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Title: Combined industrial olive oil extraction plant using ultrasounds, microwave, and heat exchange: Impact on olive oil quality and yield

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Abstract: In this study, an industrial combined plant (ICP) constructed from a low-frequency ultrasound device, microwave apparatus and heat exchanger is employed to investigate the real possibility of introducing these innovative technologies to the olive oil extraction process and evaluating their influence on olive oil quality and yield. The novelty of this study lies in the simultaneous use of these three technologies to condition the olive paste in a real olive oil extraction plant. Different olive paste treatments were compared in order to define the effects on the olive oil quality and yield. The use of a spiral heat exchanger in addition to the malaxer reduced the malaxation time to 20 min, and with the microwave apparatus it was possible to obtain an entirely continuous process, without interruptions, from the milling phase to the solid-liquid separation phase. The internal spiral aids in moving the paste from the input to output section, resulting in limited operating pressure. Using the ICP device led to an average increase in extractability ranging from 2.30 to 3.85% with respect to the control thesis, for the Arbosana and Arbequina varieties, respectively, but this difference was not statistically significant. Regarding the virgin olive oil (VOO) quality, the use of the ICP did not affect the marketable parameters and total phenol content, while in terms of the process efficiency, the ICP obtained a higher value than the conventional process and improved the extraction yield.

Highlights

1. A spiral coil heat exchanger was developed to condition the olive paste
2. An industrial combined plant (ICP) was developed to condition the olive paste
3. Using combined plant an increase in extractability was found
4. Superior results in phenolic and volatile compounds were found using ICP

1 **Combined industrial olive oil extraction plant using ultrasounds, microwave,**
2 **and heat exchange: Impact on olive oil quality and yield**

3

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14

15 **Abstract**

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17 device, microwave apparatus and heat exchanger is employed to investigate the real possibility of
18 introducing these innovative technologies to the olive oil extraction process and evaluating their
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29 respect to the control thesis, for the Arbosana and Arbequina varieties, respectively, but this
30 difference was not statistically significant.

31 Regarding the virgin olive oil (VOO) quality, the use of the ICP did not affect the marketable
32 parameters and total phenol content, while in terms of the process efficiency, the ICP obtained a
33 higher value than the conventional process and improved the extraction yield.

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39 **1. Introduction**

40 Ultrasound, microwave and heat exchangers are used for food processing in a variety of industrial
41 and everyday applications. Increasing the thermal performance of virgin olive oil (VOO) plants,
42 saving on process time and improving the olive oil efficiency and quality have become research
43 goals in recent years. For this reason, researchers and private companies have conducted numerous
44 studies to increase olive oil plant performance (Jmenéz & Beltran, 2007; Juliano et al., 2013;
45 Almeida et al., 2016; Bejaoui et al., 2015; Bejaoui et al., 2016b; Bejaoui et al., 2017; Chemat et al.,
46 2017; Iqdiam et al., 2017; Leone et al., 2016; Juliano, 2017; Leone et al., 2017; Leone et al., 2018).

47 The application of ultrasound treatment in olive oil extraction plants has been studied for the past 10
48 years, particularly in terms of aiding malaxation operations. Within this time, the application of
49 low-frequency ultrasound (that is, 20 to 80 kHz) in olive oil extraction plants in order to condition
50 the olive paste has been also investigated, using mainly laboratory-scale equipment and research
51 conducted in full-scale plants (Jiménez et al., 2007, Almeida et al., 2016). The impact on the olive
52 oil quality and yield has been the main subject of investigation (Bejaoui et al., 2015; Bejaoui et al.,
53 2016a; Bejaoui et al., 2017). The latest studies have been highlighted the positive impact of high-
54 power ultrasounds on the oil extraction yield and extractability. These parameters resulted in higher
55 than conventional malaxation (Bejaoui et al., 2016b) and did not affect the fatty acid and phenolic
56 composition; however, the green sensorial attribute was increased significantly (Bejaoui et al.,
57 2018).

58 In recent years, the impact of high-frequency ultrasound standing waves (megasonics) (that is, 300
59 to 400 or 800 kHz), applied before and/or after the malaxation process or by combining low- and
60 high-frequency ultrasound, on olive oil extractability has also been studied (Leone et al., 2017;
61 Juliano et al., 2017). These studies demonstrate the enhancement of olive oil separation, thereby
62 increasing the extraction yield. Several of these researches have also highlighted the instantaneous
63 and homogeneous heating of olive paste, under continuous conditions, compare to traditional
64 malaxation. Process time saving has been demonstrated by means of applying an industrial

65 microwave prototype to olive oil extraction equipment, with significant potential to become a real
66 alternative technique to continuous conditioning of the olive paste (Leone et al., 2015; Leone et al.,
67 2017), also in combination with megasonics (Leone et al., 2018). Tamborrino et al. (2014)
68 highlighted the higher concentration of volatile compounds, without compromising the high olive
69 oil quality, when using a continuous microwave-assisted system for paste malaxation.

70 Regarding the application of heat exchange to the olive oil extraction process, the literature has
71 reported one of the first researches conducted by Amirante in 2006, in which a positive influence on
72 improving the malaxation efficiency was determined by introducing a heat exchanger between the
73 crusher and malaxer. In recent years, the introduction of tubular heat exchangers has been studied
74 experimentally during the olive oil extraction process, in order to evaluate its influence on the
75 process rapidity of as well as the yield and olive oil quality (Esposito et al., 2013; Leone et al., 2015;
76 Veneziani et al., 2015). In fact, the use of a heat exchanger plays an important role in transferring
77 heat between the fluids circulating inside the pipes, thereby reducing the process time (Veneziani et
78 al., 2015). The same study demonstrated a positive influence on the olive oil phenolic
79 concentrations and volatile compounds. Thereafter, the introduction of a tubular heat exchanger to
80 the mechanical olive oil extraction process was studied, evaluating the effect of olive paste cooling
81 treatment on the yield and olive oil quality. The results demonstrated an insignificant difference
82 regarding the yield and olive oil quality parameters, but a significant improvement in the phenolic
83 compounds was observed (Veneziani et al., 2017).

84 In this research, a combination of innovative technologies including ultrasounds, microwaves and a
85 horizontal spring heat exchanger was studied. The novelty of this study lies in the use of these three
86 technologies assembled in a pilot plant. The industrial combined plant (ICP) used was a full-scale
87 size, and was designed and assembled in order to gain increased knowledge concerning the use of
88 innovative means of conditioning olive oil paste. The ICP was capable of operating by using all
89 three technologies implemented simultaneously or individually. The study constituted the first

90 experience of an ICP pilot plant using three implemented technologies proven in an operational
91 environment, and evaluated the influence on olive oil quality and yield.

92

93 **2. Materials and methods**

94 *2.1. Industrial olive oil extraction plant equipped with ICP*

95 The experimental tests were performed in an industrial olive oil extraction plant located in Foggia
96 (Italy). The mill included a leaf remover, washing machine (Special Automatic, Alfa Laval
97 Corporate AB, Lund, Sweden), single-grid hammer crusher (75 hp Hammer Crusher, Alfa Laval
98 Corporate AB, Lund, Sweden), six malaxer machines sealed on the top, each with a capacity of 700
99 L, three-phase solid/liquid horizontal centrifugal decanter (NX X32, Alfa Laval Corporate AB,
100 Lund, Sweden), and a liquid/liquid vertical plate centrifuge (UVPX 507, Alfa Laval Corporate AB,
101 Lund, Sweden).

102 The processing plant included the ICP for olive paste conditioning.

103

104 *2.1.1. ICP system layout*

105 The ICP was equipped with the following units: (i) continuous microwave machine (reverberant
106 tunnel, generator head, power supply, water-cooled magnetron head and polypropylene pipe); (ii)
107 ultrasonic pilot device (generator, transducer and vessel); and (iii) spiral-coil heat exchanger (with
108 modular tubular units including helicoids). These three technologies were controlled by a
109 programmable logic controller (PLC), capable of commanding simultaneous use or any possible
110 combination among the components.

111 In the following, the most relevant components of this plant are described in detail, with focus on
112 the design specifications, constructive materials, suppliers and operational regimes.

113 The spiral-coil heat exchanger (SCHE) consisted of six units, each operating on tube-in tube, as
114 illustrated in Figure 1. The olive paste flowing through a straight tube was moved by a cavity pump
115 placed in the output section of the grid hammer mill, while the hot fluid service flowed in counter-

116 currently outside the tube in the heating jacket. Each module length was 2 m and heat exchange area
117 of 0.56 m², and these were connected by flanges welded at their extremities and bolted. Inside the
118 straight tube of each module, a spiral coil was connected to an electric motor equipped with a
119 mechanical speed reducer. The SCHE was assembled by EMITECH s.r.l. (Corato, Italy).

120 The high-power ultrasound pilot device (UPD) was capable of producing electromagnetic waves at
121 20 kHz. It was equipped with an adjustable power supply, allowing the power output to range from
122 50 to 3000 W. The pilot equipment consisted of a stainless steel cylindrical tank, which was
123 connected to the heat exchange and MW pilot device to ensure process continuity. An ultrasound
124 transducer was placed inside the steel cylindrical tank.

125 The continuous microwave (CMW) system (Emitech s.r.l., Molfetta, BA, Italy), was constituted by
126 a reverberant chamber constructed from AISI 304 stainless steel, equipped with a TM060 generator
127 head (Alter s.r.l., Reggio Emilia, Italy) and coupled to a YJ1600C magnetron (Alter s.r.l., Reggio
128 Emilia, Italy). The generator head was connected to a SM1180T power supply (Alter s.r.l., Reggio
129 Emilia, Italy). This system had a maximum power of 6.0 kW at 2.45 GHz. The magnetron was
130 water-cooled. Inside the reverberant chamber there was a polypropylene tube with a diameter of
131 0.0654 m and length of 2 m.

132 A PLC included in the main electrical panel allowed for controlling the electrical power provided
133 by the magnetrons and, consequently, the output temperature.

134 2.2 *Experimental process description*

135 The ICP was tested by selecting five different operational modes to investigate the optimal
136 combinations for improving the oil extractability (*E*) and olive oil quality.

137 An experimental design was developed, using five replicates per treatment and the same olive
138 batch.

139 Tests were carried out using olives of the Arbosana (*Olea europaea* L.) cultivar, with a maturity
140 index of 2.6, and Arbequina (*Olea europaea* L.) cultivar, with a maturity index of 2.7. The fruit

141 ripeness was determined according to the method proposed by the International Olive Council
142 (IOOC, 2001).

143 All tests were carried out at a mass flow rate of $1300 \pm 10 \text{ kg h}^{-1}$, with 8.6% water added to the
144 decanter.

145 The ICP operational modes were as follows:

146 *Operation mode 1 (control test): malaxer machine (MM)*

147 The ICP was bypassed during the process.

148 The olive paste that was just crushed was malaxed using a traditional MM at $25 \pm 1 \text{ }^\circ\text{C}$ for 40 min
149 and after being transferred to the decanter.

150 *Operation mode 2: SCHE*

151 The ICP was operated by the SCHE device only, and the MM was bypassed.

152 The olive paste obtained was pumped by the main pump throughout the SCHE. The olive paste
153 temperature was $25 \pm 1 \text{ }^\circ\text{C}$ following SCHE processing, and the operating time was 3.61 min.

154 *Operation mode 3: SCHE and MM (SCHE-MM).*

155 The ICP was operated by the SCHE only, followed by malaxation treatment.

156 The olive paste that was just crushed was pumped throughout the SCHE. The olive paste output
157 temperature was $23 \pm 1 \text{ }^\circ\text{C}$ and the operating time was 3.61 min. Subsequently, the olive paste was
158 transferred to the MM and processed for 20 min at $25 \pm 1 \text{ }^\circ\text{C}$.

159 *Operation mode 4: SCHE and CMW machine (SCHE-CMW).*

160 The ICP was operated by the SCHE, followed by the CMW machine, and the malaxer was
161 bypassed.

162 The olive paste that was just crushed was pumped throughout the SCHE, reaching a temperature of
163 $21 \pm 1 \text{ }^\circ\text{C}$, and then throughout the microwave device, reaching a temperature of $25 \pm 1 \text{ }^\circ\text{C}$.

164 The operating time was 3.61 min for the heat exchanger processing and 0.6 min for the microwave
165 device processing.

166 *Operation mode 5: SCHE, UPD and CMW machine (SCHE-UPD-CMW).*

167 The ICP was operated by all three devices and the MM was bypassed.
168 The olive paste that was just crushed was pumped throughout the SCHE, reaching 21 ± 1 °C, and
169 subsequently throughout the UPD followed by the CMW until reaching 25 ± 1 °C.
170 The operating time was 3.61 min for the heat exchange processing, 1.3 min for UPD processing and
171 0.6 min for the microwave device processing.
172 The specific energy transferred was 15.60 kJ kg^{-1} when the CMW machine was used and 8.31 kJ
173 kg^{-1} when the UPD was used.

174

175 2.3. *Sampling*

176 The following samples were collected for each run:

- 177 • N. 1 sample of olives to determine oil and water content;
- 178 • N. 1 sample of pomace from the decanter. The sample was composed of a small fraction of
179 pomace sampled from the decanter at regular time intervals of 1 min;
- 180 • N. 1 sample of wastewater from the decanter. The sample was composed of a small fraction
181 of wastewater sampled from the decanter at regular time intervals of 1 min;
- 182 • N. 1 sample of olive oil. At the end of each run the oil was stored in a steel tank and one
183 sample was taken.

184 All of the samples were stored at 4 °C until analysis.

185

186 2.4. *Extractability (E)*

187 The extractability (*E*) was calculated using the following equation:

$$188 \quad E = \frac{W_{oil}}{W_{total\ oil}} \times 100, \quad (1)$$

189 where W_{oil} is the extracted oil mass (kg) and $W_{total\ oil}$ is the oil mass of the olives (kg).

190

191 2.5. *Oil and water content in olives, pomace and wastewater*

192 The total oil content was determined for a sample that was previously dehydrated until reaching a
193 constant weight, and the moisture removed from the sample was recorded. The oil in the dried
194 sample was extracted by hexane in an automatic extractor (Randall 148, Velp Scientifica, Milan,
195 Italy), following the analytical technique described by Cherubini et al. (2009). The results were
196 expressed as the percentage of oil in wet and dry matter.

197

198 2.6. *VOO analysis*

199 The VOO quality was assessed considering only the oils obtained from the Arbosana cultivar. The
200 free fatty acids, peroxide number and spectrophotometric absorptions were determined and sensory
201 analyses were carried out according to official methods (Regulation ECC 2568/91). Phenolic
202 compounds were extracted by means of a methanol-water mixture (70:30, v/v) and the methanolic
203 extracts were used to determine the total phenolic content (TPC) of the oil, as previously reported
204 by (Caponio et al., 2018; Squeo et al., 2016). For the HPLC analysis, the extraction procedure was
205 the same as that of TPC, with slight modifications. In particular, 5 g of oil and 2 mL of MeOH-H₂O
206 mixture were used, and 250 µL standard solution of gallic acid (100 ppm in MeOH-H₂O) was added
207 for quantification. All of the reagents used were HPLC grade. The chromatographic system and
208 conditions are reported in (Tamborrino et al., 2017). The identification of single phenolics was
209 carried out by comparing the retention times with those of the reference standards, or literature data
210 where no standards were available. The tocopherols were analysed as previously reported in
211 (Tamborrino et al., 2017), using the same chromatographic system. The carotenoids were
212 spectrophotometrically determined by weighting approximately 0.25 g of a sample in a 10-mL
213 volumetric flask and bringing it to volume with hexane. The absorbance was read at 449 nm against
214 a blank of pure hexane. Quantification was performed by means of an external calibration curve of
215 β-carotene (R^2 equal to 0.9987). The chlorophylls were determined in accordance with the IUPAC
216 method (1995). All spectrophotometric analyses were performed on a Cary UV 60
217 spectrophotometer (Agilent Technologies, Inc., Santa Clara, CA, USA).

218 2.7. *Statistical analysis*

219 Each industrial treatment was performed seven times, and all laboratory analyses were performed in
220 triplicate. All experimental data were analysed using the ANOVA test and Tuckey's multiple range
221 test ($\alpha = 0.05$), using the MATLAB[®] statistics toolbox (The Mathworks Inc., Natick, MA, USA).
222 The data on the VOO quality were analysed by means of Minitab 17 software (Minitab Inc., State
223 College, PA, USA). One-way ANOVA was applied, followed by Fisher's LSD post-hoc test for
224 multiple comparisons, at a significance level of 5%. Principal component analysis (PCA) was
225 carried out on the quality data using the same software.

226

227 **3. Results and discussion**

228 *3.1 Effects of ICP system on quantitative parameters*

229 Tables 1 and 2 display the results of the dry matter in the wastewater, oil content in the pomace (dry
230 matter), oil extractability (*E*) and operating pressure, respectively, for the Arbosana and Arbequina
231 cultivars.

232 For both cultivars, the dry matter content in the wastewater was statistically higher when only
233 SCHE was used for the olive paste conditioning. This means that the sedimentation of solids in the
234 decanter took place with lower effectiveness than in other conditions, owing to the poor
235 conditioning of pastes when only the SCHE was used. This aspect did not affect the oil lost in the
236 wastewater. The value was less than 1% in all experimental tests (data not shown).

237 The water content in the pomace did not exhibit statistically significant differences among the tests
238 for both varieties.

239 When only the SCHE was used, the oil content in the pomace was 24.38% (d.m.) and 21.82%
240 (d.m.) for the Arbosana and Arbequina cultivars, respectively. This value is statistically higher than
241 those obtained by other theses, which exhibited no significant differences.

242 Regarding the extractability, for both cultivars there were no significant differences between the
243 conditions, except when only the SCHE was used, where the extractability value was statistically

244 lower than that for the other four test conditions, with no significant differences between them.
245 Based on the data analysis of the extractability and oil content in the pomace, it is possible to assert
246 that the use of only the SCHE is not sufficient to provide high efficiency in olive paste
247 conditioning.

248 The use of the SCHE in addition to the malaxer reduced the conditioning time, as also reported in
249 previous papers such as those of Esposto et al. (2013) and Leone et al. (2016).

250 It should be emphasised that, by using the spiral heat exchanger, it was possible to work with
251 pressures under 3.4 bar, which is significantly lower than the value of approximately 8 bar generally
252 used in similar heat exchangers without the internal coil.

253 By comparing the MM, SCHE, SCHE-MM and SCHE-CMW, it is possible to analyse the
254 individual effects of the microwave system on the plant performance. As reported in Table 1 and 2,
255 the use of the CMW machine in addition to the SCHE significantly increased the extractability and
256 significantly decreased the oil content in the pomace. Moreover, the SCHE-MW condition exhibited
257 no significant differences in extractability and oil content in the pomace compared to the control
258 (MM) and SCHE-MM treatment. However, it was possible to achieve a significant reduction in the
259 conditioning time, obtaining an entirely continuous process, without interruptions, from the milling
260 phase to the solid-liquid separation phase. This confirmed previous conclusions drawn by Leone et
261 al. (2014) and Tamborrino et al. (2014). The positive effect of CMW on E was demonstrated in this
262 study, with a 4 °C thermal increase and a specific energy transfer of approximately 15.60 kJ kg⁻¹.

263 The impact of ultrasound on the plant quantitative performance can be assessed by comparing
264 SCHE-UPD-CMW and SCHE-CMW. The use of the UPD (8.31 kJ kg⁻¹ at 20 kHz) resulted in an
265 increase of 2.01 and 2.80% for the Arbosana and Arbequina cultivars, respectively.

266 Comparing the use of the ICP system when operated with all three active devices, with respect to
267 the control (MM), it can easily be observed that the increase in E was equal to 2.30 and 3.85% for
268 the Arbosana and Arbequina cultivars, respectively. Although the extraction results are not
269 statistically significant, it appears that the increasing E is inversely proportional to the data for the

270 control thesis extraction. This means that a greater amount of oil contained in the pomace in the
271 control test resulted in a greater effect of the combination of the three technologies on the decanter
272 efficiency.

273 This result is in accordance with Bejaoui et al. (2016b), Iqdiam et al. (2017) and Jiménez and
274 Beltran (2007).

275

276 *3.2 Effect of ICP system on VOO quality*

277 Considering the basic analytical parameters, all of the VOOs obtained from the experimental trials
278 were below the maximum limits set for the extra virgin category, and no significant differences
279 were highlighted owing to the paste conditioning process (data not shown). The fruity note median
280 was higher than 0 in all cases, and no sensory defects were reported.

281 Table 3 displays the total phenolic, tocopherol and pigment contents of the samples, along with the
282 statistical analysis results. The amount of phenolic compound in the Arbosana oils ranged from
283 approximately 130 to 200 mg kg⁻¹ oil, similar to the values previously reported for the same
284 cultivar, which is recognised as belonging to the class of low-phenolic olive cultivars (Scarafia,
285 2013). Certain significant differences were highlighted as a result of the technological treatments.
286 The superior performances in terms of the absolute phenolic amount were attributed to the
287 combined SCHE-UPD-CMW treatment, which was not statistically different from the malaxation
288 trial (MM). In the other cases, the oils were progressively poorer in phenolics, with the SCHE trial
289 being most inferior. When the heat exchanger was coupled with malaxation or microwaves, the
290 results were intermediate to the two conditions described above and not significantly different. In
291 2006, the first paper reporting on the use of a heat exchanger during the olive extraction process, by
292 Amirante et al., stated that the oil phenolic content was positively influenced by the heat exchanger.
293 However, in that case it was only used as a rapid tool for paste conditioning, and the malaxation
294 conditions was similar to that of other tests (30 min at 27°C). Subsequently, Leone et al. (2016)
295 reported that a heat exchanger allowed for a reduced malaxation time from 40 to 10 min, without

296 affecting the phenolic content, using olives of Peranzana cultivar. These results are not confirmed
297 by this study, in which by using olives of Arbosana and Arbequina cultivars the phenolic content
298 was lower even when coupling the heat exchanger with the traditional malaxation step for 20 min.
299 In contrast, Esposito et al. (2015) found that similar behaviour was exhibited by a phenolic reduction
300 when a heat exchanger was used. Similarly, studies on the application of microwaves have
301 demonstrated that this treatment alone could not extract a comparable amount of hydrophilic
302 antioxidant to that of the common process, even if the effect of such technology is clear and
303 correlable with the reduction of the olive paste's conditioning time (Leone et al., 2018; Tamborrino
304 et al., 2014). To the best of the authors' knowledge, no previous studies have taken into account the
305 impact of new conditioning technology on lipophilic antioxidants and pigments. Considering such
306 compounds, once again the SCHE-MM and SCHE-CMW trials provided very similar results, with
307 the sole significant exception being the carotenoids content, which was higher when the heat
308 exchanger was coupled with microwaves. Among the other trials, significant differences were only
309 exhibited in the contents of the β - γ -tocopherols and chlorophylls, with MM increasing the content of
310 the former and decreasing that of the latter. Overall, this is noteworthy as the different conditioning
311 treatments did not negatively affect the lipophilic antioxidant contents compared to those observed
312 for the hydrophilic ones. However, traditional malaxation was proven to be the superior treatment in
313 terms of tocopherol extraction.

314 Table 4 reports the oil HPLC phenolic profile of the identified compounds. Overall, the data were in
315 agreement with the TPC results and, considering the total amount, the same pattern could be
316 concluded. Indeed, the highest values were determined in the SCHE-UPD-CMW and MM oils, with
317 the lowest amount again detected in the SCHE oils. Considering the phenolic classes, the most
318 abundant were the secoiridoids, arising from oleuropein and ligstroside (Bendini et al., 2007), while
319 remarkable amounts of flavones and lignans were also detected. Tyrosol was the only phenyl ethyl
320 alcohol found in the samples, with the highest significant value in the SCHE-MM trial. The SCHE-
321 UPD-CMW and MM trials exhibited very similar phenolic profiles, with few significant

322 differences, owing to the tyrosol and dialdehydic forms of secoiridoid contents. In particular, the
323 SCHE-UPD-CMW oils were poorer in tyrosol but significantly richer in 3,4-DHPEA-EDA, 3,4-
324 DHPEA-EA-dialdehyde form and *p*-HPEA-EDA. The compound *p*-HPEA-EDA, also known as
325 oleocanthal, was indicated as the main factor responsible for the pungent notes of the oil (Bendini et
326 al., 2007). The effect of the heat exchanger differed in the coupled treatment function. Indeed, when
327 associated with microwaves (SCHE-CMW), higher amounts of secoiridoid derivatives and lignans
328 were found (3,4-DHPEA-EDA, 3,4-DHPEA-EA-dialdehyde form, *p*-HPEA-EDA, pinoresinol and
329 3,4-DHPEA-EA), while, on the contrary, the association with traditional malaxation (SCHE-MM)
330 provided significantly higher amounts of flavones. The treatment using only the heat exchanger
331 exhibited the worst results, and almost all of the phenolic compounds were significant lower than
332 those of the other trials. Considering the traditional extraction process, three main factors affected
333 the olive paste preparation following crushing: malaxation time, temperature and air contact. In this
334 study, the processing temperatures in the different trials were almost the same and the observed
335 difference could not be a result of this parameter. The same statement could be made regarding the
336 air with which the olive paste was in contact, which was not modified. The only processing factor
337 that was significant different among the treatments was the paste conditioning time. It is well
338 known that, during malaxation, several enzymatic reactions take place, and wall-degrading enzymes
339 (pectinases, cellulases and hemicellulases) along with endogenous glycosidases (β -glucosidase) and
340 oxidoreductases (polyphenol oxidase and peroxidase) can affect the release of phenolic compounds
341 as well as their profiles and contents (Cirilli et al., 2017; García-Rodríguez et al., 2011; Montedoro
342 et al., 2002). In light of these considerations, as suggested by Esposto et al. (2015), the observed
343 differences could be owing to the different times available for the enzymes to work. A shorter time
344 resulted in less enzyme action and thus phenolic reduction. In fact, in the SCHE-MM, SCHE-CMW
345 and SCHE trials, the paste conditioning was faster and the phenolic compounds were lower than in
346 the common malaxation (MM). However, these considerations are not fully supported by the
347 experimental results, in which the most superior were determined in the SCHE-UPD-CMW trial,

348 where the paste conditioning time was also very short and lasted for approximately 6 min. In this
349 case, the additional effect of the ultrasound treatment appeared to overcome the issue owing to the
350 short paste conditioning time. Indeed, acoustic cavitation owing to ultrasound may disrupt
351 biological cell walls, facilitating the release of minor compounds (Chemat et al., 2017). In
352 summary, from a quality point of view, the heat exchanger and microwave have been proven as
353 useful technologies for rapid olive paste conditioning, but they need to be associated with a
354 common malaxation step or ultrasound treatment, which can respectively exert the mechanical
355 effect required for enhancing phenolic release.

356 Moreover, paste malaxation was found to be the principal step in which volatile compounds were
357 formed, through the activity of other fundamental enzymes, those of the so-called lipoxygenase
358 (LOX) pathway. It is possible that modifications in the paste conditioning duration will alter the
359 activity extent of such enzymes and, consequently, change the oil volatile profile. Further studies
360 are necessary to investigate this aspect.

361 Figure 2 illustrates the score plot (A) and loading plot (B) of the PCA performed on the oil quality
362 data. Overall, the first two principal components accounted for approximately 70% of the total
363 variance. The treatments were effectively separated within the space of the principal components,
364 with the only exception being the MM and SCHE-UPD-CMW trials, which were overlapped. This
365 is in accordance with previous discussions, from which it emerged that the oils from these trials
366 exhibited very similar characteristics. The PCA clearly depicted that when only the heat exchanger
367 was used (SCHE), the oils were poorer in both lipophilic and hydrophilic antioxidants. The SCHE-
368 CMW and SCHE-MM trials lay in different areas on PC2, mainly owing to the differences in the
369 carotenoids and the 3,4-DHPEA-EDA content.

370

371 **4. Conclusions**

372 The experimentation highlighted the importance of using the spiral heat exchanger, which, in
373 addition to the malaxer, reduces the conditioning time by half. Furthermore, the internal spiral aids

374 in moving the paste from the input to output section, resulting in limited operating pressures.
375 Therefore, the heat exchanger is a useful device for rapid temperature adjustment of the olive paste
376 following crushing, thereby improving the malaxation effect, making its implementation within the
377 oil extraction process necessary.

378 The research emphasises that, by using a microwave apparatus in addition to a heat exchanger, it is
379 possible to obtain a continuous conditioning process of the olive paste with only several minutes of
380 treatment. Finally, although the use of a low-frequency ultrasonic apparatus for conditioning the
381 olive paste led to an average extractability increase of 2.30% and 3.85% with respect to the control
382 thesis for the Arbosana and Arbequina varieties, respectively, this difference was not statistically
383 significant. Although the use of ultrasound did not result in significant extractability increases, in
384 light of the average differences obtained, it is probable that this difference could be significant when
385 increasing the number of comparative tests.

386 Regarding the oil quality, the use of alternative conditioning technologies, individually or in
387 combination, may save the lipophilic antioxidant furniture, while resulting in a hydrophilic
388 antioxidant reduction. The ultrasound cavitation effect is capable of overcoming this drawback.

389

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393

394

395 **References**

396 Almeida, B., Valli, E., Bendini, A., Gallina Toschi, T., 2016. Semi-industrial ultrasound-assisted
397 virgin olive oil extraction: impact on quality. *Eur. J. Lipid Sci. Technol.* 119
398 <http://dx.doi.org/10.1002/ejlt.201600230>.

399

400 Amirante, P., Clodoveo, M. L., Dugo, G., Leone, A., & Tamborrino, A. (2006). Advance
401 technology in virgin olive oil production from traditional and de-stoned pastes: Influence of the
402 introduction of a heat exchanger on oil quality. *Food chemistry*, 98, 797-805.

403

404 Bejaoui, M.A., Beltran, G., Sanchez-Ortiz, A., Sanchez, S. Jimenez, A., 2015. Effect of continuous
405 high power ultrasound before malaxation to virgin olive oil quality criteria and yield. A laboratory
406 scale approach. *Eur. J. Lipid Sci. Technol.* 10.1002/ejlt.201500020.

407

408 Bejaoui, M.A., Beltran, G., Sanchez-Ortiz, A., Sanchez, S. Jimenez, A., 2016a. Continuous high
409 power ultrasound treatment before malaxation, a laboratory scale approach: effect on virgin olive
410 oil quality criteria and yield. *Eur. J. Lipid Sci. Technol.*, 118, pp. 332-336.

411

412 Bejaoui, M.A., Beltran G., Aguilera M.P., Jimenez, A. (2016b). Continuous conditioning of olive
413 paste by high power ultrasounds: Response surface methodology to predict temperature and its
414 effect on oil yield and virgin olive oil characteristics *LWT - Food Science and Technology* 69
415 (2016) 175-184.

416

417 Bejaoui, M.A., Sanchez-Ortiz, A., Aguilera M.P., Ruiz-Moreno, M.J., Sánchez, S., Jiménez, A.,
418 Beltrán, G. (2017). High power ultrasound frequency for olive paste conditioning: Effect on the
419 virgin olive oil bioactive compounds and sensorial characteristics. *Innovative Food Science &*
420 *Emerging Technologies*, Volume 47, Pages 136-145.

421

422 Bendini, A., Cerretani, L., Carrasco-Pancorbo, A., Gómez-Caravaca, A. M., Segura-Carretero, A.,
423 Fernández-Gutiérrez, A., & Lercker, G. (2007). Phenolic molecules in virgin olive oils: a survey of
424 their sensory properties, health effects, antioxidant activity and analytical methods. An overview of
425 the last decade. *Molecules*, 12, 1679-1719.

426

427 Beycan Ibrahimoglu, M. Zeki Yilmazoglu, Disposal of olive mill wastewater with DC arc plasma
428 method, *Journal of Environmental Management*, Volume 217, 2018, Pages 727-734,

429

430 Caponio, F., Squeo, G., Curci, M., Silletti, R., Paradiso, V.M., Summo, C., Crecchio, C., &
431 Pasqualone, A. (2018). Calcium carbonate effect on alkyl esters and enzymatic activities during
432 olive processing. *Italian Journal of Food Science*, 30, 381-392.

433

434 Chemat, F., Rombaut, N., Sicaire, A-G., Meullemiestre, A., Fabiano-Tixier, A-S., Abert-Vian, M.
435 2017. Ultrasound assisted extraction of food and natural products. Mechanisms, techniques,
436 combinations, protocols and applications. A review. *Ultrasonics Sonochemistry* 34, 540-560

437

438 Cirilli, M., Caruso, G., Gennai, C., Urbani, S., Frioni, E., Ruzzi, M., Servili, M., Gucci, R., Poerio,
439 E., & Muleo, R.M. (2017). The role of polyphenoloxidase, peroxidase, and β -glucosidase in
440 phenolics accumulation in *Olea europaea* L. fruits under different water regimes. *Frontiers in plant*
441 *science*, 8, 717. doi: 10.3389/fpls.2017.00717

442

443 Esposto, S., Veneziani, G., Selvaggini, R., Urbani, S., et al., Flash thermal conditioning of olive
444 pastes during the olive oil mechanical extraction process: impact on the structural modifications of
445 pastes and oil quality. *J. Agric. Food Chem.* 2013, 61, 4953–4960.

446

447 Iqdiam, B. M., Mostafa, H., Goodrich-schneider, R., Baker, G. L., Welt, B., Marshall, M. R., &
448 Marshall, M. R. (2017). High Power Ultrasound : Impact on Olive Paste Temperature , Malaxation
449 Time , Extraction Efficiency , and Characteristics of Extra Virgin Olive Oil. *Food and Bioprocess*
450 *Technology*, 11(3), 634-644.

451

452 García-Rodríguez, R., Romero-Segura, C., Sanz, C., Sánchez-Ortiz, A., & Pérez, A.G. (2011). Role
453 of polyphenol oxidase and peroxidase in shaping the phenolic profile of virgin olive oil. *Food*
454 *Research International*, 44, 629-635.

455

456 IUPAC (1995). Determination of chlorophyll pigments in crude vegetable oils. *Pure and Applied*
457 *Chemistry*, 67, 1781–1787.

458

459 Jiménez, A., G. Beltran, M. U. (2007). High-power ultrasound in olive paste pretreatment. Effect on
460 process yield and virgin olive oil characteristics. *Ultrasonics Sonochemistry*, 14(6), 725– 731.
461 doi:10.1016/j.ultsonch.2006.12.006

462

463 Juliano, P., Swiergon, P., Lee, K.H., Gee, P.T., Clarke, P.T., Augustin, M.A. 2013. Effects of pilot
464 plant-scale ultrasound on palm oil separation and oil quality, *J. Am. Oil Chem. Soc.* 90, 1253–1260.

465

466 Juliano, P.*, Augustin, M.A., Mawson, R., Xu, X-Q, Knoerzer, K. 2017a. Advances in high
467 frequency ultrasound separation of particulates from biomass. *Ultrasonics sonochemistry*. 577-590.

468

469 Juliano, P., Balczyk, F., Swiergon, P., Supriyatna, M. I. M., Guillaume, C., Ravetti, L.,
470 Canamasas, P., Cravotto, G., Xu, X. 2017b. Extraction of olive oil assisted by high-frequency
471 ultrasound standing waves. *Ultrasonics Sonochemistry*, 38, 104-114.
472 doi:10.1016/j.ultsonch.2017.02.038

473

474 Leone, A., Tamborrino, A., Romaniello, R., Zagaria, R., Sabella, E., Specification and
475 implementation of a continuous microwave-assisted system for paste malaxation in an olive oil
476 extraction plant. *Biosyst. Eng.* 2014, 125, 24–35.

477

478 Leone, A., Esposito, S., Tamborrino, A., Romaniello, R., Taticchi, A., Urbani, S., & Servili, M.
479 (2016). Using a tubular heat exchanger to improve the conditioning process of the olive paste:
480 Evaluation of yield and olive oil quality. *European Journal of Lipid Science and Technology*,
481 118(2), 308–317. <http://dx.doi.org/10.1002/ejlt.201400616>.

482

483 Leone, A., Romaniello, R., Tamborrino, A., Xu, X., & Juliano, P. (2017). Microwave and
484 megasonics combined technology for a continuous olive oil process with enhanced
485 extractability. *Innovative Food Science and Emerging Technologies*, 42, 56-63.
486 doi:10.1016/j.ifset.2017.06.001

487

488 Leone, A., Romaniello, R., Tamborrino, A., Urbani, S., Servili, M., Amarillo, M., Juliano, P.
489 (2018). Application of microwaves and megasound to olive paste in an industrial olive oil
490 extraction plant: Impact on virgin olive oil quality and composition. *European Journal of Lipid
491 Science and Technology*, 120(1) doi:10.1002/ejlt.201700261

492

493 Leong, T. Knoerzer, K. Trujillo, F.J. Johansson, L. Manasseh, R. Barbosa-Canovas, G.V. and
494 Juliano, P. 2015. Megasonic separation of food droplets and particles: design considerations, *Food
495 Engineering Reviews*, 7, 298-320.

496

497 Montedoro, G., Baldioli, M., Selvaggini, R., Begliomini, A.L., Taticchi, A. & Servili, M. (2002).
498 Relationships between phenolic composition of olive fruit and olive oil: the importance of the
499 endogenous enzymes. *Acta Horticulturae*, 586, 551-556.
500

501 Official Journal of the European Communities. 1991. European Community Regulation No.
502 2568/1991, N. L. 248 of September 5th.
503

504 Romaniello R., Leone A., Tamborrino A. Specification of a new de-stoner machine: evaluation of
505 machining effects on olive paste's rheology and olive oil yield and quality. *J Sci Food Agric* (2017).
506

507 Scarafia, L. (2013). Olive cultivars and oil polyphenols. Published on West Coast Olive Guide.
508 Summer 2013. © 2013 Agbiolab, Inc, 530 924-4789, www.agbiolab.com.
509

510 Squeo, G., Silletti, R., Summo, C., Paradiso, V.M., Pasqualone, A., & Caponio, F. (2016). Influence
511 of calcium carbonate on extraction yield and quality of extra virgin oil from olive (*Olea europaea*
512 L. cv. Coratina). *Food Chemistry*, 209, 65-71.
513

514 Tamborrino, A., Romaniello, R., Zagaria, R., & Leone, A. (2014). Microwave-assisted treatment for
515 continuous olive paste conditioning: Impact on olive oil quality and yield. *Biosystems Engineering*,
516 127, 92–102. <http://dx.doi.org/10.1016/j>.
517

518 Tamborrino, A., Squeo, G., Simone, F., Paradiso, V.M., Summo, C., Bianchi, B., Leone, A., &
519 Caponio, F. (2017). Industrial trials on coadjuvants in olive oil extraction process: effect on

520 rheological properties, energy consumption, oil yield and olive oil characteristics. *Journal of Food*
521 *Engineering*, 205, 34-46.

522

523 Trujillo, F.J., Juliano, P. Barbosa-Cánovas, G. and Knoerzer, K. Separation of suspensions and
524 emulsions via ultrasonic standing waves. A review, *Ultrasonics Sonochemistry*, 21 (2014) 2151-
525 2164.

526

Figure 1
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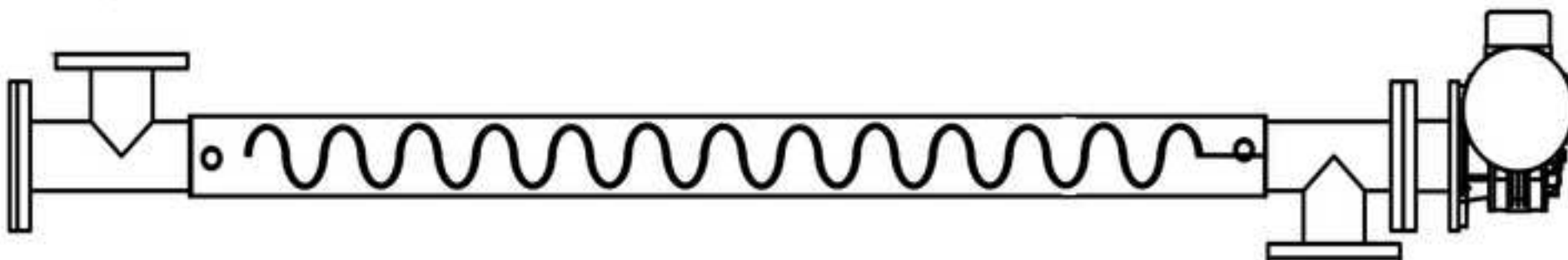


Figure 2a
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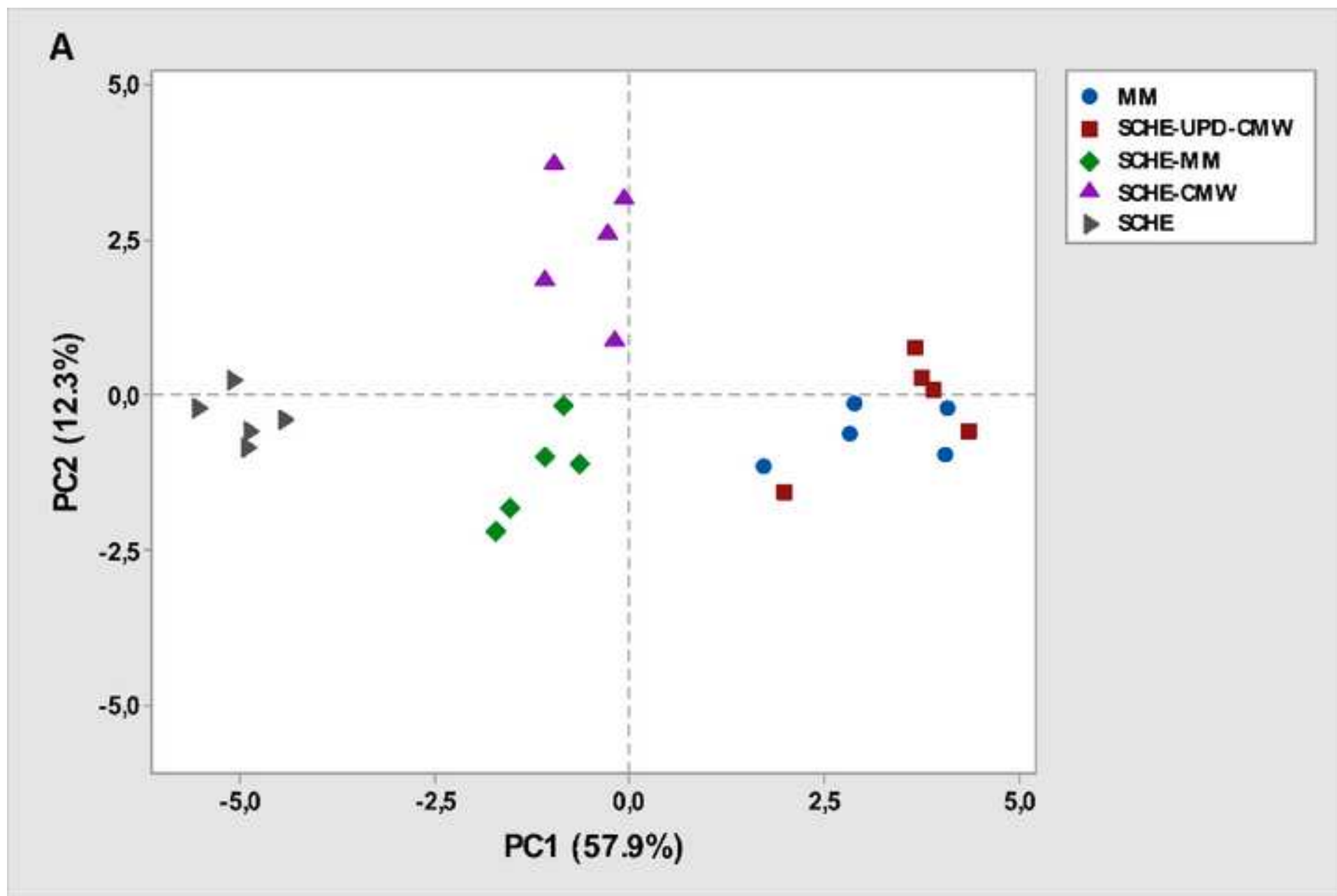


Figure captions.

Fig. 1 – Module of spiral-coil heat exchanger (SCHE).

Fig. 2 – Score plot (A) and loading plot (B) of the Principal Component Analysis performed on the oil's quality data.

Table 1. Quantitative results for the processing of the Arbosana olives

	(MM)	(SCHE)	(SCHE - MM)	(SCHE-CMW)	(SCHE-UPD-CMW)
Dry matter in the waste water (%)	6.88 ± 0.17 b	17.32 ± 0.18 a	6.96 ± 0.14 b	6.82 ± 0.13 b	6.72 ± 0.19 b
Water content in the pomace (%)	55.30 ± 1.19 a	55.27 ± 0.94 a	55.66 ± 0.73 a	55.55 ± 0.65 a	55.27 ± 0.91 a
Oil content in the pomace (% d.m.)	13.54 ± 0.83 ab	24.38 ± 1.71 a	13.46 ± 1.12 ab	13.56 ± 1.72 ab	11.91 ± 1.23 b
E (%)	83.21 ± 1.13 ab	71.42 ± 1.69 b	83.20 ± 1.58 ab	83.50 ± 1.74 ab	85.51 ± 1.24 a
*Operating pressure (bar)	1.8	3.2	3.2	3.2	3.7

Data represent mean value ± standard deviation. Different letters in rows denote statistical significant differences ($p < 0.05$).

*The pressure value was measured after the cavity pump placed after the hammer crusher

Table 2. Quantitative results for the processing of the Arbequina olives

	(MM)	(SCHE)	(SCHE - MM)	(SCHE-CMW)	(SCHE-UPD-CMW)
Dry matter in the waste water (%)	7.24 ± 0.24 b	16.92 ± 0.20 a	6.74 ± 0.22 b	6.60 ± 0.07 b	6.72 ± 0.05 b
Water content in the pomace (%)	56.38 ± 0.71 a	55.26 ± 0.47 a	55.88 ± 0.76 a	55.26 ± 0.66 a	55.45 ± 0.68 a
Oil content in the pomace (% d.m.)	12.92 ± 1.44 ab	21.82 ± 1.73 a	13.28 ± 1.14 ab	12.78 ± 1.26 ab	10.67 ± 0.96 b
E (%)	80.00 ± 2.14 ab	67.56 ± 3.23 b	80.09 ± 1.40 ab	81.05 ± 1.31 ab	83.85 ± 1.12 a
*Operating pressure (bar)	1.8	3.2	3.2	3.2	3.7

Data represent mean value ± standard deviation. Different letters in rows denote statistical significant differences ($p < 0.05$).

*The pressure value was measured after the cavity pump placed after the hammer crusher.

Table 3. HPLC phenolic profile of Arbosana oil samples and results of the one-way ANOVA followed by Fisher's LSD test for multiple comparisons. Data are expressed as mean ± standard deviation (mg kg^{-1} ; $n=5$).

Identified compounds	SCHE-UPD-CMW	SCHE	MM	SCHE-MM	SCHE-CMW
Tyrosol	0.583±0.140 c	0.076±0.083 e	0.845±0.194 b	1.025±0.154 a	0.376±0.024 d
Vanillic acid	0.175±0.021 a	0.048±0.046 c	0.209±0.031 a	0.120±0.023 b	0.120±0.052 b
Syringic acid	0.150±0.014 ab	0.091±0.012 c	0.168±0.017 a	0.092±0.024 c	0.141±0.006 b
<i>p</i> -cumaric acid	0.138±0.023 ab	0.000±0.000 d	0.144±0.022 a	0.092±0.020 c	0.118±0.015 b
<i>t</i> -ferulic acid	0.151±0.049 a	0.029±0.040 b	0.099±0.092 a	0.162±0.030 a	0.010±0.022 b
3,4-DHPEA-EDA	0.283±0.024 b	0.205±0.022 c	0.219±0.015 c	0.265±0.012 b	0.340±0.017 a
3,4-DHPEA-EA-dialdehyde form	0.243±0.020 a	0.119±0.012 d	0.218±0.011 b	0.160±0.008 c	0.205±0.008 b
<i>p</i> -HPEA-EDA	3.054±0.198 a	1.800±0.046 d	2.822±0.283 b	1.928±0.037 d	2.337±0.014 c
Pinoresinol	2.101±0.122 a	1.376±0.066 c	2.056±0.148 a	1.437±0.035 c	1.660±0.087 b
3,4-DHPEA-EA	7.535±0.562 a	5.857±0.165 c	7.369±0.388 a	6.641±0.338 b	7.309±0.254 a
Luteolin	2.752±0.338 a	0.511±0.209 d	2.471±0.340 a	2.128±0.134 b	1.033±0.218 c
<i>p</i> -HPEA-EA	1.821±0.087 a	1.388±0.051 d	1.786±0.076 ab	1.578±0.055 c	1.707±0.022 b
Apigenin	3.367±0.135 a	0.818±0.355 c	3.217±0.085 a	3.173±0.181 a	1.553±0.277 b
Total	22.354±0.842 a	12.318±0.582 d	21.623±0.724 a	18.801±0.481 b	16.909±0.495 c

3,4-DHPEA-EDA, dialdehydic form of elenolic acid linked to hydroxytyrosol; *p*-HPEA-EDA, dialdehydic form of elenolic acid linked to tyrosol; 3,4-DHPEA-EA, oleuropein aglycon; *p*-HPEA-EA, ligstroside aglycon.

Different letters on the same row indicate significant differences ($p \leq 0.05$).

Table 4. Total phenolic, tocopherols and pigments content of Arbosana oil samples and results of the one-way ANOVA followed by Fisher's LSD test for multiple comparisons. Data are expressed as mean \pm standard deviation (mg kg^{-1} ; $n=5$).

Compounds	SCHE-UPD-CMW	SCHE	MM	SCHE-MM	SCHE-CMW
TPC	195 \pm 15 a	129 \pm 15 c	194 \pm 16 a	169 \pm 13 b	158 \pm 14 b
α -tocopherols	4.73 \pm 0.14 b	4.35 \pm 0.09 b	5.83 \pm 0.13 a	4.33 \pm 0.08 b	4.56 \pm 0.68 b
γ -tocopherol	260.73 \pm 1.36 ab	262.46 \pm 3.15 ab	273.64 \pm 3.64 a	255.23 \pm 2.74 b	255.33 \pm 30.14 b
Total tocopherols	265.45 \pm 1.38 ab	266.81 \pm 3.16 ab	279.48 \pm 3.72 a	259.56 \pm 2.72 b	259.89 \pm 30.71 b
Carotenoids	8.07 \pm 1.79 ab	8.25 \pm 0.99 ab	8.55 \pm 1.64 ab	6.45 \pm 1.51 b	8.98 \pm 2.77 a
Chlorophylls	5.18 \pm 0.06 a	4.23 \pm 0.03 b	4.12 \pm 0.02 c	3.80 \pm 0.05 d	3.79 \pm 0.03 d

TPC, total phenolic content.

Different letters on the same row indicate significant differences ($p \leq 0.05$).

Figure 2b
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