Evaluation of Critical K Concentration Expressed on a Tissue Water Basis in Spinach¹

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Summary

Potassium fertilizer experiments of spinach were carried out to clarify the effects of growing season and plant age on the critical K concentration expressed as % dry weight or % tissue water.

The relationships between the dry weight of tops and K concentration of the youngest leaf were usually delineated by the second or the third degree polynomials.

At the sixth leaf stage, the critical K concentration of the youngest leaf ranged from 4.3 to 6.7% when expressed on a dry weight basis. When expressed on a tissue water basis, it was not variable with about 0.7%, except after plants had been chilled.

In early summer, the critical K concentration of the youngest leaf was kept constant at about 0.7% on a tissue water basis, but that on a dry weight basis decreased from the fifth to the seventh leaf stage.

Introduction

Plant analysis has long been used as a diagnostic tool for assessing the nutritional status of the crop. As a basis to interpret the result of plant analysis, we have to determine the critical nutrient concentration below which growth responses to the application of the nutrient are observed. The critical concentrations are desired to be constant throughout the growing season and under different environmental conditions(1). However, plant age(5) and environmental conditions such as temperature(13) and soil moisture supply(9) altered the critical K concentration on a dry weight basis.

The nutrient concentrations have usually been expressed on a dry weight basis in plant analysis(6), but some authors(2, 3, 4, 8, 10) disapproved of this conventional method of expression, especially for expressing K concentration. Cassidy(2, 3) stated that the concentrations of water soluble nutrients such as Cl and K should be expressed on a tissue water basis to reveal their true

distribution in plants. Leigh *et al.* (8) found that fresh weight : dry weight ratio and K concentration relative to dry weight decreased with aging of spring barley, but K concentration relative to tissue water was substantially constant throughout its vegetative growth. They presumed that the critical K concentration on a tissue water basis would be constant irrespective of plant age.

The purposes of this study were to determine whether the critical K concentration on a tissue water basis would be more constant than that on a dry weight basis or not, regardless of the growing season and plant age.

Materials and Methods

K fertilizer experiments with a randomized block design were carried out in 1979, 1981 and 1982. Pots of 15 l capacity were used, each containing 6.5 kg (oven-dry basis) of the volcanic ash soil. The soil had a pH of 6.0 in 1 : 2.5 water suspension, a CEC of 29.7 me/100 g and a exchangeable K of 135 ppm. K treatments, which were replicated 4 times, consisted of 6 rates of K₂SO₄ (0, 0.5, 1, 2, 4 and 8 g per pot). All pots

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received 10 g of $(\text{NH}_4)_2\text{SO}_4$, 40 g of super-phosphate and 10 g of $\text{Ca}(\text{OH})_2$.

Seeds of spinach (*Spinacia oleracea* L.) cv 'Yuho' were sown and the seedlings were thinned to 3 per pot after establishment. Plants were watered as needed throughout the growing season.

In 1979, plant tops were harvested at the sixth leaf stage in each of the 4 experiments. Sowing dates were May 12, May 24, Oct. 26 and Nov. 2. Corresponding harvest date for each sowing date was June 7, June 16, Dec. 13 and Dec. 26, respectively. In 1981, seeds were sown on Oct. 12 in 12 pots each in 6 K treatments, totalling 72 pots. Plant tops from each of 4 pots in each K treatment were harvested separately on Nov. 21, Nov. 30 and Dec. 12 (the fifth, the sixth and the seventh leaf stage, respectively). 1982 experiment was the same as 1981 experiment except that seeds were sown on May 18 and plant tops were harvested on June 9 and 13 (the fifth and the seventh leaf stage, respectively).

Plant tops were harvested at dawn and separated into the youngest leaf, the largest leaf (usually the fourth leaf from the base) and the residual parts. The leaf of which petiole begins to appear is called the youngest leaf in this paper. After determination of fresh weight, leaf samples were dried at 70°C in a forced-air oven and weighed. Then they were ground, dry-ashed, taken up in HCl and analyzed for K by flame-photometry. K concentration of the leaf was expressed both on a dry weight basis and a tissue water basis. Tissue water was calculated as the difference between fresh and dry weight.

Air temperature at a 1.2 m height above-ground was measured by a thermally sensitive resistor.

Data were subjected to a factorial analysis of variance before the least significant differences were calculated. Regressions up to the fifth order were calculated to relate the dry weight of tops to K concentration of the youngest leaf and the largest leaf. The adequate-fitted polynomials of the lowest degree were found according to the method

of Snedecor and Cochran(11). If the non-linear polynomials were obtained, K concentration corresponding to 90% of the maximum dry weight was calculated and designated as the critical concentration.

Results

1979 experiment

Mean temperatures during the experiments

Table 1. Effects of K application on the dry weight of spinach tops (g/plant) grown in early summer and late autumn.

K ₂ SO ₄ applied (g/pot)	Experimental no. ²			
	1	2	3	4
0	0.33 a A ^y	0.20 a B	0.33 a A	0.42 a A
0.5	0.54 b A	0.44 b B	0.59 b A	0.60 b A
1	0.78 c A	0.56 c C	0.65 bc BC	0.69 bc AB
2	1.08 d A	0.72 d B	0.75 cd B	0.79 cd B
4	1.24 e A	0.87 e B	0.80 d B	0.86 d B
8	1.21 e A	0.85 e B	0.80 d B	0.85 d B

² Growing periods for experiment 1, May 12-June 7; for experiment 2, May 24-June 16; for experiment 3, Oct. 26-Dec. 13; for experiment 4, Nov. 2-Dec. 26.

^y Mean separation in columns (a-e) or in rows (A-B) by LSD test, 5% level.

Table 2. Effects of K application on K concentration of the youngest leaf (% dry weight and % tissue water) in spinach grown in early summer and late autumn.

K ₂ SO ₄ applied (g/pot)	Experimental no. ²			
	1	2	3	4
% dry weight				
0	3.19 a B ^y	2.68 a BC	2.25 a C	3.95 a A
0.5	4.41 b A	3.43 b B	2.87 b C	4.36 ab A
1	4.98 b A	4.04 c B	3.35 b C	4.73 b A
2	6.23 c A	5.30 d B	4.47 c C	6.21 c A
4	7.68 d A	7.04 e B	5.60 d C	7.10 d B
8	8.82 e A	8.01 f B	6.60 e C	7.62 d B
% tissue water				
0	0.40 a B	0.36 a B	0.41 a B	0.58 a A
0.5	0.51 b B	0.43 b C	0.49 b B	0.63 ab A
1	0.57 b B	0.50 c C	0.57 c B	0.69 b A
2	0.65 c BC	0.62 d C	0.71 d B	0.82 c A
4	0.77 d C	0.76 e C	0.86 e B	0.92 d A
8	0.86 e B	0.85 f B	0.96 f A	0.99 e A

² Growing periods for experiment 1, May 12-June 7; for experiment 2, May 24-June 16; for experiment 3, Oct. 26-Dec. 13; for experiment 4, Nov. 2-Dec. 26.

^y Mean separation in columns (a-f) or in rows (A-C) by LSD test, 5% level.

Fig. 1. The relationship between the dry weight of tops and K concentration (% dry weight) of the youngest leaf in spinach grown in early summer and late autumn.

Symbols: ■—■ Experiment 1; ▲—▲ Experiment 2; □—□ Experiment 3; △—△ Experiment 4. Arrows indicate the critical K concentration. Regression equations for each experiment together with their critical concentrations (%) are:

Experiment	Equation	R ²	Critical conc.
1	$Y = -0.00885 X^3 + 0.133 X^2 - 0.416 X + 0.607$	0.87	6.73
2	$Y = -0.0298 X^2 + 0.435 X - 0.725$	0.96	5.60
3	$Y = -0.0343 X^2 + 0.402 X - 0.343$	0.86	4.31
4	$Y = -0.0288 X^2 + 0.433 X - 0.764$	0.81	5.79

Table 3. The relationship between the dry weight of tops (Y, g) and K concentration (X, % dry weight) of the largest leaf in spinach grown in early summer and late autumn.

Experimental no. ^z	Regression equation	R ²	Critical conc. ^y
1	$Y = -0.00212 X^3 + 0.0358 X^2 - 0.0409 X + 0.182$	0.86	8.52
2	$Y = -0.013 X^2 + 0.242 X - 0.268$	0.95	6.73
3	$Y = -0.0141 X^2 + 0.216 X - 0.019$	0.88	5.26
4	$Y = -0.0103 X^2 + 0.207 X - 0.181$	0.81	7.16

^z Growing periods for experiment 1, May 12-June 7; for experiment 2, May 24-June 16; for experiment 3, Oct. 12-Dec. 13; for experiment 4, Nov. 2-Dec. 26.

^y K concentration (% dry weight) corresponding to 90% of the maximum dry weight.

Fig. 2. The relationship between the dry weight of tops and K concentration (% tissue water) of the youngest leaf in spinach grown in early summer and late autumn.

Symbols: ■—■ Experiment 1; ▲—▲ Experiment 2; □—□ Experiment 3; △—△ Experiment 4. Arrows indicate the critical K concentration. Regression equations for each experiment together with their critical concentrations (%) are:

Experiment	Equation	R ²	Critical conc.
1	$Y = -18.4 X^3 + 31.2 X^2 - 14.5 X + 2.33$	0.87	0.68
2	$Y = -2.50 X^2 + 4.25 X - 0.949$	0.90	0.66
3	$Y = -1.90 X^2 + 3.36 X - 0.665$	0.82	0.68
4	$Y = -2.75 X^2 + 5.21 X - 1.62$	0.82	0.77

Table 4. The relationship between the dry weight of tops (Y, g) and K concentration (X, % tissue water) of the largest leaf in spinach grown in early summer and late autumn.

Experimental no. ^z	Regression equation	R ²	Critical conc. ^y
1	$Y = -10.1 X^3 + 18.6 X^2 - 8.98 X + 1.61$	0.89	0.77
2	$Y = -1.54 X^2 + 2.87 X - 0.47$	0.91	0.69
3	$Y = -0.925 X^2 + 1.94 X - 0.216$	0.87	0.75
4	$Y = -1.26 X^2 + 2.73 X - 0.633$	0.83	0.82

^z Growing periods for experiment 1, May 12-June 7; for experiment 2, May 24-June 16; for experiment 3, Oct. 12-Dec. 13; for experiment 4, Nov. 2-Dec. 26.

^y K concentration (% tissue water) corresponding to 90% of the maximum dry weight.

1, 2, 3 and 4 were 20, 22, 13 and 12°C, respectively. Daily minimum temperatures dropped often to nearly 0°C after Dec. 14.

The dry weight of tops increased with increasing rates of K_2SO_4 , but levelled off at 4 g in early summer and at 2 g in late autumn (Table 1). The dry weight of tops receiving K_2SO_4 above 2 g was greater in the experiment 1 than in the others, but the reason for this difference is not clear.

K concentration of the youngest leaf, either expressed on a dry weight basis or a tissue water basis, increased with increasing rates of K_2SO_4 in both seasons (Table 2). For the same rate of K_2SO_4 , K concentration on a dry weight basis was lower in the experiment 3 than in the others. When expressed on a tissue water basis, however, K concentration of the youngest leaf was higher in late autumn, especially in the experiment 4, than in early summer. K concentration of the largest leaf showed the similar trends although data were not presented in this paper.

The relationships between the dry weight of tops and K concentration of the youngest leaf, either expressed on a dry weight basis or a tissue water basis, are satisfactorily delineated by the second or the third degree polynomials (Figs. 1 and 2). The critical K concentrations of the youngest leaf on a dry weight basis ranged from 4.3 to 6.7%, but those on a tissue water basis were constant between 0.66 and 0.68% except in the experiment 4. Except in the experiment 4, the critical K concentration of the largest leaf ranged from 5.3 to 8.5% when expressed on a dry weight basis (Table 3), and from 0.69 to 0.77% when expressed on a tissue water basis (Table 4).

1981 experiment

Mean temperature during the experiment was 11°C. After the first harvest, daily minimum temperatures dropped often to nearly 0°C.

The dry weight of tops increased with increasing rates of K_2SO_4 , but levelled off at 2 g at all 3 stages (Table 5). K concentration of the youngest leaf, either expressed on a dry weight basis or a tissue

Table 5. Effects of K application on the dry weight of spinach tops (g/plant) at 3 stages of growth.

K_2SO_4 applied (g/pot)	Harvest date ^a			
	Nov. 21	Nov. 30	Dec. 12	Average
0	0.35	0.75	1.14	0.75 a ^y
0.5	0.47	0.85	1.61	0.97 b
1	0.52	1.02	1.77	1.10 b
2	0.70	1.18	2.10	1.33 c
4	0.66	1.18	2.04	1.30 c
8	0.69	1.20	2.09	1.33 c
Average	0.56 A	1.03 B	1.79 C	

^a Plants were sown on Oct. 12 and harvested at the 5th, 6th and 7th leaf stage, respectively.

^y Mean separation for average values by LSD test, 5% level. $K \times$ Harvest date interaction is not significant.

Table 6. Effects of K application on K concentration of the youngest leaf (% dry weight and % tissue water) at 3 stages of growth.

K_2SO_4 applied (g/pot)	Harvest date ^a			
	Nov. 21	Nov. 30	Dec. 12	Average
% dry weight				
0	3.35	3.72	2.88	3.32 a ^y
0.5	3.98	4.52	3.83	4.11 b
1	4.77	5.40	4.57	4.91 c
2	5.29	6.33	5.25	5.62 d
4	6.22	6.60	5.76	6.19 e
8	6.41	7.08	6.07	6.52 f
Average	5.00 B	5.61 A	4.72 C	
% tissue water				
0	0.49	0.56	0.49	0.51 a
0.5	0.58	0.68	0.62	0.63 b
1	0.66	0.79	0.73	0.73 c
2	0.73	0.90	0.83	0.82 d
4	0.82	0.94	0.91	0.89 e
8	0.88	0.98	0.96	0.94 f
Average	0.69 C	0.81 A	0.76 B	

^a Plants were sown on Oct. 12 and harvested at the 5th, 6th and 7th leaf stage, respectively.

^y Mean separation for average values by LSD test, 5% level. $K \times$ Harvest date interaction is not significant.

water basis, increased with increasing rates of K_2SO_4 at all 3 stages (Table 6). For the same rate of K_2SO_4 , K concentration on a dry weight basis was the highest at the second stage and the lowest at the third stage. K concentration on a tissue water basis was the highest at the second stage and the lowest at the first stage. Although data were not presented, K concentration

Fig. 3. The relationship between the dry weight of tops and K concentration (% dry weight) of the youngest leaf at 3 stages of growth in spinach.

Symbols: Δ — Δ Nov. 21; $\circ$ — $\circ$ Nov. 30; $\blacksquare$ — $\blacksquare$ Dec. 12.

Arrows indicate the critical K concentration. Regression equations for each harvest together with their critical concentrations (%) are:

Harvest date	Equation	R ²	Critical conc.
Nov. 21	$Y = -0.0293 X^2 + 0.388 X - 0.599$	0.74	5.09
Nov. 30	$Y = -0.0123 X^3 + 0.188 X^2 - 0.782 X + 1.69$	0.74	5.90
Dec. 12	$Y = -0.0827 X^2 + 1.03 X - 1.11$	0.70	4.57

Table 7. The relationship between the dry weight of tops (Y, g) and K concentration (X, % dry weight) of the largest leaf at 3 stages of growth in spinach.

Harvest date ^z	Regression equation	R ²	Critical conc. ^y
Nov. 21	$Y = -0.0125 X^3 + 0.195 X^2 - 0.87 X + 1.53$	0.79	6.07
Nov. 30	$Y = 0.101 X + 0.357$	0.72	— ^x
Dec. 12	$Y = -0.0257 X^2 + 0.474 X - 0.0417$	0.70	6.34

^z Plants were sown on Oct. 12 and harvested at the 5 th, 6 th, and 7 th leaf stage, respectively.

^y K concentration (% dry weight) corresponding to 90% of the maximum dry weight.

^x Critical K concentration can not be calculated because the regression is linear.

Fig. 4. The relationship between the dry weight of tops and K concentration (% tissue water) of the youngest leaf at 3 stages of growth in spinach.

Symbols: Δ — Δ Nov. 21; $\circ$ — $\circ$ Nov. 30; $\blacksquare$ — $\blacksquare$ Dec. 12.

Arrows indicate the critical K concentration.

Regression equations for each harvest together with their critical concentrations (%) are:

Harvest date	Equation	R ²	Critical conc.
Nov. 21	$Y = -5.31 X^3 + 9.67 X^2 - 4.73 X + 0.998$	0.74	0.73
Nov. 30	$Y = -7.89 X^3 + 18.3 X^2 - 12.8 X + 3.54$	0.72	0.85
Dec. 12	$Y = -20.3 X^3 + 40.7 X^2 - 24.2 X + 5.69$	0.67	0.75

Table 8. The relationship between the dry weight of tops (Y, g) and K concentration (X, % tissue water) of the largest leaf at 3 stages of growth in spinach.

Harvest date ^z	Regression equation	R ²	Critical conc. ^y
Nov. 21	$Y = 0.748 X - 0.006$	0.78	— ^x
Nov. 30	$Y = -4.16 X^3 + 10.7 X^2 - 7.94 X + 2.59$	0.71	0.97
Dec. 12	$Y = -1.25 X^2 + 3.36 X - 0.119$	0.67	0.93

^z Plants were sown on Oct. 12 and harvested at the 5 th, 6 th and 7 th leaf stage, respectively.

^y K concentration (% tissue water) corresponding to 90% of the maximum dry weight.

^x Critical K concentration can not be calculated because the regression is linear.

of the largest leaf showed the similar patterns to those of the youngest leaf with the progress of growth stage.

The relationships between the dry weight of tops and K concentration of the youngest leaf, either expressed on a dry weight basis or a tissue water basis, are satisfactorily delineated by the second or the third degree polynomials (Figs. 3 and 4). The critical K concentrations of the youngest leaf on a dry weight basis ranged from 4.6 to 5.9%, and those on a tissue water basis ranged from 0.73 to 0.85%.

At the second stage, the dry weight of tops related linearly to K concentration of the largest leaf on a dry weight basis so that the critical concentration could not be determined (Table 7). At the first stage, a linear relationship also existed between the dry weight of tops and K concentration of the largest leaf on a tissue water basis (Table 8).

1982 experiment

Mean temperature during the experiment was 21.5°C and daily minimum temperatures seldom dropped below 10°C.

Although data were not presented, the dry weight of tops increased with increasing rates of K_2SO_4 , but levelled off at 2 g at both stages. The critical K concentrations of the youngest leaf on a dry weight basis decreased from 7.0 to 5.9% with the progress of growth stage, but those on a tissue water basis changed little (0.71 and 0.74% at the first and the second stage, respectively). The critical K concentrations of the largest leaf, either expressed on a dry weight basis or a tissue water basis, could not be determined at both stages because the relationships between the dry weight and K concentration were better fitted by linear regressions.

Discussion

The nutritional status of the crop could be diagnosed easily if the critical concentration of the nutrient was constant regardless of environmental conditions and plant ages. Therefore the choice of the basis for expressing the nutrient concentration should

depend not only on the closeness of the correlation between the plant growth and the nutrient concentration, but also on the constancy of the critical concentration.

As for the closeness of the correlation, there was no difference in the squared multiple correlation coefficients (R^2) whether K concentration was expressed on a dry weight basis or a tissue water basis. However, the critical K concentrations of the youngest leaf on a tissue water basis varied to a lesser extent than those on a dry weight basis. Thus tissue water provides a better basis than dry weight for expressing K concentration in plant analysis. This result confirms the opinion of Leigh *et al.* (8). Clements(4) recognized that K concentration on a tissue water basis reflected better the yield responses of sugarcane to potash application than that on a sugar-free dry weight basis.

Even in tissues of the similar physiological age (for example, the youngest leaf), concentration of soluble carbohydrate on a dry weight basis increases with the progress of growth stage, resulting in the dilution of K concentration on a dry weight basis(7). This would be the reason why the critical K concentration of the youngest leaf on a dry weight basis decreased with plant age in the present study. Cox(5) has also reported that the critical K concentration of lupins on a dry weight basis decreased with plant age.

The effect of plant age on the critical K concentration was overcome by expressing K concentration on a tissue water basis in the 1982 experiment. Possibly tissue water content changed little with plant age. Sugiyama *et al.* (14) found that tissue water content at dawn was stable within a few day when plants were well-watered. After plants were chilled, however, tissue water content decreased rapidly, resulting in the increase of K concentration on a tissue water basis(12, 14). Thus the critical K concentration of the youngest leaf on a tissue water basis varied with temperature. Namely, the critical K concentration was about 0.7% for the plants grown in early summer, and above

0.75% for the chilled plants (experiment 4 in 1979, and the second and the third stage in 1981). In contrast, the critical K concentration on a dry weight basis varied within the same season. Its variation due to the growing season was obscure.

The relationships between the dry weight of tops and K concentration of the youngest leaf were fitted by the second or the third degree polynomials. On the other hand, the dry weight of tops sometimes related linearly to K concentration of the largest leaf. If the relationships were fitted by linear regressions, the critical K concentrations could not be calculated. Therefore, the youngest leaf seems to be suitable for use in plant analysis, as compared with the largest leaf.

In conclusion, we could better assess the K status of the crop if the youngest leaf were sampled and its K concentration were expressed on a tissue water basis.

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体内水分当たり及び乾物重当たりのホウレンソウ葉中限界カリ濃度の比較

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摘 要

乾物重または体内水分当たりで表した葉中の限界カリ濃度が栽培の季節や生育段階によって変化するかどうかを明らかにするため、初夏と晩秋にカリ施肥量を変えてホウレンソウを栽培し、葉分析を行った。

地上部乾物重と最も若い展開葉のカリ濃度との関係は、多くの場合2次式または3次式によくあてはまった。

6葉期には、最も若い展開葉における乾物重当たりの限界カリ濃度は最低で4.3%、最高で6.7%であった。

これに対して、体内水分当たりの限界カリ濃度は約0.7%とほぼ一定であったが、低温遭遇後には0.75%以上となった。

初夏に行った生育段階に関する実験では、最も若い展開葉における体内水分当たりの限界カリ濃度は5葉期から7葉期までは約0.7%で、比較的一定に保たれた。一方、乾物重当たりの限界カリ濃度はこの間に低下した。