

# Soil-foliar Biofortification of Wheat with Zinc and Iron - Self-explanatory Model

Marija Menkinoska<sup>1</sup>, Angelina Stredovska Bozhinov<sup>1</sup>, Sasko Martinovski<sup>2</sup>,  
Tatjana Blazevska<sup>2</sup>, Zlatin Zlatev<sup>3</sup>, Goce Menkonoski<sup>4</sup>

<sup>1</sup>International Balkan University, Faculty of Dental Medicine Makedonsko-Kosovska Brigada bb 1000,  
Skopje/North Macedonia

<sup>2</sup>University St Kliment Ohridski Bitola, Faculty of Technological and Technical Sciences – Veles, Dimitar  
Vlahov bb str., 1400 Veles, North Macedonia

<sup>3</sup>Trakia University, Faculty of Technics and technologies, 38 Graf Ignatiev str., 8602, Yambol, Bulgaria

<sup>4</sup>University St Kliment Ohridski Bitola, Faculty of Economics, Prilep, Adress: St. „Prilepski Braniteli“ No:143  
/ 7500 Prilep, North Macedonia

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**ABSTRACT:** Bio-fortification is a process that increases the concentration of micronutrients in plants while they are in the growth stage. Adding nutrients to food during their processing is less natural in terms of biofortification. Zinc and iron as micronutrients are particularly important for human health, and common wheat (*Triticum aestivum*) is a major component of the human diet, and therefore all research to increase zinc and iron concentrations during the cultivation of wheat, and afterwards in the flour, is quite significant. The first goal, is to develop a model that will enable obtaining information on wheat and flour. The second goal is to raise the model to a higher level, i.e. to incorporate the self-explanatory aspect. Some results are given from the outputs of model obtained from this research on the production of wheat and flour with zinc and iron of the variety Treska. New modelling concept for Soil-foliar biofortification of wheat with zinc and iron is obtained. The benefits can be threefold: a benefit for companies through greater profit, a benefit for citizens through the consumption of healthy and quality food products and finally, a benefit for the state. The model will allow companies engaged in the production of wheat and flour to obtain a wealth of information on methods for increasing zinc and iron in wheat and flour, as well as other important information.

**KEYWORDS** - biofortification, model, entity-relationship model, self-explanatory, wheat

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Date of Submission: 05-01-2024

Date of Acceptance: 17-01-2024

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## I. INTRODUCTION

Today, research by modern science leads to the realization that certain nutrients are essential, but also that they are needed in certain amounts for optimal human health [1].

However, unlike the pharmaceutical industry in which medicines have a targeted approach to restoring health, the food industry is a multi-parameter approach to preserving and/or improving health. In fact, the diet consists of a multitude of nutritional and chemical molecules, each of which can regulate different biological processes, and thus a pharmaceutical industry-like approach, i.e., "one medicine, one goal" is not applicable [2].

Plant-based foods provide a range of nutrients that are essential for human nutrition, and thus promoting good health. Therefore, the interest in using functional foods in the prevention and/or treatment of various diseases keeps increasing [3]. However, in most of the world, the main raw materials obtained from agricultural crops are often deficient in some of these nutrients.

Traditional agriculture technology can slightly improve the nutritional value of some foods, but rapid advances in molecular biology should be used to produce crops with increased mineral, amino acid and antioxidant content, as well as improved fatty acid composition.

At least 22 minerals are needed to maintain human health [4] which can be provided by proper nutrition. However, it is estimated that over 60% of the world's population is deficient in iron, over 30% in zinc, 30% iodine, 15% selenium, then calcium, magnesium and copper are common deficiencies not only in developing countries but also in developed countries [5]. This is due to the increased consumption of plant foods and cereals with low concentration of minerals, because they are grown in areas with low content of available minerals, and also an additional reason is the insufficient intake in the diet of animal-based products [6]. Lack of zinc in soils also reduces the concentration of Zn in wheat grain, thus reducing the nutritional quality [7]. A comprehensive study [8] shows that in average, one third of the world's population is affected by Zn deficiency.

The main solution to reduce the deficiency of Zn and other micronutrients in the cultivation of cereals is biofortification (biological enrichment).

In creating models for enrichment of wheat with zinc and iron through soil-foliar biofortification (but also for other purposes) it is necessary to include scientific methods and concepts using advanced technology. For better development and use of these models, it is necessary to present them in a relationship-entity format, and then, to be able to provide the so-called self-explanatory aspect of the model or automatic explanation of each of its outputs. Also, the use of modern technology is necessary for building such models, such as information technology, and one such technology is the Geographic Information System (GIS) [9] [10], which can help in the production of wheat by creating systemic models (spatial and analytical) that can describe the current situation and project the future (GIS contains a wide range of tools for spatial analysis).

## II. MATERIALS AND METHODS

The research on soil-foliar bio-fortification of wheat with zinc and iron was completed with the use of a model obtained with an original modeling concept. In modeling, one of the key factors is the correct determination of the stages (sub-stages) and their order. One characteristic typical for this model is in stage 2, where the entity OUTPUT is first defined and based on that, the entities METHODOLOGY and INPUT are defined, in many models, when modeling, the order is reversed. Second, the model we propose is not classically mathematical or statistical. It includes several different methods such as: mathematics, statistics, logical relations, information and economic methods, etc., and all these methods are defined as entities in a logical relationship. Specific methods used in the proposed model are methods required for soil-foliar biofortification; geographical methods for spatial and analytical analysis using GIS; and economic methods. The third important feature of the model is the ability for each output of the model to create an explanation for its acquisition. Certainly, with that, its theoretical and applied applicability and development will be drastically improved.

According to the structure of the model, it can be concluded that it resembles an input-output model but has specifics that make it original and different.

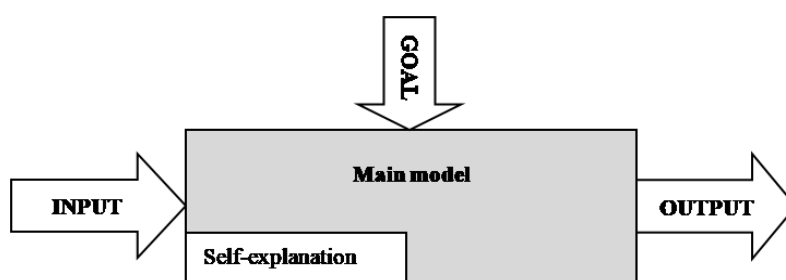
Further on in the text we will show an original modeling concept for model creation, with built-in self-explanation. Fig.1 generally shows the main model consisting of four main entities:

GOAL: entering the goals and interests of the main model;

INPUT: entering the necessary input data, which can be spatial and non-spatial (analytical);

OUTPUT: obtained output data on entering the necessary input data for wheat and flour which can be spatial and analytical. Outputs for this model are: concentration of iron and zinc; morphological and production characteristics; minerals; quality properties (proteins, gluten, fats, phenols and antioxidant capacity, moisture, etc.); economic parameters for costs and revenues in the production of wheat and flour; and spatial data and analyzes required for wheat production.

Self-explanation: Another important feature of this model is to provide self-explanation, which means that for each output it will automatically create an explanation of the relationships and methods and input used to obtain it.



**Figure 1. Self-explanatory model for fortification of wheat with zinc and iron**

In the first phase of modeling the entity GOAL is defined, where two sub-entities are defined: Goal and Interest which are in a relationship (equation 1):

$$I_n = R(G_n) \tag{1}$$

where  $G_n$ -Goals,  $I_n$ -Interests.

The following goals and interests are defined for this model:

$G_1$ : Self-explanatory model for enrichment of wheat with zinc and iron through soil-foliar biofortification.

$G_2$ : Obtaining wheat and flour enriched with zinc and iron that will be used in the production of wheat products (bread, pastries, etc.) with higher nutritional properties.

$G_3$ : Obtaining wheat and flour enriched with zinc and iron at prices that can be accepted by the market.

$I_1=R(G_1, G_2, G_3)$ : Economic interest of companies to produce wheat products (bread, pastries, etc.) with higher nutritional properties (zinc and iron).

$I_2=R(G_1, G_2, G_3)$ : Interest of consumers to consume wheat products (bread, pastries, etc.) with higher nutritional properties (zinc and iron).

$I_3=R(G_1, G_2, G_3)$ : Interest of the country for the consumption of healthy and quality products.

What is specific for the second stage is that a relational entity relationship is established in the following order: first, the outputs are defined (equation 2), second, based on the outputs, the required methodology for their acquisition is defined and finally, based on the methodology, the required inputs and its data sources are defined (Fig.2). So, the output is related to the methodology, and can be in analytical or graphical form.

$$Ou_n=R(M_n) \tag{2}$$

where  $Ou_n$ - Outputs,  $M_n$ -methodologies.

The required methodology used to obtain the outputs is an important segment in modeling. This section defines the methodologies to be used in the model. This is necessary in building models and is an important step to get quality outputs. The methodology is related to the inputs in the model (equation 3):

$$M_n=R(In_n) \tag{3}$$

where  $In_n$ - Inputs,  $M_n$ -methodologies.

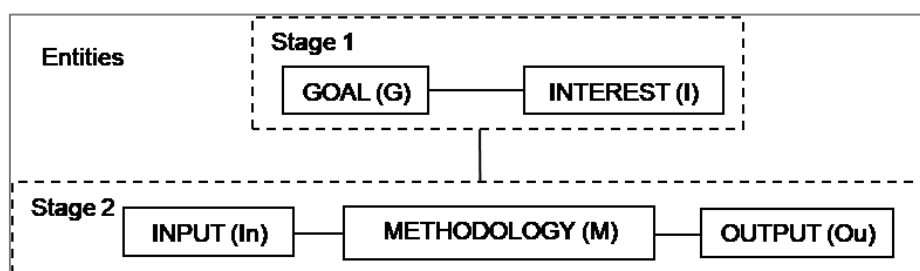


Figure 2. Relationship of the main entities from stage 1 and stage 2

The model outputs are shown in Table 1.

Table 1. Model outputs

Output	Explanation
$Ou_1(M)$	Morphological and production characteristics of wheat: number of ears in the main area, number of grains per wheat ear, grain weight, main wheat ear weight (Eg.2 for all outputs)
$Ou_2(M)$	Comparison of mean values of morphological traits in wheat - statistical method is used - LSD test with analysis of values $p < 0.05$ - statistically significant value
$Ou_3(M)$	Classification of produced wheat
Quality characteristics of wheat and flour	
$Ou_4(M)$	Wet gluten content
$Ou_5(M)$	Raw protein content
$Ou_6(M)$	Fat content
$Ou_7(M)$	Minerals in wheat and flour (ash)
$Ou_8(M)$	Concentration of iron and zinc
$Ou_9(M)$	Farinological analysis of quality characteristics
$Ou_{10}(M)$	Moisture content
$Ou_{11}(M)$	Total phenol content and antioxidant capacity

The methodologies needed to obtain the outputs are shown in the following table (Table 2).

Table 2. Research methodology

Methodology	Explanation
$M_1$	Research method (Eg.3 for all methodologies) 1. fertilization methods 2. fertilization variants 3. repetitions 4. fortifying of wheat 5. varieties of wheat 6. types of fertilizer and amount of fertilizer 7. distance between parcels 8. seed 9. sowing period 10. number of parcels
$M_2$	Stages of fertilization of the wheat
$M_3$	Statistical method - LSD test
$M_4$	Method for determination of morphological characteristics of wheat - Biometric methods for analysis of morphological and production characteristics (height of stem (cm), length of ear, etc.)

M <sub>5</sub>	Laboratory tests - agrochemical analysis of soil
M <sub>6</sub>	Classification of wheat
M <sub>7</sub>	Minerals in wheat and flour (ash)- Determination of the amount of ash in grains and mill products by gravimetric method
M <sub>8</sub>	Quality characteristics of wheat and flour - Wet gluten content, procedure for determination of wet gluten content in grain and mill products
M <sub>9</sub>	Quality characteristics of wheat and flour - Crude protein content by determination of total nitrogen
M <sub>10</sub>	Quality characteristics of wheat and flour- Fat content as determined by the Soxlet method with Diethyl ether extraction
M <sub>11</sub>	Quality characteristics of wheat and flour- Concentration of iron and zinc in wheat and flour, determined with an atomic absorption spectrometer
M <sub>12</sub>	Quality characteristics of wheat and flour –Farinological testing and Brabender farinograph
M <sub>13</sub>	Quality characteristics of wheat and flour- Moisture content (%)
M <sub>14</sub>	Quality characteristics of wheat and flour- Total phenol content and antioxidant capacity

The model inputs are shown in the following table (Table 3).

**Table 3. Model inputs**

Inputs	Explanation
In <sub>1</sub>	Variants included in the research
In <sub>2</sub>	Characteristics of several types of fertilizers
In <sub>3</sub>	Pre-culture
In <sub>4</sub>	Soil samples, soil composition and chemical and mechanical composition of the soil
In <sub>5</sub>	Wheat variety
In <sub>6</sub>	Seed characteristics
In <sub>7</sub>	Climate conditions
In <sub>8</sub>	Costs and planned revenues for obtaining fortified wheat and flour

Based on all the entities from the previous phases, the creation of the logical model begins with the establishment of relations between them in the ER model. This concept of modeling will allow the business model to be self-explanatory of the process embedded in it. This means that for each output there will be a specific explanation of how it is obtained, which methods are used, and which data are used. Also, further development and its use (applied and theoretical) will be much easier.

The following are the relations between all the outputs and the methodology (equations 4):

$$\begin{array}{ll}
 \text{Ou}_1 = R(\text{M}_1, \text{M}_2, \text{M}_4, \text{M}_5) & \text{Ou}_6 = R(\text{M}_1, \text{M}_2, \text{M}_5, \text{M}_{10}) \\
 \text{Ou}_2 = R(\text{M}_1, \text{M}_2, \text{M}_3, \text{M}_5) & \text{Ou}_7 = R(\text{M}_1, \text{M}_2, \text{M}_5, \text{M}_7) \\
 \text{Ou}_3 = R(\text{M}_1, \text{M}_2, \text{M}_4, \text{M}_5, \text{M}_6) & \text{Ou}_8 = R(\text{M}_1, \text{M}_2, \text{M}_4, \text{M}_5, \text{M}_{11}) \\
 \text{Ou}_4 = R(\text{M}_1, \text{M}_2, \text{M}_5, \text{M}_8) & \text{Ou}_9 = R(\text{M}_1, \text{M}_2, \text{M}_5, \text{M}_{12}) \\
 \text{Ou}_5 = R(\text{M}_1, \text{M}_2, \text{M}_5, \text{M}_9) & \text{Ou}_{10} = R(\text{M}_1, \text{M}_2, \text{M}_5, \text{M}_{13}) \\
 \text{Ou}_{11} = R(\text{M}_1, \text{M}_2, \text{M}_5, \text{M}_{14}) &
 \end{array} \tag{4}$$

The following are the relations between the methodology and inputs (equations 5):

$$\begin{array}{ll}
 \text{M}_1 = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) & \text{M}_8 = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) \\
 \text{M}_2 = R(\text{In}_1, \text{In}_2) & \text{M}_9 = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) \\
 \text{M}_3 = R(\text{In}_1, \text{In}_2, \text{In}_3) & \text{M}_{10} = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) \\
 \text{M}_4 = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4) & \text{M}_{11} = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) \\
 \text{M}_5 = R(\text{In}_1, \text{In}_4) & \text{M}_{12} = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) \\
 \text{M}_6 = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) & \text{M}_{13} = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) \\
 \text{M}_7 = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7) & \text{M}_{14} = R(\text{In}_1, \text{In}_2, \text{In}_3, \text{In}_4, \text{In}_5, \text{In}_6, \text{In}_7)
 \end{array} \tag{5}$$

Fig. 3 shows an entity relationship in the Ou<sub>8</sub> output model (for Fe and Zn concentration) and how to obtain the self-explanation.

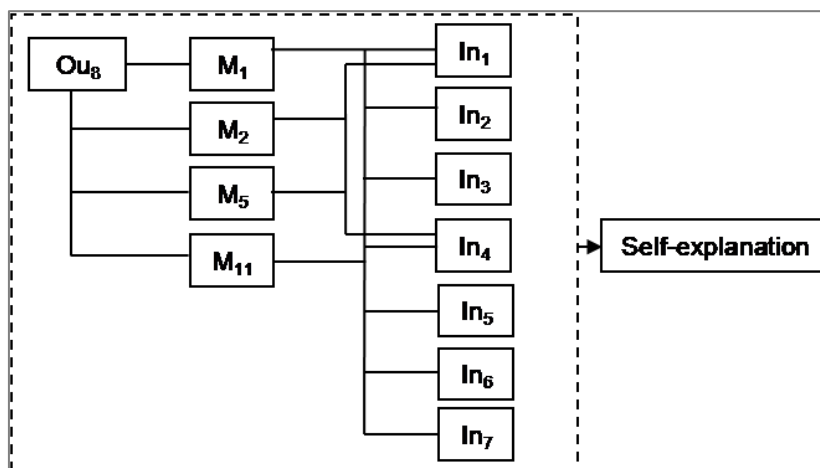


Figure 3. Entity relationships OUTPUT-METHODOLOGY-INPUT

The physical model is the stage of implementation of the logical model in a software solution (Fig. 3). The creation is done in a software development environment, with integration of DBMS and other necessary software. GIS software can also be used, and we did our research in the software of the company ESRI - ArcMap, where GIS models are created using the Model Builder (graphic tool). DBMS software can be used for non-GIS databases, and we used SQL.

In the verification phase, the model with the test data is checked. The output data is analyzed, and now this can easily be done because the physical model is self-explanatory, i.e. there is an explanation for obtaining each output. Model verification can cause general changes: smaller changes in model entities to stage 4 and larger changes starting with stage 1.

### III. RESULTS AND DISCUSSION

The main research was conducted on two varieties *Treska* and *Radika*, but here results are given from the outputs obtained from our research on the production of wheat and flour with iron and zinc of the variety *Treska*.

Input values in the model:

In<sub>1</sub>: Variants included in the research:

- Fe application in soil (variant 1)
- Fe application in soil and foliar (variant 2)
- Fe foliar application (variant 3)
- Control – without fertilizing (variant 4)
- Zn application in soil (variant 5)
- Zn application in soil and foliar (variant 6)
- Zn foliar application (variant 7)

In<sub>2</sub>: Characteristics of several types of fertilizers:

- four types of fertilizer: NPK (9:15:15), Fe EDTA, Zn EDTA, YARA VERA Amidas (40% nitrogen, 35% urea, 5% ammoniacal nitrogen, 5.5% sulphur, 14.5% other)

In<sub>3</sub>: Pre-culture:

- potatoes

In<sub>4</sub>: Soil samples, soil composition and chemical and mechanical composition of the soil

- depth of 0-30 cm, water pH (8.1), pH of nKCl (7.1), CaCO<sub>3</sub> (6.68 %), humus (3.34%), total nitrogen (0.35%), easily available mg/100g soil for P<sub>2</sub>O<sub>5</sub> (25.46) and K<sub>2</sub>O (26.65), available ppm for Fe (13.54) and Zn (1.62)

In<sub>5</sub>: Wheat variety:

- The inputs of In<sub>6</sub> are presented in Table 4.

Table 4. In<sub>6</sub>: Seed characteristics – variety *Treska*

Variant	1	2	3	4	5	6	7
Hectoliter mass (kg/hl)	76.05	76.05	76.05	75.5	76.65	76.85	76.45
Weight of 1000 grains (g)	44.2	45.9	49.6	43.9	44.38	44.08	44.73

In<sub>7</sub>: Climate conditions:

- temperature, precipitation, etc. and additional irrigation (average monthly temperatures (average 12.4 C<sup>0</sup>) and monthly precipitation amounts (total 398 mm) for the period from November 2012 to July 2013 for the Skopje region - Dolno Lisice

In<sub>8</sub>: Costs and planned revenues for obtaining fortified wheat and flour:

- cost of materials (seeds, fertilizers, soil cultivation, etc.), prices for hiring human resources and machinery (soil cultivation, sowing, harvesting, etc.), projections for planned revenues and sales prices, analysis of the market for wheat and flour and their products

Methods used in the model:

M<sub>1</sub>: Research method:

1. fertilization methods
  - 3 fertilization methods: soil, foliar and soil + foliar
2. fertilization variants
3. methods for the seven variants:
  - Fe soil, Fe soil + foliar, Fe foliar, Reference - soil fertilization, Zn soil, Zn soil +foliar, Zn foliar
4. repetitions:
  - 3
5. fortifying of wheat:
  - in 6 stages
6. varieties of wheat:
  - 2 varieties of wheat: Treska and Radika
7. types of fertilizer and amount of fertilizer:
  - 4 types of fertilizer
8. distance between parcels:
  - 50 cm
9. seed:
  - 600-650 germinable seeds/m<sup>2</sup>
10. sowing period:
  - from 19 to 20 November
11. number of parcels:
  - total: 3 repetitions \*7 variants \*2 types=42 parcels (P) or 21 parcel per type

M<sub>2</sub>: Stages of fertilization of the wheat:

- 6 stages:germination, tillering and booting, booting, booting, booting and heading, heading

M<sub>3</sub>: Statistical method - LSD test: Comparison of the mean values of the morphological characteristics of wheat with LSD test [11].

M<sub>4</sub>: Method for determination of morphological characteristics of wheat - Biometric methods for analysis of morphological and production characteristics (height of stem (cm), length of ear, etc.) [11] [12]

M<sub>5</sub>: Laboratory tests - agrochemical analysis of soil:

- The carbonate content in the soil is determined volumetrically using a Scheibler apparatus
- The humus content is determined on the basis of total carbon, using Simakov's modification of the Turin method
- The pH of the soil solution is determined electrometrically, with a glass electrode in water suspension and in nKCl suspension
- The humus content is determined on the basis of total carbon, using Simakov's modification of the Turin method
- The total N is determined according to the Kjeldahl method
- Easily accessible forms of P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O are determined according to the Al method
- The research also covers the trace elements Fe and Zn in the soil and they are determined according to the DTPA method with AAS - atomic absorption spectrometry, type Varijan Spektra AA800

M<sub>6</sub>: Classification of wheat:

- 1 class - min. 78 (kg/hl), 2 class - min. 76 (kg/hl), 3 class - min. 72 (kg/hl)

M<sub>7</sub>: Minerals in wheat and flour (ash)- Determination of the amount of ash in grains and mill products by gravimetric method:

Ash content in the wheat and flour [13a] [13b].

M<sub>8</sub>: Quality characteristics of wheat and flour - Wet gluten content, procedure for determination of wet gluten content in grain and mill products:

$$WGC = m \cdot 100$$

where WGC is wet gluten content in (%), m - amount of gluten, tests done in a farinological laboratory:



- Control index  $M_8 \text{Gluten} W = 100 * W / RF$   
where W is wheat in (%), RF is ref. value (4 var.) (some for  $M_9$ ,  $M_{10}$  and  $M_{11}$ )
  - Control index  $M_8 \text{Gluten} F = 100 * F / RF$   
where F is flour wheat in (%), RF is ref. value (4 var.) (some for  $M_9$ ,  $M_{10}$  and  $M_{11}$ )
  - $M_9$ : Quality characteristics of wheat and flour - Crude protein content by determination of total nitrogen:
    - Control index  $M_9 \text{Protein} W = 100 * W / RF$
    - Control index  $M_9 \text{Protein} F = 100 * F / RF$
  - $M_{10}$ : Quality characteristics of wheat and flour- Fat content as determined by the Soxhlet method with Diethyl ether extraction:
    - Control index  $M_{10} \text{Fat} W = 100 * W / RF$
    - Control index  $M_{10} \text{Fat} F = 100 * F / RF$
  - $M_{11}$ : Quality characteristics of wheat and flour- Concentration of iron and zinc in wheat and flour, determined with an atomic absorption spectrometer:
    - Control index  $\text{Iron} = 100 * W / RF$
    - Control index  $\text{Iron} = 100 * F / RF$
    - Control index  $\text{Zn} = 100 * W / RF$  Control index  $\text{Zn} = 100 * F / RF$
  - $M_{12}$ : Quality characteristics of wheat and flour –Farinological testing of quality characteristics of flour with Brabender farinograph [14]:
    - Determination of physical properties of wheat flour with farinograph
    - Determination of physical properties of wheat flour with extensograph
  - $M_{13}$ : Quality characteristics of wheat and flour- Moisture content (%) in wheat and flour:
    - Moisture content in flour is one of the most important indicators of quality. Flour moisture usually ranges between 12.5 and 15%
  - $M_{14}$ : Quality characteristics of wheat and flour- Total phenol content and antioxidant capacity in wheat and flour:
    - Total phenol content and antioxidant capacity (CUPRAC) of whole grain flour Obtained model outputs from 1 to 11 (showing values of several outputs).
- $Ou_5(M)$ : Content of wet gluten in grain and flour (Table 5)  
 $Ou_6(M)$ : Protein (%) content in grain and flour (Table 6)  
 $Ou_7(M)$ : Fat content in grain and flour (Table 7)

**Table 5. Content of wet gluten in grain and flour**

VARIANT	Wheat	Index from control	Flour	Index from control
1	33	106,45	34	117,24
2	35	112,90	36	124,14
3	34	109,68	35	120,69
4	31	100	29	100
5	32	103,23	33	113,79
6	34	109,68	35	120,69
7	33	106,45	34	117,24

The content of wet gluten is an important indicator of the quality of the grain because it affects the quality of the bread. Our results suggest that the greatest impact on the wet gluten content has the used fertilizer, because in the grain and the flour was measured the highest value of the wet gluten in variant 2, 3 and 6 compared with control. According to Keram at.al. [15] the application of N, P and K with doses of 120 N: 60  $P_2O_5$ : 40  $K_2O$   $kg.ha^{-1}$ , in combination with Zn as  $ZnSO_4$  with doses of 0, 1.25, 2.50, 5, 10 and 20  $kg.ha^{-1}$  during the sowing of wheat leads to increased content of wet gluten for 12,37 % with application of 20  $kg.ha^{-1}$   $ZnSO_4$  compared to control and other variants.

**Table 6. Protein content in grain and flour**

VARIANT	Wheat	Index from control	Flour	Index from control
1	13,00	107,52	12,5	110,89
2	11,85	98,02	11,29	100

3	11,85	98,02	10,77	95,4
4	12,09	100	11,29	100
5	10,72	88,67	11,29	100
6	12,09	100	12,05	106,73
7	11,61	96,03	11,81	104,6

The analysis of the protein content has scientific and practical significance. Spare proteins are located in the endosperm in cereal crops. The results shown in Table 6, show some differences that result not only from hereditary traits but also on climatic factors and applied fertilizers. The highest protein content in flour exists in variant 1 by 10.89% in terms of control. In this case, the effect of Fe EDTA as a chelating compound stabilizing metal ions (in this case iron) is confirmed, and protecting them from oxidation and deposition, leading to an increase in the percentage of proteins in variant 1. Also, Abbas et al. [16] concluded that the soil application of Fe at doses of 0, 4, 8, 12 and 12 kg.ha<sup>-1</sup> in the form of FeSO<sub>4</sub> leads to an increase in the percentage of proteins and an increase in Fe concentration in wheat. According to Habib (2009) [17] application of 150 g.ha<sup>-1</sup> iron in the form of Fe<sub>2</sub>O<sub>3</sub>, increases iron content in the grain leads to an increase in the percentage of proteins.

**Table 7. Fat content in grain and flour**

VARIANT	Wheat	Index from control	Flour	Index from control
1	1,38	115	1,02	40,48
2	1,57	130,83	2,23	88,49
3	1,06	88,33	1,02	40,48
4	1,2	100	2,52	100
5	1,73	144,16	1,07	42,46
6	1,12	93,33	1,09	43,26
7	1,53	127,5	2,26	89,68

Wheat flour contains a certain amount of fat, which has a beneficial effect on the technological quality of the flour, improving the interaction of the flour starch and gluten [17]. Analyzes of % of fats have generally shown that soil application of zinc and soil + foliar application iron results in a significant increase % of fat in the grains while the flour in all variants % of fat is reduced compared to control. It can be concluded that the application of Zn and Fe EDTA chelating fertilizers have no effect on the percentage of fat in flour.

Ou<sub>8</sub>(M): Concentration of iron and zinc (Table 8)

**Table 8. Concentration of iron and zinc**

Variant	Fe (mg.kg <sup>-1</sup> )		Zn(mg/kg <sup>-1</sup> )	
	Wheat	Flour	Wheat	Flour
1	40.6	28.9	-	-
2	47.1	28.7	-	-
3	65.4	24.2	-	-
4	44.8	13.2	26.6	10.8
5	-	-	27.1	11.9
6	-	-	26.6	17.9
7	-	-	33.7	26.1

In analyzing the presented results in Table 8 it may be stated that the biggest increase of Fe in wheat was obtained with the variants 3 and 2 while it was decreased for -10,6 % with the variant 1 in relation to the variant 4\* control, but they are higher with 118,9% in the flour. Also from among the obtained results it is established the highest concentration of Zn in wheat and flour obtained by foliar fertilization of 26.83% as compared to the control.

All of the plants, nevertheless, firstly absorb the iron from the soil in a form of Fe (III) which then transforms into Fe (II). By means of specific transporters, the mineral substances firstly absorb in the root and then, in the complexes such as nicotinamide, the metals are transported and mobilized in other parts of the plants [18]. Also according to the bio-fortification in the majority cases is an expensive strategy and its efficiency is often limited by the soil conditions [19]. As a alternative solution, it is suggested a nutrition with nitrogen which may have a significant influence on the re-translation of Fe and Zn in the cereal crops. It is supposed that the



nutrition with nitrogen has a positive effect since it is needed for the biosynthesis of nicotianamine (NA) [20]. The additional provision of nitrogen leads to improvement of the accumulation of Fe and Zn in the wheat grains  $Ou_{11}(M)$ : Total phenol content and antioxidant capacity of wheat (Table 9)

**Table 9. Total phenol content and antioxidant capacity of wheat**

Variant	Total phenol		CUPRAC (mmol trolox equivalents/g DW)	
	Wheat	Flour	Wheat	Flour
1	1,29	0,79	5,48	2,10
2	0,99	0,75	4,58	1,79
3	0,92	0,89	5,11	2,18
4	0,92	0,82	3,32	1,90
5	0,99	0,86	4,03	2,19
6	1,12	0,82	4,68	2,17
7	1,01	0,92	4,51	2,09

From the results present in Table 9 we can concluded that the concentration of TP in wheat samples was higher than the flour samples. In whole wheat grain samples the content of TP was increased after treatment with Fe and Zn fertilizers. After the Fe and Zn fertilizers treatment TP content in flour samples compared to the control group was increased but not significantly. The CUPRAC values obtained from wheat grains measurement varies from 3.32 and 5.48  $\mu\text{mol trolox equivalents/g DW}$ . The CUPRAC values in wheat flour shows lover values that range between 1.79-2.19  $\mu\text{mol trolox equivalents/g DW}$ . The Fe-chelates and Zn-chelates fertilizers applied on soft wheat in different ways have influence on the TP content improvement and also improve antioxidant activity.

First, the results obtained through the model outputs ( $Ou_9$ ) showed a significant enrichment of the zinc and iron concentrations in wheat and flour using soil-foliar biofortification methods. Second, through the self-explanatory model, users of the model can obtain information about each output as it is received. The concept a self-explanatory model is the future of creating other models in this and other areas.

#### IV. CONCLUSION

Biofortification, unlike regular fortification, focuses on the internal enrichment with micronutrients in plant parts used for food while the plants are in the growth phase, rather than adding nutrients to the food when it is processed. Agronomic biofertilization imposes as an alternative cost-efficient solution nowadays, which increases the productivity and concentration of micronutrients in grain and flour, and this will contribute to improving human health. In this paper, it is concluded that agronomic biofortification with zinc and iron leads to improved nutritional characteristics of wheat and flour.

A novelty in this paper is the concept of modeling to create a model for enrichment of wheat with zinc and iron through soil-foliar biofortification which includes self-explanation. The use of advanced scientific methodologies and advanced technologies such as GIS, DBMS and advanced data analysis is important in model building. The concept of modeling, which is presented in five stages in which all important functions are defined as entities in the E-R model, will allow raising the models to a higher level, and specifically for the model presented in the paper, the incorporation of the so-called self-explanation of the modeling process itself, will enable its better theoretical and applied use, but also easier further development.

The benefit of enriching wheat with zinc and iron through soil-foliar biofertilization can be threefold: benefits for companies through the development of new food products that would satisfy consumers, and thus obtaining greater profit, benefits for the people through the consumption of healthier and safer food products and benefits for the state.

Further research can continue to include new determinants that can significantly affect the model.

#### REFERENCES

- [1]. Mutch, D., Wahli, W., Williamson, G., Nutrigenomics and nutrigenetics: the emerging faces of nutrition. *FASEB Journal*, 19 (12), 2005, 1602-16. DOI: <https://doi.org/10.1096/fj.05-3911rev>
- [2]. Roberts, M., Mutch, D., German, J., Genomics: food and nutrition. *Current Opinion on Biotechnol*, 12, 2001, pp. 516-522.
- [3]. Newell-McGloughlin, M., Nutritionally improved agricultural crops. *Plant Physiology*, 147, 2008, pp. 939-53. DOI: <https://doi.org/10.1104/pp.108.121947>
- [4]. White, P., Broadley, M., Bio-fortifying crops with essential mineral elements. *Trends in Plant Science*, 10, 2005, pp. 586-593.
- [5]. Welch, R., Graham, R., Breeding crops for enhanced micronutrient content. *Plant and Soil*, 245, 2009, pp. 205-214.

- [6]. Bayram, M., Korkut, K., Effect of the glutenin genes on quality parameters in common wheat. *Journal of Central European Agriculture*, 21 (1), 2020, 62-76. DOI: <https://doi.org/10.5513/JCEA01/21.1.2422>
- [7]. Dăcu, A-D., Ianovici, N., Sala, F., A method for estimating nitrogen supply index in crop plants: case study on wheat. *Journal of Central European Agriculture*, 21 (3), 2020, 569-576. DOI: <https://doi.org/10.5513/JCEA01/21.3.2760>
- [8]. Hotz, C., Brown, K., Assessment of the risk of zinc deficiency in populations and options for its control. *Food Nutritional Bulletin*, 25, 2004, pp. 94-204.
- [9]. Mladenov, M., Penchev, S., Deyanov, M., Complex assessment of food products quality using analysis of visual images, spectrophotometric and hyperspectral characteristics. *International Journal of Engineering and Innovative Technology IJEIT*, 4 (12), 2015, 23-32.
- [10]. Martinovski, S., GIS modelling for strategic planning of the urban environment. Lambert Academic Publishing, (2017).
- [11]. Menkinoska, Gjorgoski I., Manasievska-Simic S., Pavlova V., Popeska Z., Stanoev V., Blazevska T., Poposka H., Gjorgovska N., Levkov V., The Influence of Fe-chelates Fertilizers on the Soft Wheat's Quality *International Journal of Emerging Technology and Advanced Engineering* Volume 4, Issue 9, 2014.
- [12]. Menkinoska M., Blazevska T., Pavlova V., Improving the quality of soft wheat through biofortification with Zn chelating fertilizers The teacher of the future. Ninth International Scientific Conference, Durres, Republic of Albania. *International journal* Vol.13,3, 2016, pp 335
- [13]. Menkinoska M, Stanoev V, Pavlova V., Blazevska T., Stamatovska V., Gjorgovska N., Determination of some physical-chemical properties of biofortification soft wheat and flour of the varieties "Treska" Original scientific paper *Journal of Engineering & Processing Management* Vol.9 No.1 DOI 10.7251/JEPM1709033M UDC 664. 2017, 641.12
- [14]. Menkinoska M, Pavlova V., Blazhevska T., Gjorgovska N., Stamatovska V., Stanoev V., Qualitative properties of zinc bio-fortified soft wheat and flour, *Proceedings of 12<sup>th</sup> symposium, novel technologies, and economic development*, 34-41, Leskovac, 2017, 20-21 ISBN 978-86-89429-25-1.
- [15]. Menkinoska M., Blazhevska T., Stamatovska V., Comparison of the rheological characteristics of bio-fortified flour obtained from soft wheat varieties Treska and Radika *JOURNAL OF AGRICULTURE AND PLANT SCIENCES, JAPS*, vol 17, no. 1, UDC: 664.641.12.017 Original scientific paper In print: ISSN 2545-4447 On line: ISSN 2545-4455, 2019.
- [16]. K. S. Keram, B. L. Sharma and S. D. Sawarkar, Impact of Zn application on yield, quality, nutrients uptake and soil fertility in a medium deep black soil (vertisol). *International Journal of Science, Environment and Technology*, Vol. 1, No 5, 2012 , 563-571
- [17]. Abbas G., M. Q. Khan, M. J. Khan, F. Hussain and I. Hussain, Effect of iron on the growth and yield contributing parameters of wheat (*triticum aestivum* L.). *The Journal of Animal & Plant Sciences* 19(3): Pages: 135-139 ISSN: 1018- 7081 139, 2009.
- [18]. Habib, M., Effect of foliar application of Zn and Fe on wheat yield and quality. *Afr. J. Biotechnol.*, 8, 6795-6798, 2009.
- [19]. Grotz N, Guerinet ML., Molecular aspects of Cu, Fe and Zn homeostasis in plants. *Biochim. Biophys. Acta* 1763:595-608, 2006.
- [20]. White P. J., Broadley M. R., Biofortification of crops with seven mineral elements often lacking in human diets - iron, zinc, copper, calcium, magnesium, selenium and iodine. *New Phytol.* 182, 49-84.10.1111/j.1469-8137.2008.02738, 2009.
- [21]. Krüger C., Berkowitz O., Stephan U. W., Hell R., A metal-binding member of the late embryogenesis abundant protein family transports iron in the phloem of *Ricinus communis* L. *J. Biol. Chem.* 277, 25062-25069.10.1074/jbc.M201896200, 2002,