DEVELOPMENT OF A NOVEL FOLIAR FERTILIZER WITH ZINC

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ABSTRACT

Zinc deficiency, resulted from various stress factors, limits agricultural production worldwide. Therefore, research into developing sustainable methods to alleviate this type of deficiency should be a priority. This study presents the development of a new foliar fertlizer, focusing on its formulation, characterization, and testing. The fertilizer formula associates a classic NPK and micronutrients (Cu, Zn, Fe, Mn) matrix with organic substances having bio-stimulating effect: humic acids and algae extracts. The composition of the experimental fertilizer (8.2% N, 3.9% P, 3.6% K, 0.8% Zn) is formulated to prevent and correct imbalances in plant mineral nutrition, especially those caused by zinc deficiency. A set of field trials were conducted in order to assess the effect of the fertilizer on grapevine (cv Chasslas dore) and maize (DKC 4590). Experimental data indicated that application of the foliar fertilizer had signifficant contribution to the improvment of yield, leaf nutrient and chlorophyll content, and crop quality parameters.

INTRODUCTION

Zinc (Zn) is one of the 8 essential micro-nutrients involved in the normal development of plants. When the amount of available zinc is inadequate, metabolic dysfunctions of the enzyme systems in which it participates appear. Zinc is recognized as a constituent of over 70 metallo-enzymes, is involved in protein metabolism, in ribonucleic acid and carbohydrate synthesis, and participate in the synthesis of growth phytohormones – low content of auxins, and thus limited growth processes, is a specific symptom of zinc deficiency [8].

Zinc deficiency has been reported after War Word II on a wide range of soils throughout the world. In Romania, the phenomenon was observed in 1966 at corn cultivated on alkaline soils. Subsequently, the phenomenon spread to other soils, being favored by high doses of mineral fertilizers, especially phosphorus. In present, zinc deficiency occurs on an area of approximately 1.5 million ha in the east and south-east of the country [10].

The factors favoring zinc deficiency are numerous, their action manifesing most often in connection. The soil has the primordial role, especially by means of the properties that affect the mobility of zinc. The effect of soil reaction on zinc mobility is due to the difference in solubility of pH dependent zinc forms. Lindsay (1979) [13] found that by increasing the pH with a unit the zinc solubility drops 100 times. The pH range at which zinc deficiency was frequently reported (pH>7.4) corresponds to the minimum solubility of most zinc compounds in the soil. An important factor involved in zinc deficiency is the high level of phosphorus in soil, zinc nutrition disorder being most often determined by high phosphorus (P) values. Mechanisms through which phosphorus interferes with reducing zinc accessibility for plants are not fully understood, although the subject is often addressed in the literature. Currently, it is widely accepted that P interferes with the absorption, translocation and use of Zn in plants [14]. Susceptibility of plants to zinc deficiency is very different, with distinctions between species, as well as within the same species - between varieties and hybrids. Maize, some bean varieties, soybean, flax, hops, vines are particularly sensitive to zinc deficiency. Many fruit trees such as apples, peaches and pears are also sensitive to zinc deficiency [12,18].

Zinc deficiency is corrected by soil fertilization, foliar spraying or seed treatment. Introduction of new technologies for a competitive agriculture and diversification of foliar fertilizers has led to an increase of the market share of this type of fertilizers up to 30-35%. The use of foliar fertilizers is in line with the current European Community political agenda: to reduce the use of conventional fertilizers and, implicitly, of soil and groundwater pollution.

Worldwide, there is an increasing trend for the production of fertilizers with natural biostimulators such as protein hydrolysates, algae extracts, as well as fulvic or humic acids. Applied to plants at low concentrations, sometimes even in the order of mg/kg, these substances favor the absorption of nutrients, as well as their protection under climate and/or technologic stress, and lead to the development of the root system, flowers and fruits [7].

MATERIAL AND METHOD

A. Experimental fertilizer

The process for obtaining the experimental fertilizer (at laboratory scale) consists of the following steps:

- phosphoric acid 85% w/w is neutralized with potassium carbonate under continuous stirring and at a constant temperature of 28-30°C. Urea - representing the amide nitrogen source, then ammonium nitrate – as the source of ammoniacal and nitric nitrogen, are gradually added, keeping the temperature at 28-30°C. Thus results the macronutrient solution (A), which is cooled to a temperature of 20-25°C;
- meso- and micronutrients (Cu-EDTA, Zn-EDTA, Fe-EDTA, Mn-EDTA, $Mg(SO_4)\cdot 7H_2O$, $Na_2B_4O_7\cdot 10H_2O$) are disolved in water at a temperature of 20-25°C, thus resulting solution (B);
- the macronutrient solution (A) and the meso- and micronutient solution (B) are mixed under continuous stirring at 20-25°C;
- the potassium humate and the algae extract are gradulally added, under continuous stirrig for 1-1,5 hours, over the macro-, meso- and micronutrient solution.
- demineralized water is added up to a final volume of 1000 ml, thus obtaining the experimental fertilizer – **Fertil Zinc Plus**.

B. Field trials

In order to study the effect of Fertil Zinc Plus on grapevine and maize two experiments in factorial format based on randomized complete block design were conducted at:

- experimental field from the V. Adamachi horticultural farm of the Faculty of Horticulture - University of Agronomic Sciences and Veterinary Medicine of Iaşi - for the tests on grapevine (cv *Chasselas dore)*;
- Ezereni experimental field of the Faculty of Agronomy – University of Agronomic Sciences and Veterinary Medicine of Iaşi - in the case of tests on maize crop *(DKC 4590)*.

In the agricultural year 2016-2017, the sum of precipitation was 504.9 mm, which was lower compared with the mean annual precipitation (517.8 mm) and the mean temperature was 10.1^{o}C, higher than the normal mean recorded in the region (9.6 ^{o}C).

The trial locations for grapevine and maize were on soils classified as hortic anthrosol and cambic chernozem respectively.

The main physical and chemical properties of the studied soils are presented in Tables 1 and 2. As shown in the tables, the two soils have similar characteristics with favorable values for the growth and development of crops under field conditions.

Table 1

Physicochemical properties of soil samples from V. Adamachi horticultural farm

Characteristic	Genetic horizons / Depth (cm)			
	Amp (0-20 cm)	**Am (20-40 cm)**	**AB (40-60 cm)**	**Bv (60-100 cm)**
Clay (%)	38.1	39.8	40.0	45.6
pH (H_2O)	6.21	6.68	7.05	7.24
Organic matter (%)	3.181	1.311	1.106	0.351
N_t (%)	0.151	0.140	0.071	0.021
Available P (mg/kg)	44	40	40	41
Available K (mg/kg)	220	210	145	156

Table 2

Physicochemical properties of soil samples from Ezereni field farm

Characteristic	Genetic horizons / Depth (cm)			
	Amp (0-20 cm)	**Am (20-40 cm)**	**AB (40-60 cm)**	**Bv (60-100 cm)**
Clay (%)	36.1	38.8	39.9	43.8
pH (H_2O)	6.24	6.31	7.15	7.31
Organic matter (%)	3.281	2.312	1.015	0.084
N_t (%)	0.135	0.142	0.131	0.071
Available P (mg/kg)	34	35	40	42
Available K (mg/kg)	222	211	178	142

The experiments were carried out on a soil without irrigation and fertilization ($N_0P_0K_0$). Three foliar treatments were performed during the vegetative season. Dilute sprays (0.5% solution) of the fertilizer (dose of 2.5 l/ha) were applied using a manual pump. The spraying of the fertilizing solution was made in the morning, in a wind-free atmosphere, at below 25^{o}C.

In order to assess the influence of the experimental fertilizer on the photosynthesis indicators and plant mineral nutrition, mature fresh leaves were harvested one week after application of the last foliar treatment, at fruit developing stage.

The extraction of chlorophyll pigments was performed in the laboratory according to the method described by Lancaster et al (1994) [11] with 85% acetone and their concentration was determined by spectrophotometric method at different wavelengths: 645 nm for chlorophyll a and 663 nm respectively for chlorophyll b.

The extraction of carotenoid pigments was performed according to the method described by Bunea et al (2008) [4] and their concentration was determined by spectrophotometric method (450 nm).

C. Statistical analysis

The collected data were subject to statistical analysis using a randomised factorial design. Each experiment was designed with three replicates. The mean values were compared using the least significant difference (LSD) test.

RESULTS AND DISCUSSIONS

Composition of the experimental fertilizer

The chemical composition and physical characteristics of the experimental fertilizer are shown in Table 3. The experimental fertilizer – Fertil Zinc Plus – associate the classic matrix of NPK type and micronutrients (Cu, Zn, Fe, Mn) with organic substances with bio-stimulating effect: humic acids and algae extracts.

Table 3

Chemical composition and physical characteristics of Fertil Zinc Plus

Component	Value
Total nitrogen (N, %)	8.22
Phosphorus (P_2O_5, %)	3.85
Potassium (K_2O, %)	3.59
Sulfur (S, %)	1.33
Copper (Cu, %)	0.023
Zinc (Zn, %)	0.814
Iron (Fe, %)	0.039
Manganese (Mn, %)	0.132
Magnesium (MgO, %)	0.052
Boron (B, %)	0.015
Organic matter (%)	13.71
pH	7.15
Density (g/cm^3)	1.23

Effects of the experimental fertilizer on grapevine

The experimental data from table 4 shows that foliar fertilization has led to a significant increase in grape production (11239 kg/ha) as compared to unfertilized control (9179 kg/ha). The foliar treatments helped the plants to increase zinc acquisition, which may induce the indol acetic acid synthesis, a plant growth regulator responsible for pollen and berry formation. Also, the yield increase can be partially explained by the bio-stimulating effect of the humic acids and algae extracts present in the composition of the experimental fertilizer.

Table 4

Effect of Fertil Zinc Plus on grapevine (cv *Chasselas dore*) yield

Variants	Yield (kg/ha)	Productive efficiency		
		Dif. kg/ha	Dif. %	Sign.
Control	9179	-	100	-
Fertil Zinc Plus	11239	2060	122.44	***
LSD 5% - 865 kg/ha LSD 1% - 1213 kg/ha LSD 0.1% - 1759 kg/ha				

The sugar content in grape fruits had significantly increased with application of the experimental fertilizer Fertil Zinc Plus as shown in table 5. The increase in sugar content is possible as a result of the increase in the enzymatic activities of fructose-1 and 6- bis phosphatase. Also, the aldolase enzymes will increase along with sufficient availability of zinc, contributing to an accelerated sugar synthesis [5].

Table 5

Effect of Fertil Zinc Plus on the sugar content of grapes (cv *Chasselas dore*)

Variants	**Yield (g/L)**	**Productive efficiency**		
		Dif. g/L	**Dif. %**	**Sign.**
Control	145.00	-	100	-
Fertil Zinc Plus	157.73	9.73	106.71	***
LSD 5% - 2.33 g/L LSD 1% - 4.06 g/L LSD 0.1% - 5.15 g/L				

Improvement of yield and quality parameters of the grapevine by application of foliar fertilizers with zinc was reported by other authors: [6, 9, 15].

Applying the three foliar treatments with Fertil Zinc Plus in critical periods for plant nutrition had a positive effect on the nutrient concentration in leaves. The data in Table 6 shows that the experimental fertilizer has led to a significant increase in the nitrogen, phosphorus and potassium content of the grapevine leaves as compared to the control.

The nitrogen content of grapevine leaves treated with the foliar fertilizer (2.79%) is comparable to the results obtained in similar studies: 1.93 - 3.10% [1, 2, 16]. Although potassium content increases as a result of three foliar treatments (0.44%), it is lower than the values reported by other authors that ranged between 0.56 and 1.35% [1].

Table 6

Effect of Fertil Zinc Plus on grapevine (cv *Chasselas dore*) leaf macronutrient content

Nutrient/Variant	**Control**	**Fertil Zinc Plus**
Nitrogen (N, %)	2.15	2.79
Difference (%)	-	0.64
Phosphorus (P, %)	0.27	0.34
Difference (%)	-	0.07
Potassium (K, %)	0.34	0.44
Difference (%)	-	0.10
Nitrogen LSD 5% - 0.243% LSD 1% - 0.368% LSD 0.1% - 0.521%	Phosphorus LSD 5% - 0.032% LSD 1% - 0.044% LSD 0.1% - 0.064%	Potassium LSD 5% - 0.043% LSD 1% - 0.066% LSD 0.1% - 0.084%

The use of the experimental foliar fertilizer on grapevine stimulated the photosynthetic assimilation process leading to significant increases for each assimilatory pigment (Table 7).

It is well-accepted that iron is an essential element for physiology of plants and its deficiency causes a significant decrease of the photosynthesis activity. Algae extracts increase the chlorophyll content in leaves. The glycine betaine in the algae extracts delays the loss of photosynthetic activity by inhibiting chlorophyll degradation [17].

The fertilizer developed in this present study contains, besides zinc, other elements such as iron and algae extracts. The latter act synergistically and lead to the increase of fotosynthesis activity in plant leaves. This results are in accordance with the findings of Blunden et al (1997) [3].

Table 7

Effect of Fertil Zinc Plus on grapevine (cv *Chasselas dore*) leaf pigments content

Pigment/Variant	Control	Fertil Zinc Plus
Chlorophyll a (mg/g)	1.48	1.88
Difference (%)	-	0.40
Chlorophyll b (mg/g)	0.93	1.18
Difference (%)	-	0.25
Carotene (mg/g)	0.70	0.90
Difference (%)	-	0.20
Chlorophyll a LSD 5% - 0.173 mg/g LSD 1% - 0.249 mg/g LSD 0.1% - 0.332 mg/g	Chlorophyll b LSD 5% - 0.104 mg/g LSD 1% - 0.163 mg/g LSD 0.1% - 0.202 mg/g	Carotene LSD 5% - 0.092 mg/g LSD 1% - 0.136 mg/g LSD 0.1% - 0.179 mg/g

Effects of the experimental fertilizer on maize

Applying the three foliar treatments with Fertil Zinc Plus on maize plants had a positive effect on yield . Data in table 8 show a significant yield increase (1516 kg/ha) as compared to the unfertilized control.

Table 8

Effect of Fertil Zinc Plus on maize (DKC 4590) yield

Variants	Yield (kg/ha)	Productive efficiency		
		Dif. kg/ha	Dif. %	Sign.
Control	5148	-	100	-
Fertil Zinc Plus	6664	1516	129.45	***
LSD 5% - 721 kg/ha LSD 1% - 989 kg/ha LSD 0.1% - 1427 kg/ha				

The experimental data from table 9 show that the macronutrients content in maize leaves had significantly increased with application of the foliar fertilizer Fertil Zinc Plus.

Table 9

Effect of Fertil Zinc Plus on maize (DKC 4590) leaf macronutrient content

Nutrient/Variant	Control	Fertil Zinc Plus
Nitrogen (N, %)	0.88	1.20
Difference (%)	-	0.32
Phosphorus (P, %)	0.24	0.32
Difference (%)	-	0,08
Potassium (K, %)	0.56	0.78
Difference (%)	-	0.22
Nitrogen LSD 5% - 0.142% LSD 1% - 0.193% LSD 0.1% - 0.288%	Phosphorus LSD 5% - 0.040% LSD 1% - 0.059% LSD 0.1% - 0.076%	Potassium LSD 5% - 0.101% LSD 1% - 0.147% LSD 0.1% - 0.203%

As in the case of grapevine, the use of the experimental foliar fertilizer on maize plants stimulated the photosynthetic assimilation process leading to significant increases for all three assimilatory pigments (Table 10).

Table 10

Effect of Fertil Zinc Plus on maize (DKC 4590) leaf pigments content

Pigment/Variant	Control	Fertil Zinc Plus
Chlorophyll a (mg/g)	1.33	1.80
Difference (%)	-	0.47
Chlorophyll b (mg/g)	0.89	1.21
Difference (%)	-	0.32
Carotene (mg/g)	0.66	0.89
Difference (%)	-	0.23
Chlorophyll a LSD 5% - 0.211 mg/g LSD 1% - 0.349 mg/g LSD 0.1% - 0.435 mg/g	Chlorophyll b LSD 5% - 0.143 mg/g LSD 1% - 0.189 mg/g LSD 0.1% - 0.302 mg/g	Carotene LSD 5% - 0.101 mg/g LSD 1% - 0.154 mg/g LSD 0.1% - 0.204 mg/g

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CONCLUSIONS

The fertilizer developed in this study contains, besides zinc, macronutrients, micronutrients, and organic substances. They all act synergistically and lead to the stimultation of plant physiological processes, e.g. increase of fotosynthesis activity in leaves. Overall investigations revealed that experimental fertilizer **Fertil Zinc Plus** had significant contribution to the improvement of yield, leaf nutrient and pigment content in the tested crops, i.e. grapevine (cv Chasselas dore) and maize (DCK 4590 hybrid). Therefore, this product can be used to treat zinc deficiencies but also to optimize the mineral nutrition of plants, especially then when soil available nutrients present a low level.

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