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This is the final peer-reviewed author's accepted manuscript (postprint) of the following publication:

*Published Version:*

J. Ortuño, L. Mateo, M. T. Rodríguez-Estrada, S. Bañón (2021). Effects of sous vide vs grilling methods on lamb meat colour and lipid stability during cooking and heated display. MEAT SCIENCE, 171(January 2021), 1-12 [10.1016/j.meatsci.2020.108287].

*Availability:*

This version is available at: <https://hdl.handle.net/11585/776823> since: 2020-10-30

*Published:*

DOI: <http://doi.org/10.1016/j.meatsci.2020.108287>

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1 **Effects of *sous vide* vs grilling methods on lamb meat colour and lipid stability during cooking and**  
2 **heated display**

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9

10 **ABSTRACT**

11 The present study compared the effect of grilling (150°C until 72°C core temperature) and *sous-vide*  
12 (SV) cooking (75°C for 35 min in a water bath under vacuum) on lamb patties immediately after  
13 cooking and after 4 h display at 65°C. Both methods produced patties with similar ( $P>0.05$ ) weight  
14 loss, and moisture and fat contents. SV-cooking prevented ( $P<0.05$ ) the formation of thiobarbituric  
15 acid reactive substances (TBARS) and oxysterols compared to grilled patties, which showed a larger  
16 proportion of highly peroxidisable polyunsaturated fatty acids. Heated display induced dehydration,  
17 surface darkening and a reduction in the hexanal/3-methylbutanal ratio, suggesting the progression  
18 of Maillard reactions. Moreover, TBARS and some lipid oxidation-derived volatiles increased  
19 ( $P<0.001$ ), while cooked-meat aroma compounds were reduced ( $P<0.001$ ). SV-cooking inhibited  
20 ( $P<0.05$ ) the formation of malondialdehyde, and 7 $\alpha$ - and 7 $\beta$ -hydroxycholesterol, and lowered the  
21 cholesterol oxidation ratio during heated display. Overall, SV-cooking may be considered a healthier  
22 way of cooking when lamb meat is to be kept warm for considerable periods before consumption.

23 **Keywords**

24 *Sous vide* cooking, grilling, catering, lipid oxidation, cholesterol oxidation, volatile compounds

25

## 26        **1. INTRODUCTION**

27    Modern lifestyles promote food consumption outside the home, and catering represents a relevant  
28    subsector in this market (Calderón et al., 2018). Catering offers a broad range of services, including  
29    schools, hospitals, hotel buffets, restaurants, take-aways and in-flight meals, thus covering a  
30    considerable share of the daily food intake for a large part of the population. Therefore, the nutritional  
31    quality of food offered by catering facilities is gaining increasing attention. Traditional catering  
32    methods are based on 'making-to-order,' whereby there is no delay between cooking and food  
33    consumption, or bulk production, when food may be held warm for considerable periods before being  
34    served, as in hospitals and canteens (Smith & West, 2003). In the latter case, according to good  
35    hygiene practices, it is recommended that food prepared in advance is kept at a core temperature  
36    above 65°C until consumption (UK Food Safety Agency, 2016; Spanish Royal Decree 3484/2000), which  
37    may in fact represent a prolonged mild cooking treatment. Although recommendations suggest that  
38    this period should be as short as possible to avoid sensory and nutritional losses, heated display times  
39    can be extended for a few hours depending on the service. While meat cooking enhances its flavour  
40    and improves tenderness, it may result in excessive desiccation and lipid oxidation (Grau et al., 2001),  
41    processes that subsequent hot display and exposure to air may aggravate, leading to undesirable  
42    odours, rancidity, texture modification, loss of essential fatty acids (FAs) or the formation of toxic  
43    compounds, including oxysterols and carbonyl compounds (Domínguez et al., 2014). High plasma  
44    concentration of secondary lipid and cholesterol oxidation products (COPs) in humans have been  
45    linked to cytotoxic, mutagenic and carcinogenic effects, and are considered to be involved in  
46    neurodegenerative disorders, and to act as a primary factor in triggering atherosclerosis (Malaguti et  
47    al., 2019; Sousa et al., 2017). Therefore, the use of any strategy able to prevent oxidation in  
48    cooked/hot-displayed meat is highly recommended to improve its sensory traits, nutritional value and  
49    safety.

50 Meat cooking conditions strongly determine the resulting transformations that occur in muscle.  
51 Prolonged cooking times and temperatures above 140°C promote Maillard reactions, crucial for the  
52 development of volatile organic compounds (VOCs), related to the well-appreciated roasty and meaty  
53 flavours, and the toasted colour of meat (Mottram, 1998). Nevertheless, higher cooking temperatures  
54 are also associated with increased lipid and cholesterol oxidation and the reduction of essential FAs  
55 when compared with milder treatments (Rasinska et al., 2019; Rodriguez-Estrada et al., 1997;  
56 Sabolová et al., 2017). The traditional methods used for meat cooking, such as grilling or pan-frying,  
57 involve high temperatures in an aerobic environment during a limited time to enhance flavour  
58 development, while preventing juice losses that may damage the meat texture (Bejerholm & Aaslyng,  
59 2004). *Sous-vide* (SV) is a culinary technique that involves the vacuum-sealing of food inside a plastic  
60 bag before cooking at a mild, controlled temperature for a comparatively long time. In Spain, SV has  
61 become one of the most common methods for preparing lamb meat in the catering industry, using  
62 temperatures of 75-85°C to ensure internal pasteurisation at 65–75°C (Roldán et al., 2015). The SV  
63 method enables juicy, tender and flavourful products to be obtained, whereas the removal of air may  
64 prevent excessive oxidation in the cooked/hot-displayed meat without need for antioxidants.  
65 However, to our knowledge, there is limited information regarding the quality benefits of SV  
66 compared with traditional cooking in catering-like systems involving heated display. For this reason,  
67 the present work aimed to assess the effects of grilling (G) and *sous-vide* (SV) cooking on the oxidative  
68 stability (lean colour, malondialdehyde, COP and VOC formation and FA retention) of cooked/hot-  
69 displayed lamb meat.

## 70 **2. Materials and methods**

### 71 **2.1 Meat sampling and preparation**

72 For the analysis, meat from ten *Segureño* lambs (N=10) was obtained from a local abattoir. After  
73 carcass ageing at 2°C for 48 h, the right lamb legs were removed, deboned by a professional butcher,

74 vacuum packed, frozen and stored at -20°C in darkness until processing. The frozen legs were brought  
75 to -5°C and minced in an atmospheric mincer using a 3-mm plate (Mainca PM98, Barcelona, Spain)  
76 after the removal of the external fat. Salt (2 g NaCl per 100 g of minced meat) was added and  
77 aerobically mixed. From each deboned leg (ca. 1000 g), about forty 25-g patties were prepared and  
78 randomly allocated to the different cooking groups: (i) grilling was carried out in a double-sided griddle  
79 (Media Liscia, Silanos, Milan, Italy) at 150°C until the meat reached a core temperature of 72°C for 20  
80 s; and (ii) *sous-vide* patties were vacuum-packed in polyamide-polypropylene pouches (200 x 295 mm,  
81 thermal resistance -40°C/120°C, oxygen permeability 7 cm<sup>3</sup>/m<sup>2</sup> for 24 h at 4°C and 80% R.H., and water  
82 steam permeability 0.8 g/m<sup>2</sup> for 24 h) (Wipack, Hamburg, Germany) and cooked at 75°C for 35 min in  
83 a water bath to reach a similar final inner temperature. The meat temperature was monitored with a  
84 portable T200 thermometer (Digitron Instrumentation Limited, Hertfordshire, UK). Patties were then  
85 randomly split into two subsets, one composed of freshly-cooked patties (G0/SV0) and the other of  
86 freshly-cooked patties placed on individual covered glass plates and held at 65°C, 800 lux and 80-85%  
87 R.H. for 4 h in a climatic cabinet Climacell 404L (MMM Medcenter Einrichtungen GmbH, München,  
88 Germany) to simulate catering conditions (G4/SV4). In both cases, patties were allowed to cool at  
89 room temperature for 30 min before colour evaluation and further stored at -80°C until lipid and  
90 cholesterol oxidation analysis.

## 91 **2.2 Weight losses, moisture content and CIEL\*a\*b\* colour analysis**

92 The difference in patty weight before and after cooking, and after cooking and catering display was  
93 calculated with the following formula: Weight loss (%) =  $((W_i - W_0)/W_i) \times 100$ ; where  $W_i$  is the weight  
94 of the patty before cooking/immediately after cooking; and  $W_0$  is the weight of the patty immediately  
95 after cooking/after 4 h of the heated display. Moisture content was determined by dehydrating the  
96 samples (5 g) at 102°C (AOAC method 930.15; 2000). Colour before and after hot display was  
97 measured using a CR-200/08 Chroma Meter II (Minolta Ltd., Milton Keynes, UK) with illuminant D65,  
98 2° observer angle, 50-mm aperture size and calibrated against a standard white tile. Reflectance

99 measurements were taken directly on the patty surface. Nine replicate measurements were taken for  
100 each sample (three measurements of three different patties). Lightness ( $L^*$ ), redness ( $a^*$ ) and  
101 yellowness ( $b^*$ ) were determined and expressed as CIEL\*a\*b\* units.

### 102 **2.3 Lipid extraction**

103 Lipids were extracted from the patties following a modified version of the Folch method (Boselli et al.,  
104 2005). Briefly, the lipid fraction was extracted using a chloroform:methanol solution (1:1, v/v) followed  
105 by the addition of another aliquot of chloroform. After mixing with 1 M KCl, the organic phase was  
106 separated and taken to dryness; the fat content was determined gravimetrically. Two independent  
107 replicate measurements were made for each sample.

### 108 **2.4 Determination of TBARS, cholesterol and oxysterols (COPs)**

109 Secondary lipid oxidation was assessed in the patties as thiobarbituric acid reactive substances  
110 (TBARS) (mg MDA/kg meat) according to Botsoglou et al. (1994). Sample absorbance was measured  
111 at 532 nm using a UV2 (Pye Unicam, Cambridge, UK) spectrophotometer and a malondialdehyde  
112 (MDA) standard calibration curve ranging from 0.1 to 10  $\mu$ M ( $y = 0.148x + 0.007$ ;  $R^2 = 0.999$ ).

113 Cholesterol and COPs were extracted and purified as described previously by Cardenia et al. (2015).  
114 Briefly, the lipid extract containing internal standards (142.8  $\mu$ g of betulinol (Sigma Chemical, St. Louis,  
115 USA) and 12.8  $\mu$ g of 19-hydroxycholesterol (Steraloids, Newport, Rhode Island, USA) for cholesterol  
116 and COPs, respectively) were subjected to cold saponification. One-tenth of the unsaponifiable matter  
117 was used to determine the sterol composition, while the remaining 9/10 were purified by SPE-NH<sub>2</sub> for  
118 COPs quantification. Both cholesterol and COPs fractions were silylated by adding 1 mL of  
119 pyridine:hexamethyldisilazane:trimethylchlorosilane (5:2:1, v/v/v), left to stand at 40°C for 20 min,  
120 and then taken to dryness under a nitrogen stream, re-dissolved in *n*-hexane and injected into a Fast  
121 gas-chromatograph/mass-spectrometer (GC/MS) (Inchingolo et al., 2014). Cholesterol and COPs were

122 identified and quantified by comparing their mass spectra and retention times with those of the  
123 corresponding chemical standards (Sigma Chemical; Steraloids (Newport, Rhode Island, USA); Avanti  
124 Polar Lipids (Alabaster, Alabama, USA)) in the SIM acquisition mode, using calibration curves built for  
125 each chemical compound. Cholesterol and COPs were expressed as g/100 g fat and mg/100 g fat,  
126 respectively. The proportion of total cholesterol oxidation (%OR) was also determined according to  
127 the following formula: %OR = (Total COPs/Total cholesterol) x 100 (Cardenia et al., 2015). Two  
128 independent replicate measurements were made for each sample.

## 129 **2.5 Determination of total FA**

130 FA methyl esters were identified and determined from the extracted lipids as described in Cardenia et  
131 al. (2015). About 20 mg of lipid extract were treated with 200  $\mu$ L of diazomethane, before adding  
132 undecanoate methyl ester (CAS 1731-86-8, Sigma-Aldrich) (internal standard, 1 mg/mL) and  
133 transmethylated with 40  $\mu$ L of 2 N KOH in methanol. A 6890N series gas chromatograph (Agilent  
134 Technologies, Madrid, Spain) coupled to a flame ionisation detector (GC-FID), an Agilent HP-88  
135 capillary column (60 m x 250  $\mu$ m x 0.2  $\mu$ m), an He flow rate of 1.4 mL/min and 1:1 split, were used.  
136 The sample injection volume was 1  $\mu$ L. The working temperatures of the injector and FID detector  
137 were 250°C and 260°C, respectively. The oven temperature ramp used was: 125°C initial temperature;  
138 125°C to 145°C at 8°C/min; 145°C for 26 min; 145°C to 220°C at 2°C/min; and 220°C for 1 min. The two  
139 standard mixes used were: (i) FAME mix C4–C24 (Supelco, Bellefonte, PA, USA); and (ii) N<sup>o</sup> 05632  
140 linoleic acid methyl ester, *cis/trans*-isomers mix (Sigma-Aldrich). The results were expressed as relative  
141 abundance (g FA/100 FA).

142 The peroxidation index (PI) of polyunsaturated fatty acids (PUFA) was calculated according to Luciano  
143 et al. (2013) using the following equation:

$$144 \text{ PI} = (\% \text{dienoic} \times 1) + (\% \text{trienoic} \times 2) + (\% \text{tetraenoic} \times 3) + (\% \text{pentaenoic} \times 4) + (\% \text{hexaenoic} \times 5)$$

## 145 2.6 Determination of volatile organic compounds (VOCs)

146 VOCs were determined by headspace-solid phase microextraction (HS-SPME) using an SPME device  
147 (Supelco Co., Bellefonte, PA, USA) containing a fibre coated with carboxen-poly(dimethylsiloxane)-  
148 divinylbenzene (CAR-PDMS-DVB) (50/30  $\mu\text{m}$  thickness). Prior to analysis, the SPME fibre was  
149 preconditioned at 270°C for 60 min in the GC injection port. Semi-frozen muscle samples were ground  
150 with a commercial grinder for 20 s with 20  $\mu\text{L}$  of an aqueous solution of 600 mg/L cyclohexanone as  
151 internal standard, used to rule out any problems during the sample preparation and injection steps  
152 (Rivas-Cañedo et al., 2013). Four grams of meat were weighed into a 20 mL screw-capped amber vial.  
153 Sample vials were flushed with helium (99.9%) for 5 s at 275 kPa to minimise lipid oxidation and  
154 volatile production by residual oxygen. In order to achieve equilibrium between each sample and its  
155 headspace before HS-SPME extraction, vials were kept in a water bath for 10 min at 40°C. The  
156 extractions were performed in an autosampler with a water bath thermostated at 40°C for 45 min and  
157 with continuous stirring (250 rpm) (Gerstel MPS 2XL; Gerstel, Mülheim an der Ruhr, Germany).  
158 Analyses were performed using an Agilent 7890A series gas chromatograph (Agilent, Avondale, AZ,  
159 USA) coupled to an IonTrap GC-MS (Agilent). Analytes were separated using a VF-WAXMS (30 m x 0.25  
160 mm i.d. x 0.5  $\mu\text{m}$  f.t.) column operating at 45 kPa of column head pressure, resulting in a flow of 1  
161 mL/min at 40°C. For analyte separation and identification, the SPME fibre was desorbed in the  
162 injection port at 250°C for 5 min. To improve the recovery of the highly volatile components, the  
163 injection port was operated in splitless mode for the initial 0.6 min, after which a 50:1 split ratio was  
164 established. The initial oven temperature programme was 40°C, which was raised to 130°C at a rate  
165 of 2.5°C/min and then raised to 200°C at a rate of 15°C/min, and maintained at this temperature for  
166 5 min. The transfer line to the mass spectrometer was held at 280°C. The mass spectra were obtained  
167 by electronic impact at 70 eV and a multiplier voltage of 1756 V, collecting data at a rate of 3 scans/s  
168 over the  $m/z$  range 30–300. For identification of the volatile components, *n*-alkanes (Sigma R-8769)  
169 (C5-C20) were analysed under the same conditions to calculate the retention indices (RI). Individual

170 peaks were identified by comparison of their retention indices and mass spectra with those obtained  
171 from commercial reference compounds (Acrōs Organics – Geel, Belgium; Sigma-Aldrich – Steinheim,  
172 Germany). At the same time, mass spectra from the NIST/EPA/NIH Mass Spectral Database (NIST 11,  
173 National Institute of Standards and Technology, Gaithersburg, USA) were used to verify identification.  
174 The results are expressed as mean abundance values multiplied by  $10^{-5}$ . Two independent replicate  
175 measurements for each sample were made.

## 176 **2.7 Statistical analysis**

177 The normal distribution of the data was tested ( $P>0.05$ ) using the Shapiro–Wilk method. The results  
178 were reported as the mean and standard error of the mean (SEM). An analysis of variance (Repeated  
179 Measures Model) was used to ascertain the effect of the cooking method and heated display on the  
180 dependent variables. When necessary, the degrees of freedom were adjusted among the repeated  
181 measures using the Greenhouse and Geisser correction. The least-square differences (LSD) means test  
182 was used to compare the least-square means (LSM), which were considered statistically different at  
183  $P<0.05$ . A principal component analysis (PCA) was performed to represent overall data variability.  
184 Convex hulls were calculated and displayed for each group (G0/SV0/G4/SV4) in the PCA plots. The  
185 data were analysed using the IBM SPSS Statistics 25 software (IBM Software Group, Chicago, IL, USA),  
186 while the correlation and PCA were run with PAST 4.01 (Paleontological statistics software package,  
187 University of Oslo, Norway) (Hammer et al., 2001).

## 188 **3. Results and discussion**

### 189 **3.1 Weight loss, selected nutrients and instrumental colour parameters**

190 **Table 1** shows the effects of cooking/heated display on the weight loss, moisture content, total fat  
191 content and instrumental colour of the lamb patties. Weight losses were similar ( $P>0.05$ ) for the G0  
192 (26.8 g/100g) and SV0 patties (29.1 g/100g), leading to similar relative percentages of moisture and

193 fat. Further dehydration occurred in both treatments during heated display, the total lipid content of  
194 both G4 and SV4 increasing ( $P<0.05$ ) as a result of the lower moisture content. Cooking juice loss  
195 depends on mass transfer during heating, which is directly related with the meat's characteristics, pre-  
196 processing and the cooking conditions applied (Domínguez et al., 2014). Previous studies reported  
197 similar weight losses, moisture and fat contents in meat patties and sausages cooked at 72°C by  
198 conventional procedures (Naveena et al., 2017; Suleman et al., 2020). In contrast, SV cooking (72.5°C  
199 x 2.5 h) was seen to reduce juice loss in rabbit meat compared to roasting (180°C x 60 min) and boiling  
200 (100°C x 20 min) (Rasinska et al., 2019). Likewise, Modzelewska-Kapituła et al. (2019) recorded a  
201 reduction in cooking loss and shear-force in SV-cooked beef compared with steam-cooked meat,  
202 although this might be related to the higher internal core temperature of steaming (75°C) than in SV  
203 (60°C). Meat cuts which suffer similar levels of cooking loss lead to similar tenderness and juiciness  
204 values (Baldwin, 2012). Subsequent heated display for 4 h dried the meat samples, probably due to  
205 the sustained denaturation of sarcoplasmic proteins and the parallel shrinkage of myofibrils  
206 (Tornberg, 2005).

207 As regards colour, the SV0 patties had higher ( $P<0.05$ )  $L^*$  and  $a^*$  values than their G0 counterparts,  
208 while the  $b^*$  values were similar. Subsequent heated display reduced the  $L^*$  and  $b^*$  to reach similar  
209 ( $P>0.05$ ) values in both types of patty, whereas the  $a^*$  value remained higher ( $P<0.05$ ) in SV4 patties.  
210 The denaturation of proteins that occurs above 60°C reduces the typical red colour of meat (provided  
211 by myoglobin) and increases surface light reflectance. However, the final lean colour depends on  
212 factors such as a dry/moist environment, oxygen access and the endpoint temperature reached. Dry-  
213 heat methods, such as grilling, lead to a darker surface colour than moist methods as a result of the  
214 greater dehydration and denaturation caused by direct contact with the heating surface. In the case  
215 of SV, the moist environment and the limited oxygen availability produced by the vacuum-packaging  
216 seems to prevent surface dehydration, myoglobin oxidation and protein denaturation (Naveena et al.,  
217 2017). Meat browning causes a reduction in the  $L^*$ ,  $a^*$ , and  $b^*$  values as the cooking temperature and

218 time increase (Bejerholm & Aaslyng, 2004), which would agree with our findings. The further  
219 maintenance of the meat at 65°C intensified the darkening process of the meat surface. Similar  
220 reductions in L\* and b\* values, associated with meat surface darkening, have been described  
221 previously during prolonged cooking (Kumar et al., 2006), desiccation probably reducing light  
222 reflectance and promoting Maillard reactions (Shahidi et al., 2014).

### 223 **3.2 Lipid and cholesterol oxidation**

224 **Figure 1** represents the effects of cooking/heated display on the levels of malondialdehyde (MDA)  
225 (Fig. 1a) and total COPs (Fig. 1b) in lamb patties. The G0 patties had similar MDA values to those  
226 mentioned previously in lamb patties grilled under comparable conditions (Serrano, Jordán, et al.,  
227 2014; Serrano, Ortuño, et al., 2014). However, SV-cooking led to lower ( $P<0.05$ ) MDA levels than those  
228 obtained after grilling. The increase ( $P<0.05$ ) of the MDA level during heated display was three times  
229 smaller in SV cooked patties (from 0.33 to 0.59 mg MDA/kg meat) than in the grilled patties (from 0.48  
230 to 1.26 mg MDA/kg meat). Thus, the suggested threshold of 1 mg MDA/kg meat for the development  
231 of off-flavours (Domínguez et al., 2014) was exceeded in the G4 patties. The effects of heating on meat  
232 lipid oxidation are well-known: cooking enhances the disruption of cell membranes, promoting  
233 contact between PUFA and pro-oxidant compounds and inducing the protein denaturation that leads  
234 to the loss of antioxidant enzyme activity (Grau et al., 2001). TBARS development has been described  
235 in both grilled meat (Broncano et al., 2009; Serrano, Jordán, et al., 2014) and SV-cooked meat  
236 (Rasinska et al., 2019; Roldán et al., 2014), any difference between the methods generally being  
237 attributed to both the heat transfer process and the time-temperature combination. For instance, SV  
238 rabbit (72.5°C x 2.5 h) presented intermediate MDA values between boiled (100°C x 20 min) and  
239 roasted (180°C x 1 h) meat (Rasinska et al., 2019). Likewise, the MDA concentration increased during  
240 SV-cooking at different temperatures up to 6 h (Roldán et al., 2014). Nevertheless, in foods in which  
241 haem pigments, free ionic iron or salt are present, as in this case, the most critical factor for lipid  
242 oxidation processes to occur is considered to be oxygen availability (Ahn & Kim, 1998). Indeed, lower

243 MDA levels were observed in cooked meat when oxygen availability during cooking was restricted  
244 (Andreo et al., 2003). Moreover, Naveena et al. (2017) found that aerobically boiled pork sausages  
245 contained double the MDA of that measured in SV-cooked sausages at the same temperature and  
246 time after 20 days under similar storage conditions. Our results seem to agree with the above, and the  
247 reduced oxygen availability during SV cooking might have partially limited secondary lipid oxidation  
248 during cooking and prevented its further development during heated display.

249 Six COPs were identified in cooked lamb meat (Table 2): 7 $\alpha$ -hydroxycholesterol (7 $\alpha$ -HC), 7 $\beta$ -  
250 hydroxycholesterol (7 $\beta$ -HC), 5 $\alpha$ ,6 $\alpha$ -epoxycholesterol ( $\alpha$ -EC), 5 $\beta$ ,6 $\beta$ -epoxycholesterol ( $\beta$ -EC),  
251 cholestanetriol (CT) and 7-ketocholesterol (7-KC). After cooking, the most abundant oxysterol in both  
252 types of sample was  $\beta$ -EC, followed by 7-KC, 7 $\beta$ -HC  $\alpha$ -EC, 7 $\alpha$ -HC and CT, this COPs relative abundance  
253 pattern remaining unchanged after the heated display period. COPs formation seemed to follow a  
254 similar trend to TBARS during cooking, since SV-cooking led to lower ( $P<0.05$ ) levels of  $\beta$ -EC, 7 $\alpha$ -HC,  
255 7 $\beta$ -HC and 7-KC than grilling, which was reflected in a lower total COPs content and lower cholesterol  
256 oxidation proportion (%OR). Previous studies made in grilled patties made from light lamb leg  
257 identified the same COPs at similar concentrations (Morán et al., 2012; Ortuño et al., 2020). Cooking  
258 usually favours COPs formation, but their relative composition and accumulation much depend on the  
259 initial oxidative status of the meat, the cooking method applied, and the time-temperature conditions  
260 used (Hur et al., 2007). Different cooking methods have been seen to yield different levels of COPs in  
261 beef (Rodríguez-Estrada et al., 1997), although Broncano et al. (2009) found a similar COPs content  
262 whether chicken meat was fried, roasted, grilled or microwaved. However, to our knowledge, this is  
263 the first study to compare the effect of SV and another cooking method on COPs formation. The  
264 differences found in COPs development might be explained by the lower level of oxygen available  
265 under vacuum-cooking conditions, and/or the higher temperature reached by the outer parts of the  
266 patties in contact with the grilling plate (Rodríguez-Estrada et al., 2014). Subsequent heated display  
267 hardly increased the total COPs content, even though the %OR increased in both G and SV samples.

268 Indeed, 7 $\alpha$ -HC and 7 $\beta$ -HC only increased in the G4 patties, pointing to continued oxysterol formation,  
269 unlike in the SV4 patties. Oxysterols in position 7 are formed by a monomolecular mechanism, in which  
270 cholesterol hydroperoxides undergo dismutation, generating 7 $\alpha$ -HC and 7 $\beta$ -HC, together with 7-KC.  
271 Due to its rapid generation and accumulation, 7-KC is often used as a marker of cholesterol oxidation.  
272 However, no significant increase in this oxysterol was recorded after heated display, which could be  
273 attributed to its dehydration or reaction with an amino group from amino acids, peptides, or proteins  
274 to generate Schiff bases (Rodriguez-Estrada et al., 2014). On the other hand, epoxy derivatives, which  
275 are formed through a bimolecular reaction mechanism between a hydroperoxyl radical and  
276 cholesterol, give rise to CT due to oxirane ring-opening in the presence of H<sub>2</sub>O in an acid environment  
277 (Rodriguez-Estrada et al., 2014). Only slight amounts of CT were detected in both samples after  
278 cooking and heated display, reflecting the usual trend found in meat products (Boselli et al., 2012;  
279 Ortuño et al., 2020). Our results suggest that a certain degree of preliminary cholesterol oxidation may  
280 occur in raw minced meat, probably as a result of aerobic grinding and the addition of salt (Mariutti &  
281 Bragagnolo, 2017), while hydroperoxide dismutation is promoted at 65°C. Total cholesterol was higher  
282 in the G0 than in the SV0 patties but only decreased in the G4 patties, in which %OR increased. Studies  
283 in model systems indicate that cholesterol thermo-oxidation is maximal at around 150°C, just above  
284 its melting point of 147-148°C, when its contact with pro-oxidant agents is promoted (Derewiaka &  
285 Molińska, 2015). At lower temperatures, cholesterol seems to be quite stable when it is  
286 unaccompanied. However, its autoxidation is enhanced by the presence of unsaturated FA, even when  
287 it is not heated. Since the reduction observed in the cholesterol content did not match the quantitative  
288 increase in COPs, it is possible that cholesterol and COPs were also dehydrated or broken down at the  
289 side-chain site, giving rise to conjugated dienes and volatile compounds, respectively. Moreover, as  
290 stated above, a portion of the generated COPs might have reacted with amine-containing compounds  
291 and consequently produced Schiff bases, and/or reacted with Maillard reaction products (Rodríguez-  
292 Estrada et al., 2014).

### 293 3.3 Fatty acid profile

294 **Table 3** shows the major FAs (>5% total FA – TFA) of the cooked/hot-displayed lamb patties. In  
295 descending order, the most abundant FAs present in the cooked lamb fat were C18:1 n-9, C18:0,  
296 C16:0, C18:2n-6 and C14:0, totalling around 90% of TFA. This profile agrees with previous studies on  
297 cooked meat obtained from light lamb legs (Campo et al., 2013; Ortuño et al., 2020). In brief,  
298 cooking/heated display did not alter ( $P>0.05$ ) the proportions of any individual FA, SFA, MUFA or the  
299 n6/n3 ratio. However, highly peroxidisable (HP) PUFA and the peroxidation index (PI), which accounts  
300 for the increasing susceptibility of PUFA to peroxidation as the unsaturation degree of their molecules  
301 increases, were higher in G0 than in SV0 patties. Also, although not significant, a trend ( $P=0.067$ ) was  
302 observed towards an increase in the PUFA concentration in G0. In contrast, no differences were found  
303 for any FA or index after heated display. Changes in the FA profile during cooking may occur either  
304 through losses of melted adipose lipids or as a result of PUFA oxidation (Gerber et al., 2009). Overall,  
305 conventional cooking methods have been reported as having little impact on FA oxidation in ruminant  
306 meat (Campo et al., 2013; Rodriguez-Estrada et al., 1997). Moreover, the protective effect of SV  
307 against PUFA degradation is not a certainty. In rabbit meat, SV cooking prevented PUFA degradation  
308 while roasting did not (Rasinska et al. 2019), although Modzelewska-Kapituła et al. (2019) obtained  
309 the opposite results when comparing PUFA levels in SV and steam-cooked beef. HP-PUFA are generally  
310 associated to cell membrane phospholipids and thus more restricted in movement during heating. In  
311 contrast, triacylglycerides containing SFA and MUFA are more abundant in neutral lipids, and  
312 predominant in intermuscular fat (Gerber et al., 2009; Rodriguez-Estrada et al., 1997). Therefore, the  
313 heat applied during cooking would promote drip loss of the SFA and MUFA present in adipose tissue  
314 rather than of HP-PUFA in a temperature-dependent way. The thermal shock caused by the higher  
315 temperatures of grilling seemingly induced a quantitatively higher release of the neutral lipids present  
316 in the adipose tissue of the patties than the uniform, but more prolonged, heating applied by SV-  
317 cooking. The oxidative degradation of FA involves a free radical mechanism, whose reaction rate is

318 determined by the formation of alkyl radicals from unsaturated FA. Such radicals are formed much  
319 more readily from HP-PUFA than from C18:1 and C18:2. Once such radicals are formed, oxidation  
320 progresses via a chain reaction that is less dependent on the nature of the unsaturated FA, and in  
321 which the more abundant oleic and linoleic acids are involved (Elmore et al., 1999). The findings of the  
322 present study seem to agree with this proposed mechanism. During heated display, the higher PI in  
323 G0 would lead to the increased formation of free radicals capable of attacking other fatty acids that  
324 are less susceptible to oxidation, such as oleic acid, thus justifying the more intense formation of MDA  
325 and 7-hydroxy COPs in grilled meat.

### 326 **3.4 Volatile organic compounds**

327 **Figure 2** shows the different VOCs families present in cooked/hot-displayed patty headspace. This VOC  
328 profile agrees with other profiles reported for cooked lamb meat (Almela et al., 2010; Gravador et al.,  
329 2014; Nieto et al., 2011; Rivas-Cañedo et al., 2013; Roldán et al., 2015), in which lipid-derived  
330 compounds, particularly saturated aldehydes (6-10 carbons), predominated over Maillard-derived  
331 compounds. The proportion of alcohols to aliphatic hydrocarbons in our study was high compared  
332 with other studies in whole intact cuts (Gravador et al., 2014; Roldán et al., 2015). Since alcohols  
333 predominate over alkanes during raw meat oxidation, this fact might be related to the oxidizing effect  
334 of aerobic grinding and salt addition (Ortuño et al., 2016). The VOC profile ( $P>0.05$ ) was similar in the  
335 G0 and SV0 patties, while heated display increased ( $P<0.05$ ) the proportion of aldehydes (from 67 to  
336 80% of total area) and furans, and decreased ( $P<0.05$ ) the proportion of the rest of the VOC families,  
337 except alcohols ( $P>0.05$ ). The formation of aldehydes during heated display was coherent with the  
338 progression of the secondary lipid oxidation rate measured by TBARS. Indeed, lipid oxidation  
339 promoted by mild cooking (60-70°C) of meat for up to 6 h is generally reflected in higher values of  
340 MDA and volatile saturated aldehydes (Del Pulgar et al., 2013; Roldan et al., 2014).

341 **Table 4** shows the relative abundance of individual VOCs in the cooked/hot-displayed patty  
342 headspace. A total of 32 VOCs were identified, all of them previously detected in cooked lamb meat  
343 (Frank et al., 2016; Roldán et al., 2015). The cooking method had a low impact on the VOC profile of  
344 freshly cooked patties since the SV0 patties only presented higher ( $P<0.05$ ) levels of 2-heptenal and  
345 1-pentanol and a lower abundance of limonene compared with their G0 counterparts. Hexanal was  
346 the most abundant compound at both sampling times, representing up to 50% of total VOCs in the  
347 headspace of the cooked patties. However, its content did not increase ( $P>0.05$ ) during heated display.  
348 The increased level ( $P<0.05$ ) of aldehydes was related to the octanal and 3-methyl-butanal contents,  
349 and the neo-formation of benzaldehyde, 2-nonenal and 2,4-decadienal. In this context, the increase  
350 in benzaldehyde and decanal was less pronounced in the SV4 patties. Of the alcohols, 1-octen-3-ol  
351 was the most abundant in the G0 and SV0 patties, increasing during heated display to reach  
352 approximately 10% of total VOCs. The rest of the alcohols, except octanol, decreased (1-pentanol) or  
353 disappeared (1-penten-3-ol, 1-pentanol, 1-hexanol, 1-heptanol) in the G4 and SV4 patties. Similarly,  
354 most of the alkanes, aromatic hydrocarbons, terpenoids (limonene), ketones and sulphur compounds  
355 (carbon disulphide) decreased after cooking ( $P<0.05$ ) or disappeared ( $P<0.05$ ) after heated display,  
356 with some exceptions (hexane, octane and toluene). The most pronounced decrease was that shown  
357 by 2,3-octanedione, from 10.3% in the headspace of freshly cooked patties to 4% after display time.

358 The predominance of hexanal can be attributed to its multiple synthesis pathways, as it can be  
359 generated from the oxidation of oleic, linoleic and arachidonic acids. Other VOCs formed in the  
360 cooked/hot-displayed meat would be related to more specific oxidation routes: octanal from oleic  
361 acid, and benzaldehyde and 2-pentylfuran from linolenic acid, whereas the dialkenals (2-nonenal and  
362 decadienal) are by-products of linoleic acid, and 1-octen-3-ol is partly produced by arachidonic acid  
363 autoxidation (Elmore et al., 1999; Ortuño et al., 2016). As mentioned above, our results suggest that  
364 heating promoted the formation of free radicals from HP-PUFA, which are able to attack other FA that  
365 are less susceptible to oxidation, such as oleic acid, in a more advanced stage of the lipid oxidation

366 process (Elmore et al., 1999). In this sense, octanal and octanol, the most abundant compounds  
367 directly derived from lipid oxidation, have been suggested to come from the  $\beta$ -scission of oleic acid  
368 from 11- or 10-hydroperoxides. The high content of oleic acid ( $\approx 40\%$  TFA) would agree with this  
369 mechanism. Also, the higher PI of the FA profile of G4-patties might justify the greater formation of  
370 benzaldehyde.

371 The VOCs profile allows lipid oxidation and Maillard reactions, concomitant reactions during cooking  
372 that determine flavour development (Almela et al., 2010; Mottram, 1998; Ortuño et al., 2016), to be  
373 assessed in the cooked meat. At low levels, lipid-derived aliphatic aldehydes might play an important  
374 role in cooked lamb flavour, especially when the meat is cooked at mild temperatures, such as during  
375 boiling or SV-cooking (Roldán et al., 2015). However, if the increase exceeds the relatively high  
376 threshold value, unpleasant rancid odours and flavours would be expected (Elmore et al., 1999). At  
377 the same time, a prolonged cooking time would involve VOCs formation from amino acids and  
378 thiamine even at mild temperatures (Del Pulgar et al., 2013). Branched aldehydes (3-methylbutanal)  
379 and alcohols (1-octen-3-ol), compounds formed from the Strecker reaction (Domínguez et al., 2019),  
380 increased after heated display. Indeed, the hexanal/3-methyl-butanal ratio (HX/3MB) has been  
381 proposed for assessing the balance between lipid oxidative reactions and amino acids degradation in  
382 meat products (Del Pulgar et al., 2013). In our study, the HX/3MB ratio was similar ( $P > 0.05$ ) in the G0  
383 (206.2) and SV0 (193.5) patties, but substantially decreased during heated display, implying a higher  
384 ( $P < 0.05$ ) rate of Strecker degradation in G4 (12.6) than in the SV4 (16.3) patties. Despite the fact that  
385 the Maillard reaction rate increases with temperature, the activation energy of the different phases  
386 may vary depending on other factors such as pH, moisture level, the presence of dicarbonyl  
387 compounds and time (van Boekel, 2001). Our results indicate that meat browning and Strecker  
388 reactions can progress when meat is kept at mild temperatures after cooking during prolonged time,  
389 probably due to surface desiccation and the formation of dicarbonyl compounds (derived from lipid  
390 oxidation) (Whitfield & Mottram, 1992). Indeed, the prolonged formation of Strecker aldehydes was

391 already observed in SV-meat kept at similar temperatures (Del Pulgar et al., 2013) and even in long-  
392 ripened meat products (Domínguez et al., 2019).

393 In contrast to Strecker-derived products, other desirable odour-active VOCs, such as carbon  
394 disulphide, limonene, benzene, 2-heptanone and 2,3-octanedione (Calkins & Hodgen, 2007; Frank et  
395 al., 2016; Machiels, 2004), decreased during heated display of the cooked patties, whereas 2-nonenal  
396 and 2,4-decadienal, two top odour-active VOCs that may negatively affect cooked lamb flavour (Bueno  
397 et al., 2011), were formed. Therefore, the VOC profile, TBARS content and instrumental colour  
398 suggested a concomitant progression of both lipid oxidation and Maillard reactions during heated  
399 display. Interactions between both pathways are well-documented and may be interconnected by the  
400 involvement of carbonyl and  $\alpha$ -dicarbonyl compounds from lipid oxidation in the formation of Schiff  
401 bases and the Strecker degradation. However, no common VOCs resulting from this interaction  
402 (pyrazines, pyridines, pyrroles, oxazoles, thiazoles, thiophenes) were found in our study, probably due  
403 to the mild heating applied (Whitfield & Mottram, 1992). A higher degree of protein oxidation, which  
404 is positively correlated with lipid oxidation in meat, may increase the amount of amino acid reactive  
405 carbonyls. This might participate in the initial attack on the amine group of the amino acids and may  
406 explain the differences seen between the SV4 and G4 patties (Gatellier et al., 2010). The increased  
407 abundance of aldehydes observed after heated display to the detriment of other aromatic VOCs,  
408 suggests a possible loss of meatiness (associated with rancidity), which may be supported by the high  
409 level of MDA formation in these samples. Sensory analyses would be necessary to evaluate and  
410 confirm these findings.

### 411 **3.5 Principal component analysis**

412 In order to better understand which parameters were the most relevant for assessing the effect of  
413 cooking treatment and heated display on the lamb patties, the data for colour, lipid and cholesterol  
414 oxidation, FA and VOCs were subjected to principal component analysis (PCA, Fig. 3). The first two

415 principal components accounted for 89.5% of the total variance. PC1 (54.7%) seemed to explain the  
416 changes produced during heated display, as freshly cooked samples (G0/SV0) were clearly separated  
417 from those kept at 65°C for 4 h (Fig. 3b). According to the loadings plot, stearic acid, ketones, alkanes,  
418 aromatic alkanes, limonene, carbon disulphide and some aldehydes (pentanal, 2-octenal) and alcohols  
419 (1-penten-3-ol, 1-pentanol, 1-hexanol, 1-heptanol) showed a strong negative correlation ( $>0.80$ ) with  
420 PC1, forming a cluster of markers of freshly cooked samples. By contrast, 3-methylbutanal, hexanal,  
421 benzaldehyde, octanal, 2-nonenal, 2,4-decadienal, 1-octanol, palmitic acid and  $b^*$  were among the  
422 variables most positively ( $>0.80$ ) correlated with the changes resulting from heated display. The high  
423 positive values in PC1 of both  $L^*$ ,  $b^*$ , lipid-derived aldehydes and 3-methylbutanal may be an indicator  
424 of the joint development of lipid oxidation and Maillard reactions. PC2 (34.8%) best explained mostly  
425 the effects derived from cooking treatment, allowing the two groups (G vs. SV) to be distinguished at  
426 both sampling times (Fig. 3b). All COPs (except CT) and PUFA, HP-PUFA and PI were the most strongly  
427 correlated variables with the G-samples, while, at the opposite extreme, SFA,  $a^*$ ,  $n6/n3$  ratio allowed  
428 separation of the SV-patties. The PCA results reflected the two different patterns explaining the  
429 changes that occurred in the patties: (i) the effect of the cooking treatment, which affected cholesterol  
430 oxidation and the relative increase of peroxidisable FA; and (ii) the effect of heated display, which was  
431 characterised by the pronounced desiccation the meat, and the subsequent formation of Maillard  
432 reaction products, and incipient rancidity to the detriment of the cooked-meat aroma. TBARS was  
433 positively correlated with both PC1 (+0.76) and PC2 (+0.53), reflecting the evolution of oxidation  
434 during the heated display, probably influenced by the higher peroxidability of G-samples.

#### 435 **4. Conclusions**

436 The heated display of cooked meat is increasingly widespread in catering, even though the practice  
437 may impair the cooked meat quality by promoting dehydration, meat surface darkening, lipid  
438 oxidation and the loss of desirable aromatic compounds. Compared to grilling, SV cooking at low  
439 temperatures and for longer time produces a lighter surface colour, does not prevent juice loss, and

440 hinders the formation of lipids and cholesterol oxidation compounds. Most of these positive effects  
441 of SV remain when the cooked meat is displayed on hot plates for a long time. The SV cooking method  
442 was seen to be effective in preventing lipid oxidation during cooking and heated display, as reflected  
443 by the TBARS, COPs and VOCs contents, which may be related to the lower oxygen availability and  
444 lower release of SFA during cooking. Therefore, SV seems to be a healthier cooking method for  
445 catering purposes when food is to be held warm for considerable periods before consumption.

#### 446 **Acknowledgements**

447 We thank Stefano Savioli and Dario Mercatante for their support in the laboratory tasks at the  
448 University of Bologna.

#### 449 **Funding sources**

450 This work was supported by the INIA (Instituto Nacional de Investigación y Tecnología Agraria y  
451 Alimentaria) [RTA2013-00104-C02-00].

452 **Declarations of interest:** none.

453 **Authors contribution:** **JO:** formal analysis, data curation, writing – original draft, visualisation; **LM:**  
454 investigation, data curation; **MTRE:** resources, writing – review & editing; **SB:** conceptualisation,  
455 methodology, validation, resources, writing – review & editing, supervision, project administration,  
456 funding acquisition.

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629 **Table 1.** Effects of cooking method and heated display on the moisture (g/100 g meat), total fat (g/100  
 630 g meat), weight loss (g/100 g) and CIELab colour (L\*a\*b\*) of cooked lamb patties.

		Treatments		SEM	P		
		Cooking	Hot-display		Cooking	Time	CxT
Moisture	G	64.2 <sup>a</sup>	53.6 <sup>b</sup>	0.706	NS	***	NS
	SV	63.7 <sup>a</sup>	54.1 <sup>b</sup>				
Total fat	G	8.89 <sup>b</sup>	12.7 <sup>a</sup>	0.414	NS	***	NS
	SV	9.07 <sup>b</sup>	13.3 <sup>a</sup>				
Weight loss	G	26.8	24.9	0.771	NS	***	NS
	SV	29.1 <sup>b</sup>	23.2 <sup>a</sup>				
L*	G	51.2 <sup>a,y</sup>	37.5 <sup>b</sup>	0.983	**	***	*
	SV	55.9 <sup>a,x</sup>	38.5 <sup>b</sup>				
a*	G	8.72 <sup>y</sup>	9.25 <sup>y</sup>	0.199	***	NS	NS
	SV	10.2 <sup>x</sup>	11.0 <sup>x</sup>				
b*	G	13.0 <sup>a</sup>	10.6 <sup>b</sup>	0.777	NS	**	NS
	SV	13.4 <sup>a</sup>	10.4 <sup>b</sup>				

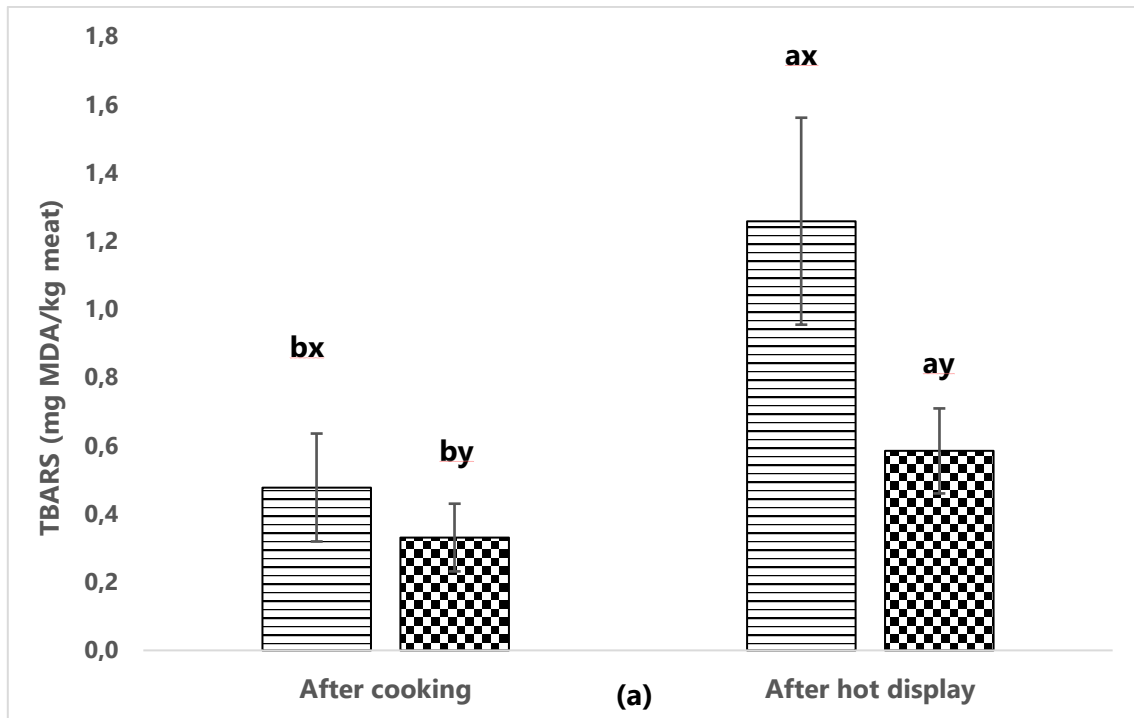
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632 Abbreviations: SV, sous vide; G, grilling; C, cooking; T, time; SEM, standard error of the mean; P,  
 633 probability.

634 CIELab coordinates = L\* (lightness), a\* (redness), b\* (yellowness).

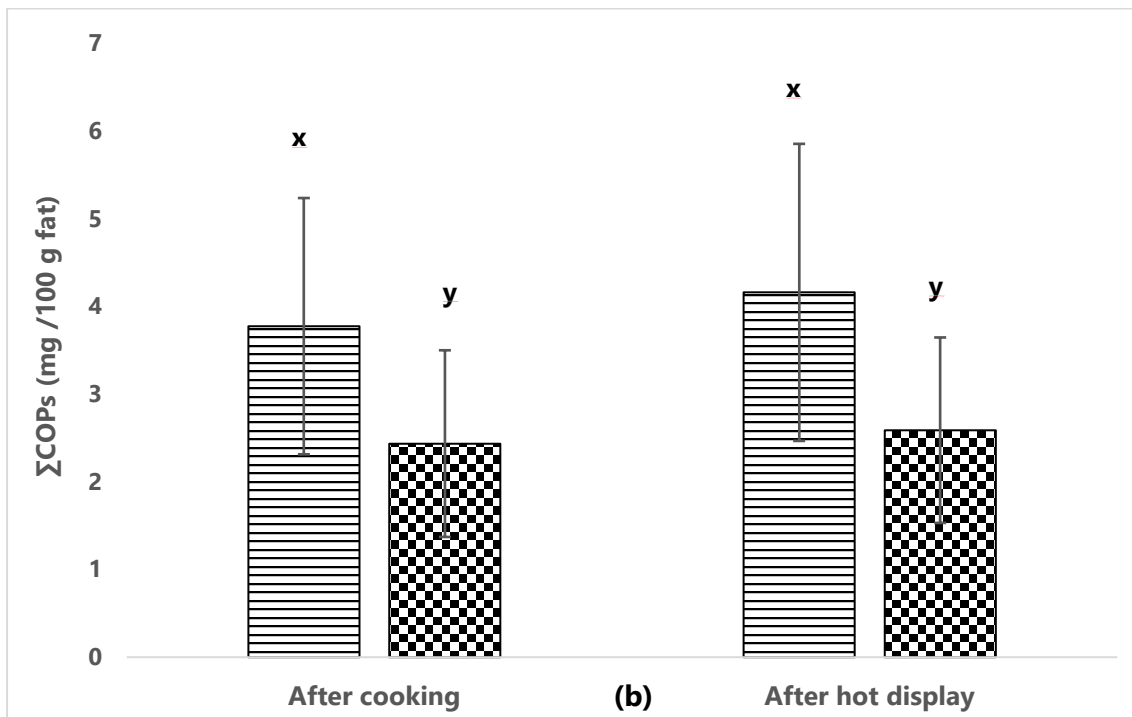
635 <sup>a, b</sup> Heated display time effects ( $P < 0.05$ ); <sup>x, y</sup> Cooking method effect ( $P < 0.05$ ). P: probability values. \*\*\*  
 636  $p < 0.001$ ; \*\*  $p < 0.01$ ; \*  $p < 0.05$ ; NS  $p > 0.05$ .

637 **Fig. 1.** Effect of cooking method and hot display on (a) secondary lipid oxidation (TBARS; mg MDA/kg  
 638 meat) and (b) total cholesterol oxidation products ( $\Sigma$ COPs; mg/100 g fat) in grilled (lines) and *sous*  
 639 *vide* (grid) lamb patties. Mean value  $\pm$  standard deviation.



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643

644 <sup>a, b</sup> Heated display time effects ( $P < 0.05$ ); <sup>x, y</sup> Cooking method effects ( $P < 0.05$ ).

645 **Table 2.** Effects of cooking and heated display time on the mean values of cholesterol oxidation  
 646 products (COPs) (mg/100 g fat), total cholesterol (g/100 g fat) and cholesterol oxidation proportion  
 647 (%OR) in lamb patties.

		Treatment		SEM	P		
		Cooking	Hot-display		Cooking	Time	CxT
7 $\alpha$ -HC	G	0.49 <sup>bx</sup>	0.60 <sup>ax</sup>	0.072	*	*	NS
	SV	0.28 <sup>y</sup>	0.33 <sup>y</sup>				
7 $\beta$ -HC	G	0.80 <sup>bx</sup>	0.95 <sup>ax</sup>	0.107	*	*	NS
	SV	0.49 <sup>y</sup>	0.55 <sup>y</sup>				
$\alpha$ -EC	G	0.50	0.47	0.079	NS	NS	NS
	SV	0.42	0.34				
$\beta$ -EC	G	1.10 <sup>x</sup>	1.01 <sup>x</sup>	0.117	*	NS	NS
	SV	0.65 <sup>y</sup>	0.67 <sup>y</sup>				
CT	G	0.14	0.10	0.029	NS	NS	NS
	SV	0.10	0.15				
7-KC	G	0.83 <sup>x</sup>	0.93 <sup>x</sup>	0.104	*	NS	NS
	SV	0.52 <sup>y</sup>	0.54 <sup>y</sup>				
Total cholesterol	G	1.52 <sup>ax</sup>	1.29 <sup>b</sup>	0.070	*	*	NS
	SV	1.23 <sup>y</sup>	1.19				
OR	G	0.25 <sup>b</sup>	0.32 <sup>ax</sup>	0.029	*	*	NS
	SV	0.19	0.21 <sup>y</sup>				

648  
 649 Abbreviations: SV, sous vide; G, grilling; C, cooking; T, time; SEM, standard error of the mean; 7 $\alpha$ -  
 650 HC, 7 $\alpha$ -hydroxycholesterol; 7 $\beta$ -HC, 7 $\beta$ -hydroxycholesterol;  $\alpha$ -EC, 5 $\alpha$ ,6 $\alpha$ -epoxycholesterol;  $\beta$ -EC,  
 651 5 $\beta$ ,6 $\beta$ -epoxycholesterol; CT, cholestanetriol; 7-KC, 7-ketocholesterol.  
 652 <sup>a, b</sup> Heated display time effects ( $P < 0.05$ ); <sup>x, y</sup> Cooking method effect ( $P < 0.05$ ). P: probability values. \*\*\*  
 653  $p < 0.001$ ; \*\*  $p < 0.01$ ; \*  $p < 0.05$ ; NS  $p > 0.05$ .

654 **Table 3.** Effects of the cooking and heated display time on the mean values of the major fatty acid  
 655 profile (g/100 g FA) and technological indexes of cooked lamb patties.

		Treatment		SEM	P		
		Cooking	Hot-display		Cooking	Time	CxT
C14:0	G	5.02	5.34	0.303	NS	NS	NS
	SV	5.01	5.34				
C16:0	G	25.0	25.7	0.354	NS	NS	NS
	SV	25.0	25.7				
C18:0	G	12.5	12.0	0.598	NS	NS	NS
	SV	12.6	12.2				
C18:1 n-9	G	40.1	40.3	0.813	NS	NS	NS
	SV	41.2	40.2				
C18:2 n-6	G	6.17	6.17	0.280	NS	NS	NS
	SV	5.90	5.90				
∑ SFA	G	46.2	46.3	0.851	NS	NS	NS
	SV	46.3	47.1				
∑ MUFA	G	43.6	44.2	0.765	NS	NS	NS
	SV	44.7	43.9				
∑ PUFA	G	9.55	9.49	0.311	NS	NS	NS
	SV	8.93	8.99				
∑ HP-PUFA	G	2.56 <sup>x</sup>	2.69	0.103	*	NS	NS
	SV	2.31 <sup>y</sup>	2.39				
n6/n3	G	7.81	7.62	0.809	NS	NS	NS
	SV	8.31	8.35				
PI	G	14.6 <sup>x</sup>	14.8	0.464	*	NS	NS
	SV	13.4 <sup>y</sup>	13.6				

656  
 657 Abbreviations: SV, sous vide; G, grilling; C, cooking; T, time; SEM, standard error of the mean; SFA,  
 658 saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; HP-  
 659 PUFA, highly peroxidizable PUFA; PI, peroxidation index.

660 ∑ SFA= Sum of C10:0, C12:0, C13:0, C14:0, C15:0, C16:0, C17:0, C18:0, C20:0, and C22:0.

661 ∑ MUFA = Sum of cis and trans isomers of C14:1, C16:1, C17:1, C18:1, and C20:1.

662 ∑ PUFA = Sum of cis and trans isomers of C18:2, C18:3, C20:4, C20:5, C22:4, C22:5, and C22:6.

663 ∑HP-PUFA: Sum of PUFA with three or more unsaturated bonds.

664 n-6/n-3 = ∑n-6 PUFA/∑n-3 PUFA.

665 PI = (%dienoic×1) + (%trienoic×2) + (%tetraenoic×3) + (%pentaenoic×4) + (%hexaenoic×5)

666 <sup>a, b</sup> Hot-display time effects ( $P < 0.05$ ); <sup>x, y</sup> Cooking method effect ( $P < 0.05$ ). P: probability values. \*\*\* p  
 667 < 0.001; \*\* p < 0.01; \* p < 0.05; NS p > 0.05.

668 **Table 4.** Effects of cooking method and heated display on the relative abundance (as AUx10<sup>5</sup>) of lipid  
 669 oxidation volatiles in lamb patties.  
 670

Compound		Treatment		SEM	C	T	CxT		
		Cooking	Hot-display						
<i>Aldehydes</i>									
Pentanal	G	12.2	a	9.32	b x	0.545	*	***	NS
	SV	11.5	a	8.29	b y				
3-Methylbutanal	G	1.04	b	9.44	a	0.412	NS	***	NS
	SV	1.32	b	8.29	a				
Hexanal	G	115		120		6.948	NS	NS	NS
	SV	114		136					
Heptanal	G	14.3		16.4		1.370	NS	NS	NS
	SV	15.4	a	12.2	b				
2-Heptenal(E)	G	0.05	y	0.03		0.018	***	**	*
	SV	0.16	a x	0.06	b				
Octanal	G	4.68	b	63.9	a	5.991	NS	***	NS
	SV	5.08	b	63.9	a				
2-Octenal(E)	G	0.03	a	nd	b	0.008	NS	***	NS
	SV	0.04	a	nd	b				
Nonanal	G	9.04	a	0.97	b	0.596	NS	***	NS
	SV	9.59	a	0.86	b				
Benzaldehyde	G	nd	b	1.02	ax	0.049	***	***	**
	SV	nd	b	0.54	ay				
2-Nonenal(E)	G	nd	b	0.03	a	0.004	NS	***	NS
	SV	nd	b	0.04	a				
Decanal	G	<0.01	b	0.01	a	0.001	NS	*	NS
	SV	<0.01		<0.01					
2,4-Decadienal(E)	G	nd	b	<0.01	a	<0.01	NS	***	NS
	SV	nd	b	<0.01	a				
<i>Alcohols</i>									
1-Penten-3-ol	G	2.13	a	nd	b	0.063	NS	***	NS
	SV	2.54	a	nd	b				
1-Pentanol	G	5.79	ay	1.52	b	0.177	*	***	NS
	SV	7.76	a x	1.17	b				
1-Hexanol	G	1.73	a	nd	b	0.075	NS	***	NS
	SV	1.96	a	nd	b				
1-Heptanol	G	0.50	a	nd	b	0.025	NS	***	NS
	SV	0.60	a	nd	b				
1-Octen-3-ol	G	17.6		25.4		1.111	NS	**	NS
	SV	20.8	b	35.2	a				
1-Octanol	G	<0.01	b	0.24	a	0.009	NS	***	NS
	SV	<0.01	b	0.23	a				
<i>Furans</i>									
2-Pentylfuran	G	1.22	b	1.88	a	0.285	NS	**	NS
	SV	1.12	b	1.54	a				
<i>Linear alkanes</i>									
Hexane	G	nd	b	1.17	a	0.118	NS	***	NS
	SV	nd	b	1.54	a				
Heptane	G	9.63	a	1.30	b	0.184	NS	***	NS
	SV	9.44	a	1.27	b				
Octane	G	3.66		4.11		0.223	NS	NS	NS
	SV	3.92		4.34					

Decane	G	0.82	<sup>a</sup>	nd	<sup>b</sup>	0.083	NS	***	NS
	SV	1.08	<sup>a</sup>	nd	<sup>b</sup>				
Dodecane	G	0.02	<sup>a</sup>	nd	<sup>b</sup>	0.001	NS	NS	NS
	SV	0.01	<sup>a</sup>	nd	<sup>b</sup>				
<i>Aromatic hydrocarbons</i>									
Benzene	G	0.40	<sup>a</sup>	nd	<sup>b</sup>	0.015	NS	***	NS
	SV	0.48	<sup>a</sup>	nd	<sup>b</sup>				
Toluene	G	1.18	<sup>a</sup>	1.56	<sup>b</sup>	0.096	NS	***	NS
	SV	0.89	<sup>a</sup>	1.22	<sup>b</sup>				
Ethylbenzene	G	0.12	<sup>a</sup>	nd	<sup>b</sup>	0.008	NS	***	NS
	SV	0.11	<sup>a</sup>	nd	<sup>b</sup>				
o-Xylene	G	0.12	<sup>a</sup>	nd	<sup>b</sup>	0.008	NS	***	NS
	SV	0.11	<sup>a</sup>	nd	<sup>b</sup>				
<i>Terpenoids</i>									
Limonene	G	0.08	<sup>a x</sup>	nd	<sup>b</sup>	0.006	*	***	NS
	SV	0.04	<sup>a y</sup>	nd	<sup>b</sup>				
<i>Ketones</i>									
2-Heptanone	G	1.00	<sup>a</sup>	nd	<sup>b</sup>	0.021	NS	***	NS
	SV	0.97	<sup>a</sup>	nd	<sup>b</sup>				
2,3-Octanedione	G	24.5	<sup>a</sup>	11.9	<sup>b</sup>	0.917	NS	***	NS
	SV	25.2	<sup>a</sup>	11.9	<sup>b</sup>				
<i>Sulphur compounds</i>									
Carbon disulphide	SV	2.69	<sup>a</sup>	0.62	<sup>b</sup>	0.205	NS	***	NS
	G	3.52	<sup>a</sup>	0.82	<sup>b</sup>				

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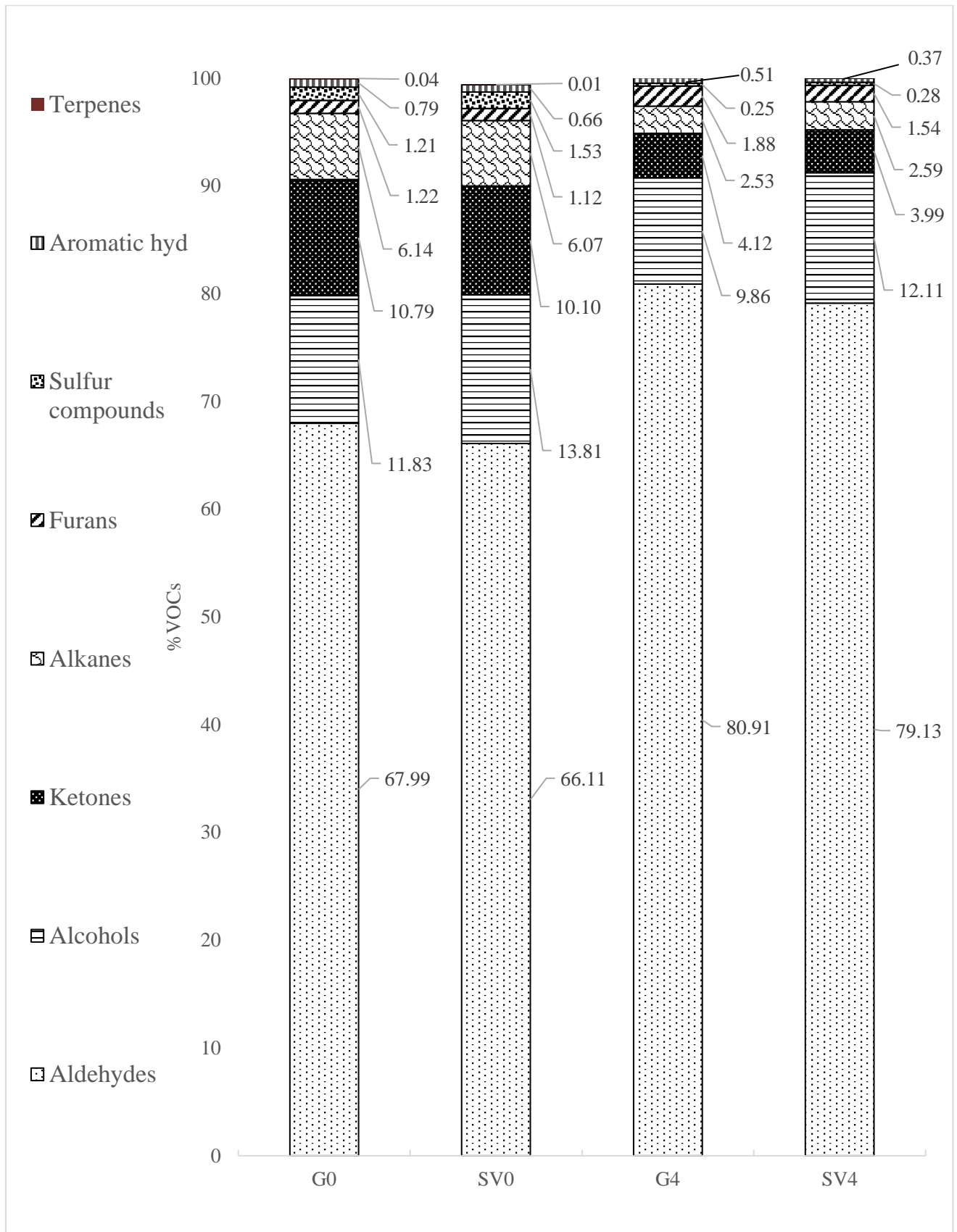
672 Abbreviations: SV, sous vide; G, grilling; C, cooking; T, time; M: Mean; SEM, standard error of the  
673 mean.

674 <sup>a, b</sup> Heated display time effects ( $P < 0.05$ ); <sup>x, y</sup> Cooking method effects ( $P < 0.05$ ). P: probability values.

675 \*\*\*  $p < 0.001$ ; \*\*  $p < 0.01$ ; \*  $p < 0.05$ ; NS  $p > 0.05$ .

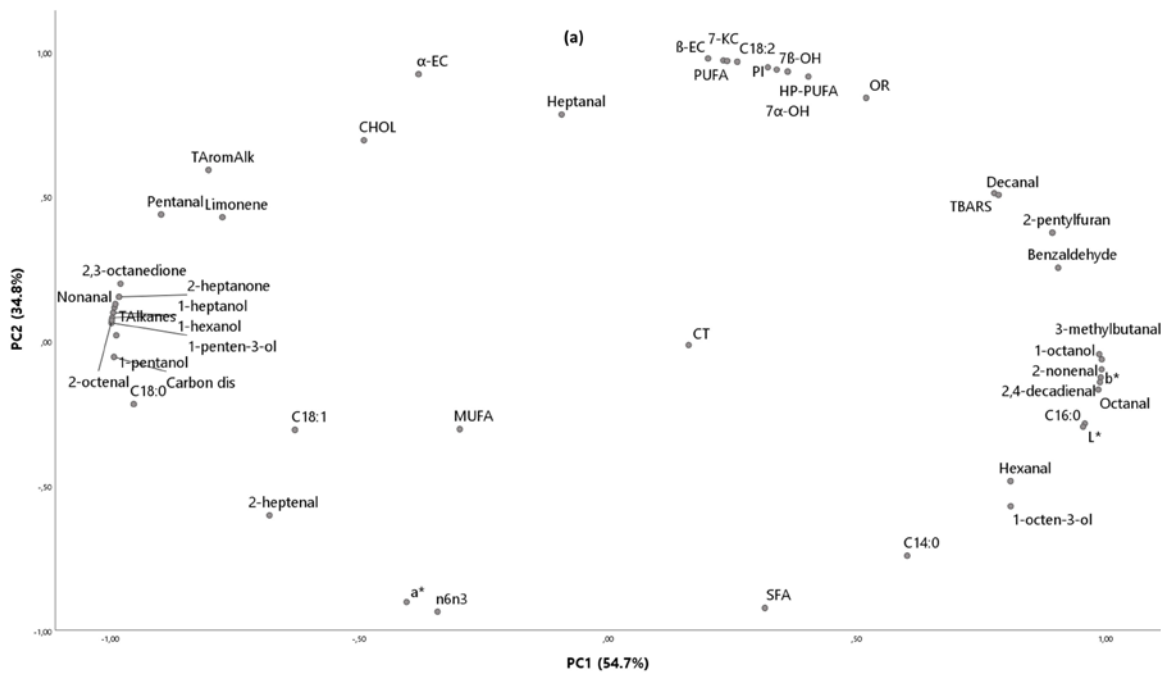
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677 **Fig. 2.** Relative abundance of VOC families (% of total area) in the headspace of grilled (G) and sous  
 678 vide (SV) lamb patties after cooking (0) and heated display (4).



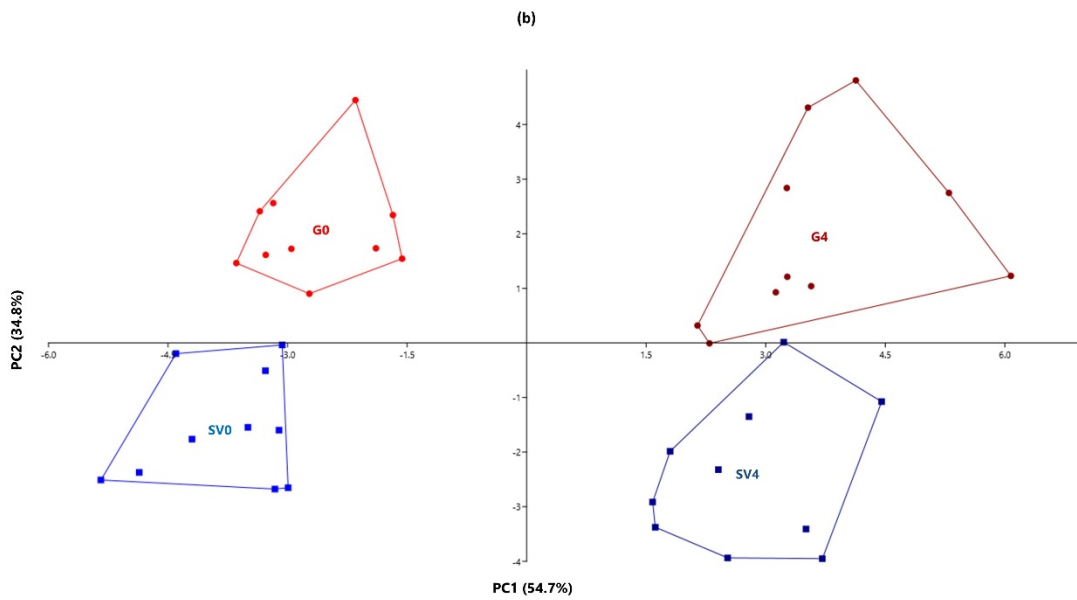
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681 **Fig 3.** Graphical representation of the loadings (a) and scores (b) of the first two Principal  
 682 Components (PC).



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