FRUIT-INDUCED & APICAL SENESCENCE IN PISUM SATIVUM L.[1,2]

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Annual plants normally grow vegetatively for a limited time, flower, fruit, and then die. Rapid stem elongation of the Alaska pea plant continues until several fruit have begun development. During fruit development growth slows, and has stopped by the time fruit development is complete. Death of the vegetative parts soon follows (cf. 20).

It was Reichart (17) apparently, who first showed that flowering and fruiting are antagonistic to vegetative growth. He found that when flower buds were removed from the Vienna wall flower, normally an annual plant, this plant would continue vegetative growth for two or more years. When flowers were allowed to develop, the plant would rapidly set fruit and die. Numerous workers have substantiated and extended these results with other species (e.g. 6, 8, 9, 11, 12, 13, 19, etc.).

Various lines of evidence suggest that fruit may not act simply by diverting organic reserves from vegetative growth. Rather, the reproductive structures appear to cause death of the plants by some other mechanism.

Preliminary experiments indicated that periodic gibberellin treatments would substantially delay senescence in the Alaska pea. Further experiments were undertaken to study in greater detail the effect of gibberellin and fruiting on senescence of this plant. The evidence presented here indicates that the effect of gibberellin on senescence is indirect. Further experiments revealed a separate apical senescence in Pisum. The nature of this senescence is described.

Senescence will be considered here to mean those progressive, deleterious changes common to all organisms of a given species which ultimately lead to the death of the organism (18).

METHODS & MATERIALS

Plants of *Pisum sativum* L., var. Alaska, and in one experiment var. Alderman Improved (Ferry-Morse Seed Co., Los Angeles) were grown in the Earhart and Campbell Plant Research Laboratories as described previously (7).

Seeds were soaked in deionized water for 4 to 5 hours, then several seeds were sown in each of a number of 0.9 liter plastic containers in a 50-50 volume mixture of Vermiculite (expanded mica) and crushed rock. After germination for 4 days at 26° C in darkness, all except two vigorous seedlings were discarded from each pot. The two remaining seedlings were immediately moved to the appropriate greenhouse room and experimental treatments were begun. The plants were watered three times weekly with Hoagland's nutrient solution and with deionized water as required. The plants were grown throughout the year under solar radiation with the photoperiod extended to 16 hours by incandescent illumination. Substantial differences in growth rates were observed in the different experiments conducted during different seasons of the year. These effects can be ascribed principally to differences in solar radiation.

Growing temperatures are expressed as: day temperature (0800-1600 hr)/night temperature (1600-0800 hr). High growing temperatures (30°/24° C) were used when practical as a matter of convenience. With this temperature regimen, plants mature rapidly, are short, and easy to handle. The results indicate the same factor limits growth at high and at more moderate temperatures. Since plants grown at lower temperatures can be grafted much more successfully, plants for grafting experiments were always grown at 23°/17° C.

Gibberellin A_3 (Merck & Co., Rahway, N. J. & Eli Lilly Co., Indianapolis, Ind.) was applied where indicated as a single 4 μl ethanolic drop to the stem apex or to the youngest leaf. Three micrograms of gibberellin A_3 were applied weekly to each plant, beginning the day the plants were transferred to the greenhouse and terminating when growth had ceased. This gibberellin treatment is saturating so far as stem growth is concerned. Indole-3-acetic acid (IAA) and kinetin treatments were applied in a similar manner.

For sprays, solutions of 2 % sucrose, 1 % casein hydrolysate (Nutritional Biochemical Co., enzymatically hydrolyzed) and 1×10^{-3} M cobaltous chloride were made up in distilled water with 0.1 % Tween–20 (polyoxyethylene sorbitan monolaurate, a detergent produced by Atlas Powder Co.). Spraying was repeated twice weekly from germination until growth had ceased. Controls were sprayed with Tween–20 solution on the same schedule.

When root temperatures were to be different from top temperatures, seeds were planted directly in 1-gallon glazed crocks in which aluminum water coils had been placed previously. The seeds were germinated in the dark at 26° C and moved to the greenhouse rooms as usual. Thus, germination occurred under uniform conditions and temperature treatments were started only after the epicotyls had emerged. From the time the plants were moved to the greenhouses, water at the desired temperature was passed

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continuously through the coils. Control plants in this experiment were planted in identical crocks with coils in place but with no water circulation. Temperatures within the crocks were checked periodically with mercury thermometers which had been placed in the crocks at the time of planting.

Grafting was done by the method of Paton and Barber (16). Young (2-3 weeks old) plants were cut diagonally through the uppermost extended internode and the cut trimmed to a wedge shape. Older plants were cut directly through a node, since nodal tissue has been found to be more amenable to grafting in more mature plants (Highkin, unpublished). The sharpened scion was inserted into a verticle slit cut in the stock and held in place with a small rubber band. The grafts were protected from desiccation either with cotton, moistened periodically with water, or with wax of low melting temperature (50 % beewax-50 % paraffin oil). The relative success of cotton and wax-protected grafts varied. Overall success in grafting old plants ranged from 50 to 90 %. Side-grafts were made in essentially the same manner except that the stock stem was slit diagonally downward to about a third of the way through the stem.

When comparing the influence of red and far-red radiation the following sources were used. Red radiation was provided by filtering the radiation from six 8-foot pink fluorescent tubes through red cellulose acetate, and far-red by filtering the radiation from eight 40-watt Lumiline incandescent tubes through blue cellulose acetate. Red and far-red irradiation was given for 4 hours daily, immediately following the 8-hour daily light period. Intensity of the red source was approximately 300 μwatt · cm^{-2} and of the far-red 140 μwatt · cm^{-2} (in the far-red region), as measured with an Eldorado photometer equipped with a 1P29 RCA phototube (type S-3 surface). The photometer was calibrated at various wavelengths against the known output of the monochrometer of a Beckman DU spectrophotometer.

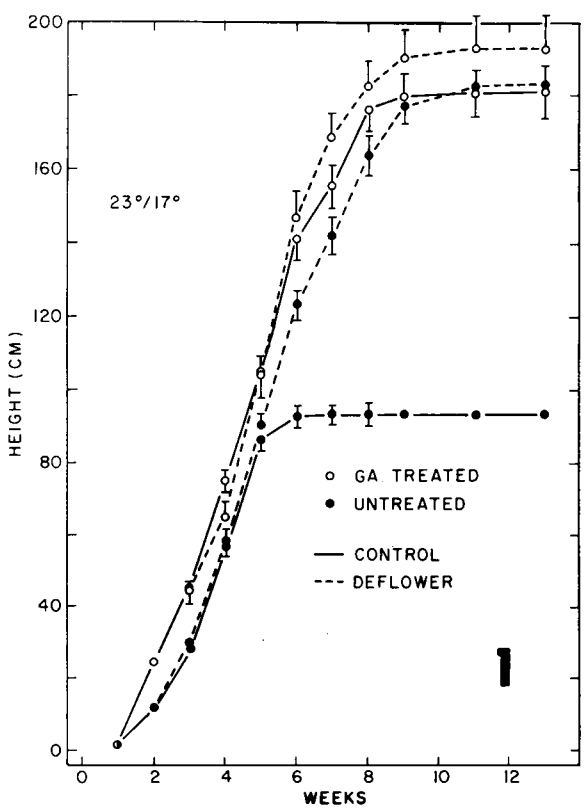


FIG. 1. Growth of Alaska peas in response to deflowering and gibberellin A_3 treatments. Control plants were allowed to develop normal fruit.

Height measurements were made weekly from germination until growth had ceased at maturity. When a plant was noticeably injured it was discarded and its previous measurements were expurgated from the data. Except where the influence of flowers and fruits was investigated all flowers were removed as soon as they became readily visible. Where statistical limits are indicated they express the standard deviation of the mean. Normally 12 plants were used per treatment.

EXPERIMENTAL RESULTS

I. FRUIT-INDUCED SENESCENCE. *Effect of deflowering & gibberellin treatments on growth*: Since fruit development has already been shown to cause cessation of vegetative growth in other species, the influence of developing fruit on stem growth was investigated in the pea plant. Growth responses to periodic applications of gibberellin A_3 were also investigated. Results of a typical experiment are presented in figure 1. Removal of flowers as they formed did not influence growth rate but prolonged the period of active stem growth for several weeks. Gibberellin treatments caused an initial promotion of stem growth (due to prevention of light inhibition) but, generally, did not affect maximum growth rate, in agreement with previous reports (7). Applied gibberellin, however, prolonged the period of active vegetative growth nearly to the same extent as removal of flowers. Total vegetative growth of plants which received either of these treatments was almost as great as that of plants which were both deflowered and treated with gibberellin.

The rate of appearance of flowers was almost completely unaffected by gibberellin treatment. However, the first several flowers of gibberellin-treated plants shriveled and abscissed while on non-treated plants these first flowers set normal fruit.

Influence of the various treatments on final dry weight of mature plants is shown in table I. Deflowering resulted in a marked increase in final dry weight, as expected. Gibberellin treatments were found to cause a similar but smaller increase. These increases in dry weight are presumably due to the increased life span. A combination of deflowering and gibberellin treatments resulted in somewhat less total dry weight accumulation than deflowering alone. The reason for this decreased weight is not clear.

The average number of nodes and flowers formed per plant is included in table I. It may be seen that increased stem length was due primarily to an increase

TABLE I

EFFECT OF DEFLOWERING & OF APPLIED GIBBERELLIN ON ALASKA PEA PLANTS

TREATMENT	DRY WEIGHT PER PLANT (g)			AVG. NO. OF NODES	AVG. NO. OF FLOWERS
	VEGETATIVE PARTS	FRUIT	TOTAL TOPS		
Control	0.9 ±0.1	1.7 ± 0.2	2.6 ± 0.2	12 ± 0.5	2.8
GA-treated	2.2 ± 0.3	1.9 ± 0.2	4.2 ± 0.5	22 ± 1.2	12.6
Deflowered	5.9 ± 0.5	...	5.9 ± 0.5	24 ± 0.4	14.8
Deflowered & GA-treated	3.9 ± 0.4	...	3.9 ± 0.4	23 ± 0.9	10.9

Plants were grown at 23°/17° C. Details of treatments are described in the text. The plants were harvested when stem elongation had completely ceased.

in number of nodes formed. The increased number of nodes was the result of the prolonged growth period rather than an increase in rate of node formation. The substantially greater number of nodes formed in both the gibberellin and deflowering treatments accounts for the greater number of flowers formed as a result of these treatments. A flower is initiated at nearly every node formed after the first flowering node.

Gibberellin treatments were shown above to substitute for the effect of deflowering on stem growth. Since gibberellin treatments have been found here to markedly delay fruit-set it may be that the principal influence of gibberellin on prolonging stem growth is due to gibberellin-induced delay in fruit-set. In a separate experiment the mean fruit-bearing node was determined for gibberellin-treated and untreated plants. The average node bearing mature fruit on the control plants was 11.9 ± 0.2, while on the gibberellin-treated plants it was 17.2 ± 0.7. Thus, the bulk of the fruit was formed at a much later time on gibberellin-treated plants, supporting the suggestion that gibberellin delays senescence by delaying fruit-set.

Effect of developmental state of reproductive structures on growth: The presence of developing flowers and fruits results in an early termination of vegetative growth. The developmental stage of the reproductive structures at which they exert an effect on stem growth has also been studied. Flowers and young fruit were removed at several stages of development and the resultant effects on vegetative growth compared. The reproductive structures were removed: A, when the first white petals became visible in the flower bud (referred to as "first white petals"); B, when the flower petals had begun to dry (drying petals), or, C, when the young pods were 1 to 2 cm long. Control plants were allowed to develop mature fruit. The results are presented in table II. As in previous experiments, deflowering markedly prolonged stem growth, shown as an increase in final stem height. Removal of the reproductive structures resulted in prolonged stem growth of a similar magnitude regardless of whether the reproductive structures were removed when they reached the stage of young flowers, older flowers, or young fruit. These results indicate that the effect of the reproductive structures on vegetative growth is initiated by older fruit rather than through some action elicited by the flowers at synapsis or fertilization. This conclusion is consistent with that of Leopold et al (see Discussion). The slight increase in stem height found when flowers were allowed to remain until drying petals is not significant in this experiment. However, this trend may well bear further investigation.

Effect of seed development on growth: A further effect of applied gibberellin was found in an examination of the fruit formed on gibberellin-treated plants. While total fruit weight was not substantially decreased as a result of gibberellin treatment, the character of the fruit was markedly changed. In three

TABLE II

EFFECT OF REMOVING REPRODUCTIVE STRUCTURES AT DIFFERENT STAGES OF FLOWER & FRUIT DEVELOPMENT ON GROWTH OF ALASKA PEA PLANTS

DEVELOPMENT OF FLOWERS OR FRUIT WHEN REMOVED	DRY WEIGHT PER PLANT (g)			NO. OF NODES	HEIGHT AT MATURITY (cm)
	VEGETATIVE PARTS	FRUIT	TOTAL TOP WT		
Control (fruit developed to maturity)	0.4	0.6	1.0 ± 0.1	12 ± 0.4	62.3 ± 3.3
First white petals	1.9	...	1.9 ± 0.3	21 ± 0.5	101.0 ± 1.6
Drying petals	2.0	...	2.0 ± 0.3	22 ± 0.9	108.2 ± 6.9
Defruit (1–2 cm)	1.8	...	1.8 ± 0.3	22 ± 1.0	99.9 ± 7.1

Plants were grown at 30°/24° C. Control plants were allowed to form and develop fruit normally. In the other treatments reproductive structures were removed at various stages of development as indicated.

experiments seeds from fruit of untreated plants constituted an average of 57 % of total fruit weight, while on gibberellin-treated plants seeds constituted, on an average, only 14 % of the fruit weight. These results suggest that seed development rather than carpel growth might be specifically responsible for cessation of stem growth in the pea.

In order to test the influence of developing seeds directly, the fruits were allowed to develop normally, but the young seeds were cut loose from the carpel wall with a fine knife when the carpels had reached full size. Thus, the carpels expanded fully but seed development was arrested at an early stage. Results of a typical experiment are presented in table III. It is clear that prevention of seed development prolonged vegetative growth almost as long as did removal of flowers. It appears that the fruit causes senescence principally through an effect by the developing seeds. Plants whose fruit received comparable injury by inserting the knife blade through the carpel wall without detaching the seeds, behaved in a manner identical to that of the seeded controls.

Developing seeds are known to secrete auxin. In order to determine whether production of auxin by the seeds was partly or wholly responsible for the influence of the seeds on vegetative growth, IAA was applied periodically to deseeded fruit. Four micrograms of IAA (in ethanol) were applied to each deseeded fruit twice weekly until two subsequent fruits had developed on the plant. The treatments were continued on newly developing fruit until growth ceased. No effect of the IAA treatment could be observed. Deseeding prolonged vegetative growth to the same extent whether IAA was applied or not.

II. Apical Senescence. Deflowering and gibberellin treatments resulted in prolonged vegetative growth. However, these treatments were effective for only a limited time (or for only a limited amount of growth). Stem growth ultimately ceased regardless of any experimental treatment so far tested. In order to identify the factor responsible for the ultimate cessation of stem growth, experiments were undertaken to determine what portion of the plant first ceases its normal function.

Morphology of apex when stem growth ceases: Termination of stem growth, in the absence of fruit, occurred with the formation of a final node consisting of a typical bract and leaf, and a single flower. No indication of the stem apex remained (fig 2). The obvious conclusion is that this is a terminal flower and we are dealing simply with determinate growth. However, this is not the case in a physiological sense. The total number of nodes (& number of flowering nodes) formed before growth terminates may be readily changed by temperature (table IV), and light treatments (table VI). Thus, the factor which determines whether stem growth shall continue or whether the remaining apical meristematic tissue shall be invested entirely in a single flower is the factor which we are investigating. An important fact verified by figure 2 is that the upper portion of the plant was still green and apparently completely healthy at the time stem elongation ceased. In deflowered plants the leaves usually turned brown and died 1 to 2 weeks or more after stem elongation had ceased.

Effect of root & shoot temperatures on growth: Growing the aerial portions and roots of the plant at different temperatures could provide evidence indicating which of these plant parts first ceases normal functions, and thus causes growth to cease. At 30°/24° C stem growth is rapid but the plant lives for only a relatively short time. At lower temperatures (e.g., 17°/11° C) growth is somewhat slower but continues for about 2-3 times as long and final stem height is much greater (20, 7). In the following experiment the aerial portions of certain plants were grown at either 30°/24° C or 17°/11° C while the roots were maintained at either 30° or 17° C (without day-night fluctuations). Flowers were removed regularly from all plants. Rates of stem growth in such

Table III

Influence of Developing Seeds on Vegetative Growth of Alaska Peas

	Control	Deseeded	Deflowered
Final stem height (cm)	68.1 ± 1.7	105.8 ± 3.7	124.4 ± 5.4
No. nodes/plant	12.9	24.1	26.5
" laterals/plant*	0	12.3	17.4
" flowers/plant	4.8	14.0	15.0
" pods/plant	1.4	5.5	...
Dry wt (g/plant)			
Vegetative parts	0.35	1.42	2.69
Pods	0.62	0.99	...
Seeds	0.49	...	...
Total fruit	1.11	0.99	...

Plants were grown at 26°/20° C. On control plants the fruit was allowed to develop normally, on deseeded plants the young seeds were detached from the carpel wall at the time the fruit had reached full size. Measurements were made after all plants had died.

* Laterals were removed as soon as possible after growth began.

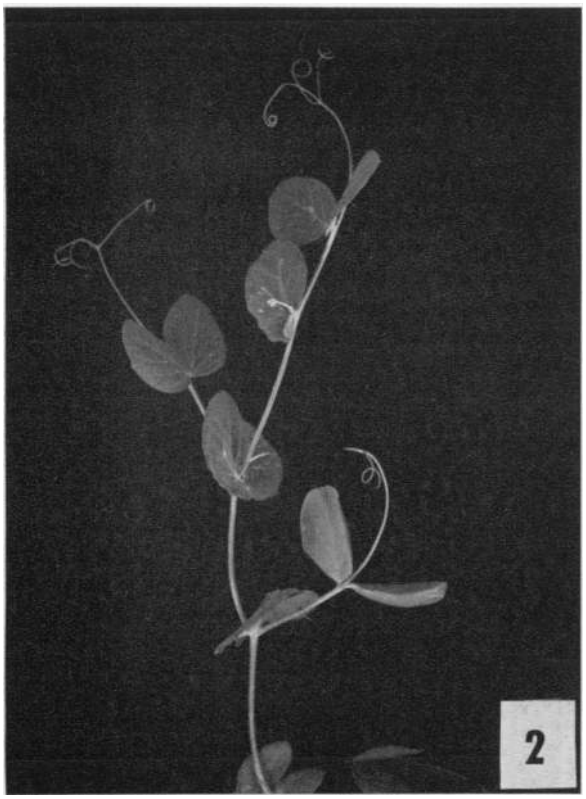


FIG. 2. Appearance of the stem apex of a deflowered Alaska pea plant shortly after differentiation of the final node. The nearest bract has been removed to show the final flower and the absence of an apical meristem. Earlier flowers were removed as they formed.

an experiment are presented in figure 3. It is obvious that rate and extent of stem elongation were determined almost entirely by the temperature of aerial portions of the plant. The influence of root and top temperatures on weight of roots and tops and on node number is shown in table IV. It may be seen that root temperatures had a substantially greater influence on dry weight than on stem elongation. Roots, then, do not limit stem elongation under the conditions used here.

This experiment also demonstrates the influence of two growing temperatures on duration of stem growth and number of nodes formed prior to senescence. At high temperatures growth had ceased and the plants were dead at harvest. At lower temperatures more nodes had been formed per plant at the time of harvest and the plants at low temperatures were still growing vigorously. It must be expected that the plants grown at 17° C would have continued growth for a substantially greater time and formed many more nodes prior to senescence if growth had been allowed to continue. The visual appearance of the stem apex makes it possible to anticipate senescence some time in advance of growth cessation. No indications of future senescence could be observed in these plants at the time of harvest.

Effect of sucrose & other chemical treatments on growth: In an effort to learn whether cessation of stem growth is related to a deficiency occurring in the photosynthetic apparatus or in nitrogen nutrition, deflowered plants growing at 30°/24° C were sprayed twice weekly with either 2 % sucrose or 1 % casein hydrolysate solution. Plants sprayed with 0.1 % Tween–20 solution served as controls. Sucrose and amino acids had almost no effect on growth, and neither affected time of senescence. The negative results here make it impossible to determine whether sucrose and amino acids were not limiting for growth or whether they did not enter the plants in sufficient amounts to influence growth. Ascorbic acid sprays also were found to be without growth-promoting effects under these conditions (2). Indoleacetic acid, applied weekly as ethanolic drops at doses of 4.0, 0.04, or 0.0004 μg per plant, was without effect on either growth rate or time of senescence, except that the highest dose resulted in a slight growth inhibition.

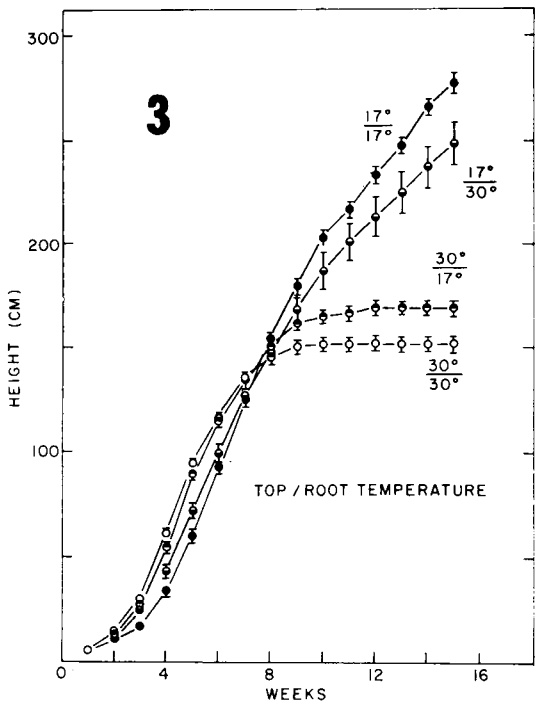
In further experiments at a growing temperature of 26°/20° C, 2 % sucrose was sprayed twice weekly onto plants which were allowed to set and develop fruit normally. Sucrose sprays were completely ineffective also in delaying fruit-induced senescence. Semi-weekly sprays with 1×10^{-3} M cobaltous chloride substantially delayed decoloration and death of the leaves, but had no effect on growth or time of either fruit-induced or apical senescence. Weekly

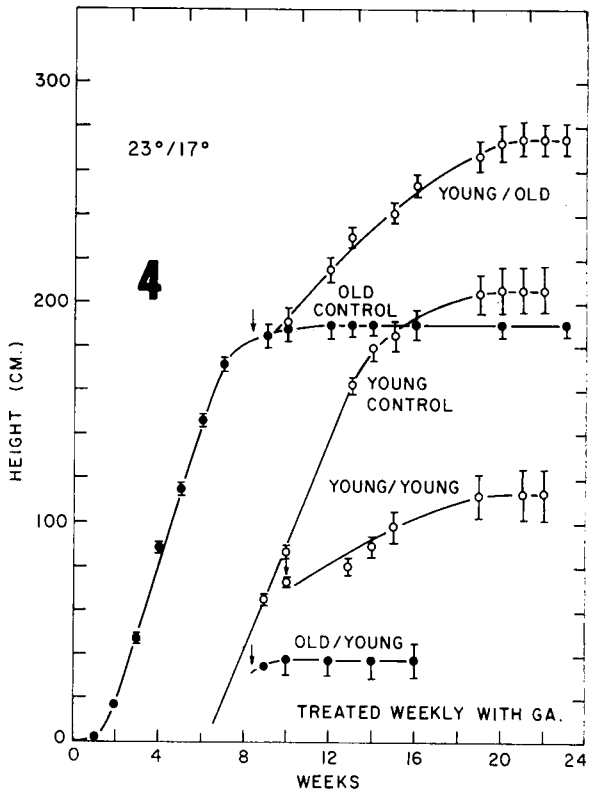
TABLE IV

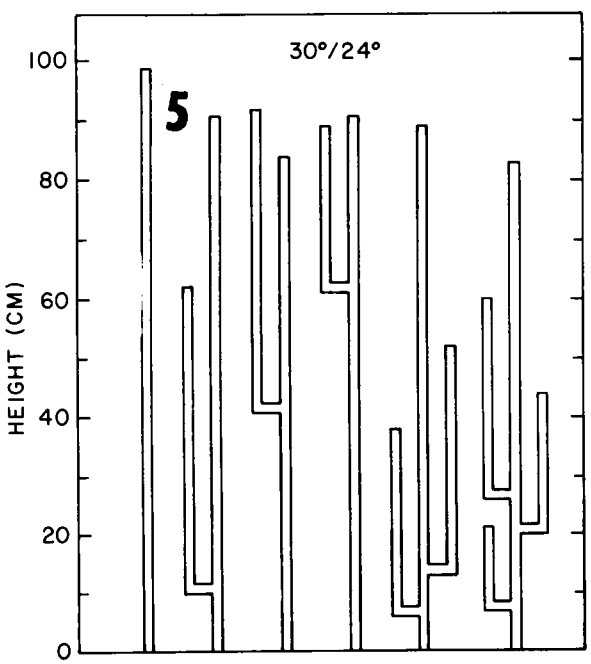
EFFECT OF ROOT & SHOOT TEMPERATURE ON GROWTH OF ALASKA PEAS AFTER 15 WEEKS GROWTH

TEMP. (C) (TOP-ROOT)	STEM HT. (cm)	AVG. DRY WT/PLANT (g)		NO. NODES
		TOP WT	ROOT WT	
17°/17°	277 ± 5	34.8 ± 2.7	9.2	34 ± 0.6
17°/30°	250 ± 10	21.3 ± 2.0	1.6	34 ± 0.8
30°/17°	172 ± 2	8.7 ± 0.4	1.7	32 ± 2.0
30°/30°	153 ± 3	6.2 ± 0.4	0.9	27 ± 0.8

Final measurements were made after 15 weeks, when plants with tops at 30° C had ceased growth, while those at 17° C were still growing vigorously. All plants were treated weekly with gibberellin A_3 and deflowered periodically.







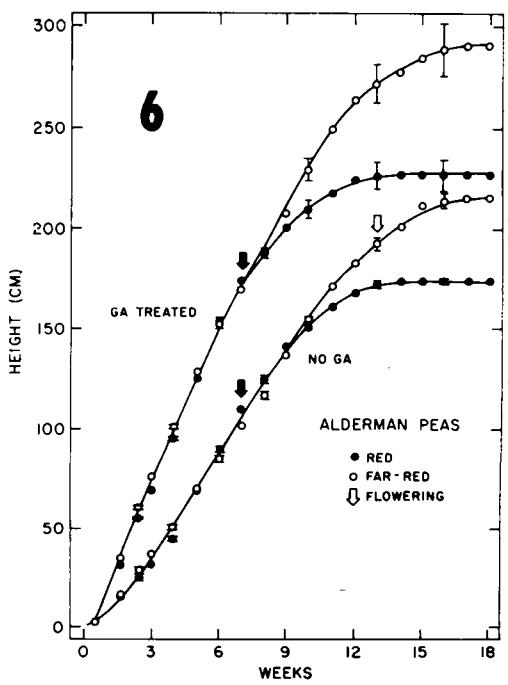


FIG. 3. The influence of root and top temperatures on stem growth of Alaska pea. Plants grown with tops at 17° C were still growing vigorously at the time of harvest while those whose top temperature was maintained at 30° C, were mature.

FIG. 4. Growth of Alaska peas illustrating the growth of young or old stem tips grafted onto young or old plants. The labels indicate scion/stock.

TABLE V

INFLUENCE OF SIDE-GRAFTED STEM TIPS ON STEM GROWTH & SENESCENCE

STOCK	ORIGINAL APEX	AGE OF GRAFTED APEX	HEIGHT OF STOCK AT TIME OF GRAFTING	GROWTH OF STOCK STEM AFTER GRAFTING	GROWTH OF GRAFTED SHOOT
Old plants	Control		112 cm*	17 ± 4.3 cm	
	Removed	Young	107		42 ± 4.7 cm
	Present	Young	101	16 ± 3.4	18 ± 6.5
Young plants	Control		36*	159 ± 18.0	
	Removed	Young	21		12 ± 2.0
	Removed	Old	23		1 ± 0.5
	Present	Young	17	113 ± 25	20 ± 9.0
	Present	Old	25	117 ± 8.4	2 ± 0.5

Plants were grown at 23°/17° C under 16 hour photoperiod. All plants were regularly deflowered.
* For the ungrafted controls, height at the time grafts were made is shown.

treatments with ethanolic solutions of kinetin at 100 and 1,000 ppm were also without effect.

Grafting experiments: A number of grafting experiments were conducted to determine if the stem apex itself is primarily responsible for cessation of apical growth. Stem tips from young plants were grafted to the tops of older stems and, reciprocally, the stem tips from the older plants were grafted to seedling plants. Results of several such experiments have proven quite conclusive.

In a typical experiment, stem tips were cut from nearly mature plants (i.e., plants which could be expected to cease stem growth within 2-3 weeks). Young (2-week-old) stem apices were grafted to the tops of the old stems in their places. The young stem tips resumed growth after 10 to 12 days and grew at a moderately rapid rate (fig 4). They continued growth for 8 to 10 weeks beyond the time the old plants normally matured. It may be seen from the figure that young tips grafted to old plants grew as fast and for as long a time as young tips grafted back to young plants. While growth rate of grafted tips was less than that of young intact plants, the grafted tips continued growth for fully as long as did the intact plants.

Tips from old plants may be grafted to young seedlings with good success, judged by the strength of tissue union and the fact that grafted tips, with several leaves present, remain completely turgid and green for several weeks. However, no more than 1 to 2 cm growth occurred in these plants. Allowing 1 to 2 weeks for tissue union to occur, these old tips grew only as much as stem tips left intact on the old plant. These grafting experiments have been repeated with plants treated periodically with gibberellin and with untreated plants. The results have always been substantially the same as reported above.

The old plant, then, was fully capable of supporting stem growth for a much longer time and to a considerably greater total height than normally occurs. However, the old stem apex did not continue growth even under what would appear to be optimum conditions, i.e., when grafted onto a young, vigorously growing plant. Thus, cessation of growth of the pea stem in the absence of developing fruit is due to a decline in vigor of the growing point or immediately subjacent tissue.

Non-transmission of controlling factor: Further grafting experiments were undertaken to test for transmission of a possible juvenile or senescence factor. Nearly mature stem apices were side-grafted to stems of young plants about two to four centimeters below the tip, leaving the stem tip of the stock intact (table V). The old tip grafted to a young seedling had virtually no effect on growth of the seedling. Furthermore, young tips side-grafted near the apex of old plants had no effect on maturation of the old stem tips. Growth of young, grafted tips was initially inhibited by apical dominance of the old tips. As soon as the old tips ceased growth, the young side-grafted shoots resumed normal growth. In both cases, then, presence of a stem tip of a different age had no effect on growth of the intact plant. Thus, neither an aging factor nor a juvenile factor capable of being translocated across a graft union seems to be responsible for apical senescence in this plant.

Lateral stem growth: On plants which had been deflowered, lateral buds began growth shortly before

FIG. 5. Final length of main stem and laterals allowed to develop on deflowered Alaska peas. Both length of laterals and position of insertion on the main stem are to scale.

FIG. 6. Growth of Alderman Improved peas in response to gibberellin A_3 treatment and daily red or far-red irradiation. The arrows indicate the time of appearance of the first visible flowers. No flowers ever became visible on the gibberellin-treated, far-red irradiated plants. All flowers were removed as soon as they became visible.

growth of the main stem ceased. (No laterals grew out when fruit was allowed to develop.) Normally, in the experiments described here, these laterals were removed as soon as they began growth. In a separate experiment growth of selected laterals was followed to provide further information on the nature of apical senescence. After the laterals began to grow, plants were trimmed to the main stem and a single lateral branch originating near the bottom, middle, or top of the main axis. Other plants were allowed to develop either two or three laterals from near the base of the old stem. No other laterals were permitted to develop. The laterals continued growth for several weeks. Duration of growth of laterals was approximately equal to the duration of growth of the main stem, although growth rate was not as rapid as had been that of the main axis. Final height of the main stem and laterals is illustrated diagrammatically in figure 5. The average position of insertion of lateral shoots on the main stem, as well as their final length is represented to scale. Total growth of the laterals was about equal, whether the shoot originated near the base or middle of the old stem. Lateral shoots from near the top of the old stem grew less. While total growth of upper laterals was least, their rate of growth was the most rapid. Thus, they could hardly have been deficient in water or nutrients as a result of difficulties of translocation up the long stem. When two or three shoots were allowed to develop on a single plant, each shoot grew less than if only one lateral was allowed to develop. Apparently in this case competition for nutrients or water occurred.

Growth of lateral shoots, after the stem apex had ceased growth, confirmed the conclusions reached as a result of the grafting experiments. Cessation of growth of the stem apex is the result of some effect localized in the stem tip itself. Furthermore, growth of laterals indicates that meristematic cells within the old plant are capable of continued growth. Thus, not all the meristematic tissue of the stem becomes senescent at the same time.

III. Flowering in Relation to Apex Senescence. Certain biennial plants, and plants whose flowering is strictly controlled by photoperiod, will grow far longer than their usual life cycle when kept strictly vegetative (e.g. 3, 4, 5). In many of the cases studied, growth appeared to be capable of continuing indefinitely. The initiation of flowers, even though all flowers are removed as soon as they are formed, may have some physiological effect on the stem apex, causing the gradual loss of some essential capacity. Experiments described below indicate that the onset of flowering does not initiate or substantially promote apical senescence in Pisum.

Alaska and Alderman Improved varieties of pea were grown in 8 hours natural light per day, supplemented with either red (low intensity red fluorescent) or far-red (blue & far-red radiation from incandescent bulbs) radiation for 4 hours at the beginning of the daily dark period. Day temperature was 26° C and night temperature 23° C. All flowers were removed as they formed, and gibberellin treatments were included in each light condition. (Aqueous solutions of gibberellin were used on the Alderman peas since previously it had been found that ethanol, over a period of time, caused considerable injury to this variety.)

Results for the Alderman variety are presented in figure 6. The far-red treatment alone did not completely prevent flowering in this variety, but it did delay the onset of flowering for several weeks. The average first node to flower in far-red treated plants was 30.2 compared to 22.4 for red-treated plants. Weekly treatments with gibberellin had no effect on flowering time of red-treated plants, but completely prevented flowering of far-red-treated plants. In spite of the marked delay in flowering, senescence was only slightly delayed by far-red as compared to red treatment and took place regardless of whether or not flowering occurred.

An identical experiment with Alaska peas gave generally similar results (table VI). With Alaska peas, surprisingly, no flowering occurred on far-red irradiated plants regardless of whether or not they received gibberellin treatments. Senescence occurred at nearly the same time in all treatments, whether the plants had begun flowering or not. Neither red nor far-red irradiation had any effect on growth rate (tall peas are, apparently, virtually saturated with respect to endogenous gibberellin), but red irradiation resulted in a substantial increase in rate of node formation compared to far-red treatment. Plants irradiated

Table VI

Influence of Red/Far-Red Radiation & Gibberellin Treatments on Growth, Flowering, & Senescence of Alaska Pea

Treatment	Height at senescence (cm)	No. nodes at senescence	Age at senescence	1st Flowering node
Red	71.5 ± 3.4	22.6 ± 0.8	ca. 70 days	19.4 ± 0.7
Red + Ga	123.4 ± 2.2	22.0 ± 0.5	"	17.1 ± 0.3
Far-red	84.6 ± 3.3	17.4 ± 0.7	"	No flowers
Far-red + Ga	109.4 ± 3.4	15.6 ± 0.7	"	No flowers

Plants were grown at 26°/23° C under 8 hours solar radiation followed by 4 hours red or far-red radiation daily. All flowers were removed as they formed. Gibberellin was applied weekly to indicated treatments.

with far-red generally ceased growth before forming those nodes which, in red-treated plants, were the first to initiate flowers. No examination was made for the presence of microscopic flower primordia. However, the fact that in the Alaska variety senescence occurred in the far-red-irradiated plants before formation of what would have been the earliest flowering nodes indicates that no flower primordia were present in this variety, at least. Thus, apex senescence appears to be independent of flower initiation in this species.

Discussion

Two separate factors can result in termination of stem growth in the Alaska pea. The first factor, which may be expected to limit growth during normal plant development, is a result of some action of the developing fruit. As a result of fruit development the entire plant dies, with the exception of the seeds. Two general mechanisms can be envisaged: fruits may divert nutrient or hormonal materials from the rest of the plant, thus terminating stem growth; alternatively, fruits may secrete some factor which inhibits stem growth.

Leopold et al (6) have reported that the longer fruit is allowed to develop before removal, the greater its effect on leaf senescence; but they state that even when fruit is allowed to attain full size, its removal delays senescence. Our results suggest that young fruits have a relatively small influence on senescence in the pea as measured by cessation of stem growth. Removal of young fruit is nearly as effective as removal of flowers in delaying senescence. The greatest inhibitory effect appears to be exerted by developing seeds. When seed development was prevented surgically, stem growth was markedly prolonged, continuing nearly as long as that of deflowered plants. The seeds alone do not completely control vegetative growth, since in a number of species parthenocarpic fruit will also retard growth (e.g. 15). The effect of parthenocarpic fruit and deseeded fruit may well be due to diversion of major nutritional factors. However, the apparently specific influence of the seeds tends to support the conclusions of many workers that diversion of organic reserves cannot account completely for the effect of developing fruit on senescence. Leopold et al (loc. cit.) have shown that staminate plants of Spinacia die following maturation of the reproductive structures, just as pistillate plants die following fruit maturation. Pollen development could hardly be expected to divert sufficient reserves from the rest of the plant to cause death by starvation.

Periodic gibberellin treatments delay senescence in the pea by delaying fruit-set and inhibiting seed development. Whether or not gibberellin has any additional effect on fruit-induced senescence cannot be determined, since the effects of developing fruit and gibberellin are both terminated by apical senescence. Thus, whether or not their effects are additive cannot be established. It is clear that gibberellin has no effect on apical senescence.

Even when flowers are continually removed from the Alaska pea plant, or gibberellin applied, or both, growth of the main stem is extended only for a limited time. Under these conditions senescence of the stem apex is evidently determined by a second factor. It has been demonstrated here that cessation of growth of the stem apex results from some degenerative change localized within the apex itself. Neither common nutritional factors (sucrose, amino acids), nor the temperature condition under which the roots grow, significantly affect the onset of apical senescence. (Apical senescence is markedly delayed by lowering the growing temperature of the tops.) The roots and stem have been shown to be capable of supporting growth for at least several weeks beyond the onset of apical senescence. It is also apparent that whatever factor controls apical senescence is not readily translocated within the plant. Thus, neither a translocatable juvenile factor nor a senescence factor appears to be involved here. The important point here is not that some growth factor normally supplied by another part of the plant is missing from the apex, but that the capacity of the apex to produce some factor essential for its own continued growth has been lost. For example, the possibility that some nutrient factor becomes exhausted has been eliminated by the present experiments. Such a nutrient factor would be restored to old stem apices grafted to young plants.

Developing fruits have a systemic effect, completely inhibiting growth of all lateral buds as well as the main axis. Apical senescence, however, is localized to the degenerate tip, since other tissue in the stem is still capable of vigorous growth. Lateral buds arise from the apical meristem some time prior to senescence, when it is younger and has undergone fewer mitotic divisions. Perhaps the greater growth of lower lateral branches compared to laterals originating from the upper portion of the plant is a reflection of the origin of lower buds from younger meristems, that is, from meristematic cells which have previously undergone fewer total cell divisions. Degeneration of vegetative cells as a result of continued cell multiplication has been observed in various organisms (Lemna, 1; fungi, 10, & protozoa 14).

Many plants when kept strictly vegetative (for example, by appropriate photoperiodic treatment) apparently are able to continue growth far longer than comparable plants which form flowers, even if no fruits are allowed to develop on the flowering plants. This has been directly demonstrated only with spinach, but in this case a complete change in growth habit is associated with floral induction. In other cases, no flowering-deflowered control has been included. It has been possible to keep Alaska and Alderman Improved peas vegetative (non-flowering) by certain light and gibberellin treatments. Even though these plants remain vegetative, apical senescence occurs at almost the same time as in flowering (but deflowered) plants. Senescence of the apex, then, may occur even in the absence of the flowering condition.

The senescence problem studied here is distinct from the problem of leaf senescence studied by various workers. Leaf senescence occurs also in the Alaska pea, progressing from base to tip. However, all our observations indicate that leaf senescence is an independent phenomenon, with no direct relationship to the apical senescence studied here. As pointed out earlier, apical senescence is often complete while most of the leaves are still green and healthy. When the developing fruits cause senescence the leaves die at the same time as the stem apex. Developing fruit thus may have the effect of markedly hastening senescence of the leaves.

It is not known if the phenomenon of apical senescence is widespread among higher plants. Certainly many species do not seem to show comparable behavior. On the other hand, there are other species in which one or more lateral buds assume dominance after the main stem has made the early growth. In rosette plants, too, lateral tillers often become more vigorous than the primary apex. These cases may prove to be caused by a degeneration of the stem apex, comparable to the case of Pisum, reported here.

Summary

Alaska peas, like many other annual plants, normally die as a result of fruit development. Gibberellin treatments delayed senescence by delaying fruit formation. Detachment of the seeds was almost as effective as removing the entire fruit in prolonging vegetative growth. When fruit development was prevented by deflowering or by gibberellin treatments, growth of the main stem continued only for a limited time. Ultimately the stem apex differentiated into a final flower and leaf and the apical meristem disappeared.

The stem apices of young plants, grafted to the tips of old plants, grew as rapidly and for as long a time as when grafted back to a young plant. The old tips ceased growth even when grafted to a young plant. An old or young stem apex side-grafted to another plant did not substantially affect growth of the plant to which it was grafted.

When fruit was allowed to mature on the Alaska pea the entire plant died. However, when flowers were continually removed, and apical senescence occurred, numerous laterals began growth. When individual lateral branches were allowed to develop, their growth rate was less than that of the old stem but duration of growth was substantially as long. It is concluded that apical senescence is due to some degenerative change taking place within the apex itself and is not translocated within the plant.

Apparently, senescence of the apex is not directly related to the physiological onset of flowering. Apical senescence occurred even though no flowering took place.

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