Influence of cooking conditions on the migration of silicone oligomers from silicone rubber baking molds to food simulants

Yi-Qi Liu [a], Magdalena Wrona [b], Qi-Zhi Su [b], Paula Vera [b], Cristina Nerín [b,*], Chang-Ying Hu [a,*]

[a] *Department of Food Science & Engineering, Jinan University, Huangpu West Avenue 601, Guangzhou City 510632, Guangdong, China*

[b] *Department of Analytical Chemistry, Aragon Institute of Engineering Research I3A, CPS-University of Zaragoza, Torres Quevedo Building, María de Luna 3, 50018 Zaragoza, Spain*

ABSTRACT

The stability, surface micromorphology, and volatile organic compounds (VOCs) of silicone rubber baking molds (SRBMs) were tested while using the molds under severe conditions: baking at 175 ◦C, microwaving at 800 W, and freezing at −18 ◦C. Moreover, migration tests of SRBMs to food simulants (isooctane, 95% ethanol, and Tenax®) at 70 ◦C for 2 h (accelerated conditions) were performed. The initial total VOCs concentration was 2.53% higher than that recommended by BfR Recommendations on Food Contact Materials. Therefore, the SRBM samples were considered as badly tempered materials, and 18 different types of silicone oligomers were identified during the migration tests. The following percentage of silicone oligomers with a molecular weight lower than 1000 Da in isooctane, 95% ethanol, and Tenax® were detected: 70.7%, 91.8%, and 97.2%, respectively. It has been proven that previous baking treatments effectively reduced the content of silicone oligomers migrating from SRBMs.

Keywords: Silicone rubber baking molds Migration Silicone oligomers Severe processing conditions Volatile organic compounds Gas chromatography

1. Introduction

Silicone rubber can be characterized by good high-temperature resistance, high-pressure resistance, flexibility and a non-sticky surface (Akbar, Ullah, & Alam, 2019; Helling, Mieth, Altmann, & Simat, 2009), so it is widely used in food contact materials, such as baking molds, pressure cooker seals, baby bottle teats and food packaging (Gross, 2015). In silicone rubber, the silicon atom is connected with organic groups (Rücker & Kümmerer, 2015). This chemical structure means that the silicone rubber has good thermal resistance, both at very high or low temperatures. Silicone rubber products can be obtained by curing treatment after the polymerization of non-natural linear and circular siloxane molecules using a crosslinking agent. Due to the imperfect production process of silicone rubber products, oligodimethylsiloxanes appeared as the reaction side-products from the polymerization process (Helling, Seifried, Fritzsche, & Simat, 2012). Moreover, most of the production process of silicone rubber requires an additional step at high temperature, over 200 ◦C, for at least 6 to 8 h to eliminate the unreacted side-products. In the absence of this step, when the silicone rubber product is used, the migration of oligomeric dimethylsiloxane to food will be greater. Therefore, these substances can be classified as non-intentionally added substances (NIAS) (Wrona &

Nerin, 2019). Over the last few decades, the market demand for silicone rubber baking molds (SRBMs) has increased (Mojsiewicz-Pienkowska, Jamr´ogiewicz, Szymkowska, & Krenczkowska, 2016), but neither the authorities nor consumers know if the process of eliminating the unreacted side-products has been applied. The largest markets for silicone rubber are China and Western Europe (Mojsiewicz- Pie´nkowska & Krenczkowska, 2018). Volatile methylsiloxanes are lipophilic and environmentally persistent substances. When the molecular weights of methylsiloxanes increase, their volatility decreases (Rücker & Kümmerer, 2015). Silicone rubber readily releases silicone oligomers after prolonged contact with fats (Gross, 2015), which could be harmful to human health, particularly low molecular weight silicone oligomers (Mojsiewicz-Pienkowska et al., 2016; Onnekink, Kappel, Boelens, & Pruijn, 2020). For example, octamethylcyclotetrasiloxane (D4) can affect estrus (Burns-Naas et al., 2002) and according to EU classification can be toxic to reproduction. Moreover, silicone oligomers from SRBMs can migrate to food during the baking process (Y. Liu et al., 2019; Y. Q. Liu et al., 2020). SRBMs are used in a variety of kitchenware, such as baking and microwave ovens, and refrigerators (Deleu et al., 2019). Cooking in silicone bags is a fashion in some developed countries. These bags are reusable, which makes them attractive for consumers. and save energy and time when using microwave heating (Flores-Ch´avez & Monta˜nez-Saenz, n.d.; Yolacaner, Sumnu, & Sahin, 2017). When silicone rubber is used in a baking mold, severe food processing conditions, such as high temperature, low temperature, microwave, even long-time reuse, may affect its characteristics, resulting in the migration of chemicals from SRBM to food especially silicone oligomers. In a particular case study, silicone molds were used daily in a commercial pizza bakery up to 1700 times; even so the detected concentration of VOCs in them was around 0.14% (Helling et al., 2012). Therefore, these materials should be evaluated regarding the migration limits to ensure food safety (Magdalena Wrona & Nerín, 2020). It should be highlighted that silicone oligomers are not present in EU regulation No. 10/2011 and its amendments (Union Europea, 2011) for food contact materials. Therefore, silicones commercialized in the EU market must be covered by European national legislation and must comply with the Frame Regulation 1935/2004/CEE (European Parliament and the Council, 2004). Resolution ResAP (2004) 5 on silicones (Council of Europe, 2004) to be used for food contact applications released by the Council of Europe, does not contain a restriction on the migration of polydimethylsiloxane, but provides acceptable daily intake or tolerable daily intake limited to 1.5 mg/kg bw. German Federal Institute for Risk Assessment (BfR) Recommendations on Food Contact Materials (BfR-German Federal Institute for Risk, 2019) for silicone rubber products requested that silicone rubber products must release no more than 0.5% volatile organic compounds and no more than 0.5% extractable components. The specific migration limit of the silica coating (SiOx) treatment agent consisting of the monomers hexamethyldisiloxane and hexamethyldisilazane on the PET surface is limited to 0.05 mg/kg (measured as hexamethyldisiloxane) in Spain RD847/2011 (Ministerio de Sanidad Política Social e Igualdad, 2011), and for the substances without specific migration restrictions or any other restriction, the specific migration limit generally applicable is 60 mg/kg. Nevertheless, there is a lack of clear and agreed (between enforcement authorities and testing laboratories) guidelines based on toxicity data provided by the European Chemicals Agency (ECHA) and the Threshold of Toxicological Concern (TTC) models for risk assessment of not officially evaluated substances. The purpose of this investigation was the study of the migration of silicone oligomers from SRBMs to food simulants. SRBMs were subjected to severe conditions simulating the real use of baking molds by consumers. Behind this aim, there was a hypothesis that SRBMs are not safe materials. The stability, surface micromorphology, and total volatile organic compounds (VOC) of the used SRBMs were studied. The qualitative analysis of silicone oligomers was performed by gas chromatography–atmospheric pressure chemical ionization-quadrupole time of flight mass spectrometry (GC–APCI-QTOF-MS), while the quantitative analysis was performed by GC–MS.

2. Materials and methods

2.1. Chemicals

Hexadecane (CAS 544-76-3) and decamethylcyclopentasiloxane (CAS 541-02-6) were purchased from Sigma-Aldrich Química S.A (Madrid, Spain). Isooctane (CAS 540–84-1) and ethanol (HPLC grade, CAS 64-17-5) were from Scharlau Chemie S.A. (Sentmenat, Spain). Tenax® 80/100 mesh was from Supelco (Bellefonte, PA). Wasserlab Ultramatic GR system (Barbat´ain, Spain) was used to obtain ultrapure water.

2.2. Materials

Purple silicone rubber baking molds with size 32 cm × 24 cm × 4 cm and thickness equal to 1.50 ± 0.05 mm were purchased in the same batch in the Chinese market in 2019. Four silicone rubber molds were used. Selection of samples was based on results from our previous study (Y. Q. Liu et al., 2020).

2.3. Sample preparation

In daily life, SRBMs are often used in the oven at 175 ◦C, in the microwave oven at 800 W, and in the freezer at − 18 ◦C. Therefore, these harsh conditions of application of SRBMs can be used for the simulation of real usage of baking molds by consumers. Therefore, the cleaned and dried SRBMs were kept in a UF110 universal stove (Memmert GmbH, Germany) at 175 ◦C for 30 min, 150 min, and 300 min. Also, they were kept in a microwave oven (LG Electronics Inc, South Korea) set to 800 W power for 5 min, 25 min, and 50 min. Finally, the SRBMs were kept in a − 18 ◦C Liebherr Combi Fridge CNPES4858 (Liebherr, Kirchdorf an der Iller, Germany) for 60 min, 300 min, and 600 min. The untreated mold was used as a control sample (blank).

2.4. Attenuated total reflection-Fourier transform infrared (ATR-FTIR) analysis

The stability of SRBMs was tested using an FTIR spectrometer (Thermo Fisher Scientific) equipped with an ATR (Thermo Fisher Scientific) with a diamond crystal. This technique was used to assess the structural changes in different types of analyzed materials (Zhu, 2019). The scanning range was 500–4000 cm− 1.

2.5. Atomic force microscopy analysis (AFM)

The SRBM samples were cut into pieces (1 cm × 1 cm) and their surface micromorphology was analyzed by AFM (Ntegra Prima; NT-MDT Spectrum Instruments, Moscow, Russia). NanoScope Analysis software was used to obtain 3D topographic pictures, the average roughness (Rα), and root mean square roughness (Rq) of the surfaces of SRBMs. Six replicates of each sample were analyzed.

2.6. Determination of total volatile organic compounds

The determination of total VOCs was performed according to the Chinese National Standard GB 28482-2012 method (GB 28482-2012, 2012): 10 g of SRBMs were cut into 2 × 2 cm squares and stored over dried CaCl2 for 96 h. Because silicone rubber has a certain ability to absorb moisture, it needs to be fully dried. Thereafter, 10 g were weighed into a glass bowl and heated for 4 h at 200 ◦C, conditioned in a desiccator, and weighed again. The loss of weight, expressed as a percentage, gives the amount of volatile organic compounds.

2.7. Migration study The migration studies were carried out according to EU Regulation No. 10/2011 (European Union, 2011), and the guidelines for testing kitchenware tools written by the Joint Research Center (JRC) from the EU Commission, which indicates the application of isooctane and 95% ethanol as fatty food simulants and Tenax® as a solid food simulant. Briefly, 0.0113 dm2 SRBMs were placed on the lid of a 20-mL screw-cap glass vial with direct contact to food simulant. SPME headspace vials with screw cap and white PTFE/red silicone septum prevented loss of VOCs. The following amounts of food simulants were added to the glass vial: 1.88 mL (1.48 g) of 95% ethanol, 1.88 mL (1.30 g) of isooctane, and 0.0452 g of Tenax®. Then, the vials were sealed and stood upside-down. Migration was carried out at 70 ◦C for 2 h. Pure liquid simulants were used as blanks and the empty glass vial (blank for solid food simulant). After migration exposure, 12 µL of 3560 µg/g of hexadecane standard solution in hexane were added as an internal standard for 95% ethanol and isooctane. The final concentration of hexadecane in the migration solutions was 15 µg/mL (23 µg/g), while Tenax® was extracted with 1.88 mL (1.23 g) of *n*-hexane and ultrasonicated for 30 min. The extract was filtered using a 0.22-µm PTFE syringe filter. Then 12 µL of 3560 µg/ g of hexadecane standard solution in hexane were added to Tenax® extract. Hexane blank was also filtered using a 0.22-µm PTFE syringe filter. Finally, 95% ethanol, isooctane, and Tenax® extract in hexane were injected directly into the GC–MS and GC–APCI-QTOF-MS. It should be explained that according to previous experiments, *n*-hexane had a better extraction ability for D4–D6 in silicone products than methanol. That is why *n*-hexane was chosen as an extracting solvent in this case. Moreover, due to the physical properties of Tenax® (very fine powder), the extracting solvent cannot be transferred well, and some losses can be observed. Therefore, it was considered that ultrasonic extraction for 30 min is sufficient, and no second extraction was performed as suggested in the literature (Chakraborty & Reddy, 2017).

2.8. Gas chromatography–mass spectrometry (GC–MS)

To perform chromatographic separation an Agilent Technologies 7820A system gas chromatograph coupled to a mass spectrometer (5977B MSN) from Agilent Technologies (Madrid, Spain) was used. The capillary column HP-5MS (30 m × 0.25 µm × 250 µm) from Agilent Technologies (Madrid, Spain) was used. The oven program was as follows: 40 ◦C for 2 min, with a rate of 20 ◦C/min up to 315 °C, maintained for 8 min. The injection type was splitless, injection volume was 1 µL and the helium flow was 2.02 mL/min. The injector temperature was 250 ◦C. The acquisition was done in electron ionization (EI) mode. The mass detector was set in scan mode (in the range *m*/*z* 50–800).

2.9. Gas chromatography–atmospheric pressure chemical ionization-quadrupole time of flight mass spectrometry (GC–APCI-QTOF-MS)

Chromatographic separation was performed using a CTC Analytics CombiPal autosampler together with a 6890 N chromatograph from Agilent Technologies (Madrid, Spain). An HP-5MS capillary column (30 m × 0.25 µm × 250 µm) from Agilent Technologies (Madrid, Spain) was used. The oven program was as follows: 40 ◦C for 2 min, with a rate of 20 ◦C/min up to 315 ◦C, maintained for 8 min. The injection type was splitless, injection volume was 1 µL and the helium flow was 1.2 mL/ min. Quadrupole-time of flight analyzer (QTOF) Xevo G2 from Waters (Milford, MA) was used as a detector in MSE mode. The following parameters of analysis were applied: API positive polarity; sensitivity mode; collision energy ramp from 15 to 40 V; temperature of the probe was 200 ◦C; the corona voltage 2.2 kV; sampling cone voltage was 30 V; extraction cone voltage was 3 V; the source temperature was 150 ◦C and the desolvation gas flow was 200 L/h. MassLynx software from Waters was applied for the collection and processing of obtained data.

2.10. Quantitative analysis

Since silicone oligomers standards were difficult to obtain, a semi-quantification analysis was performed using decamethylcyclopentasiloxane (D5) as calibration standard. Moreover, hexadecane in hexane was applied as an internal standard (IS) solution. The final concentration of IS in D5 was 15 µg/mL (23 µg/g). The calibration curve was plotted using standards in the range of 0.013–48 µg/g. All standards were directly injected into the GC–MS. Analytical parameters of the GC–MS method, such as limits of detection and quantification (LOD and LOQ), linearity, and correlation coefficient, were determined (Bratinova, Raffael, & Simoneau, 2009). It should be highlighted that the sensitivity of GC–APCI-MS is better than that of GC–MS, and the background is smaller. It is a good quantitative analysis method, but analytes such as siloxane oligomers present in the detection solvent vary greatly. GC–MS equipment can effectively analyze and process this kind of sample. *2.11. Statistical analysis* All data were presented as the mean ± standard deviation for three independent measurements ($n = 3$). In the case of AFM analysis, six independent measurements ($n = 6$) were performed. Statistically significant differences among samples were evaluated by means of paired one-tailed t-test analysis ($p < 0.05$). Statistical analysis of data was done using SPSS (version 16.0) software.

3. Results and discussion

The main aim of this study was to determine the influence of severe food processing conditions (simulating cooking) on the migration of possible toxic volatile compounds released from SRBMs. The results of the performed analysis are presented below.

3.1. ATR-FTIR analysis

ATR-FTIR was used to test the surface of the following SRBMs: untreated, baked at 175 ◦C, microwaved at 800 W, frozen at □ 18 ◦C. Different times of treatment were checked. Supplementary Material Fig. S1 shows the infrared spectra of the four kinds of SRBMs after the longest time of exposure in each case. After analyzing the results, it can be concluded that the FTIR spectrum was basically unchanged, indicating that the chemical molecular structure of SRBMs was stable even after the application of severe conditions.

In the spectra, the following IR peaks can be seen: 2962 cm□ 1 – the symmetric stretching of the C–H bond within CH3, 1260 cm□ 1 – the asymmetric stretching of the C–H bond within CH3, 1008 cm□ 1 – Si–O–Si symmetric stretching, and 788 cm□ 1 – Si–C symmetric stretching. The obtained results are in accordance with the literature (Chakraborty & Reddy, 2017).

3.2. AFM analysis

AFM was used to obtain a 3D topographic pictures of SRBMs treated at different conditions. They are shown in Fig. 1 Fig. 2 shows the surface micromorphology of SRBMs. The surface of the untreated SRBM (control) was relatively smooth and flat. However, the micromorphology of SRBMs placed in the oven, microwave oven and refrigerator became rougher and had unevenness. Consequently, comparing the root mean square of roughness (Rq) and the average roughness (Ra) the results showed that the Rq and Ra of SRBMs were increased after being used in baking, microwaving, and freezing. This behavior can be related to the fact that under high temperature and microwaving, the SRBMs expanded and released small molecules from the fine pores on the mold surface, making the SRBM rougher. In the case of the SRBM placed in the freezer for a long time, the temperature of SRBM changed from low to high, causing its shrinking and then its expansion. As a result, the formation of fine pores on the mold surface occurred. The obtained value of Ra of SRBM was 0.48 μm, which was similar to the Ra value of commercial SRBM (0.50 μm) reported by Chakraborty and Reddy (2017). To sum up, the surface roughness of SRBMs changed after treatment under severe conditions. These results are in accordance with those that showed that UV radiation increased the surface roughness of SRBMs (Mishra, Shimpi, & Mali, 2013).

3.3. Determination of volatile organic compounds (VOCs) The safety assessment of silicone food contact materials is connected with the determination of the concentration of VOCs (%). Thus, the study of the total VOCs is an effective indicator of the potential mobility of molecules. A limit of 0.5% for the release of VOCs was adopted to distinguish tempered materials and non-tempered materials (Oliveira, Monsalve, Nerin, Padula, & Godoy, 2020). The initial value of total VOCs of SRBM was 2.53% (Fig. 3), which exceeded the limit of 0.5% established in the BfR Recommendations on Food Contact Materials for silicone rubber products. It should be highlighted that the SRBMs used by consumers should be non-tempered material. The results showed that when SRBMs were baked at 175 ◦C for 30–300 min and microwaved at 800 W for 25–50 min, the total VOCs were significantly reduced ($p < 0.05$). This can be attributed to the fact that at high temperatures the swollen SRBM releases volatile substances, leading to their reduction in SRBMs, showing the importance of the tempering process in silicone rubber products. In turn, freezing did not significantly change the total amount of VOCs in SRBM. Nevertheless, it has been shown that the total VOCs increased slightly after freezing of SRBM for 300–600 min. This performance can be explained by the strong adsorption capacity of silicone rubber, which can adsorb the water droplets condensed on its surface. Even if SRBM is dried, there is still a certain adsorption capacity for moisture, which is not easy to remove. In Shanghai city, it was found that VOC concentrations in four different SRBMs analyzed in 2019 were within the range of 1.9–2.4% (JianPing, LinYan, & LiuJie, 2019). The literature showed that for the new SRBMs, the release of total VOCs decreased from 0.44% to 0.14% after about 1700 times reuse (Helling et al., 2012). The results suggested that baking and microwaving could reduce VOCs in SRBM. Moreover, the addition of an appropriate annealing process to the actual production process of SRBMs could effectively reduce the amount of total volatile substances released from the SRBMs. This means that in terms of safety, it is essential that a post-processing step is applied to silicone rubber materials, heating them at over 200 ◦C for several hours prior to launching on the market as the final products.

3.4. Identification of VOCs by GC–MS and analysis GC–APCI-QTOF-MS

There are different migration abilities of silicone oligomers from SRBMs to food simulants. In this experiment, it was found that there is a greater migration ability of silicone oligomers from SRBMs to isooctane than to 95% ethanol and Tenax®, and the most complete profile of silicone oligomers from D4 to D21 was found in the case of isooctane and 95% ethanol (Table 1). Subsequently, the silicone oligomers migrating from SRBM to food simulants were detected by both GC–MS and GC–APCI-QTOF-MS, and the chromatograms are shown in Supplementary Material (Fig. S2). GC–APCI-QTOF-MS can be more accurate to determine the quality of particle fragments, which is conducive for qualitative analysis of substances, while GC–MS can be used for quantitative analyses of batch samples. The chromatograms of methylcyclosiloxanes are characteristic, as they belong to the same family of compounds as their chromatographic peaks appear at the same interval. The mass difference between each compound is 74, corresponding to C2H6OSi. The results show that 18 kinds of silicone oligomers migrated from SRBMs to food simulants with the molecular formula of [Si(CH3)2–O]n (n = 4…21) and molecular weights from 296.6160 to 1557.2340 (Table 1). What is more, the types of silicone oligomers detected during the migration are in accordance with previous research on the composition of silicone molds analyzed by DART and reported by Gross (2015).

3.5. Migration of silicone oligomers

Migration is a mass transfer process in which chemical substances with a low molecular weight are transferred from packaging into food (Gavril et al., 2019; Poças, Oliveira, Pereira, Brandsch, & Hogg, 2011). The migration results of silicone oligomers were quantified after the method validation using the standard of D5 with IS of hexadecane in hexane. The method showed LOD = 0.0039 mg/kg, LOQ = 0.013 mg/kg and $r = 0.9997$. Fig. 4 compares the results of the migration of different SRBMs with a different number of carbons subjected to different conditions (different food simulants and different treatments). High temperature is beneficial to the volatilization of silicone oligomers from SRBMs, which in turn reduces the contents of silicone oligomers in SRBMs. Fig. 4 shows that the migration of the silicone oligomers after baking, microwaving, and freezing of SRBMs was reduced. The highest decrease was observed in the case of the baking process. When the food simulant was isooctane, 18 different silicone oligomers (D4–D21) were detected. The highest migration was obtained in the case of D6, followed by D5, D7, D8, and D4. The results indicated that when SRBM was baked at 175 ◦C for 30 min, the relative migration of D4–D13 to isooctane was significantly ($p < 0.05$) reduced. The relative migration of D4–D17 was significantly ($p < 0.05$) reduced after baking for 150 min, and among them, the D8–D11 decreased until they could not be detected. The relative migration of D4–D18 decreased significantly ($p < 0.05$) during baking for 300 min, when D7–D12 decreased until undetected. In the case of treating the samples in a microwave oven at 800 W for 5 min, the relative migration of D4–D7 decreased significantly ($p < 0.05$). D4–D8 decreased significantly ($p < 0.05$) after 25 min, and D4–D12 decreased significantly ($p < 0.05$) after 50 min. Finally, when the samples were frozen at □ 18 ◦C for 60 min or 300 min, the relative migration of D4 and D5 decreased significantly ($p < 0.05$), while D4–D6 decreased significantly ($p < 0.05$) after the SRBM was frozen for 600 min. All the described results of migration into isooctane show that the severe processing conditions of SRBMs reduced the content of silicone oligomers in the analyzed samples, especially the silicone oligomers with low molecular weight. When 95% ethanol acted as a food simulant, playing the same role as isooctane, 18 different silicone oligomers (D4–D21) were detected. The concentration profiles of migrants were different after severe processing conditions. After baking, the migration amounts of the silicone oligomers in SRBMs dropped rapidly. Specifically, when the SRBM was baked for 300 min, the relative migration of D4–D17 decreased significantly ($p < 0.05$), while D7–D13 decreased to be undetected. However, when the SRBM was microwaved for 50 min, only the relative migration of D4–D7 decreased significantly ($p < 0.05$). When the SRBM was frozen for 600 min, only the relative migration of D4 and D6 decreased significantly ($p < 0.05$). When the food simulant was Tenax®, only 14 types of silicone oligomers (D4–D17) were detected as migrants. The results showed that after baking of SRBMs for 30 min, the relative migration of D4–D12 decreased significantly ($p < 0.05$). After baking for 150 min, D4–D14 decreased significantly ($p < 0.05$), with D6–D12 dropping below the LOD. After baking for 300 min, D4–D15 decreased significantly ($p < 0.05$), while D6–D14 decreased to be undetected. The relative migration of D4–D7 decreased significantly ($p < 0.05$) after microwaving for 50 min and the relative migration of D4–D6 decreased significantly ($p < 0.05$) after freezing for 600 min. Comparing these three food simulants, it was found that silicone oligomers are migrate more easily to isooctane than 95% ethanol, with Tenax® migration being more difficult. Simultaneously, compared to isooctane and 95% ethanol, fewer types of oligomers migrated to Tenax®. This implies that there are different migration capabilities of silicone oligomers in different food simulants, chemical states, and types of food. Extensive study published by Zhang et al. (Zhang, Wong, Begley, Hayward, & Limm, 2012) showed migration of three cyclic silicone oligomers (D4, D5 and D6) to 95% ethanol after 2 h at 40 ◦C. The highest concentration of migrant was obtained in the case of D6 (155 ng/mL) after of 48 h of assay. The experiment was designed to overestimate results of migration assay and simulate worst-case scenarios in realistic situations. Authors strongly highlighted that future studies were necessary to reflect real migration of chemical compounds using food simulants.

3.6. Effect of severe processing conditions on the migration of silicone oligomers

As mentioned before, silicones are not included in the plastic legislation, and silicone oligomers are not present in the EU legislation 10/ 2011 (European Union, 2011) for food contact materials. However, silicones, like any other food contact material, must comply with the frame regulation 1935/2004/CEE (European Parliament and the Council, 2004). In addition, there are some specific legislations for silicones from several European member states. The main principle in most of them is that in addition to some specific compounds, the positive list existing in the plastics regulation https://doi.org//10/2011/EU also applies to silicones. However, none of them includes silicone oligomers as potential migrants. The toxicity of molecules was assigned with toxicity classes (I–III) for chemicals and proposed a maximum daily intake by the TTC decision tree and Cramer classification. According to Cramer rules, the toxicity level of the detected silicone oligomers can be checked and they belong to class III that represents the most toxic substances with a maximum estimated daily intake of 0.09 mg/person/day, i.e., theoretical maximum migration amount is equal to 0.09 mg/kg (Wrona & Nerin, 2019). The obtained results show that silicone oligomers are released from SRBMs and possibly migrate into food. When checking quantitative analysis, all samples exceeded the established SML of 0.01 mg/kg for non-listed substances. Even if the theoretical maximum migration limit established by Cramer rules for Class III substances is considered (that is nine times higher than SML), all analyzed samples do not comply with current legislation EU 10/2011 for plastics in contact with food. After analyzing the results (Supplementary Material – Fig. S3) it can be concluded that the highest migration rate of silicone oligomers can be observed in the case of isooctane, then 95% ethanol and then Tenax®. In all simulants, the migration of both low and high molecular weight silicone oligomers decreased drastically during the baking process, and their migration decreased to half of their initial value after microwaving for 50 min, but their least effective migration was in the freezing process. The migrated concentration of silicone oligomers was 2670 mg/kg in isooctane including 70.7% of silicone oligomers with molecular weight less than 1000, 884 mg/kg in 95% ethanol with 91.8% of silicone oligomers having molecular weight less than 1000, 284 mg/ kg in Tenax® including 97.2% of silicone oligomers with a molecular weight less than 1000. It should be highlighted that SRBM swelled in isooctane as was also observed by Zhang et al. (2012). The migration of siloxane oligomers in isooctane is the highest because swelling promotes migration. The amount of migration of siloxane oligomers can represent the use of silicone rubber molds in the most severe use environments, which has a certain reference significance. At the same time, the purpose of this study was to compare the migration of siloxane oligomers in different food simulants without calculating the diffusion coefficient. Therefore, isooctane was chosen as one of the food simulants. In the case of the SRBMs for the longest baking time (300 min), microwaving time (50 min), and freezing time (600 min), the migration of silicone oligomers with lower than 1000 Da to food simulants was reduced. Within these three methods used, the baking is the most efficient one to reduce them, as Table 2 shows. However, under microwaving or freezing, the migration of silicone oligomers with a molecular weight higher than 1000 Da to 95% ethanol and Tenax® were increased. Also, it is possible to change the microstructure of the SRBM after microwaving or freezing, which increased its surface roughness, and then increased the ability of silicone oligomers with a molecular weight higher than 1000 to migrate. To sum up, the surface structure of the silicone rubber baking mold changes during use, and the migration of siloxane oligomers decreases as the use time increases. Simulants such as 95% ethanol and isooctane are used to substitute vegetable oil containing<1% unsaponifiable matter (simulant D2) as they are easier to analyze. Application of oil in contact with silicone rubber also cause swelling and high release of silicone oligomers (Y. Q. Liu et al., 2020). There is advice against high-fat food preparation in silicone molds due to high migration (Helling, Mieth, et al., 2009). Moreover, some investigations showed that after multiple usage silicone molds tend to absorb fat which cannot be removed by washing. However, it was sweated out after baking cycles. Fat incorporated into silicone molds can be easily oxidized and causes an unpleasant smell (Helling, Kutschbach, & Simat, 2009).

4. Conclusion

In this study, under different conditions of usage including baking at 175 ◦C, microwaving at 800 W, and freezing at □ 18 ◦C, the migration (70 ◦C for 2 h) of silicone oligomers from SRBMs to food simulants (isooctane, 95% ethanol, and Tenax®) was determined. As a result, 18 kinds of silicone oligomers were identified by GC–MS and GC–APCI-QTOF-MS. Migration tests indicated that isooctane allowed the highest migration for silicone oligomers from SRBM, 95% then ethanol and finally Tenax®. After analyzing the quantitative results, it can be concluded that migration into isooctane, 95% ethanol, and Tenax®, of silicone oligomers with a molecular weight lower than 1000 Da was 70.7%, 91.8%, and 97.2% respectively of all silicone oligomers. The baking treatment is the most effective way to reduce the contents of silicone oligomers in SRBMs, while 95% ethanol acts like the second, and Tenax® is the third. It should be highlighted that during migration into isooctane, swelling of samples was observed. Under the severe processing conditions, the chemical structure of SRBMs was unchanged but the roughness of the surface of SRBMs was increased. Besides, the initial VOCs value was 2.53% higher than those that were recommended by BfR Recommendations on Food Contact Materials. Therefore, it could be considered that SRBMs studied were made of non-tempered materials. It should be highlighted that baking treatment significantly ($p < 0.05$) reduced the VOCs value in SRBMs and microwaving treatment had the same tendency as baking treatment, but was less effective, while freezing had no effect on VOC value. Furthermore, qualitative results of migration have shown that all analyzed samples do not comply with current legislation EU 10/2011 for plastics in contact with food, nor the BfR recommendation. The results obtained above emphasize the importance of the tempering process for silicone rubber products. Non-tempered SRBMs contains numerous silicone oligomers, especially silicone oligomers with a molecular weight<1000 Da which are potentially toxic. Unfortunately, during the use of the SRBMs, silicone oligomers easily migrate to food simulants and food, which can pose a health risk for consumers and could affect food properties such as odor and taste. Ç

CRediT authorship contribution statement **Yi-Qi Liu:** Methodology, Investigation, Conceptualization, Validation, Formal analysis, Data curation, Writing - original draft, Writing - review & editing. **Magdalena Wrona:** Methodology, Conceptualization, Validation, Data curation, Writing - review & editing. **Qi-Zhi Su:** Methodology, Conceptualization, Investigation, Data curation. **Paula Vera:** Methodology, Conceptualization, Writing - review & editing. **Cristina Nerín:** Conceptualization, Resources, Writing - review & editing, Supervision, Project administration, Funding acquisition. **Chang-Ying Hu:** Conceptualization, Resources, Supervision, Project administration, Funding acquisition.

Acknowledgments This work was supported by the National Key R&D Program of China (Grant No. 2018YFC1603205/2018YFC1603200), Key R&D Program of Guangdong province, China (Grant No. 2019B020212002), National food safety standard establishment plan of China (Grant No. spaq-2019- 013), and Jinan University's first batch of short-term overseas research projects for graduate students in 2019. The authors from the GUIA group also acknowledge the funding provided by Gobierno de Arag´on (Spain), FEDER – Fondo Europeo de Desarrollo Regional – funds given to GUIA research group: T53_20R and RTI2018-097805-B-I00 project obtained from Ministerio de Ciencia e Innovaci´on, Spain.

References

Akbar, M., Ullah, R., & Alam, S. (2019). Aging of silicone rubber-based composite insulators under multi-stressed conditions: an overview. *Materials Research Express*. https://doi.org/10.1088/2053-1591/ab3f0d.

BfR-German Federal Institute for Risk. BfR Recommendations on Food Contact Materials XIV. Polymer Dispersions. , (2019).

Bratinova, S., Raffael, B., & Simoneau, C. (2009). *Guidelines for performance criteria and validation procedures of analytical methods used in controls of food contact materials*. In A CRL-NRL-FCM Publication.

Burns-Naas, L. A., Meeks, R. G., Kolesar, G. B., Mast, R. W., Elwell, M. R., Hardisty, J. F., & Thevenaz, P. (2002). Inhalation toxicology of octamethylcyclotetrasiloxane (D4) following a 3-month nose-only exposure in Fischer 344 rats. *International Journal of Toxicology*. https://doi.org/10.1080/10915810252826000.

Chakraborty, R., & Reddy, S. B. (2017). Performance of silicone rubber insulators under thermal and electrical stress. *IEEE Transactions on Industry Applications., 53*(3), 2446–2454.

Council of Europe. Resolution ResAP(2004)2 on cork stoppers and other cork materials and articles intended to come into contact with foodstuffs. , (2004).

Deleu, L. J., Luyts, A., Wilderjans, E., Van Haesendonck, I., Brijs, K., & Delcour, J. A. (2019). Ohmic versus conventional heating for studying molecular changes during pound cake baking. *Journal of Cereal Science, 89*, 102708. https://doi.org/10.1016/j.jcs.2019.01.008.

European Parliament and the Council. Regulation (EC) No 1935/2004 of the European Parliament and of the Council of 27 October 2004 on materials and articles intended to come into contact with food and repealing Directives 80/590/EEC and 89/109/ EEC. , (2004).

Flores-Ch´avez, H., & Monta˜nez-Saenz, J. (n.d.). Baking Process in Oven and Microwave-oven in Sourdough Enriched with Chickpeas and Dietary Fiber of Prickly pear and Oats. International Journal of Engineering Research and Applications, Vol. 4, pp. 300–305. Retrieved from https://www.ingentaconnect.com/content/doaj/22489622/2014/00000004/00000004/art00224.

Gavril, G. L., Wrona, M., Bertella, A., ´Swieca, M., Rˆap˘a, M., Salafranca, J., & Nerín, C. (2019). Influence of medicinal and aromatic plants into risk assessment of a new bioactive packaging based on polylactic acid (PLA). *Food and Chemical Toxicology, 132*, 1–11. https://doi.org/10.1016/j.fct.2019.110662. GB 28482-2012. Safety requirements of soothers for babies and young children. , (2012).

Gross, J. H. (2015a). Polydimethylsiloxane Extraction from Silicone Rubber intO Baked Goods Detected by Direct Analysis in Real-Time Mass Spectrometry. *European Journal of Mass Spectrometry, 21*(3), 313–319.

Gross, J. H. (2015b). Analysis of Silicones Released from Household Items and Baby Articles by Direct Analysis in Real Time-Mass Spectrometry. *Journal of the American Society for Mass Spectrometry, 26*(3), 511–521.

Helling, R., Kutschbach, K., & Simat, T. (2009). Migration behaviour of silicone moulds in contact with different foodstuffs. *Food Additives & Contaminants. Part A, Chemistry, Analysis, Control, Exposure & Risk Assessment, 27*, 396–405. https://doi.org/10.1080/19440040903341869.

Helling, R., Mieth, A., Altmann, S., & Simat, T. J. (2009). Determination of the overall migration from silicone baking moulds into simulants and food using 1 H-NMR techniques. *Food Additives & Contaminants: Part A, 26*(3), 395–407.

Helling, R., Seifried, P., Fritzsche, D., & Simat, T. J. (2012). Characterisation and migration properties of silicone materials during typical long-term commercial and household use applications: A combined case study. *Food Additives & Contaminants: Part A, 29*(9), 1489–1500.

JianPing, Y., LinYan, Y., & LiuJie, S. (2019). Investigation on volatile matter content in silicone rubber products for baking in Shanghai circulation market. *Journal of Food Safety and Quality, 10*(21), 7168–7171.

Liu, Y., Hu, C., Shang, G., Zhang, H., Zeng, Y., Zhu, L., & Zhong, H. (2019). Detection method and safety evaluation of three kinds of cyclosiloxanes in cake trans-migrated from silicone rubber mold. Food and Fermentation Industries, 45(18), 228–233. 10.13995/j.cnki.11-1802/ts.021013.

Liu, Y.-Q., Yu, W.-W., Jiang, H., Shang, G.-Q., Zeng, S.-F., Wang, Z.-W., & Hu, C.-Y. (2020). Variation of baking oils and baking methods on altering the contents of cyclosiloxane in food simulants and cakes migrated from silicone rubber baking moulds. *Food Packaging and Shelf Life, 24*, 100505. https://doi.org/10.1016/j. fpsl.2020.100505.

Ministerio de Sanidad Política Social e Igualdad. Real Decreto 847/2011 por el que se establece la lista positiva de sustancias permitidas para la fabricaci´on de materiales polim´ericos destinados a entrar en contacto con los alimentos. , (2011).

Mishra, S., Shimpi, N. G., & Mali, A. D. (2013). Effect of surface modified montmorillonite on photo-oxidative degradation of silicone rubber composites. *Macromolecular Research, 21*(5), 466–473.

Mojsiewicz-Pienkowska, K., Jamr´ogiewicz, M., Szymkowska, K., & Krenczkowska, D. (2016). Direct human contact with siloxanes (silicones) - safety or risk part 1. Characteristics of siloxanes (silicones). Frontiers in Pharmacology. 10.3389/fphar.2016.00132.

Mojsiewicz-Pie´nkowska, K., & Krenczkowska, D. (2018). Evolution of consciousness of exposure to siloxanes—review of publications. *Chemosphere, 191*, 204–217.

Oliveira, W.d. S., Monsalve, J. O., Nerin, C., Padula, M., & Godoy, H. T. (2020). Characterization of odorants from baby bottles by headspace solid phase microextraction coupled to gas chromatography-olfactometry-mass spectrometry. *Talanta, 207*, 120301. https://doi.org/10.1016/j.talanta.2019.120301.

Onnekink, C., Kappel, R. M., Boelens, W. C., & Pruijn, G. J. M. (2020). Low molecular weight silicones induce cell death in cultured cells. *Scientific Reports, 10*(1), 9558. https://doi.org/10.1038/s41598-020-66666-7.

Poças, M.d. F., Oliveira, J. C., Pereira, J. R., Brandsch, R., & Hogg, T. (2011). Modelling migration from paper into a food simulant. *Food Control, 22*(2), 303–312. Rücker, C., & Kümmerer, K. (2015). Environmental Chemistry of Organosiloxanes. *Chemical Reviews, 115*(1), 466–524.

Union Europea. Commission Regulation (EU) No 10/2011 of 14 January 2011 on plastic materials and articles intended to come into contact with food. , (2011).

Wrona, M., & Nerin, C. (2019). CHAPTER 7: Risk Assessment of Plastic Packaging for Food Applications. In Food Chemistry, Function and Analysis (Vol. 2019-Janua). 10.1039/9781788012973-00163.

Wrona, Magdalena, & Nerín, C. (2020). Analytical approaches for analysis of safety of modern food packaging: A review. Molecules. 10.3390/molecules25030752.

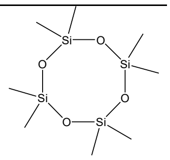
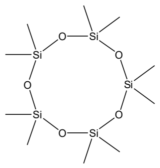
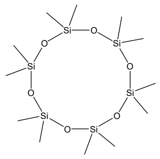
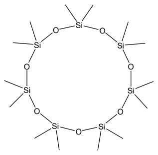
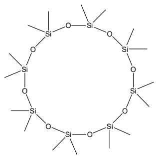
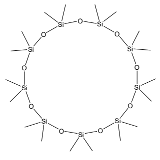
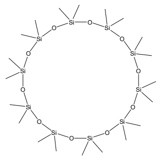
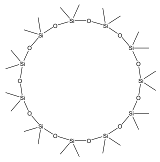
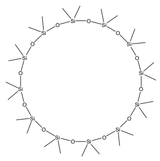
Yolacaner, E. T., Sumnu, G., & Sahin, S. (2017). In *The Microwave Processing of Foods* (pp. 117–141). Elsevier. https://doi.org/10.1016/B978-0-08-100528-6.00006-1.

Zhang, K., Wong, J. W., Begley, T. H., Hayward, D. G., & Limm, W. (2012). Determination of siloxanes in silicone products and potential migration to milk, formula and liquid simulants. *Food Additives & Contaminants: Part A, 29*(8), 1311–1321.

Zhu, Y. (2019). Influence of corona discharge on hydrophobicity of silicone rubber used for outdoor insulation. *Polymer Testing, 74*, 14–20.

Tables and Figures:

Table 1: Volatile compounds identified in SRBMs by GC–MS and GC–APCI-QTOF-MS after migration assays.

No	tR (min)	Name	Abbreviation	Degree of polymerization	Molecular formula	Molecular mass	CAS	Structure
1	6.934	Octamethylcyclotetrasiloxane	D4	4	$C_8H_{24}O_4Si_4$	296.6160	556-67-2	
2	8.255	Decamethylcyclopentasiloxane	D5	5	$C_{10}H_{30}O_5Si_5$	370.7700	541-02-6	
3	9.526	Dodecamethylcyclohexasiloxane	D6	6	$C_{12}H_{36}O_6Si_6$	444.9240	540-97-6	
4	10.635	Tetradecamethylcycloheptasiloxane	D7	7	$C_{14}H_{42}O_7Si_7$	519.0780	107-50-6	
5	11.602	Hexadecamethylcyclooctasiloxane	D8	8	$C_{16}H_{48}O_8Si_8$	593.2320	556–68-3	
6	12.441	Octadecamethylcyclononasiloxane	D9	9	$C_{18}H_{54}O_9Si_9$	667.3860	556–71-8	
7	13.192	Eicosamethylcyclodecasiloxane	D10	10	$C_{20}H_{60}O_{10}Si_{10}$	741.5400	18772-36-6	
8	13.875	Docosamethylcycloundecasiloxane	D11	11	$C_{22}H_{66}O_{11}Si_{11}$	815.6940	18766-38-6	
9	14.480	Tetracosamethylcyclododecasiloxane	D12	12	$C_{24}H_{72}O_{12}Si_{12}$	889.8480	18919-94-3	

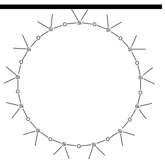
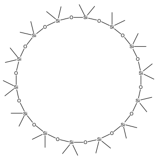
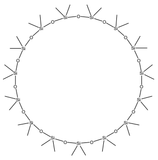
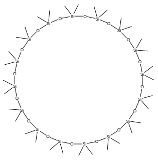
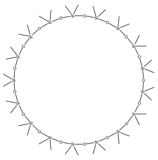
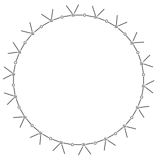
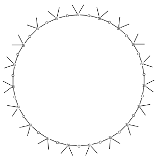
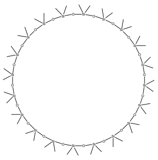
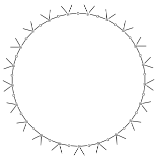
10	15.076	Hexacosamethylcyclotridecasiloxane	D13	13	$C_{26}H_{78}O_{13}Si_{13}$	964.0020	23732-94-7	
11	15.317	Octacosamethylcyclotetradecasiloxane	D14	14	$C_{28}H_{84}O_{14}Si_{14}$	1038.1560	149050-40-8	
12	15.618	Triacontamethylcyclopentadecasiloxane	D15	15	$C_{30}H_{90}O_{15}Si_{15}$	1112.3100	23523-14-0	
13	16.130	Dotriacontamethylcyclohexadecasiloxane	D16	16	$C_{32}H_{96}O_{16}Si_{16}$	1186.4640	150026-95-2	
14	16.687	Tetratriacontamethylcycloheptadecasiloxane	D17	17	$C_{34}H_{102}O_{17}Si_{17}$	1260.6180	150026-96-3	
15	17.274	Hexatriacontamethylcyclooctadecasiloxane	D18	18	$C_{36}H_{108}O_{18}Si_{18}$	1334.7720	23523–12-8	
16	17.967	Octatriacontamethylcyclononadecasiloxane	D19	19	$C_{38}H_{114}O_{19}Si_{19}$	1408.9260	150026-97-4	
17	18.750	Tetracontamethylcycloicosasiloxane	D20	20	$C_{40}H_{120}O_{20}Si_{20}$	1483.0800	150026-98-5	
18	19.728	Dotetracontamethylcyclohenicosasiloxane	D21	21	$C_{42}H_{126}O_{21}Si_{21}$	1557.2340	23523–13-9	

Table 2. Changes in the migration of silicone oligomers to food simulants in the longest used SRBMs (compared with the control group).

Food simulants	**Usage**	**All**	**Silicone oligomers**	
			molecular weight higher than 1000	**molecular weight lower than 1000**
Isooctane	Baking	13.5%	44.6%	0.6%
	Microwaving	52.4%	78.9%	41.5%
	Freezing	74.0%	80.2%	71.4%
95% Ethanol	Baking	4.0%	42.8%	0.6%
	Microwaving	56.5%	136.1%	49.4%
	Freezing	74.2%	99.2%	72.0%
Tenax®	Baking	1.4%	42.0%	0.2%
	Microwaving	46.2%	168.7%	42.6%
	Freezing	80.5%	171.3%	77.9%

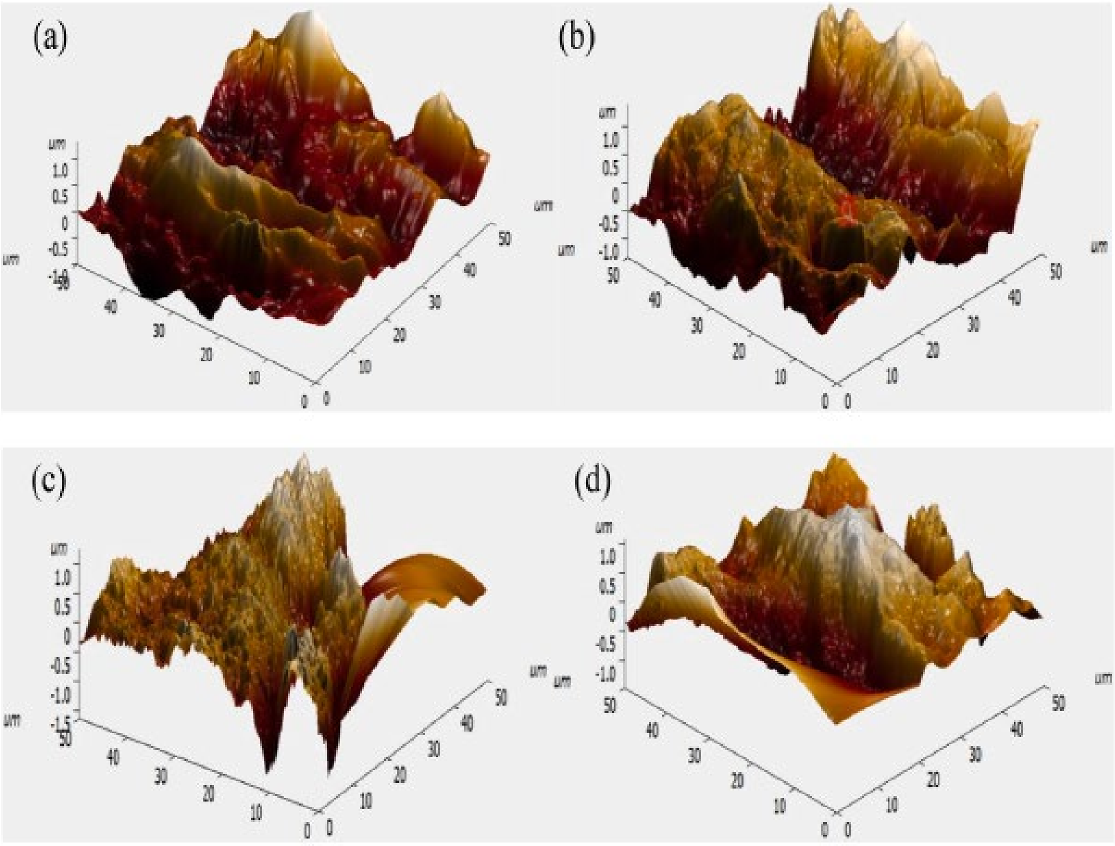


Fig. 1. The 3D morphology images of SRBMs acquired by AFM; (a): the control mold; (b): the mold baked at 175 ◦C for 300 min; (c): the mold microwaved at 800 W for 50 min; (d): the mold frozen at □ 18 ◦C for 600 min.

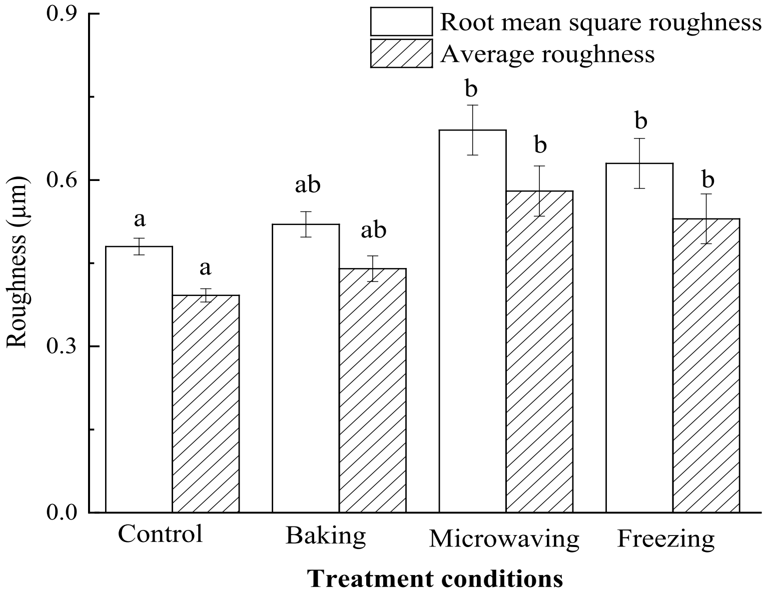


Fig. 2. The roughness of SRBMs under different treatments: control sample; baking at 175 ◦C for 300 min; microwaving at 800 W for 50 min and freezing at □ 18 ◦C for 600 min.

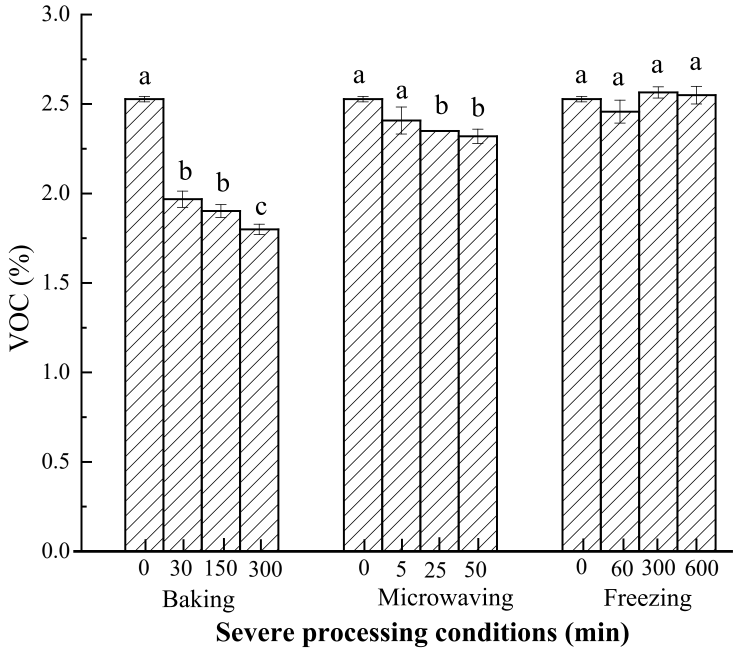


Fig. 3. The amount of initial volatile organic compounds in SRBMs after different treatments, with the processing time of 0 min as control.

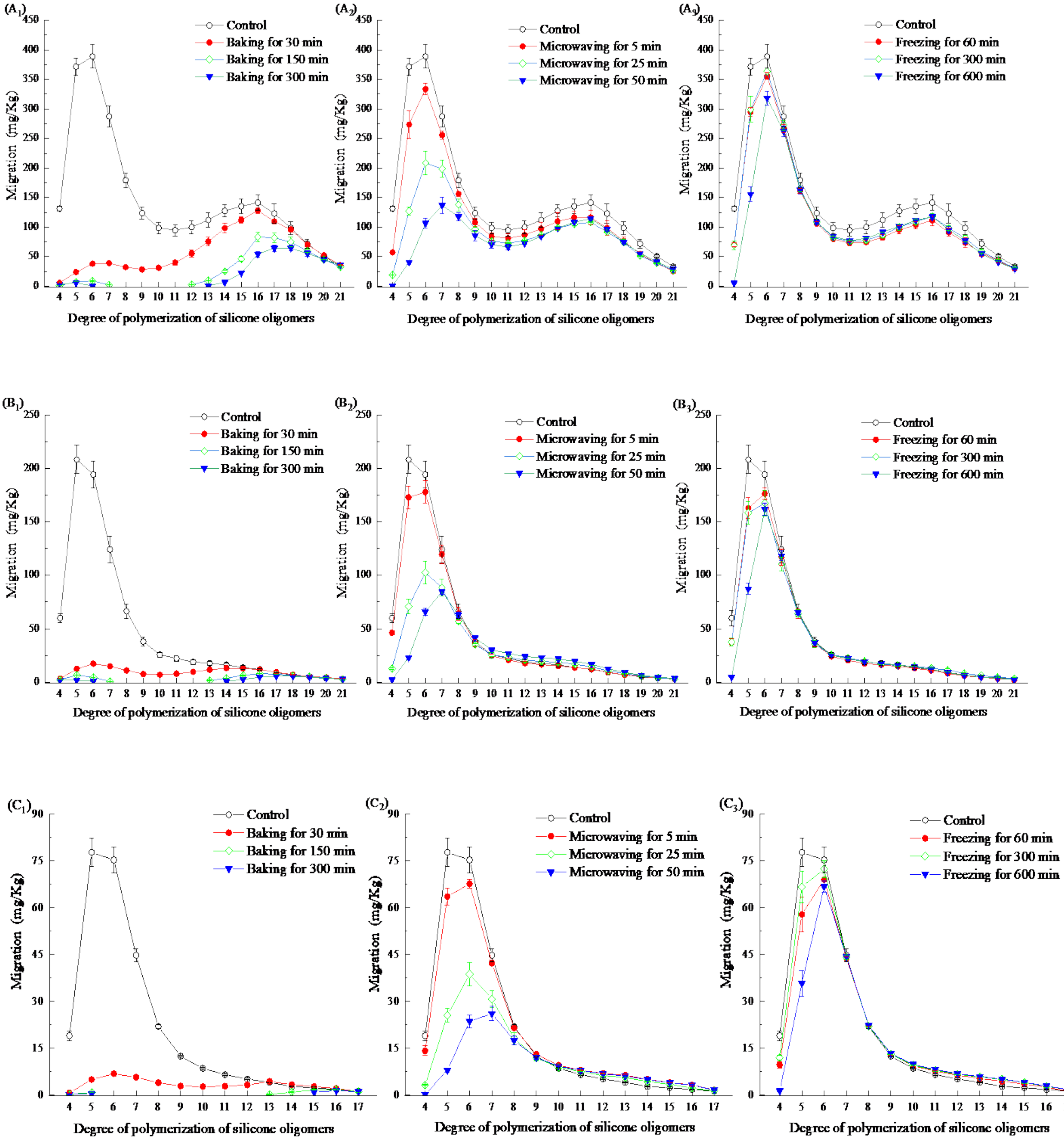


Fig. 4. Migration of silicone oligomers with different degrees of polymerization from different SRBMs to (A1-3) isooctane, (B1-3) 95% ethanol, (C1-3) Tenax®, while 1 represented the baked SRBMs, 2 represented the microwaved SRBMs, 3 represented the frozen SRBMs. Absence of point indicates result below the detection limit.