

COMPARISON OF THE TOXICITY OF HEAVY METALS TO CABBAGE GROWTH

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KEY WORDS

Cabbage Heavy metals Toxicity Translocation

SUMMARY

Cabbage plants were grown for 55 days with a nutrient solution containing 1 and 10 ppm of V, Cr(III), Cr(VI), Mn, Fe, Co, Ni, Cu, Zn, Cd, Hg(I), or Hg(II). A comparison of the plant growth and chemical analysis revealed that Cr(VI), Cu, Cd, and Hg(II) in the solution are most toxic to the plant growth (hence detrimental to the cabbage-head formation) and Mn, Fe, and Zn are less toxic than other heavy metals, and that Mn, Zn, Co, Ni, and Cd are translocated into all the plant organs while V, Cr(III), Cr(VI), Fe, Cu, Hg(I), and Hg(II) are accumulated in the roots.

INTRODUCTION

The increasing problem of environmental pollution by heavy metals necessitates study on the toxicity of heavy metals to plants as well as animals. The toxicity of heavy metals to crop plants varies from metal to metal¹ and from crop to crop^{4,5}. Effects of graded-levels of some heavy metals in the culture solution on the growth of cabbage (*Brassica oleracea* L. var. *capitata* L.) plants were investigated in a previous experiment², and the heavy metals present in excess always brought about stunted growth of the plants.

The present experiment was designed to compare the toxicity of ten heavy metals (V, Cr, Mn, Fe, Co, Ni, Cu, Zn, Cd, Hg) to the cabbage growth and head formation. For this purpose, cabbage plants were grown for a long time with the nutrient solution containing the same concentration of each heavy metal. As chromium and mercury have two stable valencies in natural conditions, the effect of both heavy metals with two chemical forms (Table 1) on the plant growth was also evaluated.

MATERIALS AND METHODS

Pre-germinated seedlings at the three-leaf stage of a Nagaoka cabbage cultivar. Soshu, were transplanted on April 17, 1976 into 4 liter pots containing a standard culture solution and grown in a greenhouse for one month by the same water culture method². The composition of the culture solution in ppm was: 50 N, 20 P, 50 K, 100 Ca, 50 Mg, 2 Fe, 0.5 B, 0.4 Mn, 0.2 Zn, 0.05 Cu, and 0.05 Mo. Their chemical forms were NaNO_3 , NaHPO_4 , K_2SO_4 , CaCl_2 , MgSO_4 , FeCl_3 , H_3BO_3 , MnCl_2 , ZnCl_2 , and Na_2MoO_4 , respectively. The culture solution was aerated for 1 h at 2 h intervals and renewed once a week, and its pH was adjusted to be 5.5. The young plants were then exposed to 1 and 10 ppm of vanadium, trivalent chromium(III), hexivalent chromium(VI), manganese, iron, cobalt, nickel, copper, zinc, cadmium, univalent mercury(I), or bivalent mercury(II) which were added, in two replications, to the culture solution (Table 1). Their chemical forms other than the above-mentioned were VCl_3 , CrCl_3 , Na_2CrO_4 , CoCl_2 , NiCl_2 , CdCl_2 , $\text{Hg}_2(\text{NO}_3)_2$ and $\text{Hg}(\text{NO}_3)_2$. For convenience, the treatment plots are later described in terms of the concentration of each heavy metal. As a control plot, the two plants receiving only the standard culture solution were allowed to grow together with the heavy metal-treated plants. After 55 days of exposure, all the plants were harvested except that the plants of the Cu 10, Hg(II) 10, Ni 10, Co 10, Cd 10, Cr(III) 10, and Cr(VI) 10 plots were sampled after 7, 14, 14, 16, 21, 44, and 44 days of exposure when they began to die before the harvest. The fresh weight of each cabbage-head of the plants were measured immediately, and the plants were separated into the inner leaves (composing the cabbage-head), outer leaves (not composing the cabbage-head), stems, and roots. After the roots were rinsed more than three times with demineralized water, all the plant samples were dried, weighed, and milled for the chemical analysis.

An aliquote of the milled samples was digested with the nitric-perchloric-sulfuric acid (5:2:1) mixture, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, and Cd in the digested solution were determined by flame atomic absorption spectrophotometry. The determination of Hg was based on the flameless atomic absorption method³ after the digestion of the milled samples with the nitric-sulfuric acid (1:1) mixture and urea. The separation of Cr(III) or Cr(VI) and of Hg(I) or Hg(II) from the total Cr or Hg was disregarded in these determinations. Thus the Cr(III) content described in this paper, for example, indicates the total Cr content of the Cr(III) treatment.

Table 1. Treatments of heavy metals in the culture solution containing all nutrients

Treatment plots	V	Cr(III)	Cr(VI)	Mn	Fe	Co	Ni	Cu	Zn	Cd	Hg(I)	Hg(II)
Control	0	0	0	0.4	2	0	0	0.05	0.2	0	0	0
V 1	1*	0	0	0.4	2	0	0	0.05	0.2	0	0	0
Cr(III) 1	0	1*	0	0.4	2	0	0	0.05	0.2	0	0	0
Cr(VI) 1	0	0	1*	0.4	2	0	0	0.05	0.2	0	0	0
Mn 1	0	0	0	1*	2	0	0	0.05	0.2	0	0	0
Fe 1	0	0	0	0.4	1*	0	0	0.05	0.2	0	0	0
Co 1	0	0	0	0.4	2	1*	0	0.05	0.2	0	0	0
Ni 1	0	0	0	0.4	2	0	1*	0.05	0.2	0	0	0
Cu 1	0	0	0	0.4	2	0	0	1*	0.2	0	0	0
Zn 1	0	0	0	0.4	2	0	0	0.05	1*	0	0	0
Cd 1	0	0	0	0.4	2	0	0	0.05	0.2	1*	0	0
Hg(I) 1	0	0	0	0.4	2	0	0	0.05	0.2	0	1*	0
Hg(II) 1	0	0	0	0.4	2	0	0	0.05	0.2	0	0	1*

* Or 10 ppm in the case of 10 ppm supply of each heavy metal.

RESULTS

Plant dry weight and cabbage-head yield

With the 1 ppm supply of each heavy metal, the total plant dry weight was more or less the same in the control, Fe 1, and Mn 1 plots, and it became smaller in the Zn, V, Cr(III), Co, Hg(I), Ni, Hg(II), Cr(VI), Cd, and Cu treatments in this order (Table 2). As the total dry weight decreased, the cabbage-head yield decreased and resulted in no yield in the Cu 1 plot. The decrease in the cabbage-head yield was greater than in the total dry weight. The total dry weight decreased as the supply of each heavy metal increased from 1 to 10 ppm, and no plot exceeded the control plot. The total dry weight in the 10 ppm supply was arranged as follows in relation to the treatments Fe > Mn > Zn > V > Hg(I) > Cr(III) · Co · Ni · Cd · Cr(VI) · Hg(II) · Cu. The cabbage-head yield decreased in the same order, and the plants in the later 7 plots, sampled before the harvest, had no cabbage-head.

Symptoms of the toxicity injury by each heavy metal observed throughout the water culture were: wilting of the leaves in the V 10 plot; chlorotic leaves in the Cr(III) 10 and Cr(VI) 10 plots; brown speckles at the leaf-margin in the Mn 10

Table 2. Effects of the supply of heavy metals on the total plant dry weight and the fresh cabbage-head yield

Treatment plots	Dry weight		Yield (g/plant)	Treatment plots	Dry weight		Yield (g/plant)
	(g/plant)	R.V.*			(g/plant)	(g/plant)	
Control	76.3	100	346	Control	76.3	100	346
V 1	66.3	87	263	V 10	21.3	28	21
Cr(III) 1	61.6	81	217	Cr(III) 10	7.6	10	0
Cr(VI) 1	36.4	48	26	Cr(VI) 10	3.9	5	0
Mn 1	75.2	99	321	Mn 10	48.1	63	118
Fe 1	77.3	101	355	Fe 10	71.3	93	314
Co 1	57.6	75	215	Co 10	6.2	8	0
Ni 1	45.7	60	92	Ni 10	4.9	6	0
Cu 1	5.1	7	0	Cu 10	1.4	2	0
Zn 1	72.4	95	304	Zn 10	31.7	42	44
Cd 1	30.7	40	16	Cd 10	4.3	6	0
Hg(I) 1	55.0	72	157	Hg(I) 10	12.3	16	4
Hg(II) 1	37.1	49	47	Hg(II) 10	1.7	2	0
LSD (5%)	2.6	—	12	LSD (5%)	2.3	—	8

* Relative values (%) of the total plant dry weight of each plot in comparison with the control plot (100%).

plot; reddish-brown roots in the Fe 10 plot; brown speckles and necrosis on the leaves in the Co 1 and Co 10 plots; brown speckles on the leaves, developing to many necrotic spots in the Ni 1 and Ni 10 plots; wilting of the leaves in the Cu 1 and Cu 10 plots; chlorosis between the leaf-veins in the Zn 10 plot; the outer leaves turning to partially purple in the Cd 1 and Cd 10 plots; and wilting of the leaves in the Hg(I) 10 and Hg(II) 10 plots. The roots of these plots were small and dark.

Contents of heavy metals and their distribution in the plants

The contents of heavy metals in the outer leaves in the 1 ppm supply of each heavy metal were in a decreasing order; Zn > Cd > Co > Mn · Fe > Ni > Cu > Cr(VI) > Hg(II) > Hg(I) > V > Cr(III) which was different from the order in the roots; Fe > Cu > Cr(III) > Cd > Zn > Co > Mn > Ni > Cr(VI) > Hg(II) > Hg(I) > V (Table 3). All the contents were much higher in the roots than in the outer leaves, and the inner leaves always had the lowest content of each heavy metal among the four cabbage organs. These contents in each organ increased as the supply of each heavy metal increased from 1 to 10 ppm. In the 10 ppm supply, the Mn, Co, Ni, Zn, and Cd contents in the outer leaves and the Fe, Cu, and Cr (III) contents in the roots were particularly higher than the other heavy metal contents.

Table 3. Effects of the supply of heavy metals on the contents of metals in the outer leaves and roots

Treatment plots	Content (ppm)		a/b	Treatment plots	Content (ppm)		a/b
	Outer leaves	Roots			Outer leaves	Roots	
	a	b			a	b	
V 1	4	438	0.009	V 10	10	1418	0.007
Cr(III) 1	2	3121	0.001	Cr(III) 10	27	12081	0.002
Cr(VI) 1	8	889	0.009	Cr(VI) 10	124	2287	0.05
Mn 1	129	1721	0.07	Mn 10	1575	9375	0.17
Fe 1	129	7493	0.02	Fe 10	143	26534	0.005
Co 1	138	1665	0.08	Co 10	1250	3235	0.39
Ni 1	125	1048	0.12	Ni 10	905	2260	0.40
Cu 1	56	3225	0.02	Cu 10	116	13000	0.009
Zn 1	358	2075	0.17	Zn 10	805	6550	0.12
Cd 1	155	2688	0.06	Cd 10	1050	5225	0.20
Hg(I) 1	6	280	0.02	Hg(I) 10	33	1882	0.02
Hg(II) 1	8	703	0.01	Hg(II) 10	48	2712	0.02

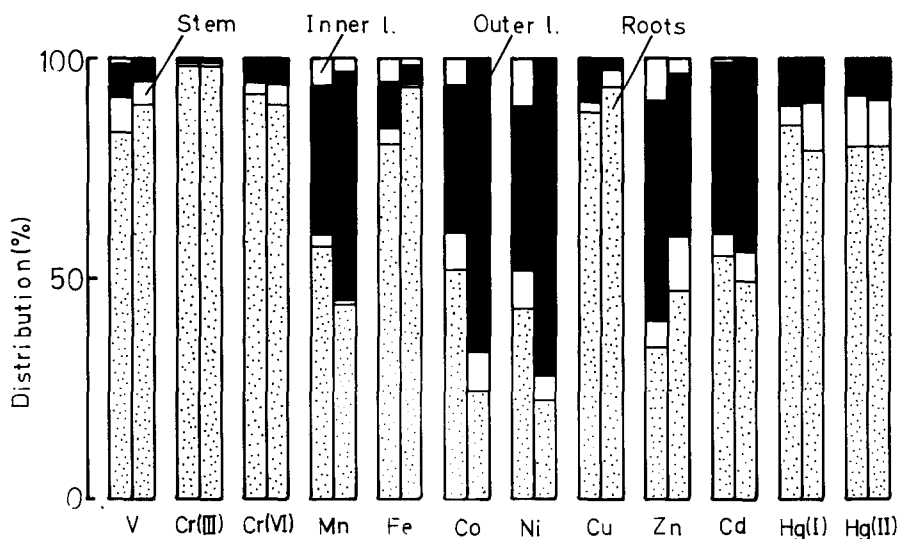


Fig. 1. Distribution of heavy metals in the various plant parts as percentages of the amount absorbed by the plant. *Left columns*: 1-ppm level. *Right columns*: 10-ppm level.

The distribution of heavy metals in the plants was computed as the percentage of the amount of each heavy metal remaining in each organ to the total amount absorbed by the plants (Fig. 1). The distribution of V, Cr(III), Cr(VI), Fe, Cu, Hg(I), and Hg(II) was quite large in the roots, and the exceptionally small distribution of Cr(III) in the plant-top organs (*i.e.* inner leaves, outer leaves, and stem) was apparent in comparison with that of Mn, Co, Ni, Zn, and Cd. The latter heavy metals were distributed mainly in the outer leaves although 20–50% of the total amount remained in the roots.

DISCUSSION

From the orders in the total dry weight of cabbage plants grown at 1 and 10 ppm of each heavy metal (Table 2), it is concluded that these heavy metals, present in the culture solution, are categorized into three groups in terms of the toxicity inducing the stunted growth of the plants: Manganese, Fe, and Zn are less toxic, and Mn and Zn at concentrations less than 1 ppm and Fe at concentrations around 1 ppm are indispensable for the plant growth as recognised before²; V, Cr(III), Co, Ni, and Hg(I) are toxic and non-essential, and each heavy metal at concentrations larger than 1 ppm apparently affects adversely the plant growth; and Cr(VI), Cd, Cu, and Hg(II) are strongly toxic, and each heavy metal at

concentrations even around 1 ppm brings about poor growth of the plants, inducing a low or no yield of the cabbage-head. The toxicity of Cu is exceptionally strong although its essentiality for higher plants is well known.

Manganese, Co, Ni, Zn, and Cd are absorbed abundantly by cabbage plants from their respective solutions and translocated from the roots to the plant-tops, and Fe is absorbed but very badly translocated. In contrast to the above heavy metals, V, Cr(III), Cr(VI), Cu, Hg(I), and Hg(II) are hardly absorbed, and their translocation from the roots to the plant-tops is small, suggesting the lower mobility of these heavy metals within the plants. A small difference in the uptake of Cr(III) and Cr(VI) by the plants indicates that more CrO_4 ions are absorbed or in other words CrO_4 ions are absorbed more easily than Cr ions. Another difference in the uptake of Hg(I) and Hg(II) is explained by their solubility in the culture solution. That is, bivalent mercury dissolves completely in the culture solution while univalent mercury precipitates in part and dissolves uncompletely. In fact, at the addition of heavy metals to the culture solution, white precipitates appeared in the Hg(I) plot (their probable chemical forms are Hg_2HPO_4 and Hg_2Cl_2) but no precipitate in any other plot.

The relationship between the relative values of the total plant dry weight and the contents of respective heavy metals in the outer leaves was examined in order to understand how the accumulation of each heavy metal reduces the plant

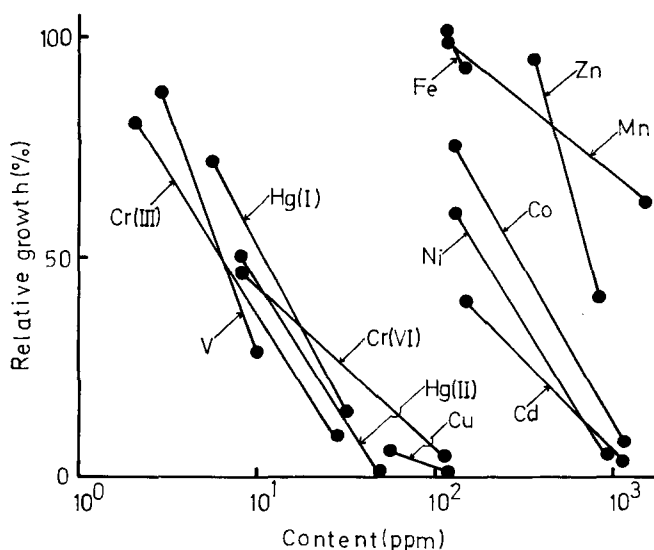


Fig. 2. Reduction of relative growth (%) in relation to the heavy metal contents in the outer leaves (ppm).

growth (Fig. 2). Manganese and Zn are accumulated abundantly not only in the outer leaves but also in the other organs, but their toxicity is weaker than that of other heavy metals. When the plant growth is markedly reduced the accumulation of Co, Ni, and Cd is much larger than that of V, Cr(III), Cr(VI), Cu, Hg(I), and Hg(II). The results of the distribution patterns (Fig. 1) and the ratio of the heavy metal contents in the outer leaves to those in the roots (Table 3) indicate that the former heavy metals are translocated into all the plant organs and affect the plant growth while the latter heavy metals injure initially the root growth. The toxicity of Fe is as weak as Mn and Zn, but the organ especially damaged by Fe is the roots, while Mn and Zn injury damages all the plant organs.

Received 17 January 1978

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