PLANT RESPONSES TO SALINE SUBSTRATES

III. EFFECT OF NUTRIENT CONCENTRATION ON THE GROWTH AND ION UPTAKE OF HORDEUM VULGARE DURING A SODIUM CHLORIDE STRESS

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Summary

Sodium chloride, at a concentration of 50 m-equiv/l, was applied to substrates with total nutrient concentrations of 0·4, 1·7, and 17 m-equiv/l. Treatment effects were studied over a 7-day period.

Sodium chloride treatment increased the chloride and sodium contents, and decreased the potassium contents, of most plant parts at all nutrient levels. These changes in ion content were usually most pronounced at the lowest nutrient concentration.

Large net losses of potassium were found from both the shoots and the roots of the sodium chloride-treated plants at the low nutrient level. Net potassium losses also occurred from the oldest leaf and sheath at the medium nutrient concentration.

Salinity reduced growth at the low nutrient treatment only, and it is suggested that this was due to the drastically changed ion relationships of these plants.

The findings emphasize the importance of the composition of the substrate to the tolerance of plants in a saline environment.

I. Introduction

Plant response to saline substrates might well vary with the ionic composition and strength of the basal nutrient solution. A knowledge of such effects would assist in a better understanding of the causal factors involved in a salinity stress.

Most salinity experiments have been carried out at a high ionic strength of the basal nutrient solution. Three concentrations of basal nutrient solution were used in the sodium chloride experiment of Reifenberg and Rosovsky (1947). It was found that the sodium chloride-treated plants absorbed the highest amounts of chloride and sodium on the substrate of low ionic strength. However, the Neubauer method was used and growth of the *Hordeum vulgare* plants used was suboptimal. Ion uptake might thus also have been abnormal. Heimann and Ratner (1961) studied the interaction between potassium chloride and sodium chloride, and found no effect of sodium concentration on potassium uptake. Heimann and Ratner did not present growth data and it is thus difficult to interpret the ion data presented. Moreover, Reifenberg and Rosovsky, as well as Heimann and Ratner, studied ion uptake

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by the whole plant only. This has serious objections because ion uptake usually varies with the individual plant part (see Lündegårdh 1951).

As far as the author is aware, there are no precise plant growth data recorded which relate to the interaction between ionic strength of the basal nutrient solution and high levels of sodium chloride. Yet, such information is not only of physiological interest, but also has bearing on the classification of species into salt-tolerant and salt-sensitive groups.

This effect of ionic strength was determined for a salt-tolerant variety of *Hordeum vulgare*. This paper describes the growth and net changes in chloride, sodium, and potassium contents for the whole plant and its individual parts.

II. Methods

The concentrations (m-equiv/l) of macroelements (apart from sodium chloride) in the high nutrient solutions used in the water-culture experiments described below were as follows: KCl, 5·28; $Ca(NO_3)_2$, 5·72; $NH_4H_2PO_4$, 2·12; $MgSO_4$, 4·24. Concentrations of microelements were those of Arnon (cf. Hewitt 1952). The saline and nutrient treatments used were as follows:

Treatment	Nutrient Strength (i.e. times high nutrient concn.)	NaCl Concn. (m-equiv/l)
Low sodium, low nutrient	1/44	0·1 (control)
Low sodium, medium nutrient	1/10	0·1 (control)
Low sodium, high nutrient	1	0·1 (control)
High sodium, low nutrient	1/44	50·1
High sodium, medium nutrient	1/10	50·1
High sodium, high nutrient	1	50·1

All solutions had a pH of 4·8. They were renewed every 24 hr and were aerated continuously. In this way, the maintenance of the specified ion concentration at the root surface was ensured.

Hordeum vulgare, cv. Bolivia (C.P.I. 8315) was transplanted into 3-l. containers with low sodium, medium nutrient solution. Each container had 22 plants and there were six containers for each treatment. By August 20 the first leaf had fully developed and treatments were imposed. Harvest 1 (H_1) was taken prior to treatment (eight plants per container). Harvest 2 (H_2) was at H_1+1 day (six plants per container), harvest 3 (H_3) at H_1+3 days, and harvest 4 (H_4) at H_1+7 days (for both H_3 and H_4, four plants per container).

At each harvest, two replicates of each treatment were obtained by pooling the plants of three containers. The plants were then separated into: leaf 1, leaf 2, leaf 3, sheath 1, sheath 2, and the roots.

Plant material was suspended in water and boiled for 1 hr. The chloride content of the filtrate was determined by the method of Best (1950) and the sodium and potassium content with an "EEL" flame-photometer. Regular potassium determinations of the substrate for the low nutrient level regimes were also made.

III. Results

(a) *Dry Weight and Relative Water Contents*

At the low sodium chloride level the dry weight curves for all nutrient treatments followed similar trends. However, at H_4, the dry weight of the whole plant increased in the order low, medium, and high nutrient level (Fig. 1), and this was mainly due

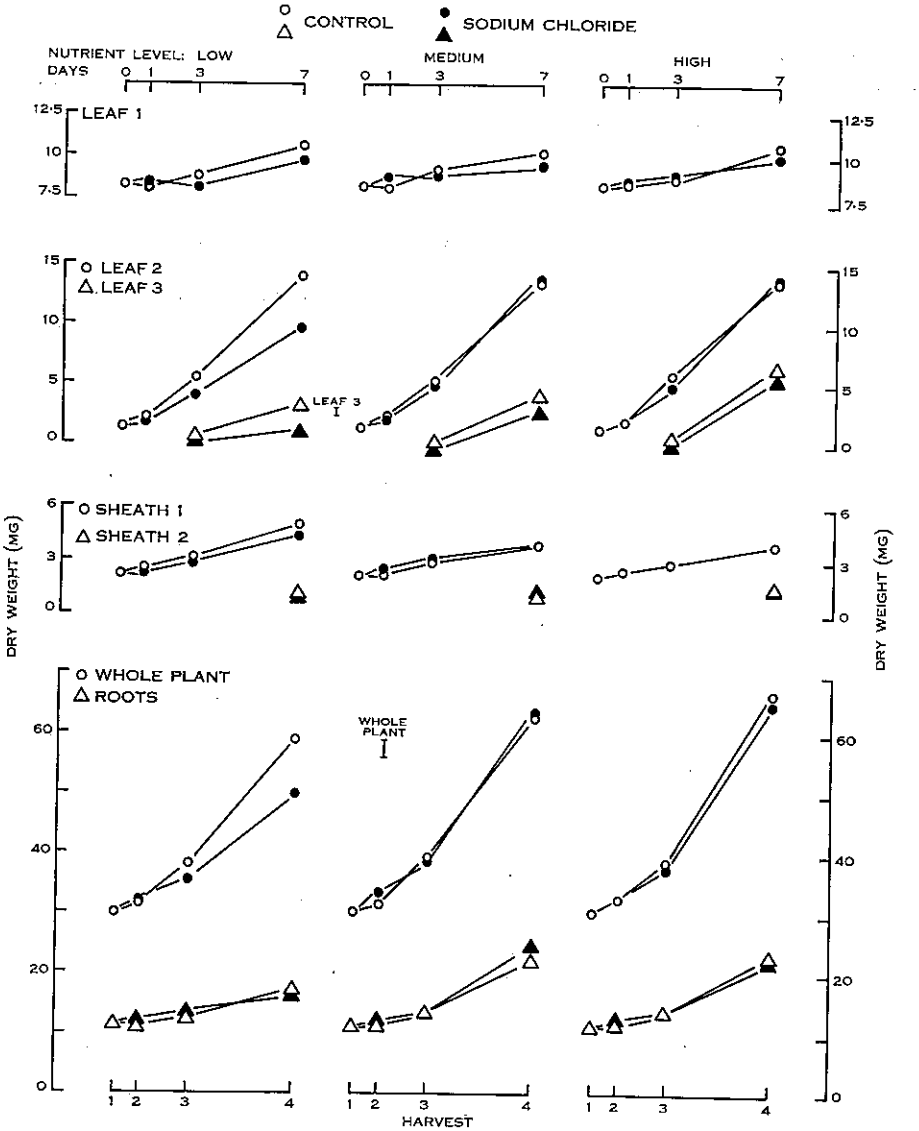

Fig. 1.—Dry weight of the whole plant and its individual parts as affected by sodium chloride treatment at three different nutrient levels. Significant differences at $P = 0 \cdot 05$ indicated for whole plant and for leaf 3. At the high nutrient level dry weights of sheath 1 are identical for sodium chloride and control treatments. Only control values are shown.

In all figures "control" refers to sodium chloride treatments at $0 \cdot 1$ m-equiv/l.

to stronger developed growth of leaf 3 and the roots at the higher nutrient level. Only at the low nutrient level was the dry weight of the whole plant reduced by high sodium chloride treatment. At H_3 and H_4, growth of all parts of the shoot was

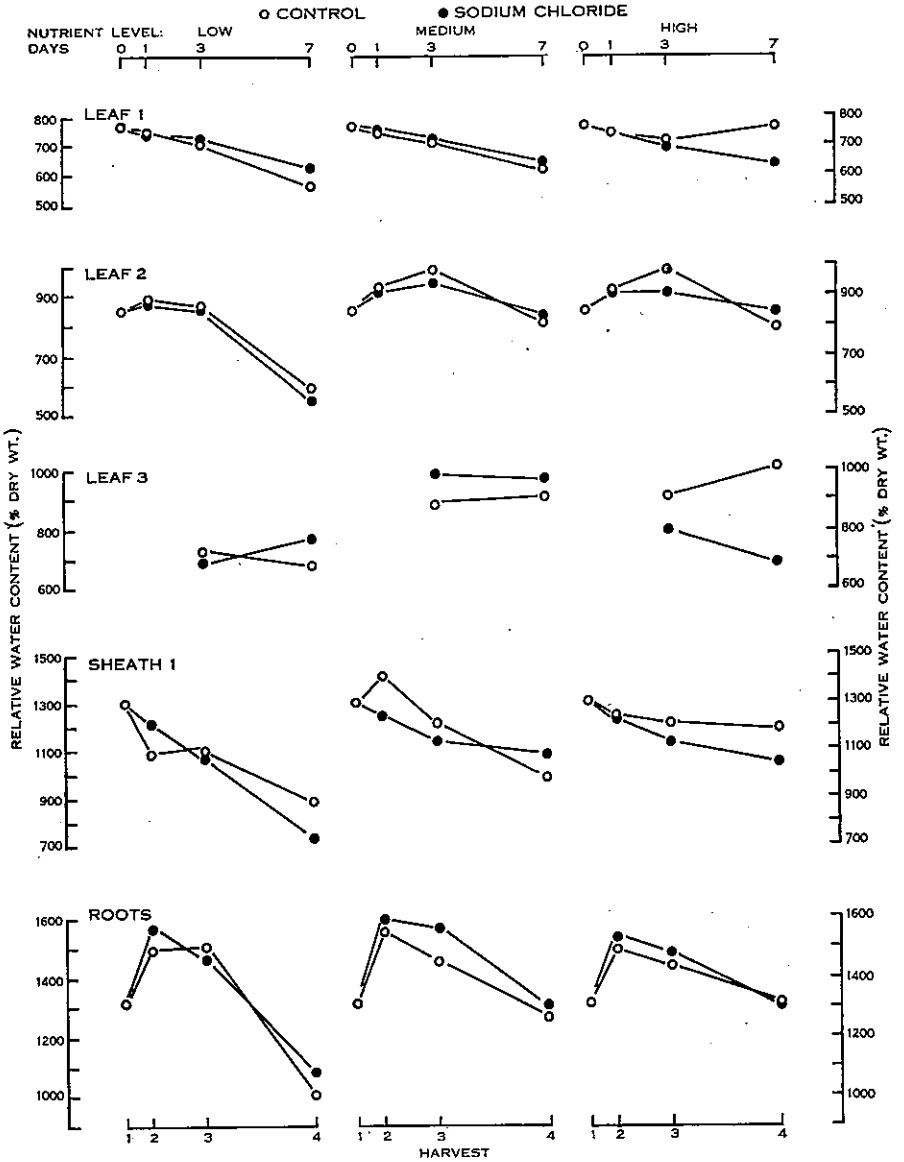

Fig. 2.—Relative water content of the individual plant parts.

retarded by this treatment, but adverse effects were relatively most pronounced in the youngest leaf (leaf 3). At the medium and high nutrient levels leaf 3 was the only plant part which was reduced somewhat in dry weight by high sodium chloride treatment.

It should be mentioned here that in the low nutrient, high sodium chloride treatment the roots developed a distinct brown discoloration after 2–3 days of treatment. Similar root symptoms were reported for green beans which were grown in soil with a high percentage of exchangeable sodium (Bernstein and Pearson 1956).

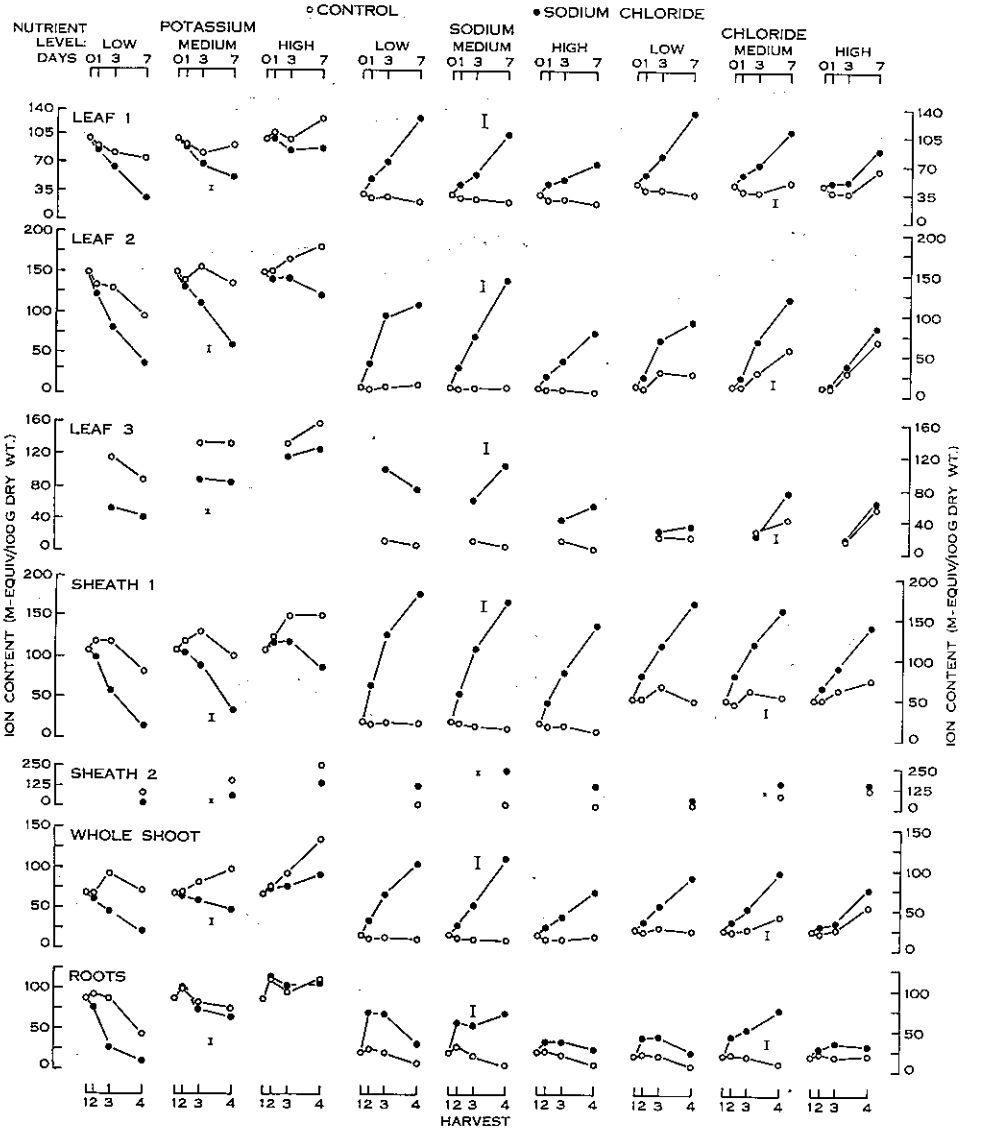

Fig. 3.—Relative amounts of potassium, sodium, and chloride as affected by sodium chloride treatment at three different nutrient levels. Significant differences at $P = 0 \cdot 05$ are indicated.

There were few consistent treatment effects on the relative water contents (Fig. 2). However, at the low nutrient level plants tended to have lower relative water contents than the higher nutrient plants.

(b) *Ion Contents*

Ion contents (in m-equiv/100 g dry wt.) of the various plant parts are shown in Figure 3, and absolute potassium contents of these parts (as μ-equiv/plant part) are presented in Figure 4. Chloride contents of plant water (in m-equiv/l) are presented in Figure 5.

(i) *Potassium Contents*

The relative potassium contents of the low sodium chloride shoots did not change consistently at low nutrient level, but they increased with time at medium nutrient and particularly at high nutrient level (Fig. 3). Potassium contents of the individual plant parts usually decreased with time at low nutrient, changed little at medium nutrient, and increased considerably with time at high nutrient level.

High sodium chloride treatment reduced the potassium contents of the whole shoot below those of the corresponding low sodium chloride treatment. In the roots, on the other hand, high sodium chloride substantially reduced potassium contents only in the case of the low nutrient plants (Fig. 3). In the sodium chloride-treated plants, potassium contents of the individual parts decreased markedly with time at the low nutrient level, but they did not change appreciably with time at the high nutrient level. Decreases at the low and medium nutrient levels were least pronounced in the youngest leaf, leaf 3 (Fig. 3).

Absolute potassium contents of the low sodium chloride shoots increased steeply throughout the experiment, and most plant parts contributed to these increases. However, little change occurred in the roots at the low nutrient level and in leaf 1 at both the low and medium nutrient level (Fig. 4).

In the high sodium chloride treatments, substantial net potassium losses occurred from both the shoots and roots of the low nutrient plants, and these losses took place throughout the course of the experiment. On the other hand, net potassium contents of both shoots and roots of the medium and high nutrient plants increased with time. In the individual parts, leaf 1 and sheath 1 showed very pronounced net potassium losses at low nutrient, smaller net losses at medium nutrient, and small net gains at high nutrient levels. Leaves 2 and 3, on the other hand, increased slightly in potassium even at the low nutrient level ($P < 0 \cdot 05$), while at the higher nutrient levels the same plant parts increased strongly in potassium content.

Potassium lost from the whole plant at the low nutrient, high sodium chloride treatment was presumably returned to the substrate. This is indicated by the following increases in the potassium contents of the solutions:

Sampling Time	Plants per Container	K^+ Increase in Solution (μ-equiv/l/day)
H_1+1 day	16	21
H_1+3 days	4	7

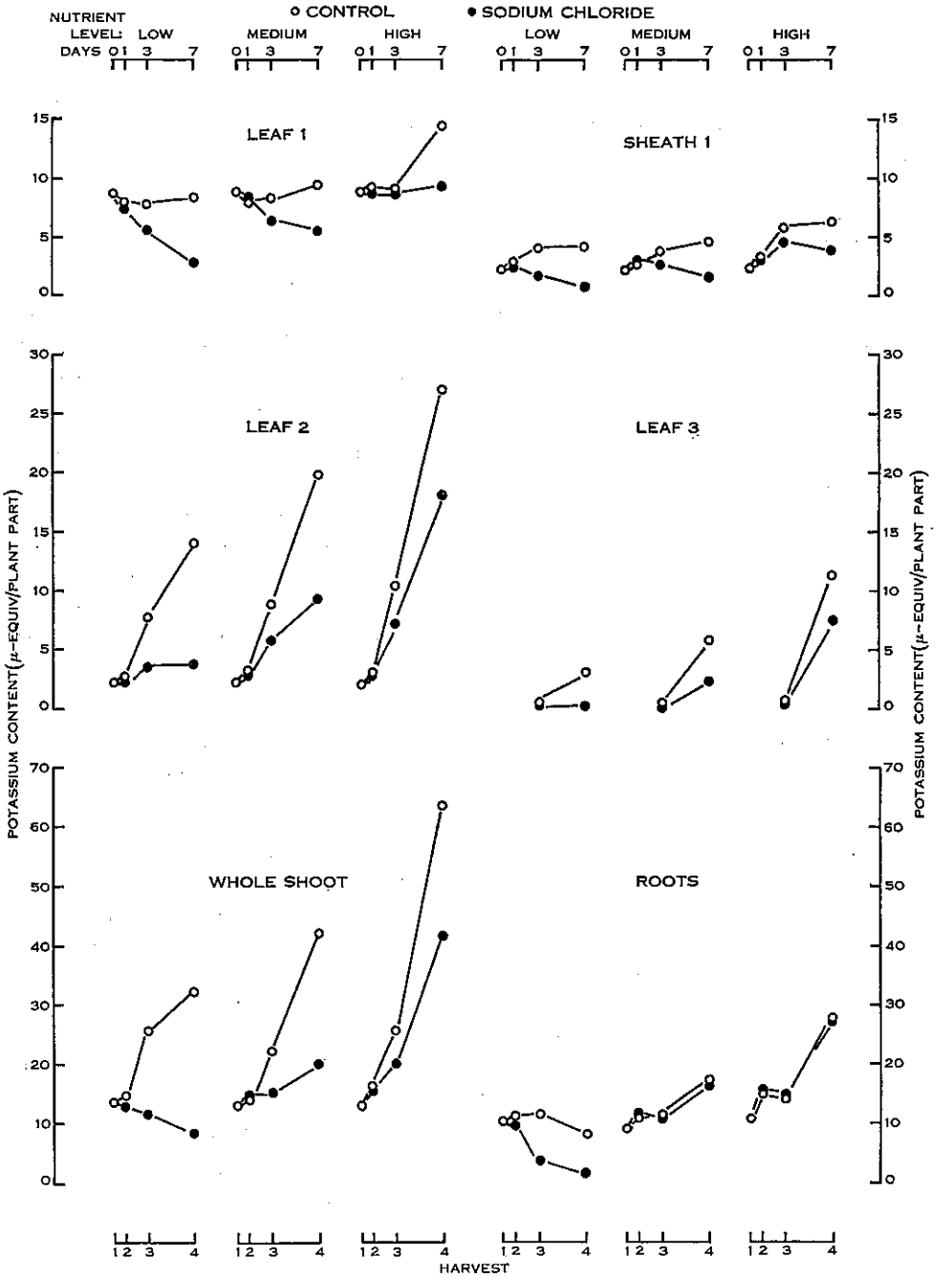

Fig. 4.—Absolute amounts of potassium in individual plant parts as affected by sodium chloride treatment at three different nutrient levels. Statistical analysis was carried out after logarithmic transformation. For the individual plant parts all decreases which were significant ($P < 0 \cdot 05$) are:

[*For continuation see opposite page*]

(ii) *Chloride and Sodium Contents*

In the low sodium chloride plants, sodium contents were very low, particularly at the medium and high nutrient levels (Fig. 3). Chloride accumulated to much higher levels than those of the substrate (Fig. 5) and at the high nutrient level chloride concentrations were often not greatly below those of the corresponding high sodium chloride plants (Fig. 3). These observations suggest a further study on the chloride uptake with a range of chloride levels at each nutrient concentration.

In the high sodium chloride treatments, chloride and sodium tended to be higher in the older than in the younger plant parts (Fig. 3) and this was in agreement with previous experiments (Greenway 1962*b*; Greenway and Rogers 1963).

Chloride and sodium contents of the shoots of the high sodium chloride plants usually increased regularly with time. However, at the low nutrient level, the rate of chloride accumulation decreased with time. Chloride and sodium contents rapidly increased during the first day of treatment, while the rate of increase was very much slower during the rest of the experiment. At the low nutrient level, chloride and sodium contents of the roots even decreased with time.

In the high sodium chloride treatments, chloride and sodium contents of all parts of the shoot generally decreased in the order low, medium, and high nutrient level. However, at H_4, leaves 2 and 3 and sheath 2 contained less chloride and sodium at low than at medium nutrient level (Fig. 3). Chloride concentrations in the plant water also usually decreased from low to high nutrient plants, the exception being leaf 3 at H_4 when chloride contents were lowest in the low nutrient plants (Fig. 5). It may be noted here that chloride contents are very low during the early stage of development of a plant part (e.g. leaf 3 of all high sodium chloride plants at H_3). It is thus likely that at H_4 the very low chloride levels in leaf 3 of the low nutrient, high sodium chloride plants were related to its strongly retarded development (see Fig. 1).

Figure 3 shows that in the high sodium chloride plants, the low nutrient level treatment always resulted in lower chloride content of the roots than the medium nutrient level treatment and at H_4 low nutrient roots were even lower in chloride content than high nutrient roots (Fig. 3). At both H_2 and H_3, sodium contents decreased in the order low, medium, and high nutrient level. However, at H_4 the sodium content of the low nutrient roots was less than that of the medium nutrient roots (Fig. 3).

Absolute amounts of sodium and chloride in the roots are presented in Figure 6. The net losses of chloride and sodium from the low nutrient, high sodium chloride roots are noteworthy, particularly in view of the high substrate concentration. It should be remembered that the health of these roots declined during the experiment and this injury might have lead to ion losses during the rinsing with distilled water at harvest.

Fig. 4 (*Continued*)

Sodium chloride, low nutrient level: $H_4 < H_3 < H_2$ for leaf 1, sheath 1, and the roots.
Sodium chloride, medium nutrient level: $H_3 < H_2$ for leaf 1; $H_4 < H_3$ for sheath 1.
Control, low nutrient: $H_4 < H_3$ for the roots.

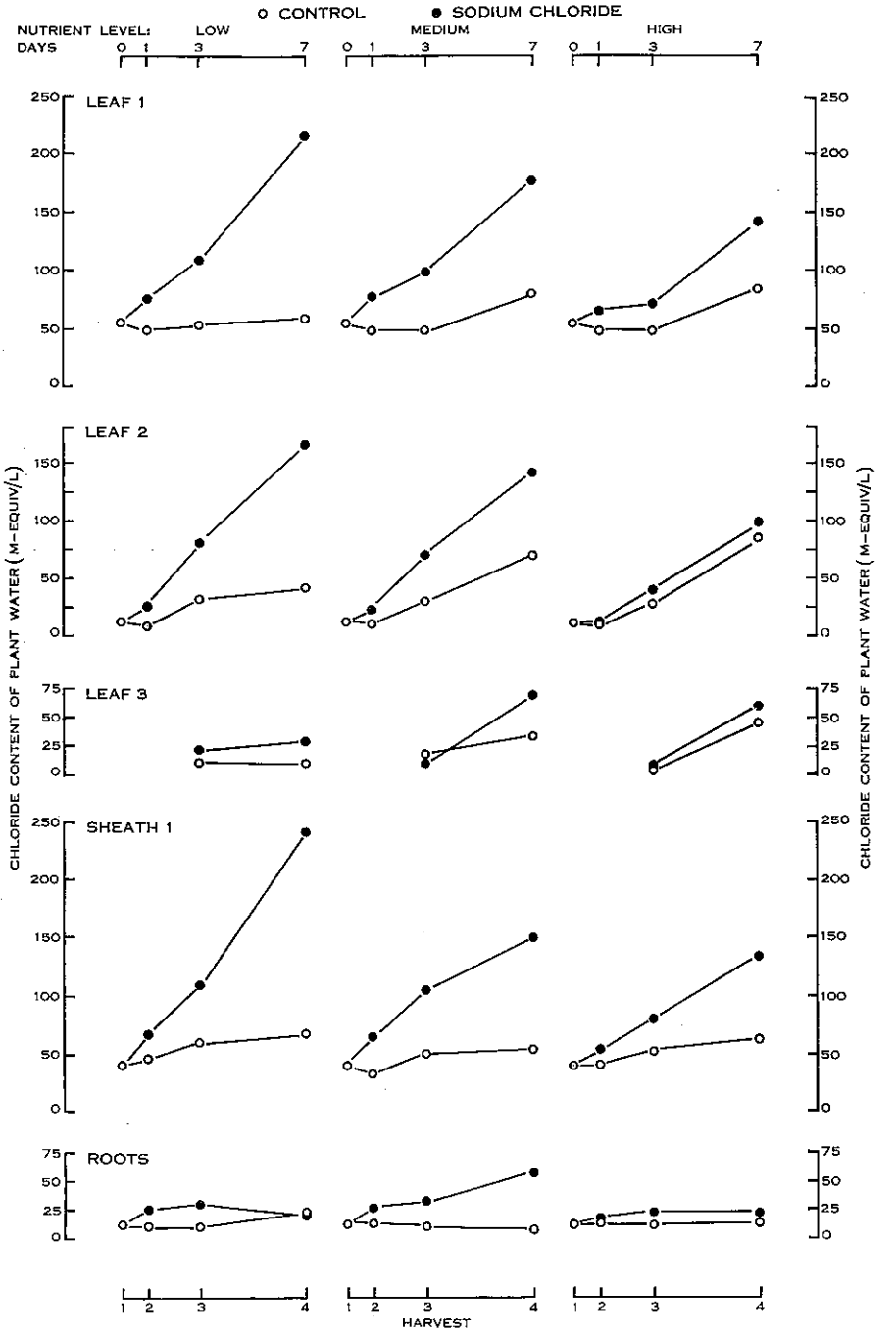

Fig. 5.—Chloride in the plant water as affected by sodium chloride treatment at three different nutrient levels.

IV. Discussion

(a) *Growth*

At the medium and high nutrient levels, growth of this salt-tolerant variety of *H. vulgare* was not appreciably affected by a sodium chloride treatment of 50 m-equiv/l (Fig. 1). This is in agreement with the reputation of *H. vulgare* as a very salt-tolerant species (Richards 1954). However, at the low nutrient level growth was substantially reduced by sodium chloride treatment. This finding emphasizes that the salt tolerance of species, or varieties, is only a relative concept and that other experimental conditions should be clearly defined.

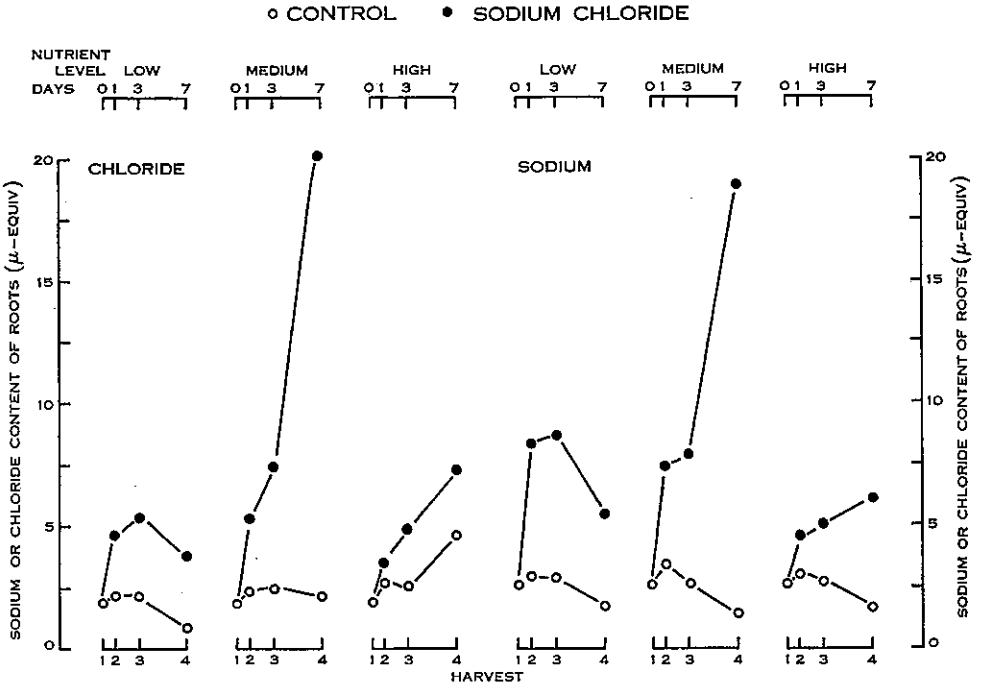

Fig. 6.—Absolute amounts of chloride and sodium in the roots of a single plant as affected by sodium chloride treatment at three different nutrient levels.

This rather large adverse effect was presumably due to the very high sodium and chloride contents, and to the very low potassium contents, of the high sodium chloride, low nutrient plants (Fig. 3). Such an adverse effect of high chloride and sodium, as well as low potassium contents, was previously suggested for a salt-sensitive variety of *H. vulgare* treated with 150 m-equiv/l of sodium chloride on a substrate of high nutrient concentration (Greenway 1962*a*). This suggestion is now supported by the fact that the shoots of that sensitive variety had similar ion relations to the shoots of the salt-treated, low nutrient plants of the present experiment. However, for the present experiment, it should be noted that the low nutrient level only contained very low levels of all the essential elements. These low concentrations of ions, other than potassium, might also have contributed to the strong adverse effect of the high sodium chloride treatment.

(b) *Chloride and Sodium Uptake*

The particularly high chloride and sodium contents of the low nutrient, high sodium chloride plants are in agreement with data for sodium presented by Heimann and Ratner (1961), and for sodium and chloride by Reifenberg and Rosovsky (1947). These workers showed that in their experiments the reduced sodium uptake was associated with an increased potassium concentration of the substrate. In all high sodium chloride treatments the chloride concentration in the plant water of the youngest leaf was well below substrate level, until it commenced its rapid development (leaf 3 at H_3 and at the low nutrient level at H_4 (Fig. 5)). The chloride concentrations of individual plant parts rapidly increased with time, so that they attained levels higher than the substrate, and particular high levels were found in leaves 1 and 2. These observations are in agreement with those during previous experiments (Greenway 1962*b*; Greenway and Rogers 1963).

These data cast some doubt on the conclusion of Slatyer (1961) that after salt application osmotic pressure of plants is adequately adjusted by rapid electrolyte absorption. Slatyer used pooled leaf samples of the whole shoot, but a more complicated picture might be expected when more individual plant parts are examined. Figure 5 indicates that in barley the osmotic pressure adjustment of the very young parts cannot be accounted for by rapid absorption of chlorides supplied in the substrate.

(c) *Potassium Uptake and Loss*

In the present experiments, considerable net losses of potassium occurred from both roots and shoots of the high sodium chloride treatment at the low nutrient level.* These net losses took place although there was a constant and readily available potassium supply, at a level sufficient to give considerable net potassium increments in the low nutrient, low sodium chloride plants.

Potassium losses from the whole plant were found by Lündegårdh, Bürstrom, and Rennerfelt (1932) for wheat and peas and by Elgabaly (1955) for barley; however, these workers grew the plants on a potassium-free substrate.

The data presented in this paper suggest strongly that, under certain conditions, a considerable circulation of potassium between shoots and substrate can occur. A similar circulation of phosphate for intact barley plants was found by Helder (1958), but he could not find evidence for a rubidium transport from the shoot back to the root. It should be added that the phosphate losses found by Helder were compensated by a much greater phosphate uptake, while in the present experiment net losses of potassium occurred.

In the present experiment net losses of potassium were also found from leaf 1 and sheath 1 of the high sodium chloride plants at the medium nutrient level. Similar net potassium losses from older parts have been found for *H. vulgare* plants at high nutrient level and treated with 100 m-equiv/l of sodium chloride (Greenway 1962*b*). Such observations suggest that potassium losses from the shoot via the roots to the

* The losses from the roots might have been, at least partly, due to the injury described earlier. The losses from the shoots are, therefore, of more significance than those from the roots.

substrate might also occur at higher nutrient levels, at least from sodium chloride-treated plants. If so, then the efflux from the shoot must have been compensated by a higher influx.

Sutcliffe (1956) grew young barley plants at low, but equivalent, concentrations of potassium and sodium and found that the preferential absorption of potassium over sodium was more pronounced in the shoots than in the roots. The comparable treatment in the present investigation (i.e. low nutrient control) also showed a higher potassium–sodium ratio in the young leaves 2 and 3 than in the roots. On the other hand, the reduction of potassium contents below the values of low sodium chloride plants due to high sodium chloride treatment was much more pronounced in the shoots than in the roots at both the medium and high nutrient levels.

In the present experiment, the reduction of potassium content, due to the presence of high sodium chloride, was rather similar in magnitude at all nutrient levels, and this was despite the large differences in sodium–potassium ratios of the substrates. Similarly, Bange (1959) found that the magnitude of sodium effects on potassium absorption of maize seedlings were independent of potassium concentration in the substrate, provided the substrate contained more than 0·2 m-equiv/l of potassium.

The presented data differ from those of Heimann and Ratner (1961) who found no marked effect of sodium chloride on potassium uptake. This difference supports the conclusion of Bange (1959) that sodium–potassium interactions are dependent on a number of experimental conditions, which would include the composition of the basal nutrient solution, the species, the age of the plant, and other factors.

V. Acknowledgments

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VI. References

Bange, G. G. J. (1959).—Interactions in the potassium and sodium absorption by intact maize seedlings. *Plant & Soil* **11**: 17–29.

Bernstein, L., and Pearson, G. A. (1956).—Influence of exchangeable sodium on the yield and chemical composition of plants. I. Green beans, garden beets, clover and alfalfa. *Soil Sci.* **82**: 247–58.

Best, R. J. (1950).—A rapid and accurate electrotitrimetric method for determining the chloride contents of soils, water, and plant materials. Trans. 4th Int. Congr. Soil Sci., Amsterdam. Vol. 3. pp. 162–4.

Elgabaly, M. M. (1955).—Specific effects of absorbed ions on plant growth. I. Effect of different combinations of calcium, magnesium, and sodium on barley seedlings. *Soil Sci.* **80**: 235–48.

Greenway, H. (1962*a*).—Plant response to saline substrates. I. Growth and ion uptake of several varieties of *Hordeum* during and after sodium chloride treatment. *Aust. J. Biol. Sci.* **15**: 16–38.

GREENWAY, H. (1962*b*).—Plant response to saline substrates. II. Chloride, sodium, and potassium uptake and translocation in young plants of *Hordeum vulgare* during and after a short sodium chloride treatment. *Aust. J. Biol. Sci.* **15**: 39–57.

GREENWAY, H., and ROGERS, A. (1963).—Growth and ion uptake of *Agropyron elongatum* as compared with a salt-tolerant variety of *Hordeum vulgare*. *Plant & Soil* **18**: 21–30.

HEIMANN, H., and RATNER, R. (1961).—The influence of potassium on the uptake of sodium by plants under saline conditions. *Bull. Res. Coun. Israel* A **10**: 55–62.

HELDER, R. J. (1958).—Exchange and circulation of labelled ions in young intact plants. In: "Radioisotopes in Scientific Research". Vol. 4. pp. 369–81. (Pergamon Press: London.)

HEWITT, E. J. (1952).—Sand and water cultures in the study of plant nutrition. Tech. Commun. Bur. Hort. E. Malling. No. 22.

LÜNDEGÅRDH, H. G. (1951).—"Leaf Analysis". (Translation of "Die Blattanalyse" by R. L. Mitchell.) (Hilger and Watts: London.)

LÜNDEGÅRDH, H. G., BÜRSTROM, H., and RENNERFELT, E. (1932).—Untersuchungen über die salzaufnahme der pflanzen. II. Die aufnahme von alkali and earth-alkali chlorides. *Svensk. Bot. Tidskr.* **26**: 271–83.

REIFENBERG, A., and ROSOVSKY, R. (1947).—Saline irrigation water and its effect on the intake of ions by barley seedlings. *Palest. J. Bot. Jerusalem* **4**: 1–13.

RICHARDS, L. A. (ED.) (1954).—"Diagnosis and Improvement of Saline and Alkali Soils." (Handb. U.S. Dep. Agric. No. 60.)

SLATYER, R. O. (1961).—Effects of several osmotic substrates on the water relationships of tomato. *Aust. J. Biol. Sci.* **14**: 519–40.

SUTCLIFFE, J. F. (1956).—The selective absorption of alkali cations by storage tissues and intact barley plants. In "Potassium Symposium". Annu. Mtg. Bd. Tech. Advisors, London, pp. 1–11. (Int. Potash Inst.: Berne.)