32	9.28	4.0
	3rd leaf sheath	3.62	5.22	1.4
	1st leaf blade	—	—	—
	2nd leaf blade	1.36	3.39	2.5
	3rd leaf blade	4.81	14.64	3.0
Epidermal cell length in μ	1st leaf sheath	73.6	112.2	1.5
	2nd leaf sheath	79.3	140.0	1.8
	3rd leaf sheath	69.2	80.3	1.2
	1st leaf blade	—	—	—
	2nd leaf blade	59.7	79.6	1.3
	3rd leaf blade	55.1	79.3	1.4
Ratio of leaf length to cell length	1st leaf sheath	134.5	197.0	1.5
	2nd leaf sheath	292.6	653.5	2.2
	3rd leaf sheath	523.1	650.1	1.2
	1st leaf blade	—	—	—
	2nd leaf blade	227.8	425.9	1.9
	3rd leaf blade	873.0	1846.2	2.2

Measurements on the seventh day after the treatment.
Plants were grown at 30°C under continuous illumination, and treated with a 1 ppm gibberellin solution.

length to 5.2 times that of the control (Table 12). The elongation of the second leaf blade of gibberellin treated plants was affected less at the higher temperature than at the lower one.

Table 12.
Leaf length, lower epidermal cell length and the ratio of leaf length to cell length of the second leaf of rice seedling.

	Part of leaf	Concentration of gibberellin in ppm			
		0	1	10	100
Leaf length in cm	Leaf sheath	2.42 (1)*	12.17 (5.2)	14.46 (6.2)	15.28 (6.5)
	Leaf blade	1.39 (1)	2.48 (1.8)	2.73 (2.0)	3.09 (2.2)
Cell length in μ	Leaf sheath	93.9 (1)	131.5 (1.4)	125.0 (1.3)	123.3 (1.3)
	Leaf blade	62.2 (1)	71.9 (1.2)	77.7 (1.3)	76.9 (1.2)
Ratio	Leaf sheath	247 (1)	925 (3.7)	1156 (4.7)	1239 (5.0)
	Leaf blade	219 (1)	345 (1.6)	351 (1.6)	401 (1.8)

* The number in parentheses indicates the ratio of the treated to the control plant. The plants were grown at 33°C.

B. Cell length

Longitudinal lengths of lower epidermal cells of the plants grown at 30°C and at 33°C are given in Tables 11 and 12, respectively. The largest growth response to the gibberellin treatment was obtained in the second leaf sheath. The lengths of the cells in the leaf blade were also increased by gibberellin treatment, but the differences between the cell lengths in the second and third leaf blade were not so distinct as those in the leaf sheaths.

The cells of the control plants grown at 33°C were longer than those of the control grown at 30°C, but the cells of the plants treated with gibberellin, grown at the former temperature, were shorter than those of the latter.

C. Ratio of leaf length to cell length

The ratios of the leaf length to the cell length are given in Tables 11 and 12. The ratios in the treated plants were higher than in the controls. This was most conspicuous in the second leaf sheath. The ratios from the treated plants grown at the higher temperature were greater than those from the plants grown at the

lower temperature. From the results, it can be said that the cell number increased strikingly in the leaf sheath and even at the higher temperature used.

D. Effects on root growth

The number of fibrous roots was scarcely affected by treatment with a 1 ppm gibberellin solution, but decreased markedly with higher concentrations (Table 13). The total length of fibrous roots from the treated plants was shorter than that of the control and for plants treated with a 10 or 100 ppm solution, the lengths were each one half that of the controls on the tenth day.

Table 13.
Number and length of fibrous roots of rice seedling.

	Concentration of gibberellin (ppm)	Days after treatment 2	4	6	8	10
Number of fibrous root	0	1	4.4	7.0	8.8	8.2
	1	1	4.8	6.6	6.8	9.2
	10	1	4.3	5.6	5.4	5.2
	100	1	4.1	6.8	5.5	4.0
Length of fibrous root in cm	0	1.5	17.9	30.5	33.4	35.8
	1	1.5	16.5	21.7	27.2	31.4
	10	1.8	13.1	15.3	17.5	18.6
	100	1.4	11.4	16.2	16.8	17.4

Table 14.
Number of cell division in root tip of rice seedling.

Days after treatment	Concentration of gibberellin (ppm)	Distance from root apex (μ) 0–100	100–200	200–300	300–400	400–500	Total
2	0	2.2	5.4	4.3	6.7	2.4	21.0
	1	3.5	5.4	9.8	8.7	5.6	33.0
	10	10.3	21.7	23.7	14.0	7.7	77.4
	100	2.5	7.0	20.5	16.5	13.0	59.5
3	0	17.3	17.5	6.5	0.5	0.7	42.5
	1	4.7	16.4	4.4	1.2	1.1	27.8
	10	14.3	29.6	24.3	6.1	5.2	79.5
	100	2.4	15.6	12.5	1.3	1.2	33.0
4	0	12.8	35.4	21.7	5.3	1.2	76.4
	1	5.6	21.4	33.4	17.2	1.5	79.1
	10	2.7	20.6	21.9	5.8	0.5	51.5
	100	1.7	6.5	2.3	0.2	0	10.7

E. Number of cell divisions in the root tips

The root tips of rice seedlings were fixed in modified NAWASHIN's fluid (Craf II) (56), dehydrated by the normal butyl alcohol procedure, and imbedded in paraffin. Serial longitudinal sections, 8 μ thick, were stained with DELAFIELD's haematoxylin.

The number of mitotic figures in the middle eight sections of root tips is given in Table 14. In the treated plants, within a short time after the treatment, more mitotic figures were found than in untreated control plants, and for the plants the higher the concentrations of gibberellin were, the greater were the observed frequencies of mitotic figures. Later, however, the frequencies in the treated plants decreased below that in the controls except for those in the case treated with a 1 ppm gibberellin solution.

F. Plant weight

The results of daily measurements of both fresh and dry weights of rice seedlings are given in Tables 15 and 16. The treated materials were heavier in both fresh and dry weights than the controls, but there were slight differences among the treated plants.

Roots of the treated plants were lighter than those of the controls in both fresh and dry weights. This inhibitory effect was more striking with the higher

Table 15.
Fresh weight of shoot and root, and shoot-root ratio of rice seedling. (mg)

Part of plant	Concentration of gibberellin (ppm)	Days after treatment				
		2	4	6	8	10
Shoot	0	3.30	10.28	18.41	25.63	28.42
	1	3.94	15.83	21.42	31.32	33.16
	10	4.14	16.80	26.04	33.05	34.62
	100	3.62	19.68	28.81	33.41	35.83
Root	0	2.56	16.38	23.92	35.10	37.84
	1	2.17	12.60	19.24	25.75	31.36
	10	2.32	10.93	15.37	21.32	22.09
	100	1.74	7.22	13.02	18.58	19.13
Shoot-root ratio	0	1.32	0.63	0.76	0.73	0.75
	1	1.81	1.26	1.11	1.23	1.06
	10	1.68	1.54	1.63	1.55	1.50
	100	2.08	2.72	2.22	1.80	1.87

Correspond to Table 16.

Table 16.
Dry weight of shoot and root, and shoot-root ratio of rice seedling. (mg)

Part of plant	Concentration of gibberellin (ppm)	Days after treatment 2	4	6	8	10
Shoot	0	0.28	0.98	2.51	4.30	5.30
	1	0.36	1.72	3.07	5.98	7.89
	10	0.38	1.71	3.80	6.12	8.01
	100	0.32	1.92	4.85	6.36	8.09
Root	0	0.21	1.26	1.88	2.50	2.92
	1	0.18	0.76	1.14	1.63	1.94
	10	0.16	0.68	0.96	1.03	1.18
	100	0.12	0.41	0.87	0.96	1.14
Shoot-root ratio	0	1.33	0.78	1.33	1.72	1.83
	1	2.00	2.26	2.59	3.67	4.06
	10	2.83	2.52	2.47	5.94	6.77
	100	2.66	4.68	3.72	6.64	7.10

Correspond to Table 15.

concentrations.

Shoot-root ratios in both fresh and dry weights are given in Tables 15 and 16. The shoot-root ratios in the treated plants were higher than in the controls, and the ratios in dry weights were higher than in fresh weights for the treated plants. These facts indicate that the photosynthates in the treated plant are utilized preferentially for the growth of aerial parts.

3. *Erigeron annuus* L.

Several plants of *Erigeron* in rosette form were collected from the field on September 18, and grown in 15 cm pots at 30°C under either long or short day conditions. On September 19 a 0.04 cc drop of a 1000 ppm gibberellin solution was given to the base of the leaves, as close to the apex as possible. Deionized water, in a drop of the same volume, was given to the control plants.

A. Cell division and cell length in the shoot apex

In order to examine the effects of gibberellin, the frequency of cell divisions in the shoot apex was investigated. Microscopic preparations of the shoot apex were made by the same methods as employed in *Pharbitis*. The cell divisions at meta-,

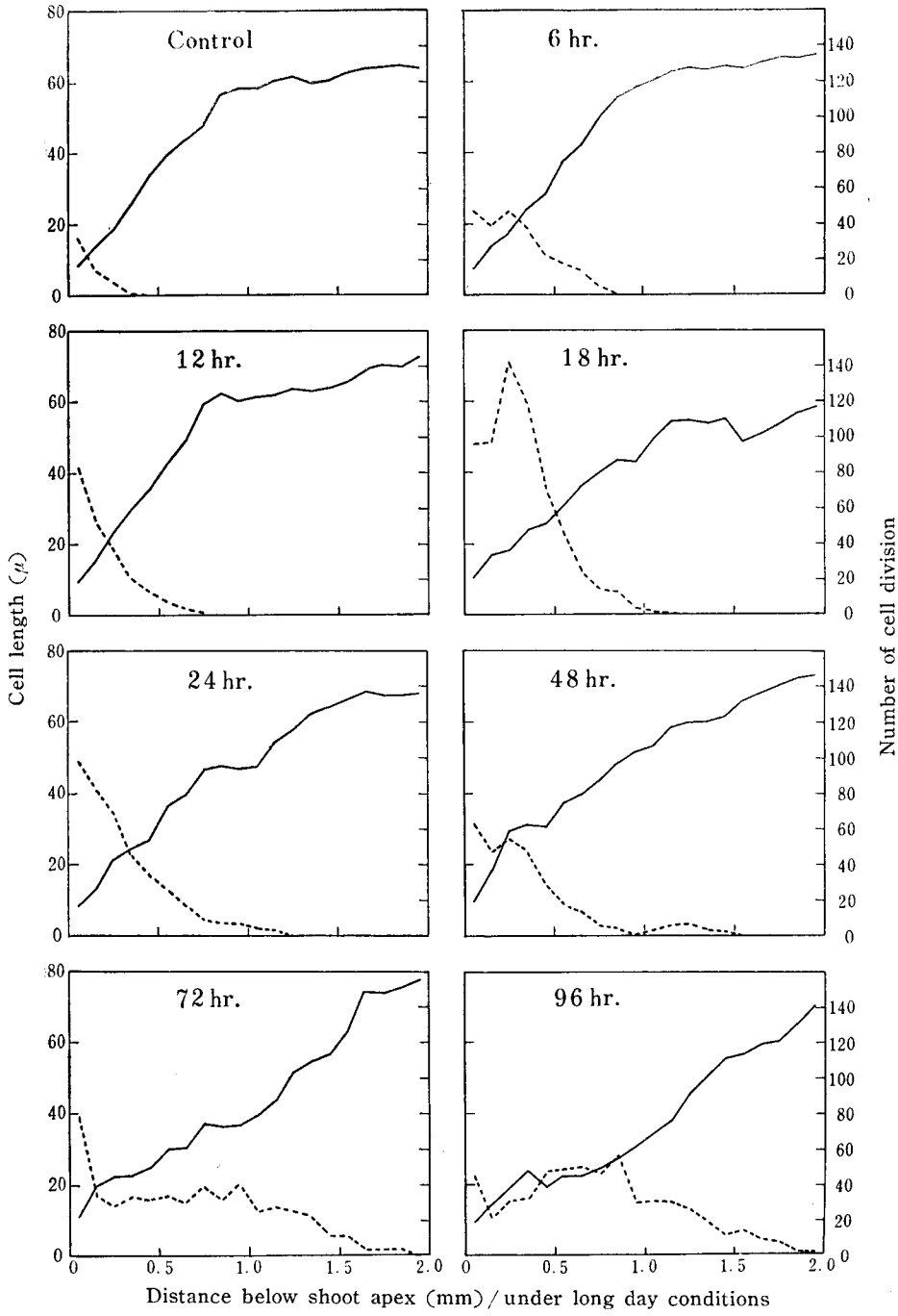
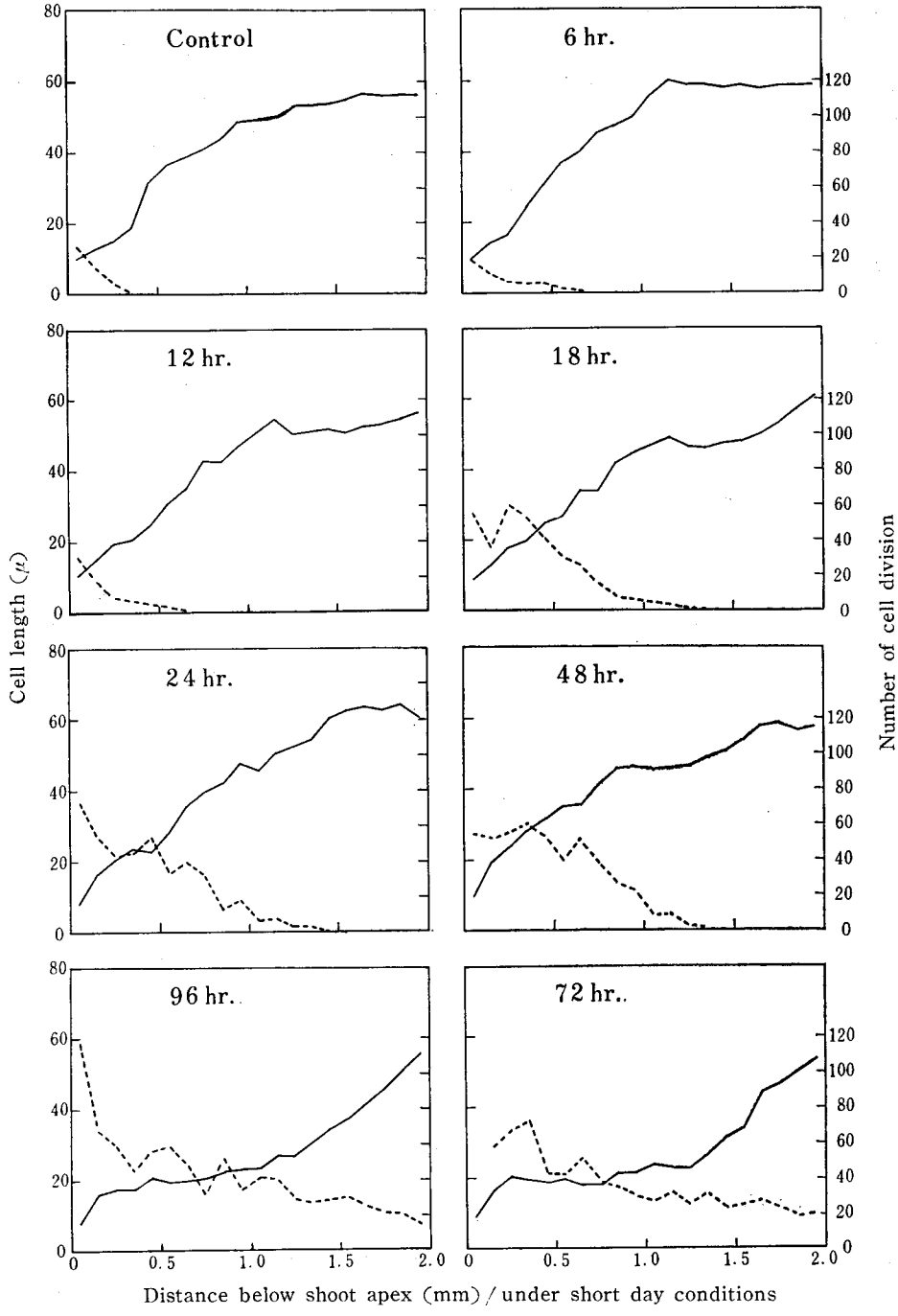


Fig. 12. Total number of cell division and cell length in shoot apex of *Erigeron annuus*. Left ordinate : Cell length in mm. Right ordinate : Number of cell division.

------- Number of cell division ——— Cell length


Control
6 hr.
12 hr.
18 hr.
24 hr.
48 hr.
96 hr.
72 hr.
Cell length (μ)
Number of cell division
Distance below shoot apex (mm) / under short day conditions
0
0.5
1.0
1.5
2.0
20
40
60
80
100
120

ana- and telophase were classified as transverse or longitudinal according to the orientation of the cell plates as in the case of *Pharbitis*. Since cells at late prophase in *Erigeron* were distinguishable from the resting stage, they were regarded as dividing cells. The results, the total number of all mitotic figures, are given in Figure 12.

Within 12 hours after treatment with gibberellin there was a marked increase in the number of cell divisions. At the twelfth hour a great number of cell divisions were found in 500 μ of the shoot apex with the maximum in the uppermost 100 μ. The active meristematic zone became wider with the lapse of time from 18 to 72 hours. As shown in Plate II, the stems of the treated plants were longer than those of the controls at the ninety-sixth hour, and many cell divisions were observed in the elongated stem. Control plants remained in rosette form and only a few cell divisions were observed. The number of transverse cell divisions in the treated plants was 33 times that in the controls under long day conditions at the ninety-sixth hour, and at the same time the number of longitudinal cell divisions was 2 times that in the controls. Under short day conditions, the numbers of transverse and longitudinal cell divisions at the ninety-sixth hour became 33 and 10 times as many as those in the controls, respectively. The number of late prophase cells was more than ten times as great as that in the controls irrespective of photoperiodic conditions. The ratios of the number of cell divisions in the treated plant to that of the control under short day conditions were 0.8, 1.4, 6.6, 7.8, 9.2, 18.1 and 16.1 after 6, 12, 18, 24, 48, 72 and 96 hours from the treatment, respectively.

At the ninety-sixth hour after the treatment, the cell length in the shoot apex of elongated plants did not differ so much from that of the controls, but the stems of treated plants were about 5 mm taller than those of the controls which did not elongate detectably during the experimental period. These facts may indicate that the stem growth caused by gibberellin is mainly due to the increase in the frequency of cell divisions.

B. Elongation of stem and flower formation

In the previous work (35) the plants in rosette form were treated in two different ways ; (1) a quantity of a 10 ppm solution of gibberellin was sprayed on leaves once every day for twenty days from June 8, and (2) a quantity of a 100 ppm solution was sprayed on leaves once every other day for three times from September 28. In each case, stem elongation was observed after about ten days from the beginning of the treatments, and flower buds were formed on the plants kept under long day conditions after about 40 days from the beginning of the

treatments as shown in Plate II. Under short day conditions the stem elongated a little, but the plants resumed the rosette habit, and no flower bud was formed. When, however, the plants were shifted from short day conditions to long day conditions within a few days after the end of the gibberellin treatment, flower buds were formed.

In the present experiment, the seeds were sown on May 30, and the seedlings were grown under natural day light. A 0.03 cc drop of a 1000 ppm gibberellin solution was applied to the base of the leaves of the plants in rosette form as close to the shoot apex as possible four times at the intervals of five days from July 2. Flower buds were observed toward the end of August on the elongated stem.

The application of gibberellin induced the abnormalities in leaf shape as shown in Plate II.

IV. Discussion

The most striking and immediate effect of gibberellin on a plant is the acceleration of shoot growth. In many early investigations it was believed that the elongation of organs promoted by gibberellin was caused mainly by cell elongation (1, 3, 15, 21, 23, 48, 52, 55). The following workers have reported that the stem elongation is due to the increase in the number of cells in the internodes (25, 30, 38, 43, 46). In his previous paper (46), the present author reported that the stem in *Pharbitis* elongates in response to gibberellin, and that the pronounced stem elongation is due to the increase in the number of cells rather than cell elongation.

According to LANG (31), gibberellin application results in flower formation, which is preceded by considerable stem elongation in *Hyoscyamus*. It has also been reported that gibberellin causes conspicuous stem elongation and flowering in numerous cold-requiring and long day plants kept under strictly non-inductive temperature or photoperiod regimes (10, 12, 13, 14, 19, 34, 36, 52); the requirements of low temperature and often long day photoperiods of the plants can be replaced by gibberellin. As reported by the present author (36), gibberellin causes an elongation of the stem and induces flowering under long day conditions in *Erigeron*, which remains in the form of a vegetative rosette if not exposed to low temperature. However, if the plant treated with gibberellin is grown under short day conditions, the shoot elongates first and then ceases to elongate, and the plant resumes the rosette form with an elongated stem. In this case gibberellin can replace only the requirement of low temperature, but not of long day photoperiods.

Pharbitis is a short day plant. Under day lengths slightly longer than the

critical, the gibberellin treated plant forms flower buds (34). This phenomenon seems to be correlated with the activating effects of gibberellin on the cell division in the shoot apex. The morphogenetic activity is accelerated by gibberellin and the flowering stimulus may play its role efficiently in the activated meristem.

When the shoot apex of a *Pharbitis* seedling is treated with gibberellin, an increase in cell division is observed in the subapical tissue within a short time after the treatment, and the length of the cells in the zone of active cell division is short. In *Erigeron* similar facts are also observed. There are, however, two noticeable differences between the reactions in *Pharbitis* and *Erigeron* ; in *Pharbitis*, the number of cell divisions in subapical tissue reaches a maximum 18 hours after the treatment, while the frequency of cell divisions increases until the end of the experiment in *Erigeron*. A large number of late prophase figures are observed in *Erigeron*, but it can not be distinguished from other phases in *Pharbitis*. From these facts it is easily understood that in the treated *Pharbitis* the rate of stem elongation decreases with the lapse of time, while in the treated *Erigeron* the elongation rate does not change for a long time.

In the second internode in *Pharbitis* ("Kidachi", a dwarf strain), the ratios of the stem length to the cell length of the treated plants are about ten times as high as those of the controls. Thus the cell number in the internode becomes about ten times as great as that found in the controls, indicating that the cells of the treated plants divided three or more times than those of the controls before they attained a permanent state. The leaf in *Oryza*, a classical material for bioassey of gibberellin, elongates markedly due to gibberellin treatment ; in the second leaf sheath the ratio of leaf length to cell length of the treated plants is 2.2 times as high as that of the control. Those facts indicate that the pronounced elongation of the organs in response to gibberellin is mainly caused by an increase in the cell number.

SACHS, BRETZ and LANG (51) pointed out in a cold-requiring biennial *Hyoscyamus*, and *Samolus*, a long day plant, that the frequency of cell divisions increases greatly in the subapical meristem. The results in *Pharbitis* ("Kidachi, a dwarf strain) or *Erigeron* (a cold-requiring biennial plant) show also an increase in the frequency of cell divisions in the subapical meristem within a short time after gibberellin treatment.

Gibberellin also accelerates hypocotyl elongation in *Pharbitis*, which is caused by an increase in cell length, but not by cell number. In gibberellin treated *Erigeron*, cells of young tissue in the portion within 2 mm from the apex are not longer than those of the control, while cells in aged tissue in the lower portion are longer than those of the control. In these cases, the elongation of the organs

is mainly caused by cell elongation.

The effects of gibberellin on cells differ according to the age of the cells. Young meristematic cells retain their dividing activity for a fairly long time. Aged cells elongate but do not recover the ability to divide.

In treated *Pharbitis* the followings are observed ; 1) high frequency of cell divisions in the subapical meristem, 2) increase in the number of leaf primordia; the plastochron age is promoted in the gibberellin treated plant. and 3) growth promotion of the differentiated organs. Gibberellin accelerates growth of the entire shoot.

The effect of gibberellin on root growth is inhibitory ; the root weight of the treated plant does not increase so remarkably as that of the control, in *Pharbitis*. In *Oryza*, both total length and weight of fibrous roots decrease with the increase in the applied gibberellin concentrations. The number of cell divisions in the root tip, however, are increased slightly by the treatments with higher gibberellin concentrations (10 and 100 ppm) in early stages, and decrease suddenly in later stages. With lower concentrations the roots are scarcely affected.

Although it was known in the previous works (4, 9, 15, 22, 61) that gibberellin inhibits root growth, MURAKAMI (32) has reported that in rice seedlings the root becomes longer than the control immediately after gibberellin treatment and becomes shorter subsequently. This may correspond to temporary activation of cell divisions in the root tip.

In *Pharbitis*, the aerial and underground parts of the treated plants are heavier than those of the control in both fresh and dry weights. As stem growth is promoted more remarkably than root growth by gibberellin, therefore, the shoot-root ratios of treated plants are higher than those of the controls, especially in water culture. In *Oryza*, shoot growth is promoted but root growth is inhibited, resulting in a remarkable increase in the shoot-root ratio.

In *Pharbitis*, the cambium activity in the stem decreases remarkably and the cell-wall thickening accompanied by lignification in the xylem and pericycle is accelerated conspicuously by gibberellin treatment.

When one side of the hypocotyl of a young seedling of *Pharbitis* is treated with gibberellin, the hypocotyl elongates pronouncedly without curvature, thereby differing in its response to IAA treatment. If gibberellin is applied to the first internode, the acceleration of cell-wall thickening is observed in the second internode as well as in the hypocotyl. These facts indicate that the translocation of gibberellin is not polar.

Although gibberellin has many effects different from IAA as reported by BRIAN *et al.* (5, 7), the decrease in cambial activity and the promotion of cell-wall thick-

ening with lignification are also the specific actions of gibberellin.

Summary

1) Studies on the physiological effects of gibberellin on three plant species with different life forms are reported here. They are a) the normal and dwarf strains of *Pharbitis nil*, b) a dwarf strain of *Oryza sativa*, and c) *Erigeron annuus*, a cold requiring biennial plant.

2) The growth of stem and leaf in *Pharbitis* and *Oryza* is promoted pronouncedly by the application of gibberellin. The effects are more remarkable in the dwarf strains and are proportional to the concentrations of gibberellin applied. The acceleration of the growth by gibberellin is mainly caused by an increase in cell number and slightly by cell elongation.

3) The differentiation and development of leaf primordia in *Pharbitis* are accelerated by gibberellin pronouncedly, so that the plastochron age is promoted remarkably.

4) A large number of cell divisions are observed in the shoot apex in the treated plants of *Pharbitis* and *Erigeron* as compared with the controls. In the hypocotyl, cells elongate but the number of cells does not increase. Consequently, it can be said that the ability of cells to divide in young tissues continues for a fairly long time, but cells in aged tissue elongate due to the application of gibberellin.

5) In *Oryza*, the number of cell divisions in the root tip reaches a maximum immediately after the treatment. The fibrous roots in the treated plants are fewer in number than those found in the controls.

6) Gibberellin causes an increase of fresh and dry weights in aerial parts, and a decrease of each in underground parts. Therefore, the shoot-root ratio of treated plants becomes higher. Consequently, it is pointed out that gibberellin generally accelerates the growth of aerial parts and inhibits the growth of underground parts.

7) By gibberellin treatment, cell-wall thickening with lignification in the xylem and pericycle is accelerated, but cambial activity decreases. It seems a plant treated with gibberellin quickly changes its form from juvenile to adult.

8) Gibberellin causes flowering in *Erigeron* without exposure to low temperature under long day conditions. This means that gibberellin replaces the low temperature requirement for flowering.

9) Gibberellin has different physiological effects than IAA ; that is, gibberellin decreases in cambial activity, but promotes the cell-wall thickening and lignification in the xylem and pericycle.

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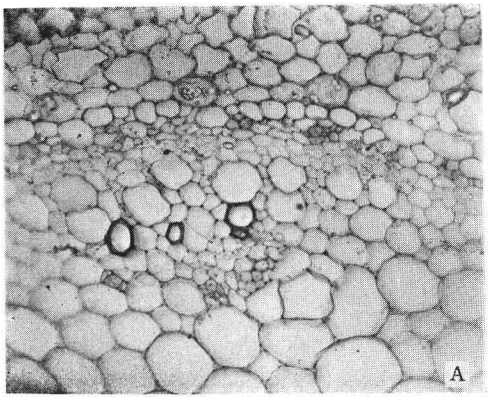
Plate I.

Transection through the vascular region in the stem of *Pharbitis nil.* A, cell-wall thickening of a few vessels in the primary xylem on the day of gibberellin treatment (three days after sowing). B, development of cell-wall thickening in the cells in one or two layers in the secondary xylem sampled four days after treatment. C, cell-wall thickening in pericycle cells, taken seven days after treatment. D, differentiation of large vessels at the side of the minor axis of the stem, ten days after treatment. E, control (no treatment), thirteen days after sowing, showing active cambium and cell-wall thickening in the metaxylem. F, control (no treatment), taken 32 days after sowing. (A–E, ×280 ; F, ×80).

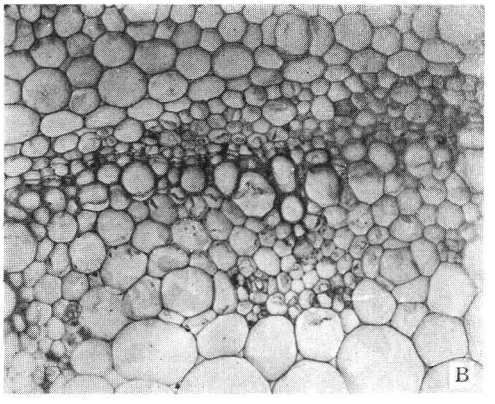
Plate II.

A, B and C ; longitudinal sections of the shoot apex of *Erigeron annuus,* taken 96 hours after treatment. A, control (no treatment). B, treated with gibberellin and kept under long day conditions. C, treated with and kept under short day conditions. D, E and F, *Erigeron annuus.* D, right, kept under long day conditions and treated with gibberellin, produced flowers on elongated stem ; left, control kept under long day conditions and not treated. E, right, kept under short day conditions and treated with gibberellin, showed the recovery of the rosette form ; left, control, kept under short day conditions and not treated. F, the plant in aerial rosette form in photograph E. G, effect of gibberellin (left) or IAA (right) on hypocotyl of *Pharbitis nil* ; middle, control. H, abnormal leaves produced in an *Erigeron* plant treated with gibberellin.

Plate I.



×280



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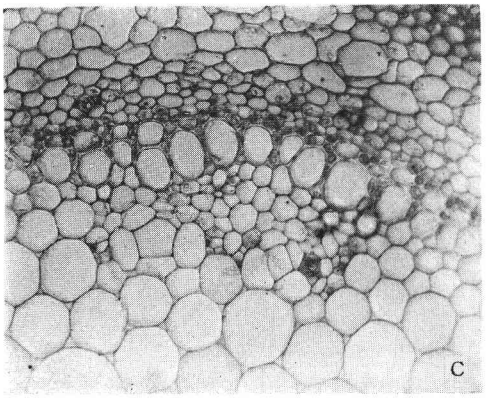

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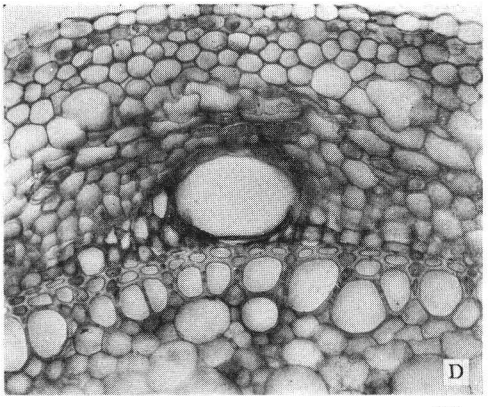

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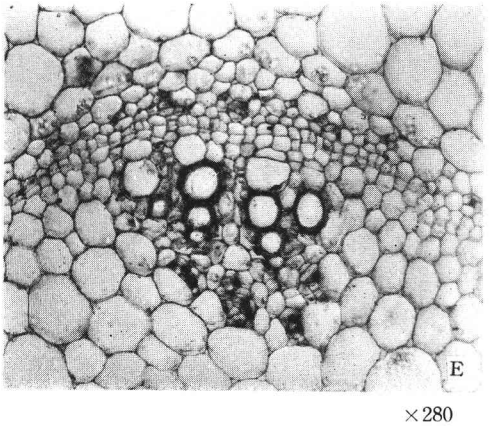

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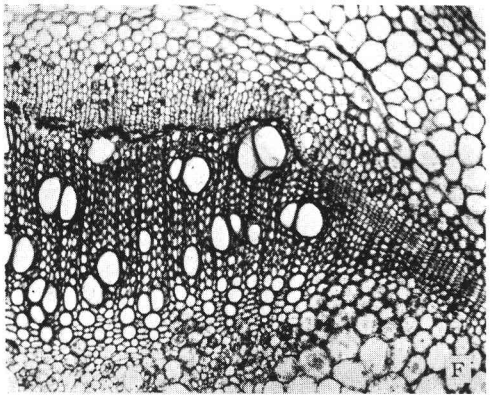

×80

Plate II.