Article

Not peer-reviewed version

Characterization of Ancient Cereals Cultivated by Intensive and Organic Procedures for Elements Content

Marta Radaelli , Elisa Scalabrin * , Marco Roman , Gabriella Buffa , Irene Griffante , Diego Streliotto , Gabriele Capodaglio *

Posted Date: 7 June 2024

doi: 10.20944/preprints202406.0446.v1

Keywords: Elemental composition; ancient cereals; major and trace nutrients; intensive agriculture; organic agriculture

Article

Characterization of Ancient Cereals Cultivated by Intensive and Organic Procedures for Elements Content

Marta Radaelli [1], Elisa Scalabrin [2,*], Marco Roman [1], Gabriella Buffa [1], Irene Griffante [3], Diego Streliotto [4] and Gabriele Capodaglio [1]

1 DAIS, University Ca' Foscari Venice, Via Torino 155, 30172 Venezia-Mestre, Italy
2 CNR-ISP, Via Torino 155, 30172 Venezia-Mestre, Italy
3 Cerba HealthCare Italia, Padova, Italy
4 Los Prados Italia Srl, Via CA' Nave 97, 35013 Cittadella, Padova, Italy
* Correspondence: elisa.scalabrin@cnr.it

Abstract: According to their nutritional value, their ability to adapt to the various environmental conditions, and their versatility, cereals are among the most cultivated plants in the world. However, the ongoing climate changes subject crops to important environmental stress that for some varieties leads to high production losses. Therefore, selection of species and varieties more versatile and adaptable to different environmental conditions can be important. However, the characteristics of some cereals are not completely known, this is a priority before to improve their cultivation. The aim of this study is to characterize select species of potential suitable for local environmental conditions, nutritional value. Elemental composition was assessed in different cereal species grown following intensive and organic agriculture. Six species were grown for this study with techniques of intensive agriculture: Tritticum monococcum L., Triticum dicoccum L., Triticum aestivum L., variety Verna, Triticum durum Desf., variety Senatore Cappelli, Triticum durum Desf., variety Claudio, and Avena strigosa Schreb.; four of these were also grown following organic procedure: Triticum monococcum L., Triticum dicoccum L., Triticum aestivum L., variety Verna, Triticum durum Desf., variety Senatore Cappelli. The study has considered 20 elements, including major nutrients (Ca, K, Mg, P and S), 7 micronutrients (B, Cu, Fe, Mn, Mo, Se and Zn) and trace elements with toxic properties (Al, Ba, Cd, Cr, Na, Rb, Sc and Sr) that can be accumulated at seeds level. The results highlighted difference in the element concentration in cereal seed in relation to the genus and species, the higher concentration of major nutrients was T. monococcum, the concentrations 6.9, 2.09, 7.2 and 2.9 mg/g for K, Mg, P and S respectively. The higher concentration of some micronutrients, B, Ca, Mo and Se (16, 785, 3.69 and 0.34 mg/g) was in A. strigosa. There is also evidence that element content can be affected by the adopted cultivation procedure; however, effects of growing procedure can be significantly different when different species are considered. T. monococcum grown by organic procedure present a lower concentration of major nutrients, while it can observe a little increase of micronutrients in the T. durum variety S. Cappelli organic, the production procedure does not affect the elemental composition of T. aestivum variety Verna. The survey highlights also that the studied species and the growing procedure, affect the capacity to accumulate and translocate trace hazardous elements for human health at seeds level.

Keywords: elemental composition; ancient cereals; major and trace nutrients; intensive agriculture; organic agriculture

1. Introduction

Plants make up most of the food eaten all over the world and cereals represents the 43% of the world food supply [1]. About half of all cultivated areas in the world are used for cereals production,

 doi:10.20944/preprints202406.0446.v1

which forms the basis of human nutrition on all continents, contributing to the energy and nutrient requirements of the populations around the world.

The estimated cereal world production in 2023 was of 2839 million tonnes with an increase of 1.1 % over 2022 [2].

Maize (*Zea mays, L.*) is the world leading produced cereal, with a harvest of more than 1,163 million ton in 2022, followed by wheat (*Triticum L.*, 808 million ton) and rice (*Oryza L.*, 776 million ton); the sum of these 3 cereals represents about 90% of world cereal production [3].

The need to feed as much people as possible and the pursuit of productivity and performance by intensive agriculture determined the loss of many nutritional and taste properties in crops, by privileging only a few cereal species and varieties [4]. This biodiversity loss highly impacted also agricultural practices, with the increased need to use water, pesticides, fertilizers [5]. Indeed, each species, have their optimal environmental conditions of growth and present different responses to environmental stresses, both biotic and abiotic [6]. Increasing evidence indicate that agricultural production, as an important field related to food security, is becoming extremely vulnerable to climate change and the need to favor sustainable cereals has been raising [7].

The ongoing climate changes, which modify the rainfall and temperature, subject crops to important environmental stress that for some varieties leads to high production losses [8]. This phenomenon is more evident for some hybrid varieties normally used in intensive production, that have a high productivity at the expense of stress tolerance [9]. Therefore, because climate change influences substantially ecological environment, it has become a great challenge to select varieties able to adapt to varying environmental conditions.

Ancient varieties, due to their adaptation that took place over a long time, proved to be more versatile than the main cereals and show a high tolerance to various biotic and abiotic stresses [6,9]. In fact, because they are more resistant to temperature variations and require less water and fertilizer, they would reduce the impact of agriculture on the environment [10–12].Removal of specific genes in crop species to improve genetic pattern is the main cause of the enhanced resistance of some ancient grain species against biotic stresses [13].

Moreover, ancient grains could be the answer to the current state of food security, meeting the nutritional needs of a growing population and satisfying individuals with special nutritional needs such as those with sensitivity to proteins such as gluten [14,15].

Our dependence on only a few crop species limits our ability to meet the challenges posed by the negative effects of climate change and the consequences of food imbalance. As indicated in several studies, ancient cereal varieties might have beneficial nutritional profile and consumers appreciate the taste and flavour of foods based on these products [16]. Moreover, ancient cereals have the potential to contribute to the improved sustainability and resilience of cropping systems, by adapting to a wide range of soil conditions and needing less cultivation inputs [7]. In addition, the production and consumption of foods based on ancient cereals indirectly encourages biodiversity, which has become a priority in environmental and organic farming circles. There is evidence that some traditional food crops, as Tef [(*Eragrostis tef* (Zucc. Trotter)], are particularly nutritious and are also more resilient to marginal soil and climate conditions than major cereals [10]. Therefore, due to their unique nutritional value and phytochemical profile, as well as their sensory characteristics, there is good potential for ancient cereals and associated products to become a part of a healthy diet [17]. Studies carried out on *Triticum turgidum ssp. dicoccoides* (wild emmer) showed high concentrations of micronutrients, Fe and Zn, significantly exceeding those of cultivated wheat [18]. High levels of Ca, Cu and K were also measured in ancient *Triticum* species [19].

However, given the low or irregular performance of ancient cereal production, careful research in this field becomes a priority to improve their cultivation. This has necessitated an alternative and robust approach to improve resilience to diverse types of stresses and increase crop yields.

The aim of this study is to characterize selected cereals species potentially suitable for various environmental conditions, in terms of resistance, productivity and nutritional value, to reduce the use of pesticides and human inputs and to identify a nutritionally superior food source.

 doi:10.20944/preprints202406.0446.v1

Different varieties of cereals, ancient grains and not, were characterized from the chemical-nutritional point of view, determining the content of nutrient elements. These parameters are of particular interest to investigate plants used both as food and forage [20–23].

Differences in the elemental composition were initially assessed in commercial samples, to highlight the main differences because element translocation capability of plants, and overall their concentration in each tissue are generally species-specific [24]. Then, since the element composition may be due to differences in soil composition and agronomic production techniques, the study was extended to larger number of species grown in the open field in a pedologic homogeneous area. In addition, species were subjected to different agricultural practices (conventional and organic) to highlight the differences related to the different production procedures.

The survey also evaluated the content of elements to verify difference of the studied species to accumulate and translocate trace elements of particular interest because phytotoxic elements and hazardous to human health [25–27].

2. Experimental

2.1. Sample Cultivation

Thirteen samples were analysed for elements content.Three commercial samples were analyzed: einkorn wheat (*Triticum monococcum* L.), oats (*Avena sativa* L.), spring wheat (*Triticum aestivum* L.). Six species were grown for this study with techniques of intensive agriculture: *Triticum monococcum* L. (FM), *Triticum dicoccum* L. (FD), *Triticum aestivum* L., variety *Verna* (V), *Triticum durum* Desf., variety *Senatore Cappelli* (SC), *Triticum durum* Desf., variety *Claudio* (C), and *Avena strigosa* Schreb. (A); four of the previous species were grown with organic agriculture: *Triticum monococcum* L. (FMB), *Triticum dicoccum* L. (FDB), *Triticum aestivum* L., variety Verna (VB), *Triticum durum* Desf., variety Senatore Cappelli (SCB). The seeds were obtained from the Rural Seed Network of Umbria and from Los Prados Company. The seeds of the tested varieties were stored at 18 °C and 40% humidity in the storage cell of the University Ca' Foscari. The growth studies following organic procedure were conducted in one greenhouse at the Department of Environmental Sciences, Informatics and Statistics, Ca' Foscari University of Venice. Open field growth following conventional procedures was carried out at the experimental station of La Battistei Farm (winter wheat season 2018-2019), located in Cittadella Municipality, 45.674 N, 11.737 E and 70 m elevation. The average annual rainfall from 1951 to 2014 at the station was 1429 mm, the average seasonal air temperature was 13.0 °C and the average sunshine duration was 2978.72 h. The soil was ploughed to a depth of 20 cm with a tractor-drawn disc plough and large clods of soil were smoothed with a harrow to create a level seedbed. The sowing was carried out on 25 October 2018. The identified varieties were sown with double replication in plots of 5 m^2 for both conventional and organic methods. Traditional (*Triticum monococcum* L., *Triticum dicoccum* L., *Triticum durum* Desf., variety Senatore Cappelli, *Triticum aestivum* L., variety Verna, and *Avena strigosa* Schreb.) and modern (*Triticum durum* Desf., variety Claudio) varieties were sown with 250 and 400 seeds per mm^2 respectively. During the growing season, particularly on 18 February 2019, 30 g of ammonium nitrate (26% N) for *Triticum durum* Desf., variety Claudio and 15 g for the traditional varieties were applied to plants grown using the conventional method. On the same day, 60 g of Super Robur (15%) for Claudio and 30 g for the traditional varieties were applied to the organically grown varieties. Nitrogen fertiliser, however, was not administered to both einkorn and emmer, as it is not necessary for cultivation, but on the contrary increases the tendency to lodging. To prevent fungal infections, the varieties were sprayed with tetraconazole on 18 March 2019 for the intensive grown varieties and organic fungicide for the organic test. The conventional method tests used a dicotyledonous herbicide for seasonal weed control, while the organic tests used mechanical weed control. The harvest took place on 1 July 2019.

Samples were crushed manually in a mort, then they have been milled more finely until a grain size of 5 mm by a ball mill (MM 400, Retsch, Verder Scientific, Haan, Germany) using Teflon gyres at a frequency of 28 Hz for 15 minutes (2 cycles). Every sample was obtained by milling at least 30 seeds.

2.2. Elements' Determination

The commercial samples einkorn wheat, spring wheat and oats (*T. monococcum, T.aestivum and A. sativa*), which were used to optimize the analytical methods, were analysed to determine major nutrient elements Ca, K, Mg, P, S and trace elements Al, B, Ba, Cu, Fe, Li, Mn, Na, Rb, Se, Sr, Zn. Samples of FM, FMB, FD, FDB, SC, SCB, C, V, VB, and A were analyzed to determine the same major nutrient elements, Li was excluded because always present at concentration similar or lower the detection limit, Mo was included because is one micronutrient of particular interest, between the toxic trace elements were also determined Cd and Cr for their environmental interest.

Samples were digested following the procedure previously described in detail [28]. In short, digestion was carried out through a microwave oven (Ethos Milestone Srl, Italy), as follows. One hundred mg of sample were placed in vessels of TFM (Modified Poly-Tetra-Fluoroethylene) with 6 ml of ultrapure HNO_3 (ROMIL Ltd.) and 4 ml of ultrapure H_2O_2 (ROMIL Ltd.). The temperature control inside the vessels was carried out by means of a probe in one reference vessel.

Before each digestion cycle, vessels were subjected to a microwave washing cycle with 10 ml of HNO_3 65% suprapur (Merck) according to the temperature and power program shown in Supplementary Material, Table S1, at the end vessels were rinsed by ultrapure water, reagent grade (ELGA, UK).

The elements determination was carried out, after the procedure validation, by ICP-OES or ICP-MS depending on concentration levels.

2.2.1. Methodology Optimization for the Element Determination

The applied analytical methodologies were ICP-OES and ICP-MS, which were optimized by analyzing a certified reference material (apple leaves SRM-1515, NIST) and commercial cereal samples.

The measurements by ICP-OES (iCap 7000 series, Thermo Scientific) involved the optimization of the plasma conditions: the geometry of the plasma for signal acquisition in both axial and radial mode, the selection of wavelength to minimize spectral interference, the optimal conditions are reported in the Supplementary Material, Table S2.

The analytical methodology referring to the ICP-MS determinations has already been reported Ranaldo et al., 2015 [28], and here it was adapted to a different instrumentation (iCap RQ ICP/MS, Thermo Fisher Scientific with QCell reaction cell), the instrumentation was calibrate by a tuning procedure conducted using a solution of Ba, Bi, Ce, Co, ln, Li and U at a concentration of 1 g/L in HNO_3 2%. It was considered the alignment of the torch, the velocity of the peristaltic pump, the cooling Ar flow and the auxiliary gas flow, the Ar flow for the nebuliser, the power transmitted to the torch and the difference of potential applied to the ionic lenses, the optimal conditions are reported in the Supplementary Material, Table S3. After the tuning procedure, preliminary performance tests were conducted to verify repeatability, both in standard acquisition mode (STD) and by applying the Qcell reaction cell (KEDS), based on these preliminary tests, to avoid any isobaric interferences, it was decided to always use the KEDS procedure with He as reaction gas.

Quantification was carried out by the external calibration curve, the determination coefficient (R^2) obtained by ICP-OES for Al, B, Ba, Ca, Fe, K, Na e P was always higher than 0.999, while for Cu, Mg, Mn, Rb, Zn were always higher than 0.99; the R^2 values for calibration curves obtained by ICP-MS were always higher than 0.999 for all the elements.

The analytical performance of both the methodologies was assessed in term of precision and accuracy (recovery %). Repeatability was assessed by repeating 4 times the measurements on the same solution obtained from the certified material SRM-1515. Reproducibility was estimated by the analysis of the commercial samples, measurements were replicated 5 times on independent portion of each sample.

Preprints.org (www.preprints.org) | NOT PEER-REVIEWED | Posted: 7 June 2024 doi:10.20944/preprints202406.0446.v1

Table 1 reports the concentrations, standard deviations and recovery obtained by the two techniques on the certified reference material, the repeatability as SD obtained by 4 measurements on aliquots of the same sample, accuracy (recovery %) is calculated as ratio % between the experimental results and the certified values. The Tables 2 and 3 report the concentrations obtained by analyses of the 3 commercial samples, using ICP-OES and ICP-MS, respectively, the standard deviation is calculated from 5 measurements carried out on independent portions of the same sample and represent the reproducibility of the method.

Table 1. Concentration in mg/g of elements in the Reference material SRM 1515, certificated value ± tolerance interval (TI); experimental results by ICP-OES and ICP-MS mean values and repeatability obtained by 4 measurements (SD) and recovery as ratio between the results and the certified value % (R%).

Element	Cert conc±TI	ICP-OES conc (SD)	R%	ICP-MS conc (SD)	R%
Al	284.5±5.8	216 (4)	76	182 (11)	64
B	27.6±2.8	--		27.9 (1.6)	101
Ba	48.8±2.3	62.7 (0.7)	128	47.4 (0.5)	97
Ca*	15.250±0.100	16.08 (0.04)	105	---	
Cu	5.69±0.13	6.3 (0.2)	112	6.0 (0.1)	105
Fe	82.7±2.6	74.7(0.6)	90	81.5(7.0)	99
K*	16.080±0.210	17.1 (0.1)	106	11.7 (0.7)	73
Mg*	2.710±0.120	3.00 (0.03)	111	2.71 (0.10)	100
Mn	54.1±1.1	58.7 (0.2)	108	56.2 (2.0)	104
Na	24.4±2.1	22.5 (1.7)	92	---	
P*	1.593±0.068	1.70 (0.16)	107	---	
Rb	10.2±1.6	---		9.8 (0.2)	96
Sr	25.1±1.1	28.9 (0.2)	115	26.2 (0.4)	105
Zn	12.45±0.43	12.2 (0.1)	98	10.5 (0.3)	84

*Concentration in mg/g.

Table 2. Concentration in mg/g and reproducibility as standard deviation by 5 measurements on independent samples (SD) of elements in commercial samples of Spelt (*Triticum monococcum*), Wheat (*Triticum aestivum*) and Oats (*Avena sativa*), determined by ICP-OES.

	Spelt		Wheat		Oats	
Element	mean	SD	mean	SD	mean	SD
Al	--	--	--	--	--	--
B	--	--	--	--	--	--
Ba	4.88	0.08	2.19	0.14	2.67	0.20
Ca*	0.505	0.008	0.345	0.009	0.764	0.049
Cu	8.83	0.13	4.11	0.19	5.01	0.46
Fe	41.2	0.8	38.8	1.9	38.0	2.6
K*	4.71	0.07	4.44	0.21	3.63	0.20
Li	--	--	--	--	--	--
Mg*	1.35	0.03	1.50	0.09	1.42	0.08
Mn	39.6	0.8	43.6	1.6	51.6	2.8
Na	5.2	1.9	9.5	1.6	11.0	1.9
P*	4.08	0.10	4.25	0.21	4.29	0.21
Rb	--	--	--	--	--	--
S*	1.75	0.03	1.26	0.02	1.60	0.09
Se	--	--	--	--	--	--
Sr	12.35	0.23	2.30	0.10	4.28	0.29
Zn	54.84	0.91	29.26	0.75	32.3	2.0

*Concentration in mg/g.

 doi:10.20944/preprints202406.0446.v1

Table 3. Concentration in mg/g and reproducibility as standard deviation by 5 measurements on independent samples (SD) of elements in commercial samples of Spelt (*Triticum monococcum*), Wheat (*Triticum aestivum*) and Oats (*Avena Sativa*), determined by ICP-MS.

	Spelt		Wheat		Oats	
Element	mean	SD	mean	SD	mean	SD
Al	2.01	0.33	1.4	0.9	4.19	0.24
B	4.3	1.6	udl		3.78	0.47
Ba	3.66	0.12	1.26	0.08	0.93	0.07
Ca	--	--	--	--	--	--
Cu	7.49	0.20	2.62	0.44	3.52	0.64
Fe	42.2	1.9	23.7	1.8	25.2	4.3
K*	4.90	0.06	3.33	0.64	2.97	0.45
Li	0.13	0.03	0.04	0.01	0.21	0.04
Mg*	1.25	0.03	0.94	0.17	1.02	0.15
Mn	35.7	0.8	27.4	5.1	35.1	5.8
Na	--	--	--	--	--	--
P	--	--	--	--	--	--
Rb	3.84	0.09	1.08	0.10	1.65	0.09
S	--	--	--	--	--	--
Se**	9	6	11	3	21	7
Sr	10.89	0.26	1.59	0.15	2.79	0.40
Zn	48.79	0.89	17.9	3.2	25.2	1.8

*Concentration in mg/g; ** Concentration in ng/g. udl=under detection limit.

On the basis of the performance of the two techniques and considering the expected concentration ranges for the various elements, the determinations were conducted by ICP-OES, for the elements Al, Ca, Fe, K, Mg, Na, P, S, and by ICP-MS for B, Ba, Cu, Mn, Rb, Se, Sr, Zn. Since the matrix is particularly complex, to avoid memory effects, the sample loading sequence involved a washing procedure by means of HNO_3 2% after each sample.

2.3. Data Elaboration and Statistical Analysis

Statistical analyses were carried out by Metaboanalyst 6.0 [29] https://www.metaboanalyst.ca/). After data normalization, ANOVA-test was applied to verify the significance of the differences in the mean values of each genotype, considering the p-value significance at level < 0.01. Hierarchical cluster analysis was performed using Ward's method and evaluating squared Euclidean distance. The number of clusters was determined in accordance with the Pseudo T-Squared grouping criterion.

A principal component analysis (PCA) was also applied to emphasize differences of plant groups based on the elemental composition.

3. Results and Discussion

The multi-element analysis on commercial samples was conducted to assess differences in the composition between different genus and species; two genera were considered, oats and wheat (Avena sativa and Triticum respectively), and two species of the same genus Triticum (T. monococcum and T. aestivum) were used as models.

The elemental composition is differentiated in major elements and trace elements in relation to their concentrations and the results are reported in Figure 1a, for major elements, and in Figure 1b, for trace elements.

Preprints.org (www.preprints.org) | NOT PEER-REVIEWED | Posted: 7 June 2024 doi:10.20944/preprints202406.0446.v1

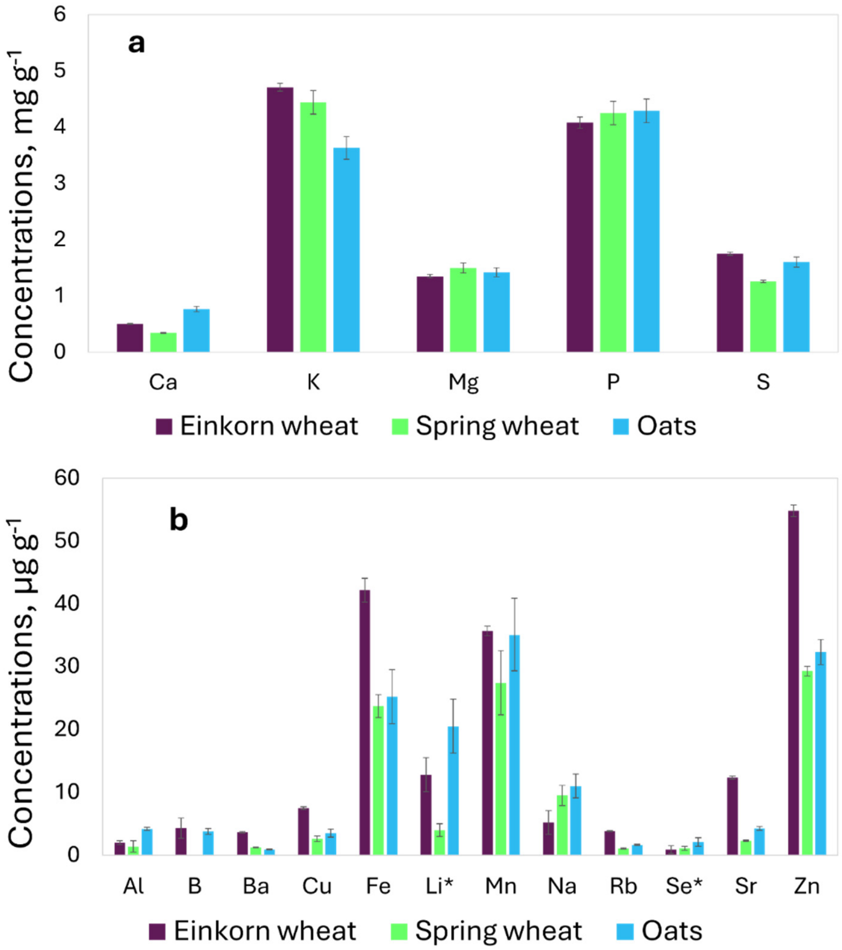

Figure 1. Element concentrations in commercial samples of einkorn wheat (*T. monococcum*), spring wheat (*T. aestivum*) and oats (*A. sativa*): (a) major elements in mg g^{-1}, Ca in mg g^{-1}; (b) trace elements in mg g^{-1}, *Li and *Se mg×100 g^{-1}.

Tests were carried out to highlight significant differences between genera (Avena and Triticum) and between the two species of the genus Triticum (T. monococcum and T. aestivum); element concentrations were considered significantly different if p value was lower than 0.01. Results did not show differences between genus and species for the content of P and Mg, while the mean concentration of K was significantly lower in oats (A. sativa), while the t-test did not show a significant difference between the two species of genus Triticum. The concentration of sulphur (S) in oats and einkorn wheat (A. sativa and T. monococcum) was comparable, while it was significantly lower in spring wheat (T. aestivum). The content of Ca was significantly different in relation to the genus and species; the highest concentration was observed in oats and differences were also significant between the two species of Triticum, with the lower concentrations in spring wheat.

The major difference observed in the commercial samples is the higher level of Ba, Cu, Fe, Rb, Sr and Zn in einkorn wheat samples in comparison to spring wheat and oats, as previously highlighted by some studies [19,30,31]. The concentration of Na (5.2 mg g-1) in einkorn wheat was about half the content in oats and wheat (11 and 9.5 mg g-1 respectively).

Differences in the elemental composition of the samples may be due to differences in soil composition and agronomic techniques; therefore, the study was extended to samples grown in open field in a homogeneous area from the pedologic point of view. The study was applied to a higher number of *Triticum* species (*T. monococcum*, *T. dicoccum*, *T. du*rum-Senatore cappelli, T. durum-Claudio and T. aestivum-Verna) and, to highlight the differences related to the production

Preprints.org (www.preprints.org) | NOT PEER-REVIEWED | Posted: 7 June 2024 doi:10.20944/preprints202406.0446.v1

techniques, the T. monococcum, T. dicoccum, T. durum-Senatore cappelli and T. aestivum-Verna species were also produced by organic agriculture.

The survey evaluated the content of additional elements (Cd, Cr, Mo and Sc) to verify difference of the studied species for their ability to accumulate trace elements of particular interest at the seeds level; Mo is an important trace element because it is a cofactor in enzymatic systems [22,23]; Cd, Cr and Sc are phytotoxic elements and are hazardous to human and animal health [25,26]. The element composition of cereals grown by conventional agriculture is reported in Figure 2 (Figure 2a for major nutrients and Figure 2b for trace elements).

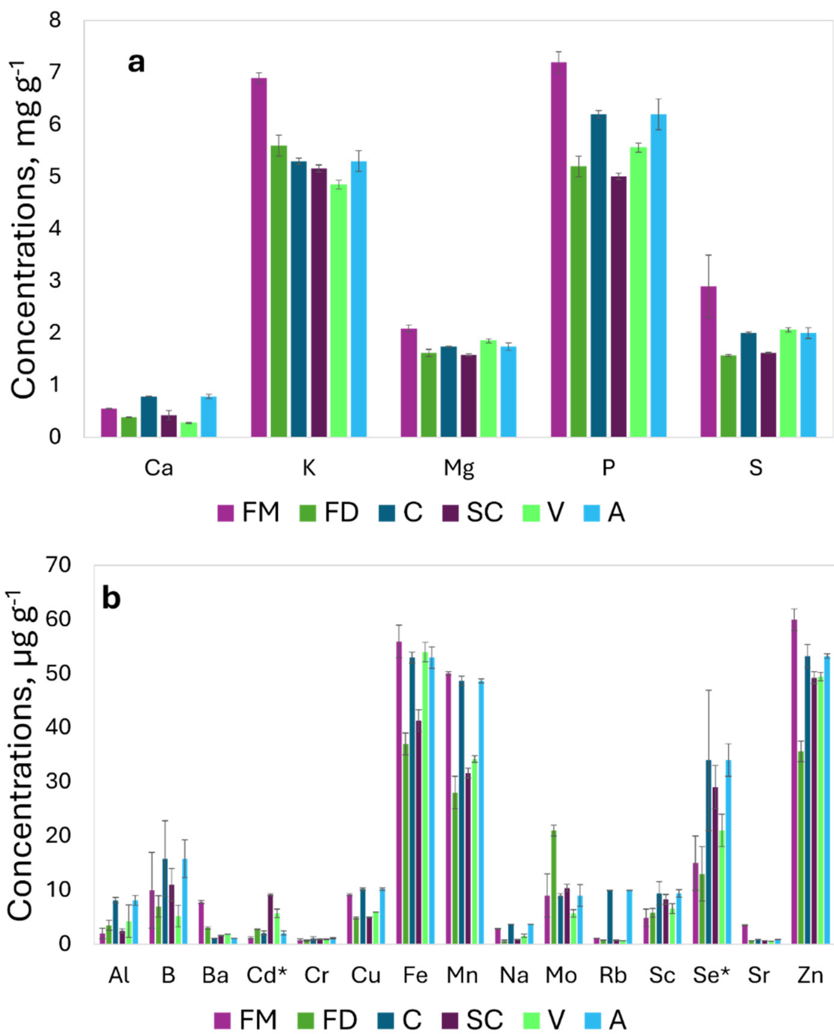

Figure 2. Element concentrations in samples of various *Triticum* species and *Avena strigosa*: (a) major elements in mg g^{-1}; (b) trace elements in mg g^{-1}, *Cd and *Se mg×100 g^{-1}.

The mean concentrations and the standard deviation obtained by 3 measurements on independent samples are reported in Table 4.

The element concentration detected in samples grown in open field are comparable to recent literature data, as reported in Table 5; in general, in this study we observed a higher concentration of B, K and P in comparison to previously reported values. Literature data of Se concentration are variable and our results are consistent with the highest concentrations while lower concentrations were observed for Na respect to literature data [18,32–39].

The highest concentration of major nutrients was observed in FM samples, while the higher concentrations of many minor nutrients (Cu, Fe, Mn, Mo and Zn) were detected both in FM and A.

Table 4. Element concentration in mg/g, mean values and standard deviation (in bracket) in the samples grown following extensive and organic agriculture.

	Triticum									Avena
Element	monococ.	monococ. organic	dicoccum	dicoccum organic	durum (Claudio)	durum (S.Cappelli)	durum org. (S.Cappelli)	aestivum (Verna)	aestivum org. (Verna)	strigosa
Al	2 (1)	1.2 (0.6)	3.5 (0.9)	1.8 (0.4)	8.6 (0.5)	2.5 (0.4)	5.2 (0.3)	4 (3)	3.9 (0.6)	8.2 (0.9)
B	10 (7)	7.5 (3.5)	6.8 (2.3)	7.1 (2.2)	13 (6)	11 (3)	12.7 (1.4)	5.2 (2.0)	8.88 (3.5)	16 (3)
Ba	7.8 (0.3)	2.5 (0.02)	3.0 (0.2)	1.3 (0.1)	1.81 (0.03)	1.58 (0.08)	1.1 (0.1)	1.87 (0.03)	1.73 (0.05)	1.13 (0.02)
Ca	550 (9)	495 (9)	384 (9)	343 (3)	340 (5)	426 (9)	504 (17)	275 (9)	364 (3)	785 (40)
Cd*	12 (2)	9 (1)	28 (1)	30 (1)	45 (4)	92 (2)	75 (10)	57 (8)	62 (3)	21 (4)
Cr	0.8 (0.2)	0.9 (0.2)	0.7 (0.1)	0.7 (0.1)	1.0 (0.3)	0.93 (0.07)	1.13 (0.09)	0.93 (0.09)	0.98 (0.09)	1.1 (0.1)
Cu	9.2 (0.2)	9.1 (0.1)	4.9 (0.2)	6.3 (0.3)	5.3 (0.2)	5.0 (0.1)	7.55 (0.5)	5.93 (0.05)	5.74 (0.08)	10.2 (0.2)
Fe	55 (3)	41.7 (1.5)	37 (2)	39.7 (1.5)	35.2 (1.0)	41 (2)	59.3 (2.4)	54.0 (1.8)	50.8 (1.3)	53.1 (0.9)
K**	6.9 (0.1)	6.6 (0.2)	5.6 (0.2)	5.1 (0.2)	5.00 (0.06)	5.16 (0.07)	5.09 (0.050)	4.85 (0.08)	5.63 (0.04)	5.34 (0.15)
Mg**	2.09 (0.07)	1.76 (0.06)	1.62 (0.07)	1.76 (0.08)	1.30 (0.02)	1.58 (0.02)	1.66 (0.01)	1.85 (0.04)	1.65 (0.01)	1.74 (0.07)
Mn	50.1 (0.3)	40.1 (0.5)	28 (3)	37.4 (2.7)	20.3 (0.9)	31.6 (0.9)	57.4 (4.1)	34.2 (0.6)	33.1 (0.5)	48.7 (0.4)
Mo	2.84 (0.09)	1.83 (0.03)	0.6 (0.2)	0.69 (0.02)	1.00 (0.06)	0.93 (0.01)	0.86 (0.03)	1.6 (0.3)	0.855 (0.008)	3.69 (0.07)
Na	9 (4)	11.0 (3.7)	21 (1)	13.7 (2.6)	9.4 (0.4)	10.4 (0.7)	12.9 (1.6)	5.7(0.7)	14.6 (2.6)	9 (2)
P**	7.2 (0.2)	6.0 (0.2)	5.2 (0.2)	5.4 (0.2)	4.20 (0.07)	5.01 (0.06)	5.28 (0.04)	5.56 (0.09)	5.39 (0.05)	6.2 (0.3)
Rb	1.12 (0.06)	7.4 (0.1	0.78 (0.05)	3.4 (0.2	0.63 (0.02)	0.74 (0.06)	2.84 (0.07	0.69 (0.04)	4.4 (0.2)	1.00 (0.03)
S**	2.9 (0.6)	2.2 (0.5)	1.57 (0.02)	1.58 (0.06)	1.36 (0.02)	1.62 (0.01)	1.81 (0.02)	2.06 (0.04)	1.90 (0.01)	2.0 (0.1)
Sc	4.9 (1.6)	6.3 (1.6)	5.8 (0.8)	6.8 (0.9)	8.1 (2.2)	8.3 (0.9)	9.0 (0.2)	6.6 (0.9)	8.0 (0.6)	9.4 (0.7)
Se	0.15 (0.05)	0.2 (0.1)	0.13 (0.05)	0.18 (0.05)	0.24 (0.13)	0.29 (0.04)	0.33 (0.02)	0.21 (0.03)	0.25 (0.03)	0.34 (0.03)
Sr	3.5 (0.1)	2.19 (0.01)	0.68 (0.03)	0.77 (0.04)	0.43 (0.02)	0.71 (0.02)	1.12 (0.07)	0.603 (0.004)	0.82 (0.04)	0.95 (0.01)
Zn	60 (2)	58.8 (0.3)	36 (2)	56.3 (3.8)	48.3 (2.1)	49 (1)	77.8 (5.2)	49.5 (0.8)	61.2 (0.6)	53.3 (0.4)

*Concentration in ng/g; **Concentration in mg/g.

doi:10.20944/preprints202406.0446.v1

doi:10.20944/preprints202406.0446.v1

Table 5. Literature data of element concentration in cereals in mg/g.

Genus	Triticum													Avena	
Species	wheat[a]	durum[b]	wheat[c]	aestivum[d]	wheat[e]	turgidum			monocc[g]	aestivum	aestivum[h]	durum[h]	tritordeum[h]	sativa[a]	sativa[i]
Variety						durum[f]	durum[g]	dicocc[g]		spelta[g]					
Al															
B			2.85												
Ba															
Ca	380	350	477		415						454	446	434	520	540
Cd*				420											
Cr				2.46											
Cu		4.0		5.63	5.10										6.00
Fe	39	50.4	19.6	0.0	37.0	46	33.0	34.1	45.9	41.8				38.0	47.0
K**	3.40	4.78	3.33		4.20									3.50	4.29
Mg**	1.20	1.03	0.87		1.35	1.34					1.19	1.12	1.31	1.10	1.77
Mn			19.7	81.6	35.0										49.0
Mo															
Na	30.0		20.0			35.0								90.0	20.0
P**		3.93	3.06			3.65									5.23
Rb															
S**			0.84			1.08									
Sc															
Se*	20			160			80.8	229.3	278.9	209.0				30	
Sr															
Zn	29	36.8	28.8	25.7	38.0	61	21.2	22.8	22.4	22.9	31.6	34.3	36.5	33.0	40.0

*ng/g; **mg/g. [a] [32]. [b] [33]. [c] [34]. [d] [37]. [e] [35]. [f] [18]. [g] [36]. [h] [38]. [i] [39].

 doi:10.20944/preprints202406.0446.v1

To highlight differences between genus, species and in relation to the grown techniques the cluster analysis was applied, both on samples and elements, assessed by heatmap plot, (Figure 3). The results highlight that FM appears particularly enriched in nutrients, with the highest levels of major nutrients and average concentration of trace nutrients higher than other Triticum species. In FM samples, only Se and B concentration showed to be lower than values determined in A and other Triticum species. The concentrations of major and trace nutrients were regularly lower in T. durum, of both varieties Claudio and Senatore Cappelli (C and SC), when grown using conventional procedures, only B and Se and other not essential elements (Sr Cr, Cd, Al) were enriched in this species. The Triticum species presenting the lower concentration of nutrients are the T. dicoccum (FD) and T. aestivum Verna (V) when grown by conventional procedure. Samples A (A. strigosa) showed to be characterized by particularly high levels of Mo, Cu and Ca while SCB have the highest levels of Mn, Fe and Zn.

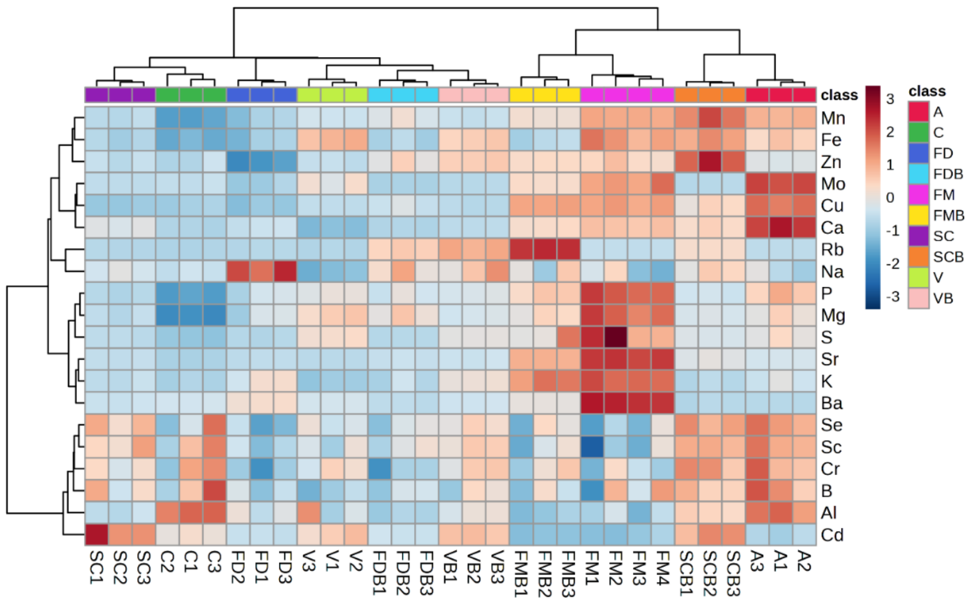

Figure 3. Heatmap of elements in crop seed samples. The color-coded matrix represents the intensities of the element concentration in each sample. Each sample is marked by the class code and the sample number, for the class codes that identify the species see the text.

The growing procedure affects the element content in *T. monococcum* and *T. durum* Senatore Cappelli, indeed FMB presents a lower content of nutrient, contrary to the results obtained for SCB which showed higher concentrations of some trace nutrients (Mn, Fe, Cu and Se) in comparison to SC samples. Previous studies showed different trends in the comparison of nutrients between organic and conventional cultivated crops, depending by the species. Ciolek et al., 2012 [40] showed an increase of Ca and Mg in conventional oat while reporting a diminution of Ca, K, Mg, Fe and Zn in conventional wheat.

Limited differences of nutrients concentrations were observed between V and FD when conventional or organic growing procedures were applied.

PCA was applied to highlight grouping of samples; explained variance \1by the first and second component was 40% and 25% respectively (Figure 4a,b). The score plot revealed principally the separation of the FM and FMB samples by the first component and a partial separation of A, C and SC while the second component highlighted the separation of A samples and SCB. FD and FDB were only partially separated by the second component. No significant differences were observed between V and VB, while limited differences were highlighted between FD and FDB.

 doi:10.20944/preprints202406.0446.v1

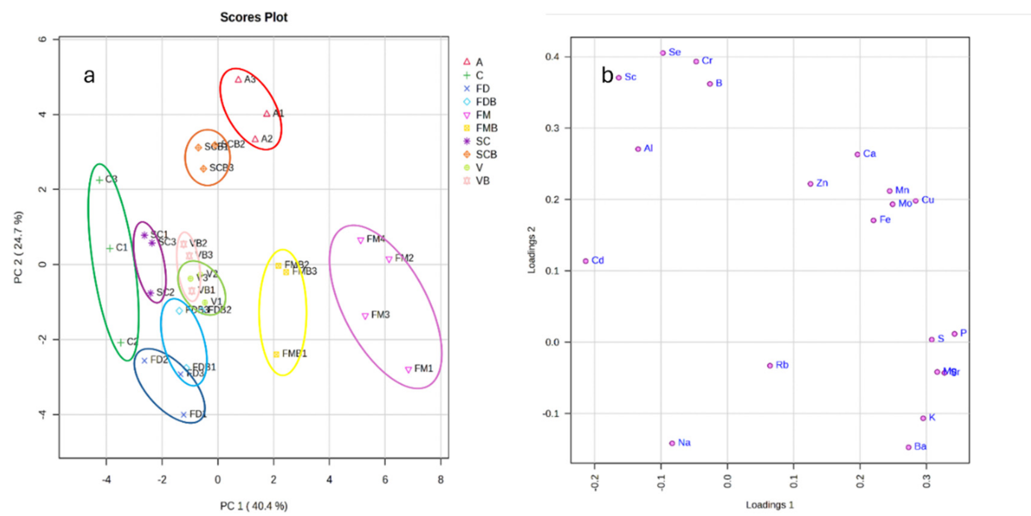

Figure 4. Principal component analysis of data for the element characterization of cereals. (a) score plot of the 30 samples; (b) loading plot based on the same data set.

The first component high loadings, responsible for the separation of FM and FMB from the other species, are mainly related to major nutrients (S, Mg, P and K), which showed high concentrations in these samples (Figure 2a). C samples showed to be characterized by the variable Cd, which showed a lowest loading value in the first component.

The second component explain the contribution of B, Se and some elements not essential for biological systems (Al, Cr and Sc), with the higher values in A and SCB samples; other trace nutrients (Mn, Fe, Cu, Mo, Ca and Zn) contribute to both the components.

Therefore, it can be stated that the difference in the element concentrations in cereal seeds is related to the genus and species, however, there are evidence that element content can be affected also by the adopted cultivation process; indeed, the results show that different cultivation procedures can affect the element composition. However, it must be highlighted that the effect of growing procedure can be significantly different when different species are considered, resulting in opposite trends; the results show, indeed, that production of *T. monococcum* by organic agriculture decrease the concentration of major nutrients, while the *T. durum* variety Senatore Cappelli grown by organic procedure (SCB) is richer in trace nutrients respect to the same species grown by conventional procedure. Effect of fertilization on the concentration of trace nutrients was studied, Stroud et al. [41] studied the effect of S fertilization on the Se and Mo concentration emphasizing the decrease of Se and Mo when S fertilization was applied. The grown procedures on other species composition (*T. aestivum* variety Verna or *T. dicoccum*) seem have not effects or have little effects.

4. Conclusions

Elemental composition was assessed in different cereal species grown following conventional and organic procedures. Six species were grown for this study with techniques of conventional agriculture: *Triticum monococcum* L., *Triticum dicoccum* L., *Triticum aestivum* L., variety Verna, *Triticum durum* Desf., variety Senatore Cappelli, *Triticum durum* Desf., variety Claudio, and *Avena strigosa* Schreb.; four of these were also grown with organic agriculture: *Triticum monococcum* L., *Triticum dicoccum* L., *Triticum aestivum* L., variety Verna, *Triticum durum* Desf., variety Senatore Cappelli.

The study has considered 20 elements, including major nutrients (Ca, K, Mg, P and S), 7 micronutrients (B, Cu, Fe, Mn, Mo, Se and Zn) and trace elements with toxic properties (Al, Ba, Cd, Cr, Na, Rb, Sc and Sr) that can be accumulated at seeds level.

The results highlighted difference in the element concentration in cereal seed in relation to the genus and species, the higher concentration of major nutrients was T. monococcum, while the higher concentration of some micronutrients, B and Se, and some toxic elements, (Cr, Sc and Al) were in *A. strigosa*. There is also evidence that element content can be affected by the adopted cultivation procedure; however, it must be highlighted that the effect of growing procedure can be significantly different when different species are considered. *T. monococcum* grown by organic procedure present a lower concentration of major nutrients, while the production procedure do not affect the elemental composition of *T. aestivum* variety Verna; sometimes the effect can be also contrasting among the same species as observed for *T. durum* variety Senatore Cappelli grown by organic agriculture which resulted richer in some trace nutrients in comparison to conventional procedure.

The survey highlights also that the studied species and the growing procedure affect the ability to accumulate and translocate trace hazardous elements for human health at seeds level.

It must be borne in mind that the response of plants, such as the ability to assimilate elements from the soil, is greatly influenced by environmental conditions, as well climatic characteristics and soil composition; therefore, also taking into account the variability of biological systems, the study is expected to be extended to different environmental plant growth conditions and to a higher number of samples.

Supplementary Materials: The following supporting information can be downloaded at the website of this paper posted on Preprints.org. Table S1. Microwave hoven program for seeds digestion. Table S2. Instrumental parameters for the ICP-OES measurements. Table S3. Instrumental parameters for the ICP-MS measurements

Author Contributions: Conceptualization: Buffa G., Streliotto D., Capodaglio G.; Experimental: Radaelli M., Roman M., Griffante I.; data elaboration: Scalabrin E.; writing—original draft preparation: Radaelli M., Scalabrin E. and Capodaglio G..; writing—review and editing: Radaelli M., Scalabrin E., Capodaglio G.; funding acquisition: Buffa G., Streliotto D., Capodaglio G. All authors have read and agreed to the published version of the manuscript.

Acknowledgement: Study was co-funded by the Los Prados Company. Authors thank the Rural Seed Network of Umbria and Los Prados Company to provide seeds, the La Battistei Farm which supported this study by allowing to use the facilities of their experimental station.

Conflicts of Interest: The authors declare no conflicts of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

References

1. FAO Agrifood solutions to climate change; 2023;
2. FAO Crop Prospects and Food Situation - Triannual Global Report; 2024; ISBN 978-92-5-138352-0.
3. FAO, R. FAO Stat Available online: http://www.fao.org/faostat (accessed on May 21, 2024).
4. Biel, W.; Jaroszewska, A.; Stankowski, S.; Sobolewska, M.; Kępińska-Pacelik, J. Comparison of yield, chemical composition and farinograph properties of common and ancient wheat grains. *Eur. Food Res. Technol.* **2021**, *247*, 1525–1538, doi:10.1007/s00217-021-03729-7.
5. Gregory, P.J.; George, T.S. Feeding nine billion: The challenge to sustainable crop production. *J. Exp. Bot.* **2011**, *62*, 5233–5239, doi:10.1093/jxb/err232.
6. Woldeyohannes, A.B.; Iohannes, S.D.; Miculan, M.; Caproni, L.; Ahmed, J.S.; de Sousa, K.; Desta, E.A.; Fadda, C.; Pè, M.E.; Dell'acqua, M. Data-driven, participatory characterization of farmer varieties discloses teff breeding potential under current and future climates. *Elife* **2022**, *11*, 1–25, doi:10.7554/eLife.80009.
7. Balakrishnan, G. The Role of Amaranth, Quinoa, and Millets for the Development of Healthy, Sustainable Food. *Foods* **2022**, *11*, 2442, doi:https://doi.org/10.3390/foods11162442.
8. Zhang, Z.; Lu, C. Assessing influences of climate change on highland barley productivity in the Qinghai-Tibet Plateau during 1978–2017. *Sci. Rep.* **2022**, *12*, 1–13, doi:10.1038/s41598-022-11711-w.
9. Colombo, F.; Franguelli, N.; Licheri, G.; Ghidoli, M.; Cassani, E.; Castelli, L.; Pasquali, M.; Bresciani, A.; Marti, A.; Dell'Anno, M.; et al. Agriculture in Marginal Areas: Reintroduction of Rye and Wheat Varieties for Breadmaking in the Antrona Valley. *Agronomy* **2022**, *12*, doi:10.3390/agronomy12071695.
10. Numan, M.; Khan, A.L.; Asaf, S.; Salehin, M.; Beyene, G.; Tadele, Z.; Ligaba-Osena, A. From traditional breeding to genome editing for boosting productivity of the ancient grain tef [eragrostis tef (Zucc.) trotter]. *Plants* **2021**, *10*, doi:10.3390/plants10040628.

11. Radzikowska, D.; Sulewska, H.; Bandurska, H.; Ratajczak, K.; Szymańska, G.; Kowalczewski, P.Ł.; Głowicka-Wołoszyn, R. Analysis of Physiological Status in Response to Water Deficit of Spelt (Triticum aestivum ssp. spelta) Cultivars in Reference to Common Wheat (Triticum aestivum ssp. vulgare). *Agronomy* **2022**, *12*, doi:10.3390/agronomy12081822.
12. Zamaratskaia, G.; Gerhardt, K.; Wendin, K. Biochemical characteristics and potential applications of ancient cereals - An underexploited opportunity for sustainable production and consumption. *Trends Food Sci. Technol.* **2021**, *107*, 114–123, doi:10.1016/j.tifs.2020.12.006.
13. Yao, G.; Zhang, J.; Yang, L.; Xu, H.; Jiang, Y.; Xiong, L.; Zhang, C.; Zhang, Z.; Ma, Z.; Sorrells, M.E. Genetic mapping of two powdery mildew resistance genes in einkorn (Triticum monococcum L.) accessions. *Theor. Appl. Genet.* **2007**, *114*, 351–358, doi:10.1007/s00122-006-0438-4.
14. Leoncini, E.; Prata, C.; Malaguti, M.; Marotti, I.; Segura-Carretero, A.; Catizone, P.; Dinelli, G.; Hrelia, S. Phytochemical Profile and Nutraceutical Value of Old and Modern Common Wheat Cultivars. *PLoS One* **2012**, *7*, doi:10.1371/journal.pone.0045997.
15. Valli, V.; Taccari, A.; Di Nunzio, M.; Danesi, F.; Bordoni, A. Health benefits of ancient grains. Comparison among bread made with ancient, heritage and modern grain flours in human cultured cells. *Food Res. Int.* **2018**, *107*, 206–215, doi:10.1016/j.foodres.2018.02.032.
16. Hidalgo, A.; Brandolini, A. Nutritional properties of einkorn wheat (Triticum monococcum L.). *J. Sci. Food Agric.* **2014**, *94*, 601–612, doi:10.1002/jsfa.6382.
17. Arzani, A.; Ashraf, M. Cultivated Ancient Wheats (Triticum spp.): A Potential Source of Health-Beneficial Food Products. *Compr. Rev. Food Sci. Food Saf.* **2017**, *16*, 477–488, doi:10.1111/1541-4337.12262.
18. Cakmak, I.; Torun, A.; Özkan, H.; Millet, E.; Feldman, M.; Fahima, T.; Korol, A.; Nevo, E.; Braun, H.J. Triticum dicoccoides: An important genetic resource for increasing zinc and iron concentration in modern cultivated wheat. *Soil Sci. Plant Nutr.* **2004**, *50*, 1047–1054, doi:10.1080/00380768.2004.10408573.
19. Golea, C.M.; Stroe, S.G.; Gâtlan, A.M.; Codină, G.G. Physicochemical Characteristics and Microstructure of Ancient and Common Wheat Grains Cultivated in Romania. *Plants* **2023**, *12*, doi:10.3390/plants12112138.
20. Noulas, C.; Torabian, S.; Qin, R. Crop Nutrient Requirements and Advanced Fertilizer Management Strategies. *Agronomy* **2023**, *13*, 1–10, doi:10.3390/agronomy13082017.
21. 2Kaur, H.; Kaur, H.; Kaur, H.; Srivastava, S. The beneficial roles of trace and ultratrace elements in plants. *Plant Growth Regul.* **2023**, *100*, 219–236, doi:10.1007/s10725-022-00837-6.
22. Bjornsson, R.; Neese, F.; Schrock, R.R.; Einsle, O.; Debeer, S. The discovery of Mo(III) in FeMoco: Reuniting enzyme and model chemistry. *J. Biol. Inorg. Chem.* **2015**, *20*, 447–460, doi:10.1007/s00775-014-1230-6.
23. Van Stappen, C.; Davydov, R.; Yang, Z.Y.; Fan, R.; Guo, Y.; Bill, E.; Seefeldt, L.C.; Hoffman, B.M.; Debeer, S. Spectroscopic Description of the E1 State of Mo Nitrogenase Based on Mo and Fe X-ray Absorption and Mössbauer Studies. *Inorg. Chem.* **2019**, *58*, 12365–12376, doi:10.1021/acs.inorgchem.9b01951.
24. Subramanian, D.; Subha, R.; Murugesan, A.K. Accumulation and translocation of trace elements and macronutrients in different plant species across five study sites. *Ecol. Indic.* **2022**, *135*, 108522, doi:10.1016/j.ecolind.2021.108522.
25. Shtangeeva, I. Accumulation of scandium, cerium, europium, hafnium, and tantalum in oats and barley grown in soils that differ in their characteristics and level of contamination. *Environ. Sci. Pollut. Res.* **2022**, *29*, 40839–40853, doi:10.1007/s11356-021-18247-y.
26. Chen, Z.; Imran, M.; Jing, G.; Wang, W.; Huang, B.; Li, Y.; Zhang, Y.; Yang, Y.; Lu, Q.; Zhang, Z.; et al. Toxic elements pollution risk as affected by various input sources in soils of greenhouses, kiwifruit orchards, cereal fields, and forest/grassland. *Environ. Pollut.* **2023**, *338*, doi:10.1016/j.envpol.2023.122639.
27. Fieve, R.R.; Meltzer, H.L.; Taylor, R.M. Rubidium chloride ingestion by volunteer subjects: Initial experience. *Psychopharmacologia* **1971**, *20*, 307–314, doi:10.1007/BF00403562.
28. Ranaldo, M.; Toscano, G.; Radaelli, M.; Scalabrin, E.; Capodaglio, G. *Nicotiana langsdorffii* wild type and genetically modified exposed to chemical and physical stress: changes in element content. *Int. J. Environ. Anal. Chem.* **2015**, *95*, 349–365, doi:10.1080/03067319.2015.1016013.
29. Pang, Z.; Chong, J.; Zhou, G.; De Lima Morais, D.A.; Chang, L.; Barrette, M.; Gauthier, C.; Jacques, P.É.; Li, S.; Xia, J. MetaboAnalyst 5.0: Narrowing the gap between raw spectra and functional insights. *Nucleic Acids Res.* **2021**, *49*, W388–W396, doi:10.1093/nar/gkab382.
30. Zencirci, N.; Karakas, F.P.; Ordu, B. Macro-Micro Element Variation in Traditionally Grown Einkorn (Triticum monococcum L. subsp. monococcum) and Emmer Wheat (Triticum dicoccon Schrank). *Int. J. Second. Metab.* **2021**, *8*, 227–245, doi:10.21448/ijsm.778596.
31. Van Boxstael, F.; Aerts, H.; Linssen, S.; Latré, J.; Christiaens, A.; Haesaert, G.; Dierickx, I.; Brusselle, J.; De Keyzer, W. A comparison of the nutritional value of Einkorn, Emmer, Khorasan and modern wheat: whole grains, processed in bread, and population-level intake implications. *J. Sci. Food Agric.* **2020**, *100*, 4108–4118, doi:10.1002/jsfa.10402.
32. McKevith, B. Nutritional aspects of cereals. *Nutr. Bull.* **2004**, *29*, 111–142, doi:10.1111/j.1467-3010.2004.00418.x.

Preprints.org (www.preprints.org) | NOT PEER-REVIEWED | Posted: 7 June 2024 doi:10.20944/preprints202406.0446.v1

33. Nowak, V.; Du, J.; Charrondière, U.R. Assessment of the nutritional composition of quinoa (Chenopodium quinoa Willd.). *Food Chem.* **2016**, *193*, 47–54, doi:10.1016/j.foodchem.2015.02.111.
34. Sushree Shyamli, P.; Rana, S.; Suranjika, S.; Muthamilarasan, M.; Parida, A.; Prasad, M. Genetic determinants of micronutrient traits in graminaceous crops to combat hidden hunger. *Theor. Appl. Genet.* **2021**, *134*, 3147–3165, doi:10.1007/s00122-021-03878-z.
35. Santos, D.; Pintado, M.; Lopes da Silva, J.A. Potential nutritional and functional improvement of extruded breakfast cereals based on incorporation of fruit and vegetable by-products - A review. *Trends Food Sci. Technol.* **2022**, *125*, 136–153, doi:10.1016/j.tifs.2022.05.010.
36. Zhao, F.J.; Ma, J.F.; Meharg, A.A.; McGrath, S.P. Arsenic uptake and metabolism in plants. *New Phytol.* **2009**, *181*, 777–94, doi:10.1111/j.1469-8137.2008.02716.x.
37. Kerketta, A.; Kumar, H.; Powell, M.A.; Sahoo, P.K.; Kapoor, H.S.; Mittal, S. Trace Element Occurrence in Vegetable and Cereal Crops from Parts of Asia: A Meta-data Analysis of Crop-Wise Differences. *Curr. Pollut. Reports* **2023**, *9*, 1–21, doi:10.1007/s40726-023-00248-9.
38. Shewry P.R., Brouns F., Dunn J., Hood J., Burridge A.J., America A.H.P., Gilissen L., Proos-Huijsmans Z.A.M., van Straaten J.P., Jonkers D., Lazzeri P.A., Ward J.L., Lovegrove A. (2023). Comparative compositions of grain of tritordeum, durum wheat and bread wheat grown in multi-environment trials. Food Chemistry, 423, 136312.
39. https://doi.org/10.1016/j.foodchem.2023.136312
40. USDA, “US Department of agriculture, agricultural research service” FoodData central, oat.
41. Ciołek, A.; Makarska, E.; Wesołowski, M.; Cierpiała, R. Content of selected nutrients in wheat, barley and oat grain from organic and conventional farming. *J. Elem.* **2012**, *17*, 181–189, doi:10.5601/jelem.2012.17.2.02.
42. Stroud, J.L.; Zhao, F.J.; Buchner, P.; Shinmachi, F.; McGrath, S.P.; Abecassis, J.; Hawkesford, M.J.; Shewry, P.R. Impacts of sulphur nutrition on selenium and molybdenum concentrations in wheat grain. *J. Cereal Sci.* **2010**, *52*, 111–113, doi:10.1016/j.jcs.2010.03.011.