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EFFECTS OF DAYLENGTH UPON GROWTH  
AND DEVELOPMENT OF SPINACH  
(*SPINACEA OLERACEA* L.)

by

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INTRODUCTION

The initiation of flower primordia in long day plants is promoted by the alternation of long days and short nights. This effect can be obtained by extending the main irradiation, consisting of a high light intensity, with a low light intensity so that only very little photosynthesis takes place. Such photoperiodic effects have been studied extensively.

Although less amply, the influence of daylength upon the vegetative growth has also been investigated. WITHROW and WITHROW (1940) have observed that spinach plants, grown under conditions favourable for flower formation (supplementary irradiation with a low intensity of red or yellow-green light) after one month of treatment had attained higher dry weights than such plants at short days. From other data available in literature, those of BONNER (1940) may be mentioned. He not only observed a positive effect of long days upon the dry weight of long day plants, but also upon that of short day plants (*e.g. Xanthium*), and intermediates (*Lycopersicum*).

Formerly, in this laboratory indications had been obtained that a supplementary irradiation at low light intensity after the main irradiation may cause considerably larger spinach plants. A further investigation of the effects of daylength upon both the vegetative growth, and the reproductive development of spinach plants has been made. The experiments to be described were already carried out in 1951, but various reasons so far prevented publication. Moreover, the interest of some of the observations may appear to have increased in connection with more recent data in literature.

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## MATERIAL AND METHODS

In these experiments spinach plants (*Spinacea oleracea* L.), variety Nobel, and to a limited extent variety Viroflay, have been used. The seeds were sown in fertile soil in wooden boxes of 60 × 30 cm area (Expts. 1, 2, 3), or in pots (Expt. 4). Until emergence the plants were in a greenhouse with an average temperature of 18°C; afterwards thinned, and transferred to a temperature controlled basement room, in which the experiments were performed.

All plants at different daylengths received the same basic irradiation during 12 hours of a 24 hour cycle. This has been realised by the use of eight 40 Watt daylight type fluorescent tubes, and four 15 Watt incandescent lamps, mounted on two side walls of each light cabinet (dimensions: 120 × 65 × 65 cm), yielding a light intensity of 15.000 ergs.cm<sup>-2</sup>.sec<sup>-1</sup> ( $\lambda < 700 \text{ m}\mu$ ) at the height of the plants. In experiments 3 and 4, however, the four incandescent lamps in each light cabinet were omitted. In order to obtain an equal distribution of the light intensity to the plants, the place of the boxes or pots in the light cabinets was alternated every two days.

The supplementary irradiation has been given directly after the main irradiation. In experiments 1 and 2 this irradiation was obtained by the four 15 Watt incandescent lamps yielding a light intensity of 1000 ergs.cm<sup>-2</sup>.sec<sup>-1</sup> ( $\lambda < 700 \text{ m}\mu$ ), and in experiments 3 and 4 with the aid of one fluorescent tube per light cabinet, mounted on a height of 1.50 m above the plants, and yielding a light intensity of 500 ergs.cm<sup>-2</sup>.sec<sup>-1</sup> ( $\lambda < 700 \text{ m}\mu$ ).

The temperature during the period of main irradiation amounted to 18°C ( $\pm 1^\circ\text{C}$ ), while for the rest of the 24 hour cycle a temperature of 15°C ( $\pm 1.5^\circ\text{C}$ ) was maintained. In experiments 3 and 4 these temperatures were 20°C ( $\pm 1^\circ\text{C}$ ), and 18°C ( $\pm 1^\circ\text{C}$ ) respectively. During the supplementary irradiation period the temperatures in the light cabinets did not rise above that of the then dark cabinet.

Part of the plants of each treatment was harvested at different intervals during the experimental period (Expts. 1, 2, 3). This involved further thinning of the plants at regular distances in the rows. The growing points of the harvested plants were examined, and the stem length and leaf dimensions were measured. Ultimately, the fresh and dry weights (dried at 105°C) were determined. The number of plants per harvest date varied in the first three experiments. Initially, 10 or even 30 plants per treatment have been harvested, while in later growth stages (after 50 days) only 4–6 plants of each treatment were available. In experiment 4 all plants (10 per treatment) were harvested simultaneously.

## RESULTS

In a preliminary experiment (Nr. 1) two varieties, viz., Nobel and Viroflay have been used. Three daylengths: 12 hours, 14 hours, and 16 hours, were applied. Plants were harvested after 25, 42, and 54 days of treatment. The appearance of flower primordia, and shooting in Viroflay plants was earlier and at shorter daylengths than in Nobel plants. This difference between these varieties is also known in practice. The different daylengths hardly resulted in any significant difference in fresh and dry weights of the shoots of the Viroflay plants. The plants of the variety Nobel apparently need longer daylengths for formation of flower primordia, and for shooting. In this variety, daylength caused signifi-

cant differences in fresh and dry weight of the shoots. After 25 days of treatment, the plants under a daylength of 16 hours had attained higher weights than those at daylengths of 12 and 14 hours, while after 54 days of treatment the plants at daylengths of 14 and 16 hours also showed significantly higher weights than those at 12 hours. Although the same trend has been observed at an earlier growth stage (42 days), these differences were not significant.

In the other experiments (Nr. 2, 3, and 4), only the more pronounced long day variety Nobel has been used, so that also more plants per treatment were available. In experiment 2, the following daylengths were applied: 12 hours, 15 hours, and 18 hours. Plants were harvested after various periods of growth between 22 and 90 days.

Figure 1A shows the course of stem length of the plants grown at the three daylengths; the values presented in this figure are averages of the harvested plants. The plants at 18 hours showed fast shooting, those at 15 hours elongated more gradually, whereas the plants at 12 hours remained in the rosette stage. These differences between the treatments were significant. Examination of the growing points showed that, at the daylength of 18 hours, already 40 per cent of the plants had flower primordia after 22 days, while all plants were in the reproductive stage after 28 days. Flower buds were visible after 35 days, and flowers appeared in the period between 43 and 62 days. At the daylength of 15 hours all plants still were vegetative after 22 days, 61 per cent of them were in the reproductive stage after 28 days, and all were so after 35 days. Flower buds were visible after 50 days; flowering started after 57 days of treatment, and still was not finished at the end of the experiment (76 days). At the daylength of 12 hours all plants remained vegetative up to 57 days; after 90 days 50 per cent had growth primordia which looked like an early reproductive stage.

In figure 1B, the average fresh weight of the shoot is plotted against duration of treatment. The same values, together with their mean errors, are given in table 1. In the period between 22 and 35 days, the fresh weights of the plants

TABLE 1. The effect of daylength on fresh and dry weight of the shoot in *Spinacea oleracea* L. after various periods of growth.

The figures represent averages, and mean errors in grams per plant (Experiment 2).

Days	12 Hours		15 Hours		18 Hours	
	Fresh	Dry	Fresh	Dry	Fresh	Dry
22	0.36 ± 0.03	0.03 ± 0.003	0.63 ± 0.07	0.03 ± 0.004	0.67 ± 0.07	0.04 ± 0.003
28	0.83 ± 0.06	0.06 ± 0.007	1.82 ± 0.08	0.13 ± 0.003	1.77 ± 0.06	0.12 ± 0.007
35	1.97 ± 0.16	0.18 ± 0.01	3.27 ± 0.15	0.23 ± 0.01	3.28 ± 0.33	0.22 ± 0.03
50	5.60 ± 0.61	0.50 ± 0.06	8.86 ± 1.05	0.82 ± 0.09	12.00 ± 0.73	1.20 ± 0.07
57	8.45 ± 1.64	1.02 ± 0.16	15.12 ± 1.59	1.55 ± 0.12	12.82 ± 1.44	1.39 ± 0.16
62	-	-	-	-	13.04 ± 0.70	1.44 ± 0.14
76	-	-	18.20 ± 2.48	2.43 ± 0.27	-	-
90	20.35 ± 2.50	2.75 ± 0.20	-	-	-	-

at 15 and 18 hours were practically equal, and significantly higher than those of the plants at 12 hours. This is also demonstrated in Plate 1, in which average plants of the three daylengths after 28 days of treatment are presented at the same scale. The plants, grown at 15 and 18 hours, are distinctly larger. After 50 days of growth, the plants at the 18-hour daylength were significantly higher

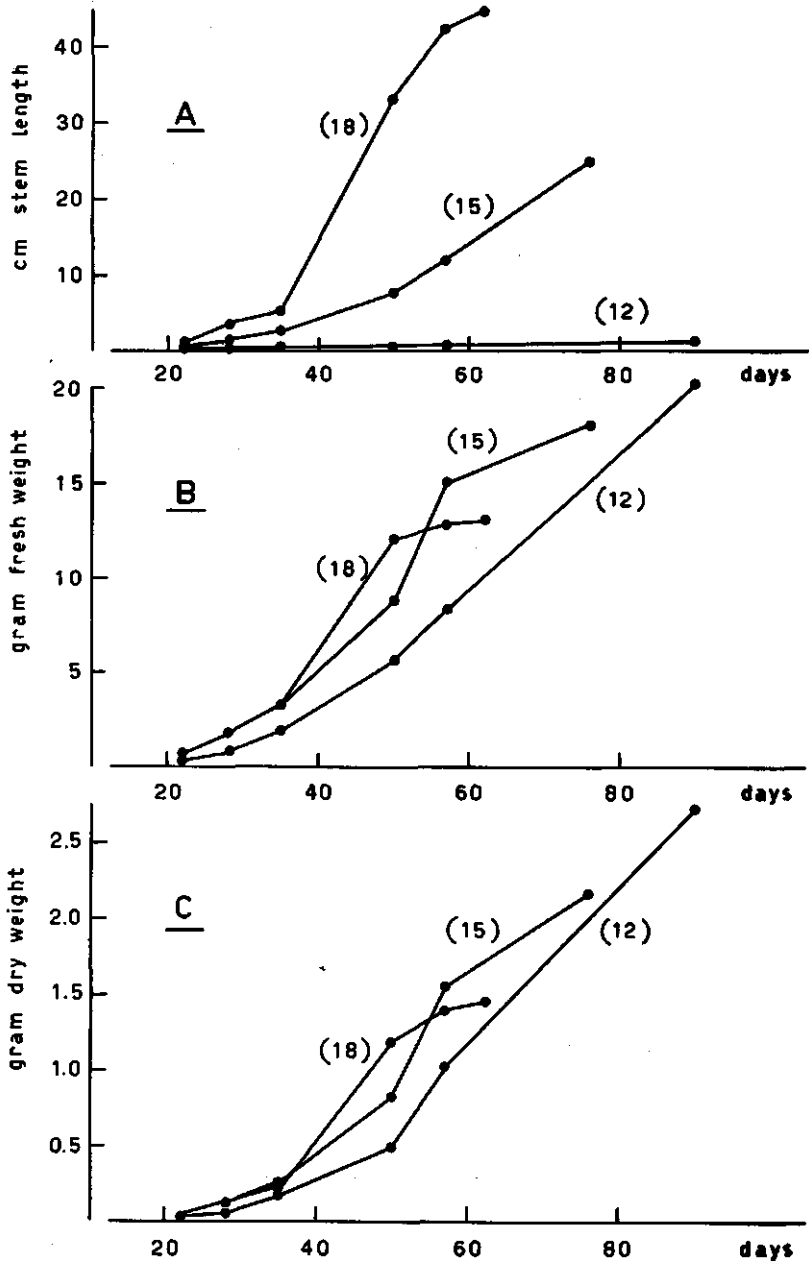


FIG. 1. The effect of three different daylengths (12, 15, and 18 hours, as indicated with the curves) upon stem length (A), fresh weight (B), and dry weight (C) of spinach plants during growth (Expt. 2).

in fresh weight than those at 15 hours. Shortly afterwards, the plants at 18 hours had attained their maximum fresh weight under these conditions, whereas those at 15 hours still increased in fresh weight. The fresh weights of the latter plants finally seem to be surpassed by those at the daylength of 12 hours, although the experimental period would have to be further extended for fully conclusive data in this respect.

The average dry weights of the shoot are plotted in figure 1C, and combined with their mean errors shown in table 1. The general pattern of the curves for the three different daylengths is similar to that of the fresh weights, given in figure 1B. Significant differences in dry weight between the plants at these daylengths were found. Already after 28 and 35 days, such differences appeared between the plants at 15 and 18 hours at one side, and those at 12 hours at the other side. As with the fresh weights, the plants at 18 hours reached a maximum dry weight earlier than those at 15 hours, but the final dry weight, reached by the latter plants, was higher. The highest dry matter production seems to be attainable by the plants at 12 hours.

The average number of unfolded leaves after various duration of treatment is given in table 2. Already after 22 days, the number of unfolded leaves is higher at longer daylengths, and the differences between the three treatments subsisted with time. As has been indicated before, the formation of flower primordia was completed after 28 and 35 days at daylength of 18, and 15 hours respectively, thus at the 6 to 7 leaf stage (Table 2). The plants at 12 hours, however, remained vegetative during nearly the whole experiment, although a higher number of leaves had been formed.

TABLE 2. Average numbers of unfolded leaves in *Spinacea oleracea* L. after various periods of growth as affected by daylength (Experiment 2).

Days	12 hours	15 hours	18 hours
22	2.0 ± 0.0	2.6 ± 0.2	3.3 ± 0.3
28	4.0 ± 0.1	4.7 ± 0.2	6.0 ± 0.2
35	5.8 ± 0.3	6.9 ± 0.4	8.3 ± 0.3
50	9.3 ± 0.3	11.8 ± 0.8	23.3 ± 1.3
57	11.8 ± 0.5	17.3 ± 2.3	27.0 ± 2.1
62	-	-	29.0 ± 2.4
76	-	25.5 ± 3.9	-
90	19.8 ± 0.3	-	-

The leaf area per plant has been estimated from the number of unfolded leaves, the length and the width of the leaf-blades, measured at the harvest times, considering the shape of the leaf-blade approximately as an ellipse. The curves of the average leaf area calculated in this way paralleled those of the fresh weights given in figure 1B. This may be explained as a formative effect of daylength upon leaf area, so that in plants at long days the leaf area is increased above those at short days.

The incident energy per cm<sup>2</sup> leaf area per 24 hours varies little with the daylength applied; the plants at a daylength of 15 hours, and those at 18 hours received 1.7, and 3.4 per cent more light energy respectively per cm<sup>2</sup> leaf area per 24 hours than those grown at 12 hours. So, the differences in incident energy per plant are primarily determined by the differences in leaf area. Considering these data, and those of the dry weights after the same growth periods, one

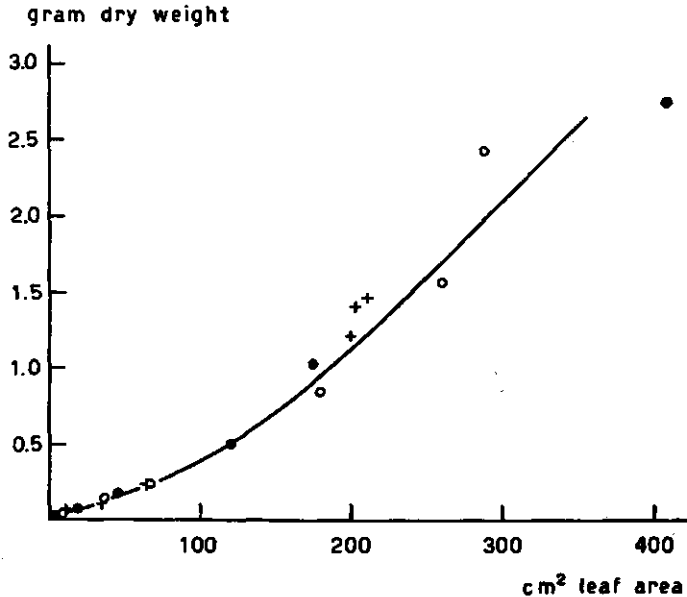


FIG. 2. The relation between shoot dry weight and leaf area of spinach plants, during various periods of growth at daylengths of 12 hours (●), 15 hours (○), and 18 hours (+) (Expt. 2).

wonders whether the plants under longer days more efficiently convert the light energy incident during growth. Possible differences in efficiency of light energy conversion per  $\text{cm}^2$  leaf area may be shown by plotting the dry weight attained after different periods of growth against the leaf area calculated at the same harvests, and comparing the values for the three daylengths. This has been done in figure 2. The amount of dry weight per  $\text{cm}^2$  leaf area appears to be independent of daylength, although at later stages (after 76–90 days) some variation occurs. Thus, the efficiency of light energy conversion is approximately the same at daylengths of 12, 15, and 18 hours. This efficiency seems higher at later growth stages of the plants. The reason for this increase so far has not been analyzed; several explanations seem possible.

From the results of experiment 2 we probably may conclude that increase in daylength results in a larger leaf area of the plants, and this implies that plants under long days are able to absorb more light energy, and consequently, produce more dry weight than plants under short days in the same period of growth.

In another experiment (Nr. 3), dry weight of leaf-blades, and dry weight of stem and petioles have been determined separately. The same daylengths were applied, although one fluorescent tube instead of the four small incandescent lamps has been used for realizing the daylengths of 15 and 18 hours. After 44 days of growth, the plants at 12 hours still were vegetative, those at 15 and 18 hours were then shifted to the reproductive phase. Some results are given in figure 3. The differences in dry weight of stem and petioles between the three daylengths after 44, 59, and 73 days of growth are significant. After 44 days of growth, the difference in dry weight of the leaf-blades at 15 and 18 hours at one side, and

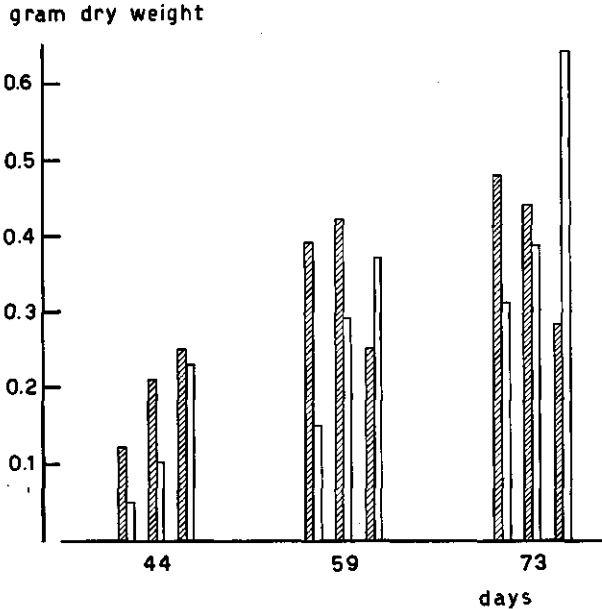


FIG. 3. The effect of daylength upon dry weight of leaf-blades (hatched bars), and stem + petioles (blank bars) of spinach plants after some periods of growth. At each date, from left to right: 12 hours, 15 hours, and 18 hours (Expt. 3).

those at 12 hours at the other side were significant, while after 59 and 73 days the differences in dry weight of the leaf-blades at 12 and 15 hours at one side, and those at 18 hours at the other side were also significant. Figure 3 shows that the dry weight of the leaf-blades of the plants at 18 hours is fairly constant from 44 till 73 days, while that of the plants at 15 hours reaches a constant level after 59 days. The dry weight of the leaf-blades of the plants at 12 hours then still increases, and is highest of all, after 73 days. The dry weight of the stem and petioles increases with time at the three different daylengths, and reaches higher values the longer the days are. Consequently, the plants under shorter days store relatively more light energy in the leaf-blades than in the stem and petioles, as compared to those under longer days. This is especially observed in later growth stages (e.g., after 73 days).

TABLE 3. The effect of daylength on stem length, and dry weight of shoot in *Spinacea oleracea* L. after 44 days of growth. The figures represent averages of 10 plants. (Experiment 4).

Daylength hours	Stem length cm	Dry weight grams
12	2.0 ± 0.2	0.54 ± 0.07
15	10.9 ± 1.8	0.57 ± 0.06
18	22.0 ± 2.7	0.82 ± 0.09
21	31.0 ± 4.1	1.04 ± 0.20
24	25.0 ± 4.0	0.71 ± 0.10

In experiment Nr. 4, five different daylengths were applied; 10 plants were available for each treatment. The length of the photoperiod ranged from 12 to

24 hours. The plants were harvested after 44 days; stem length and shoot dry weight have been determined. Table 3 gives the average values, and the calculated mean errors. The longest plants and the highest dry weights were found at a daylength of 21 hours. The plants at 18 hours were not significantly lower in dry weight, however. Continuous irradiation caused lower dry weights than cycles of 24 hours interrupted by 3 hours dark.

## DISCUSSION

The application of long days including low intensity supplementary light caused early flower initiation and flowering in the long day plant *Spinacea oleracea* L. (Fig. 1A). Under the same conditions, the vegetative growth in early growth stages (*i.e.* production of fresh and dry weight, increase in number of leaves, and in leaf area) proceeded more rapidly than under short day conditions (Fig. 1B and C, Tables 1 and 2). WAGENAAR (1954) also found a higher dry weight and a larger leaf area of spinach plants under long days in early growth stages. WITHROW and WITHROW (1940) observed higher final dry weights of spinach in long days, as compared to those in short days, both harvested after an arbitrary period of growth. With other long day plants, similar effects have been reported. BONNER (1940) mentioned the effect of long day upon the dry weight of *Brassica*. WASSINK and STOLWIJK (1953), and BODLAENDER (1958, 1959) observed higher weights, and a higher leaf number in potato after a certain period of growth in long days.

The increase in leaf number (Table 2) and leaf area under long days in early growth stages probably were responsible for the higher weights, for the growth per unit leaf area seemed independent of the daylength (Fig. 2). This increase in leaf area may be considered as a formative reaction upon the daylength, while the larger leaf area under the same light intensity results in a higher weight in the same period of growth. Indications of the effect of daylength upon leaf growth in literature are divergent. BORTHWICK, PARKER, and SCULLY (1949) observed larger plants of *Taraxacum kok-saghyz* L. (long day plant) under long day conditions. ARNEY (1956) found higher leaf area (and higher number of cells) in strawberry plants (short day plant) in long days. However, in both cases a relatively high supplementary irradiation was applied. BÜNNING (1956) observed lower leaf areas of the long day plant *Plantago lanceolata* L., and higher ones of the short day plant *Perilla ocymoides* L. under long day conditions. In his experiments no flower primordia were observed. BLACKMAN (1956) has shown that reduction in light intensity may result in increase in leaf area, depending on the plant species. The low intensity of the supplementary light in our experiments may partly be responsible for the pronounced effect upon leaf area.

From the growth curves in figures 1B and 1C, it is evident, however, that, under short days, the weight of spinach plants ultimately increases above that under long days, owing to the extension of the growth period. The higher final weights of potato under long days in the experiments of WASSINK and STOLWIJK (1953) and of BODLAENDER (1958) are also due to a longer growth period, because potato behaves as a short day plant for tuber growth which thus is retarded under long days. SMILDE (1960) demonstrated that with the short day plant *Sesamum indicum* L. final dry weight, leaf number, and leaf area were highest under long days as a result of the suppression of flowering, and the extension



of the growth period. Thus, the effects upon this short day plant are opposite to those upon spinach. These results may explain the positive effects of long days upon the final dry weight of some other short day plants, viz. *Xanthium* and *Cosmos* (BONNER (1940)), and *Soja* (WITHROW AND WITHROW (1940)).

Naturally, long day plants, like spinach, growing under long day conditions, and thus developing towards flowering, store relatively more dry weight in the stem than they do under short day conditions, as can be seen in figure 3. With potato plants under long days, BODLAENDER (1959) also found relatively higher dry weights of stems with respect to leaves as compared with plants under short days; it should be noted that in this case the stems belong to the "vegetative growth" which, in potato, is promoted by long days.

A practical implication for spinach might be the possibility to obtain higher plant weights during early stages of growth, at which the harvest for consumption takes place. Extension of the short days during winter months (e.g. under glass) with a low light intensity would then result in higher plant weights. It is, however, doubtful whether in the usual dense stand of plants in practice, the increase in leaf area per plant would result in higher yields per unit soil area, since the amount of light energy available during the winter may well restrict further increase in growth.

#### SUMMARY

Some experiments on the effects of daylength upon vegetative growth, and reproductive development of *Spinacea oleracea* L. have been described. Different daylengths have been applied, making use of supplementary irradiation of low intensity. The growing points have been examined, while stem length, leaf dimensions, and weight of the plants have been determined after various periods of growth.

In early growth stages, the plants under long days showed higher values of stem length, fresh weight and dry weight, leaf number, and leaf area than those under short days (Figures 1A-1C; Tables 1, 2; Plate 1). The maximum effect was observed at daylengths of 18 to 21 hours (Table 3).

The growth per unit leaf area appeared to be independent of daylength (Figure 2). The increase in leaf number and in leaf area as a formative reaction upon extension of the day with low intensity light may be responsible for the effect on plant weight.

Under long day conditions the plants earlier reached their maximum size (leaf area and weight), and growth stopped, whereas under shorter days growth proceeded, and the plants finally reached a larger size (Figure 1B, 1C).

Plants in long days accumulated relatively more dry weight in stems and petioles than those under short days (Figure 3).

These results are discussed in connection with other data on short and long day plants. Possible practical implications for spinach growing are mentioned.

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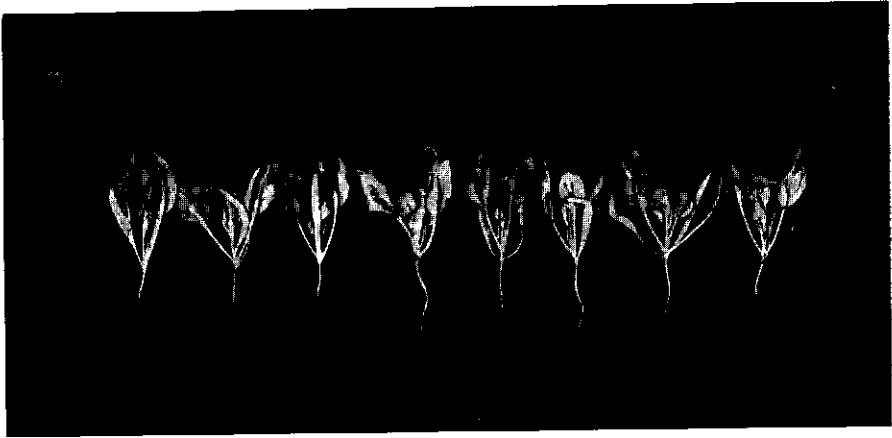


PLATE 1. The size of spinach plants after 28 days of growth at daylengths of 12 hours (A), 15 hours (B), and 18 hours (C) respectively (Expt. 2).