5. Application of organogels in a sunflower oil cake

The attempt of substituting saturated traditional fats with structured fatty materials has been thoroughly studied in the past 10 years (Pernetti et al., 2007; Hughes et al., 2009). To the best of our knowledge, however, only few tentative applications of these materials have been performed, in particular in baked products.
Nakano and Masaki (1989) patented an oil-structuring agent described as disaturated-monounsaturated mixed triglycerides featuring a minimum of one long chain (20 to 24 carbons) saturated fatty acid; this compound, when dissolved at a concentration of 10% in oil and slowly cooled, uniformly solidifies to thereby inhibit the migration of the liquid oil in food products (Nakano & Masaki, 1989). The material has solid-like properties and affects the crystallizing behaviours of other solid fats, imparting a migration-inhibiting effect at 0.5-1% in a baked product. This study indicates that the inhibition of oil migration via organogelation could be considered by the food industry, especially for products with fillings or fatty layers, validating the development and optimization of this promising technology (Hughes et al., 2009). More recently, Goldstein and Seetharaman (2011) studied the effect of a novel monoglyceride stabilized oil on cookie properties. They produced an emulsified shortening and found interesting benefits on cookie performances, especially on texture, compared to traditional fats. Another study, presented by Manzocco et al. (2012), investigated the properties of a short dough pastry in which the fat used was palm oil or a hydrogel, formed by a palm oil-water emulsion with saturated monoglyceride. The substitution of traditional fat with the hydrogel allowed a reduction in fat content with minor effects on pastry quality characteristics, although increasing acrylamide formation and staling tendency.
The aim of this part of the research was to employ sunflower oil organogels, produced with β-sitosterol and γ-oryzanol as gelators, in a plum cake formulation and evaluate their impact on cake final characteristics. Besides technological aspects, the chosen organogel is characterised by interesting nutritional benefits: anti-oxidant and anti-cholesterolemic activity, carried by the two gelators, and a polyunsaturated fatty acid rich profile, thanks to the use of sunflower oil. The evidence of phytosterols nutritional properties has been reported in chapter 1.5.3. In particular, Quìlez et al. (2003) added sterol esters in two different bakery products (muffin and croissant) and using a randomized, double-blind, placebo-controlled design, they provided 3.2 g/day of phytosterols, for 8 weeks. Total and LDL-cholesterol decreased in the tested group (by 5.5% and 10.4%, respectively), while in the control no changes were registered, concluding that bakery products are an excellent carrier for phytosterols.
Plum cake characteristics during storage were also studied by monitoring changes in texture and in both primary and secondary oxidation products. It is not unlikely, in fact, that structured oil could have an oxidative behaviour different from that of liquid oil. Actually, Da Pieve et al. (2011) found that a cod liver oil organogel formed by a monoglyceride gelator was ineffective on first steps of oxidation but seemed a consisting hurdle against the formation of secondary oxidation products. Considering the anti-oxidant properties of phytosterols, a slowdown in oxidation reactions could be a consequence of the organogel presence in the product, in order to produce a health-value added food.

5.1. Materials and Methods

5.1.1. *Materials*

Refined sunflower oil (Salov SpA, Massarosa, Italy) was bought in a local supermarket; fatty acids profile corresponded to 10.4% saturated, 27.6% mono-unsaturated and 53.8% polyunsaturated fatty acids. β-sitosterol was purchased from Novachimica Srl (Cinisello

Balsamo, Italy) and contained 10% campesterol; γ-oryzanol (minimum 95% pure) was kindly supplied by Sochim International SpA (Milano, Italy) for preliminary trials and by Prodotti Gianni SpA (Milano, Italy) - Jan Dekker International BV (Wormerveer, Netherlands) for rheological evaluations and cake preparation.
Sugar, liquid whole egg product, flour and baking powder were purchased from the same suppliers cited in § 3.1.1.
Chemicals used for PV analysis (chloroform, sodium thiosulfate, glacial acetic acid, potassium iodide, starch) and carbonyl compounds analysis (diethyl-ether, 1-butanol for spectrophotometric use, potassium hydroxide in pellets, 2,4-decadienal standard solution, di-nitro phenyilhydrazine - 50% moisture, chloridric acid) were purchased from Sigma-Aldrich (Sigma-Aldrich Co., Saint Louis, MO, USA).

5.1.2. *Organogel preparation*

Organogels (OG) at different sterol concentrations were prepared by weighing β-sitosterol and γ-oryzanol in different quantities but maintaining a 2:3 ratio (w/w) between them. The weighed compounds were dispersed in warm sunflower oil (≅ 75°C), in order to achieve the complete melting of the gelators (Bot et al., 2009). The mix was stirred until obtaining a clear solution. The material was then poured in small screw cap containers (25 g each) and stored at 4°C for exactly 1 week. OG were conditioned at 25°C for 30 minutes before using them in the cake formulation or performing the rheological evaluations.

5.1.3. *Experimental design*

A two-factor, five-level Central Composite Design (CCD) was used to plan the experiments. The two factors considered were the quantity of gelled sunflower oil in the cake batter and the percentage of gelators in the oil. The ±1 levels of the two considered factors were 8-22% for OG content in the batter, 3.50-5.50% for gelators concentration in the OG. The centre point cake (named CC) corresponded to a formulation with 15% OG content in the batter and 4.50% gelators in the OG. Cakes were prepared following the process previously described (§ 3.1.5). In Table 5.1, run order, factor levels and plum cake formulations are reported.
CC sample, produced following the centre point formulation of CCD, contained around 2 g of total phytosterols in a 240 g cake. A supposed cake consumption of 120 g/day (half cake) could provide around 1 g of phytosterols. This value is definitely higher than the average daily supply in western diet (0.15 – 0.4 g) and complies with the maximum daily consumption suggested by EFSA (3 g, as previously reported in § 1.3.3).

Table 5.1. Run order of the Central Composite Design, corresponding coded and uncoded factor levels and batter formulations.

Run	FACTORS		INGREDIENTS (% in batter)					
	OG (coded)	Gelators (coded)	OG	Gelators [a]	Flour	Sugar	Egg product	Baking Powder
1	-1	-1	8.00	3.50	41.34	24.75	24.75	1.16
2	0	0	15.00	4.50	38.21	22.86	22.86	1.07
3	-√2	0	5.10	4.50	42.66	25.53	25.53	1.18
4	0	-√2	15.00	3.09	38.21	22.86	22.86	1.07
5	0	0	15.00	4.50	38.21	22.86	22.86	1.07
6	+1	+1	22.00	5.50	35.07	20.98	20.98	0.97
7	+√2	0	24.90	4.50	33.76	20.20	20.20	0.94
8	0	0	15.00	4.50	38.21	22.86	22.86	1.07
9	0	0	15.00	4.50	38.21	22.86	22.86	1.07
10	-1	+1	8.00	5.50	41.34	24.75	24.75	1.16
11	0	0	15.00	4.50	38.21	22.86	22.86	1.07
12	0	+√2	15.00	5.91	38.21	22.86	22.86	1.07
13	+1	-1	22.00	3.50	35.07	20.98	20.98	0.97

[a] expressed as percentage in the OG (w/w)

5.1.4. *Reference samples*

In addition to the runs scheduled in the experimental design, a cake produced with 15% sunflower oil in the batter (named SC) was produced, following the process reported in § 3.1.5. Moreover, another cake was prepared with the same process, using 15% sunflower oil in the batter and adding 4.5% gelators directly in the flour, as powders, thus maintaining the same percentage used for CC but without using a pre-formed OG; this last cake will be named SPC (sunflower powder cake). Both SC and SPC cakes were produced in double and analysed as previously reported (§ 3.1.6).

5.1.5. *Cake storage*

In order to monitor product changes during shelf life, SC and CC cakes were packed in OPP bags and stored at 25°C for 24 days. Texture, moisture and oxidation indexes (PV and carbonyl compounds) were measured as described in § 5.1.7.

5.1.6. *Rheological measurements*

Rheological behaviour of organogels and selected batters was evaluated using a Physica MCR 300 rheometer (Anton Paar, Graz, Austria), supported by Rheoplus/32 software (v3.00, Physica Messtechnik GmbH, Ostfildern, Germany), and equipped with a plate-plate geometry (d=25 mm, rough surface), gapped to 1 mm.

OG produced for preliminary trials and the five OG produced for the Experimental Design were studied through frequency sweep (FS) tests, performed at 25°C, with γ=0.01% and f=0.01-10 Hz. These tests conditions were within the linear viscoelastic range, previously determined by strain sweep (SS) tests carried out at 25°C, with f=1 Hz and γ=0.001-100%.

Batters of CC, SC and SPC cakes were evaluated. Temperature Sweep (TS) tests (25°C to 70°C; heating rate, 2.5°C/min) were performed with γ=0.01% and f=1 Hz. FS tests were performed at 25°C, with γ=0.01% and f=0.01-100 Hz. The test conditions were within the linear viscoelastic range, previously determined by SS tests performed at 25°C, with f=1 Hz and γ=0.001-100%. The exposed edges of the sample were covered with a thin layer of paraffin oil to prevent dehydration during measurements.

5.1.7. *Analytical measurements*

Cake texture, moisture, volume, colour and apparent specific gravity were measured following the methods described in § 3.1.6.

Oxidation indexes considered were Peroxide Value (PV) and carbonyl compounds content.

PV was determined following the standard method, as reported in § 4.1.3.

Carbonyl compounds were determined following the tentative method 13:2003 reported in "The Standard Methods for the Analysis of Fats, Oils and Related Materials" by Japan Oil Chemists' Society. This method, proposed by Endo and co-workers to analyse pure oil (Endo et al., 2003) and afterwards proposed for food products evaluation (Usuki et al., 2009), employs 1-buthanol as a solvent, instead of toxic benzene (as normally used in the Standard Method 2.5.4-1996 by JOCS). In this research, oil was extracted by milling two slices of cake using a rotating blade mixer; 10 g of the milled cake (in duplicate) were weighted, 100 mL of diethyl ether was added and the suspension was stirred for 15 min. The ethereal extract was then filtered under vacuum with rough filter paper (60 g/m^3, 0.14 mm thickness and capillary rise > 79 mm, ALBET-Hahnemuehle, Barcelona, Spain). The material was re-extracted twice (70 mL of solvent for 10 minutes each time), recovering the powder possibly fallen on the filter during filtration. The filtered solution was then evaporated under vacuum and, after removing last traces of solvent by flowing N_2, oil was weighed (0.10 g or 0.15 g). The analysis then followed the protocol reported by Endo et al. (2003). A calibration line was obtained plotting increasing concentrations of 2,4-decadienal standard solutions, from 50 to 400 µmol, versus the corresponding measured adsorbance, and calculating the best fitting equation. Results were calculated as µmol of 2,4-decadienal on kg of oil/OG, and afterwards expressed as mmol of 2,4-decadienal on 100 g of dry cake. 2,4-decadienal was chosen as the reference compound, since it is the most abundant secondary oxidation product in sunflower oil, as reported by Guillén and Uriarte, 2011.

5.1.8. *Statistical analyses*

CCD generation, data analysis and optimization were performed using Design Expert 8 (Stat Ease Inc., Minneapolis, USA). Data were treated by the same software, as described in § 3.1.4.
In order to determine the significant difference among samples, the one way analysis of variance (ANOVA) was carried out, followed by Fisher's Least Significant Different (LSD) test in order to discriminate samples. Data was elaborated using the Statgraphics Plus 5.1 (Statistical Graphics Corp., 1994-2001, Princeton, NJ, USA).

5.2. Results and Discussion

5.2.1. *Preliminary trials*

The choice of gelators percentages in the CCD was made considering various aspects. From literature, several studies (Bot & Agterof, 2006; Duffy et al., 2009; Sawalha et al., 2011) considered percentages ranging from 6% to 25%, obtaining gels whose properties resembled those of a block of fat. Bot et al. (2009) reported that 4% of structurants is needed to form a gel at 5°C, but in this case its characteristics strongly depended on the ratio between gelators.
In a first explorative stage of the research, rheological characterization of organogel was done. It resulted that an OG with 5% gelators showed storage and loss modulus higher than butter (Fig. 5.1); however, when 4% gelators concentration was used, fat texture was much softer than butter. In this range of gelators concentration, a large variation of both G' and G'' was thus evidenced, as it can be seen in Fig. 5.1. Considering the results obtained and the information found in the literature, concerning mostly OG made with sunflower oil, chosen gelators percentages in sunflower oil were from 3.09% to 5.91% (CCD star points). Those organogels included structured and weakly structured materials, in order to highlight how different levels of gel structuration could influence cake characteristics. Moreover, the use of soft OG in cake formulation could have a positive impact on final product characteristics, as it was seen from the optimized formulation produced with palm oil-palm olein blend (§ 3.2.3). Finally, a reduction in the quantities of phytosterols used could be desirable also from an industrial point of view, as these compounds are high cost materials (on the marketplace, the average cost is 80€/kg).

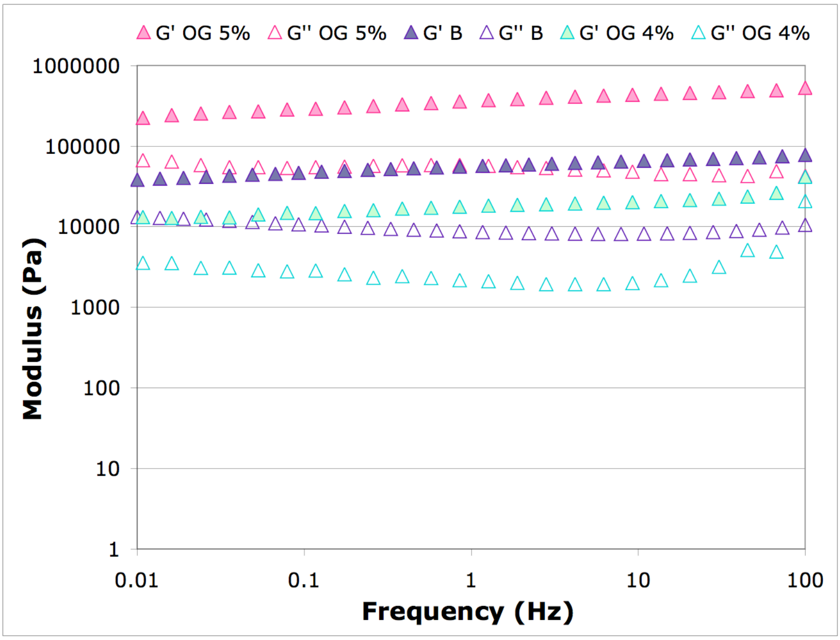


Figure 5.1. Mechanical spectra for storage and loss modulus for butter (B) and OG with 4% and 5% gelators in sunflower oil; each spectra is the average of 3 replicates.

5.2.2. *Experimental Design and reference cakes*

Table 5.2 shows the results of the response variables considered in the thirteen runs of the Central Composite Design, while in Fig. 5.2 some examples of cakes corresponding to selected CCD runs are shown.

Table 5.2. Mean values of the response variables considered in the Central Composite Design.

RUN	RESPONSES							
	Slice Moisture (%)	Crumb Moisture (%)	Specific Volume (cm^3/g)	Young Modulus (kPa)	Load at 25% Strain (N)	L* Crust	a* Crust	b* Crust
1	20.56	23.70	2.65	64.65	6.55	69.53	3.47	35.21
2	18.11	21.44	2.80	70.10	6.48	67.11	5.31	36.82
3	20.47	24.48	2.92	74.62	7.32	72.12	3.31	35.01
4	18.28	21.77	2.53	71.56	6.66	68.34	3.60	36.09
5	18.31	21.67	2.63	71.06	6.77	65.31	5.33	36.50
6	16.65	19.83	2.18	71.83	6.46	63.77	4.51	36.65
7	16.22	21.69	1.49	156.41	15.69	59.69	4.59	37.29
8	18.18	21.59	2.55	74.62	6.77	69.04	3.95	37.41
9	18.31	21.81	2.55	72.93	6.63	67.11	4.99	37.15
10	20.79	23.78	2.62	88.09	8.32	71.57	3.22	36.87
11	18.37	21.47	2.53	79.81	7.08	68.51	4.37	36.88
12	18.29	21.58	2.51	75.06	6.97	69.75	3.41	36.99
13	16.83	19.65	2.32	71.86	6.63	66.04	5.15	37.81

Table 5.3. Analysis of effects for selected responses of the Experimental Design.

	Moisture	Specific Volume	L* Crust	b* Crust	Load 25% strain
OG	- - - ($p<0.001$)	- ($p<0.05$)	- ($p<0.05$)	- ($p<0.05$)	- ($p<0.05$)
Gelators	NS	NS	- ($p<0.05$)	NS	+ ($p<0.05$)

Results obtained from the experimental design demonstrated that OG quantity in the batter showed significant effects for moisture, volume, load at 25% strain and some colour parameters (Table 5.3). The role of gelators percentage appeared to be less clear, as their impact was significant only for Crust L* and Load at 25%. Unfortunately, significant models were found for Crust L*, Crust a*, Crust b* and cake (both slice and crumb) moisture, only for OG quantity in the cake. Nevertheless, colour variables do not add information on final product characteristics and cannot be considered important cake quality indicators; regarding moisture, it could be easily predicted that its variations would be directly linked to fat quantity in the cake, as it was previously discussed (§ 3.2.2). For all the other response variables considered in the CCD, models were not significant for both factors, due to the high lack of fit, indicating their inability to predict response values. Thus, optimization of organogel cake was not feasible on the basis of CCD outcomes. In Fig. 5.3, two examples of response surfaces are showed.

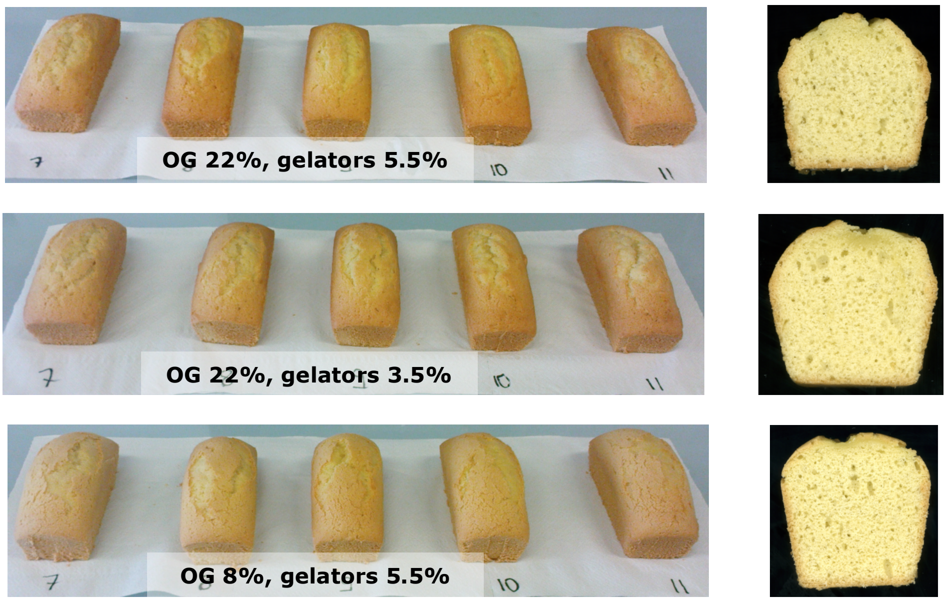


Figure 5.2. Selected CCD runs: five baked cakes produced for each run (left) and central slice of the cake (right).

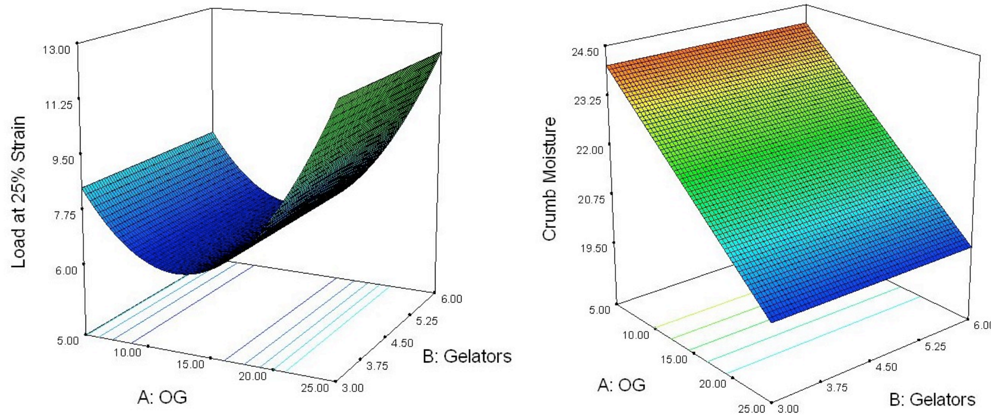


Figure 5.3. Response surfaces for a not significant model for both factors (load at 25% strain, left) and a significant model for OG factor (crumb moisture, right).

In order to see if the range of the two factors considered could give noticeable response variations, the standard deviation of the overall mean of the design points (except CCD centre points) was compared to that of the mean of the five CCD centre points, for all response variables; selected variables are shown Table 5.4. Standard deviation and coefficient of variation of the five centre points were, for all variables, smaller than that of the rest of the runs, thus showing that the effects are present but not displayed along a response surface. In other words, it was possible to appreciate effects of the two factors on cake characteristics but they did not follow a systematic distribution. This is confirmed by the high lack of most response surface models.

Table 5.4. Mean values, standard deviation and coefficient of variation of the five centre points and of the remaining runs of CCD for selected variables.

	Cube and Star Points			Centre Points		
	Mean	**SD**	**CV**	**Mean**	**SD**	**CV**
Slice Moisture (%)	18.51	1.88	10.19	18.26	0.11	0.58
Crumb Moisture (%)	22.07	1.80	8.14	21.57	0.11	0.52
Specific Volume (cm^3/g)	2.40	0.43	17.97	2.61	0.11	4.21
Young Modulus (kPa)	84.26	29.88	35.46	73.70	3.83	5.19
Load at 25% Strain (N)	8.07	3.14	38.85	6.74	0.22	3.28
Crust a*	3.90	0.73	18.76	4.79	0.60	12.73
Crust b*	36.49	0.99	2.70	36.95	0.34	0.93
Crumb a*	-5.86	0.41	6.96	-5.74	0.31	5.45
Crumb b*	27.38	1.17	4.26	26.40	0.48	1.84

5.2.3. *Organogel rheological characteristics*

Our results obtained from the rheological characterization of the five different OG used in the DoE confirmed what it is already reported in the literature (Bot & Agterof, 2006; Sawalha et al., 2011): consistency of OG (measured as storage modulus) increased with increasing gelators concentration. In particular, the gap of G' values between gels with gelators concentration of 3.50% and 4.50% resulted broad (Fig. 5.4).

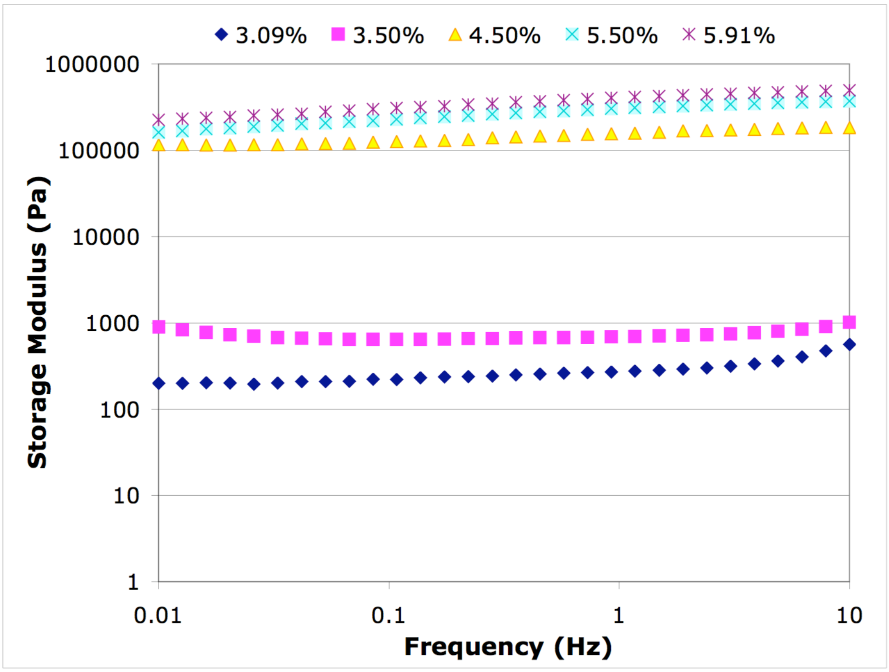


Figure 5.4. Storage modulus values for organogels at concentrations used in CCD; each curve is the average of 3 replicates.

It was previously stated that different concentrations of phytosterols do influence the rheological properties but not the effective formation of the nano-tubules, the base components of these materials (Bot et al., 2009). Observing the mechanical spectra of the samples in Fig. 5.5, in fact, all the gels showed a solid-like structure, with storage modulus higher than loss modulus, highlighting the fact that in all the runs of the experimental design an effectively structured fat was employed. Furthermore, the gels showed a shear thinning behaviour: as frequency increased, complex viscosity decreased (Fig. 5.6).

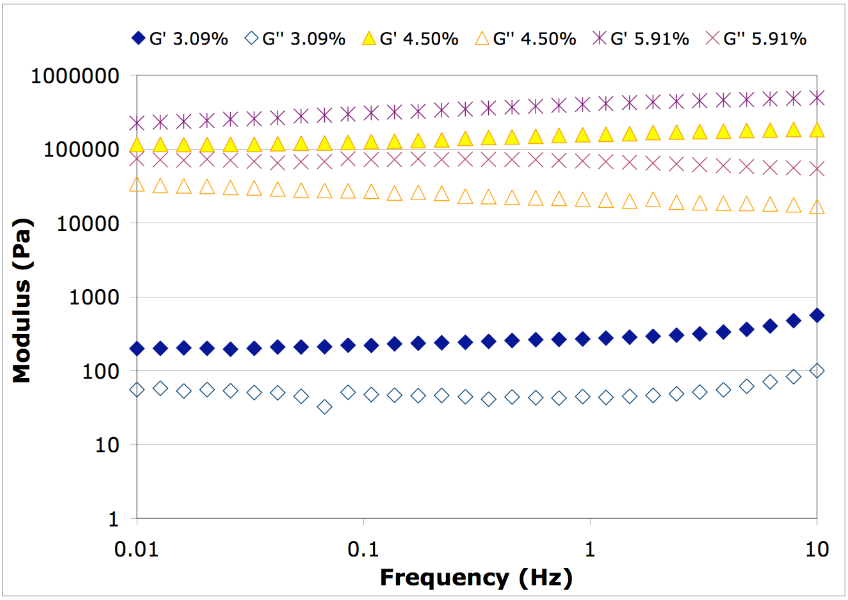


Figure 5.5. Mechanical spectra for selected gelators concentrations used in CCD; each spectra is the average of 3 replicates.

It is thus possible to conclude that formation of gel occurred in all the concentrations considered and that the samples had different textural properties. Since the effect of OG on cake properties was not significant, it could be worthwhile investigating more deeply on the causes of these phenomena.

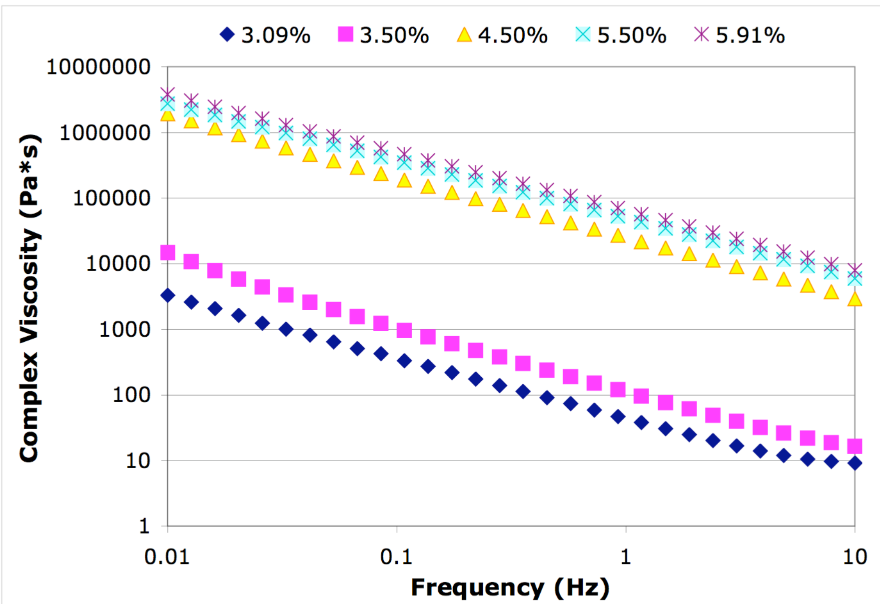


Figure 5.6. Complex viscosity values for gelators concentrations used in CCD; each spectra is the average of 3 replicates.

In order to perform the CCD runs, the production in different days of two or more replicates of OG with the same gelators concentration was required. Comparing replicates of storage modulus of OG with 3.5% gelators produced in two different days (Fig 5.7), a different rheological profile was evidenced. Furthermore, the required time for performing the analysis and replacing the sample is around 2 hours, so also the replicates of gel rheological analyses performed in the same day, were carried out after different times from gel preparation. This time lap between replicates allowed the gel to evolve through time, probably reorganizing itself and creating more bounds among tubules, as the second spectra of the OG (in the same day) showed a higher storage modulus. A hypothesis for this behaviour is a disturbance due to sampling. In fact, it has been reported that OG formation is influenced by the amplitude of oscillating applied stress. Bot & Agterof (2006) reported that increasing shear stress amplitudes resulted in faster gel formation, thus the handling of the sample could (unavoidably) influence and, eventually, arouse OG structuring, by increasing connections among tubules. Moreover, an OG with the same gelator percentage but produced in a different day resulted in a material with different rheological characteristics. Thus, one could conclude that the standardization of organogel production (storage for exactly 1 week at 4°C) could not guarantee the same outcomes in terms of gel rheological characteristics. Nevertheless, it is interesting to notice that the gap between OG replicates appears clearly in mechanical spectra only for certain gelators concentrations.

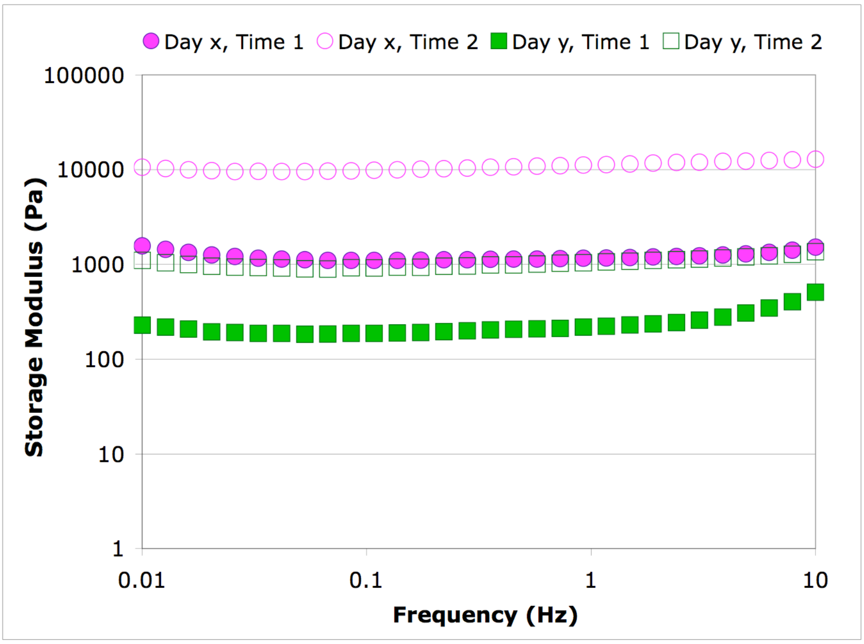


Figure 5.7. Storage modulus for OG at 3.5% gelators concentration, measured on day x and y, at time 1 and 2.

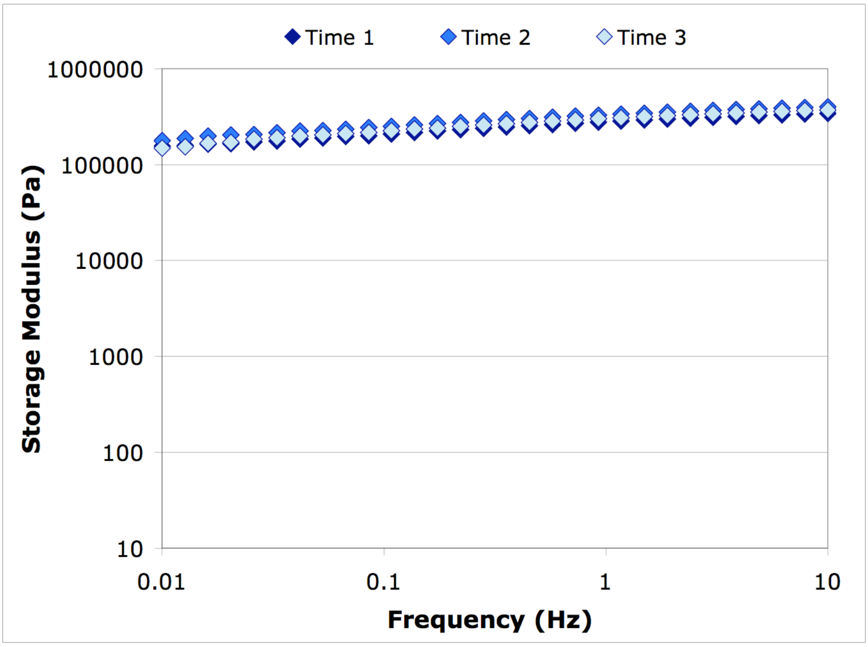


Figure 5.8. Replicated values of storage modulus for OG at 5.5% gelators concentration, measured on the same day.

In Fig. 5.8, storage modulus values (obtained from FS tests) for OG with 5.5% gelators are reported. In this case, unlike the high variability of the different replicates found for OG at low concentration, the small differences among the three mechanical spectra could be attributed to the experimental error (CV of 8%). In the literature, in fact, none of the studies (Bot & Agterof, 2006; Bot et al., 2008; Bot et al., 2009; Sawalha et al., 2011) that produced OG in non-controlled shear conditions reported discrepancies among replicates of the same sample or differences between the characteristics of the same gel produced in controlled and non-controlled shear conditions. This was probably due to the fact that in the literature works the percentages of gelators used were higher than 6% At these concentrations, the OG is more structured and not subjected to perturbations or variations during production or storage, as also confirmed by our findings. Moreover, in most research papers (Bot & Agterof, 2006; Sawalha et al., 2011) in which organogel rheological characteristics were studied, the OG were formed directly in the rheometer (either by cone-plate, plate-plate or Couette system), undress strictly controlled conditions.

5.2.4. *Reference cakes characteristics*

Experimental design outcomes showed that different gelators concentrations (in the range of 3.09-5.91% in the OG) did not significantly influence cake characteristics. In order to better understand if the structuring of oil in the form of organogel had an impact on batter and cake properties, three formulations at equal 15% fat content were compared: SC, with liquid sunflower oil; SPC, with liquid sunflower oil and powder gelators dispersed directly in the flour (no pre-formed OG used); CC, centre point formulation of the CCD (pre-formed OG used).
Regarding batters characteristics, it was possible to observe that the presence of OG in the formulation did not affect mechanical characteristics of the batters. In fact, as shown in Fig. 5.9, the storage modulus curves of the three batters (CC, SPC, SC) are overlapped.

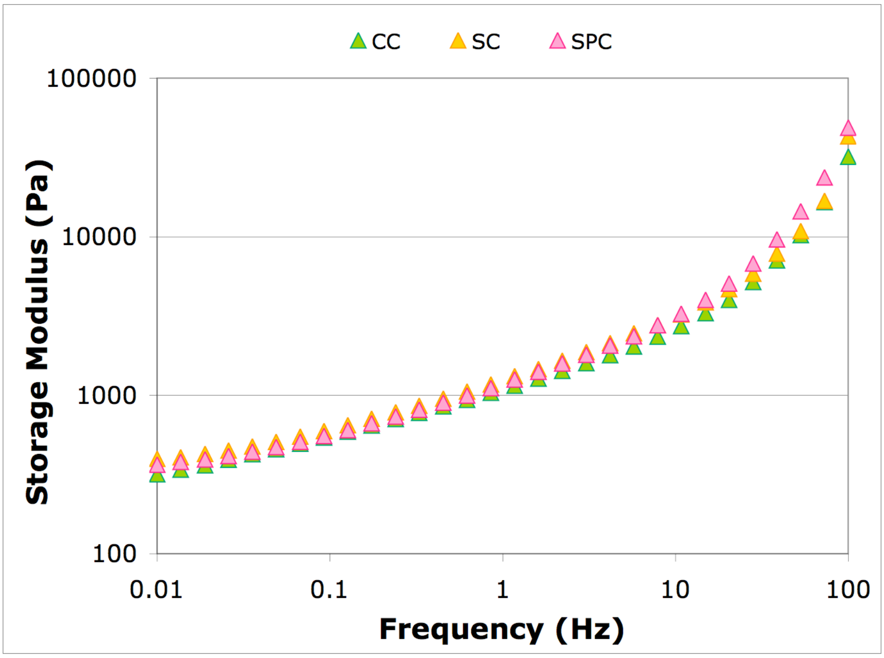


Figure 5.9. Storage modulus values in mechanical spectra for CC, SC and SPC batters.

It is likely that the OG could not keep its structure during batter preparation, as the mechanical action of the beater during creaming breaks down the gelly solid without enhancing retention of air. This is confirmed by the evaluation of the creaming properties of the three batters: they were not significantly different ($p>0.05$) in terms of apparent specific gravity (SG), with an average value of 1.08 ± 0.03.

Comparing the TS curves of the three samples, a similar behaviour for the batters was found, with a gradual decrease of both moduli (data not shown) and viscosity, as the temperature reached higher values (Fig. 5.10).

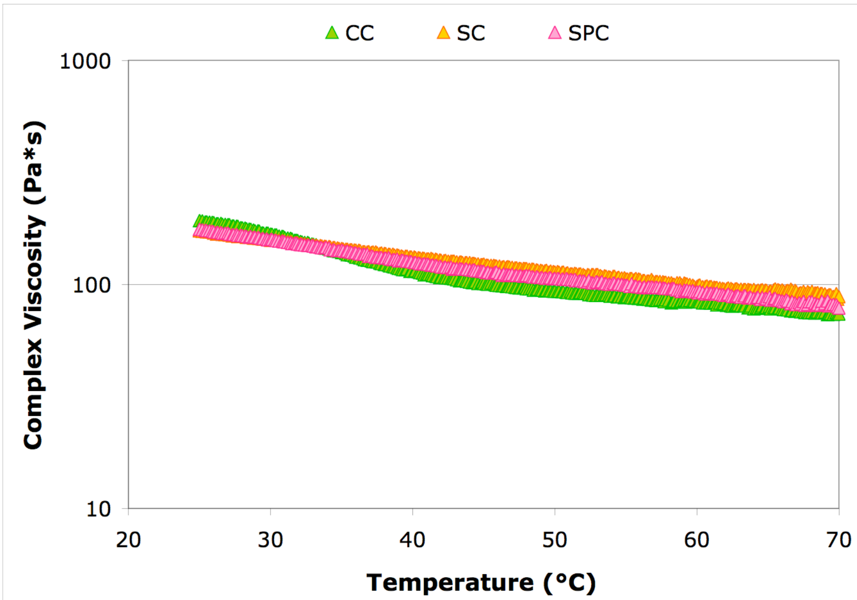


Figure 5.10. Complex viscosity values in TS test for CC, SC and SPC batters.

Observing the curves of complex viscosity, a slightly different trend of CC curve is noticeable. In fact, there is a steeper viscosity decrease at 30-35°C, that could be related to the melting of the OG and thus to a small loss in structuration in the whole batter. However, the difference among the three samples, calculated by applying ANOVA to mean values taken from each curve every 5°C, was always not significant ($p>0.05$). The similarity of batter viscosities yielded similar specific volume values in final cakes, according to findings of Shelke et al. (1990); nonetheless, it was possible to observe interesting differences in terms of texture of the final product (Table 5.5). SC and SPC resulted significantly ($p<0.05$) harder than the CCD centre point (CC), as shown by the highest values of Young Modulus and Load at 25% strain, highlighting that fat structuration through organogelation had a positive impact on final cake characteristics. Moreover, no significant difference ($p>0.05$) was shown between SC and SPC cakes; this means that addition of β-sitosterol and γ-oryzanol as powders does not influence, neither positively nor negatively, standard cake performances. Goldstein and Seetharaman (2011) found the same behaviour in a cookie produced with a monoglyceride (MAG) gel: the sample containing the structured fat was softer than the one with the reference shortening or with dry MAG in the flour. This outcome could seem in contrast with the fact the specific gravity (SG) of the batters was not significantly different ($p>0.05$); however, it is worth considering that SG of batters measures the total air holding capacity but gives little information about bubble size or dispersion (Sanz et al., 2008), and air retention properties of the batter over time. Regarding colour measurements, CC crust and crumb presented a significantly higher red parameter (a*), while crumb had a minor yellow component (b*). In terms of specific volume and moisture, the three formulations studied were not significantly different ($p>0.05$). Furthermore, it is worth observing slice images in Fig 5.11: CC crumb appears characterized by a more homogeneous structure, thus probably explaining the softer consistency of the product.

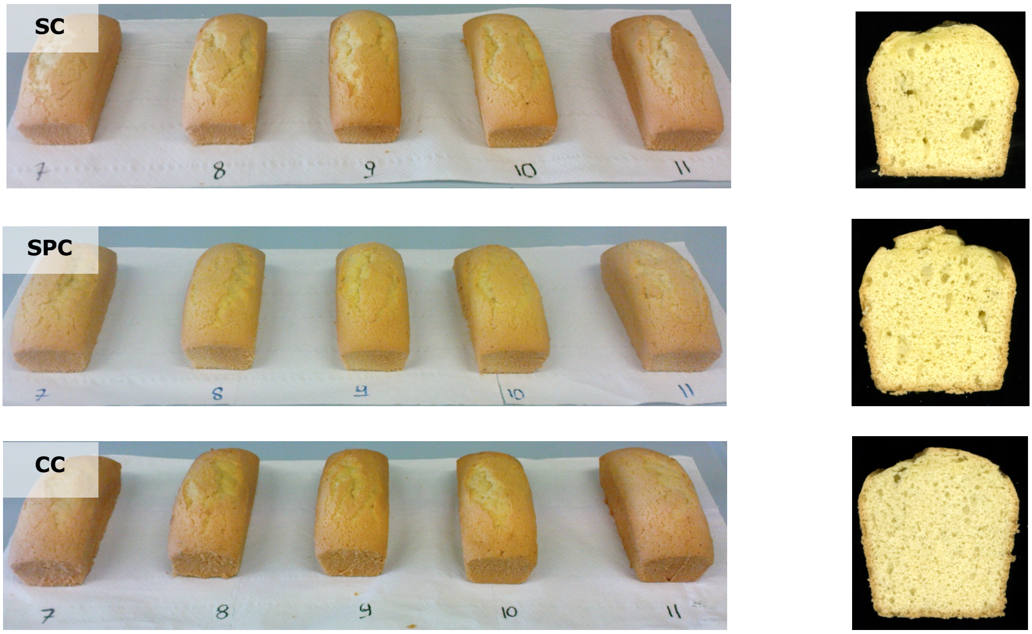


Figure 5.11. Baked samples (left) and central slice (right) for SC, SPC and CC cakes.

Table 5.5. Values of selected responses for SC, SPC and CC.

	Slice Moisture (%)	Crumb Moisture (%)	Specific Volume (cm^3/g)	Young Modulus (kPa)	Load at 25% Strain (N)	Crust		Crumb	
						a*	b*	a*	b*
SC[1]	18.31±0.09[a]	21.49±0.07 [a]	2.65±0.04[a]	88.44±0.33[a]	8.23±0.04[a]	3.96±1.43[a]	37.72±0.47[a]	-6.82±0.02[a]	29.43±0.83[a]
SPC[1]	18.12±0.15[a]	21.52±0.01[a]	2.52±0.01[a]	94.22±3.83[a]	8.71±0.50[a]	3.05±0.34[a]	37.26±0.77[a]	-6.51±0.06[a]	29.22±0.26[a]
CC[2]	18.26±0.11[a]	21.60±0.15[a]	2.61±0.11[a]	73.70±3.83[b]	6.74±0.22[b]	4.79±2.02[b]	36.95±0.87[a]	-5.74±0.36[b]	26.40±0.88[b]

[1] average and SD calculated on two technological replicates; [2] average and SD calculated on five technological replicates
[a,b] Mean values with different letters within the same column are statistically different ($p<0.05$)

5.2.5. *Cake storage*

Even though it was not observed a clear trend of the influence of gelators concentration on cake characteristics, it has been shown that the presence of OG in the formulation enhances product texture (§ 5.2.4). With this in mind, it was worth investigating if a positive influence could be maintained during storage, since the structured fat could inhibit some migration phenomena linked to staling and overall quality depletion of the cake (as previously reported in § 5).

SC and CC formulations were thus newly produced and monitored during 24 days storage at 25°C.

As previously observed (§ 4.2.2), moisture gradually decreased both in slice and crumb, with crumb moisture values higher than those of the whole slice (Fig. 5.12). The initial difference between crumb and slice moisture (around 3%) gradually decreases, until converging to similar moisture values after 15-20 days of storage.

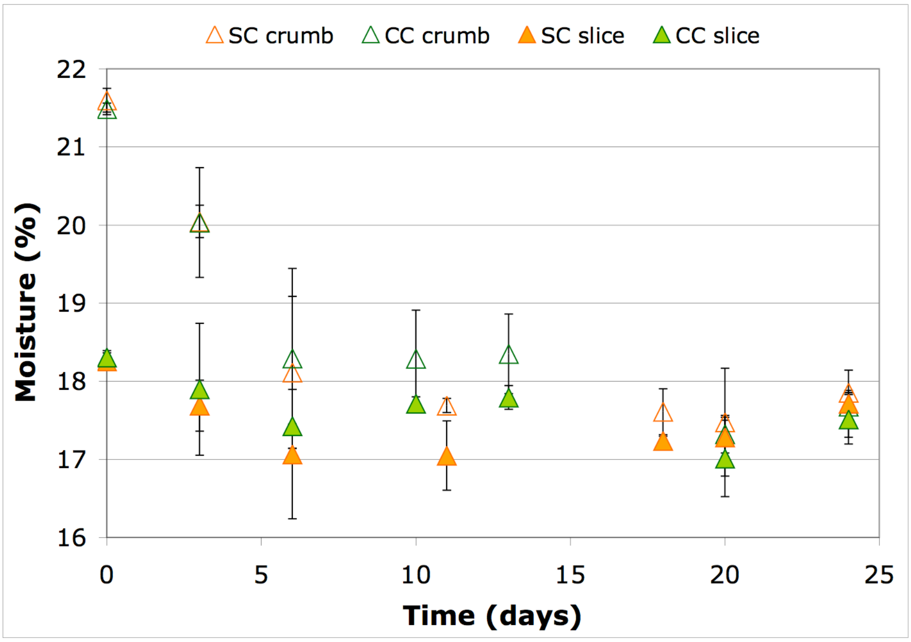
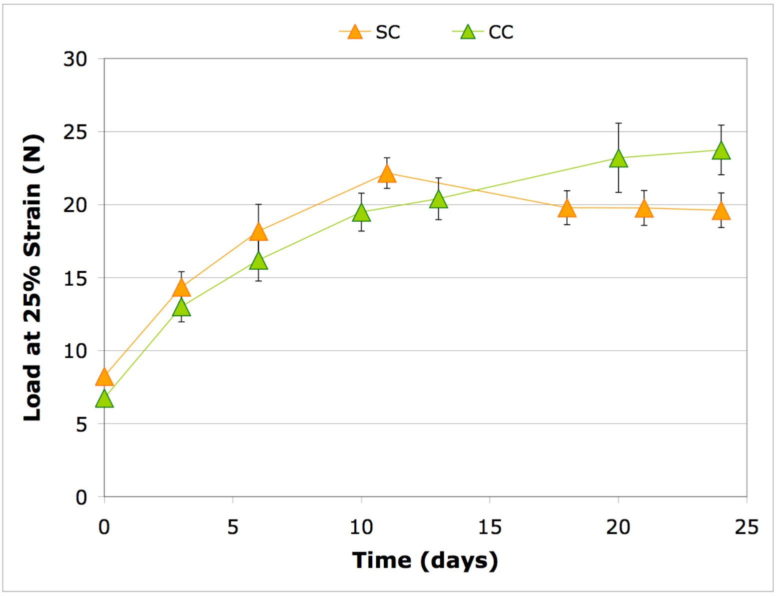


Figure 5.12. Slice and crumb moisture values for CC and SC cakes during storage.

Regarding texture characteristics, in the first 10-15 days of storage CC formulation maintained the initial lower firmness showed by the fresh product (Fig. 5.13); after 15 days of storage, load and Young Modulus values for SC cake do not increase further, in contrast with the trend observed for CC. This behaviour can be linked to moisture loss; load at 25% strain and crumb moisture are in fact highly correlated, both for SC and CC cakes ($R = -0.972$ and $R = -0.964$, respectively). Moreover, the initial higher softness of CC could be attributed to a lubricating effect of organogels, as the presence of a structured fat may improve water retention in the cake for a longer time. Kumari et al. (2011) found that the use of sunflower oil (compared to saturated coconut oil) in pound cakes decreased the moisture content of the product, indicating less water binding capacity of cake containing oil.



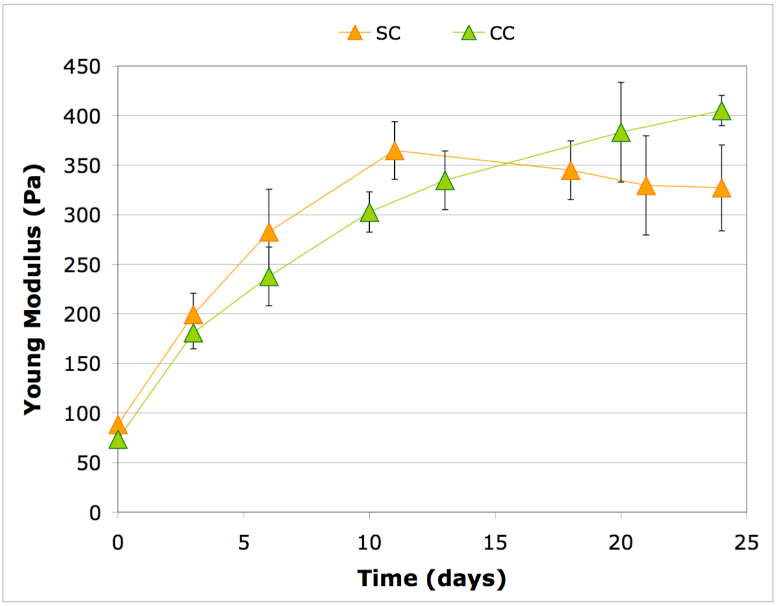


Figure 5.13. Load at 25% strain (top) and Young Modulus (bottom) values for SC and CC cakes during storage.

Regarding oxidation profile, PV values for CC and SC cakes are shown in Fig. 5.14. For both samples, it is noticeable an initial lag-phase, followed by a steeper increase rate of PV. Higher values of PV were measured for CC cake, due to initial higher values of the OG itself (12.71 ± 0.47 meq O_2/kg oil), comparable to those of the fresh CC cake (12.29 ± 0.28 meq O_2/kg oil). This was reasonably due to the use of oxidised phytosterols in the formulation. Moreover, the slope of the solid line is three times higher for CC sample, indicating a more rapid oxidation in presence of organogel. This was confirmed by the analysis of sunflower oil used to produced

OG, that showed a PV of only 3.46 ± 0.22 meq O_2/kg oil. To clarify this point, an OG prepared with a "fresher" batch of plant sterols was analysed, yielding a PV of 11.22 ± 1.40 meq O_2/kg oil, only slightly lower than the value previously found. These compounds seem thus extremely sensible to oxygen (even if stored at 4°C in the dark). It can be also hypothesised that the extraction process (from their natural source) altered phytosterols properties or even that the samples received were old -even though within their shelf life- or kept in unsuitable conditions by the producer.

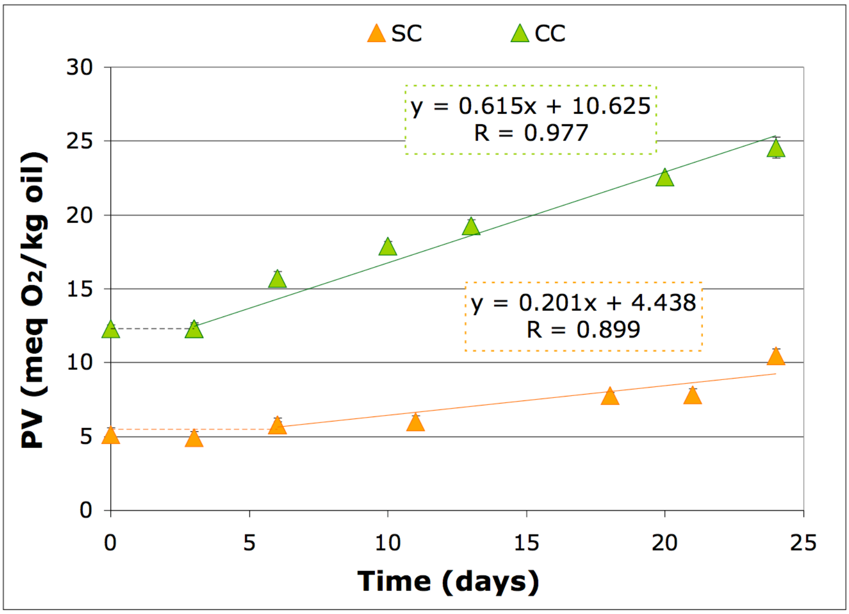


Figure 5.14. PV for CC and SC cakes during storage, expressed as meq O_2/kg oil; the dotted line represent the induction period, while equations in the graph refer to the regression curve (solid line).

Carbonyl compounds analysis, calculated on the basis of the regression line in Fig. 5.16, showed an initial higher value in CC sample, confirming the poor stability of plant sterols (Fig. 5.15). During 25 days storage at 25°C, in a dark environment and in oriented OPP packages, no further secondary oxidation reaction can be observed in both samples. Accordingly, an informal sensory evaluation could not perceive any off-odour.

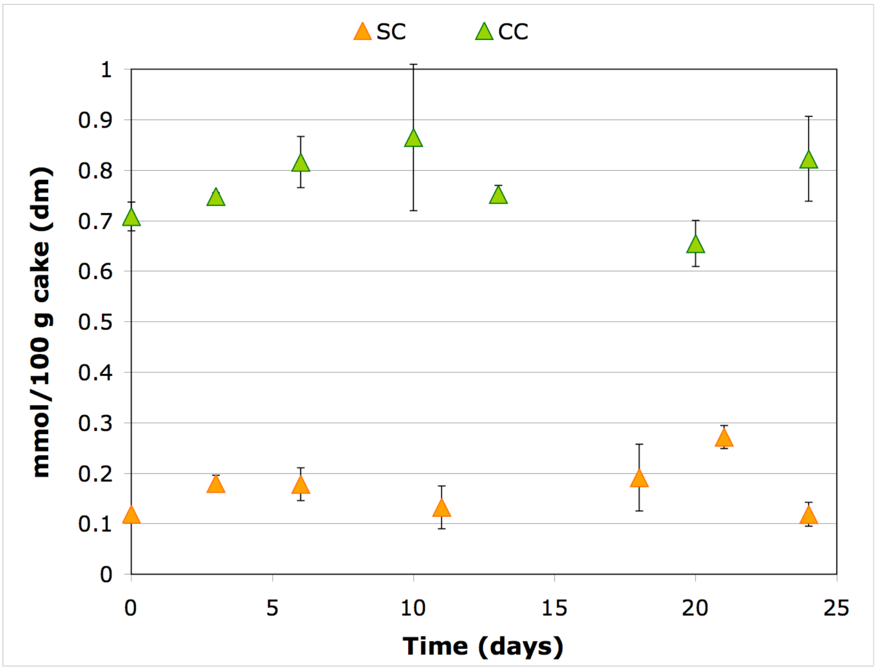


Figure 5.15. Carbonyl compounds values for CC and SC cakes, expressed as mmol of 2,4-decadienal in 100 g of cake (dry basis).

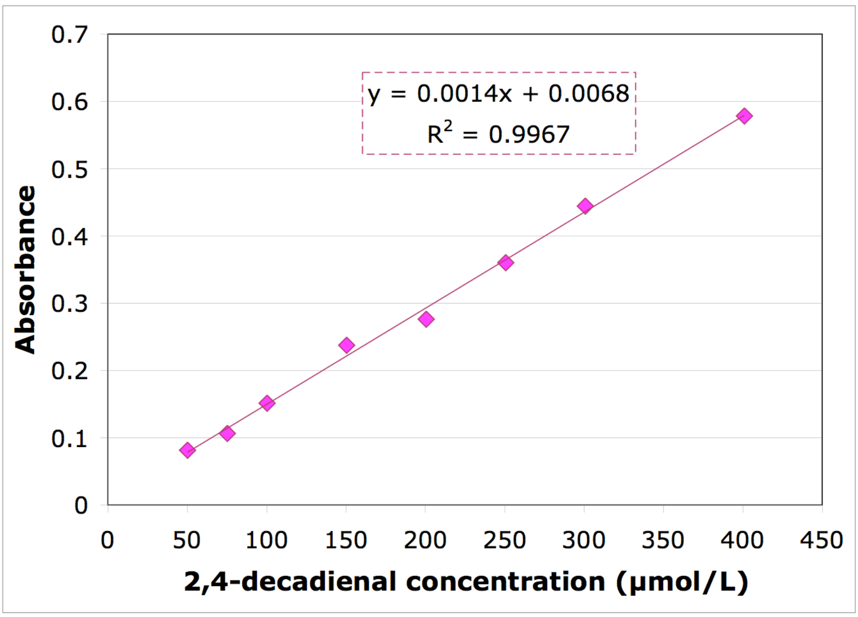


Figure 5.16. Calibration line obtained for carbonyl compound analysis.

5.3. Conclusions

Rheological characteristics of organogels formed using γ-oryzanol + β-sitosterol were affected by the concentration of gelators in sunflower oil, with increasing consistency corresponding to higher concentrations. Even though all the gels considered in the experimental design showed different mechanical spectra, they did not clearly affect cake performances. However, the presence of the structured oil, in comparison to the use of liquid sunflower oil, influenced positively product characteristics, producing cakes with a softer texture. This difference is maintained in the first phase of storage, suggesting that organogels probably interact with cake major components (starch, proteins, water) and inhibit the staling phenomena responsible of its quality depletion. The analysis of oxidation products in fresh and stored cakes highlighted the sensitivity to oxidation of plant sterols used in organogel production, yielding an increase of oxidation indexes of the oil in which they are dissolved.

5.4. References

Bot A, Agterof WGM, 2006, Structuring of edible oils by mixtures of γ-oryzanol with β-sitosterol or related phytosterols. J Am Oil Chem Soc 83:513-521.

Bot A et al., 2008, Fibrils of γ-oryzanol + β-sitosterol in edible oil organogels. J Am Oil Chem Soc 85:1127–1134.

Bot A et al., 2009, Effect of sterol type on structure of tubules in sterol + γ-oryzanol-based organogels. Food Biophys 4:266–272.

Da Pieve S et al., 2011, Effect of monoglyceride organogel structure on cod liver oil stability. Food Res Int 44:2978–2983.

Duffy N et al., 2009, Organogel-based emulsion systems, micro-structural features and impact on in vitro digestion. J Am Oil Chem Soc 86:733-741.

Endo Y et al., 2003, A modified method to estimate total carbonyl compounds in frying oils using 1-butanol as a solvent. J Oleo Sci 52(7):353-358.

Goldstein A, Seetharaman K, 2011, Effect of a novel monoglyceride stabilized oil in water emulsion shortening on cookie properties. Food Res Int 44:1476–1481.

Guillén MD, Uriarte PS, 2012, Aldehydes contained in edible oils of a very different nature after prolonged heating at frying temperature: Presence of toxic oxygenated α, β unsaturated aldehydes. Food Chem 131:915–926.

Hughes NE et al., 2009, Potential food applications of edible oil organogels. Trends Food Sci Technol 20:470-480.

Kumari R et al., 2011, Use of vegetable oils, emulsifiers and hydrocolloids on rheological, fatty acid profile and quality characteristics of pound cake. J Texture Stud 42:377–386.

Manzocco et al., 2012, Use of monoglyceride hydrogel for the production of low fat short dough pastry. Food Chem 132(1):175-180.

Nakano A, Masaki N, 1989, Migration inhibitor for fats and oils for baked cakes and process for producing composite baked cake with the use of the same. US Patent 4855152.

Pernetti M et al., 2007, Structuring of edible oils by alternatives to crystalline fat. Curr Opin Colloid In 12:221–231.

Quìlez J et al., 2003, Bakery products enriched with phytosterol esters, α-tocopherol and β-carotene decrease plasma LDL-cholesterol and maintain plasma β-carotene concentrations in normocholesterolemic men and women. J Nutr 133:3103–3109.

Sanz T et al., 2008, Evaluation of four types of resistant starch in muffin baking performance and relationship with batter rheology. Eur Food Res Technol 227:813–819.

Sawalha H et al., 2011, The influence of concentration and temperature on the formation of γ-oryzanol+β-sitosterol tubules in edible oil organogels. Food Biophys 6:20–25.

Shelke K et al., 1990, The dynamics of cake baking as studied by a combination of viscosimetry and electrical resistance oven heating. Cereal Chem 67(6):575-580.

Uzuki R et al., 2009, Estimation of total carbonyl compounds in roasted and dried foods by the tentative method using 1-butanol as a solvent. J Oleo Sci 58(1):23-5.

6. Conclusions and future perspectives

The role of lipids in a baked product appeared to be complex and variable, as many factors have to be taken into consideration. From our studies, the role of fat melting characteristics and fat quantity on quality attributes of a model cake has been highlighted, as well as the possibility to predict cake behaviour through empirical modelling. In particular, the possibility to use more unsaturated oils without decreasing, but rather improving, the quality features of the cake has been demonstrated. Since a baked product is a complex system, composed of macro-components such as flour, eggs, sugar, water, a deeper study on the interaction of the fatty fraction with the other ingredients could help in understanding the role of lipids in the whole batter matrix and in the baked product, especially at a micro-structural level.

Regarding the employment of novel structured fatty matrix in the form of an organogel, more research is needed, both on the theoretical and applicative side. The studies reported in the literature mainly concern the production and characterization of relatively high concentrated sterols organogels. having consistency unfit for food applications, at least in bakery. Thus, the technological gaps in organogel production and storage conditions should be filled, to obtain a standardized production able to give reliable results in final cakes. Moreover, also considering the results obtained by Rush et al. (2009), who found a tempered postprandial insulin response after a mono-glyceride gel consumption, thus suggesting structure-dependent metabolic response, a monitoring of anti-cholesterolemic and insulin effects of phytosterols involved in an organogel could be of interest, in order to establish if the structuring of the oil has a metabolic benefit, in addition to the potential improvement of texture of the final products. In addition to this, it is relevant what is reported by Quìlez et al. (2003), who demonstrated that ingestion of phytosterols has not to be simultaneous to that of cholesterol; on the contrary, the intake of bakery products between meals could stimulate production of endogenous cholesterol.

Finally, the use of different types of baked product as models could be considered in a further phase of structured fat application. For instance, in naturally leavened cake/bread, the retention of air during proofing is strongly influenced by fat physical state and thus organogels could have a role in enhancing textural and volume properties.

References

Quìlez J et al., 2003, Bakery products enriched with phytosterol esters, α-tocopherol and β-carotene decrease plasma LDL-cholesterol and maintain plasma β-carotene concentrations in normocholesterolemic men and women. J Nutr 133:3103–3109.

Rush JWE et al., 2009, Acute metabolic responses to butter, margarine, and a monoglyceride gel-structured spread. Food Res Int 42:1034-1039.