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# Influence of sodium humate on the uptake of some ions by tomato seedlings

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Abstract

The influence of sodium humate on the uptake of $K^+$, $Rb^+$ $NH_4^+$, $Mg^{2+}$, $Fe^{3+}$, $PO_4^{3-}$, $NO_3^-$ and $Cl^-$ ions from equilibrated aqueous solutions was investigated. By varying transpiration and applying dinitrophenol "passive" and "active" uptake were distinguished. Ion decrease in the solution and accumulation of labelled elements in roots and shoots were determined. Humate strongly stimulated $K^+$ and $Rb^+$ uptake and strongly inhibited the uptake of $Cl^-$, while it slightly enhanced $Mg^{2+}$ and $PO_4^{3-}$ uptake. It, moreover, facilitated iron transport from the roots to shoots. The interaction of humate with dinitrophenol is discussed.

## INTRODUCTION

A synthetic discussion of the influence of humic compounds on the mineral nutrition of plants may be found in the monograph of Trojanowski (1973) and papers by Gumiński (1968, 1972) and Lisiak (1976). The influence of humic compounds on the accessibility of the particular mineral components of the soil to plants, their content in plants and on the rate of mineral salt ions uptake were investigated. The latter problem has been tackled by but few authors. B. Niklewski and Wojciechowski (1938), Wojciechowski and Kużdowicz (1939) and Duda (1947) studied ion uptake by roots of intact plants, Dell'Agnola and Ferrari (1971) followed the influence of humate on sulphate ions uptake by detached roots and Vaughan and MacDonald (1971) experimented on tissue disks cut out of beet roots. Recently Tichý (1979) investigated the effect

of treatment of maize roots with humate on iron uptake by seedlings after their transfer from aqueous humate solution to a mineral medium, These studies revealed that humic acids used in appropriate concentrations enhance as a rule mineral salts cation and anion uptake, chloride anions excepted, their uptake being even inhibited.

At present similar investigations were undertaken with the application of modern methods which enable to simultaneously study uptake and accumulation of certain elements in the roots and shoots, and to test whether humic acids affect the so-called "active" or "passive" processes.

## MATERIAL AND METHODS

Three-week-old tomato seedlings variety "Mory 33" from water cultures were used for the experiments. The composition of the nutrient solution was as follows: 1) macroelements (grams per $dm^3$ of water): $Ca(NO_3)_2 \cdot 4H_2O$ — 0.59, $KNO_3$ — 0.25, $KH_2PO_4$ — 0.07, $MgSO_4 \cdot 7H_2O$ — 0.24, iron citrate — 0.025; 2) microelements (milligrams per $dm^3$ of medium): $H_3BO_3$ — 1.43, $MnCl_2 \cdot 4H_2O$ — 0.9, $ZnSO_4 \cdot 7H_2O$ — 0.111, $H_2MoO_4$ — 0.042, $CuSO_4 \cdot 5H_2O$ — 0.039.

The plants were grown in summer in a well aerated glasshouse under natural light, four specimens in 1 — $dm^3$ glass jars. After 10 days of vegetation the medium was changed. Plants of similar length were chosen from the material, the roots were washed in distilled water and they were placed for two or three days on medium deprived of the element the uptake of which was to be tested. After this period of "starving" the plants were placed in porcelain jars, four in each, containing 200 $cm^3$ of an equilibrated solution of several mineral salts.

For investigation of $K^+$, $Rb^+$ and $Ca^{2+}$ uptake the 3-component solution of Broyer and Hoagland (1943) was used, consisting of $Ca(NO_3)_2 \cdot 4H_2O$ — 1.18, $KNO_3$ — 0.26, and $KH_2PO_4$ — 0.068 g/$dm^3$ of water at pH 6.4 · $^{86}Rb^+$ relative $^{45}Ca^{2+}$ — traces.

In tests of $Mg^{2+}$ uptake the following solution was used: $Ca(NO_3)_2 \cdot 4H_2O$ — 1.18, $KNO_3$ — 0.51, $KH_2PO_4$ — 0.14, $MgCl_2 \cdot 6H_2O$ — 0.4 g/$dm^3$ of water at pH 6.4. $^{86}Rb^+$ relative $^{45}Ca^{+2}$ — traces.

Two solutions were applied for iron testing: without and with phosphorus. The former solution consisted of $Ca(NO_3)_2 \cdot 4H_2O$ — 1.18, $KNO_3$ — 0.51, $MgSO_4 \cdot 7H_2O$ — 0.49, $FeCl_3$ — 0.025, KCl — 0.07; the latter contained $Ca(NO_3)_2 \cdot 4H_2O$ — 1.18, $KNO_3$ — 0.51, $KH_2PO_4$ — 0.14, $MgSO_4 \cdot 7H_2O$ — 0.49, $FeCl_3$ — 0.025 g/$dm^3$ water, pH of both solutions was 6.4.

For phosphorus uptake experiments the solution: $Ca(NO_3)_2 \cdot 4H_2O$ — 1.18, $KNO_3$ — 0.51, $KH_2PO_4$ — 0.14, $MgSO_4 \cdot 7H_2O$ — 0.49 g/$dm^3$ was applied. When the isotope $^{32}P$ was used, the solution contained addi-

tionally 0.025 g $FeCl_3$ and 0.068 g $KH_2{}^{32}PO_4$. pH was varied, 5, 6 and 7, with the isotope 6.4.

The solution for determination of ammonium ions uptake contained: $(NH_4)_2SO_4$ — 0.82, $KH_2PO_4$ — 0.068, $CaCl_2$ — 0.552, KCl — 0.188; for uptake of nitrate ions: $Ca(NO_3)_2 \cdot 4H_2O$ — 1.46, $KH_2PO_4$ — 0.068, KCl — 0.188 g/dm$^3$ of water. The total nitrogen amount was the same in both solutions. The nitrogen compounds contained 50 per cent of $^{15}N$. The pH used was 5, 6 and 7.

In experiments with $Cl^-$ the same solution was used as for $Mg^{2+}$.

After 24, 48 or 72 h the plants were removed from the solution, the roots were washed with distilled water or (in iron uptake tests) with sodium citrate solution.

Ion uptake and accumulation of the elements in roots and shoots was investigated by two methods. A "chemical one" consisting in analysis of the ions remaining in the solution after removal of the plants, and a "physical" one based on determination of the labelled elements in the plants.

In the "chemical" method the washings from the roots were combined with the solution in which the roots were previously immersed and the solution was made up to 500 cm$^3$. Potassium and calcium were determined with a flame photometre, magnesium colorimetrically with the use of titanium yellow, phosphorus also colorimetrically after Fiske-Subbarow (1925) with the use of Eikonogen, ammonium ions by Kjeldahl's method, nitrate ions colorimetrically with phenolsulphonic acid, chloride ions by titration with silver nitrate against dichlorofluorescein (Struszyński 1957, Marczenko 1968, Mejbaum-Katzenellenbogen and Mochnacka 1966).

The amount of ions absorbed was calculated as the difference between their initial content in the solution and that after removal of the plants.

The radioactive elements were determined in the plant material with a Geiger-Müller counter, and $^{15}N$ with a mass spectrometre (Tölgyessy and Varga 1974).

To the solution from which the plants took up the tested ions sodium humate was added or not alternately. The humate dose was 100 mg/1 dm$^3$ of salt solution, this dose having been found optimal in water cultures in earlier experiments. Sodium humate was obtained from leaf compost by application of HCl and NaOH solution (Gumiński and Sulej 1967). It contained 8.7 per cent ash.

The initial pH of the salt solutions was adjusted with HCl or NaOH.

Two methods were used for distinguishing "passive" and "active" ion uptake. The first consisted in that part of the plants were left to transpire without limitation and part were covered with glass beakers.

The second way consisted in that the salt of the phosphorylation inhibitor, 2,4-dinitrophenol (DNP), was introduced into the solution in a $10^{-5}$ concentration. This concentration was chosen after Stachurska (1967) and on the basis of our preliminary trials.

In the "chemical" method four replications were run for each treatment. The single replicate consisted of four plants in one vessel. Each plant was a replication in radioactivity determination. The results are given as means obtained from the replications with mean deviation. The methods used for isotope determination did not allow statistical elaboration, it can only be said that the measurement error did not exceed 8 per cent.

## RESULTS

### POTASSIUM AND RUBIDIUM

It was ascertained by the chemical method that the amount of $K^+$ in the solution diminished under the influence of humate both in free and limited transpiration conditions. DNP reduced the $K^+$ loss in conditions of free transpiration, and when it was limited, DNP led to a negative exchange balance of these ions between the roots and the salt solution. Humate attenuated the effect of DNP in limited transpiration, but did not influence this effect in free transpiration (Table 1).

Table 1

Potassium uptake by tomato seedlings. Duration of uptake 48 h, solution pH 6.4

| Treatment | $K^+$ loss from solution, mg | |
|---|---|---|
| | free transpiration | restricted transpiration |
| Control | 3.8±0.08 | 2.5±0.06 |
| DNP | 2.0±0.07 | —4.0±0.06 |
| Humate | 5.3±0.03 | 3.5±0.04 |
| Humate+DNP | 2.0±0.09 | —1.0±0.04 |

Investigations on labelled rubidium uptake gave similar results as those with potassium when the chemical method was used. They, moreover, revealed that humate stimulates $^{86}$rubidium accumulation both in roots and shoots. It was found that DNP did not completely abolish the uptake of this ion, although it was greatly limited. Humate reduced inhibition of rubidium accumulation evoked in roots and shoots by DNP (Table 2).

Table 2

Uptake and accumulation of radioactive rubidium ($^{86}Rb$) by tomato seedlings. Duration of uptake 48 h, pH of solution 6.4

| Treatment | Free transpiration | | | |
|---|---|---|---|---|
| | radioactivity, KBq×g$^{-1}$ dry mass | | $^{86}Rb$ content in relation to amount added to pot per gram of dry mass, % | |
| | shoots | roots | shoots | roots |
| Control | 20.78 | 56.63 | 1.28 | 3.62 |
| DNP | 8.66 | 22.65 | 0.53 | 1.40 |
| Humate | 25.84 | 68.93 | 1.59 | 4.25 |
| Humate+ DNP | 11.59 | 39.64 | 0.71 | 2.90 |
| | Restricted transpiration | | | |
| Control | 13.95 | 33.83 | 0.86 | 2.09 |
| DNP | 5.49 | 21.96 | 0.34 | 1.35 |
| Humate | 14.07 | 38.04 | 0.86 | 2.35 |
| Humate+ DNP | 7.94 | 24.65 | 0.49 | 1.52 |

Total radioactivity of medium in pot = 1617 KBq.

CALCIUM

The chemical method did not reveal any influence of humate on $Ca^{2+}$ uptake in free transpiration. When the latter was limited only a tendency to enhanced uptake of these ions was noted under the influence of humate. Strictly speaking, this result concerns the balance of ion exchange. Inhibition of calcium accumulation in plants caused by DNP was slight and more pronounced in limited than free transpiration; interaction with humate was not noticeable (Table 3).

Table 3

Calcium uptake by tomato seedlings.
Duration of uptake 48 h, pH of solution 6.4

| Treatment | $Ca^{2+}$ loss from solution, mg | |
|---|---|---|
| | free transpiration | restricted transpiration |
| Control | 8.9±0.03 | 8.5±0.04 |
| DNP | 8.0±0.03 | 7.0±0.05 |
| Humate | 9.1±0.12 | 9.0±0.07 |
| Humate+DNP | 7.5±0.10 | 7.4±0.03 |

Application of labelled calcium showed that in the case of free transpiration, humate enhanced calcium accumulation both in roots and shoots, whereas, when transpiration was limited, stimulation was noted only in roots. DNP in free transpiration distinctly inhibited calcium accumulation in shoots and enhanced it in roots. In limited transpiration dinitrophenol also caused increased calcium accumulation in roots and had no noticeable influence on accumulation in the shoots. Humate neutralised stimulation of Ca accumulation in roots due to DNP in the case of free transpiration, and reduced accumulation in roots in limited transpiration in the presence of DNP. Apart from this, humate showed no interaction with DNP (Table 4).

Table 4

Uptake and accumulation of radioactive ($^{45}Ca$) by tomato seedlings. Duration of uptake 24 h, pH of solution 6.4

| Treatment | Free transpiration | | | |
|---|---|---|---|---|
| | radioactivity, KBq×g$^{-1}$ dry mass | | $^{45}Ca$ content in relation to amount added to pot per gram of dry mass, % | |
| | shoots | roots | shoots | roots |
| Control | 0.251 | 1.107 | 0.103 | 0.456 |
| DNP | 0.177 | 3.334 | 0.073 | 1.370 |
| Humate | 0.388 | 1.498 | 0.160 | 0.612 |
| Humate+ DNP | 0.175 | 0.947 | 0.072 | 0.393 |
| | Restricted transpiration | | | |
| Control | 0.052 | 0.090 | 0.021 | 0.032 |
| DNP | 0.066 | 0.137 | 0.016 | 0.056 |
| Humate | 0.060 | 0.157 | 0.024 | 0.065 |
| Humate+ DNP | 0.043 | 0.116 | 0.018 | 0.042 |

Total radioactivity of medium in pot=242 KBq.

## MAGNESIUM

Two experiments were performed in conditions of free transpiration with the use of the chemical method. It was noted that magnesium ions were somewhat more intensively taken up at pH 5 than at pH 7. Humate in general showed a tendency to stimulation of $Mg^{2+}$ uptake, while DNP distinctly inhibited it. When they were applied jointly the effect in nearly all cases was intermediate (Table 5).

Table 5

Magnesium uptake by tomato seedlings. Duration of uptake 24 h

| Treatment | Loss of $Mg^{2+}$ from solution, mg | | | | | |
|---|---|---|---|---|---|---|
| | pH 5 | | pH 6 | | pH 7 | |
| | total loss | mg×g$^{-1}$ dry mass | total loss | mg×g$^{-1}$ dry mass | total loss | mg×g$^{-1}$ dry mass |
| **Experiment 1** | | | | | | |
| Control | 3.58±0.09 | 1.33±0.04 | 3.33±0.10 | 1.23±0.05 | 2.92±0.08 | 1.17±0.06 |
| DNP | 2.21±0.06 | 0.81±0.04 | 2.08±0.10 | 0.73±0.03 | 1.54±0.07 | 0.55±0.02 |
| Humate | 3.55±0.04 | 1.41±0.04 | 3.53±0.06 | 1.23±0.06 | 3.31±0.04 | 1.20±0.03 |
| Humate+ DNP | 2.10±0.03 | 0.78±0.03 | 2.66±0.05 | 1.00±0.02 | 2.58±0.10 | 0.93±0.03 |
| **Experiment 2** | | | | | | |
| Control | 1.58±0.15 | 0.62±0.03 | 1.52±0.10 | 0.57±0.03 | 1.53±0.15 | 0.56±0.03 |
| DNP | 0.93±0.07 | 0.44±0.03 | 1.05±0.06 | 0.41±0.01 | 1.11±0.06 | 0.37±0.02 |
| Humate | 1.81±0.02 | 0.71±0.04 | 1.78±0.11 | 0.68±0.03 | 1.46±0.04 | 0.55±0.01 |
| Humate+ DNP | 1.33±0.04 | 0.53±0.02 | 1.54±0.05 | 0.63±0.04 | 1.53±0.07 | 0.54±0.01 |

### IRON

Iron uptake was measured by the isotope method in free transpiration conditions in two variants — without and with phosphorus.

The plants took up larger quantities of iron in the solution without phosphorus than in that containing phosphate ions. In both cases humate distinctly inhibited iron accumulation in the roots, but rather favoured it in the shoots. No major difference was found in the influence of humate on iron accumulation in both the tested solutions.

Dinitrophenol markedly inhibit iron accumulation in the shoots, but did not have a distinct influence on its accumulation in the roots. Humate enhanced the inhibitory action of DNP on iron accumulation in shoots, and in roots DNP reduced the inhibitory effect of humate (Table 6).

### PHOSPHORUS

The chemical method revealed that humate enhances phosphate ions uptake from solutions with pH 5, 6 and 7 which do not contain iron, whereas DNP neutralised this influence. DNP itself showed a weak tendency to stimulation of phosphate accumulation (Table 7).

Enhanced by humate phosphate ion uptake from the solution free of iron was demonstrated also by the isotopic method. Accumulation

Table 6

Uptake and accumulation of radioactive iron ($^{59}Fe$) by tomato seedlings. Duration of uptake 24 h, pH of solution 6.4

| Treatment | Solution without phosphorus | | | |
|---|---|---|---|---|
| | radioactivity, KBq×g$^{-1}$ dry mass | | $^{59}Fe$ content in relation to amuunt added to pot per gram of dry mass, % | |
| | shoots | roots | shoots | roots |
| Control | 1.85 | 83.22 | 0.081 | 3.63 |
| DNP | 0.55 | 73.07 | 0.02 | 3.19 |
| Humate | 1.87 | 38.92 | 0.08 | 1.70 |
| Humate+ DNP | 0.40 | 41.69 | 0.02 | 1.82 |
| | Solution with phosphorus | | | |
| Control | 1.06 | 40.79 | 0.04 | 1.78 |
| DNP | 0.56 | 45.93 | 0.02 | 2.00 |
| Humate | 1.19 | 26.32 | 0.05 | 1.15 |
| Humate+ DNP | 0.32 | 36.07 | 0.01 | 1.57 |

Total radioactivity of medium in pot = 2290 KBq.

occurred mainly in the roots. When, however, the solution contained $FeCl_3$, no stimulation was observed, but a slight decrease of accumulation under the influence of humate both in shoots and roots. DNP distinctly inhibited phosphorus accumulation in shoots and depressed it somewhat in roots if the solution did not contain iron. When $FeCl_3$ was present in the solution, DNP slightly inhibited phosphorus accumulation in shoots, and in the roots even showed a tendency to stimulation. DNP completely abolished the effect of humate if there was no iron in the solution. In the presence of $FeCl_3$ inhibition of P accumulation was

Table 7

Phosphorus uptake by tomato seedlings. Duration of uptake 24 h

| Treatment | Loss of $PO_4^{3-}$ from solution, mg | | | | | |
|---|---|---|---|---|---|---|
| | pH 5 | | pH 6 | | pH 7 | |
| | total loss | mg×g$^{-1}$ dry mass | total loss | mg×g$^{-1}$ dry mass | total loss | mg×g$^{-1}$ dry mass |
| Control | 2.44±0.16 | 0.94±0.06 | 2.31±0.06 | 0.97±0.01 | 2.26±0.04 | 0.91±0.01 |
| DNP | 2.28±0.09 | 1.02±0.04 | 2.37±0.10 | 1.01±0.02 | 2.34±0.16 | 0.97±0.02 |
| Humate | 2.97±0.12 | 1.12±0.04 | 3.14±0.13 | 1.29±0.05 | 3.16±0.11 | 1.15±0.03 |
| Humate+ DNP | 2.08±0.06 | 0.96±0.01 | 2.46±0.05 | 1.02±0.04 | 2.48±0.13 | 1.03±0.03 |

more pronounced in the combination DNP+humate than when each of these substances was applied separately. Accumulation of phosphorus in roots was the same as when DNP was used alone (Table 8).

Table 8

Uptake and accumulation of radioactive phosphorus ($^{32}P$) in tomato seedlings. Duration of uptake 24 h, pH of solution 6.4

| Treatment | Solution without iron | | | |
|---|---|---|---|---|
| | radioactivity, KBq × g$^{-1}$ dry mass | | $^{32}P$ content in relation to amount added to pot per gram of dry mass, % | |
| | shoots | roots | shoots | roots |
| Control | 35.58 | 73.14 | 0.62 | 1.29 |
| DNP | 27.57 | 71.00 | 0.48 | 1.25 |
| Humate | 38.75 | 93.26 | 0.68 | 1.64 |
| Humate+ DNP | 26.78 | 71.01 | 0.48 | 1.25 |
| | Solution with iron | | | |
| Control | 35.68 | 84.36 | 0.62 | 1.48 |
| DNP | 34.83 | 87.94 | 0.61 | 1.55 |
| Humate | 33.73 | 81.29 | 0.59 | 1.43 |
| Humate+ DNP | 29.92 | 87.99 | 0.52 | 1.53 |

Total radioactivity of medium in pot = 5690 KBq.

NITROGEN

Uptake and accumulation of $NH_4^+$ and $NO_3^-$ ions with free transpiration was tested by determining the loss of ions in the solution and $^{15}N$ content in whole plants. Two experiments were performed: in the first DNP was not added to the salt solution and in the second it was added. It results from these experiments that in the presence of humate there was only a tendency to taking up $NH_4^+$ and $NO_3^-$ ions (Table 9). Addition of DNP to the salt solution abolished this doubtful effect of humate (Table 10).

CHLORINE

Uptake of chloride ions was investigated only by determination of the loss of the ions in the salt solution. The experiments were performed twice with free transpiration. Humate inhibited strongly chloride ion uptake, the effect was relatively weaker at pH 5 than at 6 and 7.

DNP alone did not exert any major influence on chloride uptake, it weakened, however, the inhibitory influence of humate (Table 11).

Table 9

$NH_4^+$ and $NO_3^-$ ions uptake by tomato seedlings. Duration of uptake 24 h

| Treatment | $(NH_4)_2SO_4$ solution | | | | $Ca(NO_3)_2$ solution | | | |
|---|---|---|---|---|---|---|---|---|
| | nitrogen content in plant, $^{15}N$ | | nitrogen taken up from solution, $N—NH_4^+$ | | nitrogen content in plant, $^{15}N$ | | nitrogen taken up from solution, $N—NO_3^-$ | |
| | $mg \times g^{-1}$ dr.m. | mg per jar | $mg \times g^{-1}$ dr.m. | mg per jar | $mg \times g^{-1}$ dr.m. | mg per jar | $mg \times g^{-1}$ dr.m. | mg per jar |
| Solution without humate | | | | | | | | |
| pH 5 | 2.15 | 3.80 | 2.95 | 5.34±0.29 | 3.98 | 7.24 | 6.42 | 11.78±0.30 |
| pH 6 | 1.98 | 3.74 | 2.98 | 5.65±0.14 | 3.63 | 7.07 | 6.24 | 12.27±0.56 |
| pH 7 | 2.27 | 4.42 | 3.15 | 6.06±0.25 | 3.60 | 6.94 | 6.15 | 11.85±0 35 |
| Solution with humate | | | | | | | | |
| pH 5 | 2.41 | 4.27 | 3.05 | 5.45±0.17 | 3.95 | 7.29 | 7.68 | 14.37±0.31 |
| pH 6 | 2.20 | 4.24 | 2.92 | 5.68±0.12 | 3.73 | 6.95 | 7.38 | 13.37±0.61 |
| pH 7 | 2.42 | 4.62 | 3.20 | 6.10±0.14 | 3.68 | 7.52 | 6.25 | 12.75±0.52 |

dr. m. — dry mass.

Table 10

$NH_4^+$ and $NO_3^-$ ions uptake by tomato seedlings in the presence of 2,4-dinitrophenol (DNP). Duration of uptake 24 h

| Treatment | Solution with $(NH_4)_2SO_4$+DNP | | | | Solution with $Ca(NO_3)_2$+DNP | | | |
|---|---|---|---|---|---|---|---|---|
| | nitrogen content in plant, $^{15}N$ | | nitrogen taken up from solution, $N—NH_4^+$ | | nitrogen content in plant, $^{15}N$ | | nitrogen taken up from solution, $N—NO_3^-$ | |
| | $mg \times g^{-1}$ dr.m. | mg par jar | $mg \times g^{-1}$ dr.m. | mg per jar | $mg \times g^{-1}$ dr.m. | mg per jar | $mg \times g^{-1}$ dr.m. | mg per jar |
| Solution without humate | | | | | | | | |
| pH 5 | 1.83 | 6.77 | 2.27 | 8.42±0.18 | 2.54 | 10.36 | 4.43 | 18.27±0.20 |
| pH 6 | 1.75 | 6.06 | 2.40 | 8.36±0.16 | 2.48 | 10.72 | 4.40 | 17.75±0.30 |
| pH 7 | 1.28 | 4.81 | 1.70 | 6.38±0.20 | 2.38 | 9.65 | 4.32 | 18.38±0.12 |
| Solution with humate | | | | | | | | |
| pH 5 | 1.38 | 4.95 | 1.99 | 7.10±0.18 | 2.65 | 10.41 | 4.66 | 18.35±0.15 |
| pH 6 | 1.50 | 5.29 | 2.04 | 7.20±0.15 | 2.20 | 9.26 | 4.18 | 17.58±0.22 |
| pH 7 | 1.52 | 5.58 | 2.00 | 7.32±0.14 | 2.60 | 10.33 | 4.42 | 18.70±0.20 |

dr. m. — dry mass.

Table 11

Chlorine uptake by tomato seedlings. Duration of uptake 24 h

| Treatment | Loss of $Cl^-$ from solution, mg | | | | | |
|---|---|---|---|---|---|---|
| | pH 5 | | pH 6 | | pH 7 | |
| | total loss | mg×g$^{-1}$ dry mass | total loss | mg×g$^{-1}$ dry mass | total loss | mg×g$^{-1}$ dry mass |
| **Experiment 1** | | | | | | |
| Control | 2.05±0.08 | 0.76±0.05 | 2.65±0.09 | 0.96±0.05 | 3.07±0.10 | 1.22±0.04 |
| DNP | 2.45±0.10 | 0.90±0.04 | 2.32±0.12 | 0.82±0.04 | 2.65±0.13 | 0.97±0.04 |
| Humate | 0.90±0.04 | 0.37±0.01 | 0.71±0.03 | 0.25±0.01 | 0.80±0.04 | 0.33±0.01 |
| Humate+ DNP | 1.90±0.09 | 0.70±0.02 | 1.50±0.08 | 0.55±0.03 | 1.80±0.09 | 0.65±0.03 |
| **Experiment 2** | | | | | | |
| Control | 2.10±0.01 | 0.83±0.05 | 2.30±0.10 | 1.00±0.04 | 2.35±0.11 | 1.04±0.03 |
| DNP | 2.05±0.02 | 0.92±0.06 | 2.41±0.02 | 0.93±0.03 | 2.80±0.07 | 0.92±0.04 |
| Humate | 1.27±0.07 | 0.61±0.04 | 1.43±0.04 | 0.54±0.02 | 1.62±0.06 | 0.55±0.05 |
| Humate+ DNP | 2.01±0.10 | 0.80±0.03 | 1.80±0.06 | 0.70±0.02 | 2.30±0.02 | 0.80±0.03 |

## DISCUSSION

Comparison of the present results with those of B. Niklewski and Wojciechowski (1938), Wojciechowski and Kuždowicz (1939) and Duda (1947) shows in general good agreement. These authors, however, recorded a much stronger stimulation of ammonium, nitrate and phosphate ions uptake by humic acids than it results from the present studies. If we consider that these authors noted a stronger stimulation at lower concentrations of humic acids than those used at present and that the concentrations applied by us weakened the stimulating effects in the studies of earlier authors, the agreement of our results with theirs seems satisfactory. It should be added that in the present experiment higher humate concentrations were used purposely because they gave the best effects as regards plant growth in water cultures (Gumiński 1972). As seen, there is no parallelism between optimal humate concentration for plant growth in long lasting water cultures and optimal concentration for $NH_4^+$, $NO_3^-$ and $PO_4^{3-}$ ion uptake in experiments of short duration.

The use by us of the method of labelled elements detection in plants, beside the method of analytical determination of the ion loss from the solution, allowed to distinguish the influence of humate on ion uptake from its effect on the exchange between roots and the external solution; the possibility of such a distinction was facilitated by the application of DNP, a phosphorylation inhibitor. The use of the latter and variation

of transpiration conditions made possible the obtention of information, whether ion uptake or accumulation in roots and shoots is of "active" or "passive" character.

It results from the present studies that sodium humate in optimal concentration for plant vegetation in water cultures stimulates markedly $K^+$ or $Rb^+$ ion uptake and accumulation in roots and shoots and prevents "passive" $K^+$ outflow from the roots. The reduction by humate of efflux of potassium ions caused by DNP may be interpreted as blocking by humate of $K^+$ efflux owing to its tannin-like action on the cytoplasm (Gumiński and Sulej 1979).

The strong inhibition by humate of chloride ion uptake noted by us is a complete confirmation of the investigations of Vaughan and MacDonald (1971) performed on different material and by different techniques.

The present results concerning potassium and chloride ions explain to some extent the results of investigations by Gumińska and Gracz-Nalepka (1972) which indicate that, in the presence of humate in the medium, increased doses of potassium chloride have a more favourable effect on the yield than when the medium does not contain humate. The view of M. Niklewski et al. (1972) on the influence of humate in relation to potassium fertilisation might be interpreted similarly on the basis of our results. It should, namely, be borne in mind that the main component of the potash salt is KCl. It should be added here that Tan (1978) found that humic acids release potassium bound in the soil in aluminosilicates.

DNP inhibited uptake and accumulation by plants not only of potassium (or rubidium), but also of magnesium and iron. The interaction of dinitrophenol with humate here seems complicated to us. No doubt the formation humate chelates with metal cations and the possibility of their migration in the plant should be taken into account. Chelates do not form with potassium or rubidium, but they form with calcium, magnesium and iron. Chelation, uptake and transport of these elements in plants are interrelated and dependent on external and internal pH. It is noteworthy that humate greatly reduced iron accumulation in roots, whereas it did not restrict this process in the shoots. It may be concluded therefore that humate facilitates transport of the taken up iron from roots to shoots. This conclusion agrees with the results of Tichý (1979) who used a different method. Since in the present investigations humate did not reduce the inhibition of iron accumulation in the shoot caused by DNP, on the contrary, it enhanced this inhibition, it would seem that humate facilitates only "passive" transport of iron to the shoots, and is rather an obstacle in the "active" process. The results concerning calcium may be similarly interpreted.

In experiments with magnesium only the loss of ions from the solution was determined, therefore, it can only be affirmed that humate favoured magnesium accumulation in the whole plant. This action was weak and prompted us to repeat the experiment. Since DNP lowered magnesium accumulation, this process in the plant must have been partly of "active" character. The intermediate value of the results of analyses for the combination humate+DNP as compared with those when humate alone or DNP alone were used seem to indicate that humate enhanced only "passive" uptake.

Since humate favoured phosphorus accumulation when iron was not present in the solution and did not give such an effect in its presence, it could be inferred that calcium binding by humate facilitates phosphorus accumulation, whereas iron binding does not facilitate it. It should be mentioned that at pH 6.4 iron chelation by humate is much stronger than calcium chelation (Aso and Takenaga 1975). It seems the phosphate-iron-humic complexes are performed. Dinitrophenol inhibited phosphate ions accumulation (mainly in shoots) only when there was no iron in the salt solution, and humate had no influence on this effect involved no doubt in the "active" process. These observations supplement the interpretation of the results of Lisiak (1978) concerning the effect of sodium humate on phosphorus utilisation by plants in water cultures at various iron and calcium doses.

Interaction of DNP with humate in relation to chloride ion accumulation remains obscure, however, a repeated experiment gave the same result. DNP alone did not show any distinct influence on accumulation of these ions, although their uptake is considered to be "active". Humate inhibited strongly no doubt $Cl^-$ ion uptake (or strongly stimulated their efflux) and dinitrophenol greatly reduced the effectiveness of humate. To elucidate this special studies should be carried out taking into account the hypothesis of Vaughan and MacDonald (1971) who suggest the possibility of humic acids blocking the special chloride ion carriers.

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## *Wpływ humianu sodowego na pobieranie niektórych jonów przez siewki pomidorów*

Streszczenie

Badano wpływ humianu sodowego na pobieranie przez trzytygodniowe siewki pomidorów jonów $K^+$, $Rb^+$, $NH_4^+$, $Mg^{2+}$, $Ca^{2+}$, $Fe^{3+}$, $PO_4^{3-}$, $NO_3^-$ i $Cl^-$ ze zbalansowanych roztworów wodnych soli. Różnicując transpirację i stosując dwunitrofenol rozróżniano pobieranie "bierne" i "czynne". Oznaczano ubytek jonów w roztworach stosując zwykłe metody analizy chemicznej oraz badano akumulację pierwiastków znakowanych w korzeniach i pędach mierząc radioaktywność lub zawartość izotopu ciężkiego. Humian silnie stymulował pobieranie $K^+$ i $Rb^+$ oraz silnie hamował pobieranie $Cl^-$ a lekko wzmagał pobieranie $Mg^{2+}$ i $PO_4^{3-}$. Ponadto ułatwiał transport żelaza z korzeni do pędów. Nie wywierał wyraźnego wpływu na pobieranie i akumulację azotu. Efektywność humianu odnosiła się raczej do pobierania i transportu "biernego" niż do "czynnego". Przedyskutowano współdziałanie humianu z dwunitrofenolem.