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# Concentration of natural pigments and other bioactive components in pulp oils from de-stoned olives

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#### Abstract

De-stoned olives (Gentile di Chieti, Caroleo and Coratina varieties) were processed in comparison with non-stoned olives (traditional extraction). Since the de-stoned oily pastes are not easy to process, a depolymerising pectocellulolytic enzyme preparation (in combination or not with draining micronised food talc) was added to them. These processing aids significantly improved the lower oil yields given by the new extraction system. Destoning increased the hourly plant processing potential of approximately 20%. In addition, it allowed to obtain separated by-products (better recyclable in chemical or feedstuff industries) and to produce highly nutraceutical oils, characterised by higher contents of hydrophilic biophenols, tocopherols and volatiles. On the contrary, the de-stoned oils had lower concentrations of pigments (both carotenoids and chlorophylls). They were chiefly valuable for the marked and harmonic aroma, the green fruitiness notes and the potentially higher preservability (shelf-life) and resistance to autoxidation. This could lead to economically prefer the new extraction system to the classical industrial processing cycles.

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*Industrial relevance:* Because of high bioactivity level of the de-stoned oil and of its high market price, the new olive oil extraction system, consisting in processing de-stoned olives, is already being used by some European industrial oil mills even though the oil yields are lower with respect to the traditional systems. It is expected that through incorporation into the de-stoned oily paste of olive processing aids, such as the officially recognised micronised food talc and/ or the exogenous pectocellulolytic enzymes (whose official recognition could be near), a regular industrial application of the innovative extraction cycle will be made.

#### 1. Introduction

Extraction technology is a major factor affecting the compositional quality and the bioactivity level of virgin olive oil (VOO). The endocarp (stone) of the olive fruit is known to be an extremely hard wood kind, so that its violent crushing during olive processing causes a great consumption of electrical energy and a mechanical and thermal stress of olive paste. This favours the chemical and enzymatic degradation of triglycerides (Ranalli, Gomes, Delcuratolo, Contento, & Lucera, 2003) and of some valuable unsaponifiable oil components, particularly biophenols.

According to some authors (Lavelli & Bondesan, 2005), another problem in olive oil extraction would be the high level of endogenous oxidoreductase enzymes, in particular peroxidase, occurring in the kernel present inside the woody stone, which could enhance the risk of oil oxidation degradations. Such a problem, however, was not pointed out by other authors (Patumi, Terenziani, Ridolfi, & Fontanazza, 2003).

Concerning the inferior oil contained in the kernel, it arises no problems since it represents a minimal proportion of the total fruit oil and moreover, according to some authors, it would not or minimally be extracted under the operating pressures or

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centrifugal forces currently used in olive mills (Ranalli, Malfatti, Lucera, Contento, & Sotiriou, 2005). Also, some authors report that the olive cultivars have a non-negligible proportion of empty stones (not containing the kernel), which for some of them touches values as high as 50–60% (Ranalli et al., 2003).

In the past, de-stoned olive paste also caused stoppages during experimental processing tests, but at present this problem appears to be overcome because of advances in the extracting plant engineering. A major technical advantage of destoning in olive processing could be the improved rheological characteristics of olive paste as a result of its increased moisture percentage that results in reduced apparent viscosity of the oil phase. Also, the formation of solid colloids should decrease as the separation of the liquid phases, by means of the hydrostatic pressure, is lower (Amirante, Clodoveo, Dugo, Leone, & Tamborrino, 2006).

Thus, it has been suggested to remove, by means of destoner machines, the stone from olive fruit and to softly crush and process the sole fruit pulp. This, in addition to increasing both quality and nutraceutical level of the produced oils, should also lead to increase of approximately 20% (a value corresponding to the stone proportion in the olive fruit) the amounts of olives processed per hour (Del Caro, Vacca, Poiana, Fenu, & Piga, 2006).

However, the stone fragments dispersed into the oily paste resulting from olive crushing are an important draining system, without which downflow and separation of the liquid phases (oil and water) from the solid phase during extraction are quite difficult. This is a major elaiotechnic problem that could at least in part be solved using suitable draining adjuvants (such as micronised food talc) during olive processing. Such angular and sharp stone fragments also contribute to breaking the residual uncrushed olive pulp cells during malaxing (a technological step following crushing). The lack of these actions in de-stoned olive pastes could partly be solved by treating them with enzyme processing aids (Ranalli et al., 2003; Ranalli, Lucera, Contento, Simone, & Del Re, 2004) even though these have not yet been officially recognised by EU (Reg. EC 1513, 2001).

Destoner, unfortunately, breaks only a part of the vegetal cells making up the parenchymatic olive fruit tissue and it cannot substitute the crusher machine, so it is advisable to couple to it a finisher, that is, a hammer or a disk crusher type exerting a softly grinding action. However, also in this instance, such an innovative procedure of olive paste preparation does not lead to a satisfactory grinding of olive pulp, since there are none of the above effects of the stone fragments (Patumi et al., 2003).

In spite of the above drawbacks, which results in lower oil yields, owing also to the fact that destoning does not thoroughly remove the pulp from the stone, the innovative extraction procedure is currently considered by olive oil producers, manufacturers and researchers for possible industrial applications. In fact it might lead to produce more valuable and functional extravirgin olive oil and to a higher final income. Even Columella and other ancient writers supported this theory (Lavelli & Bondesan, 2005). There are at present some olive growers who already produce de-stoned virgin olive oil (DVOO), which are usually marketed in small beautiful bottles (100 to 500 ml) at very high prices. This could be the determinant factor leading to

decide for regular industrial employment of the new olive processing cycle (Luaces, Pérez, & Sanz, 2004).

In addition, it should also be considered the increased industrial exploitability and the increased market value of the by-products arising from the new extraction technology. In particular, the oil recovered from the fruit kernel, owing to its richness in oleuropein, squalene and nüzenide could profitably be employed in cosmetic and pharmaceutical industries. Yet, the de-stoned olive residue (husk), containing no or minimal amounts of minute stone fragments, could be used as a good ingredient of complex animal feeds or of some human foods, as well as for recovery of the contained residual oil (up to 40%, dry matter base) (Luaces et al., 2004).

Due to these positive aspects, the new olive processing technology is currently being studied in-depth in research centres, which even special projects are implementing. Unfortunately, no univocal results have been obtained so far in these scientific investigations. Moreover, some analytical parameters have not or poorly been studied in the new products (Lavelli & Bondesan, 2005). There are even some authors who obtained de-stoned oils showing no substantial compositional differences with respect to non-stoned oils (Patumi et al., 2003).

The aim of this work was thus to point out the actual analytical composition (and the nutraceutical level) of DVOO, a natural product that could potentially have a role in increasing the competitivity of the olive oil sector (Luaces et al., 2004). Special attention we paid to the highly bioactive pigments, which have not or little been investigated so far in DVOO.

#### 2. Material and methods

### 2.1. Chemicals

Most of the solvents, reagents, and equipments used for the analytical characterisation of the oil samples have been given in earlier works (Ranalli et al., 2003; Ranalli et al., 2004, 2005). Chemicals were mostly of chromatographic grade and were commercially available from Carlo Erba (Milan, Italy), Fluka (Buchs, Switzerland) and Sigma-Aldrich Chemical (St. Louis, MO, USA).

## 2.2. Characteristics of the processing aids used

The depolymerising enzyme complex tested, *Clarex 8XL* (Gist-Brocades, Seclin City, France), is essentially made up of pectolytic, cellulolytic and hemicellulolytic enzyme species. It has a different relative enzyme composition with respect to other similar ones but its activity is likewise not less than 2000 units/ml. One unit of activity is defined as the amount of enzyme complex liberating 1  $\mu$ mol of reducing sugars per minute from pectins. It degrades the vegetable colloids (pectins, hemicelluloses, proteins, etc.) emulsifying the minute oil droplets. Also, the oil–water emulsions are removed since it also contains an endopolygalacturonase enzyme species. In addition, the rheological features of the olive paste are improved. Such an enzyme preparation is water-soluble and thoroughly comes out in the liquid effluent (waste water) during the final step (oily must centrifugation) of the

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