Rev. Cubana Quím.
Vol.30, no.2, mayo-agosto, 2018, págs. 232-242, e-ISSN: 2224-5421

Determinación fitoquímica de frutos de S*olanum lycopersicum L.* Irrigados con agua tratada con campo magnético estático

Phytochemical determination of Solanum lycopersicum L. fruits irrigated with water treated with static magnetic field

***Dra. C. Albys E. Ferrer-Dubois* [I], *Dra. C. Yilan Fung-Boix* [I], *Dra. C. Elizabeth Isaac-Alemán* [I], *Dra. C. Natalie Beenaerts* [II], *Dra. C. Ann Cuypers* [II]**

albys@uo.edu.cu; albysf@gmail.com

[I] Centro Nacional de Electromagnetismo Aplicado, Universidad de Oriente, Santiago de Cuba, Cuba; [II] Center for Environmental Sciences. Hasselt University. Belgium

Recibido: 6 de noviembre de 2017 Aprobado: 10 de enero de 2018

Resumen

Se determinaron los compuestos fitoquímicos de los frutos de *Solanum lycopersicum L.* provenientes de plantas irrigadas bajo la acción de agua tratada con un campo magnético estático (CME), entre 20 y 200 mT. Se utilizaron dos grupos experimentales con 25 plantas cada uno. El grupo control recibió el riego con agua no tratada y el otro grupo fue irrigado con agua tratada con CME. Se cosecharon los frutos maduros y se prepararon extractos acuosos y etanólicos. A estos extractos se les realizó el tamizaje fitoquímico. Los extractos acuosos preparados a partir de los frutos irrigados del grupo con CME revelaron cualitativamente una marcada presenciade carotenoides, fenoles, taninos, carbohidratos y flavonoides. Estos resultados mostraron que el CME utilizado favorece la mayor extracción de estos metabolitos los cuales tienen un gran valor nutricional y potencial antioxidante, lo que contribuye a las propiedades farmacológicas de los frutos de *S. lycopersicum L.*

Palabras clave: tomate, tamizaje fitoquímico, extractos, campos magnéticos, metabolitos.

Abstract

The phytochemical compounds of *Solanum lycopersicum L.* fruits from irrigated plants under the action of water treated with a static magnetic field (SMF), between 20 and 200 mT were determined. Two experimental groups with 25 plants each were used. The control group received irrigation with untreated water and the other group was irrigated with water treated with SMF. The ripe fruits were harvested and aqueous and ethanolic extracts were prepared. These extracts were subjected to phytochemical screening. The aqueous extracts prepared from the irrigated fruits of the group with SMF showed qualitatively a marked presence of carotenoids, phenols, tannins, carbohydrates and flavonoids. These results showed that the SMF used favors the greater extraction of these metabolites which have a high nutritional value and antioxidant potential, which contributes to the pharmacological properties of the fruits of *S. lycopersicum L.*

Keywords: tomato, phytochemical screening, extracts, magnetic fields, metabolites

Introduction

Dietary recommendations in recent years suggest an increase in the consumption of foods containing phytochemicals. They provide beneficial effects for human health and play an important role in the prevention of chronic diseases [1].

One of the world's most consumed plants is *Solanum lycopersicum L.* (tomato), which is considered to nutraceuticaldue to their antioxidant activity [2]. It is an important source of chemical compounds, specifically secondary metabolites with antioxidant action, such as polyphenols, hydroxycinnamic acids, carotenoids and vitamins, among others. These compounds modulate metabolic processes in the human body and show recognized beneficial effects on human health.In turn, they can prevent the generation of reactive species that affect, among other causes, the onset of cardiovascular diseases and neurodegenerative diseases [3] and some types of cancer [4].

In addition, due to its genetic and molecular advantages, the tomatois considered a biological model for morphological, physiological and genetic studies [5]. Thanks to the experience derived from its use, it can be extrapolated to other horticultural species with some similarity [6]. An aspect that has justified its use to evaluate the effects of the MF. This plant has been treated with MF to improve plant morphology and physiology [7]. A high content of polyphenols and carotenoids was evidenced by using high-intensity pulsating electric fields in processed fruit juices from tomato [8]. The experimental studies provide evidence of the benefits related to the phytochemical content of the fruit of this species; however, any variation under the action of water treated with SMF between 20 and 200 mT has not been recorded.Therefore the objective of this research was to determine the phytochemical compounds of the aqueous and ethanolic extracts of fruits of *Solanum lycopersicum* L. from plants irrigated with water treated with a SMF between 20 and 200 Mt.

Materials and methods

The research was carried out at the National Center for Applied Electromagnetism (CNEA, Spanish acronym) in Santiago de Cuba and at the Center for Environmental Sciences at the Hasselt University, Belgium. The Tomato, *Solanum lycopersicum L.*, Aegean hybrid, was used as biological model.

Seeds with good phytosanitary status and certified by the Provincial Laboratory of Seeds attached to the Ministry of Agriculture (MINAGRI, Spanish acronym) in

Santiago de Cuba, were used. The plants were cultivated under semicontrolled conditions at the "Veguita" greenhouses (a suburban area of Santiago de Cuba) and all care required for the cultivation of tomato wasgiven in compliance with MINAGRI specifications [9].The temperature in the experimental growth area were in the ranged from 27 to 30 °C. The approximate value of precipitation was 68.04 mm and the relative humidity 70.32 % during the entire period of the experiment.A Voucher species was deposited and stored at the Eastern Center for Ecosystems and Biodiversity (BIOECO, Spanish acronym*)*, Santiago de Cuba, with registration: BSC 21509.

Experimental design

A completely randomized experimental design including two experimental groups were used:

WMT (control): *S. lycopersicum L.* irrigated with water without treatment with MF

WTSMF: *S. lycopersicum L.* irrigated with non-uniform or heterogeneous SMF (20-200 mT)

A magnetic device composed of permanent magnets wereused to apply the magnetic treatment to the irrigation water during plant cultivation. This equipment was designed, constructed and characterized in CNEA [10]. Thetotal sample size was 50 plants, 25 from each experimental group, in a completely randomized experimental design with three replicates for each determination.

Localized irrigation was performed by aerial and drip microjet, depending on the plants' stage of cultivation. During the first and second cultivation phase, that includ the establishment, vegetative growth and flowering, the irrigation was performed once a day for 20 min. In the third cultivation phase, which incorporates fruit ripening, the irrigation was conducted twice a day during 20 min. The irrigation schedule was completed in 120 days.Treatments were carried out at the time of irrigation of the plants during the entire crop cycle until the development of the fruits.

Irrigation was carried out twice a day through an aerial microjet system for 30 min, which has ITUR pump accessories and a distributor system, controlled by valves that ensured that the irrigation was done in sections. The characteristics of the irrigation system were as follows:

Water speed: 1,4 – 1,6 ms^{-1}

Pump Flow: 2,54 – 2,91 $m^3 h^{-1}$

Crop natural conditions

The substrate characterization was performed, which was composed of a mixture of soil (brown without carbonate) and organic matter (3.5 %). The pH was 7.0. Pathogens, bacteria, and fungi were determined in collaboration with the Center for Industrial Biotechnology Studies (CEBI, *Spanish acronym*), Universidad de Oriente.

Collection and selection of fruits

In the two experimental groups the harvest of the ripe fruits was carried out in the fifth harvest according to established protocols [11]. After cultivation, 10 units of fruits were harvested at random from the two experimental groups. The fruits were dried in a stove (MWL-200, VEB, Germany) at 60 °C for 72 h. Then, they were pulverizedin an electric mill of blades (IKA M 20, China).Operations facilitated the extraction of the active principles and were carried out according to Branch Standard of the Ministry of Public Health (MINSAP, *Spanish acronym*) 310/91 [12].

Preparation of aqueous and ethanolic extracts

The dried fruits were weighed on a digital analytical balance (Sartorious® BS 124S, China) with an accuracy of 0,1 mg and the aqueous extracts were prepared at 50 mgL^{-1} by infusions with distilled water during 5 - 10 min. The extracts were cooled at room temperature and centrifuged (2 000 rpm) for 10 min in a centrifuge (Centribio, MLW T24 D, Germany). The supernatant was filtered through a Whatman No. 4 filter paper until the clarified aqueous extracts were obtained.

The ethanolic extracts were prepared from the pulverized dried fruits. They were obtained according to the technique described in advance [13]. Solutions were prepared at a concentration of 0.1 gmL^{-1} with 70 % ethanol, which were macerated in the dark for 7 days and with periodic stirring at room temperature (24 ± 2 °C). These mixtures were concentrated to total dryness in a rotary evaporator (Heidolph 4011, Canadá). With the resulting solid, infusions were prepared in distilled water until a concentration of 50 μgmL^{-1} was obtained. The extracts were centrifuged (2000 rpm) for 10 min. The supernatants were filtered with Whatman No. 4 filter paper until clarified.

Phytochemical determination

The extracts were subjected to a phytochemical screening to obtain a qualitative criterion of the chemical composition, according to the protocol of Miranda *et al.* [14].

This method allowed to detect groups of secondary metabolites present in the extracts by specific chemical tests using different reagents, where the appearance of a certain color or precipitate colored or not, is indicative of the presence of the compound. Table 1 shows the metabolites that were determined and the tests performed for their identification.

TABLE 1. METABOLITES AND TESTS EVALUATED IN THE PHYTOCHEMICAL SCREENING PERFORMED ON AQUEOUS AND ETHANOLIC EXTRACTS OF *S. lycopersicum L.*

Metabolites	**Tests**
Alkaloids	Mayer
Quinones	Legal
Saponins	Foam
Essential oils and fatty substances	Sudan III
Carotenoids	Carr-Price
Phenols and tannins	Ferric chloride
Carbohydrates	Molisch
Flavonoids	Concentrated sulfuric acid
Reducing sugars	Benedict

For the description of the trials, the crossing system used by García [15] was used to indicate the presence or absence of metabolites in the experimental groups.

Results and discussion

With the phytochemical screening, the qualitativedetermination of the chemical compounds of *Solanum lycopersicum L*. fruits was carried out and a high variability was found. Nine types of metabolites were identified in the aqueous and ethanolic extracts of the two experimental groups: alkaloids, quinones, saponins, essential oils and fatty substances, carotenoids, phenols, tannins, carbohydrates, flavonoids and reducing sugars (table 2).

TABLE 2: PHYTOCHEMICAL SCREENING TO AQUEOUS AND ETHANOLIC EXTRACTS OF *S. lycopersicum L.* FRUITS OBTAINED BY USING DIFFERENT TREATMENTS IN IRRIGATION WATER

		Experimental Groups			
Metabolites		**WMT**		**WSMT**	
		AE	**EE**	**AE**	**EE**
Alkaloids		+	+	+	+
Quinones		++	++	++	++
Saponins		++	+	++	+
Essential oils and fatty substances		-	-	-	-
Carotenoids		+	+	+	++
Phenols and tannins		+	+	++	+
Carbohydrates		+	+	++	+
Flavonoids		+	+	++	+
Reducing sugars		+	+	++	++

Legend: AE: Aqueous Extract, EE: Ethanolic Extract, WMT (control): *S. lycopersicum* L. irrigated with water without treatment with MF. WTSMF: *S. lycopersicum* L. irrigated with non-uniform or heterogeneous SMF (20-200 mT). Response ++ (very positive: very marked presence of the secondary metabolite), Response + (positive: presence of the secondary metabolite), Response - (negative: absence of secondary metabolite)

In the aqueous and ethanolic extracts of both experimental groups, was evidenced the presence of all identified metabolites. Carotenoids, phenols, tannins, carbohydrates, flavonoids and reducing sugars showed a very positive response in the WSMT group, in relation to the WMT group. In the aqueous extracts of both experimental groups were demonstrated, the higher presence of saponins, that could be because they present a polar zone in their structure that consequently showed greater water solubility (table 2).

The presence of alkaloids was a positive response in the aqueous extracts of the two experimental groups. It did not occur in the same manner in the ethanolic extracts, where a weak presence of this type of secondary metabolite was obtained.

Carotenoids were more pronounced in the ethanolic extract compared to the aqueous extract (table 2). The higher presence of the carotene in the ethanolic extract of the WSMT group could have its cause in the chemical nature of these compounds. They are apolar, therefore they dissolve better in ethanol, which is less polar than water. Ethanol has a lypophilic fragment that supports the solvation of organic substrates [16].

Phenolic compounds with a marked response in the aqueous extracts of the WSMT group were determined. It was evidenced that polyphenols are more soluble in aqueous solutions than in ethanolic [16].The polyphenol group includes flavonoids, which also had a marked presence, as did the carbohydrates and sugars in the group where extracts were obtained fromthe fruits of the WSMT group.

The absence of fats could be because *S. lycopersicum L.* is composed of 0,2 g per 100 g of fresh fruit approximately so it is not a considerable amount [17]. Saponins were not found either. They are glycosides, capable of producing foam, but they are not very common in fruits of this species.

In the four extracts analyzed, the aqueous extracts had a higher content of secondary metabolites in relation to the ethanolic extracts. These results are consistent with those obtained by others authors [18]. Phenols, tannins, carbohydrates and flavonoids showed a marked presence in the aqueous extract, with respect to the ethanolic extract in the experimental WSMT group. Therefore, the aqueous extract was richer in metabolites. Due to the polar nature of these compounds, using water as solvent allowed for better extractions.

Water has two hydrogen atoms that make its polarity stronger, however ethanol has two carbon atoms that do not transfer their electrons so easily. The hydrogen-oxygen bond, having greater electronegativity of oxygen. It generates a dipole moment, in the tetrahedral structure of water, which is maintained or increased. There is a lower polarity of the carbon-hydrogen bond of the carbon chain.

One of the reasons, for which a better extraction and concentration of metabolites was obtained in the aqueous extracts, could be that the applied SMF reduced the mobility of the solvent in the solution, to improve the reaction. According to other investigations, the MF reduces the volume of the solvent and increases the strength of the hydrogen bonds. By maximizing the electrostatic attraction and the interactions, caused by the changes in the radial distribution function of the solvents. The hydrogen bonding characteristics of the solvents investigated by molecular dynamics simulations were evidence of the hydrogen bond strength of oxygen with hydrogen. It is a more efficient intermolecular hydrogen bond compared to nitrogen and hydrogen [19]. In addition, it has been shown that aqueous extractions increase the content of phenolic compounds, because water can improve the turgor of the cells in the contact surface area between the plant and the solvent, with an increase in extraction yield [20].

The predominant presence of these metabolites in the aqueous extract of the WSMT group could be due to the influence of the static magnetic treatment in the irrigation water of the *Solanum lycopersicum L.* plants. This could contribute to favor the synthesis of these secondary metabolites, leading to its marked presence in the fruits of this experimental group.

The SMF was possibly able to alter the irrigation water and affect its physicochemical properties. Different researchers have verified that with the application of MF to water, changes occur, in addition toalterations in the hydration of the ions. When water circulates through a MF, in each charged particle a Lorentz force isexerted in the opposite direction. The fact that protons or ions hydrogen are transferred along the hydrogen ring junction chains to form a proton stream under the action of this MF force, is controversial [21].

It is suggested that this water is better assimilated or absorbed by cells, and can affect the different potentials that are generated in that process. Therefore, it was possible to favor the intensity of the flow of water and minerals to the interior of the plant, which occurred faster that it, did under normal conditions by activation of the osmosis mechanism. All of which probably favored the transport routes of nutrients and soluble substances in plants. As a result, the stimulating effect of magnetically treated water between 20 and 200 mT was evident against the active compounds.

Literature provides evidence for multiple studies that support the effects of irrigation water with SMFin different magnetic induction ranges. It was achieved an increase in the concentration of carbohydrates and phenols in leaves of *Solanum lycopersicum L*., *Triticum sp*. (wheat) and *Pisum sativum* (pea), using a magnetizer (Magnetron model UT1, from Magnetic Technologies LCC) [22]. An increase in the quercetin concentration, carotenoids and ascorbic acid, it has been obtained with the influence of SMF [23].

The phytochemical composition of the extracts of plants, as well as the methods for extraction, the duration and the polarity of the organic solvents, have influence for to determine the antioxidant potentials of the plant species [24]. The results of this research could to be related the increase of the antioxidant capacity in *Solanum* *lycopersicum L.* [25]. It could to contribute to raise awareness of one of the possible ways to improve the bioactive compounds and additionally, the nutritional and medicinal properties under the action of SMF between 20 and 200 mT. Therefore, this plant could be investigated for this purpose, since theoretical contributions to the knowledge were obtained.

Conclusions

The phytochemical screening showed that the aqueous extracts prepared from the fruits of S. lycopersicum L. irrigated with the water treated with SMF between 20 and 200 mT, revealed a marked presence of carotenoids, phenols, tannins, carbohydrates and flavonoids. The results provide scientific evidence about the positive incidence of the application of MF in the secondary metabolites of fruits. The presence of a variety of secondary metabolites in S. lycopersicum L. fruits contributes to the biological and pharmacological effects associated with the consumption of this plant.

Acknowledgements

Authors are grateful to the Center for Environmental Sciences of the Hasselt University in Belgium for the VLIR- IUC-UOs project and P1 of the Universidad de Oriente, and to the BIOECO researchers in Santiago de Cuba.

References

1. ZHANG, Y. J. *et al.* "Antioxidant Phytochemicals for the Prevention and Treatment of Chronic Diseases". *Molecules.* 2015, **20** (12), 21138 – 21156. ISSN 1420-3049.

2. RAY, S.; SAHA, R.; RAYCHAUDHURI, U.; CHAKRABORTY, R. "Different quality characteristics of tomato (SOLANUM LYCOPERSICUM) as a fortifying ingredient in food products: A review". *Technical Sciences/University of Warmia and Mazury in Olsztyn.* 2016, **19** (3), 199 – 213. ISSN 1505-4675.

3. CHEN, W. *et al.* "Lycopene attenuates Aβ1–42 secretion and its toxicity in human cell and *Caenorhabditis elegans* models of Alzheimer disease". *Neuroscience Letters.* 2015, **608**, 28 – 33. ISSN 0304-3940.

4. MARTÍ, R.; ROSELLÓ, S.; CEBOLLA, J. "Tomato as a Source of Carotenoids and Polyphenols Targeted to Cancer Prevention". *Cancers.* 2016, **8** (6), 58. ISSN 2072-6694.

5. DE VOS, Ric CH; HALL, R. D.; MOING, A. "Metabolomics of a model fruit: tomato. *Annual Plant Reviews Volume 43: Biology of Plant Metabolomics*. 2011, 109 – 155. ISSN 144433994X.

6. HIWASA TANASE, K. Fruit Ripening in Tomato and Its Modification by Molecular Breeding Techniques. En *Functional Genomics and Biotechnology in Solanaceae and Cucurbitaceae Crops*. Springer, Berlin, Heidelberg, 2016, 155 – 174. ISBN 978-3-662-48535-4.

7. JEDLIČKA, J.; PAULEN, O.; AILER, Š. "Research Of Effect Of Low Frequency Magnetic Field On Germination, Growth And Fruiting Of Field Tomatoes". *Acta Horticulturae et Regiotectuare.* 2015, **18** (1), 1 – 4. ISSN 1338-5259.

8. VALLVERDÚ, A. *et al.* "Effects of Pulsed Electric Fields on the Bioactive Compound Content and Antioxidant Capacity of Tomato Fruit". *J Agric Food Chem.* 2012, **60** (12), 3126 – 3134. ISSN 0021-8561.

9. CASANOVA, A. *et al. Manual para la producción protegida de hortalizas.* 2da edición. La Habana: Inst. Inv. Hortícolas "Liliana Dimitrova", CNSV, MINAGRI, 2007. ISBN 9597111373.

10. GILART, F. *et al.* "High flow capacity devices for anti-scale magnetic treatment of water". *Chemical Engineering and Processing: Process Intensification.* 2013, **70** (1), 211 – 216. ISSN 0255-2701.

11. FERREIRA, S.; FREITAS, R.; LAZZARI, E. "Padrão de identidade e qualidade do tomate (*Lycopersicon esculentum* Mill.) de mesa". *Ciência Rural.* 2004, **34** (1), 329 – 335. ISSN 0103-8478.

12. MINSAP *Medicamentos de origen vegetal: Droga cruda. Especificaciones generales*. La Habana, Cuba, 1991.

13. PEREIRA, R. *et al.* "Antioxidant efects of different extracts from *Melissa officinalis, Matricaria recutita* and *Cymbopogon citrates*". *Neurochem Res.* 2009, **34**, 973 – 983. ISSN 1573-6903.

14. MIRANDA, M. M.; CUÉLLAR, C. A. *Farmacognosia y Productos Naturales.* La Habana. Cuba: Ed. Varela Félix, 2001. ISBN 959-258-129-0.

15. GARCÍA, D.; OJEDA, F.; MONTEJO, I. "Evaluación de los principales factores que influyen en la composición fitoquímica de *Morus alba* (Linn.). I Análisis cualitativo

de metabolitos secundarios". *Pastos y Forrajes.* 2003, **26** (4), 335 – 346. ISSN 2078-8452.

16. SIDDHURAJU, P. "Antioxidant activity of polyphenolic compounds extracted from defatted raw and dry heated Tamarindus indica seed coat". *LWT - Food Science and Technology*. 2007, **40** (6), 982 – 990. ISSN 0023-6438.

17. PINELA, J.; BARROS, L.; CARVALHO, A.; FERREIRA, I. "Nutritional composition and antioxidant activity of four tomato (*Lycopersicon esculentum* L.) farmer' varieties in Northeastern Portugal homegardens". *Food Chem Toxic*. 2012, **50** (3–4), 829 – 834. ISSN 0278-6915.

18. ILAHY, R. *et al.* "Fractionate analysis of the phytochemical composition and antioxidant activities in advanced breeding lines of high-lycopene tomatoes". *Food & Function.* 2016, **7** (1), 574 – 583. ISSN 2042-6496.

19. MOOSAVI, F.; GHOLIZADEH, M. "Magnetic effects on the solvent properties investigated by molecular dynamics simulation". *Journal of Magnetism and Magnetic Materials.* 2014, **354**, 239 – 247. ISSN 0304-8853.

20. PINELA, J. *et al.* "Microwave-assisted extraction of phenolic acids and flavonoids and production of antioxidant ingredients from tomato: A nutraceutical-oriented optimization study". *Separat Purificat Techn.* 2016, **164** (1), 114 – 124. ISSN 1383-5866.

21. PANG, X. F.; DENG, B.; TANG, B. "Influences of magnetic field on macroscopic properties of water". *Modern Physics Letters B.* 2012, **26** (11), 1250069. ISSN 0217-9849.

22. SELIM, A.; ZAYED, M.; ZAYED, M. "Magnetic field treated water effects on germination, growth and physio-chemical aspects of some economic plants". *Acta Botanica Hungarica.* 2013, **55** (1-2), 99 – 116. ISSN 0236-6495

23. DUBOIS, A. F. *et al.* "Evaluación de la irrigación con el uso de campos magnéticos estáticos sobre la síntesis de metabolitos secundarios en *Solanum lycopersicum* L. (tomate)". *Investigación y Saberes.* 2012, **1** (1), 54 – 61. ISSN 1390-8073.

24. NETO, J. D. R. L. *et al.* "Phytochemical screening, total phenolic content and antioxidant activity of some plants from Brazilian flora". *Journal of Medicinal Plants Research.* 2016, **10** (27), 409 – 416. ISSN 1996-0875.

25. DUBOIS, A. E. F.; LEITE, G. O.; ROCHA, J. B. T. "Irrigation of *Solanum lycopersicum* L. with magnetically treated water increases antioxidant properties of its tomato fruits". *Electromagnetic Biology and Medicine.* 2013, **32** (3), 355 – 362. ISSN 1536-8378.