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Evolution of carotenoids and volatile compounds in microwave-dried fruits of three different loquat cultivars (*Eriobotrya japonica* Lindl.)

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Abstract:	The consumption of loquat fruits is highly appreciated for their carotenoid content and valuable sensory notes, but it is limited due to the low shelf-life. An on-line temperature controlled microwave system based on infrared thermography was used to dry three different loquat cultivars at 60 °C. The time to reach the target value of 23% moisture content was about 105 min in Claudia fruits and 162 min in Virticchiara and Peluche. Seven carotenoids were identified in loquat fruits, among these the major were all-trans-β-carotene in Virticchiara and Claudia. Virticchiara had the major total carotenoid content (206 µg/g dry basis), followed by Peluche (158 µg/g d.b.) and Claudia (41 µg/g d.b.). The loss of carotenoids after drying ranged between 24% (Peluche) and 41% (Claudia). Carotenoids that showed a higher loss were on average: lutein (70%) and zeaxanthin (51%). Thirty-six volatile compounds were identified in fresh and dried loquats: the aldehydes were the most abundant class. After drying, aldehydes declined slightly, with alcohols falling more sharply. From the sensory analysis, Virticchiara was the most appreciated cultivar. The shortened times by using temperature-controlled microwave heating with infrared thermography have guaranteed a fair quality of the dried loquats from the nutritional and sensory point of view, variable among the three cultivars.
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Dear Editors-in-Chief: **Octavio Paredes-López**

We submit the following original manuscript: “Evolution of carotenoids and volatile compounds in microwave-dried fruits of three different loquat cultivars (*Eriobotrya japonica* Lindl.)”

authors: Vittorio Farina , Luciano Cinquanta*, Francesco Vella, Serena Niro, Gianfranco Panfili, Antonio Metallo, Gennaro Cuccurullo, Onofrio Corona,

for publication in “PFHN – Plant Foods for Human Nutrition” as original research paper.

Loquat fruits are rich in bioactive compounds, but their consumption in fresh form is limited due to the low shelf-life. Till date, loquat microwave dried processing and its effect on fruit quality and bioactive compounds have not been investigated. In this research we evaluated the effects of microwave drying at 60°C, controlled by infrared thermography, on the quality of three cultivars of loquat fruit, by analyzing carotenoid compounds, antioxidants activity, sensorial characteristics and volatile compounds. We think the article is interesting for the journal because of its interdisciplinary character, including arboriculture, food technologies (microwaves, infrared), nutritional and sensory analysis.

The undersigned, prof. Luciano Cinquanta, attests that:

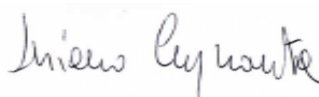
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no portion of the work has been or is currently under consideration for publication elsewhere

no portion of the manuscript, other than the abstract, has been previously published or posted in the Internet

The authors declare that they have no conflict of interest.

Yours sincerely



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Evolution of carotenoids and volatile compounds in microwave-dried fruits of three different loquat cultivars (*Eriobotrya japonica* Lindl.)

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Abstract

The consumption of loquat fruits is highly appreciated for their carotenoid content and valuable sensory notes, but it is limited due to the low shelf-life. An on-line temperature controlled microwave system based on infrared thermography was used to dry three different loquat cultivar at 60 °C. The time to reach the target value of 23% moisture content was about 105 min in *Claudia* fruits and 162 min in *Virticchiara* and *Peluche*. Seven carotenoids were identified in loquat fruits, among these the major were all-trans- β -carotene in *Virticchiara* and *Claudia*. *Virticchiara* had the major total carotenoid content (206 $\mu\text{g/g}$ dry basis), followed by *Peluche* (158 $\mu\text{g/g}$ d.b.) and *Claudia* (41 $\mu\text{g/g}$ d.b.). The loss of carotenoids after drying ranged between 24% (*Peluche*) and 41% (*Claudia*). Carotenoids that showed a higher loss were on average: lutein (70%) and zeaxanthin (51%). Thirty-six volatile compounds were identified in fresh and dried loquats: the aldehydes were the most abundant class. After drying, aldehydes declined slightly, with

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4 alcohols falling more sharply. From the sensory analysis, *Virticchiara* was the most appreciated cultivar.
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6 The shortened times by using temperature-controlled microwave heating with infrared thermography have
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8 guaranteed a fair quality of the dried loquats from the nutritional and sensory point of view, variable among
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10 the three cultivars.
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14 **Key-words:** *Eriobotrya japonica*; carotenoids; drying; microwave; volatile compounds
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17 **1 Introduction**

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21 Loquat (*Eriobotrya japonica* Lindl.) is a subtropical evergreen tree belonging to the *Rosaceae* subfamily
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23 *Maloideae*. Currently, the main producers of loquat are China and Spain but the fruit is commercially
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25 grown in many countries. It is cultivated extensively in the Mediterranean basin especially in the citrus-
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27 growing regions, in Italy, its cultivation is done 90% in Sicily, mainly in the province of Palermo (1).
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29 Inappropriate harvesting can lead to unmarketable fruits; about 20–30% of fresh fruits produced in China
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31 are lost during post-harvest owing to decay and quality decline, producing a thoughtful economic loss (2).
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33 An operation to increase its availability to the consumer is drying, prolonging shelf-life. Pitted dried
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35 loquat can be a healthy snack appreciated by consumers (3), owing to their carotenoid and phenolic
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37 contents and valuable sensory notes, or ingredient in sweets (4). Since a long time is required to obtain
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39 dried products with hot air heating, microwave (MW) processing was proposed as an efficient technique
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41 for moisture removal. In fact, MW processing considerably reduces the time needed to reach the set point
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43 temperature and hence, the entire drying period. Till date, loquat microwave dried processing and its effect
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45 on fruit quality and bioactive compounds have not been investigated. The purpose of this research was to
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47 evaluate the effects of microwave drying at 60°C, controlled by infrared thermography, on the quality of
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49 three cultivars of loquat fruit, by analyzing carotenoids, antioxidants activity, sensorial characteristics and
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51 volatile compounds.
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60 **MATERIALS AND METHODS**

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4 **Plant material**
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7 Loquat fruits were harvested in a commercial farm located near Palermo, Italy (38°04' N, 13°22' E, 150 m
8 a.s.l.) in 2018. The orchard was managed according to the organic farming protocol. Three cultivars were
9 studied: two yellow-fleshed cultivars (*Peluche* and *Virticchiara*) and one white-fleshed (*Claudia*). *Peluche*
10 is a commercial cultivar diffused in many Mediterranean areas, whereas, *Virticchiara* and *Claudia* are local
11 ecotypes. Three uniform trees grafted on loquat seedling rootstock were selected for each cultivar, planted
12 in single rows (north–south oriented), spaced at 5 × 5 m and trained to full globe branched at about 1.2 m
13 from the ground, while reaching 3 to 3.5 m in height. Fruits were hand-picked at the ripe stage suitable for
14 the fresh fruit market using peel color as a maturity index. A sample of 45 fruits per cv (15 fruits x 3 tree x
15 cv.) were used the for physicochemical and sensory analysis on fresh, while other 15 fruits were dried.
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30 **Microwave prototype and drying test**
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33 Drying trials were carried out by means of a lab scale microwave (MW) plant (1 m³), equipped with a
34 magnetron operating at 2.45 GHz with a power output of 2 kW (5). A rotary stirrer has been installed
35 inside the oven, a teflon grid turning table (550 mm diameter) allowed MW drying, while the turntable
36 rotated at 7 rpm (Fig. 1). The destoned and peeled halved fruits (about 300 g) were placed in one layer of
37 the rotating grid for heating, combined with a balance (Gibertini EU-C 1200 RS, Novate Milanese, Italy)
38 located in the upper part of the oven for the online measurement of moisture loss. Temperature control
39 during the complete drying process were detected by IR thermography system (ThermaCAM Flir P65,
40 Canada) located on the upper surface. Based on previous researches (6-7-8), it was decided to dry fruits by
41 MW at 60 °C, because the best results were obtained under similar conditions (*i.e.* on apricots and apples).
42 Just before commencement of the drying process at 60°C, loquat samples were MW blanched at 90°C x 1
43 min to inactivate enzymes and reduce microorganisms. The end of MW drying point (about 23% moisture
44 content, corresponding to water activity = 0.69) was chosen after several preliminary tests, in order to
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4 guarantee at the same time: microbial stability and even appreciable sensory characteristics in dried fruits.
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6 After drying, the samples were vacuum packed in high barrier plastic films and stored in the dark at 4 °C
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8 before being analyzed.
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10 11 **Physico-chemical analyses**

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15 The weight of the different parts of the fruits (Fig. 2) were determined by an analytical balance. The water
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17 activity measurement on dried loquats was done by a water activity meter (Testo 650, Testo Inc., USA) at
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19 25 °C. Total Soluble Solids Content (TSSC) was measured on juice (Brix°) using digital refractometer
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21 Atago Palette PR-32 (Atago Co., Ltd, Tokyo, Japan) and total acidity (TA) (g/L of citric acid) was
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23 measured using a CrisonS compact titrator (Crison Instruments, SA, Barcelona, Spain). Total reducing
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25 compounds, mainly phenolic (TPC), were analysed according to the Folin-Ciocalteu procedure (9). Color
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27 was measured using a CR-300 Chroma Meter Minolta. All determinations were obtained from different
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29 samples in triplicate.
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35 **Sensory analysis**

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38 A panel of 10 judges (4 male and 6 female, aged between 25 and 37 years) defined the sensory profile. All
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40 panelists had a broad expertise in sensory evaluation of fruits and were trained using different samples of
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42 loquat to enable them recognize the qualitative characteristics to be assessed and to generate the attributes
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44 (1-10). The fresh and dried samples were evaluated using twenty attributes, the intensity of each descriptor
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46 was assessed by categorical scores from 1 (absence of sensation), to 9 (extremely intense).
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50 **DPPH radical scavenging method**

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52 Antioxidant activity (AA) has been evaluated by the DPPH method. Loquat samples were homogenised in
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54 25 mL of distilled water and subjected to centrifugation at 4000 g for 10 min, after the supernatant was
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56 filtered by a 0.45 μ m filter (Millipore). The AA analysis was carried out according to reports by (7).
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60 **Volatile organic compounds (VOCs)**

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4 Loquat fruits were subjected to GC/MS analysis to identify the volatile organic compounds (VOCs). The
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6 GC/MS equipment, column, conditions used for analysis and the identification of the compounds were
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8 described elsewhere (11-12).
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10 11 **Carotenoid analysis**

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14 The carotenoid analysis, performed by HPLC, was carried out as reported in a previous paper (13).
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16 17 **Statistical analysis**

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19 All data were tested for differences between the cvs using the one way analysis of variance (ANOVA;
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21 general linear model) followed by Tukey's multiple range test for $P \leq 0.05$ using XlStat® software version
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23 9.0 (Addinsoft, Paris, France). Data of the total phenols, DPPH and groups of volatile compounds were
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25 tested for differences between fresh and dried loquats using Student's t-test for mean comparison ($P \leq$
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27 0.05).
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30 31 **Results and Discussion**

32 33 34 **Physicochemical composition of fresh fruits**

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37 The cv with the highest TSSC content in fresh fruit was *Virticchiara* (14.3 °brix), followed by *Peluche*
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39 (10.4 °brix) and *Claudia* (10.3 °brix). As far as titratable acidity is concerned, the cv that recorded the
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41 highest value was *Peluche* (13.1 g l⁻¹), followed by *Claudia* (11.3 g l⁻¹) and *Virticchiara* (7.5 g l⁻¹).
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43 TSSC/TA is the true index of the fruit's gustative balance: the cv that has shown this highest value was
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45 *Virticchiara* (1.91), followed by *Claudia* (0.89) and *Peluche* (0.80).
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50 51 **Microwave heating**

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53 Drying time at 60 °C, including the blanching, to reach the target value of 23% moisture content, was about
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55 105 minutes in *Claudia* fruits (Fig. 3), while both in *Virticchiara* and *Peluche*, was equal to about 162 min
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57 (Fig. 3). These data are certainly interesting, considering that a convective vacuum drying test at 60 °C
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59 took about 800 minutes to reach around the same moisture content (14). Microwave (MW) drying
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4 significantly reduces the time to reach the selected temperature and therefore the heating period, due to
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6 volumetric MW heating. In these trials, the increasing rate of the initial heating period to reach 90 °C (for
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8 blanching) was just below 3 min and after about one minute, the temperature was reduced to 60 °C.
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10 11 **Phenols and DPPH**

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15 The concentration in total hydrophilic reducing compounds, mainly phenols (TPC), was highest in *Claudia*,
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17 while no significant variations between the other two cultivars were ascertained. A significant decrease was
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19 found ($P < 0.05$) in the TPC after drying: the loss of TPC in dried samples was 40% in *Virticchiara* and
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21 about 25% in the other fruits. Accordingly, the antioxidant activity measured as inhibition of DPPH
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23 scavenger activity showed that *Claudia* had the highest inhibition value (21.5%) and *Peluche* had the
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25 lowest (17.1%). A significant ($P < 0.05$) increase in antioxidant activities (AA) after drying was observed
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27 in all samples: AA had almost tripled in *Claudia* and *Peluche* and slightly less than doubled in *Virticchiara*
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29 fruits (Table 2). This behavior could be explained by oxidation of polyphenols that leads to the formation
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31 of stable intermediates with great antioxidant capacity and the formation of Maillard Reaction Products
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33 (MRPs) known to exhibit various antioxidant properties (15).
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40 **Carotenoids**

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43 HPLC analysis identified seven carotenoids in loquat fruits. *Virticchiara* showed the major total
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45 carotenoids (TC) content: 206.1 µg/g dry basis (d.b.), followed by cultivar *Peluche* (158.1 µg/g d.b.) and
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47 *Claudia* (41.7 µg/g d.b.). In two cultivars, the major carotenoids were all-trans-β-carotene; while β-
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49 cryptoxanthin was predominant in *Peluche* (85.1 µg/g d.b.). Similar results were already observed for
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51 different loquat cultivars (16-17) and are comparable with those found in apricots (18). Taking into account
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53 the Recommended Daily Allowance (RDA) for vitamin A, which is 800 µg/day (19), 100 g of loquats
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55 contribute 33 % of the RDA in *Peluche*, 30% in *Virticchiara* and only 6% in *Claudia*. Significant losses of
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57 carotenoids were found after drying (Table 3), ranging between 24% (*Peluche*) and 41.5% (*Claudia*).
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4 Carotenoids that showed a higher loss were: lutein with an average of 70% and zeaxanthin with an average
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6 of 51%, both being xanthophyll. The high susceptibility at thermal treatment of xanthophyll was also
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8 observed in previous papers (20-21). Regarding *cis* isomers, several studies reported that different results
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10 after thermal treatment could be due to degradation and concomitant increase in *cis*-isomers 24, in our
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12 case, 9-*cis*- β -carotene and 13-*cis*- β -carotene showed a loss of about 43 % and 71%, respectively.
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16 17 **Color**

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20 The color of peeled fruits did not differ significantly among the three cvs for L and b* values (Fig. 4a).
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22 *Claudia* showed, as expected based on the lowest carotenoid content, a lower a* value, corresponding to a
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24 color typical of white fleshed cv. Comparing values of fresh fruit with dried fruit, a reduction of all the
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26 parameters in the three cvs was observed (Fig. 4b). Significant differences between cvs remained only for
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28 the lowest value of the red index (a*) in *Claudia* samples. The development of browning of samples after
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30 drying may be related to carotenoids destruction, as well as enzymatic and non-enzymatic browning (22).
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34 35 **Volatile compounds**

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38 Thirty-six volatile compounds (VOCs) were identified in fresh and dried loquats (data not reported). These
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40 included nine aldehydes, eight organic acids, seven alcohols, five furans, four ketones, two esters and one
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42 hydrocarbon. The aldehydes were the most abundant class (Fig. 5), varying between almost 60% of the
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44 total VOCs in *Virticchiara* to about 40% in *Peluche*, while the amount of aldehydes were greatest in
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46 *Claudia* fruits (about 1402 $\mu\text{g/g}$ d.b.). The other classes of VOCs in order of importance were alcohols,
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48 represented mainly in *Claudia* samples by C6-compounds: *cis*-3-hexeno-1-ol, *trans*-2-hexenol and 1-
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50 hexanol, which have been reported to contribute to the green aromas of loquat (23). Esters, mainly
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52 represented by γ -butyrolactone, prevailed in *Claudia*, as well as organic acids, responsible for floral and
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54 fruity notes (24). MW drying led to an evolution in VOCs: alcohols decreased in all samples, esters
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4 diminished in *Claudia* and *Peluche*, while they were constant in *Virticchiara* dried fruits. The aldehydes
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6 have undergone less variation in the dried products, with a decrease of about 20% only in *Claudia*.
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9 **Sensory analysis**

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12 The results of the sensory analysis on fresh fruits showed significant differences for the descriptors; sweet,
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14 sour, loquat flavor and overall evaluation. The panel's preferences were for the cv *Virticchiara* (Fig. 6). The
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16 greater intensity of the bitter taste perceived in *Claudia* was correlated with the higher content in phenolic
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18 substances, while *Peluche* fruits were characterized by a lower “loquat flavor”, as well as a weak intensity
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20 of sweet. *Claudia* was judged by panelists to be more yellow and less brown in contrast to the *Virticchiara*,
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22 which was the most appreciated cultivar. Although, the differences between the different dried fruits were
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24 much smaller than for the fresh loquats (Fig. 7).
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30 **Conclusions**

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33 The use of temperature-controlled microwaves with infrared thermography (60 °C) has shown good results,
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35 using short period of time (from 105 to 162 min) and ensuring a fair quality of dried loquats, from a
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37 nutritional and sensory point of view. Carotenoids reduction after drying was comprise between 24 and
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39 41%, depending on the cultivar. The study also demonstrated the different behavior of the loquat cultivars
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41 tested using microwave drying. In particular, all the dried cvs showed a good level of appreciation by the
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43 panel test, while the cv *Peluche* was poorly evaluated as a fresh product.
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17 **Figure Legends**

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20 **Figure 1.** Pilot microwave plant.

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23 **Figure 2.** Whole fruit fresh weight (FrW), flesh weight (FIW), skin weight (SkW), seed weight (SdW) in
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25 different loquat cultivars: *Peluche*, *Claudia* and *Virticchiara*. Values represented as mean \pm SD.

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28 (*footnote*) For each column, within the same series, different lowercase letters indicate significantly
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30 different at $P \leq 0.05$ as measured by Tukey's multiple range test. Letter "a" denotes the highest value.
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34 **Figure 3.** Drying curve of pitted halved *Claudia* and *Peluche* loquat fruits.

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37 **Figure 4.** Color CIE L a*b* coordinates in fresh (A) and dried (B) loquat fruits of the three observed cv.

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39 Values represented as mean \pm SD.

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42 (*footnote*) For each column, within the same series, different lowercase letters indicate significantly
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44 different at $P \leq 0.05$ as measured by Tukey's multiple range test. Letter "a" denotes the highest value, ns
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46 means no significant differences.
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49 **Fig. 5.** Amount of volatile compounds ($\mu\text{g/g}$ d.b.) united by chemical groups, in fresh (F) and dried (D)
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51 by microwave at 60 °C in different loquats fruits

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54 For each column, within the same series, different lowercase letters indicate significantly different at P
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56 ≤ 0.05 as measured by Tukey's multiple range test. Letter "a" denotes the highest value; ns means no
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58 significant differences.
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Figure 6. Fresh fruit sensory profile of the three observed loquat cultivars. For each descriptor * indicate significantly different at $P \leq 0.05$.

Figure 7. Fresh fruit sensory profile of the three observed loquat cultivars. For each descriptor * indicate significantly different at $P \leq 0.05$.

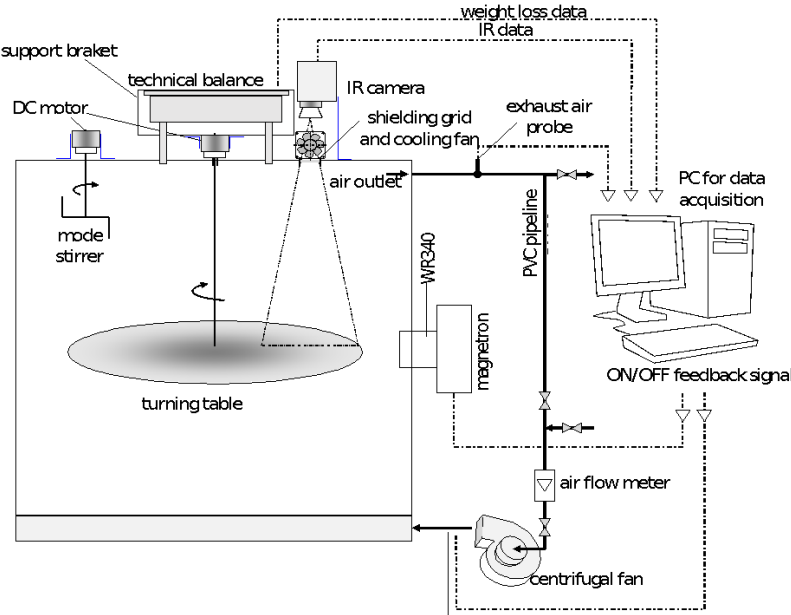


Fig.1

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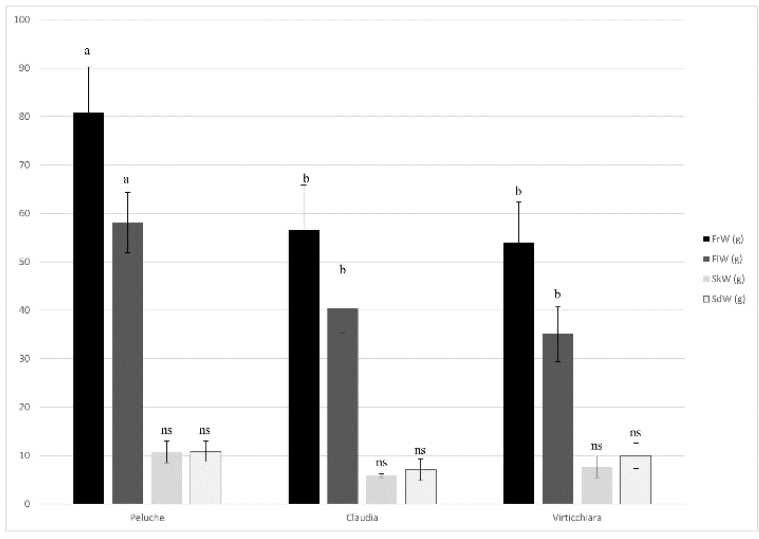


Fig.2

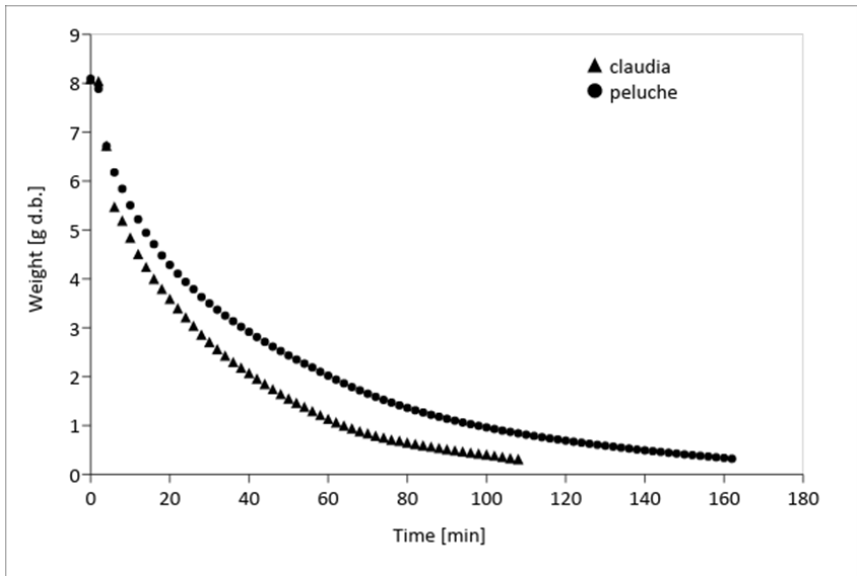
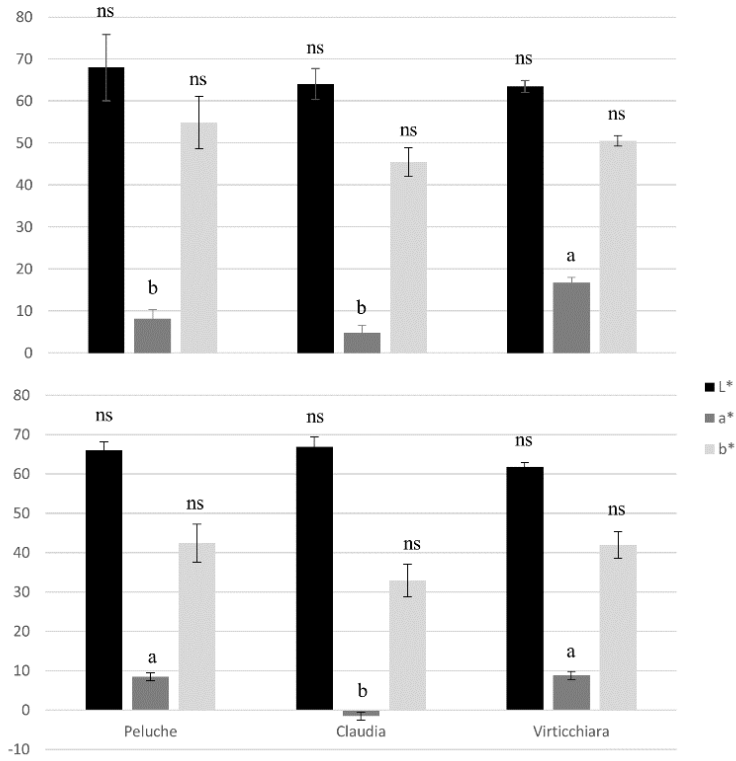


Fig. 3

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Fig. 4

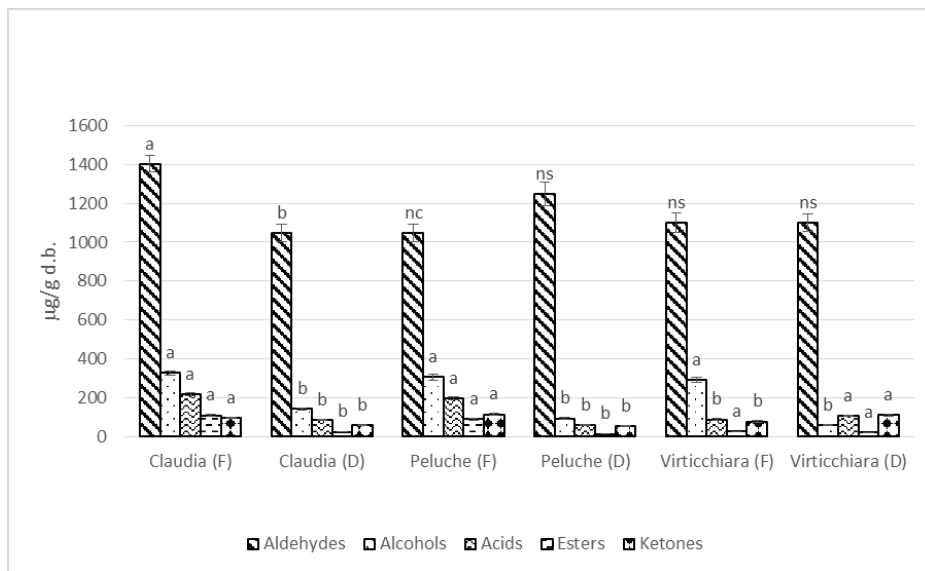


Fig.5

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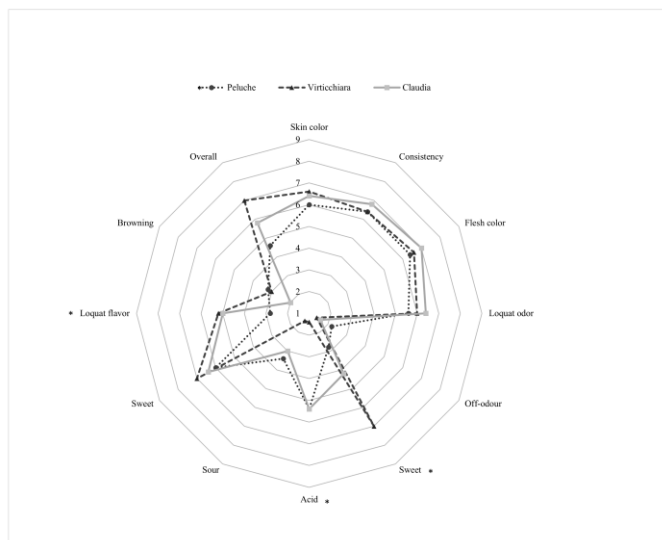


Fig. 6

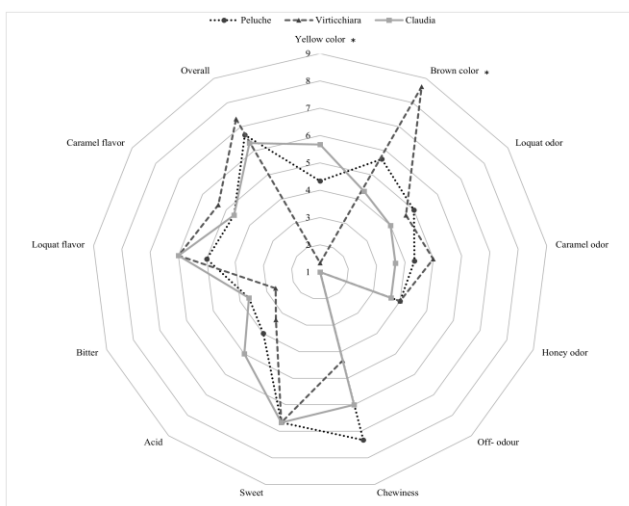


Fig. 7

Table 1

Total Phenols and DPPH in fresh and dried loquats fruit

Fruits	Total Phenols ((+) catechin) (mg/kg d.b)	DPPH %	Total Phenols ((+) catechin) (mg/kg d.b.)	DPPH %
Fresh		Dried		
Claudia	2252.77 ± 80.74 ^a	21.47 ± 1.63 ^b	1706.68 ± 22.21 ^b	58.62 ± 0.97 ^a
Peluche	1720.10 ± 24.44 ^a	17.08 ± 1.38 ^b	1277.87 ± 57.59 ^b	45.34 ± 1.00 ^a
Virticchiara	1688.45 ± 72.25 ^a	20.20 ± 0.79 ^b	1011.86 ± 66.17 ^b	35.94 ± 0.97 ^a

For each column, within the same series, different lowercase letters indicate significantly different at $P \leq 0.05$ as measured by Tukey's multiple range test. Letter "a" denotes the highest value

Table 2Carotenoid composition and content ($\mu\text{g/g}$ d.b.) in fresh and dried loquats.

	<i>Claudia</i> (Fresh)	<i>Claudia</i> MW (60°C)	loss %
lutein	5.9 ± 0.26 ^a	1.2 ± 0.07 ^b	80.3
zeaxanthin	2.3 ± 0.30 ^a	1.3 ± 0.17 ^b	42.7
β -criptoxanthin	13.1 ± 0.21 ^a	10.4 ± 1.82 ^b	20.5
α -carotene	2.5 ± 0.07 ^a	1.60 ± 0.17 ^b	34.0
13cis β -carotene	0.8 ± 0.11 ^a	0.47 ± 0.04 ^b	41.4
all trans β -carotene	15.2 ± 0.26 ^a	8.7 ± 2.30 ^b	42.8
9cis β -carotene	1.9 ± 0.20 ^a	0.7 ± 0.14 ^b	62.2
total carotenoids	41.7 ± 0.26 ^a	24.4 ± 4.58 ^b	41.5
	<i>Virticchiara</i> (Fresh)	<i>Virticchiara</i> MW (60°C)	loss %
lutein	6.9 ± 0.15 ^a	3.04 ± 0.38 ^b	56.1
zeaxanthin	23.5 ± 2.49 ^a	12.2 ± 3.84 ^b	48.1
β -criptoxanthin	69.3 ± 9.83 ^a	68.1 ± 4.02 ^a	1.7
α -carotene	11.3 ± 0.77 ^a	6.7 ± 0.32 ^b	40.4
13cis β -carotene	4.7 ± 0.21 ^a	2.7 ± 0.17 ^b	42.4
all trans β -carotene	77.6 ± 3.86 ^a	47.5 ± 0.48 ^b	38.8
9cis β -carotene	12.8 ± 0.20 ^a	2.6 ± 0.03 ^b	79.4
total carotenoids	206.1 ± 16.79 ^a	142.9 ± 15.35 ^b	30.7
	<i>Peluche</i> (Fresh)	<i>Peluche</i> MW (60°C)	loss %
lutein	3.0 ± 0.38 ^a	0.7 ± 0.02 ^b	74.4
zeaxanthin	7.3 ± 0.58 ^a	2.8 ± 0.08 ^b	61.6
β -criptoxanthin	85.1 ± 13.38 ^a	69.9 ± 0.12 ^b	17.9
α -carotene	7.2 ± 0.87 ^a	6.3 ± 0.18 ^b	12.3
13cis β -carotene	3.2 ± 0.22 ^a	1.8 ± 0.09 ^b	45.1
all trans β -carotene	45.7 ± 7.23 ^a	36.3 ± 1.47 ^b	20.6
9cis β -carotene	6.6 ± 1.11 ^a	2.5 ± 0.28 ^b	61.4
total carotenoids	158.2 ± 23.77 ^a	120.4 ± 3.15 ^b	23.9

For each line, within the same series, different lowercase letters indicate significantly different at $P \leq 0.05$ as measured by Tukey's multiple range test.