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Design of Roll-In Margarine Analogous by Partial Drying of Monoglyceride-Structured Emulsions

(Article begins on next page)

14 April 2025

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Journal:	*European Journal of Lipid Science and Technology*
Manuscript ID	Draft
Wiley - Manuscript type:	Research Article
Date Submitted by the Author:	n/a
Complete List of Authors:	Calligaris, Sonia; University of Udine, Department of Agricultural, Food, Environmental and Animal Sciences Plazzotta, Stella; University of Udine, Department of Agricultural, Food, Environmental and Animal Sciences Barba, Luisa; Institute of Crystallography National Research Council Trieste Branch Manzocco, Lara; University of Udine, Department of Agricultural, Food, Environmental and Animal Sciences
Keywords:	fat substitution, monoglyceride, rheological properties, laminated products, puff pastry
Additional Keywords (select from list):	

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Graphical Abstract

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Monoglyceride-structured emulsion

- 47.6% water
- Soft

Partial gentle drying

Partially-dried emulsion

- 20% water
- Plastic and laminable

Baking trials

Puff pastry

- Comparable to roll-in-margarine puff pastry

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Legend Graphical Abstract

The application of a gentle drying procedure at 30 °C was used to turn soft monoglyceride-structured O/W emulsions into plastic and laminable systems. The latter showed a water content and rheological properties analogous to those of a conventional roll-in margarine and similar performances in baking trials of puff pastry.

1 **Title: Design of roll-in margarine analogous by partial drying of monoglyceride-structured**

2 **emulsions**

3 **Running Title: Novel monoglyceride-emulsions for margarine replacement**

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9 **Abstract**

10 A novel process is proposed to turn soft gelled emulsions containing 4 or 9 g/100 g saturated

11 monoglyceride (MG4 and MG9) and 52.4 g/100 g water into a fat material with the typical water content

12 (20 g/100 g) and rheological properties of roll-in margarines. A gentle drying procedure at 30 °C was

13 found to allow partial water removal from the emulsions, without causing phase separation. As compared

14 to the initial emulsions, the partially dried systems (MG4-dried and MG9-dried) showed a more solid-

15 like appearance and a microstructure with oil droplets tightly packed into the MG-network. Calorimetric

16 (DSC) and crystal organization (XRD) analyses showed that drying modified neither the amount of

17 crystallized material nor induced MG transition toward a different lamellar organization. Moreover, the

18 MG9-dried emulsion showed rheological properties (G' = 4.6×10^5 Pa, critical stress = 634 Pa)

19 comparable to those of commercial roll-in margarine and was successfully used in baking trials of puff

20 pastry.

21 **Practical applications**: The last years have seen an increasing demand of strategies for the nutritional

22 improvement of lipid-containing foods, due to the emerging health concern related to the consumption

23 of high quantities of saturated and trans fatty acids. However, the unique technological and sensory

24 properties of fats make this task very challenging, especially considering the production of laminated

25 products requiring the use of plastic fat materials with peculiar rheological properties. The results

26 obtained in this study are very promising, as they show that the MG9-dried fat can be used as a trans-

27 free alternative to traditional laminating margarine.

28 Key-words: fat substitution/monoglyceride/rheological properties/laminated products/puff pastry

29 Abbreviations: MG, monoglycerides

1 Introduction

31 Food fats, including animal fats (e.g. butter and lard), tropical oils (e.g. palm oil, palm kernel oil and

32 coconut oil) as well as margarine and shortenings containing them, are multi-purpose ingredients used

33 to modulate food structure (aeration, lightness), rheological properties (plasticity, texture), and sensory

34 characteristics (taste, colour, flavour, crispiness, creaminess) [1]. They have the common characteristic

35 to be spreadable at room temperature thanks to the presence of a fat crystal network made of

36 triacylglycerols [2]. In this context, saturated and trans fatty acids play a determining role being

37 crystalline at room temperature. Despite unique fat technological functionalities, the last years have seen

38 an increasing demand for strategies for the nutritional improvement of lipid-containing foods. This is due

39 to the emerging health concern related to the consumption of high quantities of saturated and trans fats.

40 The latter are well known to be associated with an increased risk of diet-related diseases and pathological

41 conditions, including cardiovascular diseases, obesity and type II diabetes [3,4]. The World Health

42 Organization (WHO) recommends the amount of total, saturated and trans fats to be less than 30% and

43   10% of the total energy intake, respectively [5]. At the same time, the European Commission in 2019

44   imposed a limitation of trans fats to 2 g per 100 g of total fats [6].

45   In this context, the most challenging fats to be substituted are those used to produce laminated products,

46   such as puff pastry and croissants, called roll-in fats or laminating fats [7]. They mainly include

47   margarines that are water-in-oil emulsions containing at least 80 g/100 g fat, of which more than 50% is

48   represented by saturated and/or trans fatty acids [8,9]. Upon lamination, these roll-in-fats are shaped into

49   several thin sheets which inter-lie among an equal number of dough layers. Laminated products owe their

50   peculiar physical and sensory properties to this laminated structure: during baking, in fact, dough water

51   evaporates, causing the expansion of the dough layers, which bake separately, due to the alternate fat

52   layers [10]. Roll-in-fats should present specific characteristics, as described by Blake and Marangoni

53   [11]. They need to be plastic enough to withstand the lamination process without losing continuity. To

54   this aim, their rheological properties should be similar to the dough ones and their melting temperature

55   higher than that of the lamination process, to avoid fat melting and prevent dough layers from coming in

56   contact. Moreover, it should be underlined that the presence of water in margarine is required for optimal

57   lubrication, plasticity and leavening [12]. Water is entrapped in the continuous matrix of fat crystals as

58   tiny droplets, presenting a diameter of about 1 μm, which accounts for $5\text{-}10\times10^9$ droplets per mL of

59   product [13]. Although finely dispersed, water droplets are combined loosely enough for the emulsion to

60   break easily upon melting of the crystalline continuous phase, which favours puff during product baking

61   [14].

62   Different Authors proposed roll-in-fat mimetics with low saturated fat content [15–20]. However, these

63   strategies resulted in fat materials which are not comparable to margarines, since not containing water

64   [21].

65   An alternative strategy could be based on the use of MG-structured emulsions, which have actually

66   shown good performances as fat substitutes in different baked goods, including short dough pastry [22],

67 sweet bread [23], cookies [24]; ice-cream [25]. The use of MG is not new and ternary gelled systems

68 containing saturated monoglycerides where firstly proposed by Marangoni and co-workers in 2007 [26–

69 28]. At high water content, the system appears as a liquid emulsion in which MG crystalline bilayers

70 surround and stabilize oil droplets. Increasing the MG content, the MG walls extend from one droplet to

71 the next one, leading to a continuous solid network, which generates a gel-like material with the

72 rheological features typical of spreadable fats. The final gel structure is affected by several compositional

73 and processing factors, such as MG and other ingredient type and concentration, pH, temperature and

74 application of shear. In any case, it exists a defined range of compositional variables in which the system

75 equilibrium is not assured and phase separation could occur [27,29,30]. It should be noted that it is not

76 possible to prepare systems simulating margarine rheological properties and fat/water ratio by simply

77 increasing the MG content. In fact, the relative increase in MG requires more water and a concomitant

78 decrease in the oil content to form a water-continuous phase in which the MG interact giving a stable gel

79 [27,28]. In this regard, Blake and Marangoni [11] affirm that to ensure proper hydration and sufficient

80 oil-droplet encapsulation, the water content of these emulsions must not be lower than 30 g/100 g. To

81 improve the rheological properties of MG emulsions, waxes were added but the final water content (35-

82 40 g/100 g) was in any case higher than that expected in margarine [11].

83 Based on these considerations, in this work, a novel process is proposed to turn soft MG gelled emulsions

84 into fat materials with the typical water content (20 g/100 g) and rheological properties of roll-in

85 margarines. In particular, we propose to apply a gentle drying to remove part of the water from the

86 aqueous network surrounding oil droplets in MG emulsions, without disrupting their ternary structure.

87 The partially dried structured emulsions were analyzed for microstructure, calorimetric (DSC), crystal

88 organization (XRD) and rheological properties. Finally, the best performing sample was selected and

89 used in baking trials of puff pastry.

90 ## 2 Materials and Methods

91 *2.1   Materials*

92 Myverol™ saturated monoglyceride (MG) were kindly supplied by Kerry Ingredients and Flavour

93 (Bristol, UK), palmitic, stearic acid, NaOH were purchased from Sigma Aldrich (Milan, Italy),

94 ingredients for puff pastry were purchased in a local market; commercial margarines were selected

95 among those typically used for the industrial preparation of laminated baked goods and kindly provided

96 by local food companies.

97 *2.2   Structured emulsion preparation*

98 MG structured emulsions were prepared according to Calligaris, Da Pieve, Arrighetti, and Barba [31].

99 Briefly, the oil phase was composed of MG (4 and 9 g/100 g), a co-surfactant mixture (0.8 and 1.8 g/100

100 g) of palmitic and stearic acid (weight ratio 1:1) and sunflower oil (47.6 and 41.6 g/100 g). The water

101 phase (52.4 g/100 g) was composed of 1 mM NaOH (pH 11) in deionized milli-Q water to promote the

102 partial ionization of the co-surfactant mixture and obtain a properly swollen phase. The samples were

103 prepared by mixing the water solution and the oil phase previously heated at 78 °C in a water bath and

104 homogenizing by using a high-speed homogenizer DI 25 (Ika-Werke, Staufen, Germany) at 59,000g for

105 1 min. Finally, the mixture was cooled at 4 °C in an ice bath and then stored at 4 °C for 24 h before usage.

106 The obtained structured emulsions were called MG4 and MG9.

107 *2.3   Structured emulsion drying*

108 The drying process was conducted according to the method patented by our research group [32]. The

109 structured emulsions were spread in a thin layer (3.0 ± 0.5 mm), as measured by a CD-15APXR digital

110 calliper (Absolute AOS Digimatic, Mitutoyo Corporation, Kanagawa, Japan) and dried at 30, 40 and 50

111 °C using a professional oven (Air-o-steam, 10 GN 1/1, Electrolux, Pordenone, Italy), with an airflow set

112 at the medium level. The drying process was continued until the structured emulsions reached a lipid

113 phase weight concentration of 80 g/100 g, which was determined by weighting the samples at defined

114 time intervals. The partially dried structured emulsions were called MG4-dried and MG9-dried.

2.4   Puff pastry preparation

116 Puff pastry was prepared using commercial laminating palm margarine (Control) and the MG9-dried

117 sample (36.9 g/100 g). Plain flour (47.7 g/100 g) was mixed with fine salt (0.7 g/100 g) and water (14.7

118 g/100 g) using a domestic mixer (Kenwood Chef KVC3110S, Milan, Italy) until obtaining homogeneous

119 dough (250 g). The latter was placed on a paper-lined sheet pan dusted with flour, covered, and left to

120 rest for 30 min at 4 °C. Then, the dough was rolled into a square of 45×45 cm. Similarly, the commercial

121 margarine and the MG9-dried sample were shaped into squares of about 31×31 cm, and puff pastry was

122 produced as described by Patient [12], with 2 h rest at 4 °C between subsequent folding steps. Puff pastry

123 was finally rolled up to 3-mm thickness and cut into 4×5 cm rectangles, which were baked at 180 °C for

124 9 min (Air-o-steam, 10 GN 1/1, Electrolux, Pordenone, Italy) and cooled at 20 °C before analyses.

2.5   Analytical determinations

2.5.1   Image acquisition

127 Sample images were acquired using an image acquisition cabinet (Immagini & Computer, Bareggio,

128 Italy) equipped with a digital camera (EOS 550D, Canon, Milano, Italy). The digital camera was placed

129 on an adjustable stand positioned 45 cm above a black or white cardboard base where the samples were

130 placed. The light was provided by 4 100 W frosted photographic floodlights, in a position allowing

131 minimum shadow and glare. Images were saved in jpeg format resulting in 3456×2304 pixels.

2.5.2   Optical microscopy

133 Samples were placed on a glass slide, covered with a cover slide and observed using a Leica DM 2000

134 optical microscope (Leica Microsystems, Heerbrugg, Switzerland). The images were taken at 200×

135 magnification using a Leica EC3 digital camera and elaborated with the Leica Suite Las EZ software

136 (Leica Microsystems, Heerbrugg, Switzerland).

2.5.3 Differential scanning calorimetry

138 The calorimetric analysis was carried out using a TA4000 differential scanning calorimeter (Mettler-

139 Toledo, Greifensee, Swiss) connected to a GraphWare software TAT72.2/5 (Mettler-Toledo, Greifensee,

140 Swiss). Heat flow calibration was achieved using indium (heat of fusion 28.45 J/g). Temperature

141 calibration was carried out using hexane (m.p. –93.5 °C), water (m.p. 0.0 °C) and indium (m.p. 156.6

142 °C). Samples were prepared by carefully weighing 15–20 mg of the sample in 160 μL aluminium DSC

143 pans, closed with hermetic sealing. An empty pan was used as a reference. Samples were heated under

144 nitrogen flow (0.5 mL/min) from 20 to 80 °C at 5 °C/min. The start of the melting transition was taken

145 as on-set (T_{on}) points of transition, that are the points at which the extrapolated baseline intersects the

146 extrapolated tangent of the calorimetric peak in the transition state. Results were normalized to account

147 for the weight variation of the samples. Total peak enthalpy was obtained by integration. The program

148 STAR ever. 8.10 (Mettler-Toledo, Greifensee, Swiss) was used to plot and analyze the thermal data.

2.5.4 X-ray diffraction

150 X-ray diffraction patterns were recorded at the X-ray Diffraction beamline 5.2 at the Synchrotron

151 Radiation Facility Elettra located in Trieste (Italy). The X-ray beam emitted by the wiggler source on the

152 Elettra 2 GeV electron storage ring was monochromatized by a Si(1 1 1) double crystal monochromator,

153 focused on the sample and collimated by a double set of slits giving a spot size of 0.2×0.2 mm. A drop

154 of the sample was lodged into a nylon pre-mounted cryoloop 20 micron for crystallographic experiments

155 (0.7–1.0 mm) (Hampton Research HR4-965, Aliso Veijo, CA, USA). Sample temperature was controlled

156 using a 700 series cryo-cooler (Oxford Cryosystems, Oxford, UK) with an accuracy of 1 °C. Analyses

157 were performed at 20 °C. Data were collected at a photon energy of 8.856 keV (λ = 1.4 Å), using a 2M

158 Pilatus silicon pixel X-ray detector (DECTRIS Ltd., Baden, Switzerland). Bidimensional patterns

159 collected with Pilatus were calibrated by means of a LaB6 standard and integrated using the software

160 FIT2D [33] obtaining a powder-like pattern for every sample experiment. The angular range (two theta)

161 of observed diffraction ring spanned from 1.3° to 21°, corresponding to interplanar distances d from 60

162 Å to 3.9 Å and to reciprocal lattice spacings q from 0.1 Å^{-1} to 1.62 Å^{-1}.

2.5.5 Rheology

164 The viscoelastic properties (moduli G' and G'') of the samples were tested using an RS6000 Rheometer

165 (Thermo Scientific RheoStress, Haake, Germany), equipped with a Peltier system for temperature

166 control. Measures were performed using a parallel plate geometry at 15 °C with a gap of 1.0 mm.

167 Oscillatory sweep tests to identify the linear viscoelastic region (LVR) were performed increasing stress

168 from 0.1 to 3,000 Pa at 1 Hz frequency. Critical stress (Pa) was identified as the strain value

169 corresponding to a 10% drop in G' value.

2.5.6 Colour

171 The puff pastry surface colour was measured using a tristimulus Chromameter-2-Reflectance

172 colourimeter (Minolta, Osaka, Japan) with a CR-300 measuring head, standardized against a white tile,

173 and data were expressed in L*, a* and b* scale.

2.5.7 Leavening capacity

175 The leavening capacity of puff pastry was determined by measuring the volume gain (%) before and after

176 baking (cm^{3}g^{-1}) by using the rapeseed displacement method [34].

2.5.8 Sensory analysis

178 A group of 13 judges was used for the sensory evaluation of puff pastry samples. Judges were not trained

179 on sensory analysis of puff pastry but they were experts in the use of the selected sensory method. Control

180   puff pastry prepared with commercial margarine and the sample prepared with the MG9-dried emulsion

181   were evaluated. Judges were asked to evaluate the intensity of firmness, crispiness and oiliness of the

182   puff pastry samples, which were identified by 3-digit random codes. The selected descriptors were

183   evaluated on a 1-9 point hedonic scale, in which 1 corresponded to "extremely low descriptor intensity",

184   and 9 to "extremely high descriptor intensity" [35].

185   *2.5.9   Data elaboration*

186   All determinations were expressed as the mean ± standard error of at least three repeated measurements

187   from two experiment replicates. Statistical analysis was performed by using R v. 3.0.2 (The R Foundation

188   for Statistical Computing). Student's t-test was used to determine statistically significant differences

189   between means (p<0.05).

190   The kinetics constants of the drying processes (k, h^{-1}) were obtained by fitting the drying data, expressing

191   the moisture weight percentage as a function drying time (t, h), by using the model proposed by Lewis

192   [36] (eq. 1):

193    $$\frac{X - X_e}{X_0 - X_e} = \exp(-kt) \qquad\qquad (\text{eq. 1})$$

194   where X, X_e and X_0 are the moisture content (g/100 g) at any time, equilibrium and initial, respectively.

195   Data fitting was performed by using TableCurve2D software (Jandel Scientific, ver. 5.01). The goodness

196   of fit was evaluated based on statistical parameters of fitting (R^2, p).

3   Results and Discussion

3.1   Structured emulsion drying

Water-in-oil emulsions containing 47.6 g/100 g water and 4 or 9 g/100 g monoglycerides were prepared

and characterized. Taking into consideration the MG content, the samples were named as MG4 and MG9,

respectively. As expected and in agreement with the literature, the obtained systems were self-standing

white emulsions, as well visible from sample images and micrographs reported in Figure 1. This peculiar

structure is due to the self-assembling of saturated monoglycerides, which form walls surrounding oil

droplets [26–28,31]. The latter resulted smaller in the case of the emulsion containing 9 g/100 g MG, due

to the higher amount of MG available for the stabilization of the increased surface area associated with

smaller oil droplets [31].

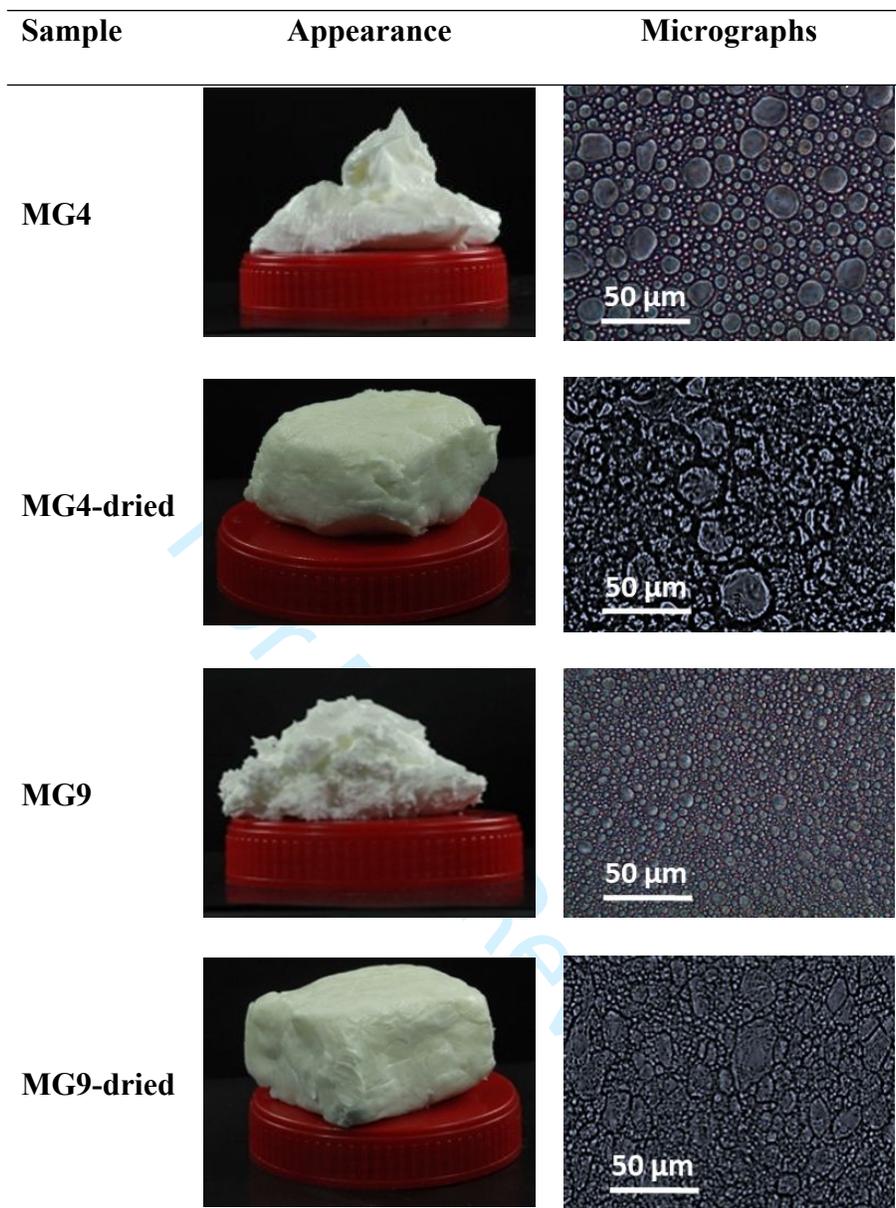| Sample	Appearance	Micrographs
MG4		
MG4-dried		
MG9		
MG9-dried		

Figure 1. Appearance and microscopic images of structured emulsions containing 4 and 9 g/100 g monoglycerides before (MG4, MG9) and after drying (MG4-dried, MG9-dried).

209 These systems were dried in a thin layer at 30, 40 and 50 °C to partially remove water and obtain systems

210 with 80 g/100 g lipid content, analogous to that of traditional margarine [21]. As previously mentioned,

211 it was not possible to prepare systems containing such lipid ratio by simply following the emulsion

212 preparation procedure. In this case, in agreement with the literature, a non-homogeneous liquid system

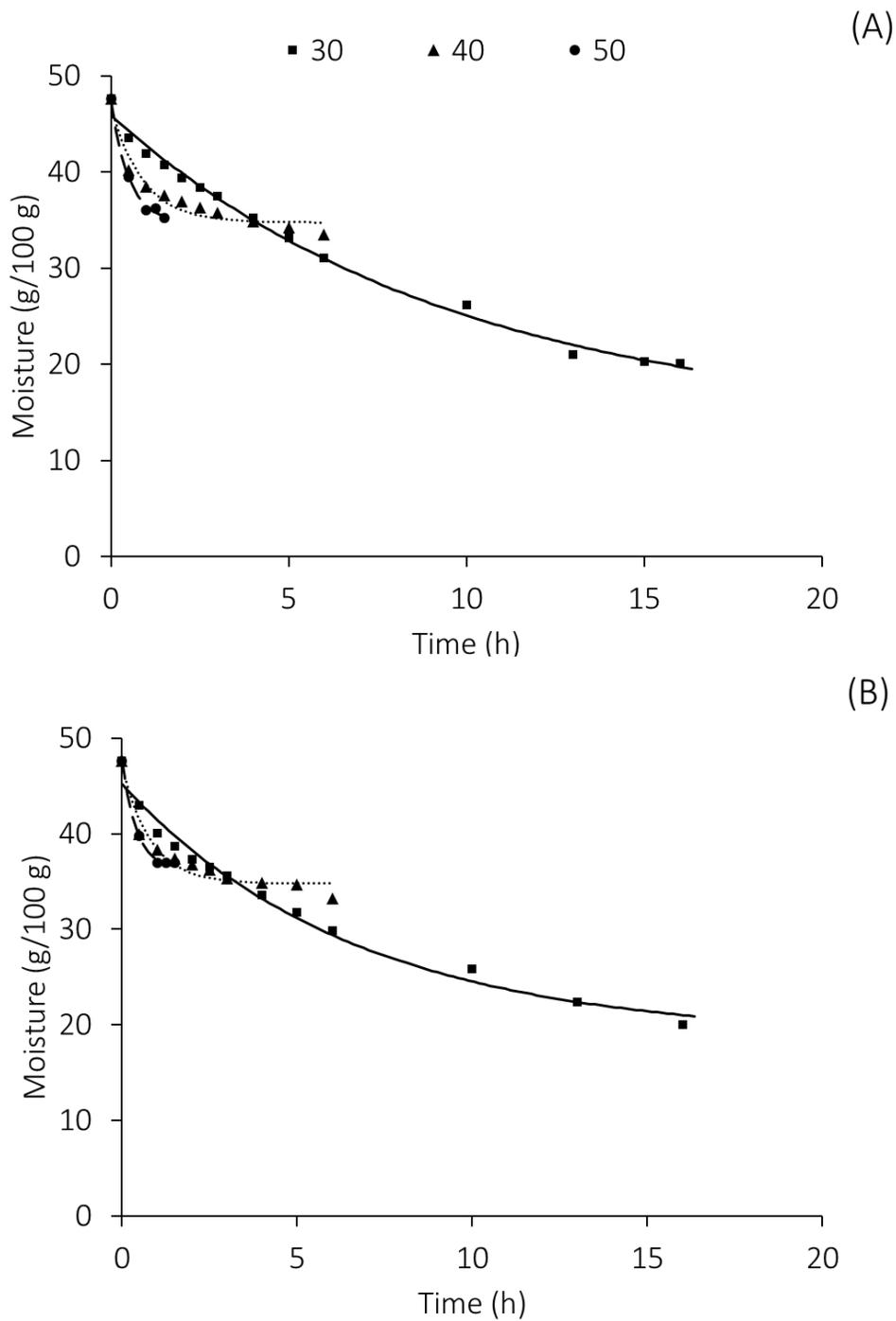213 was obtained, in which the lipid and water phase were separated [11].

Figure 2. Moisture of structured emulsions containing 4 g/100 g (A) and 9 g/100 g (B) monoglycerides as a function of drying time at 30, 40 and 50 °C (symbols). Fitting curves of drying model are also reported (lines).

218 Figure 2 reports the moisture (g/100 g) of the emulsions as a function of drying time at the different

219 temperatures. During drying, water content showed an exponential decrease, with an initial fast decrease-

220 phase followed by a tendency to reach a plateau. Experimental data were fitted to a classical exponential

221 decay model developed for thin-layer drying [36] to obtain the kinetic constants reported in Table 1. The

222 model well-explained experimental data, as shown by the R^2 values. Moreover, independently on the

223 MG content, as expected, the increase in drying temperature provoked an increase in the drying rate,

224 resulting in higher kinetic constant values (k, Table 1). However, it was not possible to take the samples

225 at the desired 20 g/100 g moisture value at both 40 and 50 °C. At these temperatures, independently on

226 the MG content, the systems broke down and leaked oil at moisture values of 33 and 35 g/100 g,

227 respectively, corresponding to a lipid content of 67 and 65 g/100 g. These results show that a drying

228 temperature higher than 30 °C is associated with the destruction of the MG network responsible for

229 emulsion structuration. By contrast, gentle drying at 30 °C allowed obtaining emulsions with 80 g/100 g

230 lipid phase after about 16 h. It must be underlined that a further water removal at 30 °C from both MG4

231 and MG9 sample resulted in emulsion break-down and oil leaking. These results suggest that the stability

232 of the samples during drying was affected by both the drying rate and the water removal extent. In this

233 regard, it is known that water is required to induce MG self-assembling into a supra-molecular

234 organization able to encapsulate oil and water among the MG lamellae [27,28]. Moreover, as evidenced

235 in the literature [27], the temperature should be strictly controlled during both emulsification and cooling

236 phase to properly structure the gel. In particular, the increase in temperature during emulsification has

237 been reported to affect the hydration state of the MG, decrease the electrostatic charge magnitude and

238 also induce changes in the mesomorphic phase, leading to a greater tendency to aggregation and thus

239 phase separation [27]. This evidence supports the hypothesis that also during drying, an accurate

240 optimization of temperature and thus of water removal rate, is required. Based on the acquired results, a

241 slow drying rate seems to allow the reorganization of the MG network surrounding oil droplets, while

242 preventing phase separation. By contrast, an increase in drying rate probably induced MG aggregation,

243 thus favouring system break-down.

244 Based on these results, the temperature of 30 °C was selected to partially dry the structured emulsions

245 MG4 and MG9 until reaching a water content of 20 g/100 g, comparable to that of conventional

246 margarine. Upon drying, the MG concentration of the emulsions initially containing 4 and 9 g/100 g MG

247 reached a concentration of 6.1 and 13.7 g/100 g, respectively. The obtained samples, called MG4-dried

248 and MG9-dried, are shown in Figure 1. As compared to the initial MG4 and MG9 emulsions, the partially

249 dried systems showed a more solid-like appearance and a microstructure with oil droplets tightly packed

250 into a network consisting of MG (Figure 1). The oil droplets present in the original structure were

251 deformed after drying, evolving from a spherical shape to a polygonal one. These results agree with

252 literature studies in which the emulsion-template method is used for oil structuring. This approach is

253 based on complete water evaporation from highly concentrated oil-in-water emulsions, which results in

254 the trapping of the oil in a physical network of biopolymers including polysaccharides and proteins or

255 their combination, originally forming the oil-water interface [37–40].

256 Differential scanning calorimetry measurements were carried out on the emulsions before and after

257 drying to identify the effects of drying on the melting temperature and the enthalpy of the transition.

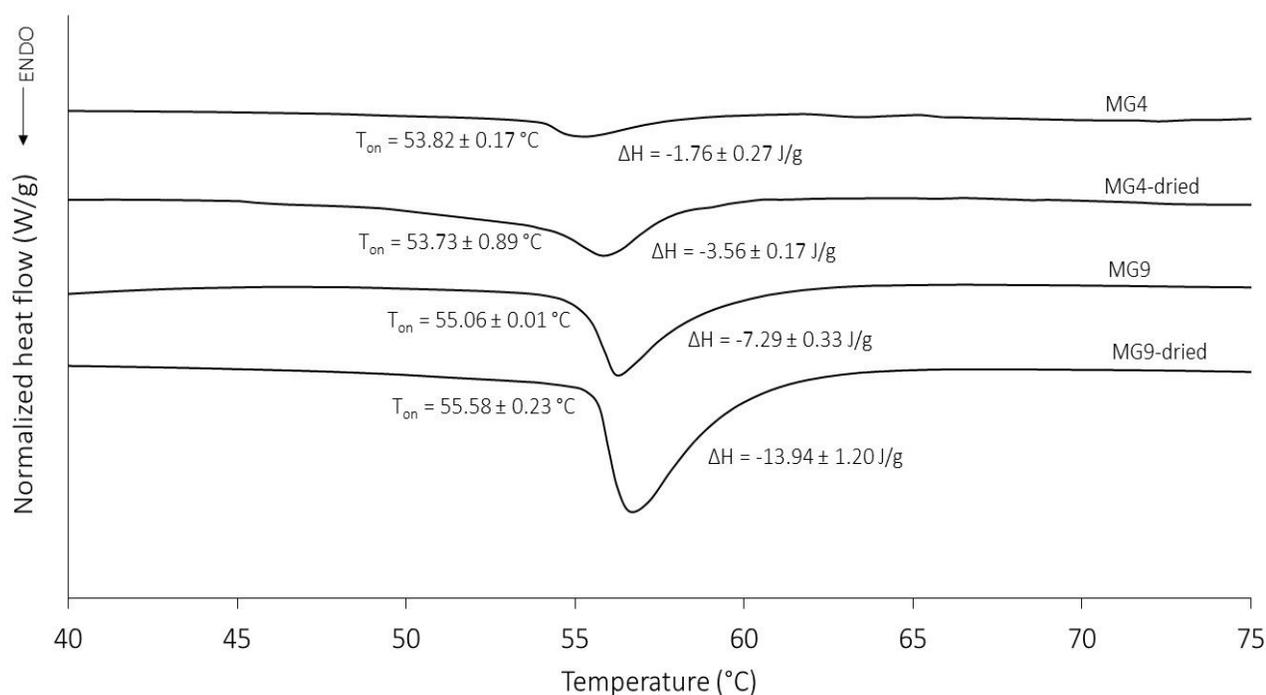258 Figure 3 shows the heating curves of the emulsions before and after drying.

Figure 3. Calorimetric melting curves of structured emulsions before (MG4, MG9) and after (MG4-dried, MG9-dried) drying. Onset melting temperature (T_{on}) and melting enthalpy (ΔH) are also reported.

All systems showed one endothermic peak corresponding to the disruption of the network among MG lamellar bilayers, followed by the melting of the crystalline MG [30]. The melting enthalpy (ΔH) expressed per g of emulsion resulted well correlated with the MG and co-surfactant content ($R^2 = 0.993$), being proportional to the quantity of crystallised material in the matrix [26,27,30]. By contrast, the melting enthalpy per g of saturated fatty acids was not significantly different in the MG4 and MG9 emulsions (data not shown), neither was affected by drying ($p \geq 0.05$). Moreover, no significant differences between sample melting temperature (T_{on}) before and after drying ($p \geq 0.05$) were observed. These results could be associated with the fact that drying did not significantly modify the amount of crystallized material in the samples.

272 Synchrotron XRD diffraction patterns were also acquired to obtain information on the effect of drying

273 on MG crystal packing in the emulsions. The patterns, reported in Figure 4, resulted similar to those

274 reported in the literature for analogous samples [27,28,30,31]. The diffraction signal superimposes onto

275 two large amorphous halos centred respectively about d = 23 Å (q = 0.27 Å $^{-1}$) and d = 4.42 Å (q = 1.4

276 Å^{-1}). Two lamellar forms appear to coexist, whose lower angle visible peaks correspond to average

277 interplanar distances of d = 62 Å (q = 0.1 Å $^{-1}$) and d = 48 Å (q = 0.13 Å^{-1}), respectively (Figure 4). All

278 other stakes around 4 Å are characteristic of MG high-angle diffraction patterns, the most intense being

279 at 4.54 and 4.12 Å. All the samples presented similar patterns in the wide angle region, due to the in-

280 plane chain ordering of MG crystals in the $L\beta$ phase [28,31]. Based on the fact that there were no

281 differences in the d-spacing of the samples in the wide-angle region, it can be concluded that they

282 presented the same in-plane molecular ordering [31]. In the small angle region, the diffraction signals of

283 the two lamellar phases appear to be much more intense with respect to the amorphous phase in sample

284 MG9 than in the sample MG4. In both cases, no structural differences were noted before and after drying,

285 and no transition toward a different lamellar organization occurred upon drying.

286

Figure 4. Powder X-ray diffraction spectra of structured emulsions before (MG4, MG9) and after (MG4-

288 dried, MG9-dried) drying. Patterns plotted as a function of reciprocal lattice spacing q, where q = 2π/d =

289 (4π/λ)·sinθ, d is the lattice spacing, λ is the X-ray wavelength and 2θ is the Bragg scattering angle,

290 represented as log-log plots opportunely scaled and shifted for clarity.

291

292

293 Finally, the rheological characteristics of the samples before and after drying were studied

294 (Supplementary Figure 1; Table 2).

Supplementary Figure 1. Amplitude sweep test of structured emulsions before (MG4, MG9) and after (MG4-dried, MG9-dried) drying. Error bars of triplicate measurements were typically not larger than the symbols and were left out for clarity.

In agreement with the literature, the emulsions before drying presented a gel-like behaviour (G'>G''), due to the elasticity of crystalline MG network, with critical stress and G' values increasing with the MG content [27,30]. As expected, the elastic modulus (G') and critical stress of the emulsion containing 9 g/100 g MG resulted significantly higher than that of the MG4 sample, due to the higher amount of solid fat content [11]. Drying led to a significant increase in both G' and the critical stress of the MG4 and MG9 emulsions (Table 2). This agrees with literature data showing that a lower water amount increases the packing of the MG, generating a system showing enhanced structural properties [11]. The values of

308 G' and critical stress have been previously indicated as key parameters for comparing the rheological

309 properties of MG structured emulsions with those of traditional solid fats [11]. Thus, rheological

310 properties of dried MG emulsions were compared to those of three commercial laminating fats with 20

311 g/100 g water content. In agreement with literature data [41,42], the latter gave G' and critical stress

312 values in the range 8.2-26.7×10^5 Pa and 263.7-995.4 Pa, respectively. With reference to these ranges, the

313 MG4-dried sample showed critical stress and G' values about 2 and 10 times lower, respectively. By

314 contrast, the MG9-dried emulsion showed rheological features comparable to those of the analyzed

315 commercial laminating fats (Table 2). Such rheological features have been previously obtained in

316 analogous MG structured emulsions by adding at least 15 g/100 g waxes or substituting sunflower oil

317 with palm oil [11]. However, the first systems showed unpleasant waxy taste, while the seconds require

318 the addition of saturated fatty acids.

3.2   Dried structured emulsions as fat alternatives in puff pastry

320 Based on its promising features, the MG9-dried sample was used in baking trials in the production of

321 puff pastry and compared to a common roll-in laminating palm margarine showing a G' and critical stress

322 of 15.7×10^5 and 995.4 Pa, respectively. Some of the preparation phases of the puff pastry using MG9-

323 dried sample are reported in the Supplementary Figure 2.

324   Supplementary Figure 2. Preparation phases of puff pastry using MG9-dried sample as laminating fat

325   substitute.

326

327

328   During both puff pastry preparation and baking, the MG9-dried emulsion showed the typical features

329   required to a laminating fat. Thanks to its plasticity, the MG9-dried sample was easily laminated into

330   thin sheets between dough layers during the folding process, during which it did not melt, preventing

331   the dough layers from touching. Therefore, upon lamination and folding, clearly defined and

332   continuous dough and MG9-dried layers were visible, confirming the optimal rheological features of

333   the MG9-dried sample. Finally, during baking, no evident oil leaking was observed, which indicates

334   that the fat phase was retained between the dough layers. This allowed the typical physical leavening of

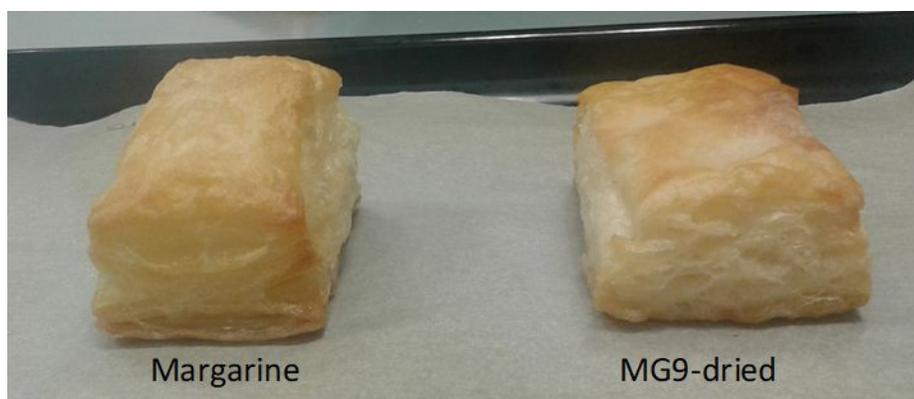335   puff pastry, as well-evident from the images reported in Figure 5.

336 Figure 5. Image of puff pastry prepared with commercial laminating margarine and MG9-dried emulsion.

337

338

339 The puff pastry prepared with the MG9-dried sample displayed the flaky golden-brown appearance of

340 puff pastry containing margarine. As shown in Table 3, the only observable difference was that MG9-

341 dried puff pastry showed a lighter colour, as indicated by the lower absolute values of the colourimetric

342 parameters a* and b* and a 16% lower leavening capacity. Moreover, it presented comparable

343 instrumental firmness, which was also confirmed by the judges during sensory evaluation (p≥0.05). The

344 latter also evidenced that the substitution of the commercial margarine with the MG9-dried fat did not

345 affect puff pastry friability (p≥0.05) but resulted in an increased oiliness mouthfeel. It must also be noted

346 that the judges did not highlight the presence of off-flavours or off-taste upon tasting the puff pastry

347 samples. These findings are very promising, as they show that MG9-dried fat can be used as laminating

348 fat replacement.

4 Conclusions

349

350 Results here reported face the timely challenge of developing substitutes of roll-in margarine able to

351 deliver a nutritional improvement of laminated products conventionally rich in saturated and trans fatty

352 acids. The process here proposed for the production of the margarine substitute requires simple unit

353 operations (e.g heating, homogenization, cooling, drying), which are commonly applied in food

354   industries, making its scaling up feasible both technically and economically. Based on the acquired

355   results, drying temperature is the key factor to be controlled to obtain a well-structured margarine

356   alternative. In fact, the desired 20 g/100 g water content, typical of margarine, is only obtained by

357   applying a gentle and gradual drying at 30 °C. Under these conditions, the MG structure entrapping both

358   water and oil is maintained, avoiding phase separation. The resulting system showed good performances

359   in both lamination and baking of puff pastry, making it an optimal candidate for the substitution of

360   margarine in the industrial production of laminated products with improved nutritional quality. It is not

361   excluded that by changing the starting emulsion formulation (e.g. oil type, other ingredients in the water

362   phase) it would be possible to further improve the range of MG-based margarine analogues.

Funding

Declaration of interest

367   The authors have declared no conflicts of interest.

Permission statements

369   The manuscript *does not* contain experiments using animals.

370   The manuscript *does not* contain human studies.

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429 Table 1. Model parameter estimates for the drying process at 30, 40 and 50 °C of structured emulsions

430 containing 4 (MG4) and 9% (MG9) monoglycerides.

Sample	Temperature (°C)	k (h^{-1})	R^2
MG4	30	0.103 ± 0.009	0.996
	40	1.119 ± 0.220	0.952
	50	2.018 ± 0.288	0.997
MG9	30	0.150 ± 0.024	0.982
	40	1.207 ± 0.234	0.953
	50	2.464 ± 0.315	0.997

431

432

433

434 Table 2. Rheological parameters of structured emulsions before (MG4, MG9) and after (MG4-dried,

435 MG9-dried) drying.

Sample	G' (×10^5 Pa)	Critical stress (Pa)
MG4	0.085 ± 0.012 [c]	22.6 ± 3.3 [d]
MG9	0.438 ± 0.029 [b]	86.0 ± 4.6 [c]
MG4-dried	0.533 ± 0.098 [b]	139.4 ± 6.4 [b]
MG9-dried	4.565 ± 0.212 [a]	633.9 ± 25.5 [a]

436 [a, b, c, d] In the same column, means indicated by different letters are statistically different ($p<0.05$)

437

438 Table 3. Leavening capacity, firmness, colour and sensory attributes of puff pastry prepared with

439 commercial roll-in margarine and MG9-dried emulsion.

Roll-in fat	Colour			Leavening capacity	Firmness (kN)	Sensory score		
	L*	a*	b*			Firmness	Friability	Oiliness
Margarine	82.84 ± 0.61	-1.22 ± 0.12	24.22 ± 0.93	12.3 ± 1.2	1.1 ± 0.2	5.0 ± 0.8	5.9 ± 1.6	4.5 ± 1.2
MG9-dried	85.66 ± 1.08	-0.50 ± 0.14	15.91 ± 0.76	10.4 ± 0.5	0.8 ± 0.1	4.9 ± 1.9	4.9 ± 2.3	7.3 ± 1.0

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