WHOLE UTILISATION OF OLIVE OIL INDUSTRY BY-PRODUCTS

G. RODRÍGUEZ, J. FERNÁNDEZ BOLAÑOS, R. GUILLĚN, A. JIMĚNEZ, R. RODRÍGUEZ, M. LAMA, A. FUENTES

Food Biotechnology Departament, Instituto de la Grasa (CSIC) Avda. PadreGarcía Tejero 4, Apartado1078, 41012 Seville, Spain; Tel: 34-954691054; Fax 34-954691262; E-mail: guirogu@cica.es

SUMMARY: A process that allows an integral recovery of solid waste from two-phase olive oil or "alperujo" has been developed. This consists on a hydrothermal pretreatment where an autohydrolysis process occurs and the solid olive by-product was partially solubilised. Due this method makes easier the solid-liquid separation, the integral recovery includes both fractions. The solid fraction is considerable reduced after the treatment and several components such oil, cellulose and proteins are concentrated. These can be used in human (refined oil) and animal (cellulosic residue) food and, finally, as substrate for composting. From the water-soluble fraction besides recovering an important part of the antioxidant hydroxytyrosol (HT), imbues olive oil stability, as well as being beneficial to health, with biological and antimicrobial properties, it will allow the recovery of mannitol and non-digestible oligosaccharides that can be used as promoters of the growing of colon bifidobacterium. Also this liquid fraction is rich in sugars, including monosaccharides (glucose) that are fermentable to bioethanol. The system of purification of hydroxytyrosol, which is under patent, allows its production to industrial scale, with high degree of purity (over 99.6%) and low economic costs from every liquid source of HT. During the purification process, in addition to this antioxidant several other interesting compounds, have also been detected and their nutritional and pharmacological properties are being studied. All these investigations will make possible the integral recovery and the revalorization of this by-product.

1. INTRODUCTION

The manufacturing process of olive oil has undergone evolutionary changes. The traditional discontinuous pressing process was initially replaced by the continuous centrifugation, using a three-phase system and later on a two-phase system. The classic production of olive oil generates three phases and two wastes: olive oil, wastewater called alpechín and waste solid called orujo with a 50-60% of moisture.

The use of a modern two-phase processing technique in which no water is added, generates oil and a new by-product that is a combination of liquid and solid waste, called "alperujo, or two-phase olive mill waste". This by-product is higher content of water than orujo, around 70% of moisture.

The olive oil industries produce large volume of wastes both solid and liquid, which represent a disposal and potentially environmental pollution problem. Nevertheless they are also promising sources of compounds that can be recovered and used as valuable substances by developing of new processes. Particularly, the bioconversion of these wastes to useful products is receiving increased attention.

The olive fruit has many high added value compounds. It has a high nutritional interest. Most of the interesting compounds remain in the alperujo. By it the alperujo has to be considered an important source of compounds of high interest such as phenolic compounds, mainly Hydroxytyrosol (HT), triterpenes, pectins and oligosaccharides, mannitol, polymeric compounds and sugars. HT is one of major phenolic compounds present in olive fruit and it has been revealed to be the most interesting, because of its remarkable pharmacological and antioxidant activity (Fabiani *et al*., 2002; Visioli *et al*., 2004). Currently, many studies on bioavailability and metabolism in human are being conducted in order to establish its health-beneficial effects (Miró-Casas *et al.,* 2003; Visioli *et al*., 2005).

2. OBJETIVES

Total recovery of the waste of two-phase oil processing pretreated by thermal reactor has been proposed in the present study. The alternative use is represented by two possible applications:

1.- Recovery of natural constituents: the major advantage is the presence of several phenolic compounds, with bioactive properties.

2.- Bioconversion into a useful products: the problem is the presence of phenolic compounds, that can be toxic.

The solution, which allows the development of both applications, is to recover those phenolic compounds, removing them from alperujo. Thus it makes easy the follow approach of the rest of compounds and the utilization of the components as a fertilizer, animal feed or biofuel.

3. EXPERIMENTAL

Prior to the bioactive compounds isolated the waste is treated by steam explosion reactor in which high temperatures (160-240 ºC) and pressures (5-40 atm) are applied for shot periods of time (2-10 minutes). Basically the pretreatment consits of submitting the lignocellulosic material to satured steam to high temperatures and pressures. Depending on the conditions used, there is a depolymerization of polysaccharides (mainly of hemicelluloses) and a breaking of the lignin-carbohydrates bonds, resulting in the solubilization of lignin fragments of low molecular weight. The steam explosion reactor used in this wok is localed in the Food Biotechnology Department within Instituto de la Grasa (C.S.I.C.) Seville (Spain).

Samples of "Alperujo" used in this study were partially destoned, being the fragments of the olive stones separated in the extraction industries; and partially deoiled, being the residual olive oil obtained after a secondary centrifugation process. After steam explosion treatment the liquid and solid fraction is separated (Fernández-Bolaños et al.,2002, 2004):

1. The liquid fraction contains useful compounds that are characterized by HPLC and GC techniques. Also the antioxidant activity of HT was performed by means of antiradical capacity, ferric reducing power and inhibition of primary and secondary oxidation (Rodríguez et al., 2007a).
2. The solid is characterized and defated, using the fat as a source of bioethanol production, and the rest of the solid as a fertilizer. The defatted solid was hydrolyzed by enzymatic way in order to increase the glucose content in the solution, and also assays to determinate the solid digestibility was carried out (Rodríguez et al., 2007b).

4. RESULTS

When the alperujo is treated by steam explosion reaction an easy solid-liquid separation is produced. It allows the utilization of both fractions separately.

4.1 Liquid fraction

4.1.1 Hydroxytyrosol

The percentage of hydroxytyrosol referred to dry matter versus treatment conditions is showed in Figure 1. It represents the evolution of content of released HT for three samples of alperujo by steam treatment. The samples were taken at the beginning, at the halfway mark and at the end of the season. The quantity of released HT increases with the severity of the treatment in three samples tested up to a value of 1,7 per cent referred to dry initial matter.

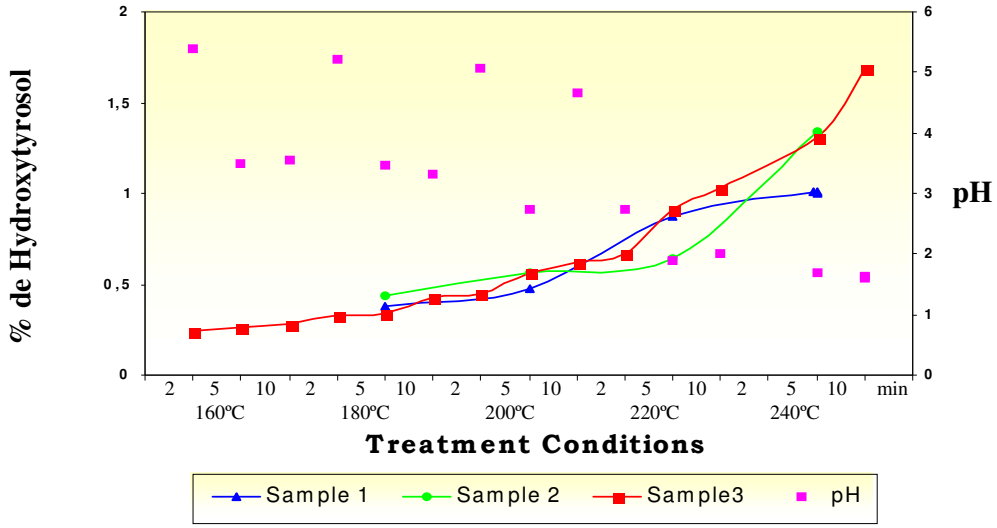

Figure 1. Evolution of the content of released hydroxytyrosol (g/100 g dry waste) at three different samples of two-phase olive waste treated by steam in function of the treatment parameter. Variations of the pH values.

The quantities of HT obtained by steam treatment without acid addition at 180 or 240 centigrade degree and with acid addition were compared against system currently used in the extraction and quantification of phenolic compounds in the olives (Figure 2). The best values were obtained by the steam treatment. There was great difference between steam treatment quantification and the rest of ordinary system. Therefore steam treatment is a good HT extraction tool and HT analytic tool from alperujo.

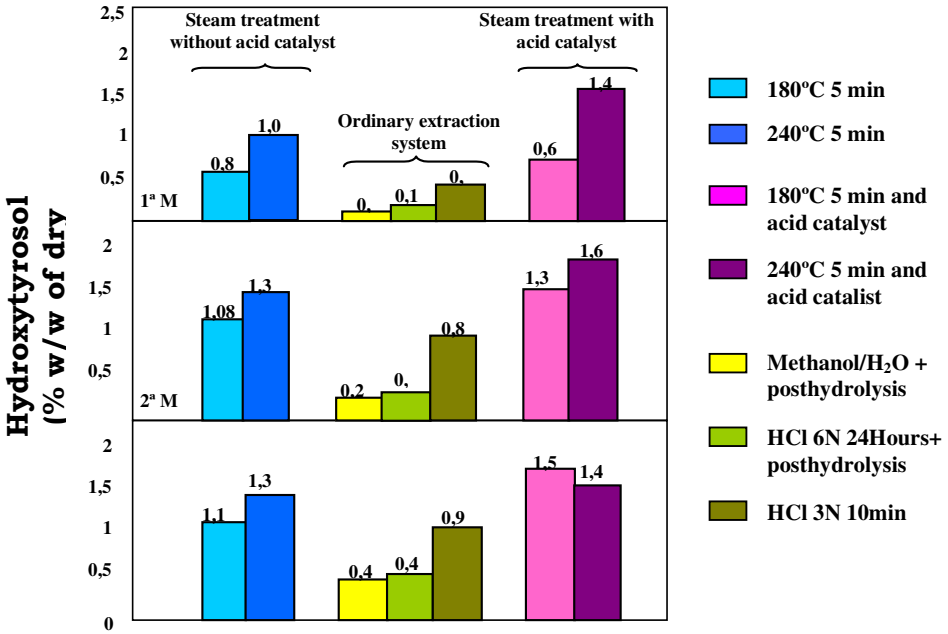

Figure 2. Comparison of HT released (free form) by different conditions of hydrothermal treatment at three different samples of two-phase olive waste, with or without acid catalyst against ordinary extraction system of phenolic compounds.

There is a new chromatographic system to purification HT under patent (Fernández-Bolaños et al., 2002a). It is a simple and inexpensive system. Nowadays it is the only system to producer natural HT (> 50% purity in the first step and >90% in the second) in large quantities within industrial sector. This system is under international patent. It could be also applied to many other sources that contain HT. The method of the invention implies the use of two stage chromatographic system. The presence and the purity of the HT purified by means of Nuclear Magnetic Resonance spectroscopy, was verified (Fernández-Bolaños et al., 2002b).

The study of the antioxidant activity of HT showed the HT has a high antiradical activity. HT has higher activity than vitamin C and similar or higher than vitamin E. In lipid assays the vitamin E presents a higher value than HT (Rodríguez et al., 2007a).

4.1.2 Soluble sugars

Sugars in the liquid fraction come from sugars that were initially soluble in alperujo or from the cell wall as cellulose and hemicellulose fundamentally. In both cases the sugars will be monosaccharide and oligosaccharide forms and the final quantity will be resultant of the balance between released-decomposition.

The steam treatment increases the quantity of total soluble sugars, some of these sugars are hydrolysed from hemicellulose, and the addition of acid increases the hydrolysed oligosaccharides. Therefore for the utilization it is necessary to take advantage of its main components such as:

- Glucose: most abundant sugar in monosaccharide form, useful to produce bioethanol.
- Oligosaccharides: mainly as a xylose. Nondigestible oligosaccharides are usually considered to enhance the growth of bifidobacteria and lactic acid bacteria in the human large intestine, with certain evidence of a preventive effect against colon cancer and other intestinal dysfunctions (Roberfroid and Slavin, 2000).
- Mannitol is used as an exicipient in pharmacy, and as anticaking and free-flow agent, lubricant, stabiliser and thickener, and low calorie sweetener in the food industry.

- Other compounds are currently being studied.

4.2 Solid fraction

This fraction is considerably reduced after the treatment and several compounds such as oil, cellulose, and proteins are concentrated.

Table1. Composition of solid fraction from alperujo before (initial) and after (final) thermal treatment, all values referred to initial dry matter.

		Sample 1 Steam treatment conditions 200°C / 5 min		Sample 2 Steam treatment conditions 200°C / 5 min		
		% (v/v) de H_2SO_4		% (v/v) de H_2SO_4		
		1	2,5	Without acid	1	2,5
% Cellulose	initial	7,47			4,95	
	final	6,58	8,50	7,97	5,55	6,83
% Lignin	initial	29,29			-	
	final	16,85	14,02	-	-	-
% Hemicellulose	initial	7,89			5,17	
	final	1,89	0,91	3,23	1,25	1,31
% Oil	initial	5,69			5,66	
	final	5,43	5,96	5,67	5,66	5,67
% Protein	initial	5,87			4,43	
	final	4,54	3,09	2,60	1,94	1,78

In Table 1 the composition of solid fraction before and after steam treatment is showed. Values are referred to the same base in order to be comparable directly. An important loss of weight happens, 66.9% and 65.2% to sample 1 and 2 respectively. Thus, some of the components are concentrated in the final solid, such as oil (16.3-18%), cellulose (19.6-25.7%) and protein (5.1-9.3%).

4.2.1 Oil

After treatment the fat remains in the solid in high concentration. It does not alter with steam treatment. The utilization of solid fraction is based in the utilization of oil after its extraction as refined oil in human feed, or as a biofuel.

4.2.2 Cellulose

The defatted rich cellulose solid can be used to produce glucose that can be fermented to obtain bioethanol, feed animal use or fertilizer use. The results showed that the steam explosion improve the cellulose accessibility in the "in vitro" digestibility and to enzymatic hydrolysis (Rodríguez et al., 2007). To obtain a utilization of the solid fraction the follow scheme can be used.

The final solid has a 20% of oil. The first step is the extraction with solvent and utilization of its oil Defatted solid has a 25% of cellulose. Different uses have been studied for the utilization of the solid defatted:

- Sacharification to obtain a fermentable glucose that can produce ethanol. Steam treatment improves the glucose solubilisation from cellulose.

- Feed Animal is other possible use of this rich cellulose solid. The studies of digestibility show that steam treatment improves the digestibility of cellulose.
- And the uses like fertilizer: the studies of detoxification show that steam treatment decreases the alperujo toxicity.

5. CONCLUSIONS

The proposed total utilization of alperujo is based in thermal pretreatment to facilitate the solid and liquid separation and the use of these phases. The aim of this work is the recovery the antioxidants and other interesting compounds, and the use of sugars, oil and rich cellulose solid.

Nowadays a new reactor has been designed in order to simplify the pretreatment, lower temperatures and pressures. It is being optimized to semi industrial level; the preliminary results show that it can be a simple and cheap tool easily applicable in the industry.

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