



# Article Hydroglycerolic Solvent and Ultrasonication Pretreatment: A Green Blend for High-Efficiency Extraction of Salvia fruticosa Polyphenols

# Spyros Grigorakis<sup>1,2</sup>, Abedalghani Halahlah<sup>1</sup> and Dimitris P. Makris<sup>3,\*</sup>

- <sup>1</sup> Department of Food Science & Nutrition, School of Environment, University of the Aegean, Mitr. Ioakim Street, 81400 Myrina, Greece; grigorakis@maich.gr (S.G.); abedh1893@gmail.com (A.H.)
- <sup>2</sup> Food Quality & Chemistry of Natural Products, Mediterranean Agronomic Institute of Chania (M.A.I.Ch.), International Centre for Advanced Mediterranean Agronomic Studies (CIHEAM), P.O. Box 85, 73100 Chania, Greece
- <sup>3</sup> Green Processes & Biorefinery Group, Department of Food Science & Nutrition, School of Agricultural Sciences, University of Thessaly, N. Temponera Street, 43100 Karditsa, Greece
- \* Correspondence: dimitrismakris@uth.gr

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**Abstract:** *Salvia fruticosa* Miller, also known as Cretan or Greek sage, is a medicinal plant with significant biological properties, which are largely ascribed to its polyphenolic composition, but there is to-date a scarcity of green and sustainable processes for efficient polyphenol extraction from this plant. The objective of this study was the implementation of an extraction process that would combine a green solvent based on glycerol, a biodiesel industry by-product, and ultrasonication pretreatment. Ultrasonication for 40 min followed by stirred-tank extraction was shown to provide significantly higher total polyphenol yield than mere stirred-tank extraction, while kinetics indicated 50 °C as the most favorable temperature, with the yield being 92 mg gallic acid equivalents (GAE) per g dry mass. Comparison of this method with a previously developed one that used methyl  $\beta$ -cyclodextrin revealed that the extracts obtained had similar antioxidant activity, and yield in major polyphenols including luteolin 7-*O*-glucuronide and rosmarinic acid was virtually equal. The current process is proposed as a sustainable and effective methodology for the generation of polyphenol-enriched extracts from *S. fruticosa*, which could be used as effective food antioxidants/antimicrobials and/or cosmetic constituents.

**Keywords:** antioxidants; extraction kinetics; glycerol; green extraction; polyphenols; *Salvia fruticosa*; ultrasonication

# 1. Introduction

Consumer awareness and demand for functional food ingredients and health-promoting supplements have boosted a great development in botanical research [1] regarding new product design and enabled the launch of a wide spectrum of formulations [2] and cosmetic ingredients [3]. *Salvia* is a genus of the Lamiaceae family and embraces more than 800 species worldwide [4]. Numerous *Salvia* species are regarded as plants with significant bioactive properties, and they have been used for centuries as folk pharmaceuticals in many countries [5]. The therapeutic potential of *Salvia* plants has been largely ascribed to principal substances, including phenolic acids and terpenoids, but in *Salvia* species a large variety of flavonoids may also occur [6,7]. *S. fruticosa*, otherwise known as *S. triloba* (family: Lamiaceae), is a sage species native to the island of Crete (southern Greece). It is regarded as a plant of great biological value [8–10], yet there is to-date no green extraction process developed for

the generation of polyphenol-enriched extracts with high antioxidant activity, which could be used as active ingredients in food supplements, cosmetics, and pharmaceuticals.

The development of green processes aimed at producing polyphenol-enriched extracts from botanicals has been of great concern to researchers, and a number of eco-friendly, reproducible, low-cost and low-energy techniques are now acknowledged as more effective alternatives to traditional extraction methodologies [11]. However, one of the major ways to comply with the principles of green chemistry is to reduce the use of toxic, volatile organic solvents, and to encourage their replacement by novel, environmentally friendly liquids. In this framework, the selection of an appropriate solvent is of paramount importance to the sustainable character of an extraction method. The ideal candidate should display high extraction efficiency, low or no toxicity, low price, and availability, and it should be produced from recyclable resources, as opposed to petroleum-derived solvents [12,13].

Glycerol (glycerine or 1,2,3-propanetriol) is a bio-liquid considered a by-product of the biodiesel industry, which is generated at about 10% by weight of the starting material (triacylglycerols) [14]. Although glycerol is a well-established sustainable solvent for various chemical processes [14–16], its use as a green solvent for effective polyphenol extraction has been introduced only within the last six years [17]. Ever since, several studies have demonstrated glycerol/water mixtures as high-performing extraction media for polyphenol recovery from various plant matrices [18–26]. This being the case, the current project was undertaken to thoroughly examine the extraction of *S. fruticosa* polyphenolic antioxidants using green glycerol/water mixtures, combined with ultrasonication pretreatment. Major polyphenolic phytochemicals in the optimally produced extracts were tentatively identified with liquid chromatography-diode array-tandem mass spectrometry (LC/MS/MS).

# 2. Materials and Methods

#### 2.1. Chemicals and Reagents

Methyl  $\beta$ -cyclodextrin chlorogenic acid ( $\geq$ 95%), luteolin 7-*O*-glucoside, and rosmarinic acid (96%) were from Sigma (St. Louis, MO, USA). Glycerol (99%) and ethanol (99.8%) were from Acros Organics (Geel, Belgium). Aluminium chloride hexahydrate and sodium acetate trihydrate were from Penta (Prague, Czeck Republic). 2,4,6-Tripyridyl-*s*-triazine (TPTZ, 99%), Folin–Ciocalteu reagent and ferric chloride hexahydrate were from Fluka (Steinheim, Germany). Anhydrous sodium carbonate was from Carlo Erba Reactifs (Val de Reuil, France). 2,2-Diphenyl-1-picrylhydrazyl radical (DPPH), gallic acid, ascorbic acid, and rutin (quercetin 3-*O*-rutinoside) were from Aldrich (Steinheim, Germany). The solvents used for chromatographic analyses were HPLC grade.

#### 2.2. Plant Material—Handling and Preparation

*Salvia fruticosa*, also known as *Salvia triloba* (Cretan or Greek sage), was purchased from a local store of certified botanicals (Chania, Greece) and further identified by the Mediterranean Plant Conservation Center (Chania, Greece). The material (250 g) consisted of dried aerial parts of the plant, and it was received in air-tight plastic packaging. Upon receipt, it was stored in a chamber of low humidity, in the dark, for no longer than a week. An amount of approximately 50 g of material was placed in a domestic blender, ground, and then sieved to yield a feed with an average particle diameter of 1.28 mm. This feed was transferred into plastic containers, stored at 7 °C, and used in all procedures.

#### 2.3. Ultrasonication Pretreatment

An exact amount of 1 g of feed was mixed with 25 mL of solvent in a 50-mL round-bottom flask, and ultrasonicated in an ultrasonication bath (Sonorex Bandeline, Berlin, Germany) with the following settings: power, 120 W; acoustic energy density, 120 W  $L^{-1}$ ; frequency, 100 Hz; temperature, 50 °C. Ultrasonication was performed for 5, 10, 20, 30, and 40 min.

#### 2.4. Batch Stirred-Tank Solid–Liquid Extraction

This procedure was implemented after ultrasonication pretreatment. The solvents tested were deionized water as well as hydroglycerolic mixtures with glycerol proportions of 20%, 40%, 60%, and 80% (w/v). Extraction was undertaken in an oil bath at a constant temperature of 50 °C with stirring at 700 rpm for 150 min. Temperature regulation and stirring were provided by a heating magnetic stirrer (VELP Scientifica, Bohemia, NY, USA). After extraction, each sample was centrifuged at 10,000× *g* for 10 min, and the supernatant was used for all analyses performed afterwards.

#### 2.5. Extraction Kinetics and Temperature Effects

Kinetics was examined by implementing the model previously proposed [27]:

$$Y_{\text{TP}(t)} = Y_{\text{TP}(0)} + \frac{Y_{\text{TP}(s)}t}{t_{0.5} + t}$$
(1)

 $Y_{\text{TP(t)}}$  is the yield in total polyphenols at any time, t,  $Y_{\text{TP(s)}}$  is the yield in total polyphenols at saturation (equilibrium),  $Y_{\text{TP(0)}}$  is a fitting parameter, and  $t_{0.5}$  represents the time at which  $Y_{\text{TP(t)}} = \frac{Y_{\text{TP(s)}}}{2}$ . According to this model, the initial extraction rate, h, and the second-order extraction rate, k, are given as:

$$h = \frac{\Upsilon_{\text{TP(s)}}}{t_{0.5}} \tag{2}$$

$$k = \frac{1}{Y_{\text{TP(s)}} t_{0.5}}$$
(3)

The effect of temperature on *k* was illustrated by performing non-linear regression between *k* and *T*. This correlation could be very effectively described using an exponential model [28]:

$$k = k_0 + ae^{-bT} \tag{4}$$

Terms *k* and  $k_0$  correspond to the second-order extraction rate and a pre-exponential factor. Determination of the activation energy (*E<sub>a</sub>*) of the process was computed as follows [29]:

$$ln(\frac{k}{k_{ref}}) = \left(-\frac{E_a}{R}\right) \left(\frac{1}{T} - \frac{1}{T_{ref}}\right)$$
(5)

 $T_{ref}$  and T are the reference temperature (K) and the temperature at which kinetics was traced,  $k_{ref}$  and k are the corresponding second-order extraction rate constants, R is the universal gas constant (8.314 J K<sup>-1</sup> mol<sup>-1</sup>), and  $E_a$  is the activation energy (J mol<sup>-1</sup>).

#### 2.6. Determinations

Total polyphenol analysis was performed using a previously described Folin–Ciocalteu methodology [30]. Yield in total polyphenols ( $Y_{TP}$ ) was given as mg gallic acid equivalents (GAE) per g dry mass (dm). Likewise, total flavonoids were determined with CH<sub>3</sub>COONa/AlCl<sub>3</sub> reagent and given as mg rutin equivalents (RtE) per g dm [31]. The antiradical activity ( $A_{AR}$ ) and the ferric-reducing power ( $P_R$ ) were estimated as reported elsewhere [27], and results were expressed as µmol DPPH per g dm and µmol ascorbic acid equivalents (AAE) per g dm, respectively.

#### 2.7. Chromatographic Determinations

Analyses were performed with a FinniganMAT P4000 pump equipped with a UV6000LP diode array detector (Thermo Scientific, Waltham, MA, USA), and a TSQ Quantum Access LC/MS/MS, coupled with a Surveyor pump (Thermo Scientific, Walltham, MA, USA), controlled by XCalibur 2.1, TSQ 2.1 software. Chromatography was run on a Superspher RP-18 column, 125 mm  $\times$  2 mm, 4  $\mu$ m,

maintained at 40 °C, employing 10-µL injections. The eluents used were (A) 2.5% acetic acid and (B) methanol, at a flow rate of 0.3 mL min<sup>-1</sup>. The elution program implemented was as follows: 0 min, 100% A; 22 min, 65% A; 32 min, 65% A; 60 min, 0% A; 65 min, 0% A. Mass spectra were acquired with negative ionization, with the following settings: sheath gas pressure 30 mTorr; collision pressure 1.5 mTorr; capillary temperature 300 °C; auxiliary gas pressure 15 mTorr. Quantification was carried out with external standard methodology, using a calibration curve of chlorogenic acid (50–1500 µg L<sup>-1</sup>,  $R^2 = 0.9986$ ), rosmarinic acid (50–3000 µg L<sup>-1</sup>,  $R^2 = 0.9985$ ), and luteolin 7-*O*-glucoside (5–1500 µg L<sup>-1</sup>,  $R^2 = 0.9982$ ). Standard solutions were prepared in HPLC grade methanol and stored in the freezer.

# 2.8. Statistical Analysis

Two repetitions were performed for each extraction and pretreatment process, and each determination was carried out in triplicate. Values given represent averages ± standard deviation. Linear correlations and kinetic model fitting were accomplished with SigmaPlot<sup>™</sup> 12.5 (Systat Software Inc., San Jose, CA, USA). Distribution analysis, at least at a 95% significance level, was done with JMP<sup>™</sup> Pro 13 (SAS, Cary, NC, USA).

## 3. Results and Discussion

# 3.1. Effect of Solvent Composition

Earlier examinations on the effect of glycerol/water proportion on polyphenol extraction employed rather low-glycerol mixtures, with glycerol percentage varying from 3.6% [32] to 9.3%–10% (w/v) [10,30,33]. However, later investigations showed that polyphenol extraction yield may increase linearly from 5% (w/v) onwards, the optimum being 20% (w/v) [12]. More thorough, single-factor studies including a wider range of glycerol/water proportions demonstrated that the optimum glycerol percentage may lie between 70% [34] and 90% (w/v) [35]. Optimum levels as high as 90% (w/v) have also been found by implementing response surface methodology [36,37]. Yet, significantly lower optimal levels of 20% (w/v) [16] and 32.5% (w/v) [19] have also been reported. Therefore, testing of the optimum glycerol/water proportion ( $C_{GL}$ ) was performed over a range varying from 0% (deionized water) to 80% (w/v) glycerol (Figure 1). Proportions > 80% were not considered because high-glycerol mixtures are very viscous and particularly problematic to handle. The assay performed indicated that a mixture with  $C_{GL}$  of 60% (w/v) provided significantly higher (p < 0.05) total polyphenol extraction yield ( $Y_{TP}$ ), which reached 66.92 ± 1.67 mg GAE g<sup>-1</sup> dm. Thus, this solution was employed to perform further experimentation.



**Figure 1.** Assay performed to identify the optimum concentration of glycerol ( $C_{GL}$ ) for *S. fruticosa* polyphenol extraction. Bars indicate standard deviation. Asterisk (\*) denotes a statistically different value (p < 0.05).

#### 3.2. Effect of Ultrasonication Pretreatment

The integration of ultrasonication as a pretreatment stage has been recently appraised, with the ultrasonication time considered ranging from 5 to 40 min [27,31,38]. On the basis of these data, the ultrasonication effect was tested within this time frame (Figure 2). As preliminary experiments showed that starting from 25 °C (room temperature), there may be an increase in temperature up to 45 °C after 40 min of ultrasonication, the assay temperature was set at 50 °C to eliminate variations arising from the ultrasonication effect. An ultrasonication temperature higher than 50 °C was not preferred to maximize the sonochemical benefit, in line with previous observations [39]. It has been proposed that ultrasound-assisted polyphenol extraction is not favored at temperatures higher than 50 °C because the collapse of cavitation bubbles, generated as a result of ultrasound irradiation, is more effective in low-vapor pressure solvents (such as glycerol/water mixtures) at lower temperatures. The collapse of cavitation bubbles is considered to enhance solute extraction because there is a release of a large amount of energy as a result of high temperature/high pressure involved in such a process. This in turn may contribute to disrupting the integrity of the solid particles, provoking an increased entrainment of solute in the liquid phase [40].



**Figure 2.** The effect of ultrasonication pretreatment on  $Y_{\text{TP}}$ , using 60% (w/v) glycerol/water mixture. Ultrasonication and subsequent stirred-tank extraction were performed at 50 °C. Bars indicate standard deviation. Asterisk (\*) denotes a statistically different value (p < 0.05).

Changes in  $Y_{\text{TP}}$  displayed an increasing progression as a function of ultrasonication time, but significantly higher  $Y_{\text{TP}}$  (p < 0.05) was achieved with 40-min ultrasonication pretreatment. From 40 to 60 min, the yields achieved with ultrasonication alone were very similar (about 7% difference), whereas ultrasonication > 60 min resulted in a slight decline (about 8%) of the yield. The combination of pretreatment and a subsequent stirred-tank extraction afforded a  $Y_{\text{TP}}$  of 79.12 ± 1.98 mg GAE g<sup>-1</sup> dm, which was 15% higher than that attained without pretreatment. This finding stressed emphatically the importance of ultrasonication pretreatment in boosting extraction efficiency. It is to be underlined that mere ultrasonication for 40 min gave a  $Y_{\text{TP}}$  of only 41.10 ± 1.03 mg GAE g<sup>-1</sup> dm, which represented approximately just 52% of the  $Y_{\text{TP}}$  reached by combining ultrasonication pretreatment and stirred-tank extraction. This fact clearly demonstrated that ultrasonication was not effective as a standalone extraction mode. This was in absolute accordance with earlier results from similar studies on grape pomace [41] and elderflowers [31].

#### 3.3. Extraction Kinetics and the Effect of Temperature

Previous studies showed that polyphenol extraction with hydroglycerolic solvents is significantly affected within a temperature spectrum ranging from 50 to 80 °C [35-37]. Thus, kinetics was traced at 50, 60, 70, and 80 °C to thoroughly investigate the influence of temperature (Figure 3).



**Figure 3.** Kinetics of *S. fruticosa* polyphenol extraction, using 60% (w/v) glycerol/water mixture. Samples were pretreated with ultrasounds prior to stirred-tank extraction for 40 min at 50 °C.

Switching *T* from 50 to 80 °C resulted in progressive acceleration of extraction, as indicated by the increase in the second-order extraction rate, *k*, from 0.369 to 1.370 g mg<sup>-1</sup> min<sup>-1</sup> (Table 1). The pattern was similar for the initial extraction rate, *h*, which increased from 1.838 to 5.194 mg g<sup>-1</sup> min<sup>-1</sup>. The correlation of *k* with *T* was portrayed by an exponential model, as previously proposed [28], which showed excellent adjustment to the experimental data (Figure 4). The fitting parameter b equaled 0.0765, and it was significantly higher than 0.0238 determined for aqueous extraction of *S. fruticosa* polyphenols using methyl  $\beta$ -cyclodextrin [42]. This finding suggested that the stirred-tank extraction using hydroglycerolic solvent was more energy-demanding.

<i>T</i> (°C)	Kinetic Parameters				
	k (×10 <sup>-3</sup> ) (g mg <sup>-1</sup> min <sup>-1</sup> )	h (mg g <sup>-1</sup> min <sup>-1</sup> )	$Y_{ m TP(s)}$ (mg GAE g <sup>-1</sup> )	t <sub>0.5</sub> (min)	
50	0.369	1.838	92.00	50.06	
60	0.528	2.400	89.27	37.19	
70	0.768	3.278	87.91	26.82	
80	1.370	5.194	84.53	16.27	

**Table 1.** Values of kinetic parameters determined for the extraction of *S. fruticosa* polyphenols, using 60% (w/v) glycerol/water mixture.

To verify this assumption, the activation energy,  $E_a$ , was estimated using Equation (5), and the value found was 47.67 kJ mol<sup>-1</sup>. This barrier was significantly higher than 5.64 kJ mol<sup>-1</sup> determined for methyl  $\beta$ -cyclodextrin-assisted extraction [42], which confirmed the higher energy requirement. However, there is an important detail that should be taken into account. In this study, stirred-tank polyphenol extraction was applied after an ultrasonication regime of 40 min, during which a significant amount of readily extractable polyphenols was recovered (Figure 2). Thus, the  $E_a$  determined represented the barrier required to extract the residual and harder-to-extract polyphenols. Such a case has been recently investigated, and it was demonstrated that the  $E_a$  required to extract polyphenols from plant material after an ultrasonication pretreatment stage was higher than that corresponding to stirred-tank extraction without pretreatment [38].



Figure 4. Non-linear regression between second-order extraction rate constant, k, and temperature, T.

YTP(s) displayed a declining trend and while its value was 92.00 mg GAE g–1 dm at 50 °C, it dropped to 84.53 mg GAE g–1 dm at 80 °C. However, distribution analysis indicated that this difference was non-significant (p > 0.05). This phenomenon has been previously reported for polyphenol extraction from onion solid wastes with hydroglycerolic mixture and attributed to polyphenol thermal instability [35]. In general, increases in T favor higher YTP because higher T usually entails higher polyphenol diffusion and solubility [43,44]. On the other hand, polyphenols are thermolabile molecules and in several cases T higher than 50 °C did not contribute to attaining increased YTP [45–47]. On the other hand, in a previous examination on cyclodextrin-aided aqueous extraction of S. fruticosa polyphenols, it was shown that polyphenol extraction yield increased constantly by raising T from 40 to 80 °C [42]. Such an effect could be attributed to the protective role of cyclodextrins against thermal degradation of polyphenols, as demonstrated by earlier studies [48].

#### 3.4. Antioxidant Properties and Polyphenolic Profile

To test the effectiveness of the method developed, a comparison was carried out with another green method established previously [42], based on characteristics pertaining to polyphenol extraction yield and antioxidant activity (Table 2). Extraction with m- $\beta$ -CD at 80 °C was proven more efficient with respect to  $Y_{\text{TP}}$ , as it afforded 108.14 ± 2.70 mg GAE g<sup>-1</sup> dm, as opposed to extraction with the hydroglycerolic solvent, which gave by 22.5% lower  $Y_{\text{TP}}$  (83.86 ± 2.10 mg GAE g<sup>-1</sup> dm). On the other hand, differences in  $Y_{\text{TFn}}$  and  $A_{\text{AR}}$  were marginal and non-significant (p > 0.05). On the contrary, the hydroglycerolic extract exhibited significantly higher  $P_{\text{R}}$ . The LC/DAD/MS/MS enabled the tentative identification of a series of polyphenolic phytochemicals (Figure 5, Table 3), based on spectral data reported earlier [42,49].

**Table 2.** Comparative evaluation of *S. fruticosa* extracts obtained with 60% (w/v) glycerol/water (GL) and methyl  $\beta$ -cyclodextrin (m- $\beta$ -CD).

Extract	$Y_{\mathrm{TP}}$ (mg GAE g <sup>-1</sup> dm)	$Y_{\rm TFn}$ (mg RtE g <sup>-1</sup> dm)	$A_{ m AR}$ (µmol DPPH g <sup>-1</sup> dm)	P <sub>R</sub> (μmol AAE g <sup>-1</sup> dm)
m-β-CD	$108.14 \pm 2.70$	$53.62 \pm 1.61$	$820.93 \pm 16.42$	$590.66 \pm 14.77$
GL	$83.86 \pm 2.10$	$51.46 \pm 2.57$	$817.58 \pm 8.18$	$709.12 \pm 17.73$



**Figure 5.** Typical HPLC traces of *S. fruticosa* polyphenol extracts, monitored at 330 nm. Extracts were produced with 60% (*w*/*v*) glycerol/water and methyl  $\beta$ -cyclodextrin (m- $\beta$ -CD) at 80 °C. Peak assignment: 1, chlorogenic acid; 2, unknown; 3, unknown; 4, 6-hydroxy luteolin 7-O-glucoside; 5, luteolin 7-O-glucuronide; 6, luteolin 7-O-rutinoside; 7, 6-methoxyluteolin 7-O-glucoside (nepitrin); 8, rosmarinic acid; 9, apigenin 7-O-glucuronide; 10, 6-methoxyluteolin derivative; 11, 6-methoxyluteolin derivative.

Table 3. Spectral	l attributes used to	tentatively identit	fy major poly	phenols in S.	fruticosa extracts.
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No	Rt (min)	UV-Vis ( $\lambda_{max}$ )	[M – H] <sup>+</sup> (m/z)	Other Ions (m/z)	Tentative Identity
1	15.77	246, 318	353	179	Chlorogenic acid
2	17.40	248, 318	253	-	Unknown
3	21.00	270, 340	593	-	Unknown
4	23.57	280, 344	477	301	6-Hydroxy luteolin 7-O-glucoside
5	25.78	256, 352	461	285	Luteolin 7-O-glucuronide
6	27.12	258, 348	593	285	Luteolin 7-O-rutinoside
7	27.90	270, 352	491	299	6-Methoxyluteolin 7- <i>O</i> -glucoside (nepitrin)
8	28.65	246, 316	359	161	Rosmarinic acid
9	29.55	264, 346	445	269	Apigenin 7-O-glucuronide
10	32.15	270, 352	475	299	6-Methoxyluteolin derivative
11	32.82	274, 332	461	299, 283	6-Methoxyluteolin derivative

In order to better demonstrate the extraction capacity of the hydroglycerolic solvent, three major constituents were considered for quantitative analysis, namely chlorogenic acid, luteolin 7-O-glucuronide, and rosmarinic acid. Other minor polyphenols that were tentatively identified in the extracts were not considered because they occurred at significantly lower levels and differences

in their content might not be indicative for reliably assessing solvent extraction capacity. The results from the quantitative assay are analytically presented in Table 4. Compared to m- $\beta$ -CD, extraction with the hydroglycerolic solvent gave a 37.5% higher yield in chlorogenic acid and a 0.57% higher yield in rosmarinic acid, but a 20.8% lower yield in luteolin 7-*O*-glucuronide. Overall, the difference in yield was only 7.4%, indicating that both extracting media performed equally in the recovery of major *S. fruticosa* phytochemicals.

**Table 4.** Quantitative information on major polyphenols considered to compare *S. fruticosa* extracts produced with 60% (w/v) glycerol/water (GL) and methyl  $\beta$ -cyclodextrin (m- $\beta$ -CD).

Compound		Yield (mg g <sup>-1</sup> dm) $\pm$ sd	
	m-β-CD	GL	% Difference
Chlorogenic acid	$0.15 \pm 0.02$	$0.24 \pm 0.05$	37.5
Luteolin 7-O-glucuronide	$6.96 \pm 1.12$	$5.51 \pm 1.57$	20.8
Rosmarinic acid	$10.57 \pm 1.37$	$10.63 \pm 0.98$	0.57
Sum	17.68	16.38	7.4

# 4. Conclusions

The approach attempted in this study aimed at (i) utilizing glycerol, a by-product of the biodiesel industry, as a green and non-volatile solvent, and (ii) integrating ultrasonication pretreatment as a step central to increasing the efficiency of the extraction methodology used. The combination of such a pretreatment with a hydroglycerolic solvent provided a high-efficiency extraction for *S. fruticosa* polyphenols. The kinetics showed that extraction at 50 °C may be the most favorable, and thus this methodology may also be energy-effective, a fact that significantly adds to the sustainable profile of the process. A prospect of this investigation would be future studies focusing on scale-up and application of hydroglycerolic extracts of *S. fruticosa* as effective food antioxidants/antimicrobials and/or cosmetic constituents. This would pave the way for the implementation of the process on an industrial scale.

**Author Contributions:** S.G. and A.H. carried out the experimentation and handled the raw data. D.P.M. conceived the idea, designed the experiment, performed statistics, handled the data, and wrote the paper. All authors have read and agreed to the published version of the manuscript.

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