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The quality of maize grains in organic farming system

COSMIN ŞONEA¹, MARIA TOADER*¹, IONELA PAULA NĂSTASE¹

¹University of Agronomic Sciences and Veterinary Medicine of Bucharest, Romania

Abstract

The paper present the quality of organic maize grains came from some Romanian organic farms, in 2016-2018 period. The quality analysis were performed in authorized laboratories to determine: Dry Matter and Moisture (DM), Carbohydrates (C), Crude Proteins (CP), Crude Fat (CF), Ash, Crude Fibers (CFB). Also, it has been calculated the Nutritional Unit (NU) and Gross Energy (GE) in kcal or MJ, Digestible Protein (DP), Non-Nitrogenous Extract Substances (NES) and Organic Substances (OS). The analysis included detection of harmful components on health of final consumer: pesticides residues, mycotoxins, nitrates and nitrites. The results are: 86.14% – DM; 13.86% – Moisture; 68.36% – C; 10.18% CP; 4.26% – CF; 1.36% – Ash; 2.39% – CFB; 1.25 – UN; 378.77 kcal or 1.58 MJ – GE, 65.68% – DP, 67.15% – NES and 83.98% – OS. Pesticides residues not find in any sample. Mycotoxins were found in only two farms, in 2016: Aflatoxin B1 (1.7-2 µg kg⁻¹) and Aflatoxin B1+B2+G1+G2 (2-3.7 µg kg⁻¹), Fumosin B1+B2 (786-992 µg kg⁻¹), Deoxynivalenol (DON) (0.7 -26 µg kg⁻¹); Zearlenon (1.2 µg kg⁻¹). In 2017 and 2018 years, these substances were not detected. For nitrates and nitrites, the results showed only nitrates in 2016, in tree farms (5.1-6 mg kg⁻¹). The micotoxin and nitrates values were below the limit allowed by the legislation in force. Following these results it can be appreciated that organic maize has good quality similar with EU standards and farmers respect the principles of organic agriculture.

Keywords

Organic maize, quality, pesticides residues, mycotoxins, nitrates and nitrites.

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✉ *Corresponding author: MARIA TOADER, University of Agronomic Sciences and Veterinary Medicine of Bucharest, Romania
E-mail: mirelatoadervali@yahoo.com

Introduction

Maize is one of the most important crops, with 193.7 million ha area harvested in the world, in 2018 year (OECD/FAO [39]). Maize grains are widely used in human nutrition, industry and animal feed (MATEI [32]).

In 2016-2018, approximately 1.7 billion tons of agricultural commodities were used as livestock feed, mainly maize and other cereals, and protein meals derived from oilseeds (OECD/FAO [39]). Around 13% of the total feed intake consisted of cereals, a value which corresponded to almost one-third of global cereal production. In this context, the maize grain is a major feed grain and a standard component of livestock diets where it is used as a source of energy (OECD/FAO [39]).

Maize is the main source of concentrates used in the feeding of animals as pigs, cattle, sheep representing up to 80% of the total concentrates, in birds up to 70% of the concentrates, and in the other categories up to 30% (youth from all species and working animals) (NICOLAE, SONEA [37]). Also, maize is one of the most versatile emerging crops having wider adaptability under varied agro-climatic conditions (KLING [30]).

On the other hand, organic production is an overall system of farm management and food production that contributes to the preservation of natural resources and applies high standards of animal welfare (WILLER & al [46]). The EU surface reached 12.6 million ha in 2017, which represents 18% of the global organic area and 7% of total EU agricultural land. Further organic area is devoted to green fodder (17%), cereals (16%) and permanent crops (11%). Regarding organic livestock, in EU, 4.85 million bovines, 5.9 million sheep and goats, almost 1.4 million pigs, and 56.5 million poultry were registered (WILLER & al [46]).

For Romania, in 2018, organic farming means about 327 thousand ha and 9008 farmers (MARD [33]). Among the crops, cereals with about 115 thousand predominate, followed by industrial crops with over 80 thousand ha. In Romania, according to the data provided by Eurostat, in 2016, there were registered over 56 thousand ha of organic maize, respectively 48% from total organic area with cereals (EUROSTAT [7]).

Organic animals must ensure their nutritional needs in good quality organic feed and in a form that allows them to manifest their natural behavior (IFOAM [21]).

The aim of this research was to study the quality and nutritional value of maize grains produced in organic farming and which could be used for human consumption, but especially, in organic animal feed. This quality analysis included both the determination of the chemical composition of maize grains and whether there are possible contaminations with certain substances such as pesticide residues, mycotoxins, nitrates and nitrites that may endanger the health of animals and consumers. Among the arguments for the importance of this research is the fact

that animals fed with certified organic feed benefit from a balanced diet, without chemicals, without additives, preservatives or synthetic dyes, without genetically modified organisms and without pesticide residues. In this way, the animals produce better quality of milk or/and meat, eggs, in which the animal welfare standards are met. In addition, the positive effects without environmental pollution are particularly important for both humans and animals. Thus, organic farms, through clean technologies for plants and animals, without contaminating water sources, habitats, soil, can create long-term sustainability. Also, the quality of organic maize grains may also depend on the conditions of cultivation, harvesting and storage. In conditions of high humidity, with many foreign bodies, mold fungi can produce mycotoxins, with risk to health of human and animals. Nitrate pollution is a current problem that affects the quality of agricultural products and thus animal feed, which can complete the overall quality of maize grains produced in organic system. Accordingly, investigations should be made into the sources of formation of these substances and identifying the administrative measures to be taken to prevent, as far as possible, their occurrence.

Materials and Methods

The design of the experiment

The research was conducted during 2016-2018 period. The analyzed samples came from four organic certified farms in Romania, from different agricultural areas: Vaslui (Vetrisoia village, Moldova Plateau), Teleorman (Merenii de Jos village, Glavacioc Valley), Arad (Pecica village, Mures Valley) and Bihor (Santimreu village, Barcau Valley). These farms were chosen because they manage large areas cultivated with maize (over 100 ha) in organic system, apply innovative cultivation technologies regarding fertiliser system and control of pest and diseases and selling the maize production grains on the Romanian market. Also, the farms have, for the most part, the same characteristics of the climate: temperate-continental climate, with maximum temperature value in July-August, in last years of 34.7°C and minimum value in January of -18.5°C and very uneven rainfall regime between 450 and 550 mm, dry periods alternating with the rainy ones, with a increasing frequency of torrential rains.

Regarding the suitability of soils for maize crop, the farms have about the same types of soil, cambic chernozem, except for the farm from Vaslui county, where the soil type was the podzolic soil (Moldova Plateau).

In terms of maize cultivation technology, it was the same in all four farms by an agreement with farmers. It was a classic technology, which included the main technological links. Thus, after harvesting the previous plant, which for each year was wheat, a plowing was carried out in August, followed by soil disking in September. In March, there was a disc harrow and then preparing the bed germinating. Prior to the preparation of the germination bed, some products were applied that stimulate the mechanisms of inducing the plants resistance against the attack of pathogens, help to mineralize the organic

substances in the soil and to improve the root system of plants. These products are: Biofos soft phosphate rock (contains phosphorus, magnesium, calcium, iron) – 250 kg ha⁻¹; Fielder + Biofertilizer (contains *Trichoderma asperellum*) – 250-750 g ha⁻¹; P soil (containing *Bacillus megaterium*) – 2 l ha⁻¹. The maize seeds were treated with Raykat Enraizador Start product – 1 l t⁻¹ of seed (contains amino acids, polysaccharides, total nitrogen and trace elements), for strong stimulation of root development and increase of nutrients absorption, especially in the germination phase. These products correspond to Annex II of the Commission Regulation (EC) No 889/2008 [14] laying down detailed rules for the implementation of Council Regulation (EC) No 834/2007 [12] on organic production. Weed control was performed by two mechanical weeding in May and one mechanical weeding in early of June. The hybrid used for sowing was DK5182, FAO group 400-450, with good tolerance to drought, heat and break and high capacity of adaptability in different growing conditions.

The density was 72,000 germinating grains ha⁻¹, the distance between rows was 70 cm, at sowing depth of 4-5 cm. The sowing was carried out in the optimal period, April 20-25, for each year of experimentation and was harvested by combine, between September 15-20. The yields productions varied between 6,300-6,800 kg ha⁻¹.

Chemical analysis and nutrition value

The chemical analysis was validated in accordance with the Quality Assurance System, ISO 17025/2005, and SANTE/11813/2017 Standards (ISO 17025 [27]; SANTE 11813 [42]). Sampling for chemical analysis was performed in accordance with the requirements of ISO 17065/2013 – Conformity Assessment, respectively, Requirements for bodies certifying products, processes and services [26] and with the requirements of Regulations (EC) 834/2007 [12] and Regulations (EC) 889/2008 [14] specifics to organic agriculture, in accredited laboratories from: Czech Republic for the various biochemical compounds (dry matter and water (moisture), carbohydrates, crude protein, crude fat, ash, crude fibers, gross energy in kcal and MJ); Germany for pesticides, mycotoxins and nitrate and nitrite residues. Samples were identified clearly and indelibly, in a way to ensure the traceability.

The results are presented by comparison with reference values of INRA (L'Institut National de Recherche pour l'Agriculture, France) standards Feeds (the most used International Standard) [23] and Order No. 369 of June 27/2019 of Romanian Ministry of Agriculture and Rural Development (MARD [34]).

Determination of Dry Matter (DM%) by Gravimetric method was in according with ISO 712/1998 [24] standards. Grains Moisture is the amount of water, expressed as a percentage that is removed from a grains sample by drying in a drying stove, at a certain temperature to the constant weight of the sample (ISO 712 [24]).

Method used for determination of Carbohydrates (C%) was High Performance Liquid Chromatography (HPLC), with refractive index detection (RID). Operational parameters of chromatographic system were: column: modified Alltima Amino 100Å, 5 µm, 250 x 4.6 mm;

mobile phase flow: 1.3 ml/min; mobile phase: acetonitrile/water (75/25; v/v); column temperature: 30°C; injection volume: 20 µl; separation time: 60 min (JOHANSEN & al [29]).

According to ISO 16634-2/2009 the crude protein (CP%), calculated as N (nitrogen) x 6.25, where N is the nitrogen obtained by mineralisation through the Dumas methods (ISO 16634 [25]).

The Crude Fat (CF%) content was determined by gravimetric methods according to AOAC 945.38F; 920.39C Gravimetric (ether extraction) (AOAC [1]).

The Ash content (%) of foods was determined by gravimetric methods, according to AOAC 923.03. The maize grains were burnt at 550°C to constant weight and the ash was determined by weighing (AOAC [1]).

The enzymatically method was used for determination of Crude Fibres (CFB%) content. It used the commercial set Megazym according to AOAC Method 985.29 and 991.43 (AOAC [1]).

Nutritional Units (NU) and Digestible Protein (DP) was made according to Order no. 369 of June 27/2019 of Romanian Ministry of Agriculture and Rural Development for approving the nutritional composition of the fodder used to feed cows, buffaloes, sheep and goats, as well as their nutritional requirements. Maize has the following nutritional composition: 866 DM g kg⁻¹, 1.25 Nutritional Units (NU) and 66 g kg⁻¹ Digestible Protein (DP%) (MARD [34]). The results obtained in these experiments were reported at these values.

Indirectly, the Gross Energy (GE) (kcal or MJ) can be determined by knowing the crude content of nutrients and the caloric coefficients of them (FAO/WHO [18]). The caloric coefficients were following: 4.1 kcal g⁻¹ for Carbohydrates (C), 5.7 kcal g⁻¹ for Crude Protein (CP) and 9.5 kcal g⁻¹ for Crude Fat (CF). The Gross Energy was calculated according to formula: GE=4.1*C%+5.7*CP%+9.5*CF%, where, GE – Gross Energy (kcal kg⁻¹); C – Carbohydrates (%), CP – Crude Protein (%), CF – Crude Fat (%) and 1 kilocalorie = 0.004184 Megajoules (FAO/WHO [18]).

Organic substances (OS) are synthesized by plants following the process of photosynthesis and represent the main component of feed. The organic substances (proteins, lipids, carbohydrates and non-nitrogenous extractive substances) in the fodder during their calcinations at 550°C are destroyed, as a result they can be calculated as the difference between Dry Matter (DM) (%) and Ash (%): OS% = DM% – Ash% (AVARVAREI [3]).

Non-nitrogenous extractive substances (NES) (%) are heterogeneous substances represented mainly by starch, soluble sugars, pectin, tannins, etc. This group could be determined by formula: NES% = OS% - (CB% + CF% + CFB%), where OS – Organic Substance (%), CP – Crude Protein (%), CF – Crude Fat (%); CFB – Crude Fiber (%) (AVARVAREI [3]).

The tested pesticides were over 250 of types. In our table we will present only 5 groups: glyphosat, glufonisat, acid aminomethylphosfonic, clormequat and mepiquat.

The using method was according the EU Standard-SANTE/11945/2015 (SANTE 11945 [40]). Determination of residues of pesticides was made by GC-MS and/or LC-MS/MS after extraction/partition with acetonitrile and purification by the QuEChERS (Quick, Easy, Cheap, Efficient, Rugged and Safe) dispersive method. QuEChERS extraction was performed according to the official AOAC method 2007.01 using Waters DisQuE™ Dispersive Solid Phase Extraction (d-SPE) product (AOAC [2]).

The method of mycotoxins analysis was the High Performance Liquid Chromatographic (HPLC) with UV (Ultraviolet) detection (Photodiode Array (PDA) detector), with purification on the immunoaffinity column (FCC [20]). The analysed mycotoxins were: B1 Aflatoxin, B1+B2+G1+G2 Aflatoxin complex, B2 Fumonisin, Deoxynivalenon (DON, vomitoxin) and Zearalenone.

For determination of nitrates and nitrites contents, the laboratory has developed and accredited its own method. High Performance Liquid Chromatography (HPLC) with UV (Ultraviolet) detector was used. The UV-Vis (Ultraviolet Visible) photodiode detector detected nitrates and nitrites ions at an absorption of 212 nm (FCC [20]).

All chemical analyses were performed in three replicates and the results were statistically analysed by the Fisher's least significant differences (LSD) test.

Results and Discussion

Chemical composition and nutrition value

The moisture content of the maize grains is a very important criterion for evaluating its quality from several points of view. Maize grains, according to the reference value (INRA [24]), must be a maximum of 14%. The storage of maize depends largely on its moisture. If the humidity exceeds 14%, a series of chemical processes related to the acceleration of respiration with heat and water production appear, followed by complex fermentation processes that lead to the alteration of the grain mass (TOADER & al [44]). The high moisture content during storage encourages the appearance of certain yeasts, molds and harmful bacteria.

Our research showed that for all samples, the average moisture values was 13.86%. It can be noticed, the samples that came from Vaslui county, that for both 2017, exceed by 1.6%, the reference value. The dry matter (DM) resulted from the difference from 100 minus moisture. These ranged in 2016-2018 period from 85.2% for farm from Vaslui to 87.8% of farm from Bihor county. The best value of moisture (12.2%) was registered in 2017, by the farm from Bihor, which exceeded the average with a distinctly significant value (Table 2).

Maize grains are generally known to be high in carbohydrate. Marta et al, in 2017, founded the content of 69.6-75.68%, for different hybrids (MARTA et al, 2017). In these research, carbohydrates had an average value of 68.38% for all four locations which expresses a value very close to reference value (69%). The value of

70.4 from 2017, obtained by the farm from Bihor county, stands out. This exceeded the experience average by 2%, which meant a very distinctly significant value compared to the average. Compared to the reference value, the value obtained for organic corn was slightly lower, by about 0.5%.

The minimum protein content of maize grains according to reference value is 8.9%. Toader at all found the protein content between 10.13 and 13.27% on organic maize grains in Fundulea Experimental Field (TOADER at al, 2017). In 2011, Imbrea and al at the Research of Experimental Field in Timisoara, found a protein content of maize around 8.2% (average value) (Imbrea et al, 2011). By comparisons with INRA standards our results are over this. In 2016, protein content varied between 9.3% in Vaslui are and 11% in Bihor county. In all the years of experimentation, the contents of the maize from Bihor County significantly exceeded the average per experiment, but also the reference value. It can also be seen that in all cases, the reference value has been exceeded. As a result, it can be highlighted that the maize obtained in organic farming conditions has a good quality in terms of protein content, being comparable to the values known for toothed maize hybrids.

In research of Suriady at all, crude fat content ranged from 2.48% up to 4.80% (SURIADY at al, 2017). The INRA standards recommends a content of 4.1%. In our research, the values of CF were slightly above the standard value, respectively between 4.0% and 4.7%. The value of 4.7, the best value, exceeded the average very distinctly, and also exceeded the reference value. This value was obtained by the farm in Teleorman, in 2018. The farm in Bihor County also obtained a good value, 4.5%, which significantly exceeded the average crude fat (CF) content.

Crude Fibre (CFB) component is one of the most important nutritional and technological factors of the maize grains. The content is directly related to digestibility, so the more feed cellulose contains, the lower the digestibility of the organic matter and vice versa. As is known, the fibre content of maize grains is between 1.4% - 3%, depending on the hybrid and growing conditions. The crude fibre of maize based on study of BeMiller at all was 1.11-4.15% (BEMILLER & al [4]) and INRA standards recommended a content of 2.6%. During this experiment the CFB oscilated on 1.8 and 2.8%, this means an average of 2.3%.

Maize grains are the basic concentrated feed for all animal species and the nutritional value according Romanian legislation is 1.25 Nutritional Units per 1 kg of grain (MARD [34]). In this research, the Nutritional Unit (NU) and Gross Energy (GE) of organic maize were in average: 1.25 NU kg⁻¹ of grains, respectively 378.88 kcal or 1.58 MJ. The value from 2018 can be highlighted, when it exceeded the average by over 10%. Among the locations, the farm from Bihor County led to obtaining values higher (390.3 kcal or 1.63 MJ), in 2016, than the reference value

(372.6 kcal or 1.56 MJ), both for 2017 and 2018 (around 381 kcal or 1.59 MJ).

In case of these experiments, the digestible protein (DP) was approximately equal to reference value, which means that organic maize grains can provide rations corresponding to the needs of animal growth and development depending on the category for which the feed is produced. The value from Bihor County is highlighted again, with 66.9% for 2017, when it significantly exceeded the average per experiment.

In the case of the other two determinations, both NES and OS values has been below the reference value.

However, the organic maize samples from Telorman and Bihor Counties were noticed, with values that exceeded the average by 1-2%. The results of 2016-2018 period showed the following values: NES%: 64.4-69.4% and OS%: 82.5-82.7%. These values demonstrate that in the optimal rations of the different categories of animals must be supplemented with other types of feed, which cover the necessary requirements of the reference value.

All results regarding the chemical composition and nutritional value and statistical processing are included in Table 1 and Table 2. The level of significance (DSL5%, 1%, 0.1%) from Table 1 has based the values of Table 2.

Table 1. Results of chemical composition and nutritional value for organic maize in 2016-2018 period

Index quality	Farm 1 (Vaslui county)	Farm 2 (Arad county)	Farm 3 (Teleorman county)	Farm 4 (Bihor county)	Average	References values of INRA and Romanian Standards
2016						
Dry matter (DM) (%)	84.7**	86.7	87.1 ^o	86.6	86.28	86.6
Moisture (%)	15.3 ^{oo}	13.3	12.9*	13.4	13.7	13.7
Carbohydrats (C) (%)	68.1	68.1	68.8	70.4***	68.9	69
Crude protein (CP) (%)	9.3 ^{oo}	10	10.1	11*	10.1	8.9
Crude fat (CF) (%)	4.2	4.5*	4.4	4.1	4.3	4.1
Crude fibre (CFB) (%)	2.6	2.8*	2.2	2,0 ^o	2.4	2.6
Ash (%)	1.7***	1.3	1.6**	1.4	1.5	1.2
Nutritional Unit (UN) (g/kg)	1.23	1.26*	1.26*	1.25	1.25	1.25
Gross Energy (kcal)	372.1 ^o	379.0	381.5	390.3***	380.7	372.6
Gross Energy (MJ)	1.56 ^o	1.58	1.59	1.63***	1.59	1.56
Digestible Protein (DP) (g/kg)	64.55 ^{oo}	66.38*	66.38*	66.00	65.83	66
Non-Nitrogenous Extractive Substances (NES) (%)	66.9	66.6	68.2	67.5	67.3	74.4
Organic Substances (OS) (%)	83.0 ^o	83.9	84.9*	84.6	84.10	87.1
2017						
Dry matter (DM) (%)	85.7	86.8	85.4	87.8 ^{oo}	86.4	86.6
Moisture (%)	14.3	13.2	14.6	12.2**	13.6	13.7
Carbohydrats (C) (%)	67.8	68.2	68.0	68.3	68.1	69
Crude protein (CP) (%)	9.1 ^o	10.8*	9.8	10.9*	10.2	8.9
Crude fat (CF) (%)	4.1	4.0 ^o	4.2	4.1	4.1	4.1
Crude fibre (CFB) (%)	1.8	2.1	2.2	2.1	2.1	2.6
Ash (%)	1.3	1.3	1.2 ^o	1.4	1.3	1.2
Nutritional Unit (UN) (g/kg)	1.24	1.26*	1.24	1.27**	1.25	1.25
Gross Energy (kcal)	368.8 ^{oo}	379.2	374.6	381.1	375.9	372.6
Gross Energy (MJ)	1.54 ^{oo}	1.58	1.57	1.59	1.6	1.56
Digestible Protein (DP) %)	65.31	66.15	65.09	66.91**	65.9	66
Non-Nitrogenous Extractive Substances (NES) (%)	69.4**	67.8	67.0	68.6*	68.2	74.4
Organic Substances (OS) (%)	84.4	84.7	83.2	85.7***	84.5	87.1
2018						
Dry matter (DM) (%)	85.20*	86.70	85.70	85.30*	85.7	86.6
Moisture (%)	14.80 ^o	13.30	14.30	14.70 ^o	14.28	13.7
Carbohydrats (C) (%)	68.40	68.30	68.40	67.50 ^o	68.2	69
Crude protein (CP) (%)	9.80	10.80*	9.80	10.80*	10.3	8.9
Crude fat (CF) (%)	4.00	4.30	4.70***	4.50*	4.4	4.1
Crude fibre (CFB) (%)	2.60	2.80*	2.70	2.80*	2.7	2.6
Ash (%)	1.20 ^o	1.20 ^o	1.50	1.20 ^o	1.3	1.2
Nutritional Unit (UN) (g/kg)	1.23	1.26*	1.24	1.24	1.24	1.25
Gross Energy (kcal)	374.30	382.44	380.95	381.06	379.7	372.6
Gross Energy (MJ)	1.56	1.60	1.59	1.59	1.59	1.56
Digestible Protein (DP) %)	64.93	66.08	65.31	65.01 ^o	65.3	66
Non-Nitrogenous Extractive Substances (NES) (%)	67.60	66.00	65.80 ^o	64.40 ^{ooo}	66.0	74.4
Organic Substances (OS) (%)	84.00	83.90	83.00 ^o	82.50 ^{oo}	83.4	87.1

Table 2. Statistical results of chemical composition and nutritional value for organic maize in 2016-2018 period

Statistical Index	DM (%)	Moisture (%)	C (%)	CP (%)	CF (%)	CFB (%)	Ash (%)	UN (g/kg)	GE (kcal)	GE (MJ)	DP (%)	NEF (%)	OS (%)
Average	86.14	13.86	68.36	10.18	4.26	2.39	1.36	1.25	378.77	1.58	65.68	67.15	83.98
Sample variance	0.87	0.87	0.52	0.43	0.05	0.13	0.03	0.00	31.85	0.00	0.54	1.83	0.874
Standard deviation	0.93	0.93	0.72	0.66	0.22	0.36	0.17	0.01	5.64	0.02	0.73	1.35	0.935
Sd	0.38	0.38	0.29	0.27	0.09	0.15	0.07	0.01	2.30	0.01	0.30	0.55	0.382
CV%	1.08	6.72	1.05	6.46	5.25	15.07	12.34	1.08	1.49	1.49	1.11	2.01	1.113
DSL 5%	0.84	0.84	0.65	0.59	0.20	0.32	0.15	0.01	5.07	0.02	0.66	1.21	0.840
DSL 1%	1.18	1.18	0.91	0.83	0.28	0.46	0.21	0.02	7.17	0.03	0.93	1.72	1.187
DSL 0.1%	1.69	1.69	1.31	1.19	0.41	0.65	0.30	0.02	10.23	0.04	1.33	2.45	1.695

Results of pesticides residues

Establishing that a plant protection product poses a low risk to human and animal health is of paramount importance in order to successfully register and market the product on a global basis (MORGERA & al [35]).

Restrictions on the use of chemicals or other substances are a key requirement for organic production methods (ZANG and BERKELEY [45]). Residue testing can provide evidence, when there is uncertainty, about the use of unauthorized substances, such as banned pesticides, genetically modified organisms, food additives or medicinal substances. Residue testing is one of the tools available to control bodies to ensure that organic production rules, as specified in the various regulations, are in line with the principles of organic farming (IFOAM [21]).

The applicable regulations do not provide for a minimum number of laboratory tests, but only the obligation to perform these tests when there are suspicions that unauthorized products have been used in organic production or the production is marketed. Traceability is mentioned as one of the important elements in ensuring consumer.

It makes it possible to verify whether all operators involved in all stages of production, preparation and distribution apply the EU requirements for organic production (REG. (EC) 834 [12]).

When non-compliance is identified, traceability makes it possible to identify the source and isolate the

problem, thus preventing the products in question from reaching consumers. At EU level, no criteria are set for pesticides to be included in these checks or the sensitivity of the methods (EU COMMISSION [13]).

Maximum pesticide residue limits (MRLs) that specify the maximum concentration of a pesticide in conventional products have been regulated by many countries in order to promote good agricultural practice guidelines (GAPs) (EU COMMISSION [13]).

Pesticide residue testing is an aspect of official controls on organic production (SANTE 8986/2006 [41]). The control authorities or control bodies must take and analyse samples for detecting products not authorised for organic production, for checking production techniques not allowed under organic production rules or for detecting possible contamination by products not authorised for organic production (SANTE 8986/2006 [41]). The number of samples to be taken and analysed by the control authority or designated control body every year shall correspond to at least 5% of the number of operators under its control (REGULATION (EC) NO 889 [14]).

In the case of our research, as shown in the following table, was not detected any pesticide residue in any sample of organic maize. These results confirm that farmers comply with the rules and principles of organic farming and only products that meet the standards approved by the legislation in force are used in maize cultivation technology.

Table 3. Results of pesticides residues for organic maize grains (mg kg⁻¹), in 2016-2018 period

Pesticides residues	Farm number 1 (Vaslui county)	Farm number 2 (Arad county)	Farm number 3 (Teleorman county)	Farm number 4 (Bihor county)	Maximum residue levels for conventional products (EU Pesticides Databases)
Glyphosate	Not detected	Not detected	Not detected	Not detected	1.0
Glufosinate-ammonium (sum of glufosinate, its salts, 1-methyl-4-phenylpyridinium (MPP) and NAG (N-Acetyl-Glufosinate) expressed as glufosinate equivalents)	Not detected	Not detected	Not detected	Not detected	0.1
Acid Aminomethylphosphonic	Not detected	Not detected	Not detected	Not detected	0.5
Chlormequat (sum of chlormequat and its salts, expressed as chlormequat-chloride)	Not detected	Not detected	Not detected	Not detected	0.01
Mepiquat (sum of mepiquat and its salts, expressed as mepiquat chloride)	Not detected	Not detected	Not detected	Not detected	0.02

Results of mycotoxins

Our research has followed the contamination of mycotoxins, over a period of three years, in organic maize samples collected at the harvest of some location with favorable conditions for the maize growing.

According to the scientific literature, the main pathogens producing mycotoxins of maize grains are: *Aspergillus* spp. and *Fusarium* spp. (DEI [6]). The mycotoxins produced by these fungi are: B₁ Aflatoxin produced by *Aspergillus flavus* and *A. parasiticus* produces complex of B₁, B₂ and G₁ and G₂ Aflatoxins; Fumonisin, Deoxynivalenol (DON) (Vomitoxin) and Zearalenone produced by several species of *Fusarium* spp., as *Fusarium graminearum*, *F. culmorum* and *F. verticillioides*, *F. proliferatum*. The maximum risk level of B₁ Aflatoxin in animal feeds is 5-50 µg kg⁻¹, set by the EU standards (EU DIRECTIVES, [15]). The maximum risk level of Fumonisin in maize grain or flour is 2000-4000 µg kg⁻¹ (FAO/WHO [16]). Droughts, supply of nutrients, monoculture or insect attack are favorable causes for the development of these very dangerous pathogens that can cause serious harm to consumers (FAO/WHO [16]). High temperatures and high humidity of maize grains after harvesting could favor the development of the fungus (ROMAN & al [38]).

The results present in Table 4 showed that no compounds were detected in the organic maize samples of Teleorman and Arad counties, in any year of research. For 2016 year, for Vaslui County farm, it was detected: B₁ Aflatoxin (2 µg g⁻¹) and B₁+B₂+G₁+G₂ Aflatoxin (2 µg g⁻¹), B₁+B₂ Fumonisin (992 µg g⁻¹), Deoxynivalenol (DON) (0.7 µg g⁻¹). Also, in 2016 year, the farm from Bihor county was detected all five types of mycotoxins: B₁ Aflatoxin – 1.7 µg g⁻¹, B₁+B₂+G₁+G₂ Aflatoxin - 3.7 µg g⁻¹, B₁+B₂ Fumonisin – 786 µg g⁻¹, Deoxynivalenol (DON) – 26 µg g⁻¹, and Zearalenone – 1.2 µg g⁻¹. The content of these mycotoxins no exceeded of the EU limits, and values have been very low. In 2017 and 2018, there are some discussions with farmers by causes which led to these results and possible measures to prevent and reduce risk of the fungi appearance. Par example, more care for: crop rotation, soil tillage and introduction into the soil of the vegetal residues with risk of contamination from the previous crops; treatment of seeds; balanced fertilisation; insect attack control; appropriate harvesting; controlling the mold infestation by cultivation technologies of maize, before harvesting, storage and processing of grains.

Table 4. Results of mycotoxins for organic maize grains (µg/kg), in 2016-2018 period

The mycotoxin	Farm number 1 (Vaslui county)	Farm number 2 (Arad county)	Farm number 3 (Teleorman county)	Farm number 4 (Bihor county)	Maximum level of mycotoxins (Commission Regulation (EC) No 1058/20 of 19 December 2006)
2016					
Aflatoxin B ₁	2	Not detected	Not detected	1.7	2.0*
Aflatoxin B ₁ +B ₂ +G ₁ +G ₂	2	Not detected	Not detected	3.4	4.0*
Fumonisin B ₁ +B ₂	992	Not detected	Not detected	786	4000**
Deoxynivalenol (DON)	0.7	Not detected	Not detected	26	1750**
Zearalenon	Not detected	Not detected	Not detected	1.2	350**
2017					
Aflatoxin B ₁	Not detected	Not detected	Not detected	Not detected	2.0*
Aflatoxin B ₁ +B ₂ +G ₁ +G ₂	Not detected	Not detected	Not detected	Not detected	4.0*
Fumonisin B ₁ +B ₂	Not detected	Not detected	Not detected	Not detected	4000**
Deoxynivalenol (DON)	Not detected	Not detected	Not detected	Not detected	1750**
Zearalenon	Not detected	Not detected	Not detected	Not detected	350**
2018					
Aflatoxin B ₁	Not detected	Not detected	Not detected	Not detected	2.0*
Aflatoxin B ₁ +B ₂ +G ₁ +G ₂	Not detected	Not detected	Not detected	Not detected	4.0*
Fumonisin B ₁ +B ₂	Not detected	Not detected	Not detected	Not detected	4000**
Deoxynivalenol (DON)	Not detected	Not detected	Not detected	Not detected	1750**
Zearalenon	Not detected	Not detected	Not detected	Not detected	350**

*All cereals and all products derived from cereals, including processed cereal products, with the exception of foodstuffs

**Unprocessed maize with the exception of unprocessed maize intended to be processed by wet milling

Results of nitrate and nitrite

Another group of contaminants analysed in organic maize samples were the nitrates and nitrites contents, which consists of the components in the excessive use of fertilizers.

Nitrates have low toxicity, but they, in the human body, following the activity of the enzyme nitrate reductase, are converted into nitrites and nitrosamines, highly toxic compounds with carcinogenic action, mutagenic and embryo toxic (EU COMMISSION [11]). According to the FAO, the

amount of nitrate tolerated by the adult human is a Maximum Risk Level (MRL) of 50 mg kg⁻¹ body weight day⁻¹ (FAO [17]). Ezeagu, in 2006, tested the level of nitrates and nitrites in three varieties of food grains, including maize grains. The results demonstrated the presence in the amount of 1000.0 mg kg⁻¹ nitrate, and 0.106 mg kg⁻¹ nitrite in maize grains (EZEAGU [8]). The source of nitrate pollution of agri-food products, which have as raw material vegetable production, are mineral fertilizers and also,

organic manure, with a high nitrogen (the main element in the composition of nitrates) content. These substances, which are considered the main nutrients for plants, when used irrationally, pollute products and endanger human health (BLUMENTHAL & al [5]).

Compliance with maximum levels of nitrates can be reasonably obtained by compliance good agricultural practices. In order to ensure safety, food products with excesses of MRL cannot be admitted to the market as such, no mixed with other products, no used as ingredients in food.

The nitrates and nitrites contents in the maize grains tested in the 4 regions of the country are shown in Table 5. It can be seen, only in 2016, in tree farms recorded nitrates values between 5.1 and 6 mg kg⁻¹. No nitrates or nitrites were detected in the other samples, in 2017 and 2018. This shows that organic farmers generally comply with the rules for the application of fertilizers in organic farming, but better management of soil nitrogen must be considered, taking into account the dynamics of this nutrient in the agricultural ecosystem of which the soil and crops are parts.

Table 5. Results of nitrates and nitrites content of organic maize grains, 2016-2018 period

Nitrates/nitrites content	Farm number 1 (Vaslui county)	Farm number 2 (Arad county)	Farm number 3 (Teleorman county)	Farm number 4 (Bihor county)	Maximum Allowed Limit (EU Nitrates Directive 91/676/EEC and WHO, 1998)
2016					
Nitrates (mg kg ⁻¹)	5.1	5.1	6.0	Not detected	50 mg kg ⁻¹
Nitrites (mg kg ⁻¹)	Not detected	Not detected	Not detected	Not detected	1 mg kg ⁻¹
2017					
Nitrates (mg kg ⁻¹)	Not detected	Not detected	Not detected	Not detected	50 mg kg ⁻¹
Nitrites (mg kg ⁻¹)	Not detected	Not detected	Not detected	Not detected	1 mg kg ⁻¹
2018					
Nitrates (mg kg ⁻¹)	Not detected	Not detected	Not detected	Not detected	50 mg kg ⁻¹
Nitrites (mg kg ⁻¹)	Not detected	Not detected	Not detected	Not detected	1 mg kg ⁻¹

Conclusion

This research has demonstrated the comparable quality of organic maize from Romania with international standards in the field.

It was also found that farmers comply with the rules imposed by the organic cultivation of maize, regarding the use of chemical fertilizers and plant protection products.

At the same time, in some years (high humidity at harvest, repeated fertilization), some aspects related to the maximum limits allowed for some harmful substances, such as mycotoxins, nitrates or nitrites, are reported. All values resulting from these substances have been shown to be much lower than industry standards. However, in the future, more attention is needed in preventing these problems and monitoring them permanently. As a result, organic corn can be successfully included in animal and human rations.

The high protein content is noticeable, which makes organic maize grains a valuable raw material that can be successfully included in animal and human nutrition.

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