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Brief Report

Effects of Weak Magnetic Fields on Plant Chemical Composition and Its Ecological Implications

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Abstract: The exposure of plants to weak magnetic fields (MFs) of various intensities and for different times is increasingly adopted to sustainably enhance plant growth in plant-based applications such as modern agriculture, phytoremediation and biogas production. However, little is known about the effects of MF exposure on plant chemical composition, and in turn on related ecosystem processes, such as the transfer of potentially toxic elements along food chains and the decomposition of organic matter. To fill this gap, the present research, through the study of the chemical composition of four edible crops (leaves of lettuce, parsley and basil, and fruits of tomato) differently exposed to weak MFs (75 Hz; 1.5 mT), aimed at evaluating the overall effects of the exposure on ecosystem processes. In particular, several essential (B, C, Ca, Cu, K, Fe, Mg, Mn, Mo, N, Ni, P, S, Zn), beneficial (Co, Na, Se, Si) and non-useful (Al, As, Ba, Cd, Cr, Li, Pb, Sr, Ti, V) elements, together with chemical compounds and derived parameters (soluble sugars, starch, chlorophylls, flavonoids, anthocyanins, nitrogen balance index), indicators of plant metabolism and health, and litter decomposability traits (C/N, C/P), were analyzed. Notwithstanding the expected variations in the observed effects among species and MF exposure conditions, the obtained results highlight a general decrease in most of the studied parameters (with the exception of those related to litter decomposability), attributable to a lower absorption/accumulation of the studied chemical elements and to a reduced synthesis of metabolites. The largest average reduction was observed for the non-useful elements, which outweighs the reduction in essential and beneficial elements and provides for an important MF-induced effect, considering their toxic, persistent and biomagnificable characteristics. Similarly, the induced increases in C/N and C/P ratios indicate the production of litter more recalcitrant to the decomposition process, suggesting that weak MF treatments may be useful to enhance soil C storage and reduce CO_2 emissions.

Keywords: low frequency MFs; edible crops; plant chemical elements and compounds; plant health; litter quality

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1. Introduction

Magneto-sensitivity is widespread in biological systems and hundreds of studies have found that weak magnetic fields (MFs) can significantly influence their dynamics in various model organisms (plants, bacteria, fungi, animals comprising human beings), although the involved mechanisms [1] and even more how they reflect on ecosystem processes [2] remain largely elusive. Weak MF exposure, up to tens of mT [3], does not emit wastes, does not generate harmful by-products and requires very little power, so it appears an environmentally and economically friendly, sustainable and highly desirable technique in modern plant-based applications [4–6].

The whole Earth biota is exposed to a natural stationary magnetic field, called the "geomagnetic field", ranging at the Earth surface from 25 to 65 μT [7], and to other magnetic fields generated by different natural and anthropogenic sources [5]. The modulation of the field to which plants are subjected, through their exposure to artificial weak MFs, has been applied in the past years to increase plant growth. The aims span improving crop yields [5–9], plant remediation capability [10,11] and efficiency of biomass conversion into biogas [12]. Indeed, it has been highlighted in different species that MFs improve not only seed germination, seedling growth [6,8,9,13–15] and fruit ripening [16], but also plant health and tolerance against environmental stresses [6,17–20].

Several mechanisms have been hypothesized to be involved [16,19], such as alterations of cell membrane characteristics and cell cycle regulation, with changes in cell metabolism and functioning, including gene expression, protein biosynthesis and enzyme activities [4,7,8,17,21,22]. At the molecular level, radical pair mechanisms have been proposed as a potential unifying explanation for the described MF effects [1]. In any case, the observed responses vary according to the plant species and age during exposure, as well as the MF intensity and exposure time [5–7,23,24].

Notwithstanding the strong effects of the geomagnetic field on plant evolution [16] and the evidence that the artificial magnetic fields employed only partly compensate for the 10% reduction in geomagnetic field of the last decades [7], the wrong modulation of MF intensities and time of exposure can turn the artificial MF treatment into a stressful factor [7,25]. In relation to the possible negative effects of MFs on plants, special focus has been directed to the genotoxic and mutagenic effects, affecting DNA integrity and spontaneous mutations [7,26]. Alterations of cell membranes can cause changes in cell electric potential, which in turn can impair the uptake and accumulation of chemical elements and then the synthesis of biomolecules, especially those directly involved in the conversion of solar energy into chemical energy [7,19]. In spite of the relevance of the topic in understanding MF effects on plants and ensuring beneficial MF exposures, little attention has been paid to the chemical composition of plants exposed to MFs, in terms of both element and biomolecule concentrations [7,9]. The relevance of these aspects goes way beyond the food safety issue and pertains to large ecological scale processes such as the transfer of potentially toxic elements (PTEs) along food chains [27] and ecosystem functioning, in relation to the modulation of decomposition and, in turn, the matter cycle by litter chemistry [28].

With the aim to clarify the overall effects of weak MFs on the chemical composition of plants and their ecological implications, in the present research several essential (B, C, Ca, Cu, K, Fe, Mg, Mn, Mo, N, Ni, P, S and Zn), beneficial (Co, Na, Se and Si) and non-useful (Al, As, Ba, Cd, Cr, Li, Pb, Sr, Ti and V) elements, together with several biomolecules (soluble sugars, starch, chlorophylls, flavonoids and anthocyanins), were analyzed in the edible organs of different species (lettuce, parsley, basil and tomato). Data were also used in calculating specific parameters involved in plant health (nitrogen balance index—NBI) and in litter decomposability (C/N and C/P), in order to outline the ecological implications of plant exposure to weak MFs.

2. Materials and Methods

To obtain realistic evaluations of the effects of weak MFs on plant chemistry and related ecosystem processes, with little bias toward particular species or exposure conditions, the experimental design involved the study of different species subjected to various MF treatments and the analysis of a large number of parameters. Specifically, the study was carried out on four plant species representing important crops (lettuce, parsley, basil and tomato) in the human diet and the most common "vegetables" in the Mediterranean diet, declared in 2010, by UNESCO, an intangible cultural heritage of humanity (http://www.unesco.org/culture/ich/index.php?lg=en&pg=00011&RL=00884; accessed 26 December 2022). Leaves of lettuce (*Lactuca sativa* L.; Asteraceae) are available and consumed worldwide as a salad crop [29,30]. Leaves of parsley (*Petroselinum crispum* (Mill.) Fuss; Apiaceae) and basil (*Ocimum basilicum* L.; Lamiaceae) are typical seasonings and are fresh-consumed in large quantities as ingredients in various dishes and food preparations [31]. Fruits of tomato (*Solanum lycopersicum* L.; Solanaceae) are one of the most popular and consumed fresh "vegetables" in the world and are also widely used by the food industry as a raw material for the production of derived products [32].

The analyses were performed on leaves of lettuce, parsley and basil, as well as fruits of tomato, from mature and healthy plants subjected to different exposure treatments with weak MFs (75 Hz; 1.5 mT) and were compared with untreated controls, exposed only to the Earth's geomagnetic field (Table 1), equal to 46.4 µT in the study area and in the growth period [33]. MFs were generated by Helmholtz coils in order to ensure the homogeneity of the field between the coils.

Table 1. MF treatments performed on seeds or plants of the studied species, with the indication of the exposure time and the sampling time after the transplanting (*L. sativa* and *S. lycopersicum*) or the first exposure (*P. crispum* and *O. basilicum*).

Species	Life Stage	Exposure Time	Sampling Time
L. sativa	seed	4 h/day for 0, 40, 110 and 150 days	70 and 150 days
P. crispum	plant	0, 1, 2, 3, 6, 9 h/day for 10 days	15 days
O. basilicum	plant	0, 1, 2, 3, 6, 9 h/day for 10 days	15 days
S. lycopersicum	seed	8 h/day for 0 and 10 days	90 days

For each species and each treatment, at least 10 plants were grown in pots at the same environmental conditions (Table 2) and on the same soil (Table 3), irrigated daily with tap water. Pots were kept away from electrical power lines and any other potential source of anthropogenic magnetic fields, to minimize interferences with the generated fields. Three independent samples, obtained by pooling together the plant material coming from three random individuals, were subjected to the laboratory analyses. Before the collection, three leaves of *L. sativa* for each treatment were analyzed, by means of a portable fluorimeter (Dualex Scientific, Force-A, France), for chlorophyll, flavonoid and anthocyanin concentrations, as well as for NBI, calculated as chlorophylls/flavonoids ratio and related to the nitrogen and carbon allocations.

Table 2. Monthly average environmental conditions at the growing site (40.5547° N, 14.9585° E, 17 m a.s.l.) during crop cultivation (Spring–Summer 2021). Data, accessed on 13 February 2023, were retrieved from the National Oceanographic and Atmospheric Administration (https://gml.noaa.gov/grad/solcalc/) and from a wheather archive (https://www.ilmeteo.it/portale/archivio-meteo/Eboli/2021).

	April	May	June	July	August
Daylight hours	13.3	14.4	15.0	14.6	13.7
Minimum temperature (°C)	13.7	19.2	24.7	27.1	27.5
Maximum temperature (°C)	17.1	22.2	28.4	30.1	30.9
Rainy days	5	1	7	1	0
Relative humidity (%)	59.7	60.1	53.0	53.0	50.3
Wind speed (Km/h)	12.2	12.5	11.0	12.1	12.7

For all the other chemical analyses, employing analytical grade chemicals and reagents only, fresh plant material was manually pulverized with liquid nitrogen in china mortars and oven-dried at 75 °C until constant weight. C and N concentrations were measured using a CHNS-O Analyzer (Flash EA1112, Thermo Fisher, USA). All the other chemical elements (Al, As, B, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Li, Mg, Mn, Mo, Na, Ni, P, Pb, S, Se, Si, Sr, Ti, V and Zn) were measured on samples obtained throughout a microwave-assisted (Ethos, Milestone, Italy) acid digestion, as reported in Baldantoni et al. [34], by means of inductively coupled plasma optical emission spectrometry (Optima 7000DV, PerkinElmer, USA), employing a PTFE Gem-Cone/Cyclonic chamber nebulizer. Standard reference material (SRM 1547 peach leaves [35]) was also analyzed in order to verify the accuracy of the obtained data, with recoveries in the range of 95–102%. Subsequently, the C/N and C/P ratios were calculated. Finally, soluble sugars and starch concentrations were determined according to the Chow and Landhäusser [36] method.

Table 3. Soil chemical and physical characteristics. Data are reported as $\bar{x} \pm 2\sigma$, where available.

	Value	Unit
Skeleton	43 ± 6	g/Kg
Coarse sand	132 ± 4	g/Kg
Fine sand	71 ± 2	g/Kg
Coarse silt	36 ± 1	g/Kg
Fine silt	452 ± 15	g/Kg
Clay	309 ± 10	g/Kg
pH_{water}	8.1 ± 1.5	–
Electrical conductivity	44.2 ± 3.0	mS/m
Total $CaCO_3$	17 ± 6	g/Kg
Active $CaCO_3$	5 ± 2	g/Kg
Organic matter	18 ± 4	g/Kg
Organic C	10.4 ± 2.0	g/Kg
Total N	1.64 ± 0.20	g/Kg
Extractable P	67 ± 13	mg/Kg P
Exchangeable Ca	3862.9 ± 229.0	mg/Kg Ca
Exchangeable Mg	459.6 ± 27.0	mg/Kg Mg
Exchangeable Na	76.1 ± 7.0	mg/Kg Na
Exchangeable K	538.0 ± 20.1	mg/Kg K
Cation exchange capacity	25	meq/100g

In order to combine the information provided by the different species/exposure settings and obtain average estimates for the effects of weak MFs, meta-analytical techniques were adopted, using the response ratio (RR) as an index of effect size. Notwithstanding the recent emphasis on a few shortcomings of RR in respect to other effect size indices such as the (standardized) mean difference [37], it is still the most adopted metric of its kind in ecology and provides for a straightforward interpretation of results, irrespective of the units of measurement [38,39]. In particular, the mean, standard deviation and number of replicates for each parameter analyzed on MF-exposed and control plants in each experimental setting were employed in calculating RR and estimating the mean effect size ($\hat{\theta}_i$) and confidence intervals through random effect models. To this end, RRs of each parameter estimated in the different exposure settings were treated as different studies and grouped into five parameter classes: essential elements (B, C, Ca, Cu, K, Fe, Mg, Mn, Mo, N, Ni, P, S and Zn), beneficial elements (Co, Na, Se and Si), non-useful elements (Al, As, Ba, Cd, Cr, Li, Pb, Sr, Ti and V), indicators of plant metabolism and health (soluble sugars, starch, chlorophylls, flavonoids, anthocyanins and NBI) and litter decomposability traits (C/N and C/P). Random effect models were calculated independently for each class, and their choice over equal effect models relied on the results of τ^2 profiling. Cochrane Q-tests were performed for each parameter group in order to evaluate the presence of heterogeneity in RRs larger than would be expected based on sampling variability. All the analyses were performed using the functions of the package "metafor" [40] for the R 4.2.2 programming language [41].

3. Results and Discussion

Considerable variations in RRs for the different parameters in the four species (Figure 1) were observed, spanning over four orders of magnitude across the null effect size, without clear trends in relation to the species and exposure conditions. Similar findings in relation to the remarkable variability in plant responses to weak MF treatments have been reported by other authors, e.g., for the flavonoid concentrations in *Helianthus annuus* [9], and confirm the previously highlighted unpredictability of weak MF effects on plants [42]. Large variations in chemical composition of the exposed plants can be explained by the modulation of MF effects by both direct (plant metabolism) and indirect (soil PTE bioavailability) mechanisms [43] and, likewise, are in common with other stimuli propagating through complex systems, such as soil fertilization [30].

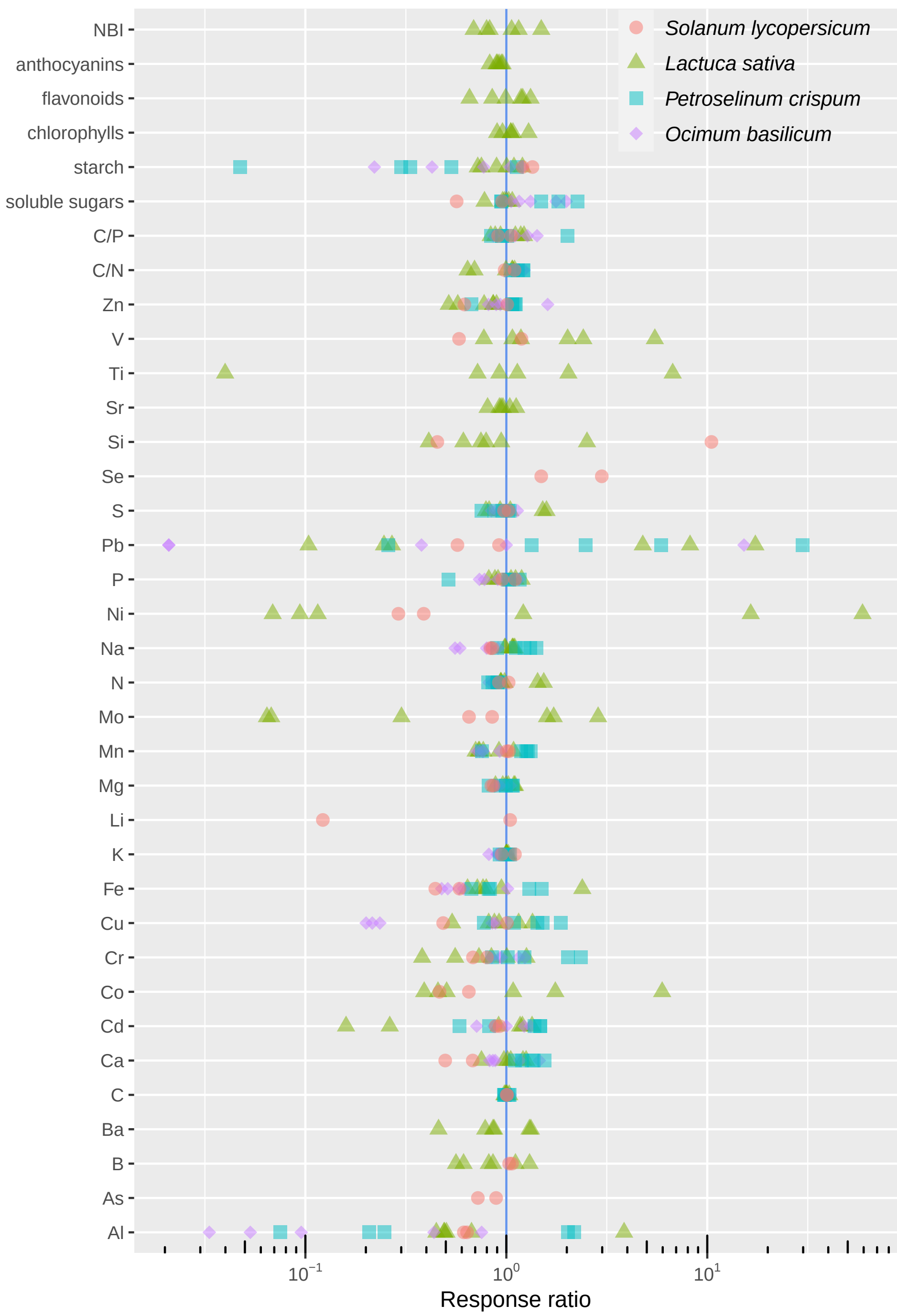

Figure 1. RRs, plotted on logarithmic scale, for each parameter measured in the different species, indicated in different colors and symbols, under the various experimental conditions.

The remarkable variability in RRs is further confirmed by the results of the Cochrane Q-tests, indicating that for the five groups of parameters, i.e., essential, beneficial and non-useful elements, as well as indicators of plant health and metabolism and of litter decomposability, the heterogeneity in the observed effect sizes is always greater ($p < 0.001$ for all groups) than the one expected based on sampling variability. Accordingly, the I^2 statistics show values from 95.52% up to 99.80% for all groups. Such a heterogeneity among parameters and species results in large confidence intervals on $\hat{\theta}_i$ estimates for most of the groups of parameters (Figure 2), especially in the case of beneficial and non-useful elements. The latter, in particular, are remarkable in showing the largest absolute effect size ($\hat{\theta}$ = −19.44%), but with an associated *p*-value that is marginally significant (p = 0.0390). Such an effect on non-useful elements contrasts with MF effects on essential elements, showing a lower absolute MF-induced reduction in concentrations ($\hat{\theta}$ = −7.60%), but with tighter confidence intervals and a lower *p*-value (p = 0.0002). Overall, for all the groups of parameters with the only exception of the litter decomposability traits, random effects models estimate negative $\hat{\theta}_i$, i.e., a reduction in parameter values induced by the weak MF exposures (Figure 2), attributable to a lower absorption/accumulation of the studied chemical elements and to a reduced synthesis of metabolites. In this context, the positive mean effect size ($\hat{\theta}$ = +5.70%, p = 0.0949) observed in the case of the litter decomposability traits is attributable to a larger relative reduction in the concentrations of N and P in respect to C.

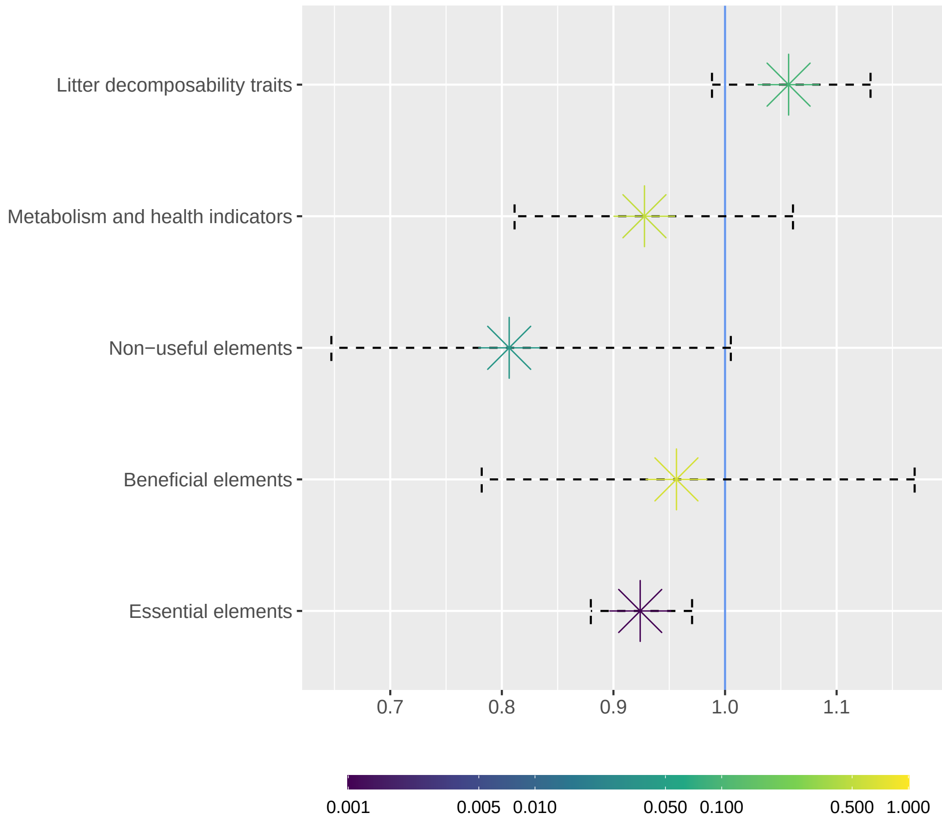

Figure 2. Effect size for the different parameter classes estimated through the random effect models, with indication of the confidence intervals. Symbols for $\hat{\theta}_i$ are colored according to the respective *p*-value, indicated by the logarithmic color bar.

In contrast to our results about the average decrease in the essential and beneficial elements (Figure 2), a positive effect of MFs on plant nutrient uptake is usually expected [7,19] and often documented. For example, Eşitken and Turan [25] reported increased concentrations of Ca, K, Fe, Mg, Mn, N and Zn, and decreased concentrations of P and S in *Fragaria* x *ananassa* leaves from plants exposed to 0.384 T at 50 Hz MF in respect to the unexposed plants, concluding that MFs play an important role in ion uptake capacity, with increases for the immobile cations and decreases for the mobile anions. Similar findings were obtained by Dhawi and Al-Khayri [44], who reported that increasing the time of exposure (from 1 to 15 min) of *Phoenix dactylifera* seedlings to 1500 mT at 10 mHz to 63 kHz MF increased the concentrations of Ca, Fe, Mg, Mn, Na and Zn, but decreased P concentration as compared to the control. Finally, Radhakrishnan and Kumari [4] found a considerable increase in Cu, K, Fe, Mg, Mn, Na and Zn concentrations (but reduced Ca concentrations) in *Glycine max* seedlings from seeds exposed for 20 days (for 5 h/day), to 1.5 µT at 10 Hz MF.

In the present research, the highest average MF-induced reduction was observed for plant non-useful element concentrations (Figure 2), reassuring consumers (at least for PTEs) on the question recently raised by Radhakrishnan [19]: "Is there any toxicity due to the consumption of MF treated foods?". In particular, two toxic non-useful elements that are potentially carcinogenic for human beings [45], Cd and Pb, always decreased in tomato fruits from plants treated at the seed stage, whereas they showed wide variations (from reduced to increased concentrations) in lettuce, parsley and basil leaves (Figure 1). In this context, it should be considered that lettuce usually has large Cd accumulation capabilities [29,30] and the MF-induced reduction in concentrations observed in several cases warrants further exploration in searching for optimal exposure conditions. In contrast to our average estimates, exposure of *Triticum aestivum* seeds to static MFs of 600 mT caused a significant increase in Cd and Pb leaf concentrations of the obtained seedlings [18]. Similar results were reported by Luo and co-workers, who found higher Cd and Pb leaf concentrations in *Eucalyptus globulus* [10] and *Noccaea caerulescens* [11] treated with static MFs before sowing, with increasing values at increasing intensities (from 30 to 400 mT).

Overall, the average reduction of the non-useful elements in the studied crops outweighs the reduction of the essential and beneficial elements and provides for an important MF-induced effect, considering the persistent and biomagnificable characteristics of most of the non-useful elements. However, the significant reduction in essential elements observed in the exposed crops (Figure 2) might still pose risks to human health in terms of intake deficiencies. Indeed, at least 25 chemical elements are essentials for humans, and plants are their main providers [46,47]. Luckily, among the main elements responsible (Ca, Cu, Fe, I, Mg, Se, Zn) for the major deficiencies [48], there are elements that marginally contribute to the observed lowering of essential element concentrations, and do so only in particular species and exposure conditions (Figure 1). Moreover, the risk of high intake toxicity from several of these elements [46] should not be underestimated and the 8% reduction in essential elements may prove influential or even beneficial, depending on the diet and exposure to environmental sources of these elements. As a future perspective, it remains to clarify if the observed findings on the reduced chemical element concentrations in treated plants would be due to direct (on plant) or indirect (soil-mediated) effects.

The general decrease observed in chemical elements reverberates with the average reduction in metabolism and health indicators (Figure 2), generally increasing in MF-treated plants. For example, Chen and co-workers [49] reported increased concentrations of proteins (and several amino acids), soluble sugars and anthocyanins in *Vigna radiata* plants from seeds exposed to 600 mT static MF. Notwithstanding the transport of carbohydrates and other metabolites from the sites of synthesis to the distant growth zones and fruits could be stimulated at low MF intensities [19,25], reduction in biomolecule synthesis or allocation has been sometimes reported in several plant species differently exposed to MFs, as in the case of total carbohydrates in *Citrus aurantifolia* [17], and of photosynthetic pigments in *Zea mays* [50] and *Robinia pseudoacacia* [51]. Generally, MF exposure disturbs biological system stability, acting as a stressful factor translating into an increase in photosynthetic pigments as a defense mechanism [7]. In the present study, however, despite the generally slower metabolism observed in the exposed plants, no increase in photosynthetic pigments nor in anthocyanins, mainly produced to develop resistance to a number of environmental stresses [52], was observed (Figure 1), suggesting no adverse effect of the applied MFs triggering defense mechanisms. Such a hypothesis is also supported by the similar or better morphological characteristics of the exposed mature plants as compared to the respective control plants (data not shown).

The induced average increases in C/N and C/P ratios (Figure 2), largely dependent on N and P reductions in the exposed plants (Figure 1), indicate the production of litter more recalcitrant to the decomposition process [53,54]. Since these traits explain the majority of the variation in decomposition rates [53] and in global C cycling [54], the obtained findings suggest that weak MF treatments may be useful to enhance soil C storage and reduce CO_2 emissions, a priority objective for the United Nation Decade of Ecosystem Restoration (2021–2030) (https://www.decadeonrestoration.org/; accessed 8 January 2023) aimed to "prevent, halt and reverse the degradation of ecosystems on every continent", also fighting climate change. In the context of sustainable agriculture, such considerations further strengthen the need for carefully evaluating plant quality, especially when emerging technologies are adopted, in order to ensure not only consumer, but also ecosystem health.

4. Conclusions

The choice of evaluating the effects of weak magnetic fields on plants through the investigation of different species and exposure conditions (life stage and time), as well as the meta-analytical approach adopted, allow drawing generalizable ecological effects. Indeed, in spite of the large variations in plant responses to weak magnetic fields, general reductions in essential, beneficial and non-useful element concentrations, as well as in biomolecule synthesis, were observed, indicating measurable effects of magnetic fields on plant chemistry. The large variability makes such effects hardly predictable, probably because of their modulation by several direct and indirect mechanisms, but they have, nonetheless, the potential to affect PTE transfer along food chains and matter cycle. An example is provided by the larger relative reduction in non-useful elements (bioaccumulable and biomagnificable) in respect to the essential elements, representing a meaningful finding for the higher trophic levels, including people using crops as food. Likewise, important ecological implications of plant exposure to weak magnetic fields regard the quality of the produced litter, which is less decomposable due to higher C/N and C/P ratios and with potential benefits for longer soil C storage and reduced CO_2 emissions.

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Abbreviations

The following abbreviations are used in this manuscript:

MF	Magnetic Field
RR	Response Ratio
PTE	Potentially Toxic Element
NBI	Nitrogen Balance Index

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