THE EFFECT OF WATER STRESS ON TRANSLOCATION IN RELATION TO PHOTOSYNTHESIS AND GROWTH

II.* EFFECT DURING LEAF DEVELOPMENT IN *LOLIUM TEMULENTUM* L.

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Summary

Photosynthetic rate, leaf and root extension, dry weight changes, and the translocation of labelled photosynthates were followed in *L. temulentum* plants subjected to water shortage at a time when the eighth leaf was expanding.

During a period of increasing water deficit, extension growth was reduced before the photosynthetic rate. However, the effects on photosynthesis were directly related to the water status of the leaf, determined as relative turgidity, and did not appear to be a consequence of the effects on growth.

The transfer of assimilates from the photosynthetic tissue into the conducting tissue was delayed in stressed leaves. However, the velocity of assimilate translocation through the conducting tissue was only slightly affected at low relative turgidity if the demand for assimilates from the labelled leaf was increased by removal of adjacent leaves.

These results suggest that effects of water stress on translocation arise indirectly from effects on growth and that the translocation pathway is either resistant to water loss or capable of functioning efficiently when under water stress.

I. Introduction

Photosynthesis, translocation of sugars, and growth have each been implicated as the first process to be affected by water stress (Wardlaw 1968). In a previous paper (Wardlaw 1967) it was shown that during the period of rapid starch deposition in the wheat grain, water stress acted initially on the leaf, reducing both photosynthesis and the transfer of sugars from the photosynthetic into the conducting tissue. However, under conditions in which the photosynthetic rate of the leaf was reduced by half there was no effect on grain growth, or on the movement of assimilates once they had entered the conducting system, and it was concluded that either translocation was insensitive to water deficit or that the phloem tissue was resistant to desiccation.

The investigation was therefore extended to a study of the effect of water stress during vegetative growth in *Lolium temulentum* L. where the response during cell division and cell extension in the leaves and roots might be expected to differ from that obtained during grain development in wheat (cf. Burström 1956; Iljin 1957).

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II. Methods and Materials

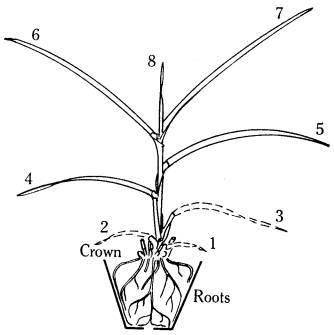
(a) *Experimental Outline*

The order of the response to water stress of leaf and root extension and leaf photosynthesis were recorded in the first two experiments [Sections III(*a*) and III(*b*)]. Measurements of the relation between leaf photosynthesis and relative turgidity [Section III(*c*)] were included in an attempt to assess whether the effect of water stress on leaf photosynthesis was associated with the water status of the leaf or whether there was a possible indirect effect of stress on leaf photosynthesis resulting from an effect on growth.

The relation between the distribution of photosynthate and water stress was studied by following the effect of stress on both the distribution of ^{14}C-labelled photosynthates [Section III(*d*)] and the dry weight of plant parts [Section III(*e*)]. The interaction between growth and conduction of assimilates in plants under water stress was determined from measurements of the velocity of ^{14}C-labelled photosynthate movement in intact and defoliated plants [Section III(*f*)].

(b) *Cultural Conditions and Imposition of Water Stress*

Plants of *L. temulentum*, Ceres strain, were grown singly in pots (10-cm diam.) containing perlite, in naturally lit phytotron units in which the day length was shortened to give an 8-hr photoperiod. They were watered once daily with standard nutrient solution and once with tap water. Temperatures were maintained at 21°C for the 8-hr light period and at 16°C for the 16-hr dark period (21/16°C).



At the time of emergence of the tip of leaf 8, all tillers and the first three leaves on the main culm were cut from all plants. The accompanying diagram shows the stage of development of the plants at that time. The plants were then transferred to an artificially lit (L.B.) growth cabinet (Morse and Evans 1962). Growth conditions, unless otherwise stated, were kept constant for the remainder of the experiment with an air temperature of 21°C and a light intensity of 2500 f.c. under high-output daylight fluorescent tubes supplemented with incandescent lamps. Light was measured by a flat EEL selenium photoelectric light meter, and 2500 f.c. was equivalent to a total radiation intensity of 13·5 g cal cm^{-2} hr^{-1} measured with a Kipp net radiometer. Relative humidity was maintained between 40 and 45%.

Water stress was induced, as previously (Wardlaw 1967), by stopping the supply of water and nutrient and allowing the pots to dry out slowly.

(c) *Growth Analysis*

The rate of extension of leaf 8 was used as a measure of the extent to which plant growth was under water stress. In the unstressed leaf there was an initial flush of growth, about 1–2 days after emergence, probably associated with loosening of a constriction caused by the older sheathing leaf. This was followed by a fairly steady rate of growth over a period of about 4 days and then a rapid decline in growth rate with the emergence of the ligule at the junction of the sheath and the blade (Fig. 1). Where possible, stress was induced during the period of most constant growth rate.

The zone of leaf extension was found, by floating excised sections on water, to be limited to the basal 4 cm of the 8th leaf (cf. Kirshin 1962; Davidson and Milthorpe 1966), and was enclosed within the sheath of leaf 7 which measured about 11 cm under the conditions of these experiments. The distal exposed photosynthetic tissue of leaf 8 was fully expanded while growth of the leaf base continued. This contrasts with the situation in dicotyledonous leaves where growth and photosynthesis occur simultaneously in the same tissue.

Leaf extension rates were calculated from direct length measurements made at set time intervals during an experiment, or were recorded continuously with an auxanometer (Williams and Biddiscombe 1965).

Root extension was measured indirectly by determining the pattern of incorporation of labelled carbon into the root following the assimilation of $^{14}CO_2$ by the leaves at set time intervals.

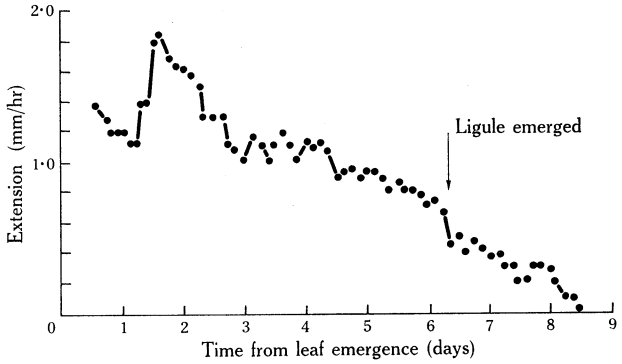


Fig. 1.—Rate of extension of leaf 8 of *L. temulentum* from the time of emergence until full expansion. Data are from one of three leaves measured with an auxanometer.

The incorporation of ^{14}C into individual roots from a control plant and from a plant under water stress following two 10-min uptake periods of $^{14}CO_2$ by leaf 6 is shown in Figure 2. Labelling was separated by 2 days and the plants were harvested 2 days after the second $^{14}CO_2$ uptake period. At harvest each root was cut into 5-mm lengths, each length was fixed to a planchet, dried at 60°C for 24 hr, and its radioactivity was measured with a Geiger–Müller counter.

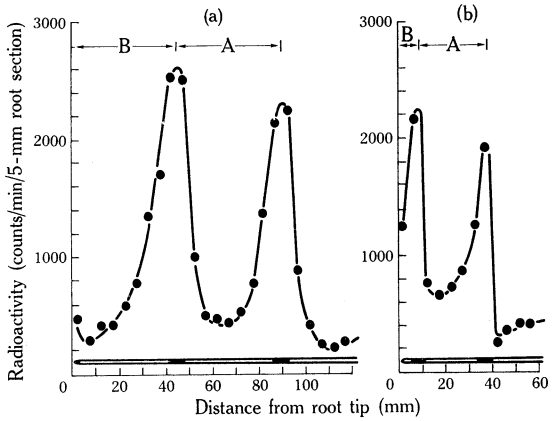


Fig. 2.—Determination of root extension in (*a*) control and (*b*) stressed plants from the pattern of ^{14}C incorporation into the roots, following the fixation of $^{14}CO_2$ by the leaves. *A* is the distance between peaks of ^{14}C activity along the root following two 10-min fixation periods 2 days apart, and indicates the growth during the first 2-day period, while *B* indicates the additional growth 2 days after the second fixation period. This is shown diagrammatically, immediately above the horizontal scale, with shading of the root to illustrate the zones of maximum activity.

Two peaks of radioactivity can be observed down the length of each root and each peak represents the zone of root tissue most actively growing following the uptake of $^{14}CO_2$ by the leaf. The very confined growth zone of the root tip makes it possible to mark a 5-mm length of root using this technique.

To determine the effect of water stress on root extension three apparently healthy roots were selected for analysis from each of four replicate plants. Growth for the three roots was averaged and the variation in response between plants was analysed statistically.

The main problem with this method of determining root growth is variation in the movement of assimilates out of the leaf and in the velocity of this movement through the conducting system. Although this is likely to be critical in experiments involving measurements over a few hours, in the present longer term experiments this error will be small. A delay in assimilate movement, which is often found under water-stress conditions (Wardlaw 1967), would in these experiments result in an underestimate of the initial stress effects on root extension and then an overestimate of the effect of stress on root extension between the second $^{14}CO_2$ assimilation and harvest. The magnitude of this error is not likely to be more than 2 hr in 48.

Dry weights of leaves and roots were measured directly, on samples taken at different stages of water stress, following drying at 80°C for 48 hr.

(*d*) *Photosynthetic Measurements*

Carbon dioxide exchange by single leaf blades, attached to the plant, was examined by enclosing the terminal 10·5 cm in a perspex chamber 2·0 by 15·5 cm in cross section. The differential in CO_2 concentration of an air stream before and after passing over the leaf was determined with a Grubb–Parsons infrared gas analyser (model SB2). Air flow rates were such that the maximum difference in CO_2 concentration was no greater than 50 p.p.m. by volume. Typically a flow rate of 1 litre per minute was used for a leaf area of 10 cm^2. All photosynthetic measurements were carried out in the artificially lit (L.B.) cabinet.

(*e*) *Relative Turgidity*

Relative turdigity was estimated, as in earlier experiments with wheat (Wardlaw 1967), by standing the basal 5 mm of the plant part in water and enclosing the whole in a water-saturated atmosphere. Light intensity was 10 f.c. and temperature 24°C throughout the uptake period. An initial experiment, in which fresh weights were recorded at hourly intervals, indicated that water uptake was complete in 4–5 hr and measurements after 5 hr were subsequently used in calculating the saturated water content of the organ.

Relative turgidity (Weatherley 1950; Hewlett and Kramer 1963) was calculated as

$$\frac{100(\text{initial fresh weight} - \text{oven dry weight})}{(\text{saturated weight} - \text{oven dry weight})}.$$

In several instances estimates of relative turgidity were made on the expanding basal tissues of the leaf. The expansion of the tissue during the uptake period was recorded and the estimate of final water content was reduced in proportion to the expansion (cf. Čatský 1959). The validity of this correction was not examined but it resulted in an increase in the estimated relative turgidity of 3% in control plants to 7% in plants under severe stress, and made little difference to the comparison between expanding and mature tissue. It appears likely that there will be a linear relationship between relative turgidity and diffusion pressure deficit for any one organ with a slope characteristic of that organ (Weatherley and Slatyer 1957; Yang and De Jong 1968), but no attempt was made here to check the relation between relative turgidity and other measurements of water stress.

(*f*) *Mannitol*

To compare the tensions required to prevent extension growth in leaves and roots, sections were floated on mannitol solutions of varying concentration, and lengths were recorded after a period of 6 hr.

(*g*) *Carbon-14 Determinations*

The methods of analysis of the distribution and velocity of movement of ^{14}C-labelled assimilates were similar to those described in earlier papers (Wardlaw 1965; Evans and Wardlaw 1966).

To follow the distribution of assimilates from leaves the terminal 10·5 cm of leaf 6, or the terminal 8·5 cm of leaf 8, of 12–16 plants were exposed to $^{14}CO_2$ in an assimilation chamber for a period of 20 min.

In the first experiment, on distribution from leaf 6, $^{14}CO_2$ containing 100 μCi of ^{14}C was generated from 100 mg of $Ba^{14}CO_3$ for each of two groups of 15 plants. Initial CO_2 concentration was 0·70%, v/v. In the second experiment, on distribution from leaf 8, $^{14}CO_2$ containing 100 μCi of ^{14}C was generated from 50 mg of $Ba^{14}CO_3$ for each of three groups of 12 plants. Initial CO_2 concentration was 0·35%, v/v. In the third experiment, on distribution from leaf 6 in relation to defoliation, $^{14}CO_2$ containing 160 μCi of ^{14}C was generated from 160 mg of $Ba^{14}CO_3$ for simultaneous feeding of 32 plants distributed between two chambers in parallel. Initial CO_2 concentration was 0·56%, v/v.

Plants were harvested either 4 or 24 hr after the uptake of $^{14}CO_2$ and were immediately cut into parts for drying and determination of the distribution of radioactivity.

For measurements of velocity of movement of ^{14}C-assimilates the terminal 15·5 cm of the 6th leaf of 30 plants was exposed to $^{14}CO_2$ over a 10-min period. $^{14}CO_2$ containing 200 μCi of ^{14}C was generated from 100 mg of $Ba^{14}CO_3$. Single plants were harvested at 10 min intervals, commencing 10 min, 13 min, and 16 min after $^{14}CO_2$ uptake for the controls, defoliated stressed plants, and intact stressed plants respectively. Immediately on harvest the assimilating leaf was cut into 5-cm lengths from below the $^{14}CO_2$ uptake area, and the remainder of the plant was divided into young shoot, older leaves (when present), crown, and roots. The parts were then dried at 80°C for 48 hr and analysed for radioactivity.

TABLE 1

ROOT AND LEAF EXTENSION IN RELATION TO WATER STRESS

Each result is the mean of four replicates ±S.E. and, for roots, each replicate represents the average in three roots/plant

Plant Part	Extension Growth (mm) after:	
	1–3 days*	3–5 days*
Leaf 8		
Control	81·0±4·1	57·2±1·8
Stressed	76·0±2·3	15·5±4·9
Percentage†	94	27
Roots		
Control	41·2±0·7	41·9±2·4
Stressed	30·6±4·0	15·2±4·1
Percentage†	74	36

* From cessation of watering.

† Stressed as percentage of control.

III. RESULTS

(a) *Root and Leaf Extension in Response to Water Stress*

A comparison of the rate of leaf extension determined by direct measurement on leaf 8 with the rate of root extension estimated indirectly by pulse feeding of the leaves with $^{14}CO_2$ (Table 1), showed that the initial effects of water stress were greater on the roots than on leaf 8, but as the stress became more severe with time leaf growth was reduced as much as root growth.

When short lengths were cut from the growing regions of the leaf base and root tip of control plants and floated on a series of mannitol solutions of different concentrations for 6 hr, it was evident (Fig. 3) that root extension was inhibited by a lower concentration of mannitol than was leaf growth. Where leaf elongation had ceased because of water stress, a 1M solution was required to inhibit extension of basal leaf sections placed in mannitol solutions. This suggests a xylem tension of about 25 atm at the stage when leaf extension ceases under water stress.

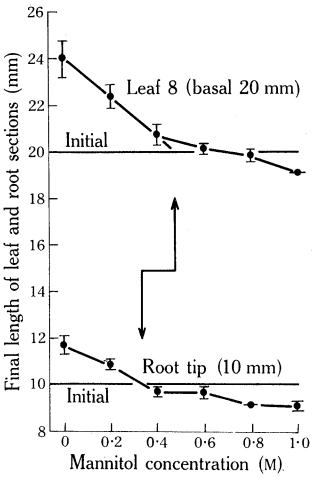


Fig. 3.—Extension growth of root and leaf sections from control plants on varying concentrations of mannitol over a period of 6 hr. Initial leaf sections were 20 mm long taken from the expanding basal zone, and initial root sections were 10 mm taken from the tip. Results expressed as final lengths are the means of three replicates (with two roots from each plant) and the vertical lines indicate $2 \times$ S.E. The arrows indicate the concentrations of mannitol that prevent leaf and root extension.

(*b*) *Photosynthesis in Relation to Leaf Extension on Plants Entering Water Stress*

Continuous recordings of the extension growth of leaf 8 and of the rate of photosynthesis of leaf 7 in plants subjected to increasing water stress (Fig. 4) indicated that leaf elongation was reduced in response to stress prior to any effect on leaf photosynthesis. In separate experiments (cf. Fig. 6) it was found that photosynthesis was reduced by water stress in leaf 7 some time before it was reduced in leaf 8. Hence, the difference in the time at which stress becomes apparent in growth response and in photosynthesis of leaf 8 will be greater than the difference shown in Figure 4. The plants were then rewatered to ensure that the reduced extension rates were related to stress. Recovery of photosynthesis was incomplete, but separate experiments indicated that the drop in photosynthesis of unstressed leaves, over this period, would be no more than 10% of the initial value and probably much less.

As relative turgidity fell below 85%, photosynthesis became less sensitive to water deficit than extension growth. Thus, while growth had almost ceased at a relative turgidity of 75%, photosynthetic activity was still about one third of its maximum rate (Fig. 5). From the results of Yang and de Jong (1968), with wheat plants 4 weeks old, a relative turgidity of about 75% would correspond to about -25 atm, which is in close agreement with the tension required to prevent growth of stressed leaves as estimated by floating leaf sections on mannitol.

(c) *Photosynthesis and Leaf Water*

A comparison of CO_2 exchange by leaves 7 and 8 in response to water stress showed there was a greater reduction of photosynthesis in the older leaf 7 than in leaf 8. However, if photosynthesis is plotted against relative turgidity of the leaf tissue [Fig. 6(*a*)] it can be seen that the greater reduction in photosynthesis of leaf 7 is generally associated with a greater leaf water deficit. The earlier appearance of stress in the older leaves of *L. temulentum* is similar to the response observed by Čatský (1965) in cabbage. Prior to the onset of stress, rates of photosynthesis in the younger leaf 8 were generally lower than those in leaf 7.

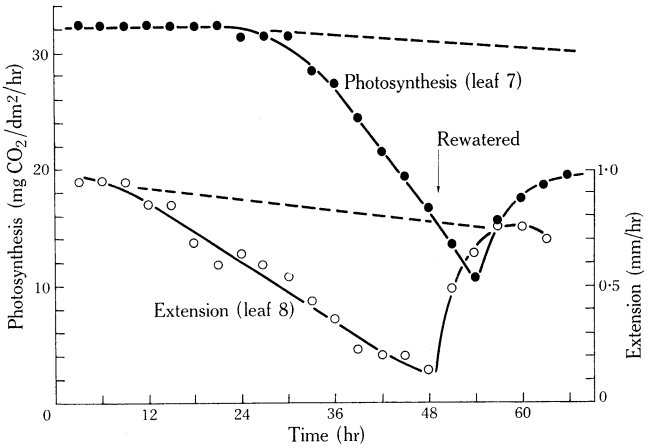


Fig. 4.—Photosynthesis of leaf 7 (●) and the rate of extension of leaf 8 (○) in response to increasing water stress and recovery following rewatering. The results presented are based on continuous recordings on one plant. The time scale is arbitrarily commenced just before the stress effects became apparent in growth, 3 days after cessation of watering. The broken lines (– – –) represent the expected response without stress for photosynthesis based on comparable control plants and for growth based on the recovery of growth following rewatering in this experiment.

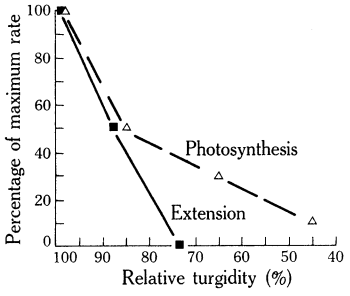


Fig. 5.—A comparison of the relation between photosynthesis or extension growth and relative turgidity of the exposed photosynthetic tissue or of the basal expanding tissue respectively of leaf 8.

An estimate of the effect of leaf sugar levels on the relation between photosynthesis and leaf water was made by growing plants at either a high (1500 f.c.) or a low (500 f.c.) light intensity for 3 days prior to the initiation of stress. Leaf photo-

synthetic rates were then measured at 2500 f.c. and their relative turgidities determined [Fig. 6(*b*)]. The following tabulation shows the results of other measurements on these leaves:

Treatment	Dry Weight (mg/5 disks*)	Extractable Weight (mg/5 disks*)	Substances Extractable in 80% Ethanol–Water (%)
High light, control	2·21	0·74	33·3
High light, stressed	2·23	0·80	35·5
Low light, control	1·65	0·63	38·1
Low light, stressed	1·73	0·65	36·7

* Disks 4 mm in diameter.

Although there were differences in leaf dry weight and 80% ethanol–water-extractable material per unit leaf area (methods as in Wardlaw and Porter 1967) between the two groups of plants, there was no obvious difference in the relation between photosynthesis and leaf water content. This suggests that, for the short duration of these experiments, the effect of water stress on the photosynthetic activity of the leaf was related to the water status of the leaf and was not the result of stress effects on growth.

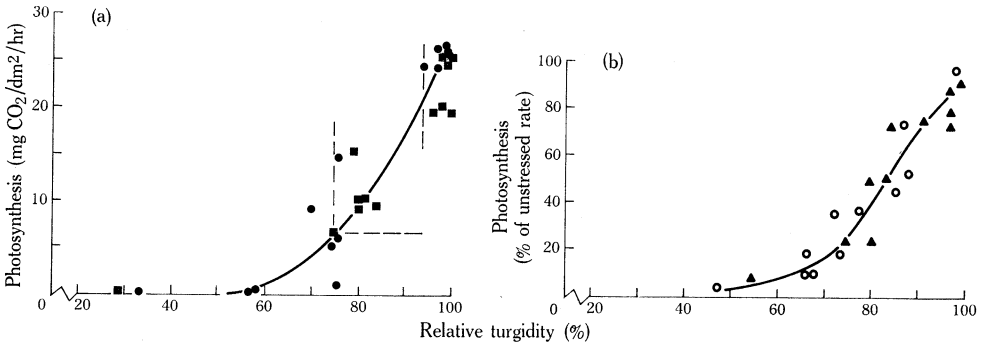


Fig. 6.—Relation between relative turgidity and photosynthesis. (*a*) A comparison between young (■, leaf 8) and mature (●, leaf 7) leaves. (*b*) A comparison between leaves previously grown under high (○) or under low (▲) light.

From light-response curves it was found that, as in wheat (Wardlaw 1967), stressed leaves in *L. temulentum* reached maximum photosynthetic rates at lower light intensities (2000 f.c.) than was the case for turgid leaves (above 3000 f.c.).

In contrast to observations made by Iljin (1957) and Kaul (1966), there was no indication of a rise in respiration rate with the onset of stress in either the expanding tissue or the photosynthetic tissue of the leaf, although a fall in rate was apparent with severe stress.

(*d*) *Water Stress and Assimilate Distribution*

To determine the effect of water stress on the distribution of assimilates from the leaves of *L. temulentum*, $^{14}CO_2$ was fed in two separate experiments to either the terminal 8·7 cm of leaf 8, which was less than half expanded at the time of labelling

(Table 2), or to the terminal 10·5 cm of leaf 6, which was fully expanded (Table 3). The distribution of ^{14}C throughout the plant was then determined either 4 or 24 hr after the commencement of $^{14}CO_2$ uptake.

(i) *Distribution from Leaf 8 (Expanding)*

Carbon assimilated by the tip of leaf 8 was largely accumulated in the growing region at the base of the leaf (Table 2), with only 19% exported to other parts of the

TABLE 2

DISTRIBUTION OF ^{14}C-LABELLED PHOTOSYNTHATE FROM LEAF 8 (EXPANDING) OF *L. TEMULENTUM* IN RESPONSE TO WATER STRESS

Each result is the mean of six replicates ± standard error

Plant Part	Percentage Distribution after 4 hr in:			Percentage Distribution after 24 hr in:		
	Controls	Mildly Stressed Plants	Severely Stressed Plants	Controls	Mildly Stressed Plants	Severely Stressed Plants
Leaf 8 (exposed part)	48·7±1·0	59·1±3·2	65·8±4·3	30·2±0·6	39·7±2·5	39·5±3·5
Leaf 8 (enclosed part)	38·4±4·5	35·2±4·0	28·2±2·9	50·5±2·5	50·9±2·4	50·7±3·4
Young leaves	2·0±1·1	0·5±0·4	0·2±0·2	3·9±0·9	1·8±0·4	1·7±0·6
Mature leaves	2·6±0·5	2·7±0·6	0·9±0·1	4·8±0·3	3·8±0·2	4·4±0·8
Crown	4·5±1·7	1·6±0·4	4·0±1·7	6·9±2·5	2·1±0·4	2·1±0·6
Roots	3·8±1·5	1·0±0·3	1·0±0·4	3·7±0·7	1·7±0·3	1·7±0·5
Total plant activity (counts/min)	1914±120	1631±107	1111±218	2067±100	1540±95	1207±152
Growth rate of leaf 8 (mm/hr)	1·35*	0·90*	0·52*	1·55†	0·65†	0·37†

* Growth rate calculated from extension for 6 hr prior to harvest.

† Growth rate calculated from extension for 16 hr prior to harvest.

plant in 24 hr (cf. Evans and Wardlaw 1966). The two stages of stress reduced both leaf extension and the rate of photosynthesis, assessed by comparing the total ^{14}C per plant in each group with that in controls. There was a clear reduction in the rate of loss of assimilates from the leaves when under stress, although differences were considerably less noticeable after 24 hr. Reduced movement of ^{14}C from the photosynthetic tissue was reflected in a much smaller export past the growing regions of the leaf, with relatively little effect on the proportion of assimilates retained in the growing zone.

Growth was reduced comparatively more than photosynthesis or translocation, except under severe stress at the 4-hr harvest. This suggests that sugars may accumulate in the base of the leaf and that the weight per unit length of growing tissue would tend to be higher in stressed than in control leaves.

(ii) *Distribution from Leaf 6 (Mature)*

The two stages of water stress examined showed reduced leaf extension growth, although the mild stress had little or no effect on photosynthesis as indicated by total

^{14}C uptake (Table 3). This confirms the sequence observed using gas analyser techniques, that stress was evident in extension growth prior to any effects on photosynthesis.

TABLE 3

DISTRIBUTION OF ^{14}C-LABELLED ASSIMILATES FROM LEAF 6 (MATURE) OF *L. TEMULENTUM* IN RESPONSE TO WATER STRESS

Each result is the mean of five replicates ± standard error

Plant Part	Percentage Distribution after 4 hr in:			Percentage Distribution after 24 hr in:		
	Controls	Mildly Stressed Plants	Severely Stressed Plants	Controls	Mildly Stressed Plants	Severely Stressed Plants
Leaf 6 (blade)	48·1±5·5	60·8±3·6	66·0±1·0	25·1±2·5	27·5±1·1	27·7±3·6
Leaf 6 (sheath)	2·7±0·2	3·5±0·7	4·6±0·8	2·2±0·5	4·9±1·2	6·1±0·9
Young leaves	2·8±0·5	2·8±0·3	3·3±0·4	5·6±0·7	10·1±1·5	12·7±0·7
Mature leaves	0·3±0·1	1·8±1·3	0·6±0·1	0·8±0·1	1·4±0·3	1·8±0·1
Crown	24·0±4·6	8·0±0·5	7·2±0·6	31·4±2·6	15·8±2·7	15·9±1·9
Roots	22·8±2·8	23·2±3·8	18·3±1·3	34·9±1·0	40·4±3·9	35·9±1·8
Total plant activity (counts/min)	1099±70	1091±42	574±129	1008±32	833±50	692±86
Growth rate of leaf 8 (mm/hr)	1·73*	1·38*	0·71*	1·93†	0·54†	0·35†

* Growth rate calculated from extension 16 hr prior to $^{14}CO_2$ uptake.

† Growth rate calculated from extension 16 hr prior to harvest.

As with leaf 8, the 4-hr analysis of ^{14}C distribution indicated a reduction in the rate of loss of assimilates from the stressed leaf. There was a small increase in retention of ^{14}C by the sheath of leaf 6 and movement of a greater proportion of the assimilated ^{14}C to the young expanding leaves of stressed plants. There was also some evidence for accumulation of assimilates in the roots when under mild stress. Even allowing for the apparent reduction in photosynthetic rate of the leaf the results indicate that there would be an initial accumulation of dry material per unit length as extension growth slowed down under stress and translocation continued.

The marked reduction in movement of ^{14}C to the crown tissues of stressed plants could be due either to the inability of these tissues to utilize a limited assimilate supply, or to the crown tissue being more sensitive to stress. The crown consisted largely of defoliated tillers as well as small buds and root bases. In a separate experiment comparing intact and defoliated shoots it was noted that the extension of leaf 8 was slower in the latter (cf. Davidson and Milthorpe 1966). A lower concentration of mannitol was required to stop extension growth in basal leaf sections from defoliated plants compared with sections from intact plants.

(e) *Dry Weight Changes in Relation to Water Stress*

Two experiments were undertaken in which the dry weights of different parts of control plants were compared with the same parts from plants under varying

degrees of water stress. The two experiments were complementary and only data from the second experiment are presented here (Table 4). Four days after cessation of watering two groups of plants were selected for severity of stress based on the rate of extension of leaf 8. Dry weight analysis of control plants over a similar period failed to show any significant trend in dry weight per unit length of leaf or root tissue with time.

TABLE 4

DRY WEIGHTS OF PLANT PARTS IN RELATION TO WATER STRESS

Each result is the mean weight of 17 replicates ± standard error

Plant Part	Dry Weight (mg) of Plant Parts in:		
	Controls	Mildly Stressed Plants	Severely Stressed Plants
Leaf 8 (expanding)			
Tip (4·0 cm)	3·9±0·11	3·8±0·11	3·6±0·09
Base (4·0 cm)	6·2±0·21	8·1±0·25	8·5±0·09
Leaf 6 (mature)			
Tip (10·5 cm)	15·7±0·56	14·6±1·54	15·5±1·01
Base (4·0 cm)	18·3±0·61	18·7±0·79	20·5±0·55
Tillers (av. per tiller)	19·6±0·98	18·9±1·29	15·7±1·36
Roots (av. per 20-mm root tip)	0·6±0·03	0·9±0·06	1·0±0·05
Growth rate of leaf 8 (mm/hr)*	1·83	1·04	0·44

* Growth rate calculated from extension for 16 hr prior to harvest.

Labelling experiments with ^{14}C had suggested that translocation of photosynthetic assimilates to the growing regions was less affected by stress than expansion. This is reflected in the dry weight per unit length of the expanding leaf base and root tip and hence confirms the observation by Gingrich and Russell (1957) that dry weight increases in roots are less affected by water stress than elongation. Also, as suggested by the labelling experiments, there was a rise in weight of the mature leaf sheath with increasing stress, which indicates that when growth is interrupted assimilates accumulate in other parts of the plant (cf. Sampson and McCarty 1930; Hartt 1967). However, there was no indication of assimilate accumulation in the photosynthetic tissue of the blades in either of these analyses.

(f) Velocity of Assimilate Movement in Relation to Assimilate Demand and Water Stress

In considering the effect of water stress on assimilate movement it is necessary to consider possible indirect effects caused by changes in growth. In wheat a reduction in grain growth caused a reduction in velocity of assimilate movement through the stem (Wardlaw 1965). If velocity of assimilate movement is reduced in plants under water stress this could be the result of either a direct effect of stress on translocation or indirectly the effect of stress on growth.

The effect of water stress on the velocity of assimilate movement in intact shoots was compared with that in shoots where the lower leaf blades had been removed, 18 hr prior to the start of the measurement, to increase the demand for assimilates from the labelled leaf (Tables 5 and 6).

TABLE 5

EFFECT OF DEFOLIATION ON ^{14}C-LABELLED PHOTOSYNTHATE DISTRIBUTION IN *L. TEMULENTUM* AS RELATED TO WATER STRESS

Each result is the mean of eight replicates ± standard error

Plant Part	Percentage Distribution after 24 hr in Controls:		Percentage Distribution after 24 hr in Stressed Plants:	
	Intact	Defoliated	Intact	Defoliated
Leaf 6 (uptake area)	24·5±3·0	20·5±3·2	35·5±2·5	30·5±1·1
Leaf 6 (residual)	3·2±0·3	1·5±0·1	18·6±0·9	11·1±1·2
Young leaves	8·0±0·5	5·7±0·4	13·7±2·2	15·6±1·2
Mature leaves	0·9±0·1	0·6±0·3	2·4±0·2	1·6±0·1
Crown	38·2±4·5	49·2±2·8	12·8±0·8	14·0±0·5
Roots	25·3±1·5	22·7±1·6	17·0±0·9	27·3±3·0
Total plant activity (counts/min)	3561±152	3816±131	2667±281	2811±227
Growth rate of leaf 8 (mm/hr)*	2·56	2·38	0·38	0·67

* Growth rate calculated from extension over the 24-hr distribution period.

TABLE 6

EFFECT OF DEFOLIATION ON THE VELOCITY OF MOVEMENT OF ^{14}C-LABELLED PHOTOSYNTHATE IN *L. TEMULENTUM* AS RELATED TO WATER STRESS

Treatment	Growth Rate of Leaf 8* (mm/hr)	Total Plant Activity (counts/min)	Velocity of Movement of Photosynthate (cm/hr)
Control (intact)	1·82	3400±166	100
Stressed (intact)	0·57	1379±216	38
Stressed (defoliated)	0·62	1946± 75	92

* Growth rate calculated from extension for 16 hr prior to harvest.

Defoliation resulted in an increased distribution of ^{14}C to the roots of stressed plants. Velocity of ^{14}C movement was greatly reduced in intact plants under stress but not in the defoliated plants. Although the degree of stress was less in defoliated than in intact plants, there was still a marked reduction in both photosynthetic rate, as judged by total plant ^{14}C, and in extension growth.

IV. Discussion

(a) *Photosynthesis in Relation to Growth*

Two distinct situations may be recognized. The present experiments showed that water stress reduced extension growth in the roots and young leaves of the vegetative plant before it had any effect on leaf photosynthesis. In contrast, earlier experiments (Wardlaw 1967) demonstrated that carbohydrate storage, during grain development, was less affected by stress than was leaf photosynthesis. Hence the sequence of response to water stress depends on the type and stage of development of the plant (cf. Iljin 1957). The explanation for a particular sequence of responses to stress appears initially to be largely related to differences in water status of the organs rather than to differences in resistance to the same water deficit.

Thus a relative turgidity of 90% resulted in a reduction in leaf photosynthesis of about 30% in leaves grown under both high and low light conditions and also resulted in a similar reduction in leaf extension. It was also noted in earlier experiments (Wardlaw 1967) that a relative turgidity of 77% in the flag leaf of wheat was associated with a reduction in photosynthesis of about 40%, which is very similar to the response obtained here with *L. temulentum*. The maintenance of rapid grain development under the above conditions of stress in wheat was associated with a high relative turgidity (92%) in the whole ear. The initial drop in ear relative turgidity has subsequently been related to the glumes and not the grain.

It was suggested by Zolkevich, Prusakova, and Lizandr (1958) that the inhibition of translocation by water stress would result in sugar accumulation in the photosynthetic tissue, which would in turn reduce photosynthesis. That is, the effect of stress on photosynthesis may be an indirect one. It has been shown that assimilate utilization can affect leaf photosynthesis (King, Wardlaw, and Evans 1967), and in *L. temulentum* growth was reduced prior to photosynthesis in plants subjected to stress. However, the close relation between photosynthesis and leaf water status under varying conditions, and a failure to observe any significant dry weight increase in the leaf tissue when under stress, suggests that such associated reactions were not part of the photosynthetic response to the range of water deficits and the short duration of stress experienced in these experiments. It is possible that under prolonged mild stress a situation might arise where the accumulation of assimilates due to reduced growth does result in reduced photosynthetic rates.

(b) *Assimilate Transfer and Translocation*

Although water stress does reduce the rate of movement of assimilates from the photosynthetic tissue into the conducting tissue, there is reason to believe that once the assimilates have entered the conducting system of the leaf they are readily translocated to other parts of the plant.

For example, there was continued movement of ^{14}C-labelled photosynthetic assimilates to the growing regions; the reduced extension growth under stress almost certainly was not the direct result of a lack of substrate. Moreover, velocity of ^{14}C-assimilate movement through a stressed leaf could be maintained at normal rates when adjacent leaves were removed to increase the demand for assimilates from that leaf. This suggests that the reduced velocity of ^{14}C-assimilate movement in plants

under stress, and with all their leaves intact, was an indirect effect resulting from the slower growth. Growth has been shown to control the velocity of assimilate movement (Wardlaw 1965), and where growth was not affected by stress, as in wheat during grain development (Wardlaw 1967), velocity of assimilate movement was not reduced.

Where stress has been shown to cause an alteration in the distribution of photosynthate, with an increased movement in one direction, it seems likely that the response to stress did not result from direct effects on conduction of photosynthate, but rather from effects on photosynthesis and the loading of assimilates into the conducting system, or on growth. This has been shown to a limited extent in these experiments with an increased movement of photosynthate from the mature leaf to the young leaves in *L. temulentum*. This redistribution of photosynthate is shown clearly by the increased upward movement of labelled assimilates in the yellow poplar under water stress (Roberts 1964), and in wheat during grain development, where water stress resulted in an increased movement of assimilates from the lower leaves to the ear (Wardlaw 1967). The stability of the conducting system in plants under water stress is well illustrated by the continued movement of assimilates from the green stem to the roots and buds in the perennial grass, *Phalaris tuberosa*, when the plant has lost its leaves as a result of a severe water deficit (McWilliam 1968).

(*c*) *Response to Water Deficit and Sugar Accumulation*

There was a difference in response to increasing water deficit between the expanding tissue and the mature photosynthetic tissue of the leaf; this difference became apparent as turgidity dropped below 85%, when photosynthesis showed less sensitivity to increasing water deficit than extension growth. The initial similarity may occur because both growth and stomatal movement are associated with cell turgor. However, when stomata are closed and stomatal movement is no longer a major variable controlling gas exchange as stress becomes more severe, the rate of CO_2 uptake is probably determined by the state of hydration of the photosynthetic apparatus. Čatský (1965) associated the change in slope of the response of the photosynthetic tissue to water stress in cabbage with the point at which the stomata were closed and no longer functional in controlling photosynthesis. This difference between tissues that only becomes apparent at low relative turgidity may be the result of a difference in response to the same water deficit as indicated above. However, the difference could be related to differences in water retention between organelles in a tissue, which were not reflected in the relative turgidity measurements of the tissue as a whole. It is also possible that relative turgidity is not a good indicator for the comparison of water status of growing and mature leaf tissue, although it is not clear why this should be in serious error at the tensions reached in these experiments.

If accumulation of sugar is an important factor in the resistance of plant organs to water stress, then continued translocation would appear to be important in survival of those organs, such as buds and roots, which are distant from the site of assimilation. However, protection of the photosynthetic apparatus (cf. Santarius 1967; Santarius and Heber 1967), would necessitate a slowing down of the transfer of assimilates away from the chloroplasts. It could be the balance between retention of sugars by the chloroplast and transfer of sugars to the conducting system that will, in part, dictate the response of the plant to water stress.

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