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ORIGINAL PAPER

EFFECT OF APPLICATION OF Mg-TYTANIT STIMULATOR ON WINTER WHEAT YIELDING AND QUANTITATIVE PARAMETERS OF WHEAT STRAW AND GRAIN*

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Abstract

The impact of titanium on plants has been tested for 100 years. Despite this long research tradition, there are few studies dedicated to the optimization of doses and application dates of titanium in cultivation of particular crops. A small-plot experiment involving winter wheat was set up to fill the gap. The aim was to determine the impact of doses and application dates of the biostimulant Mg-Tytanit (MgTi) on the aerial phytomass and underground phytomass formation, quantity and quality of winter wheat yield, and the titanium content in wheat grain and straw. The experiment consisted of 5 treatments: 0 – control without MgTi; $2xTi_{0.2}$ – two MgTi applications of a dose of 0.2 dm³ ha⁻¹; $3xTi_{0.2}$ – three MgTi applications of a dose of 0.2 dm³ ha⁻¹; 2xTi_{0.4} - two MgTi applications of a dose of 0.4 dm³ ha⁻¹; 3xTi_{0.4} - three MgTi applications of a dose of 0.4 dm³ ha⁻¹. The Mg-Tytanit was applied in spring during two or three different growth stages: BBCH 29, BBCH 32-37, BBCH 55-59. The MgTi application in the growth phases BBCH 29 and 32-37, regardless the applied doses, increased the yields of aerial phytomass and underground phytomass. All MgTi applications increased straw yield. The use of Mg-Tytanit in the total doses from 0.6 to 0.8 dm³ ha⁻¹ caused a significant increase in wheat grain yield. The MgTi did not have a significant influence on the majority of qualitative parameters of wheat grain. One tonne of grain and an appropriate quantity of straw needed about 26.2 g of titanium uptake.

Keywords: titanium, biostimulant, Mg-Tytanit, winter wheat.

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Biostimulants are substances which do not have the character of a fertilizer, although they stimulate the growth of one or several parts of plants simultaneously (root, stalk, leaf, flower, etc.), the accumulation of nutrients, or the formation of phytomass and accumulation of nutrients at the same time. Titanium is classified as a biostimulant. Titanium is not added to fertilizers for the sake of increasing its content in foodstuff because Ti is not an essential element in human nutrition. However, its positive effects on the growth of both animals and plants have been recorded (YAGHOUBI et al. 2000, ALCARAZ-LÓPEZ et al. 2003). Significantly fewer examples of positive Ti effects on animals are known in comparison with the data about its impact on plants.

In many studies, the application of titanium increased the chlorophyll content in leaves, content of vitamin C, sugar, fat, gluten and nutrients in fruits and plant seeds. It extends the shelf-life of fruits, increases the yield of cultivated crops and decreases the content of nitrates and some heavy metals (PAIS 1983, MATUŠKOVIČ et al. 1995, MALINOWSKA, KALEMBASA 2012, JABERZADEH et al. 2013, and others). On the other hand, some negative effects of Ti use in crop cultivation have been noted (Kužel et al. 2007, BOUGUERRA et al. 2016). Researchers have observed a reduction in the chlorophyll content in leaves and a lower weight of oat aerial phytomass. They have also noted the phytotoxic effect of Ti on tomatoes and lettuce. SKAWIŃSKI et al. (2012) reported that in some cases grain yield and straw yield of winter wheat decreased after an application of a titanium-containing preparation. There are several reasons explaining these results, e.g. differences in application dates and Ti doses, the strength of the solution and compound used, the sensitivity of particular plants to Ti, the way Ti was applied, i.e. separately or with other substances, the place of application, i.e. soil or leaves.

The scientists studying the impact of titanium on plants applied the element in different preparations and in various compositions of water soluble or insoluble compounds. They have found that it is suitable to use titanium chelated by organic acids, in particular ascorbic, citric and malic acids (ALCARAZ-LÓPEZ et al. 2005, KOVÁČIK et al. 2014).

The differences between plants species in their content of and needs for particular macro- and microelements also concern titanium. WALLACE et al. (1977) observed chlorotic and necrotic spots on bean leaves when their Ti content achieved around 200 mg kg⁻¹. The damage in cabbage plants apeared if the Ti content exceeded 4 mg kg⁻¹ in inner leaves and 3.000 mg kg⁻¹ in roots (HARA et al. 1976).

The plant's response to Ti also depends on its nutrition. In the cultivation of potatoes, winter wheat and spring barley under sufficient N nutrition, these crops responded negatively to increased Ti application (30 g ha⁻¹). In cultivation under N deficiency, wheat responded significantly positively to Ti application, potatoes did not respond to it, while barley yield decreased (TLUSTOŠ et al. 2005). KOVÁČIK et al. (2016a) emphasized the necessity to elaborate evaluation criteria for the content of available Ti in soil and total Ti content in plants.

The biostimulant Mg-Tytanit, one of the preparations containing titanium, is used in many countries of Europe and Asia. It is in many cases economically effective, although its optimal application doses and application dates are yet to be determined (DOBROMILSKA 2007).

The aim of the experiment has been to evaluate the impact of the biostimulant Mg-Tytanit applied on the leaves of winter wheat in two doses (0.2 and 0.4 dm³ ha⁻¹), two or three times in a season, on the dynamics of aerial and underground phytomass formation, Ti content in grain and straw, and on the quantity and quality of winter wheat yield. The quantity of Ti taken up by the yield of one tonne of winter wheat grain and an appropriate quantity of its straw was also studied because the information about the Ti uptake by field and garden crops is either missing or scarce.

MATERIAL AND METHODS

Experimental site

The field experiment was conducted during three farming years (2009/2010, 2010/2011, 2011/2012) on Haplic Chernozem in western Slovakia, near the village Bučany (48°42′ N, 17°70′ E). The locality is situated in a maize-growing region, about 165 m a.s.l., in a very warm and very dry climatic region of the temperatures sum (TS) >3,000°C and the total annual precipitation 517 mm (maximal in June – 71.76 mm and minimal in January – 28.04 mm). The warmest month is July (average daily temperature 19.49°C) and the coldest one is January (average daily temperature 1.77°C).

The parameters of soil taken from the layer 0.0-0.3 m (Table 1) were determined by the following methods: $N_{in} = N-NH_4^+ + N-NO_3^-$, $N-NH_4^+ -$ colorimertically with the Nessler's agent, $N-NO_3^- -$ colorimertically with Table 1

		N		Available forms of					G		
Years	Depth (m)	IN _{in}	Р	K	Ca	Mg	Ti		G	U _{ox}	$\mathrm{pH}_{\mathrm{KCl}}$
	(111)		(mg kg ⁻¹)								
2009		18.3	70	235	3,600	317	1.02	2,142	13.8	15.2	7.14
2010	0.0-0.3	27.3	48	250	2,450	390	0.88	1,360	41.9	13.1	6.55
2011		25.2	74	265	6,550	335	1.56	1,617	7.6	14.8	7.08

Soil parameters before experiments (dry matter)

 $\rm N_{in}$ – inorganic nitrogen, S – water soluble sulphur, $\rm N_t$ – total nitrogen, $\rm C_{ox}$ – total oxidizable carbon

phenol-2,4-disulphonic acid; P – colorimertically, K and Ca – flame photometry, and Mg by AAS – all four in Mehlich 3 extracts (MEHLICH 1984); S – spectrometrically ICP-OES in water extract (ZBÍRAL et al. 2011*b*); C_{ox} – oxidometri-

cally (DZIADOWIEC, GONET 1999); $N_t - Kjeldahl method (BREMNER 1960);$ Ti – spectrometrically ICP-MS in Mehlich 3 extract (ZBÍRAL et al. 2011*a*); pH – potentiometrically in 1.0 mol dm⁻³ KCl.

Materials

The Polish liquid biostimulant Mg-Tytanit was used, containing in 1 dm³ 8.5 g of Ti as titanium ascorbate, 40.8 g of Mg and 54.4 g of S as magnesium sulphate (MgSO₄). Mg-Tytanit (MgTi) is dark-brown liquid of the density of 1.36 kg dm³. Every year, winter wheat cultivar Šarlota was sown in the first decade of October, using 530 seeds per m². In spring, during the sampling of plant material, there were 480 plants per 1 m² (a three-year average).

Experimental design and field management

The experiment was conducted as a small-plot trial in a split-plot design with completely randomised blocks in four replications, consisting of 5 treatments (Table 2).

Table 2

Variant			Growth phase	Total dosage of		
		BBCH 29 BBCH 32-37 BBCH 55-59		Mg-Tytanit	Ti	
No.	No. Marking Mg-Tytanit dose (dm ³ ha ⁻¹)					(g ha'l)
1	0	0	0	0	0	0
2	2xTi _{0.2}	0.2	0.2	0	0.4	3.4
3	3xTi _{0.2}	0.2	0.2	0.2	0.6	5.1
4	2xTi _{0.4}	0.4	0.4	0	0.8	6.8
5	3xTi _{0.4}	0.4	0.4	0.4	1.2	10.2

Treatments of experiment

The area of one plot was 20 m^2 . The samples of the aerial and underground winter wheat phytomass were taken in the growth stages BBCH 29, BBCH 32-37, BBCH 55-59, BBCH 65-69 (Table 3). After each plant material sampling, except the last one, MgTi was applied on the same day.

The whole wheat aerial phytomass in the technological ripeness phase was taken manually from an area 1.0 m² of each plot. In the laboratory, the kernels were thrashed from ears, the weight of seeds and other vegetative mass (straw) was determined and the proportion between grain and straw was calculated. The following were determined in winter wheat grain and straw: the content of dry matter, Ti by the inductively coupled plasma atomic emission spectroscopy (ICP-AES) method (YANG et al. 2012) and nitrogen by the Kjeldahl method after mineralization in concentrated H_2SO_4 (COHEN

Growth stages of	sampling of winter	wheat plant and	l spraying Mg-Tytanit
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Treatment							
1 st sampling and subsequent 1 st Ti spray	2 nd sampling and subsequent 2 nd Ti spray	3 rd sampling and subsequent 3 rd Ti spray	4 th sampling				
Growth phase							
End of tillering BBCH 29steam elongationBBCH 32-37		earing BBCH 55-59	flowering BBCH 65-69				

1910). Other parameters of wheat grain (a thousand kernel weight – TKW, share of first-class grain, volume weight, content of gluten, Zeleny index, falling number) were also determined.

Statistical analysis

The data were processed statistically by analysis of variance (ANOVA). The differences between the variants were evaluated subsequently by the LSD test at the significance level $\alpha = 0.05$ in the PC program Statgraphics, version 5.

RESULTS

Aerial and underground phytomass formation

The first two Mg-Tytanit sprayings applied in the growth phases BBCH 29 and 32-37 (measured in the growth phases BBCH 32-37 and BBCH 55) stimulated the formation of winter wheat aerial and underground phytomass (Table 4). The stimulation of both types of phytomass was not identical but comparable.

After the first spraying, the difference in the weight of aerial phytomass compared to the control was 6.52 and 6.77%, and that of roots equalled 11.88 and 16.83%. It is evident that the foliar application of Mg-Tytanit carried out in the growth phase BBCH 29 affected the underground phytomass formation more significantly positively than that of aerial phytomass. After the second spraying, the difference in the weight of aerial phytomass compared to the control was 15.72 and 25.87%, and roots – 19.52 and 30.93%. Although the difference in the weight of roots compared to the control was higher again, we cannot claim that the second spraying affected roots more distinctly. The weight of phytomass sampled in BBCH 55 reflected also the effect of the first spraying. The impact of the second spraying was stronger on aerial phytomass than on roots. This claim is substantiated by the differences in the weight increase between the variants treated with MgTi and the control,

	Growth phase as BBCH											
Variant	29	32-37	55	65-69	32-37	55	65-69	32-37	55	65-69		
		weight o (g of 100	f 1 plant)% DM)		(%)			rise to previous growth phase (%)				
Aerial phytomass												
1	0.256	0.798	2.690	5.645^{a}	100.0	100.0	100.0	311.7	337.1	209.9		
2	0.256	0.852	3.386	6.772^{b}	106.8	125.9	120.0	332.8	397.4	200.0		
3	0.256	0.852	3.386	6.793^{b}	106.8	125.9	120.3	332.8	397.4	200.6		
4	0.256	0.850	3.113	7.405°	106.5	115.7	131.2	332.0	366.2	237.9		
5	0.256	0.850	3.113	6.496^{b}	106.5	115.7	115.1	332.0	366.2	208.7		
$\mathrm{LSD}_{0.05}$				0.374								
			U	ndergrou	und phyt	omass						
1	0.075	0.202	0.333	0.476^{a}	100.0	100.0	100.0	269.3	164.9	142.9		
2	0.075	0.226	0.436	0.628^{b}	111.9	130.9	131.9	301.3	192.9	144.0		
3	0.075	0.226	0.436	0.582^{b}	111.9	130.9	122.3	301.3	192.9	133.5		
4	0.075	0.236	0.398	0.645^{b}	116.8	119.5	135.5	314.7	168.6	162.1		
5	0.075	0.236	0.398	0.589^{b}	116.8	119.5	123.7	314.7	168.6	148.0		
$LSD_{0.05}$				0.104								

Effect of Mg-Tytanit application on dynamics of winter wheat phytomass formation (three-year average)

Key to Tables 4-7: means followed by different letters indicate significant differences between tretments at a level of 95.0%

when the weight increment achieved in the particular variant during the previous growth phase is taken into account. The differences in the aerial phytomass reached 29.15% and 60.33%. The differences in the root phytomass were 3.79% and 28.07%. The third spraying with MgTi in both of the applied doses (0.2 and 0.4 dm³ ha⁻¹) negatively but insignificantly affected the underground phytomass (comparisons of variants 2 and 3 and variants 4 and 5 in the growth phase BBCH 65-69). The dose 0.2 dm³ ha⁻¹ also insignificantly affected the aerial phytomass, but the dose 0.4 dm³ ha⁻¹ significantly depressed it.

The aerial to underground phytomass weight ratio (growth phase 65-69) was the highest in the control variant, followed by variants 1 to 5, where it equalled 11.86, 10.78, 8.28, 11.48, 11.03, respectively. This proves that the underground phytomass grew faster than roots as a result of MgTi application.

On average, most aerial and underground phytomass during the three experimental years was formed until the growth phase BBCH 65-69 in variant 4, where a total $0.8 \text{ dm}^3 \text{ ha}^{-1}$ MgTi was applied in a two sprayings of 0.4 dm³ ha⁻¹ (Table 4). The difference in the formed aerial phytomass was

31.18% and that in root phytomass equalled 35.5% in comparison with the control. The least phytomass was formed in the control variant.

The other applied MgTi doses (0.2 and 0.4 dm³ ha⁻¹) did not equally affect the both phytomasses. After the first spraying, the weight of aerial phytomasses from the variants treated with either of the two doses was almost identical, while the weight of roots was higher after the application of the higher dose. Following the second spraying, both aerial and underground phytomass was higher in the variants where 0.2 dm³ ha⁻¹ MgTi was applied than after the application of the higher dose. The third spraying did not affect positively the phytomass growth; on the contrary, the higher MgTi dose had a more negative impact than tha lower one.

Grain yield, the main product of winter wheat cultivation, was increased in all the variants where MgTi was used (Table 5), although statistically significant increases were determined only in variants 3 and 4, where 0.6 and 0.8 dm³ ha⁻¹ Mg-Tytanit was applied in total, thrice the dose 0.2 dm³ ha⁻¹ and twice the dose 0.4 dm³ ha⁻¹, respectively. The yield increased by 5.3% in variant 3, and by 5.7% in the variant 4. It is evident that total MgTi doses should not be lower than 0.6 dm³ ha⁻¹ and higher than 0.8 dm³ ha⁻¹ in order to increase grain yield.

The biostimulant MgTi increased significantly the straw yield in all the variants compared to the control (Table 5), but the differences between the variants were not significant. The highest straw yield was achieved in variant 4. The relative growth of straw yield (a three-year average) was higher than that of grain. These results and changes in the proportions between straw and grain weight show that MgTi affected the yield of grain more than that of straw. The highest grain and straw yield was achieved in variant 4, where Mg-Tytanit was applied twice in 0.4 dm³ ha⁻¹ (total 0.8 dm³ ha⁻¹).

Nitrogen and titanium content in grain and straw

Application of Mg-Tytanit significantly increased the nitrogen content in grain, while decreasing it insignificantly in straw relative to the control (Table 5), but the differences between the variants treated with MgTi were not significant.

The titanium content in grain was the lowest in the control variant. The application of MgTi tended to increase the Ti content in grain, but a significant increase in grain was only observed in variant 3 with the highest Ti content. Differences in the Ti content in grain between the variants treated with MgTi were not significant.

The titanium content in straw, unlike its content in grain, was the highest in the control variant. As the amount of applied MgTi increased, the titanium contents in straw decreased. Straw in variant 5, where the highest total MgTi dose was applied, contained the least Ti.

During the three years, the titanium content in wheat grain varied from 0.27 to 0.43 mg kg⁻¹ whereas in straw it ranged from 20.42 to 24.93 mg kg⁻¹ on average.

	Gra	Grain		Straw		Grain		Straw	
Variant	(Ma hail)	(%)	(Mg ha ⁻¹)	(%)	/ grain ratio	N	Ti	N	Ti
	(Mg na ')					(mg kg ⁻¹)			
1	6.20^{a}	100.00	6.98^{a}	100.00	1.13	$23,775^{a}$	0.27^{a}	7,385	24.93°
2	6.34^{a}	102.26	7.43^{b}	106.45	1.17	$25,353^{b}$	0.33^{ab}	7,197	23.97^{bc}
3	6.53^{b}	105.32	7.35^{b}	105.30	1.13	$25,086^{b}$	0.43^{b}	7,230	22.04^{ab}
4	6.55^{b}	105.65	7.58^{b}	108.60	1.16	$25,359^{b}$	0.38^{ab}	7,232	20.69^{a}
5	6.32^{a}	101.94	7.50^{b}	107.45	1.19	$25,806^{b}$	0.34^{ab}	7,332	20.42^{a}
LSD	0.184		0.257			986.578	0.129		2.150

Effect of Mg-Tytanit application on grain and straw winter wheat yield and N and Ti content in grain (three-year average)

Wheat yield parameters

 $\mathbf{5}$

 $\mathrm{LSD}_{0.05}$

The parameters such as grain volume weight, TKW, gluten content, Zeleny index and falling number were not influenced considerably by the biostimulant (Table 6). An exception was the significant decrease in weight

Table 6

 271.64^{b}

19.375

Effect of Mg-1 ytanit application on parameters of winter wheat grain (three-year average)								
Variant	VW	TKW	Grain of the 1 st class Gluten		Zeleny index	Falling number		
	(g dm ^{.3})	(g)	(g)	(%)	(cm ³)	(s)		
1	795.73	41.90	81.03^{b}	25.24	31.46	274.42^{b}		
2	791.27	41.75	78.54^{a}	26.09	30.63	268.17^{b}		
3	795.83	41.49	80.43^{ab}	25.99	30.84	267.42^{b}		
4	797.13	41.72	79.09^{a}	25.82	32.18	261.11^{a}		

VW - volume weight, TKW - thousand kernel weight

41.43

794.00

of first class grains in variants 2 and 4, where two sprayings with MgTi were carried out. The third spraying used in variants 3 and 5 had a slightly positive impact on this parameter.

 79.35^{ab}

1.932

26.18

31.81

Titanium uptake by grain and straw yield

The lowest titanium uptake by grain was in the variant where the lowest grain yield and the lowest Ti content in grain were noted (Tables 5 and 7). The highest uptake was demonstrated for grain from variants 3 and 4, where the highest grain yield and the highest Ti content were observed. Titanium uptake by grain reached 2.25 g ha⁻¹ and by straw -164.8 g ha⁻¹ on average. The Ti uptake by straw was on average 73.2-fold higher than by grain.

Variant	Grain	Straw	Total	Straw/ grain ratio	Uptake by the yield of 1 Mg of grains and appropriate amount of straw
		(g ha ⁻¹)			(g Mg ^{.1})
1	1.69^{a}	174.01°	175.70°	103.0	28.34
2	2.11^{ab}	178.10°	180.21 ^c	84.4	28.42
3	2.83^{c}	162.00^{b}	164.83^{b}	57.2	25.24
4	2.47^{bc}	156.83^{a}	159.30^{a}	63.5	24.32
5	2.13^{ab}	153.15^{a}	155.28^{a}	71.9	24.57
LSD _{0.05}	0.486	4.145	5.373		

Effect of Mg-Tytanit application on titanium uptake by winter wheat (three-year average)

As the applied MgTi dose increased, the titanium uptake decreased (variants 2 to 4).

The titanium uptake with one tonne of grain yield and an appropriate mass of straw varied from 24.32 to 28.34 g. On average, all variants required 26.2 g of titanium, which corresponds to 155.28 to 180.21 g ha⁻¹. The Ti uptake by grain and straw at the average grain yield achieved (6.39 Mg ha⁻¹) was 167.06 g.

DISCUSSION

The wheat response to the application of Mg-Tytanit biostimulant confirmed that the same substance used in the same doses and on the same application dates in crop cultivation often results in an unequal growth of phytomass. The first two MgTi sprays stimulated the formation of aerial and underground phytomass (Table 4). The third MgTi spray was not substantiated. These findings do not correspond with the data recorded by KovAčIK et al. (2016b), where there was an increase in both aerial and underground phytomass after the third spray of the same MgTi doses in the oilseed rape cultivation.

The increase of the formation of both phytomass types after the first and second MgTi application was relatively high (Table 4), from 15.1 to 31.2% (aerial phytomass) and from 22.3 to 35.5% (roots). However, these values are lower than recorded by LOPEZ-MORENO et al. (1995) in experiments with pepper.

Considerable differences in the formation of phytomass observed up to the growth phase BBCH 56 were not reflected equally in the differences in grain and straw yields (Table 5). The differences in yields were significantly lower and did not exceed 5.7% (grain) and 8.6% (straw) on average for the three years. These values fully correspond with the results of KOVAČIK (2014), who claims that the crop yield in Slovakia is increased on average by 6% after foliar application of substances whose concentrations in plants are at the level of microelements. Application of preparations containing Ti caused increase of fruit yields by more than 6% in some cases, or raised seed yields of the cultivated crops. Mg-Tytanit has not been tested on the mentioned crops, and wheat was not among the plants it had been applied experimentally (PRUSINSKI, KASZKOWIAK 2005). Despite the data suggesting significant increase of crop yields after Ti application, it is still claimed that Ti-containing substances can typically increase crop yields in Slovakia by 3.5 to 8.7% (TICHÝ 1990).

Farmers often rely on the information about the formed phytomass, the height of plants, thickness of stalks, etc., when they assess quantities of future yields. Our findings showing the dependence between the created aerial phytomass in the growth phase BBCH 65-69 and the wheat grain yield prove the validity of those assessments.

The finding that Mg-Tytanit determined the yield of wheat straw more positively than the grain is not a general characteristic of this preparation, because in an earlier experiment (Kováčik et al. 2016b) MgTi more positively affected winter oilseed rape seed yield than its straw. The insignificant impact of MgTi on the mechanical and qualitative parameters of wheat grain (volume weight, thousand kernel weight, gluten content, Zeleny index and falling number) is not a general characteristic of this preparation either (Table 6). On the contrary, its use in experiments improved the quality of cultivated crops (SKUPIEŃ, OSZMIAŃSKI 2007).

The tendency towards increased Ti and N content in grain after MgTi application corresponds with the results of MALINOWSKA and KALEMBASA (2012) and ALCARAZ-LOPEZ et al. (2005), who claimed that Ti application increased the potassium and sodium content in plants.

The Ti content in wheat grain and straw was comparable with its content detected in other plants, although it was not equal (Table 5). Wheat grain contains 2- to 10-fold less titanium and straw has 2- to 3-fold more of this element than the same organs of winter oilseed rape in the experiments conducted by Kováčik et al. (2016b).

Rationalization of plant fertilization means respecting the plants' demand for nutrients and nutrient pools in soil. In good agricultural practice, methods used for the calculation of nutrient doses should respect the crop demand for nutrients to create a unit of the main yield and an appropriate quantity of byproduct. In order to elaborate the methodology of plant nutrition with Ti based on the above principle, it is necessary to acquire data about plant requirements for titanium. In this context, the finding is one tonne of grain and an appropriate quantity of straw requires an uptake of 26.2 g of titanium by the plant. The Ti content in grain is over 70-fold less than in straw. Thus, when crop residues (straw) are ploughed in after harvest, it is not advisable to forgo the monitoring of available titanium in soil. And reversely, if straw is removed from a field, a farmer should supplement the supply of titanium through fertilization.

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