

Biofortification of Romanian and Egyptian Wheat with Zinc and Its Effect on Yield

Alina Laura Agapie¹, Mohamed Mostafa El-Fouly³, Marinel Nicolae Horablaga¹,
Ciprian Buzna^{1*}, Samir Hosni Shaaban³, Rozalia Kadar², Ionut Racz²,
Adel Badr El-Nasharty³, Nicolae Tritean²

¹Agricultural Research and Development Station Lovrin, Timiș County, Romania

²Agricultural Research and Development Station Turda, Cluj County, Romania

³Department of Fertilization Technology, National Research Centre, 33 El-Bohouth St. (former El-Tahrir St.)
Dokki, Giza, 12622, Egypt

*Corresponding author. E-mail: buznac@yahoo.com

ABSTRACT

The study was carried out during the winter season of 2019/2020, 2020/2021 and 2021/2022 under irrigated farming conditions in Oraby Village Mariut sector, Alexandria, Egypt, where the common wheat varieties are grown, namely Giza 168 and Giza 171. These varieties spraying with Zn sulfate at a concentration of 5 g/L of water during tillering or milk stage and (tillering + milk stage).

The results of soil test indicated that the soil has a lime texture, as the total CaCO₃% exceeded 25%, and has a high alkaline reaction, and salt and Na level is also high. In addition, this soil is low in its content of O.M% and very low of its content of Zn. Spraying zinc sulphate with 5 g/L of water showed that there is a positive effect on growth parameters in cultivars Giza 171 and 168 especially the two sprays in the tillering phase and the milky phase followed by the tillering phase compared to the control. The improvement in growth parameters reflected on the yield and its components, as the biological yield and harvest index increased.

In Romania, the experiment took place in two representative areas for the country's agriculture - Agricultural Research and Development Station Lovrin (ARDS Lovrin), in the west of Romania and Agricultural Research and Development Station Turda (ARDS Turda), in the center of the country. The results obtained following the agronomic biofortification with zinc of three autumn wheat genotypes and one spring wheat genotype, carried out in different phenophases, highlight the greatest success in spring wheat, under the conditions of foliar application of zinc sulfate in the growth phase twinning of the crop, treatment preceded of seed treatment. The foliar treatment was carried out with a 5% zinc sulfate solution, the foliar treatment with a 0.5% zinc sulfate solution, and the ground treatment with 30 kg/ha zinc sulfate. The production obtained recommends foliar treatment with zinc, causing significant and distinctly significant increases at Ciprian wheat variety. In the Pădureni spring wheat variety, both variants of seed treatment with zinc led to significant increases of production.

Keywords: zinc fertilizer, growth parameters, wheat yield.

INTRODUCTION

Zinc is well known to have an important role either as a metal component of enzymes or as a functional, structural or regulatory co-factor of a great number of enzymes (Hotz and Brown, 2004). Zinc is an indispensable nutrient for biological systems in plants, humans, and animals (Broadley et al., 2007). Considering the food demand by the increasing global population and widespread occurrence of Zn malnutrition problem, increasing grain Zn concentration in wheat with high yield capacity would be

more meaningful (Prasad et al., 2013; Chen et al., 2017; Bhatt et al., 2020). Wheat (*Triticum aestivum* L.) is an important crop in Egypt and over the world used in human food and animal feed as well as, with regard to cultivated area and total production, as well as nutritive value. It provides 37% of the total calories for the people and 40% of the protein in the Egyptian diet (El-Habbasha et al., 2015). Wheat is genetically low in Zn concentration, with reduced bioavailability (Graham et al., 2001; Cakmak, 2002).

There are several ways for increasing wheat production; one of them is the

appropriate application of zinc, especially with balance and recommendations for other nutrients. However, the concentrations of Zn depend on the size of wheat grains (Velu et al., 2011) and number of grains per spike (Nowack et al., 2008). Haslett et al. (2001) and Timsina (2014) demonstrated the role of phloem transport of Zn in wheat plants by performing stem girdling, and they showed that ⁶⁵Zn supplied on upper leaf was transported to lower leaves and root tip. Ranjbar and Bahrmaniar (2007), showed that using zinc caused increasing effects on grain yield, total dry matter, yield, 1000 grain weight, number of tiller, grain zinc content, flag leaf zinc content, plant height, number of node, protein content. Application of zinc fertilizer not only improves nutritional quality but also contributes significantly to grain production in Zn deficient soils (Peck et al., 2008). Zinc content in Egyptian wheat can be increased through zinc treatments. The expansion of zinc use in Romania will also lead to an increase in the zinc content of Romanian wheat exported to Egypt. Increasing Zn content leads to increasing the quality of wheat. This study aimed to investigate and explore practical possibilities to increase Zn content in grains of different Egyptian and Romanian wheat varieties exported to Egypt.

MATERIAL AND METHODS

Experiment I Egypt

Two wheat varieties were tested in field in Egypt. These varieties were treated with Zn in the field during two phases tillering/stem extension (1) and milk stage (2). It was tested, which of the wheat varieties are most receptive to increasing the Zn content of the yield through bio-fortification.

Treatments were as follows for each variety:

1. Without Zn
2. With Zn (stage 1)
3. With Zn (stage 2)
4. With Zn (stage 1 and 2)

The zinc fertilizer used was zinc sulphate - 27%.

The study was carried out during the season of 2019/2020, 2020/2021 and 2021/2022 to study effect of Zn sulfate (as a source of Zn) on yield and its components of the wheat varieties in the farmer's field under irrigated farming conditions in Oraby Village Mariut sector, Alexandria, Egypt (located between latitude 30°58'47"N and longitude 29°48'38"E).

Soil preparation and cultivation: soil was ploughed using a chisel plough, leveled by wooden leveler and divided into experimental units with three replicates.

Experiments design: the experimental design was randomized complete block design (RCBD) with three replicates. Plot area was 10.5 m² (3.5 m long and 3 m wide).

Soil analysis: before planting, a representative soil sample was taken before sowing to test physical and chemical properties. Soil was analyzed for: texture - pH - EC (Bouyoucos, 1954; Jackson, 1986), CaCO₃ - OM (Walkley and Black, 1934; Alison and Moodle, 1965).

Data record

Yield and yield components

Yield was determined at maturity, yield components were also reported (grain yield, straw yield, harvest index, etc). At harvest, biological yield and grain yield were determined by cutting plants from 1 m² of each plot, and bundled, then weighted, yield was calculated as kilograms/plot, and recorded in terms of ton/ha while Harvest Index (HI) was calculated as follows:

$$HI = \frac{\text{grain yield}}{\text{biological yield at maturity}} \times 100$$

Also, plant height and number of spikes/m² were recorded. At the same time, ten random spikes from each plot were sampled and the following traits were measured; One-Hundred Kernel Weight (g), number of spikelets per spike and spike length (cm).

The grains of the selected varieties were tested for their total Zn content. Zn content was calculated as mg/weight as well as mg/number of grains.

It was also tested, which of the wheat varieties are most receptive to increasing the Zn content of the yield through bio-fortification.

Statistical Analysis

The obtained data were statistically analyzed using COSTAT program and L.S.D. value at the probability levels of 5% was calculated according to Gomez and Gomez (1984).

Experiment II Romania

The experimental device established at ARDS Lovrin and ARDS Turda, includes the following factors:

Factor A:

- a1 - untreated seed;
- a2 - seed treated with zinc sulphate - 0.5% solution.

Factor B:

- b1 - Ciprian variety;
- b2 - the Glosa variety;
- b3 - variety Andrada;
- b4 - Pădureni variety.

Factor C:

- c1 - untreated control;
- c2 - ground treatment with zinc sulphate 30 kg/ha;
- c3 - foliar treatment at the end of twinning;
- c4 - foliar treatment in the bellows phase;

c5 - foliar treatment in the milk-wax phase.

The chemical fertilizer used was zinc sulfate, with an active substance content of Zn of 25.4%. Ground treatment, 30 kg s.a. Zn/ha was applied before sowing. For seed treatment and foliar administration, the concentration of the prepared solution was 0.3%.

RESULTS AND DISCUSSION

Experiment I Egypt

Results of the first season (2019/2020)

Soil testing and irrigation water

The results of the soil tests indicated that the soil has a lime texture, as the percentage of total calcium carbonate is high and exceeded 25%, and the soil has a high alkaline reaction, and the degree of salt and sodium level is also high. In addition, this soil is low in its content of organic matter and its content of nitrogen and medium content of phosphorous and potassium, perhaps due to the continued fertilization of these two elements. The soil is also low in its content of zinc (Table 1). It is known that under the influence of high soil pH, as well as salinity and calcium carbonate, most of the nutrients are in an unavailable form for the plant, especially if soil is low in organic matter.

Table 1. Soil test of Mariout before sowing (0-30cm depth)

Character		Nutrient (mg/kg)	
Texture: calcareous		Available - N	508.00 L
pH	8.4 H	Available - P	19.00 M
E.C dS/m	3.4 VH	Available - K	260.00 M
CaCO ₃ %	28.6 H	Available - Ca	493.00 L
O.M %	1.5 L	Available - Na	842.00 VH
		Available - Fe	3.63 VL
		Available - Mn	1.06 VL
		Available - Zn	0.85 L
		Available - Cu	0.70 L

VL=Very Low, L=Low, M=Moderate, H=High, VH=Very High (Ankerman and Large, 1974)
The irrigation water analyzes also showed a high salinity of the water, reaching 6 dS/m.

Growth parameters

Spraying treatments with zinc sulfate at a concentration of 5 g/L of water showed that there is a positive effect on growth parameters in both cultivars Giza 171, 168,

especially the two sprays in the tillering phase and the milky phase followed by the tillering phase compared to the control (Tables 2 and 3).

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Table 2. Effect of foliar spray with zinc sulfate at different stages on growth parameters of Giza 171

Characteristics Treatment	Plant height (cm)	Spike length (cm)	Spikelets no./spike	Spike number/m ²	100 - Kernel weight (g)
Control	100.0	13.6	17.7	280.0	4.53
At Tillering	112.0	15.0	20.0	313.3	4.92
At Milky Stage	104.0	14.5	19.3	294.3	4.76
At Tillering & Milky Stage	117.0	16.8	22.7	331.0	5.12
LSD (0.05)	3.11	0.52	1.49	8.28	0.03

Table 3. Effect of foliar spray with zinc sulfate at different stages on growth parameters of Giza 168

Characteristics Treatment	Plant height (cm)	Spike length (cm)	Spikelets no./spike	Spike number/m ²	100 - Kernel weight (g)
Control	94.7	10.7	16.3	247.7	4.12
At Tillering	102.0	12.5	18.3	283.3	4.43
At Milky Stage	99.0	11.7	17.7	271.0	4.34
At Tillering & Milky Stage	106.0	13.5	20.3	298.0	4.63
LSD (0.05)	2.58	0.68	1.29	6.67	0.07

Yield and its components

The improvement in growth parameters was reflected on the yield and its

components, as the biological yield and harvest index increased, as shown in Tables 4 and 5.

Table 4. Effect of foliar spray with zinc sulfate at different stages on yield and its components of Giza 171

Characteristics Treatment	Grain yield (ton/ha)	%	Biological yield (ton/ha)	Harvest Index (%)
Control	6.31	100	17.92	35.20
At Tillering	7.12	113	18.92	37.67
At Milky Stage	6.76	107	18.71	36.17
At Tillering & Milky Stage	7.57	120	19.57	38.63
LSD (0.05)	0.17	2	0.62	0.61

Table 5. Effect of foliar spray with zinc sulfate at different stages on yield and its components of Giza 168

Characteristics Treatment	Grain yield (ton/ha)	%	Biological yield (ton/ha)	Harvest Index (%)
Control	5.89	100	16.76	35.13
At Tillering	6.65	113	18.19	36.53
At Milky Stage	6.28	107	17.55	35.76
At Tillering & Milky Stage	7.00	119	18.92	37.00
LSD (0.05)	0.25	3	0.68	0.63

Growth parameters, yield and its components (2020/2021)

It is known that under the influence of the severe deficiency of zinc in this soil and in light of high soil pH, salinity and calcium carbonate, spraying with zinc has a high

impact on plant growth, especially because zinc has a strong impact on improving the physiological activity in plant cells. Growth Tables 6 and 7).and yield parameters also improved significantly in both cultivars, Giza 168 and 171.

Table 6. Effect of foliar spray applications with Zinc sulfate at different stages on growth parameters, yield and its components of Giza 168

Characteristics Treatment	Plant height (cm)	Spike length (cm)	Spikelets no./spike	Spike number/m ²	100 - Kernel weight (g)	Grain yield (ton/ha)	%	Biological yield (ton/ha)	Harvest Index (%)
Control	93.0	10.0	16.7	255.0	4.66	6.012	100	16.585	36.25
At Tillering	101.0	11.7	18.7	281.0	4.99	6.786	113	17.881	37.95
At Milky Stage	97.7	10.7	18.3	278.0	4.89	6.381	106	17.387	36.70
At Tillering & Milky Stage	103.7	14.0	19.3	284.2	5.12	7.050	117	18.109	38.93
LSD (0.05)	1.5	1.2	1.6	2.5	0.08	0.23	2	0.48	0.65

Table 7. Effect of foliar spray applications with Zinc sulfate at different stages on growth parameters, yield and its components of Giza 171

Characteristics Treatment	Plant height (cm)	Spike length (cm)	Spikelets no./spike	Spike number/m ²	100 - Kernel weight (g)	Grain yield (ton/ha)	%	Biological yield (ton/ha)	Harvest Index (%)
Control	98.9	12.7	17.4	262.0	4.73	6.501	100	17.806	36.51
At Tillering	111.1	14.0	19.4	288.0	5.10	7.182	110	18.510	38.80
At Milky Stage	105.0	13.2	19.1	285.4	4.99	6.874	106	18.224	37.72
At Tillering & Milky Stage	115.2	16.0	20.0	291.0	5.30	7.650	118	19.343	39.55
LSD (0.05)	2.6	0.8	1.3	2.4	0.06	0.27	3	0.25	1.05

Growth parameters, yield and its components (2021/2022)

Spraying treatments with zinc sulfate at a concentration of 5 g/L of water showed that there is a positive effect on growth parameters in both cultivars Giza 171, 168, especially the two sprays in the tillering

phase and the milky phase followed by the tillering phase compared to the control. The improvement in growth parameters was reflected on the yield and its components, as the biological yield and harvest index increased, as shown in Tables 8 and 9.

Table 8. Effect of foliar spray applications with Zinc sulfate at different stages on growth parameters, yield and its components of Giza 168

Characteristics Treatment	Plant height (cm)	Spike length (cm)	Spikelets no./spike	Spike number/m ²	100 - Kernel weight (g)	Grain yield (ton/ha)	%	Biological yield (ton/ha)	Harvest Index (%)
Control	94.7	11.1	17.1	250.2	4.76	6.70	100	18.11	37.00
At Tillering	102.0	12.0	19.5	285.2	5.09	7.40	110	19.47	38.01
At Milky Stage	99.0	12.8	18.9	270.9	4.98	7.05	105	18.83	37.44
At Tillering & Milky Stage	106.3	13.9	19.9	291.2	5.31	7.81	117	19.91	39.23
LSD (0.05)	2.87	0.71	0.89	3.40	0.15	0.31	3	0.70	0.72

Table 9. Effect of foliar spray applications with Zinc sulfate at different stages on growth parameters, yield and its components of Giza 171

Characteristics Treatment	Plant height (cm)	Spike length (cm)	Spikelets no./spike	Spike number/m ²	100 - Kernel weight (g)	Grain yield (ton/ha)	%	Biological yield (ton/ha)	Harvest Index (%)
Control	99.5	12.9	17.9	260.4	4.83	6.92	100	18.73	36.95
At Tillering	109.6	13.7	20.3	290.0	5.35	7.85	113	20.59	38.12
At Milky Stage	104.9	14.1	19.6	286.4	5.20	7.25	105	19.22	37.72
At Tillering & Milky Stage	113.5	14.9	20.9	294.0	5.69	7.98	115	20.08	39.75
LSD (0.05)	3.01	0.66	0.92	3.10	0.12	0.20	2	0.43	0.84

In this study, the effect of foliar spraying with zinc on the yield and its components of grain and straw was studied for three years using two wheat varieties. According to Ackerman and Large (1974), soils have high pH, as well as salinity and calcium carbonate, and are also low in organic matter and very low in zinc content. In this context, zinc is recommended when soil testing shows that the level of available zinc in the soil is below a critical level. The salinity level of irrigation water was also high (EC6 dS/m). It is known that under the influence of high soil pH, as well as salinity and high calcium carbonate, most of the nutrients are in an unavailable form for the plant (Alloway, 2008), especially in light of low organic matter, as is the case in dry and semi-dry lands where the rate of loss of organic matter is high due to high temperature and drought. In this context, Ramadan et al. (2020) indicated that the foliar spray method is more suitable for the availability of nutrients for plants to achieve optimal growth compared to the soil spray method and provides sufficient zinc nutrition for wheat.

However, the results in this paper show that grain Zn concentrations of high yielding cultivars can be significantly increased by foliar application of Zn fertilizers, even in some cases grain yield can be also simultaneously increased by foliar Zn application as shown in China by Karim et al. (2012) and in Turkey by Yilmaz et al. (1997). It was found in this study that spraying zinc did not have any negative effect on grain productivity. A recent research paper indicates that foliar-applied zinc meets the zinc requirements of wheat plant growth, improves grain zinc concentration and increases grain yield as shown by Cakmak et al. (1997), Yassen et al. (2011), and Hassan et al. (2019).

Experiment II Romania

The production obtained in the agricultural year 2019-2020 for the four analyzed varieties and the influences of the experimental factors on its realization are presented in the following.

Table 10. Production obtained in the agricultural year 2019-2020 of the Ciprian variety

Var.	Treatment	Treatment	Grain yield (kg)	Dif.	%	Sem.
v1	Untreated control	No seed treatment	5330	100	0.00	Control
v2	Soil treatment		5683	107	+353	-
v3	Foliar treatment - twinned completion		5830	109	+500	-
v4	Foliar treatment in the bellows phase		5667	106	+337	-
v5	Foliar treatment in the milk - wax phase		5833	109	+503	-
DL 5% - 920.12; 1% - 1241.27; 0.1% - 1611.22.						
v6	Untreated control	With seed treatment	5257	100	0.00	Control
v7	Soil treatment		5693	108	+436	-
v8	Foliar treatment - twinned completion		5737	109	+480	-
v9	Foliar treatment in the bellows phase		5307	101	+50	-
v10	Foliar treatment in the milk - wax phase		5707	109	+450	-
DL 5% - 660.12; 1% - 818.14; 0.1% - 1330.21.						

In the Ciprian variety (Table 10) maximum production obtained - 5833 kg/ha. Production increases are highlighted between the variants to which seed treatment was applied (zinc sulfate - 0.3% solution) and those to which this treatment was not applied. The highest increase is recorded when applying the foliar treatment in the wax milk phase x untreated seed - 503 kg/ha more than in the control version. In the version where the ground treatment x seed treatment was applied and the one with ground treatment x untreated seed,

the production increase is small - 454 kg/ha;

Compared to the unfertilized control, within the scheme - seed treatment x variety x soil/foliar treatment - no significant production increases are obtained;

Compared to the unfertilized control, within the scheme - untreated seed x variety x soil/ foliar treatment - a production increase is obtained only in the variant where zinc treatment was applied to the ground - 91 kg/ha, the production of the other variants being under the production of the blank.

Table 11. Production obtained in the agricultural year 2019-2020 of the Glosa variety

Var.	Treatment	Treatment	Grain yield (kg)	Dif.	%	Sem.
v1	Untreated control	No seed treatment	5680	100	0.00	Control
v2	Soil treatment		5750	101	+70	-
v3	Foliar treatment - twinned completion		5723	101	+43	-
v4	Foliar treatment in the bellows phase		5317	94	-363	-
v5	Foliar treatment in the milk - wax phase		5850	103	+170	-
DL 5% - 770.14; 1% - 900.56; 0.1% - 1200.21.						
v6	Untreated control	With seed treatment	5673	100	0.00	Control
v7	Soil treatment		6037	106	+364	-
v8	Foliar treatment - twinned completion		5747	101	+74	-
v9	Foliar treatment in the bellows phase		5797	102	+124	-
v10	Foliar treatment in the milk - wax phase		5810	102	+137	-
DL 5% - 840.36; 1% - 1127.68; 0.1% - 1668.81.						

In the Glosa variety (Tabel 11) maximum production achieved - 6037 kg/ha. Production increases are highlighted between the variants to which seed treatment was applied (zinc sulfate - 0.3% solution) and those to which this treatment was not applied. The highest production increase is recorded when applying the zinc treatment to the ground x seed treatment - 287 kg/ha more than in the ground treatment variant x untreated seed.

Between the variants to which foliar fertilization x seed treatment was applied and those with foliar fertilization x untreated seed, the production increase is 149 kg/ha.

Compared to the unfertilized control, within the scheme - seed treatment x variety x soil/ foliar treatment - no significant production increases were recorded in any of the experimental variants.

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Table 12. Production obtained in the agricultural year 2019-2020 of the Andrada variety

Var.	Treatment	Treatment	Grain yield (kg)	Dif.	%	Sem.
v1	Untreated control	No seed treatment	5333	100	0.00	Control
v2	Soil treatment		4980	93	-353	-
v3	Foliar treatment - twinned completion		4610	86	-723	-
v4	Foliar treatment in the bellows phase		5007	94	-326	-
v5	Foliar treatment in the milk - wax phase		4777	90	-556	-
DL 5% - 619.84; 1% - 991.12; 0.1% - 1478.86.						
v6	Untreated control	With seed treatment	4820	100	0.00	Control
v7	Soil treatment		5113	106	+293	-
v8	Foliar treatment - twinned completion		4623	96	-497	-
v9	Foliar treatment in the bellows phase		4747	98	-73	-
v10	Foliar treatment in the milk - wax phase		4920	102	+100	-
DL 5% - 830.11; 1% - 1111.11; 0.1% - 1665.28.						

In the Andrada variety (Table 12) maximum production obtained - 5333 kg/ha. Production increases are highlighted between the variants to which seed treatment was applied (zinc sulfate - 0.3% solution) and those to which this treatment was not applied. The highest production increase is recorded when applying the zinc treatment to the ground x seed treatment - 133 kg/ha more than in the ground treatment variant x untreated seed. Between the variants to which foliar fertilization x seed treatment was applied and those with foliar fertilization x untreated seed, the production increases are very small, insignificant.

Compared to the non-fertilized control, within the scheme - seed treatment x variety x soil/ foliar treatment - a positive production increase is obtained only in variants where ground treatment and foliar treatment in the milk-wax phase have been applied.

Compared to the unfertilized control, within the scheme - untreated seed x variety x soil/ foliar treatment - a positive production increase is obtained only in the variant to which zinc treatment was applied in the bellows phase.

Regarding the influence of the experimental factors on the production, factor A - the seed treatment with zinc sulfate - has an insignificant influence. Thus, compared to the untreated control variant, the application of the foliar treatment brings a small decrease in production, by approximately 1%, respectively, 36 kg/ha.

Factor C - zinc sulfate treatment - in the crop conditions of the 2019-2020 agricultural year, each analyzed genotype shows its productive potential. The best production results are obtained with the foliar application of zinc sulfate in the waxy milk phenophase, where a production exceeding the control, unfertilized variant by 20% is recorded. As for the productivity of varieties, in the conditions of 2019-2020, from the data presented in table above we can conclude that the production obtained in the agricultural year 2020-2021 for the four varieties analyzed and the influence of experimental factors on its realization are presented as follows.

In the 2019-2020 agricultural year, zinc applied in various forms (soil, as a seed and foliar treatment, at different stages of crop development) had a positive influence on

wheat crops, both in Romania and in Egypt. Thus, taking into account the cultivars used and the climatic conditions of the experimental areas, the production differences compared to the unfertilized control reached up to 19% in Egypt and up to 9% in Romania. In Romania, the application of soil fertilization and foliar fertilization at the end of the tillering period stands out with very good results, while in Egypt the most important treatment for increasing production and productivity elements of the crop is the combined application of zinc at tillering & milky stage.

The recorded production of the Ciprian variety (Table 13), in the climatic conditions

of the agricultural year 2020-2021, brings a difference from the control of 9.1% (without seed treatment) in the fertilized version of foliar zinc sulfate 0.3% in the milk-wax stage and 18.6% (with seed treatment) in the foliar fertilized version in the bellows phase.

When applying the seed treatment, the production varies in the range of 4907 kg/ha (control) - 5820 kg/ha (foliar treatment in the bellows phase), with a production increase of 913 kg/ha, and in the variants where no seed treatment was applied - 5342 kg/ha in the blank version and 5827 kg/ha (foliar treatment in the bellows phase), 484 kg/ha more.

Table 13. Production obtained in the agricultural year 2020-2021 of the Ciprian variety

Var.	Treatment	Treatment	Grain yield (kg)	Dif.	%	Sem.
v1	Untreated control	No seed treatment	5342	0,00	100	Control
v2	Soil treatment		5642	300	105,6	-
v3	Foliar treatment - twinned completion		5224	118	97,8	-
v4	Foliar treatment in the bellows phase		5340	2,3	100	-
v5	Foliar treatment in the milk - wax phase		5827	484	109,1	-
DL 5% - 1075.14; 1% - 1563.84; 0.1% - 2345.77.						
v6	Untreated control	With seed treatment	4907	0,00	100	Control
v7	Soil treatment		5069	162	103,3	-
v8	Foliar treatment - twinned completion		5277	371	107,6	-
v9	Foliar treatment in the bellows phase		5820	913	118,6	*
v10	Foliar treatment in the milk - wax phase		5284	378	107,7	-
DL 5% - 837.5; 1% - 1218.18; 0.1% - 1827.27.						

In the Glosa variety (Table 14), the mineral fertilization on the ground with zinc sulphate - 30 kg/ha - was noted both when applying the seed treatment and in its absence, with an increased production

increase of 24% in both cases. Compared to the control variant, there is an increase in production ranging from 124 kg/ha to 1323 kg/ha.

Table 14. Production obtained in the agricultural year 2020-2021 of the Glosa variety

Var.	Treatment	Treatment	Grain yield (kg)	Dif.	%	Sem.
v1	Untreated control	No seed treatment	5409	0,00	100	Ctrl.
v2	Soil treatment		6731	1323	124,5	*
v3	Foliar treatment - twinned completion		5551	142	102,6	-
v4	Foliar treatment in the bellows phase		5982	573	110,6	-
v5	Foliar treatment in the milk - wax phase		5778	369	106,8	-
DL 5% - 1049.84; 1% - 1527.05; 0.1% - 2290.57.						
v6	Untreated control	With seed treatment	5462	0,00	100	Ctrl.
v7	Soil treatment		6778	1316	124,1	*
v8	Foliar treatment - twinned completion		5802	340	106,2	-
v9	Foliar treatment in the bellows phase		5643	180	102,3	-
v10	Foliar treatment in the milk - wax phase		5976	513	105,4	-
DL 5% - 1060.93; 1% - 1543.17; 0.1% - 2314.76.						

In the case of spring wheat, Pădureni (Table 15) variety, there are noted increases in production under the influence of soil fertilization, with 35.9% more than in the control version and a production increase of 373 kg/ha, statistically significant increase

and the foliar fertilized variant in the development phase of milk - wax wheat, with an increase compared to the control of 502 kg/ha (48.3%), statistically assured increase as distinctly significant for the probability of transgression of 1%.

Table 15. Production obtained in the agricultural year 2020-2021 for Pădureni variety

Var.	Treatment	Treatment	Grain yield (kg)	Dif.	%	Sem.
v1	Untreated control	No seed treatment	1040	0,0	100	Ctrl.
v2	Soil treatment		1413	373	135,9	*
v3	Foliar treatment - twinned completion		1220	180	117,3	-
v4	Foliar treatment in the bellows phase		1365	325	131,2	-
v5	Foliar treatment in the milk - wax phase		1542	502	148,3	**
DL 5% - 332.56; 1% - 483.73; 0.1% - 725.6.						

The fertilization with zinc on the ground also brings in the case of the Dacic wheat variety (Table 16) the production increases superior to the control, 569 kg/ha in the case

of applying the treatment to the seed and 969 kg/ha when the seed treatment is not applied, which are statistically significant increases for the probability of transgression of 5%.

Table 16. Production obtained in the agricultural year 2020-2021 of the Dacic variety

Var.	Treatment	Treatment	Grain yield (kg)	Dif.	%	Sem.
v1	Untreated control	No seed treatment	5942	0.00	100	Ctrl.
v2	Soil treatment		6911	969	116,3	*
v3	Foliar treatment - twinned completion		6378	436	107,3	-
v4	Foliar treatment in the bellows phase		6718	776	113,1	*
v5	Foliar treatment in the milk - wax phase		6298	355	106	-
DL 5% - 727.39; 1% - 1058.02; 0.1% - 1587.03.						
v6	Untreated control	With seed treatment	6349	0,0	100	Ctrl.
v7	Soil treatment		6918	569	109	*
v8	Foliar treatment - twinned completion		6651	352	104,8	-
v9	Foliar treatment in the bellows phase		6476	127	102	-
v10	Foliar treatment in the milk - wax phase		6611	262	104,1	-
DL 5% - 531.03; 1% - 772.41; 0.1% - 1158.61.						

Considering the effectiveness of seed treatment with zinc solution for the Pădureni spring wheat variety, we chose to experiment with the new lines obtained at ARDS Turda, treating the seeds before sowing with a solution in a concentration of 0.3% zinc in the form of zinc sulphate.

The production results obtained in the new lines of spring wheat are spectacular, compared to those obtained in Pădureni, the best line T 4188-19 surpassing it with 2594 kg/ha (Table 17). Also, the quality indices required by the bakery industry have

higher values, compared to winter wheat, the highest protein content being recorded in the Pădureni variety (14.8%).

In the 2020-2021 agricultural year, the same trend of production growth under the influence of applied zinc treatments is maintained. For Romania, the soil treatment brings production increases of up to 1300 kg/ha (25%) for winter wheat and up to 370 kg/ha for spring wheat (36%). In Egypt, the foliar treatment at tillering & milky stage remains the best option, with a difference of 19-20% compared to the control.

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Table 17. Production results and quality indices of spring wheat varieties and lines with seed treatment 100% zinc solution, SCDA Turda 2020

Genotype	Yield (kg/ha)	Protein content (%)	Wet gluten content (%)	Zeleny test (ml)	Test weight (kg/hl)
Pădureni	4491	14.8	30.0	61.9	78.9
Triso	6523	13.8	27.7	52.6	80.9
T. 3935-19	6458	12.6	25.1	42.1	79.0
T. 3936-19	6358	12.8	25.5	44.9	79.5
T. 3938-19	5924	13.7	27.4	51.0	75.8
T. 3964-19	6629	12.5	24.8	41.5	79.9
T. 3965-19	6695	13.0	26.0	44.6	81.7
T. 3974-19	6428	12.6	25.0	43.4	82.6
T. 4007-19	6499	13.2	26.3	48.3	80.0
T. 4015-19	6931	12.9	25.7	43.8	81.9
T. 4066-19	6331	13.7	27.5	52.5	78.2
T. 4068-19	6686	13.3	26.7	51.9	82.1
T. 4071-19	6856	14.0	28.1	52.9	79.3
T. 4073-19	6853	13.6	27.2	51.6	80.5
T. 4074-19	6167	14.0	28.1	55.6	79.6
T. 4075-19	6752	13.8	27.7	53.0	81.6
T. 4076-19	6496	13.0	25.8	46.1	81.1
T. 4107-19	6851	13.7	27.4	52.0	78.5
T. 4162-19	6653	13.6	27.3	50.5	81.4
T. 4165-19	6841	13.2	26.3	48.0	81.9
T. 4173-19	6675	13.9	28.0	53.7	79.7
T. 4176-19	6920	13.1	26.2	47.7	80.7
T. 4183-19	6877	13.4	26.8	49.4	82.5
T. 4188-19	7085	13.5	27.0	51.0	81.2
T. 4189-19	6326	13.1	26.2	47.1	80.4
Media	6532				
DL5%	413				

Results similar to those obtained in this research project have been reported by the following researchers: Reis et al. (1982), Graham and Webb (1991), Brennan (1992), Grewal et al. (1996), Grewal (2001), Streeter et al. (2001), Cuero et al. (2003), Huber and Haneklaus (2007), Khoshgoftarmanesh et al. (2010), Helfenstein et al. (2015).

CONCLUSIONS

Applied in various forms and at different stages of wheat crop development (on the soil, as a seed and foliar treatment), zinc had a positive influence on wheat crop, both in Romania and in Egypt. Thus, taking into account the cultivars used and the climatic conditions of the experimental areas, the differences in production compared to the unfertilized control, during the period in which the experiment was carried out, reached up to 20% in Egypt and up to 17% in

Romania. Among the cultivars used, the most receptive to agronomic biofortification with zinc are the spring cultivars.

In Romania, the application of soil fertilization and foliar fertilization at the end of the tillering period stands out with very good results, while in Egypt the most important treatment for increasing production and productivity elements of the crop is the combined application of zinc at tillering and milky stage.

Regarding seed treatment with zinc sulfate, it brings modest increases in production, but contributes to increasing crop resistance, zinc being able to be successfully used as an alternative to fungicide treatment.

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REFERENCES

- Alison, L.E., Moodle, C.D., Carborate, 1965. In: C.A. Black (eds.), *Methods of Soil Analysis*. American Society of Agronomy, Madison, Wisconsin, USA: 1379-1396.
- Alloway, B.J., 2008. *Zinc in soils and crop nutrition*. 2nd ed., International Zinc Association (IZA) and International Fertilizer Association (IFA), Brussels, Belgium and Paris, France.
- Ankerman, D., and Large, R., 1974. *Soil and plant analysis*. Tech. Bull. A&L, Agricultural laboratories, Inc., New York, USA: 42-44, 74-76.
- Bhatt, R., Hossain, A., Sharma, P., 2020. *Zinc biofortification as an innovative technology to alleviate the zinc deficiency in human health: a review*. From the Journal Open Agriculture.
- Bouyoucos, H.H., 1954. *A recalibration of the hydrometer for making mechanical analysis of soils*. Agronomy Journal; 43: 343-348.
- Brennan, R.F., 1992 *The effectiveness of zinc fertilizers as measured by DTPA soil extractable zinc, dry matter production and zinc uptake by subterranean clover in relation to soil properties of a range of Australian soils*. Soil Research, 30: 45-53.
- Broadley, M., White, P., Hammond, J., Zelko, I., Lux, A., 2007. *Zinc in plants*. New Phytol., 173: 677-702.
- Cakmak, I., Ekiz, H., Yilmaz, A., Torun, B., Koleli, N., Gultekin, I., Alkan, A., Eker, S., 1997. *Differential response of rye, triticale, bread and durum wheats to zinc deficiency in calcareous soils*. Plant and Soil, 188: 1-10.
- Cakmak, I., 2002. *Plant nutrition research: priorities to meet human needs for food in sustainable ways*. Plant Soil, 247: 3-24.
- Chen, X.P., Zhang, Y.Q., Tong, Y.P., Xue, Y.F., Liu, D.Y., Zhang, W., Deng, Y., Meng, Q.F., Yue, S.C., Yan, P., Cui, Z.L., Shi, X.J., Guo, S.W., Sun, Y.X., Ye, Y.L., Wang, Z.H., Jia, L.L., Ma, W.Q., He, M.R., Zhang, X.Y., Kou, C.L., Li, Y.T., Tan, D.S., Cakmak, I., Zhang, F.S., Zou, C.Q., 2017. *Harvesting more grain zinc of wheat for human health*, Scientific Reports, 7, 7016, DOI:10.1038/s41598-017-07484-2.
- Cuero, R., Ouellet, T., Yu, J., Mogongwa, N., 2003. *Metal ion enhancement of fungal growth, gene expression and aflatoxin synthesis in Aspergillus flavus: RT-PCR characterization*. Journal of Applied Microbiology, 94: 953-961.
- El-Habbasha, E.S., Badr, E.A., Latef, E.A., 2015. *Effect of zinc foliar application on growth characteristics and Grain Yield of some wheat varieties under Zn deficient sandy soil condition*. International Journal Chemtech Research, 8(6): 452-458.
- Gomez, K.A., and Gomez, A.A., 1984. *Statistical procedures for agricultural research (2nd ed.)*. John Wiley and Sons, New York.
- Graham, D.R., and Webb, M.J., 1991. *Micronutrients and disease resistance and tolerance in plants*, In: Mortvedt, J.J., Cox, F.R., Shuman, L.M., Welch, R.M. (eds.), *Micronutrients in agriculture*. 2nd Edn, Madison, Wisconsin, USA, Soil Science Society of America Inc.: 329-370.
- Graham, R.D., Welch, R.M., Bouis, H.E., 2001. *Addressing micronutrient malnutrition through enhancing the nutritional quality of staple foods: principles, perspectives and knowledge gaps*. Adv. Agron., 70: 77-142.
- Grewal, H.S., Graham, R.D., Rengel, Z., 1996. *Genotypic variation in zinc efficiency and resistance to crown rot disease (Fusarium graminearum Schw Group 1) in wheat*. Plant Soil, 186: 219-226.
- Grewal, H.S., 2001. *Zinc influences nodulation, disease severity, leaf drop and herbage yield of alfalfa cultivars*. Plant Soil, 234: 47-59.
- Hassan, M.U., Chattha, M.U., Ullah, A., Khan, I., Qadeer, A., Aamer, M., Khan, A.U., Nadeem, F., Khan, T.A., 2019. *Agronomic biofortification to improve productivity and grain Zn concentration of bread wheat*. Int. J. Agric. Biol., 21: 615-620.
- Haslett, B.S., Reid, R.J., Rengel, Z., 2001. *Zinc mobility in wheat: uptake and distribution of zinc applied to leaves or roots*. Annals of Botany, 87: 379-386.
- Helfenstein, J., Pawlowski, M.L., Hill, C.B., Stewart, J., Lagos-Kutz, D., Bowen, C.R., 2015. *Zinc deficiency alters soybean susceptibility to pathogens and pests*. J. Plant Nutr. Soil Sci., 178: 896-903.
- Hotz, C., and Brown, K.H., 2004. *Assessment of the risk of zinc deficiency in populations and options for its control*. Food and Nutrition Bulletin, 2: 194-204.
- Huber, D.M., and Haneklaus, S., 2007. *Managing nutrition to control plant disease*. Landbauforsch. Volk., 4: 313-322.
- Jackson, M.L., 1986. *Soil Chemical Analysis*. Printice Hall, Indian Private Limited, New Delhi: 251-280.
- Karim, R., Zhang, Y., Zhao, R., Chen, X., Zhang, F., Zou, C., 2012. *Alleviation of drought stress in winter wheat by late foliar application of zinc, boron, and manganese*. Journal of Plant Nutrition and Soil Science, 175(1): 142-151.
- Khoshgoftarmanesh, A.H., Kabiri, S., Shariatmadari, H., Sharifnabi, B., Schulin, R., 2010. *Zinc nutrition effect on the tolerance of wheat genotypes to Fusarium root-rot disease in a solution culture experiment*. Soil Sci. Plant Nutr., 56: 234-243.

- Nowack, B., Schwyzer, I., Schulin, R., 2008. *Uptake of Zn and Fe by wheat (Triticum aestivum var. Greina) and transfer to the grain in the presence of chelating agents (Ethylenediaminedisuccinic acid and Ethylenediaminetetraacetic acid)*. J. Agric. Food Chem., 56: 4643-4649.
- Peck, A.W., McDonald, G.K., Graham, R.D., 2008. *Zinc nutrition influences the protein composition of flour in bread wheat (Triticum aestivum L.)*. J. Cereal Sci., 47: 266-274.
- Prasad, R., Shivay, Y.S., Kumar, D., 2013. *Zinc fertilization of cereals for increased production and alleviation of Zn malnutrition in India*. Agric. Res., 2(2): 111-118.
- Ramadan, Y., Hafeez, M.B., Khan, S., Nadeem, M., Rahman, S., Batool, S., Ahmad, J., 2020. *Biofortification with zinc and iron improves the grain quality and yield of wheat*. Crop International Journal of Plant Production, 14(3): 501-510.
- Ranjbar, G.A., and Bahrmaniar, M.A., 2007. *Effects of soil and foliar application of Zn fertilizer on yield and growth characteristics of bread wheat cultivars*. Asian J. Plant Sci., 6(6): 1000-1005.
- Reis, E.M., Cook, R.J., McNeal, B.L., 1982. *Effect of mineral nutrition on take-all of wheat*. Phytopathology, 72: 224-229.
- Streeter, T.C., Rengel, Z., Neate, S.M., Graham, R.D., 2001. *Zinc fertilisation increases tolerance to Rhizoctonia solani (AG 8) in Medicago truncatula*. Plant Soil, 228: 233-242.
- Timsina, Y.N., 2014. *Effect of Nitrogen Fertilization on Zinc and Iron Uptake and Yield Components of Wheat*. Master thesis, Norwegian University of Life Sciences, As, Akershus, 80.
- Yassen, A.A., Hellal, F.A., Doaa, M., Abo-Basha, 2011. *Influence of organic materials and foliar application of zinc on yield and nutrient uptake by wheat plants*. Journal of Applied Sciences Research, 7(12): 2056-2062.
- Yilmaz, A., Ekiz, H., Torun, B., Gultekin, I., Karanlik, S., Bagci, S.A., Cakmak, I., 1997. *Effect of different zinc application methods on grain yield and zinc concentration in wheat grown on zinc-deficient calcareous soils in Central Anatolia*. Journal of Plant Nutrition, 20: 461-471.
- Velu, G., Ortiz-Monasterio, I., Singh, R.P., Payne, T., 2011. *Variation for grain micronutrients concentration in wheat core-collection accessions of diverse origin*. Asian J. Crop Sci., 3: 43-48.
- Walkley, A., and Black, I.A., 1934. *An examination of the Degtjareff method for determining organic matter and a proposed modification of the chromic acid titration method*. Soil Science, 37(1): 29-38.