Physic-chemical, phytochemical characterization and *in vitro* antioxidant activity of *CUCUMIS SATIVUS* l. vAR. mARKET MORE with magneto priming

Caracterización físico-química, fitoquímica y actividad antioxidante *in vitro* de *CUCUMIS SATIVUS* l. vAR. mARKET MORE con magneto-priming

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ABSTRACT

The aim of this study was to evaluate the effect of a static magnetic field with a magnetic induction of 60-110 mT on the bioactive compounds and antioxidant properties of *Cucumis sativus* L. var market more. Alcoholic and aqueous extracts were used to determine the chemical-physical properties, concentration of phenolic compounds, and antioxidant activity in vitro. Seeds treated with a static magnetic field showed an increase in mineral concentrations compared to untreated seeds. The same behavior was obtained in the physicochemical properties between the aqueous and ethanolic extracts. For phenolic compounds and antioxidant activity by DPPH radical scavenging, the aqueous extracts of seeds treated with a static magnetic field presented higher values with respect to the control ethanolic extract. These results highlight the importance of using the static magnetic field in the quality and nutritional value of the seeds of this species

Keywords: *Cucumis sativus;* Alcoholic and aqueous extracts; mineral concentrations.

RESUMEN

El objetivo del estudio fue evaluar el efecto de un campo magnético estático con una inducción magnética de 60-110 mT sobre los compuestos bioactivos y propiedades antioxidantes de las semillas *Cucumis sativus* L. var. market more. Se utilizaron extractos alcohólicos y acuosos para determinar las propiedades químico-físicas, concentración de compuestos fenólicos y actividad antioxidante *in vitro*. Las semillas tratadas con un campo magnético estático mostraron un aumento en las concentraciones de minerales en comparación con las semillas no tratadas. El mismo comportamiento se obtuvo en las propiedades fisicoquímicas entre los extractos acuosos y etanólicos. Para los compuestos fenólicos y la actividad antioxidante por secuestro de radical DPPH los extractos acuosos de semillas tratadas con campo magnético estático presentaron valores superiores con respecto al

extracto etanólico control. Estos resultados destacan la importancia de utilizar el campo magnético estático en la calidad y valor nutricional de las semillas de esta especie.

Palabras clave: *Cucumis sativus;* extractos alcohólicos y acuosos; concentraciones de minerales

Introduction

Magnetic fields are only one of several factors affecting plant growth and development. Crop yields in general and uniformity are raised by the application of chemicals (such as hormones) in pre-sowing seed treatment, although they are considered to be very effective and invasive, ecologically incorrect and difficult to apply. During the last decades, the discovery of hormones and chemical fertilizers played an important factor in yield improvement in plant breeding, but the current use of chemical fertilizers and other materials has been controversial, which has led to the use of ecologically friendly alternative treatments with reduced costs, such as gamma rays, laser, electron beam, microwaves, magnetic fields, and radio frequency energies to cause seed biostimulation. [(1)]. In recent years, there has been a growing interest in the study of the effects of magnetic fields on biological systems and their potential to improve the nutritional and pharmacological properties of agricultural products.

Other authors such as [(2, 3)] demonstrated that magnetic field could reduce oxidative damage in plants due to the activities of antioxidant enzymes such as peroxidase, polyphenol oxidase (PPO), superoxide dismutase (SOD) and catalase (CAT) in plant cells. They emphasized that magnetic fields affected the antioxidant activity and increased the activity of free radical ions in plants [(1)]. Magnetic field treatment has also been used on seeds and became widely used in the agricultural sector. The application of the magnetic field is pre-sowing, and was named "magneto-priming", it is a priming treatment to dry seeds and non-destructive. It has been reported to increase the germination rate and seedling vigor of many crops. Research has also been reported on the metabolic changes that occur during germination in different types of seeds in response to magneto-priming in non-stressed environments[(1)].

In this sense, the magnetic field constitutes one of the safe and affordable pre-sowing physical seed treatments that could improve germination and seedling establishment, stimulate growth and, therefore, increase crop yields [(4)]. Shine, M. B., *et al.* demonstrated in seeds of that *Glycina max* L. (soybean) treated with static magnetic fields of 150 and 200 mT for 1 h presented an increased reactive oxygen species (ROS) production mediated by cell wall peroxidase, while ascorbic acid content, superoxide dismutase and ascorbate peroxidase activity decreased in the hypocotyl of germinating seeds[(5)]. An increase in cytosolic peroxidase activity indicated that this antioxidant enzyme had a vital role in removing the increased H_2O_2 produced in seedlings of magnetically treated seeds. Therefore, these studies contribute to our first report on the biochemical basis of enhanced germination and seedling growth in magnetically treated soybean seeds in relation to increased ROS production.

The antioxidant activity of plant products has been associated with their content of bioactive metabolites, such as polyphenols. These compounds have been shown to have antioxidant properties, which means that they can neutralize free radicals and protect cells from oxidative damage [(5,6)]. *Cucumis sativus* L. seeds are the subject of growing interest due to their potential as a source of bioactive compounds and antioxidants. Polyphenols and other antioxidant compounds present in these seeds have been the subject of studies due to their ability to combat oxidative stress and prevent oxidative stress-related diseases [(7)].

Among the secondary metabolites most relevant to health present in the *Cucumis sativus* species and in the family in general, are saponins, tannins, carotenoids, terpenoids, where cucurbitacins stand out, a metabolite to which the bitter taste of cucumber is attributed, being found in lowest concentrations in the seeds [8, 9]. In general, cucurbitacins at high concentrations show cytotoxicity, but in rights concentrations, they show potential to treat pathologies such as inflammation and autoimmune diseases [8]. In addition to antioxidant compounds, the seeds of *Cucumis sativus* L. also contain minerals important for human health, such as calcium, magnesium and zinc, which play a crucial role in various biological functions and contribute to maintaining a proper balance in the body. [7, 8, 9, 10]

In this context, the species *Cucumis sativus* L. commonly known as cucumber, its seeds have attracted special attention due to their content of phenolic compounds, which have antioxidant properties and health benefits [1]. Various previous studies have explored the chemical components and antioxidant properties of these seeds through *in vitro* assays [9], evaluating their potential to prevent diseases related to oxidative stress, and chemical analysis have been carried out to identify the types and concentrations of phenols, steroids, tannins, saponins, carotenoids, in addition to having a 50 % IC of antioxidants of 6555 ppm present in the cucumber seeds [9,10, 11, 12].The study demonstrates with scientific evidence the effect of magnetic treatment as a physical abiotic factor on the phenolic compounds and antioxidant properties of seed extracts. It can be employed in the development of innovative strategies to improve the quality and nutritional value of agricultural products, as well as to explore its potential application in the food industry and human health.

The aim of this study was to evaluate the physic-chemical, phytochemical properties and *in vitro* antioxidant activity of seeds of *Cucumis sativus* L. var. market more with magneto-priming.

Materials and methods

The research was carried out at National Center of Electromagnetism Applied, located at Avenida Las Américas s/n, Santiago de Cuba, Cuba. Laboratory of Plant Biotechnology and Laboratory of Environmental Sciences of the University of Hasselt in Belgium.

Preparation of extracts

Seeds of *Cucumis sativus* L. var. market more from Provincial Seed Company of Santiago de Cuba were used, with 80 % germination. Extracts of 100 seeds per experimental group were prepared and placed in a 100 mL erlenmeyer flask for disinfection. Disinfection was performed by washing with 1 % commercial detergent for 15 minutes in a rotary shaker, followed by three rinses with sterile distilled water for more than three minutes. The seeds were then disinfected with sodium hypochlorite (0,1 %) for 15 minutes on a rotary shaker and rinsed three times with sterile distilled water. Then, the seeds were transported to the laminar flow cabinet and immersed in ethanol (70 %) for 5 minutes, rinsed three times with sterile distilled water. For each group, four 6 cm (10,5 x 1,7 cm) Petri dishes were prepared with sterile filter paper moistened with 2 mL of sterile distilled water. Twenty-five seeds were placed per 7 cm plate.

All experiments were performed under laboratory conditions, at a constant temperature of 28 ± 2 °C and a relative humidity of 90 ± 2 %. Illumination was determined with a luxmeter (model Daylight TL-D 36W 154-765 µmol-2, China), the average value obtained was 299,75 μmol^{-2}.

Different experimental groups were:

- Group 1 (AE Control): aqueous extract of seeds not treated with static magnetic field

- Group 2 (AE SMF): aqueous extract of seeds treated with static magnetic field

- Group 3 (EE Control): ethanolic extract of seeds not treated with static magnetic field.

- Group 4 (EE SMF): ethanolic extract of seeds treated with static magnetic field.

The static magnetic field treatment was applied with a magnetic induction range of 60-110 mT, and exposure time of one hour for two days. The first application was after seed sterilization and the second was 24 hours after the first application. After completing the magnetic treatment, three grams of *Cucumis sativus* L. var. market more seeds were weighed for each experimental group, a control without magnetic treatment (CTR) and with static magnetic treatment (SMF), to be powdered and placed in Erlenmeyer flask, diluted in 100mL of 96% ethanol (group 1 and 2) and sterile distilled water (group 3 and 4). Subsequently, the seed powders diluted in sterile distilled water were boiled for three minutes and then placed in maceration for 48 hours on a rotary shaker. At the same maceration time, the diluted seed powders were placed in 96% ethanol. The extracts were then filtered and centrifuged for subsequent phytochemical characterization Albayrak, S., *et al* [13]. The extracts obtained were stored at 4ºC until use. Taking into account that cooking processes affect the stability of the extracts, they were prepared again prior to the assays.

Characterization of physic-chemical parameters of *Cucumis sativus* L. var. market more seeds

a) Physical parameters

Total solids

From the previously homogenized sample of each experimental group, 5 mL were transferred to a clean, dry and previously tared porcelain capsule; it was placed in a water bath and evaporated until the residue was apparently dry. It was then transferred to an oven, at a temperature of 105 ± 2 ºC, for 3 hours. The capsule was removed from the oven, placed in a desiccator until it reached room temperature and weighed. The previous process was repeated, but using only 60 minutes of drying, until a constant mass was obtained [14]**.**

Total solids (TS) were calculated using the equation (1):

$$TS=\frac{Pr-P}{V\cdot 100} \qquad (1)$$

Where: Pr is the mass of the capsule plus residue (g); P is the mass of the empty capsule (g); V is the volume of the test portion (mL); 100 is a mathematical factor.

Values were approximated to the nearest tenth. The amount of total solids g100 mL^{-1} was reported. The assay was performed in triplicate. The measurement of **total dissolved solids** (TDS) and **conductivity** were obtained using a portable pH conductivity meter (Mettler Toledo, China) with glass electrodes, previously calibrated.

Determination of pH

The potentiometric method was used, which is based on the arrangement of two electrodes, a reference and an indicator, and a potential measuring device (Mettler Toledo pH meter, China) was used. These, working together, can make an adjusted measurement of the potential of a cell with respect to a reference value by MINSAP [(15)].

b) Chemical parameters

Minerals assessment by Atomic Emission Spectroscopy with Inductively Coupled Plasma (ICP-OES)

The chemical characterization of *Cucumis sativus* L. seed extracts from both groups (CTR and SMF), was performed through isonomic studies according to the method of Phan-Thien, K.-Y., *et al.*[(16)], using the standard procedures Lab. Ciencias Ambientales, H [(17)]. This method allowed determining the concentration of ions present in the seeds of *Cucumis sativus* L. The analysis is performed by inductively coupled plasma optical emission spectroscopy (ICP-OES). The content of mineral elements is determined by the method of extraction of metals in heating block, through Optical Emission Spectrometry, according to standardized procedures Lab. Ciencias Ambientales, H [(17)].

Test tubes were prepared and weighed for the collection of seed extracts. 3 blank samples and 3 reference samples (spinach) were included, then the block was heated to 40°C and 1 mL of nitric acid (HNO_3) suprapur was added to each tube and allowed to stand overnight. The temperature was then increased to 60°C and then to 110°C, allowing the samples to dry for approximately 4 hours. Subsequently, 1 mL of HNO_3 suprapur was added, the samples were dried again at 110°C and this step was repeated once more. 1 mL of 37% suprapur hydrochloric acid (HCl) was added to each sample and dried at 110°C for approximately 3 hours. Finally, a 20 % HCl solution was prepared and 500 µL was added to the short tubes. The samples were transferred to sterile tubes and analyzed on the ICP.

Phytochemical screening

Phytochemical screening was performed on all the extracts of the experimental groups according to the methodology described by [(18)]. Simple, rapid and selective tests or techniques were used for the determination of the different secondary metabolites present in the extracts. The selected assays were Lieberman-Burchard, sulfuric acid (H_2SO_4) conc-Shinoda, iron (III) chloride ($FeCl_3$), Dragendorff and Fheling

Phenolic compounds assessment

The determination of phenolic compounds was determined by the Follin-Ciocalteau assay, based on the ability of phenols to act in oxidation/reduction reactions, thanks to the reducing character of the Follin-Ciocalteau reagent that uses as a mixture, phosphowolframic and phosphomolybdic acids in a basic medium [(19)]. These are reduced by oxidizing the phenolic compounds, which produces blue oxides of wolfram (W_8O_{23}) and molybdenum (Mo_8O_{23}). For this purpose, aqueous and ethanolic extracts were established at a concentration of $3mgmL^{-1}$. Gallic acid was used as a standard to establish the calibration curve. The results were shown in milligrams of gallic acid equivalent g^{-1} (mg $GAEg^{-1}$).

In vitro antioxidant activity assessment in seed extracts of *Cucumis sativus* L. var. market more with magneto-priming

Ferric Reducing Antioxidant Power Assay (FRAP)

This method measures the ability of antioxidants to reduce ferric iron. It is based on the reduction of the ferric iron complex and 2,4,6-tripyridyl-s-triazine (TPTZ) to the ferrous form at low pH. The absorbance of Fe is measured spectrophotometrically. 50 µL of the aqueous extract and leaves from each experimental group, 535 µL of HCl (37%) (40mM) are used. A solution of TPTZ (10 mM) is prepared with 10 mL of HCl (40 mM) and one of FeCl3-6H2O (20 mM). In addition to an acetate buffer (0.3 mM) at pH 3.6. FRAP is prepared daily with TPTZ in FeCl3-6H2O (20 mM) and acetate buffer. This preparation is kept during the whole process in a water bath at 37ºC. A calibration curve is performed with 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) solutions, at 15.625; 31.25; 62.5; 125; 250; 500; 1000 mM, expressed equivalent Trolox capacity (µM Troloxg-1 dry weight), according to the method of Peñarrieta, J. M., *et al* [20]. All samples are performed in triplicate.

2,2-diphenyl-1-picrylhydrazyl (DPPH) radical blotting method

The technique consisted of incubation for 30 minutes at 37 ºC of the aqueous and ethanolic extracts of each experimental group in an ethanolic solution of DPPH, at a concentration of 0.3 mM, and an absorbance reading at 515 nm [21]. The sample solution (1.0 $mgmL^{-1}$) was diluted to concentrations of 250, 125, 50, 25, 10 and 5 μgmL^{-1}. The percentage inhibition of the tested sample on DPPH radicals was calculated, converting to percentage antioxidant activity (AA), with the use of the formula given below. All samples were performed in triplicate.

$$AA\% = 100 - \frac{[(Abs_{muestra} - Abs_{blanco})100]}{Abs_{DPPH}} \qquad (2)$$

The DPPH solution was prepared on the same day as the analysis was performed, to measure a full 96-well plate (exactly 18.240 mL of 0.11 mM DPPH solution was used). 43.38 mg of DPPH (MW = 394.32 $gmol^{-1}$) was dissolved in 100 mL of methanol to obtain a 1.1 mM DPPH solution. This was then diluted 10x in methanol to obtain the 0,11 mM DPPH working solution. Preparation of the solutions for the standard curve was performed with trolox reagent (1000 µM 6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid). Six concentrations were established for the curve in µM (0, 200, 400, 600, 600, 800, 1000).

• Radical scavenging activity is calculated by the equation (3):

$$\%\ de\ inhibition\ = \left(\frac{Ab - As}{Ab}\right) \times 100\ \% \qquad (3)$$

where Ab is the absorbance of the blank sample and As is the absorbance of a treatment sample. Results were expressed as µM ascorbate equivalents (AE, µM ascorbic acid) g^{-1} FW by comparison with a standard Trolox curve.

Statistical analysis

A descriptive statistical analysis was performed, a completely randomized design was used and the Shapiro Wilk test was used for data normality. The comparison of means of the treatments was by means of a simple classificatory ANOVA with HSD test and Tukey's test with a probability of 99 %. Statgraphics 19 was used and the graphs were plotted with Prisma version 7.

Results and discussion

Determination of physic-chemical parameters of *Cucumis sativus* L. var. Market more seeds

Table 1 shows the physic-chemical characteristics of the seed extracts *C. sativus* L. Market more. The pH results were obtained values that corroborate the weak acid characteristics of the substances extracted in these extracts, such as flavonoids, phenols and tannins, terpenes, among others. It seems that compounds extracted with the same extraction method maintain the weak acid characteristics. The conductivity did not show statistically significant differences between the CTR and SMF groups of each extract, however, when comparing the co-conductivities between the aqueous extract and the ethanolic extract, the conductivity is higher in the former. This is because water has a greater capacity than alcohol to dissociate into charged ions.

Regarding total dissolved solids (TDS) and total solids (TS), the results showed no statistically significant differences between the untreated (CTR) and treated (SMF) groups. Furthermore, similar to the results obtained in conductivity, the differences between the types of extracts were marked, this may be a consequence of the extraction of the polar compounds by the aqueous extract having been more efficient than the extraction with alcohol. However, it is important to keep in mind that many relevant compounds in *Cucumis sativus* seeds with relevance to health are of oily origin, so it is important to take this into account depending on the study in which it is decided to apply an extraction method or other.

Table 1. Physic-chemical characteristics of the extract of *Cucumis sativus* L. var. market more.

Treatment		pH	Conductivity ($mScm^{-1}$)	TDS (mgL^{-1})	TS (gmL^{-1})
AE	Control	5,92 ±0,001	39,77 ±0,085	20,13 ±0,024	1,09 ±0.007
	SMF	6,13 ±0,010	41,07 ±0,062	20,77 ±0,024	1,10±0.007
EE	Control	6,05 ±0,004	1.22 ±0,001	0,61 ±0,001	1.26±0.008
	SMF	6,30 ±0,003	0.80 ±0,003	0,40 ±0,001	1.27±0.009

AE: Aqueous Extract, EE: Ethanolic Extract, total dissolved solid (TDS), Total solid (TS). The mean ± standard error is represented. ($p > 0,05$)

These results show that magnetopriming did not change the physicochemical properties of the Cucumis sativus seed extract significantly, when compared with the control group. However, differences were observed between the aqueous and ethanolic extracts, suggesting that the extraction method can influence the physicochemical properties of the extract since it is possible to extract less polar compounds using ethanolic extracts, this may be relevant in terms of applicability. and specific characteristics of each type of extract.

Phytochemical screening

Table 2 shows the results of the phytochemical screening in the aqueous and ethanolic extracts where the various metabolites are found. It should be noted that the aqueous and ethanolic extracts from seeds treated with magneto-priming showed different colorations according to the analysis. In the Lieberman-Burchard analysis, a dark green color was obtained, which showed the presence of triterpenic and/or steroidal compounds; both the aqueous ethanolic extract from the seeds that were magneto-primed showed a double presence of these biomolecules. The same behavior occurred with $FeCl_3$ where the aqueous and ethanolic extracts had a dark green color indicating the presence of phenols and tannins, with a more marked presence in the extracts with SMF. Mandey, J. S., *et al.*[12] applied a phytochemical screening of *Cucumber seeds* where they obtained the presence of flavonoids, tannins, saponins, and steroids. Except for saponins, there were coincidences with this study, while these variations could be related to the cucumber varieties used.

Table 2. Phytochemical screening of seed extracts of *Cucumis sativus L.* var market more with magneto-priming.

Metabolites	Essays	Evidence	AE Control	AE SMF	EE Control	EE SMF
Phenols and Tannins	$FeCl_3$	Dark green	(+)	(+++)	(+)	(++)
Flavonoids	H_2SO_4 conc	Intense yellow	(+)	(+)	(+)	(+)
	Shinoda	Red	(+)	(+)	(+)	(+)
Triterpenes and steroids	Lieberman-Burchard	Dark green	(+)	(++)	(+)	(+++)
Saponin	Foam	No foam	(-)	(-)	(-)	(-)
Alkaloids	Dragendorff	Brick red precipitate	(-)	(-)	(-)	(-)
Sugars	Fheling	Precipitate	(+)	(+)	(+)	(+)

AE: Aqueous Extract, EE: Ethanolic Extract. Absence (-); Marked presence (++), abundance (+++),

Minerals assessment by Atomic Emission Spectroscopy w ith Inductively Coupled Plasma (ICP-OES)

The Table 3 show the average values reflect that although there were no statistically significant differences, the seeds treated with magneto-priming have a higher average value of minerals. When comparing these results with those obtained by Murthy, H. N., *et al.*[22] In three varieties of *Cucumis sativus*, some differences in concentrations are observed. While the lowest levels were obtained in the minerals Ca, K, Mg, Na, the highest were in Cu, Fe, Mn, S, however the P was very similar to the other varieties, mainly *Cucumis sativus* var. hardwickii

Minerals	AEControl (mgkg^{-1})	AESMF (mgkg^{-1})	EE Control (mgkg^{-1})	EE SMF (mgkg^{-1})

Minerals have high relevance in the physiology of organisms. They can act as the maintenance of physical integrity, in environmental responses, forming part of complex molecules such as phospholipids, phosphorylated sugars, nucleic acids, nucleic acids. Also as cofactor of oxidative enzymes such as Superoxide Dismutase (SOD), in catalytic groups and in the transport of substances [23].

Ca	642.131 ±89.075	895.140 ±27.003	764.188 ±112.243	791.276 ±57.267
Cu	14.320 ±1.736	18.278 ±0.108	17.242 ±1.801	20.667 ±5.404
Fe	129.701 ±15.275	167.355 ±11.858	135.173 ±10.694	146.051 ±17.484
K	4397.1183 ±498.714	4852.212 ±65.113	4972.501 ±382.419	5015.062 ±439.910
Mg	3423.518 ±488.483	4383.002 ±81.436	3940.710 ±438.802	3918.323 ±324.005
Mn	25.111 ±3.281	32.320 ±0.335	29.741 ±3.600	28.774 ±2.453
Na	177.236 ±0.01	196.701 ±3.396	206.856 ±16.442	226.989 ±16.969
P	9523.653 ±1281.583	11553.850 ±231.852	11395.918 ±1191.368	11185.672 ±987.204
S	2697.011 ±358.340	3370.415 ±56.697	3171.525 ±353.094	3131.999 ±291.430
S^2	60.258 ±7.752	74.549 ±0.942	69.254 ±7.312	70.849 ±6.708

Table 3. Mineral concentration of *Cucumis*

Plasma (ICP-OES) ($p > 0.05$)The variations in the average values obtained may be due to the mobilization generated by the effect of the magnetic field inside the seeds. The ferrite oscillation hypothesis attempts to explain this process, through the effect of magnetic fields on ion mobility and absorption [(24)]. Goldsworthy's membrane potential change hypothesis proposes that the effect of the magnetic field modifies membrane permeability by selectively removing $^{Ca2+}$ and replacing it with other cations, making it more unstable and favoring greater water flux [(24, 25)].

The minerals with the highest concentrations were P, K, Mg, S, while the lowest were S^2 isotope, Cu, Mn. The highest concentrations were more frequent in aqueous extracts than in ethanolic extracts. Niyi, O. H., *et al.* [(26)] determined the minerals composition present in *Cucumis sativus L.* seeds, with slightly higher values than those obtained in the variety evaluated in this research such as Ca, Cu, K and Na, while the minerals Fe, Mg y Mn was higher in the variety Market more.

<u>Phenolic compounds assessment</u>

The results of phenolic concentration present in the cucumber seed extracts are shown in figure 1, where there were statistically significant differences for a 95 % probability, between the aqueous and ethanolic extracts, the highest concentration was found in the aqueous extracts of seeds treated with magneto-priming with 8,31 mg $GAEg^{-1}$ and the control with 7.86 mg $GAEg^{-1}$, while the ethanolic extract of the treated group was 7.06 mg $GAEg^{-1}$ and the control was the lowest of all with 3.29 mg $GAEg^{-1}$.

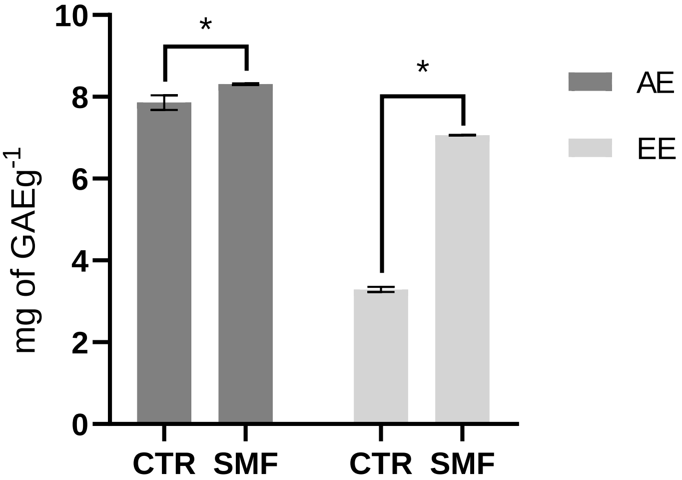

Fig. 1- Phenolic compounds in aqueous and ethanolic extracts of seeds of *Cucumis sativus* var. market more, aqueous (AE) and ethanolic (EE) extracts of treated (SMF) and untreated (CTR) seeds. Mean ± standard error ($p < 0.05$)

These results indicate that the aqueous extraction method for both the SMF treated group and the CTR control favored the release and extraction of phenolic compounds present in the seeds cucumber It is important to note that the ethanolic extract of the control group showed the lowest concentration of phenolic compounds, which represents approximately 58% lower than the aqueous control. This may be due to the influence of the extraction method used. There were also statistically significant differences between the control and treated groups in both extracts. Abdollahi, F., *et al.*(27) evaluated the effect of static magnetic fields on the antioxidant systems of almond seeds. They found that the treatment increased the production of phenolic compounds and enhanced the activity of antioxidant enzymes. These results suggest that SMFs can induce free radical production in almond seeds, which in turn triggers antioxidant responses to protect them, leading to increased production of secondary metabolites to decrease oxidative stress.

These results demonstrated that the aqueous extracts of cucumber seeds presented a higher concentration of phenolic compounds compared to the ethanolic extract of the control group. These phenolic compounds are known for their antioxidant properties and health benefits. However, it is important to note that more research is required to fully understand the profile of phenolic compounds in cucumber seed extracts and their relevance in terms of potential health applications. In addition, the use of magneto-priming significantly increased the concentration of phenols, both in aqueous and ethanolic extracts, which could have important health benefits.

In vitro antioxidant activity assessment of *Cucumis sativus* L. var. market more seeds with magneto-priming

The results of the antioxidant capacity through reduction of ferric ions and free radical scavenging activity DPPH in the extracts (aqueous and ethanolic) of Cucumis sativus seeds treated with static magnetic field (SMF) and untreated (CTR) are shown in the figures 2 and 3, respectively. Both groups showed antioxidant capacity, with the treated group showing greater reducing power and radical sequestration with statistically significant differences compared to the control group.

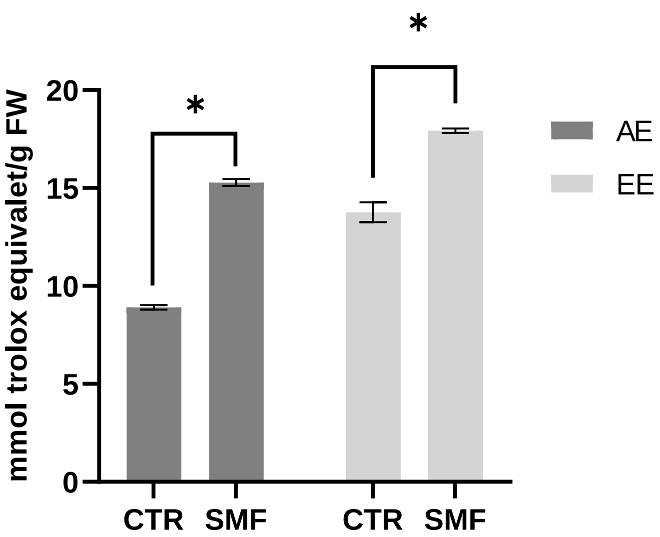

Fig. 2- Free radical reduction method by FRAP of aqueous (AE) and ethanolic (EE) extracts of treated (SMF) and untreated (CTR) seeds. Mean values ± SE. * indicate statistically significant differences. The analysis was performed in triplicate. One-way ANOVA LSD test. ($p<0,05$)

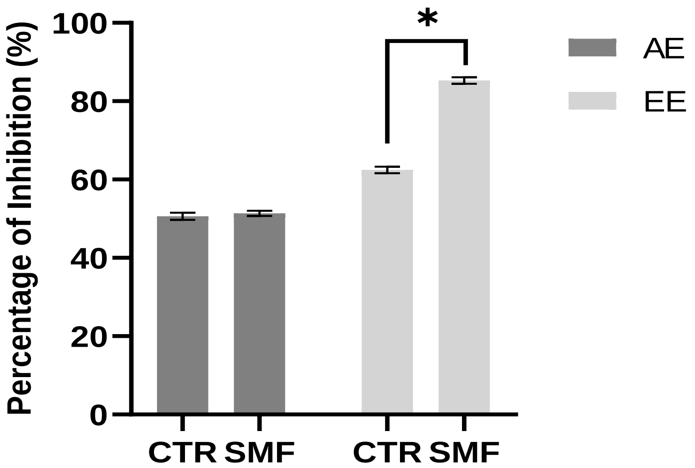

Fig. 3- Free radical scavenging method by DPPH of aqueous (AE) and ethanolic (EE) extracts of treated (SMF) and untreated (CTR) seeds. Mean values ± SE. * indicate statistically significant differences. The analysis was performed in triplicate. One-way ANOVA LSD test. ($p<0,05$)

The results showed that both aqueous and ethanolic extracts of *Cucumis sativus* var market more seeds showed antioxidant capacity as free radical reductant and scavenger. The group treated with SMF in both types of extracts exhibited higher reducing and scavenging power compared to the untreated group. These differences were statistically significant, indicating a positive impact of the static magnetic field treatment on the antioxidant capacity of the extracts by promoting the production of these metabolites as a consequence of the stress generated by the applied static magnetic field. This effect has been evidenced by other authors [28, 29].

The ethanolic extract of seeds treated with SMF, its reducing power increased by 37.54% compared to the control group. This finding suggests that the static magnetic field has the ability to enhance the antioxidant capacity of the ethanolic extract of *Cucumis sativus seeds*. However, it is interesting to note that, although the percentage increase was lower, the aqueous extract treated with SMF showed a 71.53% increase in its reducing power compared to the control group. This indicates that the aqueous extract also benefits from static magnetic field treatment, although to a lesser extent than the ethanolic extract.

Phenolic compounds are known for their antioxidant properties and other health benefits, so the higher concentration of these compounds in the aqueous extracts suggests that these extracts may have a higher antioxidant potential compared to the ethanolic extract of the control group. While the free radical scavenging power evaluated by DPPH, the ethanolic extract of treated seeds increased by 22.83% compared to the control group. While, in the aqueous extract, there were no statistically significant differences. These elements are of great value since a future formulation with ethanolic extracts may favor a greater antioxidant effect in the seed extract of Cucumis sativus of this variety. In addition, the application of the magnetic field during the first stage of germination evidence that it has effects on the production of antioxidant compounds (28, 30-32), which increases its effect evaluated against the sequestration and reduction of free radicals in vitro evaluated in this research. These results, which show the capacity to sequester and reduce radicals, are consistent with the increase in the concentration of phenolic compounds (figure 1) in the treated groups (SMF) with respect to the control groups (CTR).

These results suggest that both water-soluble and fat-soluble compounds present in Cucumis sativus seeds are susceptible to the effect of the static magnetic field, resulting in a significant increase in their antioxidant capacity. This effect can be attributed to a number of factors that attempt to be explained by several of the hypotheses described so far. The "radical pair mechanism" hypothesis explains how magnetic fields can affect enzymatic reactions involving free radical intermediates, in addition to the fact that magnetic fields can influence the production of reactive oxygen species (ROS), which can lead to the initiation of oxidative stress in cells. It also suggests that magnetic fields can affect the activity of enzymatic antioxidants and the expression of their genes. The internal magnetic fields generated by nuclear spins may influence singlet-triplet conversion of free radical pairs where the interaction between unpaired electron spins, the spins of adjacent nuclei and the magnetic field (Zeeman interaction) determines the energy involved in the recombination of radical pairs (24).

Magnetic fields can have a significant impact on cellular processes related to oxidative stress and antioxidant activity through mechanisms involving free radical pairs and their magnetic interactions. Our findings demonstrate that static magnetic field treatment significantly enhances the antioxidant capacity of Cucumis sativus seed extracts, both in their aqueous and ethanolic forms. These results support the hypothesis that static magnetic field can enhance the antioxidant properties of bioactive compounds present in Cucumis sativus seeds, which could have important implications in the food and health industry.

Conclusions

The results of this study reveal that static magnetic field treatment can have significant effects on Cucumis sativus seed extracts. A trend toward higher mineral concentrations was observed in the magnetic field-treated seeds compared to untreated seeds. In addition, the extraction method was found to influence the physicochemical properties of the extracts, with differences between aqueous and ethanolic extracts. As for phenolic compounds, a higher concentration was found in the aqueous extracts, suggesting a higher antioxidant potential compared to the ethanolic extract. Finally, both aqueous extract and ethanolic extract of seeds treated with magnetic field showed higher antioxidant capacity compared to the control group. These findings support the hypothesis that static magnetic field can enhance the antioxidant properties of bioactive compounds in Cucumis sativus seeds, which could have important implications in the food and health industry.

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Conflicts of interest

There are no conflicts to declare

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