

Garlic used as a preservative increases the reduced form of ascorbate in pepper fruits

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SUMMARY

This work is a continuation of the one initiated by our group last season which dealt on the *Ripening of pepper fruits*. The main objective of this project is to design strategies which allow post-harvest and storage practices for pepper fruits conducting to extend their shelf life without lowering their nutritional value. In this work, an innovative procedure was set up with results implying improvement of quality of fruits. The potential repercussion of the proposed process is under a broader study for future transfer to the productive sector.

INTRODUCTION (AND OBJECTIVES)

Pepper is a plant which belongs to the family of the *Solanaceae*. Pepper varieties are classified as sweet and hot, based on the culinary purposes. The main pepper's producer countries are China, Mexico, Turkey, Indonesia, Spain and USA [1,2]. According to the shape, three main types of fruits are distinguished: California (square and short), Lamuyo (square and long), and Dulce italiano (long and narrow). From all of these types of peppers, the California one has been chosen for our experiments.

In this project, we have also used garlic. The properties attributed to garlic are almost innumerable: improves the body immune system, increases the body defences and the vitality, is an anticoagulant and improves blood circulation by dilating blood vessels; it is also a great antiseptic and expectorant, so it protects the respiratory system [3].

The ingestion of vitamin C helps to prevent illnesses. In fact, vitamin C deficiency in the human diet causes scurvy, but also cardiovascular diseases [4]. Pepper fruits have been proved to be a good source of vitamin C [5]. In a former work, the effect of incubation with garlic in the physiology of pepper fruits was studied [6]. In this work, the involvement of garlic in the post-harvest nutritional features of pepper fruits has been investigated.

MATERIALS AND METHODS

Plant materials and Experimental design

Pepper fruits (California type) used in this work were provided by *Syngenta Seeds, Ltd*, El Ejido, Almería, Spain, and garlic (purple type) was obtained from the farm *El Huerto del Tito*, Casas Nuevas, Granada, Spain.

The following experimental design was developed:

1. Fruits were weighed individually and washed with distilled water and dried.
2. Then, they were numbered (1-30) for future identification.
3. Six groups of fruits were set:
 - Group 0: fruits were chopped and frozen with liquid nitrogen until we used them for analyses.
 - Group A: incubated with a master preparation of garlic (MPG).
 - Group B: peppers without garlic incubation.
 - Group C: peppers at room temperature until the end of the experiment.
4. The groups A and B were subdivided into two subgroups each one:
 - Sub-group A1*: Peppers with MPG in a cold room (6°C; 14 hours of darkness).
 - Sub-group A2*: Peppers with MPG at room temperature (22°C/14°C, day/night; 14 hours of darkness).
 - Sub-group B1*: peppers without garlic in a cold room (6°C; 14 hours of darkness).
 - Sub-group B2*: peppers without garlic at room temperature (22°C/14°C day/night; 14 hours of darkness).
5. Fruits from sub-groups A1, A2, B1 and B2 were weighed and analyzed at 11 and 25 days.

Assays

In all pepper fruits the protein concentration [7] and vitamin C content by HPLC-MS were measured. The volatile compounds in garlic were identified and determined by gas chromatography linked to mass spectrometry (GC-MS).

RESULTS AND DISCUSSION

Some of the results obtained in this work cannot be given here since they are within a broader investigation focused in the future transfer to the productive sector. Therefore, they are subjected to confidentiality as part of a potential patent.

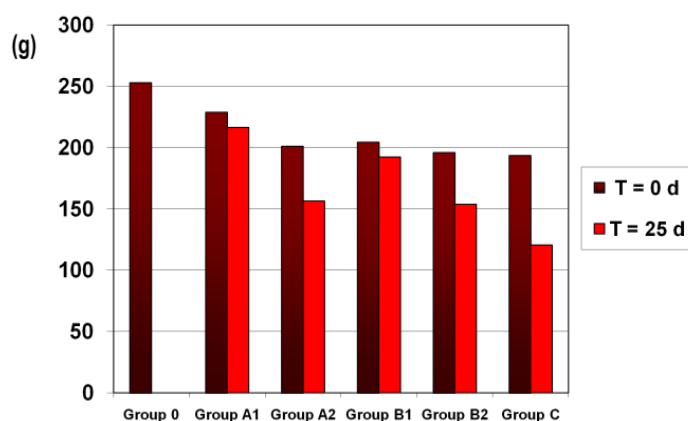
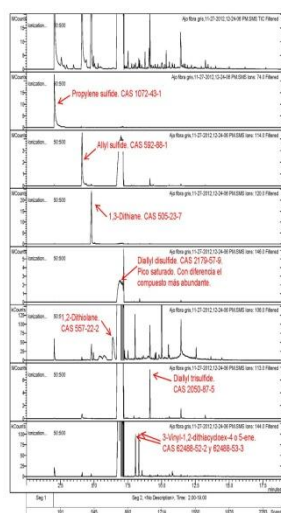


Figure 1. Effect of MPG and temperature on the fresh weight of pepper fruits.



Número	Compuestos identificados en orden de elución	Número CAS	Área (cuentas)	% referido al más abundante
1	Propylene sulfide	1072-43-1	8,154E+07	2,98
2	Allyl mercaptan	870-23-5	3,304E+06	0,12
3	Methyl allyl sulfide	10152-76-8	3,377E+06	0,12
4	Dimethyl disulfide	624-92-0	8,643E+06	0,32
5	Allyl sulfide	592-88-1	7,744E+07	2,83
6	2,4-Dimethylthiophene	638-00-6	2,095E+07	0,76
7	1,3-Dithiane	505-23-7	1,870E+08	6,82
8	2-Propenyl, methyl sulfide	2179-58-0	1,916E+06	0,07
9	1-Propenyl, methyl sulfide	5905-47-5	2,869E+07	1,05
10	Diallyl disulfide	2179-57-9	2,740E+09	100,00
11(*)	2-Mercapto-3,4-dimethyl-2,3-dihydrothiophene	NIST 322301	2,054E+07	0,75
12	3-Vinyl-1,2-dithiacyclohex-4-ene	62488-52-2	8,514E+07	3,11
13	3-Vinyl-1,2-dithiacyclohex-5-ene	62488-53-3	1,029E+08	3,76
14	Diallyl trisulfide	2050-87-5	4,499E+07	1,64

(*) No se posee el CAS, en su lugar se ofrece el NIST

Figure 2. Separation and identification of volatile compounds from garlic. The analysis was done by combination of gas chromatography to mass spectrometry (GC-MS) at the Instrument Technical Service of the EEZ.

CONCLUSIONS

1. Garlic increases the vitamin C content (mainly the reduced form of ascorbate) and this improves the conservation of peppers fruits.
2. This methodology can be also applied to other agricultural produce.
3. This project could lead to a patent.

ACKNOWLEDGEMENTS

This work was supported by the ERDF-cofinanced grant AGL2011-26044 from the Ministry of Science and Innovation, Spain. HPLC-GC and LC-MS analyses were performed at the Instrument Technical Service of the Estación Experimental del Zaidín, CSIC, Granada. The coordination of the IESs' teachers who participated in the PIISA program is also acknowledged.

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MY OWN IDEAS

Santiago Sánchez Requena

Empecé el proyecto PIIISA en 4º de la ESO, y nuestro grupo ha sido el único que ha continuado con el mismo proyecto durante 2 años dada su importancia, ya que podría desembocar en una patente.

El primer año nos dedicamos fundamentalmente al trabajo en el laboratorio. Me gustó bastante porque algunas veces parecía un científico, con la bata y los guantes, trabajando con bisturí y nitrógeno líquido. El trabajo de laboratorio es entretenido, e incluso divertido, es bastante metódico, en mi opinión; me sorprendió la excesiva limpieza para no contaminar las muestras. A lo largo de nuestra investigación hemos podido comprobar y realizar algunos de los pasos del método científico como: la elaboración de una pregunta, la creación de una hipótesis y la experimentación, siempre con la orientación de nuestro investigador José Manuel Palma.

Este último año nos hemos dedicado a la recopilación de los resultados para crear unas conclusiones que expondremos en la EEZ (Estación Experimental del Zaidín) y comunicaremos; Terminando así el ciclo del método científico. La divulgación la realizaremos mediante un Poster y la exposición, ambas en inglés. Esta parte ha sido más difícil y rigurosa, en ocasiones tediosa. Aunque creo que lo peor será cuando tenga que hablar en público y en inglés.

Esta experiencia me ha gustado bastante, ya que nos ofrece alternativas que hasta entonces desconocíamos y el hecho de que hayamos "colaborado" en la creación de una patente con solo 15 años me parece muy gratificante. Además me siento orgulloso de ser también los únicos que hemos continuado con el mismo proyecto 2 años seguidos. También ha servido para darme cuenta de lo difícil que es ser científico, ya que se necesitan muchos conocimientos, idiomas, curiosidad, dedicación, y una mezcla entre creatividad y razonamiento a la hora de la creación de las hipótesis. Quisiera darle las gracias a José Manuel Palma, Carmelo Ruiz, y otros muchos científicos por la paciencia que han tenido con nosotros.