

## The effect of sodium humate upon phosphorus nutrition of plants with variable doses of iron and calcium in tomato water cultures

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### Abstract

Studies were carried out on the effect of sodium humate upon content of P, Fe, Ca, and dry weight yield of tomato seedlings in water cultures. Nutrient solutions contained 10 times more or 10 times less P, Fe, and Ca than the control (nutrient solution according to Hampe). Sodium humate was effective only in relation to phosphorus at lowered amount of Fe, and supplied the plants with Fe when phosphorus content remained in excess. At lowered Ca content humate was not more effective in relation to phosphorus. Sodium humate protected the plants against inhibiting effects of excessive Ca content, and supplied them with phosphorus.

### INTRODUCTION

Positive effect of humic compounds upon plant growth and development is especially visible if plants remain in unfavourable conditions. Although it is very difficult to give any undoubtful proof on the penetration of large-particle, humic substances into plant cells, the effect of these substances upon plant organisms has been frequently noted. It is possible that this effect is of an indirect character. The phenomenon is extremely significant for the turnover of minerals, and especially for phosphorus nutrition of plants. Depending on the pH value in the environment, phosphorus forms insoluble (or almost insoluble) salts with bi- and trivalent metal cations. And thus, at low pH value iron and aluminium phosphates are formed, while at higher pH values — calcium and magnesium phosphates. Consequently, in many cases plants suffer from phosphorus shortage which becomes unavailable for plant roots.

Humic substances prevent the formation of such salts. They also affect (although to a limited extent) solubility of phosphorus salts, and hence favour the uptake of phosphorus by plants (Chaminade, 1944; Jurkowska, 1957; Szymański, 1962). Mechanisms of these processes are still under discussion. Results of Catsky (1958) and Gumiński et al. (1965) point to the fact that at insufficient iron content presence of large amounts of calcium decreases the efficiency of humate, the more so that in water cultures iron precipitates — among others — in form of phosphates. Humate induces formation of complex iron compounds, making it easily assimilable by plants. Furthermore, humate forms also complex compounds with calcium (Chaminade and Vistelle, 1974; Szymański, 1962). Some authors (Sinha, 1972) are also of the opinion that the presence of iron in particles of humic acids is essential for the formation of phosphoro-humic bonds, from which phosphorus is readily available for plants.

In view of the above it seemed justified to undertake the present studies on the effect of sodium humate upon the accumulation of phosphorus by plants in conditions of either shortage, or excess of  $\text{Fe}^{+3}$  and  $\text{Ca}^{+2}$  ions in the nutrient solution.

#### MATERIAL AND METHODS

Three vegetation experiments with water cultures of tomato seedlings of the 'Stonora' variety were carried out in June, July and August 1976. They were conducted in an unacclimatized greenhouse. During the second experiment climatic conditions were different due to higher number of colder, cloudy days.

Eight-days old seedlings, germinated in tap water, were transferred into 1 l glass jars containing nutrient solution. 4 seedlings were placed in each jar. After 7 days one seedling was removed from each jar, leaving 3 seedlings of the same size. Each experimental combination (object) consisted of 6 jars, with 3 seedlings in each jar. Nutrient solution described by Hampe (1938) was used. It contained (in g per 1 l of distilled water):  $\text{Ca}(\text{NO}_3)_2 \times 4 \text{H}_2\text{O}$  — 0.71,  $\text{KNO}_3$  — 0.57,  $\text{MgSO}_4 \times 7 \text{H}_2\text{O}$  — 0.284,  $(\text{NH}_4)_2\text{HPO}_4$  — 0.142,  $\text{Fe}_2\text{SO}_4 \times n \text{H}_2\text{O}$  — 0.116, and an addition of microelements:  $\text{H}_3\text{BO}_3$ ,  $\text{MnCl}_2 \times 4 \text{H}_2\text{O}$ ,  $\text{ZnSO}_4 \times 7 \text{H}_2\text{O}$ ,  $\text{CuSO}_4 \times 5 \text{H}_2\text{O}$ ,  $\text{NaMoO}_4 \times 2 \text{H}_2\text{O}$  — 0.6 mg of each compound per 1 l of nutrient solution. pH of the solution was brought to 6.4 before seedlings were introduced into it. pH value was checked every week and, if necessary, adjusted with 0.1 n HCl or 0.1 n NaOH.

Decreased amount of phosphorus in the solution was substituted with nitrogen in form of  $\text{NH}_4\text{Cl}$ . Since the amounts of phosphorus and calcium were lowered simultaneously, it was necessary to replenish nitro-

gen with sodium nitrate. At increased amounts of phosphorus  $\text{Na}_2\text{HPO}_4 \times 12 \text{H}_2\text{O}$  was used.

Humate obtained from one-year old compost (Gumiński, 1950) was dissolved in 0.01 n NaOH and added into each combination. 10 ml of humate solution, containing 100 mg Na-humate, were added into 1 l of nutrient solution.

Nutrient solutions were aerated and mixed every day by pouring them into another jars. Each experiment lasted 21 days. In the second and third experiment length of stems and roots were measured. Dry weight of seedlings was determined in every experiment. Dried and ground plant material from every combination was digested in concentrated  $\text{H}_2\text{SO}_4$  and  $\text{HNO}_3$ . Calcium in the obtained solution was determined by flame photometry, phosphorus colorimetrically with the method of Fiske and Subbarow, and iron also colorimetrically using  $\alpha$ - $\alpha'$ -dipyridyl.

## RESULTS OF THE EXPERIMENTS

Results of the first experiment are presented in Table 1. This experiment dealt with the effect of humate on dry weight yield of seedlings, growing in nutrient solution with lowered or increased (compared to the control) amount of phosphorus.

Table 1

Dry weight yield (in g) of shoots and roots of 3 plants from one jar  
Average values for 6 repetitions

Combination	-humate		+humate	
	shoots	roots	shoots	roots
$\frac{1}{100}$ P	0.04	0.01	0.05	0.01
$\frac{5}{100}$ P	0.23	0.07	0.41	0.12
$\frac{1}{10}$ P	0.90	0.22	1.14	0.31
$\frac{1}{5}$ P	2.23	0.41	2.10	0.46
$\frac{1}{2}$ P	3.00	0.45	3.20	0.47
1 P	3.31	0.42	3.19	0.51
5 P	2.50	0.46	2.46	0.44
10 P	0.90	0.14	1.68	0.33

Confidence interval for shoots 0.22, for roots 0.08.

Changed doses of phosphorus resulted in decreased dry weight of shoots and roots of tomato seedlings. The only exceptions were combination 1/2 P and 5 P with respect to root weight, and 1/2 P and humate with respect to shoots — in both cases slight increase of dry weight was noted, but its value did not exceed error range.

Table 2

Length of shoots and roots of seedlings (in cm)  
Average values from measurements of 18 plants

Combination		- humate		+ humate	
		shoots	roots	shoots	roots
$\frac{1}{10}$ P	$\frac{1}{10}$ Fe	12.1	19.6	20.2	18.3
	1 Fe	11.9	24.4	19.8	21.3
	10 Fe	11.4	25.6	13.8	27.2
1 P	$\frac{1}{10}$ Fe	19.7	20.0	26.0	19.0
	1 Fe	21.3	19.7	25.8	18.6
	10 Fe	18.3	17.7	23.9	20.3
10 P	$\frac{1}{10}$ Fe	7.0	6.4	10.7	11.4
	1 Fe	12.6	17.0	14.5	16.0
	10 Fe	20.0	17.9	19.5	17.8

Confidence interval for shoot 4.20, for roots 2.3.

Plants grown in combination with the lowest dose of phosphorus (1/100 P) were characterized by growth inhibition already at 7th day of the experiment. These plants developed only two leaves, and typical symptoms of phosphorus deficiency were noticeable rather early. They consisted at first of dark colour of both leaves, violet spots at their lower side, and violet coloration of shoots, gradually leading into necrotic changes and withering of both leaves. Similar symptoms were noticed in plants from combination 5/100 P, although they occurred later, i.e. at 14th day of the experiment. Humate in the nutrient solution lowered the rate of these changes, so that seedlings withered 3-4 days later. Favourable effect of humate was most pronounced in the combination with 1/10 P, in which yield of plant dry weight increased by 26% for shoots, and 38% for roots. Furthermore, humate liquidated visible symptoms of phosphorus deficiency, which were far less pronounced in this combination than in combinations with 1/100 P and 5/100 P. In combination with 10 times higher doses of phosphorus, humate also resulted in increased dry weight of plant shoots and roots. The differences were statistically significant.

Table 3

Dry weight yield (in g) of shoots and roots of 3 plants from one jar  
Average values from 6 repetitions

Combination		-humate		+humate	
		shoots	roots	shoots	roots
	$\frac{1}{10}$ Fe	0.19	0.05	0.74	0.18
$\frac{1}{10}$ P	1 Fe	0.19	0.06	0.91	0.26
	10 Fe	0.21	0.07	0.36	0.13
	$\frac{1}{10}$ Fe	0.75	0.15	1.47	0.33
1 P	1 Fe	0.69	0.10	1.10	0.34
	10 Fe	0.53	0.11	1.24	0.34
	$\frac{1}{10}$ Fe	0.07	0.01	0.13	0.04
10 P	1 Fe	0.29	0.08	0.37	0.10
	10 Fe	0.90	0.19	0.79	0.16

Confidence interval for shoots 0.4, for roots 0.05.

Basing on the results of the first experiment, for further studies combinations with 10 times higher or 10 times lower phosphorus doses were selected. Changed doses of phosphorus affected exogenous ratio P/Fe and P/Ca in the solution. Consequently, in the second experiment combination containing 10 times lower or higher amounts of iron was introduced. Amounts of calcium and other components of nutrient solution remained the same.

10 times lower doses of phosphorus at full dose of iron in nutrient solution (object 1/10 P, 1 Fe) resulted in a decrease of stem growth rate and dry weight of shoots (Tables 2 and 3). In this combination roots of seedlings were longer although their dry weight was lower than in control plants. Similar phenomenon was noted in combination 1/10 P, 10 Fe. Addition of humate to nutrient solution did not affect root length, with an exception of their slight decrease in object 1/10 P, 1 Fe plus humate, while significant effect was noted with respect to dry weight of roots — it increased considerably compared with combination without humate addition.

10 times lower doses of iron in this experiment did not inhibit rate of growth and accumulation of dry weight (object 1 P, 1/10 Fe, Tables 2 and 3). At lowered iron dose humate strongly stimulated the increase of dry weight (Table 3) and phosphorus content in plants (Table 4). At 10 times higher dose of phosphorus, deficiency of iron was noted (Table 5), the only exception being the combination with simultaneous increase of phosphorus and iron content.

Table 4

Phosphorus content in dry weight of seedlings (shoots) in mg P/100 mg of dry weight, and in shoots yield of 3 plants from one jar  
Average values from 3 analyses

Combination		- humate		+humate	
		mg P/100 mg of d. wt.	mg P in the yield	mg P/100 mg of d. wt.	mg P in the yield
$\frac{1}{10}$ P	$\frac{1}{10}$ Fe	0.520	0.988	0.400	2.296
	1 Fe	0.244	0.463	0.270	2.457
	10 Fe	0.280	0.588	0.214	0.770
1 P	$\frac{1}{10}$ Fe	0.596	4.470	0.700	10.290
	1 Fe	0.490	3.381	0.580	6.380
	10 Fe	0.444	2.353	0.284	3.521
10 P	$\frac{1}{10}$ Fe	1.540	1.078	1.524	1.931
	1 Fe	1.008	2.923	0.888	3.285
	10 Fe	0.644	5.796	0.625	4.934

Table 5

Iron content in dry weight of shoots (in mg Fe/100 of dry weight) and in stem yield of 3 plants from one jar

Combination		- humate		+humate	
		mg Fe/100 mg of d. wt.	mg Fe in the yield	mg Fe/100 mg of d. wt.	mg Fe in the yield
$\frac{1}{10}$ P	$\frac{1}{10}$ Fe	0.024	0.045	0.011	0.081
	1 Fe	0.018	0.035	0.023	0.209
	10 Fe	0.035	0.074	0.030	0.108
1 P	$\frac{1}{10}$ Fe	0.026	0.195	0.016	0.235
	1 Fe	0.025	0.172	0.016	0.176
	10 Fe	0.032	0.169	0.024	0.297
10 P	$\frac{1}{10}$ Fe	0.017	0.011	0.020	0.026
	1 Fe	0.019	0.055	0.018	0.066
	10 Fe	0.030	0.270	0.036	0.284

In combination with increased phosphorus content, and decreased or full iron content, plants were chlorotic. Chemical analyses showed that iron content decreased both, in dry weight and in total yield of plants compared to its content found in control plants (Table 5). Sodium humate in combination 10 P, 1/10 Fe saves the plants. It does not result in any significant increase of Fe content per unit of dry weight but it increases total amount of this element in total plant yield.

In the third experiment attention was given to the effect of Na-humate on growth, dry weight production, and phosphorus and calcium contents in tomato seedlings, grown in nutrient solution containing 10 times lower or higher phosphorus or calcium doses, or else P and Ca doses together.

Table 6  
Length of shoots and roots of seedlings (in cm)  
Average values measurements of 18 plants

Combination		-humate		+humate	
		shoots	roots	shoots	roots
$\frac{1}{10}$ P	$\frac{1}{10}$ Ca	19.4	23.0	25.0	23.5
	1 Ca	16.6	21.0	23.5	26.1
	10 Ca	12.3	20.2	15.1	24.4
1 P	$\frac{1}{10}$ Ca	26.5	26.2	25.5	28.7
	1 Ca	31.7	25.7	33.0	25.0
	10 Ca	9.7	18.8	15.1	26.2
10 P	$\frac{1}{10}$ Ca	15.5	19.5	17.0	21.0
	1 Ca	21.8	23.7	21.2	23.5
	10 Ca	15.6	30.8	15.4	26.5

Confidence interval for shoots 4.8, for roots 2.8.

Decrease of calcium content to 1/10 inhibited growth of shoots (Table 6). This decrease correlated with low dry weight of shoots (Table 7). Some differences were also noted in the growth and dry weight of roots, although these differences proved statistically insignificant. Significant decrease of growth rate and dry weight of seedlings was observed in case of increased amounts of calcium. In this object stimulating effect of sodium humate on stem and root growth was noted. Also dry weight of plants increased (Table 6 and 7). Lower content of calcium accompanied by lower content of phosphorus did not have any visible effect upon growth, contrary to the object with 10 times increased content of calcium. In the latter case statistically significant inhibition of growth and decrease of dry weight yield were observed.

Table 7

Dry weight (in g) of shoots and roots of seedlings  
Average values from 6 repetitions

Combination		-humate		+humate	
		shoots	roots	shoots	roots
$\frac{1}{10}$ P	$\frac{1}{10}$ Ca	0.64	0.20	1.02	0.28
	1 Ca	0.68	0.24	1.02	0.30
	10 Ca	0.23	0.05	0.43	0.09
1 P	$\frac{1}{10}$ Ca	1.03	0.28	1.10	0.24
	1 Ca	1.46	0.32	1.48	0.30
	10 Ca	0.13	0.02	0.32	0.05
10 P	$\frac{1}{10}$ Ca	0.30	0.07	0.54	0.14
	1 Ca	0.88	0.22	0.81	0.17
	10 Ca	0.42	0.08	0.55	0.11

Confidence interval for shoots 0.5, for roots 0.04.

Table 8

Phosphorus content in dry weight of shoots in mg P/100 mg of dry weight and in shoots yield of  
3 plants from one jar  
Average values from 3 analyses

Combination		-humate		+humate	
		mg P/100 mg of d. wt.	mg P in the yield	mg P/100 mg of d. wt.	mg P in the yield
$\frac{1}{10}$ P	$\frac{1}{10}$ Ca	0.109	0.700	0.109	1.115
	1 Ca	0.082	0.563	0.113	1.154
	10 Ca	0.213	0.489	0.289	1.246
1 P	$\frac{1}{10}$ Ca	0.528	5.446	0.546	6.012
	1 Ca	0.489	7.279	0.507	7.514
	10 Ca	0.289	0.388	0.610	1.954
10 P	$\frac{1}{10}$ Ca	1.039	3.118	0.837	4.523
	1 Ca	0.794	6.994	0.864	7.004
	10 Ca	0.502	2.110	0.451	2.483



Chemical analyses of plant material showed that combinations with excessive amounts of calcium (compared to phosphorus level) were characterized by lower phosphorus content in dry weight of seedlings (shoots) than the control ones. Addition of sodium humate to nutrient solution resulted in an increase of phosphorus content in plant dry weight (Table 8). In case of excessive amounts of calcium in the solution, its content in dry weight of shoots also increased (compared with the control), although total amount of Ca in plant yield decreased. This phenomenon was connected with inhibition of plant growth rate. Addition of humate resulted in a slight decrease of calcium in dry weight while total amount of Ca in plant yield increased by over 100% compared to the combination without humate (Table 9).

Table 9

Calcium content in dry weight of shoots in mg Ca/100 mg of dry weight and in shoots yield of 3 plants from one jar  
Average values from 3 analyses

Combination		- humate		+humate	
		mg Ca/100 mg of d. wt.	mg Ca in the yield	mg Ca/100 mg of d. wt.	mg Ca in the yield
$\frac{1}{10}$ P	$\frac{1}{10}$ Ca	1.98	12.67	1.70	17.34
	1 Ca	2.46	16.72	3.44	35.08
	10 Ca	4.53	10.41	5.09	21.88
1 P	$\frac{1}{10}$ Ca	1.62	16.68	1.85	20.35
	1 Ca	3.12	45.55	3.10	45.88
	10 Ca	5.78	7.51	5.04	16.12
10 P	$\frac{1}{10}$ Ca	0.91	2.73	1.18	6.37
	1 Ca	0.76	6.68	1.20	9.72
	10 Ca	3.97	16.67	3.70	20.35

Lowered level of calcium in nutrient solution (object 1 P,  $\frac{1}{10}$  Ca) only insignificantly increased phosphorus content in dry weight (plus 6%), while total amount of phosphorus in plant yield decreased (by 25%) compared to the control. This was due to a decrease of dry weight yield. Deficiency of calcium in this combination resulted in a decrease of calcium in plant dry weight (by 48%), as well as of its total content in plant yield. Humate addition resulted in an increase of calcium content per unit of dry weight by 14%, and of its total amount in plant yield by 19% (Table 9).

In combination in which phosphorus and calcium were simultaneously decreased (10 times), significant decrease of their content in plants was

observed compared to the control. Na-humate increased total amount of P in dry weight yield by 58% although it did not change P content per unit of dry weight, and slightly lowered Ca content in dry weight with simultaneous increase of total Ca content in seedling shoot yield (by 36%, Table 9).

#### DISCUSSION

Results presented above show that plants suffered from phosphorus deficiency at lowered content of this element in nutrient solution, but also in some specific conditions, when roots remained in the environment with excessive amounts of Fe and Ca in relation to phosphorus content. In case of phosphorus deficiency sodium humate induces better utilization of this element by plants, as supported by the results of chemical analyses of phosphorus content in plants. Although mechanism of this process has not been fully explained as yet, it is possible to suggest several facts basing on the results obtained with variable doses of Fe and Ca.

Decreased amounts of iron during experiments did not limit plant growth, only decreased the excess of this element; in this case humate is more effective toward phosphorus. Addition of humate increases phosphorus content in dry weight, but most of all it improves utilization of this macroelement in the production of plant mass. Most probably its effect is based on iron binding in complex compounds, and thus on creating more favourable conditions for phosphorus uptake by plants. Also direct effect of these bonds upon plant organisms is not to be excluded. Protective effect of humate is also noted in case of phosphorus excess, which inhibits Fe uptake by plants (Rediske, Biddulph, 1953). Complex compounds of iron with humate may constitute source of iron for plants, as supported by the results of Aso and Takenaga (1975).

10-fold decrease of calcium content in nutrient solution inhibited plant growth. At lowered Ca content in the solution sodium humate exerts slight positive effect (most probably laying within the range of error) upon the content of calcium in dry weight and in total plant yield. This result is in agreement with previous studies by Leonowicz-Babiak (1976), who showed that only fraction II positively affects plants in case of calcium deficiency, contrary to non-fractioned (total) humate and fraction I. At lowered Ca content and limited supply of phosphorus, humate was almost ineffective toward phosphorus. Most probably it formed rather stable complexes with calcium cations. Mieczyska (1976) states that lack of calcium inhibits phosphorus uptake.

Excess of calcium in case of tomato seedlings strongly inhibited plant growth, increased Ca content, and decreased P content in dry weight.

To some extent humate liquidates toxic effect of excessive amounts of calcium, most probably by causing formation of complex calcium compounds, with drawing this element from the environment (Jurajda, 1974), and hence improving conditions for phosphorus uptake (Żurawski, 1974).

Along with changes of Ca content in nutrient solution ratio between Ca and Mg becomes unbalanced. This situation also affects plant growth. Lower Ca/Mg ratio can result in some disturbances of protein metabolism (Buczek and Leonowicz-Babiak, 1971). This phenomenon, however, should constitute an object of further studies since it is known that humate forms complexes also with magnesium.

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*Wpływ humianu sodu na forsforowe żywienie roślin  
na tle zmieniających się dawek żelaza i wapnia w kulturach wodnych  
pomidorów*

#### Streszczenie

Celem niniejszej pracy było zbadanie wpływu Na-humianu otrzymanego z kompostu liściowego na akumulację fosforu przez rośliny w warunkach zmniejszonej ilości lub nadmiaru żelaza i wapnia w pożywkach kultur wodnych.

Przy zmniejszonej podaży Fe humian wpłynął na lepsze pobieranie i wykorzystanie fosforu. Dodatnie działanie humianu polegające na dostarczaniu roślinom Fe uwidoczniło się w warunkach nadmiaru P w pożywce. Przy 10-krotnie obniżonej ilości Ca, humian przejawiał niewielki dodatni wpływ na rośliny i nie wzrastała istotnie jego efektywność w stosunku do fosforu. Wyraźnie dodatni wpływ humianu, polegający na dostarczaniu roślinom fosforu, wystąpił w warunkach nadmiaru wapnia w pożywce.