AN ABSTRACT OF THE THESIS OF

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 Title:
 The Effects of L-glutamate, Mono-potassium Salt on Whole

Live Spinach Plants
Abstract Approved:

Spinach plants were grown under greenhouse conditions in hopes of determining the affects of L-glutamate, mono-potassium salt solution on whole living plants. Both the leaves and the roots were treated with the solution in this experiment. In all of the previous research only leaf discs were used under laboratory conditions. The results of the previous research revealed a significant increase in cell numbers produced when treated with the solution. In the present study, the growth conditions of whole live plants varied greatly from the previous research, even though the concentration of the solution and the amount of time in the solution were the same for both studies.

The results of this study differed greatly from all other studies. The solution was actually detrimental to the spinach plants whose root systems were bathed in the solution. They all died before the end of the forty day-treatment period.

Five measurements were taken, and two of those measurements revealed a reduction in size when compared to the control plants. The plants treated with a solution sprayed on the leaves exhibited fewer leaves per plant and a reduced fresh weight. The sprayed plants did exhibit larger leaves and a greater dry weight.

THE EFFECTS OF L-GLUTAMATE, MONO-POTASSIUM SALT ON WHOLE LIVE SPINACH PLANTS

A Thesis

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by

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Graduate Council

Approved for

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INTRODUCTION

As the human population increases so to does the need for finding ways of increasing the world's supply of food. One method of increasing that supply would be to increase the yields of the major food crops such as maize, wheat, and garden crops. That can be accomplished by improving the rate of photosynthesis. Some plants such as maize and sugarcane have exhibited large increases in yields over the past 25 years, where other crops, such as rice, wheat, and soybeans have had only partial success at yield increases (Bassham, 1977).

In 1948, the research of Calvin, Bensen, and Bassham of the University of California, Berkley, found that certain species of plants, such as grains, legumes, fruits, and vegetables, were a C_3 species type, meaning that when radioactive ${}^{14}CO_2$ is supplied to a leaf in light for a short time, the first detectable product is the C_3 compound phosphogylcerate (Zelitch, 1979). This information then led to the work, in 1965, of Kortachad, at the experimental station of the Hawaiian Sugar Planter's Association. His research identified the C_4 species of plants. The C_4 plants (in this case sugarcane) produce at the beginning of photosynthesis the C_4 compounds malate and aspartate. Other C_4 species were then discovered to be maize, sorghum, millet, and certain weeds.

As the process of photosynthesis begins in the C_3 species the phosphoglycerate of glycolate synthesized is then further oxidized by glyoxylate. This oxidation process causes some of the carbon dioxide assimilated during photosynthesis to be lost by photorespiration (Oliver and Zelitch, 1977). This loss may be as high as 50% in some species (Zelitch, 1975). The C_3 species also release carbon dioxide three to five times faster during photorespiration than during "dark" respiration (Zelitch, 1974). By blocking the path of glycolate oxidation, photorespiratory carbon dioxide is not wasted by its release into the atmosphere but is taken up by photosynthesis, thus creating a more photosynthetically efficient plant.

Glycilic acid, L-glycidate, aspartate, and glutamate have been found to inhibit or block the oxidation of glycolate. In 1975 L-glutamate was successfully used to regulate the synthesis of glycolate (Oliver and Zelitch, 1977). Since 1978 glutamate and aspartate have had the most success at blocking the oxidation process of glycolate (Oliver, 1978).

Estimates of the mean crop growth-rate (dry weight produced per square meter of land per week) in the United States for maize, sorghum, and sugarcane show that these species have two to three times the crop growth-rate of the less efficient species such as spinach, tobacco, and hay (Zelitch, 1971). This enormous difference is due to the rapid rates of net carbon dioxide assimilated, the production of the 4-carbon compounds malate and aspartate, and the reduced production of glycolate by the C_4 species. Thus the C_4 plants are more photosynthetically efficient.

The concentrations of L-glutamate as described by Zelitch (1979)

and spinach, a C₃ species also used by Zelitch, were used in this experiment. The experimental technique was varied from that of Zelitch by using whole, live spinach plants instead of leaf discs. The whole plants were grown under greenhouse conditions and the study was designed to determine the affects of L-glutamate, monopotassium salt solution on whole, live spinach plants.

METHODS AND MATERIALS

Bloomsdale Long Standing was the variety of spinach used in this study. Nine flats of "Jiffy" peat trays were used as containers and equal parts of peat moss and perlite were utilized as the growing medium. Each flat was sown with 20 spinach seeds. All nine flats were then transferred to the greenhouse and placed in a germination bed which was held at a constant temperature of 21°C. Upon germination and emergence, the flats were randomly placed in a sunlight receiving location in the greenhouse. Since the temperature in the greenhouse could not be kept constant, it varied with each The mean temperature for Test I was 19.76°C with the highest test. reading at 25°C and the lowest being 8°C. During Test II the highest reading was 22°C and the lowest being at 13°C. For Test III the average temperature was 22°C with little variation in temperature throughout the test. Those readings reflected the temperature of the greenhouse between 8:00 and 10:00 a.m.

After the spinach plants had developed a set of leaves, they were fertilized with Ortho's 12-6-6 analysis liquid fertilizer. Each flat was also thinned to ten evenly spaced plants at that time.

Three different treatments were used for each test. In Treatment I three of the nine flats were flooded with a 30mm solution of L-glutamate, mono-potassium salt. The solution was allowed to remain in the medium for approximately two hours, after which the medium was flooded with distilled water. For Treatment II the L-glutamate solution was sprayed on to the leaves of the plants and the medium was flooded with distilled water only. After two hours these plants were sprayed again with distilled water. A piece of cardboard was placed next to the flats being sprayed to prevent any drift of the L-glutamate. Treatment III was the control. This group consisted of the remaining three flats and the plants were treated with distilled water only.

Each test was replicated three times and each ran for approximately forty days. At the end of each test the plants were removed from the medium and this included removing all particles of medium from the roots. The following data were then recorded: number of leaves per plant, length of the leaf blades, width of the leaf blades (at the widest point), and fresh mass of each plant. Each plant was then individually labeled, oven dried, and the dry mass determined and recorded.

Means of the data from each of the three treatments were calculated and those means were compared statistically by using a Student t test at the .05 level of significance.

RESULTS AND DISCUSSION

The results of this study indicated that there were some differences in the growth of spinach plants after being treated with L-glutamate, mono-potassium salt solution when compared to the control plants. The most apparent difference was in the spinach plants whose root systems were bathed in the L-glutamate solution (Table I). All of those plants died in all three tests before the termination of the forty day growth period. Prior to their death the plants exhibited an unhealthy appearance after approximately 21 days of treatment. A definite difference in their growth became apparent on the 26th day of Test I, the 24th day of Test II, and the 15th day of Test III. These plants appeared dwarfed when compared to the control, and the leaves were curled and dried at the edges. The overall appearance was much like that of plants suffering from fertilizer burn.

The difference between the control and those plants sprayed with the L-glutamate, mono-potassium salt solution was not great, but was evident in some areas. Of the five measurements taken, the sprayed plants had three higher mean measurements. When the data for leaf length was averaged the plants sprayed were larger at 4.26 cm in length while the control plants were somewhat smaller at 4.07 cm in length (Table II). The mean overall difference was .56 cm. The same was true for the leaf width measurements (Table III). The sprayed plants averaging 1.75 cm and the control plants 1.55 cm. The difference here was .19 cm. The mean dry mass of all the sprayed plants was

Treatment	Test	Leaf Length cm	Leaf Width cm	Number of Leaves	Fresh Mass gr	Dry Mass gr
	1	4.81	1.58	6.20	1.65	.16
Control	2	2.69	1.48	8.50	1.75	.55
	3	4.68	1.60	6.67	1.61	.18
	1	4.75	1.67	5.40	1.46	.23
Leaves Sprayed	2	3.37	1.93	7.97	1.64	.52
With L-glutamate	3	4.93	1.64	5.90	1.48	.22
	1					
Roots Bathed In	2					
L-glutamate	3					

Table I. Mean growth of spinach plants after 40 days of treatment with L-glutamate

-- Plants that died before the end of the 40 day treatment period.

Test					Tr	eatment	:			
		Control		L	eav	es Spra	yed	Root	ts Bath	ed
	1-3	H-1	G-8	c	-2	B-7	A~5	D9	E4	F-6
	6.70	5.00	5.00	5	.30	5.30	4.50			
	5.60	4.50		4	.30	5.20				
	7.00	4.20	3.80	5	.10	4.70	6.40			
	4.10	3.90	4.20	5	.50	6.60	2.20			
I	5.00	6.00	5.90	5	.00	6.50	4.60			
	5.60	5.70	4.40	6	- 80	5.50	2.80			
	6.80	4.60	5.70	5	.50	6.70	5.00			
	6.30	4.50	5.00	5	. 54	6.30	4.30			
	3.40	4.20	3.90	3	.30	5.20	3.90			
	6.10	3.60	3.80	4	.00	5.90	3.60		~-	
<u> </u>	3.25	2.10	2.80	2	.95	2.75	3.30			
	3.25	2.75	3.00		.05		3.75			
	2.70	2.05	2.90		.65		3.20			
	3.40	1.90	2.95		.85		2.75			
11	3.10	1.35	3.75		.65		2.95			
	3.25	2.30	2.80		.75		3.45			
	3.50	2.20	3.30		.30		4.15			
	2.30	1.75	2.95		.75		3.55			
	2.75	2.05	2.70		.80		3.60			
	2.95	2.35	2.50		.00		3.00	~-		
	6.40	3.70	3.80		.34					
	4.10	4.40	5.50		.50		3.80			<u> </u>
	5.30	4.10	5.70		.90		4.90			
	6.00	4.70	3.70		.70		4.20			
III	3.40	6.10	4.90		.80		2.70			
	4.30	5.80	5.10		.73		2.10			
	3.15	3.80	4.30		.21		4.50			
	6.40	4.40	4.50		.68		6.30			
	5.70	4.10	5.00		.10		5.10			~
	3.20	5.10	3.80	5	.00	5.80	4.40			
Plar	nts that	died t	efore	the e	nđ	of the	40 day	treatment	period	

Table II. Mean leaf blade length measured in cm after 40 days of treatment with L-glutamate

Test										
	с	ontrol		Leave	s Spra	yed	Roots Bathed			
	I-3	H-1	G-8	C-2	B-7	A-5	D-9	E-4	F-6	
	2.20	1.80	1.90	2.10	2.20	1.50				
	1.60	1.30		1.50	2.30					
	2.30	1.80	1.30	1.70	2.00	1.40				
_	1.70	1.40	1.50	1.70	2.20	1.00				
L	1.70	1.60	1.90	1.60	1.70	1.50				
	1.70	1.40	1.40	2.20	2.20	1.20				
	2.00	1.20	1.80	1.70	2.40	1.30				
	1.60	1.50	1.90	1.70	1.80	1.70				
	1.20	1.40	1.10	1.30	2.30	1.40	÷			
	2.00	1.90	1.30	1.40	2.00	1.10				
	1.50	1 25	1.65	1 10	1 50	1.65				
	1.50	1.25 1.55	1.65	2.20 1.80	1.50 2.00	1.65 2.20				
	1.50	1.50	1.75	1.00	1.95	2.20				
	1.95	1.40	1.80	2.10	1.75	1.50				
II	1.40	.65	1.70	2.15	1.65	2.00				
* *	.85	1.45	2.00	1.25	1.80	2.45				
	1.55	1.60	1.85	2.00	1.90	2.80				
	1.40	1.35	1.65	1.80	2.00	2.15				
	1.35	1.30	1.60	1.22	1.75	2.70				
	1.30	1.55	1.10	1.57	1.60	2.25				
		<u> </u>		<u> </u>		<u>.</u>				
	1.50	1.80	1.20	1.50	2.20	1.10				
	1.20	1.40	1.70	1.70	2.00	1.30				
	2.30		1.80	1.30	2.30	1.50				
	1.70	1.10	1.20	1.50	1.60	1.60				
III	1.80	1.70	1.80	1.80	1.70	1.10				
	2.10	1.50	1.90	2.10	2.10	1.50				
	1.90	1.30	1.40	1.70	1.40	1.40				
	1.70	1.20	1.50	1.40	1.80	1.60				
	2.00	1.70	1.90	1.50	1.90	1.50				
	1.30	1.70	1.40	1.90	2.30	1.00				

Table III. Mean leaf blade width measured in cm after 40 days of treatment with L-glutamate

-- Plants that died before the end of the 40 day treatment period.

also greater than that of the control plants (Table IV). The former being .33 grams and the latter .29 grams. The sprayed plants averaged .03 grams more in dry mass than the control plants.

The remaining mean measurements, number of leaves and fresh mass, were somewhat higher with regards to the control plants. The mean number for the control was 7.12 leaves per plant and for those sprayed with L-glutamate it was 6.42 leaves per plant, a difference of .70 leaves per plant (Table V). The overall mean for fresh mass was 1.67 grams for the control and 1.53 grams for the plants sprayed, a difference of .14 grams (Table VI).

There were obvious differences in the measurements but when compared statistically using a Student <u>t</u> test at the .05 level of significance, no significant differences were found.

ontrol H-1 G-8 .30 .24 .10 .21 .05 .22 .07 .21 .22 .07 .07 .11 .33 .03 .13 .18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	C-2 .25 .12 .63 .09 .13 .08 .09 .28 .09 .05 .73 .72 .30 .33 .58 .93	es Spr B-7 .18 .10 .23 .18 .25 .24 .21 .18 .17 .19 .22 .51 .20 .34 .45	A-5 .45 .43 .22 .30 .39 .31 .32 .53 .33 .33 .21 .70 .41 .45 .43	Roc D-9	ets Bat	F-6
.30 .24 .10 .21 .05 .22 .07 .21 .22 .07 .07 .11 .33 .03 .13 .18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.25 .12 .63 .09 .13 .08 .09 .28 .09 .05 .73 .72 .30 .33 .58 .93	.18 .10 .23 .18 .25 .24 .21 .18 .17 .19 .22 .51 .20 .34 .45	.45 .43 .22 .30 .39 .31 .32 .53 .33 .33 .21 .70 .41 .45	D-9	 	F-6
$\begin{array}{cccccccccccccccccccccccccccccccccccc$.12 .63 .09 .13 .08 .09 .28 .09 .05 .73 .72 .30 .33 .58 .93	.10 .23 .18 .25 .24 .21 .18 .17 .19 .22 .51 .20 .34 .45	 .43 .22 .30 .39 .31 .32 .53 .33 .33 .21 .70 .41 .45			
.21 .05 .22 .07 .21 .22 .07 .07 .11 .33 .03 .13 .18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.63 .09 .13 .08 .09 .28 .09 .05 .73 .72 .30 .33 .58 .93	.23 .18 .25 .24 .21 .18 .17 .19 .22 .51 .20 .34 .45	.43 .22 .30 .39 .31 .32 .53 .33 .21 .70 .41 .45			
.22 .07 .21 .22 .07 .07 .11 .33 .03 .13 .18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.09 .13 .08 .09 .28 .09 .05 .73 .72 .30 .33 .58 .93	.18 .25 .24 .21 .18 .17 .19 .22 .51 .20 .34 .45	.22 .30 .39 .31 .32 .53 .33 .33 .21 .70 .41 .45			
.21 .22 .07 .07 .11 .33 .03 .13 .18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.13 .08 .09 .28 .09 .05 .73 .72 .30 .33 .58 .93	.25 .24 .21 .18 .17 .19 .22 .51 .20 .34 .45	. 30 . 39 . 31 . 32 . 53 . 33 . 33 . 21 . 70 . 41 . 45			
.07 .07 .11 .33 .03 .13 .18 .05 .21 .06 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.08 .09 .28 .09 .05 .73 .72 .30 .33 .58 .93	.24 .21 .18 .17 .19 .22 .51 .20 .34 .45	. 39 . 31 . 32 . 53 . 33 . 33 . 21 . 70 . 41 . 45		 	
.11 .33 .03 .13 .18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.09 .28 .09 .05 .73 .72 .30 .33 .58 .93	.21 .18 .17 .19 .22 .51 .20 .34 .45	. 31 . 32 . 53 . 33 . 21 . 70 . 41 . 45		 	
.03 .13 .18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.28 .09 .05 .73 .72 .30 .33 .58 .93	.18 .17 .19 .22 .51 .20 .34 .45	. 32 . 53 . 33 . 21 . 70 . 41 . 45		 	
.18 .05 .21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.09 .05 .73 .72 .30 .33 .58 .93	.17 .19 .22 .51 .20 .34 .45	.53 .33 .21 .70 .41 .45		 	
.21 .06 .42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.05 .73 .72 .30 .33 .58 .93	.19 .22 .51 .20 .34 .45	.33 .21 .70 .41 .45		 	
.42 .44 .76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.73 .72 .30 .33 .58 .93	.22 .51 .20 .34 .45	.21 .70 .41 .45		 	
.76 .63 .44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.72 .30 .33 .58 .93	.51 .20 .34 .45	.70 .41 .45		 	
.44 .49 .49 .73 .15 .75 .43 .68 .81 .59	.30 .33 .58 .93	.20 .34 .45	.41 .45		 	
.49 .73 .15 .75 .43 .68 .81 .59	.33 .58 .93	.34 .45	.45			
.15 .75 .43 .68 .81 .59	.58 .93	.45				
.43 .68 .81 .59	.93		. 43			
.81 .59		E /.				
	72	.54	.68			
	.45	.43	1.20			
.40 .70	. 32	. 39	.69			
.31 .43	.68	. 49	.65			
.65 .49	.60	.40	.62	<u></u>		
.05 .05	.13	.13	.44			
		.15	.33			
.21 .08	.13	.11	. 39			
.05 .07	.26	.10	.32			
.24 .22	.18	.09	. 30			
.17 .28	.14	.17	.31			_
.12 .19	.21	.19	.23			_
.13 .40	.10	.11	.45			
.15 .12	.15	.18	.41			
.09 .22	.17	. 22	.35	~		
	.05 .07 .24 .22 .17 .28 .12 .19 .13 .40 .15 .12	.34.21.20.21.08.13.05.07.26.24.22.18.17.28.14.12.19.21.13.40.10.15.12.15	.34 .21 .20 .15 .21 .08 .13 .11 .05 .07 .26 .10 .24 .22 .18 .09 .17 .28 .14 .17 .12 .19 .21 .19 .13 .40 .10 .11 .15 .12 .15 .18 .09 .22 .17 .22	.34 .21 .20 .15 .33 .21 .08 .13 .11 .39 .05 .07 .26 .10 .32 .24 .22 .18 .09 .30 .17 .28 .14 .17 .31 .12 .19 .21 .19 .23 .13 .40 .10 .11 .45 .15 .12 .15 .18 .41 .09 .22 .17 .22 .35	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$

Table IV Mean dry mass measured in grams after 40 days of treatment with L-glutamate

Test	Treatment											
		Contro	51	Leave	es Spra	ayed	Roots Bathed					
	I-3	H-1	G-8	C-2	B-7	A-5	D-9	E-4	F-6			
	6	9	6	8	7	5						
	8	8		7	6							
	6	9	4	5	7	5						
_	5	8	5	5	6	3						
I	6	7	6	6	7	4						
	6	10	4	9	7	4						
	8	5	6	6	5	5						
	7	7	7	8	5	5	~~					
	4	5 8	4	2	7	4	-*					
	7	8	5	4	6	4						
	8	8	9	9	9	6						
	9	10	9	9	9	9						
	7	7	9	6	6	8						
	6	11	9	6	8	9						
II	8	7	10	7	12	6						
	7	7	9	7	10	8						
	9	12	9	8	9	9						
	7	7	8	6	7	8						
	7	8	8	8	9	7			_			
	9	12	9	8	9	7						
	8	8	4	6	6	4						
	7	7	4 6	9	8	4						
	6	5	6	5	5	4						
	6	4	4	9	8	5						
III	9	9	6	8	7	4			_			
	7	8	5	6	4	5						
	, 7	8	4	7	5	3						
	, 7	9	7	4	8	5						
	8	7	, 7	6	7	6						
	8	, 7	6	8	6	5			_			

Table V. Number of leaves per plant after treatment with L-glutamate

-- Plants that died before the end of the 40 day treatment period.

Test			_						
	c	Control			s Spra	yed	Roots Bathed		
	I-3	H-1	G8	C-2	B-7	A-5	D ~9	E-4	F~6
	2.10	2.60	1.65	2.20	2.25	.65			
	1.90	1.40		1.35	1.35				~
	2.50	1.50	1.34	1.00	2.65	.32			
	1.30	1.00	1.53	.86	2.73	.69			
I	1.11	1.30	1.69	1.30	1.97	.70			
	2.00	1.20	1.47	4.30	2.84	.45			
	3.65	.70	1.90	1.00	2.71	.60			
	1.90	.90	1.55	2.50	1.89	.84			
	3.50	.60	1.30	.57	2.14	.70			
	2.70	1.90	1.25	.78	2.02	.51		~ -	
	1.76	1.33	1.35	2.09	1.19	1.05			
	2.61	2.76	1.87	2.00	1.65	2.55			
	1.30	1.40	1.55	.79	.89	1.34			
	1.56	1.80	2.10	1.40	1.39	1.51			
II	1.99	.68	2.12	1.65	1.46	1.50			
	1.35	1.86	1.93	2.49	1.50	2.49			
	2.00	2.74	1.71	1.79	1.29	2.99			
	1.31	1.57	2.09	1.20	1.32	1.06			
	.99	1.56	1.35	1.85	1.63	2.34			
	2.53	1.82	1.52	1.71	1.22	2.34			
<u></u>									
	1.90	.60	1.31	1.30	1.10	1.60			
	2.10	1.30	1.89	1.80	1.25	1.50			
	1.77	.70	1.67	1.40	1.05	1.74			
~	1.56		1.33	2.20	.93	1.60			
111	1.44	1.30	1.64	1.70	1.00	1.45			
	2.00	2.10	1.54	1.50	1.50	1.69			
	1.53	1.70	1.44	1.90	1.84	1.31			
	2.90	2.60	1.53	• 88	.99	1.65			
	2.60	• 90	1.55	1.30	1.77	1.53			
	1.90	1.50	1.30	1.60	2.00	1.44			

Table VI. Average fresh mass measured in grams after 40 days of treatment with L-glutamate

-- Plants that died before the end of the 40 day treatment period.

SUMMARY

This study was conducted to determine the effects of L-glutamate, mono-potassium salt on the Bloomsdale Long Standing variety of spinach.

The major difference in plant reaction occurred between the plants whose root systems were bathed with the L-glutamate and the control plants. The plants treated with L-glutamate all died after approximately 21 days of treatment. The differences were not significant between the plants that were sprayed with the L-glutamate and the control plants. One treatment was not greater than the other in all of the five measurements taken. The sprayed plants had longer and wider leaves, with a greater dry mass than the control plants. The control plants had more leaves and greater fresh mass.

From the results of this experiment it might be hypothesized that treatment with L-glutamate, mono-potassium salt does not significantly effect the leaf number nor size of spinach plants. However, a solution of L-glutamate, mono-potassium salt does have a detrimental effect on spinach plants when applied to their root systems. It also appears that a 30 mm solution of L-glutamate, mono-potassium salt does have a negative effect on the growth of spinach plants when they are grown under greenhouse conditions.

Many questions are still unanswered in this area of study and much research needs to be done on this topic in the future. LITERATURE CITED

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