Dynamics of Leaf Growth and Development in Sugarbeets

E. Ann Clark and R. S. Loomis*

Leaf growth and development are dynamic processes which vary in response to the external environment and to internal factors such as plant size and activity (20, 9). In the sugarbeet these processes are unconstrained by reproductive activities during the first year of growth and leaves are produced throughout ontogeny against a background of increasing plant size. A progressively more massive root sink is an important aspect affecting internal status of the plant.

Integration of leaf growth processes within whole-plant activities was reviewed in detail by Wetmore and Wardlaw (31). Although some of the events are controlled in part by plant hormones, it will be useful here to emphasize nutritional aspects of the problem.

Hussey (10, 11) showed with tomato that leaf initiation rate was relatively constant during ontogeny, but if old leaves were removed, the initiation rate increased and the apex enlarged. He suggested that subapical nutrition was normally limiting to leaf initiation and early growth. Wilson (32) came to a similar conclusion regarding leaf growth after removing the apical meristem of cucumber. With sugarbeet, Das Gupta (6) found that the growth of young leaves was greatly enhanced by removing the apex and the more juvenile unfolding leaves, an observation also made by Loomis and Brookings (unpublished).

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*Graduate student and Professor, Department of Agronomy, University of California, Davis, CA. Present address of the senior author: Department of Agronomy, Iowa State University, Ames, Iowa.

Of particular interest is the progression of increasing
and then decreasing mature sizes shown by the first 20 to
25 leaves of the sugarbeet. We (13, 14) were able to dup-
licate this progression with a simulation model based on
nutritional control of leaf cell division and expansion.
Allsop (1) concludes in his review that such sequences are
dependent upon carbohydrate nutritional status and associa-
ted changes in the apical meristem; he found no need to
postulate that specific juvenile hormones were involved.
Ashby (2) and Ashby and Wangerman (3, 4) came to a somewhat
similar view after establishing correlations between cell
size, mature leaf size, and the number of less mature, com-
peting leaves.

Nutritional status also is strongly affected by the general
environment. Conditions favoring photosynthesis relative
to growth sink activity will enhance carbohydrate supply
and thus alter growth balances (15). Such hypotheses can
be used in the interpretation of leaf growth responses to
temperature and radiation (16, 18, 21, 26, 29). Leaf
growth responses to moisture (24, 25) and nutrient stresses
(12, 17, 27) can be analyzed in a similar approach -- using
nitrogen deficiency, for example -- by considering separate
limiting nutritional pools of nitrogen and carbon assimi-
lates.

The simulations of juvenile leaf sequences in sugarbeet
with a dynamic model based on assimilate limits to growth
illustrated importance of variations in leaf initiation
rate and leaf morphology to the integration of shoot and
storage root activities. But an effort by Loomis, Ng and
Hung (13) to simulate responses to variations in plant
density failed, in part because that "computer genotype"
failed to duplicate the developmental and environmental
plasticity of real plants. A better understanding of the
physiological controls is needed. The experiment reported
here involves a field study of this issue. Emphasis was
given to characterization of the responses to light-limited
status imposed by variations in plant density.

MATERIALS AND METHODS

The growth and development of sugarbeet leaves as influenced by planting density were investigated in a field experiment conducted at Davis, California, during the summer months of 1974 with a high-performing commercial variety, US H10B. Four density treatments with equidistant (hexagonal) spacing patterns were established. This was accomplished by varying between-row and within-row spacing (Table 1). An excess of seeds, pretreated with PCNB, Dexon, and Lindane, was planted in rows in dry soil with the effective date of planting taken from the first irrigation on 20 May. When the plants had reached the 8- to 12-leaf stage (14-21 June), the rows were thinned carefully to single plants to establish the hexagonal pattern. The treatments were arranged replicated four times in a Latin square design with appropriate borders. Plots were 7.6 cm wide and contained 240 spaced plants. Plot length varied accordingly.

Table 1. -- Space relations used to achieve four plant densities after thinning. Alternate rows were offset to achieve a triangular relationship among adjacent plants.

		Spacing between plants	
Plant density		Within rows	Between rows
pl m^{-2}	pl ha^{-1}	cm	cm
24	240,000	22	18
12	120,000	30	26
6	60,000	42	37
3	30,000	61	53

Prior to planting, 153 kg N ha^{-1} and 90 kg P ha^{-1} from a mixture of NH_4NO_3 and ammonium phosphate fertilizer were incorporated uniformly with the soil. Additional N, totalling 52.5 kg ha^{-1}, was applied by hand during the season as needed to maintain soil resources in a nonlimiting condition. During the first 4 weeks water was applied frequently from sprinklers mounted on risers to facilitate emergence and early growth. Later irrigations were applied as needed to prevent water supply from becom-

ing limiting to growth. Weeds were a problem early in the
season and were not fully controlled until the end of June.
An incidence of powdery mildew was noted in August and con-
trolled with wettable sulphur (9.4 kg ha^{-1} in an o.5%
aqueous suspension). The disease attacked only the older
leaves so that possible influences on leaf growth were
presumably indirect. The results presented here are from
selected situations judged free from confounding influences
of the disease

Selected parameters of leaf-area expansion and of
partitioning plasticity within leaves were measured as
follows:

Leaf Appearance Rate
Leaf appearance rate (where leaf appearance was arbitrarily
defined to be attainment of 5 cm in length) was assessed
on five plants in each plot at weekly intervals between 53
and 130 days after planting. The smallest leaf greater
than 5 cm in length was tagged for reference and the number
of leaves appearing since the previous week's tag was re-
corded.

Individual Leaf Expansion
Leaf area expansion was estimated using the linear regres-
sion method of Owen (1957). As indicated in Figure 1,
estimated area, A', was taken as the product of the coor-
dinates, X and Y. Owen calculated $A' = kXY$, where k is the
coefficient of regression of estimated on actual leaf area.
In present study, a sample of 500 leaves yielded a regres-
sion coefficient (k) of 1.01 and we took $A' = XY$. The 1%
underestimate was well within the error ot planimetery.
At 1- to 2-week intervals starting on day 53, the youngest
leaf over 5 cm long was tagged on each of five plants per
plot. These leaves were then measured (X and Y) every 3 to
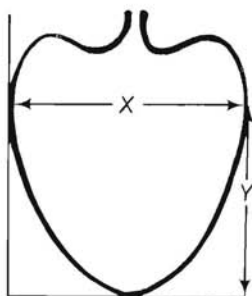5 days until they reached maturity.

    Figure 1.--Coordinates X and Y used in estimating the
area of sugarbeet leaves.

Mature Leaf Area

Ten plants in each plot were reserved for measurement of
mature leaf area. At 7 to 10 day intervals, every fifth
leaf (10, 15, 20, 25, etc.) on these plants was tagged for
reference. When these tagged leaves were fully mature, as
estimated by their age and appearance (close to senescence),
their areas were determined by the linear regression tech-
nique.

Components of Partitioning Plasticity within the Leaf

At 59, 91 and 129 days after planting, three representative
plants were harvested from each plot for partitioning
measurements. Plants were harvested prior to 9 a.m. and
were maintained within plastic bags in a cold room until
separated into components. Individual leaves were removed
from the plants in ontogenetic sequence. Blades were sep-
arated from petioles at the point where the laminar tissue
was sharply reduced. The blade area of each leaf was
estimated by regression. Individual blade and petiole
parts were then dried at $60^{\circ}C$ in a forced draft oven and
weighed.

Solar radiation and minimum and maximum daily temperatures
as measured at a standard weather station (600 m S.E.) are
shown in Figure 2. Incident radiation was relatively con-
stant through 7 August (80 days), at which time it began

a gradual decline to about 650 cal $cm^{-2}da^{-1}$ by 27 August
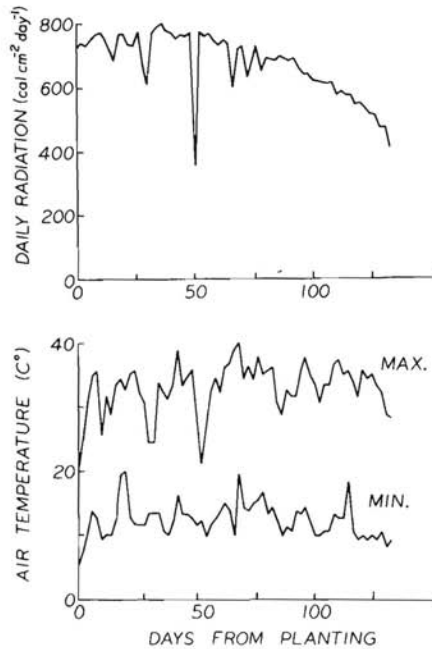(100 da) and 500 cal $cm^{-2}da^{-1}$ by mid-September (120 days).

Figure 2.--(Upper) Daily total solar radiation on a
horizontal surface (Eppley solarimeter) during the growing
season. (Lower) Two-day averages of daily maximum and
minimum air temperatures during the growing seasons.

RESULTS

Both of the high density plantings (12 and 24 pl m^{-2}) had
attained full cover by early July (estimated by visual
inspection); the next intermediate density (6 pl m^{-2})
reached full interception in early August (Table 2). With
3pl m^{-2}, interception remained incomplete throughout the
season. Lesser levels of light interception at the second
sampling date (Table 2) are attributable to a reduction
in the canopy following the powdery mildew infection
(Table 3).

Leaf Appearance Rate

The rates of leaf appearance (LAR) exhibited by the four
densities were highly significantly different during the

first 4 weeks of measurement (50 to 80 days; Fig.3) with 3 pl m^{-2} having the highest rate, decreasing sequentially to 24 pl m^{-2} having the lowest rate.

Table 2.--Percentage transmission of PAR quanta (Lambda sensor) at solar noon, measured at the ground level at 70 and 108 days from planting.

Plant Density	Day 70		Day 108	
	Transmission	$S_{\bar{x}}$	Transmission	$S_{\bar{x}}$
pl m^{-2}	%		%	
24	2.0	0.23	8.0	1.63
12	2.0	0.28	14.0	2.20
6	8.0	2.05	20.0	3.06
2	25.0	5.10	37.0	3.56

Table 3.--Leaf area per plant and leaf area index (LAI) measured at 59, 91 and 129 days from planting.

Plant density	Leaf area	Day 59		Day 91		Day 129
		LAI^b	Leaf area	LAI	Leaf area	LAI
pl m^{-2}	dm^2pl^{-1}		dm^2pl^{-1}		dm^2pl^{-1}	
24	24.5	5.9	32.4	7.8	40.3	9.7
12	39.1 $(2.8)^a$	4.7	48.6 (4.2)	5.8	48.4 (5.2)	5.8
6	55.1	3.3	77.7	4.7	63.5	3.8
3	68.7	2.1	120.0	3.6	89.5	2.7

a Standard errors of the treatment means at each harvest are shown in parentheses.
b Calculated from the leaf area (dm^2pl^{-1}) data.

Beyond 80 days, the LAR's converged to a common level and generally were not significantly different from each other for the remainder of the season.

The convergence of LAR among densities to a common level tended to parallel the trend of the various densities towards full cover. The occurrence of the lowest LAR values in each density corresponded with time of infection and buildup of the powdery mildew disease. LAR values increased following an application of sulphur on day 87.
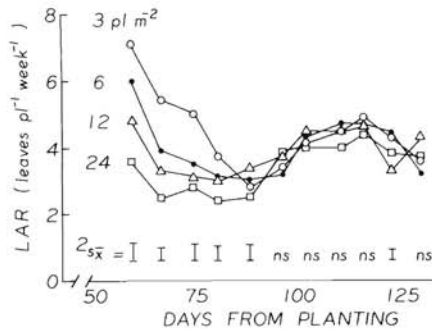
    Figure 3.--Leaf appearance rate (LAR) as a function
of time for different plant densities.

Leaf Expansion and Mature Area

Leaf expansion measurements were begun on day 53 (17- to
24-leaf stage) with 2 or 3 leaves already senescent.   The
leaf expansion process was more rapid and the area of
mature leaves greater with low densities (Fig. 4).   In
addition, the rates of expansion in all densities declined
as the season progressed.

The differences in final area attained by comparable
leaves from densities (Fig. 4) are particularly striking.
The areas attained by the leaves with 3 pl m^{-2} were the
largest, with those of the other densities following in
regular order: 6 > 12 > 24 pl m^{-2}.

The sugarbeet leaves continued to expand, albeit slowly,
until just prior to senescence (Fig. 4).   The final mature
areas (Fig. 5) were determined when the leaves were quite
old as judged by their overall appearance and chronological
age.   Differences among densities in mature leaf areas were
highly significant for each measured leaf number.   The area
reached by successive leaves followed a common pattern in
each density -- rising to a maximum, then falling through-
out the remainder of the season.   The numbered leaf with
the maximum area varied with density: leaf 15 with the two
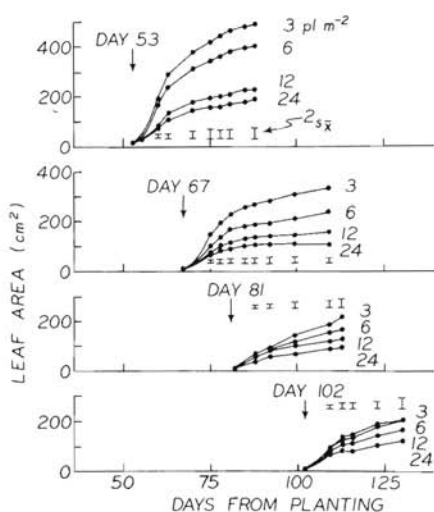higher densities, and leaf 20 at the low densities.

Figure 4.--The course of leaf blade expansion of leaves appearing on various dates for different plant densities.
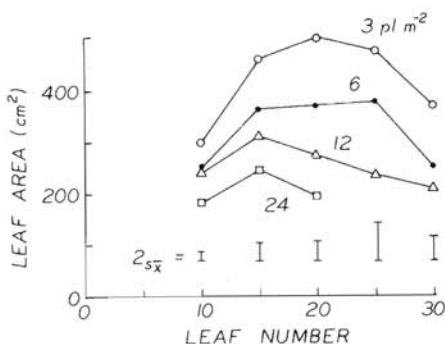
Figure 5.--Blade area of mature leaves measured just prior to their senescence for various densities. Numbering proceeds from first-appeared to later-appeared leaves.

Partitioning Within the Leaf

Dry matter partitioning within leaves was assessed using plants harvested at 50, 91, and 129 days from planting. The patterns were similar at each harvest; data obtained at 91 days are presented. The chronological leaf distributions of each plant were used to select similarly aged leaves for the construction of Figures 6 and 7, showing

the course of leaf ontogeny between day 53 and day 88.
Mean incident radiation declined from 750 to 700 cal
$cm^{-2}da^{-1}$ during this period.

Specific leaf area (SLA; cm^2 leaf surface g^{-1} blade dry
weight) was smaller with increasing leaf age and with
decreasing density (Fig. 6).   Young leaves from plants of
all densities exhibited high SLA values which were main-
tained for varying lengths of time and then declined as
the leaves increased in weight per unit area during de-
velopment.   After the first week, the SLA values of leaves
from each density assumed a consistent order, with density
24 pl m^{-2} always highest, followed by $12 > 6 > 3$ pl m^{-2}.

The allocation of dry matter to blade and to petiole parts
with leaf ontogeny is depicted in Figure 7 for the highest
and lowest densities (the intermediate densities fell be-
tween these two in all parameters measured).
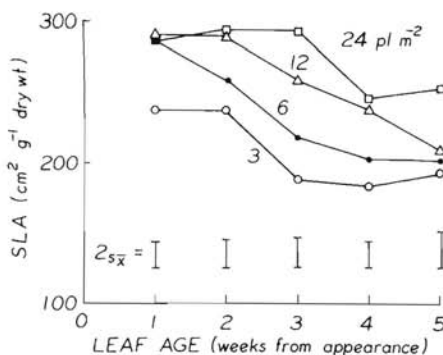
Figure 6.--Specific leaf (blade) areas (SLA) observed
on day 91 as a function of leaf age at various densities.

Resource allocation was markedly influenced by density.
Rates of dry matter accumulation were strikingly different
after 2 weeks time, when the curves for the lowest density
turn sharply upward while those of the highest density
remain essentially linear.   Blade and petiole tissues
differed in this regard.   At the lowest density, petiole
weight paralleled blade weight for the first 3 weeks, then

Figure 7.--Dry weights of blades (B) and petioles (P) on day 91 as a function of leaf age. Data for 3 and 24 pl m^{-2}.

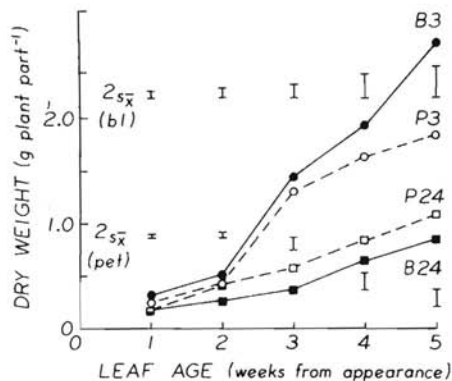leveled off while blade weight continued to increase linearly. At the highest density, however, increases in blade and petiole weight were essentially linear and parallel over the whole period, showing no tendency to level off; petiole weight exceeded blade weight at all leaf ages. At the low density, blade weight exceeded petiole weight with the magnitude of the difference increasing with leaf age.

DISUCSSION

Variations in planting density were used here to elicit growth and developmental plasticity in leaves. The microclimate and the availability of soil resources both vary with population density but with soil resources maintained at nonlimiting levels, we can attribute the observed effects to modifications of the aerial environment. The principal effect of increasing density is a reduction in the area of sunlight per plant and hence a reduction in the potential photosynthate production per plant. The attainment of full cover places a restriction on further increases in photosynthate production per plant and thus in the supply per growing point. Associated with the increasing density of the foliage canopy is increased shading of the apical meristem and the developing leaves,

and, through a greater proportion of the net radiation
going to evapotranspiration, lower midday air temperatures
within the canopy. Soil temperature fluctuations also
are moderated as canopy density increases. These effects
of density are to some extent confounded with the normal
increase in erectness and tightness of the leaf whorl
which occurs during ontogeny.

The principal vegetative response of sugarbeet plants to
light and microclimate limitations included variations in
storage root growth, leaf surface expansion, and parti-
tioning within leaves. Successively greater growth depres-
sion per plant and per organ with increasing density was
observed in all measured parameters (Figures 3, 4, 5, 6,
and 7).

Assimilate status at the apex has been implicated as a
major factor affecting the rate of leaf initiation (30,
18, 5, 11). Leaf appearance rate is determined both by
leaf initiation rate (LIR) and by the time interval be-
tween initiation and appearance of successive leaves. In
sugarbeet, LIR is reduced and the time interval increased
in low light (26) as is also the case with cucumber (18),
bean (5) and tomato (10). Assimilate status was not
measured but we would expect it to vary with density (with
light), as well as with the size or age of the plant (in-
creases in root sink activity and mutual shading).

Leaf expansion rate and the final area of mature leaves
were affected to a much greater extent by variable plant
spacing than was LAR (Figures 3, 4, and 5). This may re-
flect the relatively small absolute carbohydrate demand,
or higher priority, for LIR compared to that for the cell
division and expansion activities following initiation.
Interpretation is complicated by the fact that the ex-
panding one-third (8) to one-half (23) its mature size.
Thrower (28) noted that restricting light incidence on a
young, expanding leaf prevented full leaf expansion and
resulted in a considerably smaller mature leaf area.

Thus, both LAR and early leaf growth depend on the in-
tegrated assimilate status of the plant as a whole and
thus density.

The canopy light environment also influenced partition-
ing in young leaves between blade and petiole tissues.
Leaves produced under high density and lower SLA (Fig. 6)
and a greater proportion of total leaf weight was used in
petiole formation (Fig. 7). In spite of the striking
differences between high and low densities in partition-
ing and in final leaf size, the blade area achieved per
unit dry weight allocated to leaf growth was the same at
both densities and relatively constant during the later
stages of ontogeny (Table 4). The much greater SLA at
high density compensated for the smaller fraction of leaf
weight going to leaf blades. At the community level,
leaf-area index was greater at the high density, despite
the smaller leaf size, because of the much greater number
and weight of leaves per unit.

Table 4.--Blade area per leaf and per g total leaf
weight (blade + petiole) observed on day 91 as a function
of leaf age at 3 and 24 pl m^{-2}. (Blade and petiole weights
for these leaves are illustrated in Fig. 7.)

Blade area	Plant density	Leaf age (weeks from appearance)				
		1	2	3	4	5
	pl m^{-2}					
Per leaf (cm^2)	3	76	120	271	355	517
	24	46	76	102	157	214
Per total leaf wt (cm^{2}g^{-1})	3	133	127	99	100	113
	24	134	114	109	107	111

As the season progressed, the effects of planting density
became less clear, with a tendency for convergence in
the individual leaf parameters. Two factors may be in-
volved in controlling this pattern of development: in-
creasing tightness of the whorl which tends to place all
developing leaves in a similar environment for light and
temperature; and an increasing dominance of the storage

root which competes for a limited supply of assimilates.
As the plants approached full cover, competition for light
from adjacent plants would limit photosynthetic produc-
tion per plant to a steady-state level, varying principally
with the amount of radiation received to that area each
day. As the potential assimilate use capacity of the root
sink continued to increase, a generally stable but pro-
gressively lower internal photosynthate status would be
established. This decline in photosynthate status could
serve to restrict vegetative growth of leaves below their
genetic potential. The effect would be greater at high
densities with a much smaller photosynthate supply per
plant.

The depression in vegetative growth during the latter half
of the season also involves the increasing requirement for
maintenance respiration and the seasonal decline in inci-
dent radiation (Fig. 2). Supporting the importance of an
active root sink, however, is the fact that leaf growth
began to decline well before incident radiation began its
seasonal fall (Figures 3, 4, and 6), and the leaves of
plants at the lowest density, which never reached full
cover, demonstrated the same trends with plant ontogeny
as did those of the light-limited plants from the other
densities.

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