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# Oil yields, protein contents, and cost of manufacturing of oil obtained from different hybrids and sowing dates of canola



Tássia C. Confortin<sup>a,e</sup>, Izelmar Todero<sup>a</sup>, Luciana Luft<sup>b</sup>, Gustavo A. Ugalde<sup>a</sup>, Marcio A. Mazutti<sup>a,b</sup>, Zanandra B. Oliveira<sup>c</sup>, Eduardo L. Bottega<sup>c</sup>, Alberto E. Knies<sup>c,d</sup>, Giovani L. Zabot<sup>e</sup>, Marcus V. Tres<sup>e,\*</sup>

- <sup>a</sup> Department of Agricultural Engineering, Federal University of Santa Maria, 1000 Roraima Av., Santa Maria, 97105-900, Brazil
- <sup>b</sup> Department of Chemical Engineering, Federal University of Santa Maria, 1000 Roraima Av., Santa Maria, 97105-900, Brazil
- c Grupo de Pesquisa em Ambiência e Bioclimatologia (GPAB), Federal University of Santa Maria, 1958 Presidente Vargas Av., Cachoeira do Sul, 96506-302, Brazil
- <sup>d</sup> State University of Rio Grande do Sul, 1040 Sete de Setembro St., Center DC, Cachoeira do Sul, RS, 96508-010, Brazil
- <sup>e</sup> Laboratory of Agroindustrial Processes Engineering (LAPE), Federal University of Santa Maria, 1040 Sete de Setembro St., Center DC, Cachoeira do Sul, RS, 96508-010, Brazil

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#### ABSTRACT

Three hybrids of canola were cultivated in Brazil submitted to three sowing dates in 2018: April 21 (early autumn), May 20 (mid-autumn), and June 24 (late autumn). The samples were harvested and the grain yields were measured. Thereafter, canola oil was extracted by Soxhlet with n-Hexane and by supercritical  $CO_2$  (SFE-CO<sub>2</sub>) following a completely randomized design. The following responses were evaluated: oil yields, oil composition, protein contents, and cost of manufacturing of oil. The highest grain yield was 2807 kg ha<sup>-1</sup> and the highest oil yield was 34.7 wt.%. The main fatty acids identified in the oil were oleic acid (46.4–57.5 wt.%), linoleic acid (10.6–15.2 wt.%), and  $\alpha$ -linolenic acid (5.8–8.1 wt.%). The highest protein content in the solid coproducts was 38.2 wt.%. Furthermore, the cost of manufacturing (COM) of oil was simulated for a pilot/industrial scale dedicated to processing 180 kg of seeds per batch. The COM of oil ranged from US\$ 7.60 kg<sup>-1</sup> to US\$ 1.196 kg<sup>-1</sup> (SFE-CO<sub>2</sub>) and from US\$ 1.13 kg<sup>-1</sup> to US\$ 1.47 kg<sup>-1</sup> (Soxhlet), respectively. After an integrated evaluation of technological and economic responses, the hybrid Hyola 61 sown on May 20 seems to be the most suitable condition for being cultivated by farmers and for being processed by food-related industries.

## 1. Introduction

Canola (*Brassica napus* L.) belongs to the Brassicaceae (Cruciferae) family [1]. It is an oleaginous crop that was developed from the genetic improvement of rapeseed (*Brassica napus*) [2]. Canola is one of the most important oil crops growing worldwide [3], being responsible for 16% of vegetable oil production [4] and ranking third in the world in oil production, behind only soybean oil and palm oil [5,6]. Unlike rapeseed, canola bran can be used for animal feed because it has a low content of erucic acid [7,1]. On average (mass basis), canola bran yields approximately 37% crude protein, 10% crude fiber, 0.6% calcium, 0.3% available phosphorus, 2.0% lysine, 0.8% methionine, 1.6% methionine + cysteine, and an energy of approximately 7118 kJ kg <sup>-1</sup> [7].

In Brazil, canola crop is an alternative for farmers for the autumn and winter seasons, with a special interest in crop rotation systems. Canola provides good soil decomposition and good protection because it has deep roots [8]. According to the most recent data from CONAB [9], the cultivated area in Brazil (especially in the South region) in the last few years is approximately 44,000 ha and the production is approximately 55,000 tons. Canola is one of the major oilseeds used for edible oil production [10] and it is considered a potential raw material for biodiesel production [11]. In Brazil, the amount of oil and protein can reach up to 40 wt.% and 38 wt.%, respectively [1].

The productive performance of canola is influenced by the hybrid and sowing date. The suitable sowing date is composed of a set of environmental factors that influence the grain yields, plant architecture, and development. The need for species and the availability of environmental resources are some factors that must be taken into account when choosing a particular sowing date. The likely risks associated with canola cultivation should be identified, thus allowing the reduction of risks of low productivity and low oil content. Hybrids as Hyola 61, 433 and 571 are being tested because they are presenting promising

E-mail address: marcus.tres@ufsm.br (M.V. Tres).

<sup>\*</sup> Corresponding author.

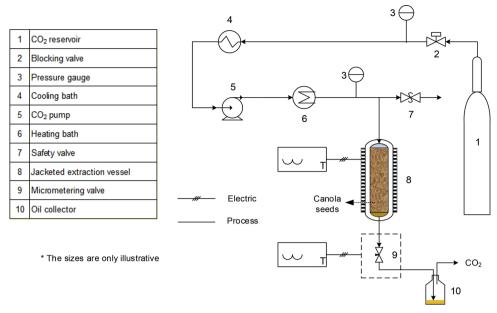


Fig. 1. Experimental apparatus for supercritical CO<sub>2</sub> extraction.

characteristics in terms of higher security to the production without additional cost to the producer [1]. In this trend, the performance of the cultivation of canola should be assessed by other responses, as the protein content and oil yields.

Organic solvents are very efficient for extracting oil from oilseeds, but the presence of solvent residues in products is still a challenge [12]. Another method that can be used for the extraction of oilseeds is the supercritical fluid extraction (SFE) with carbon dioxide (SFE-CO<sub>2</sub>), which this solvent is non-toxic, non-flammable and can be used under mild conditions of temperature [13]. SFE using supercritical carbon dioxide (SC-CO<sub>2</sub>) has been accepted as the green technology for extracting various valuable food/non-food ingredients from the natural products and pharmaceutical products, among others [14,15]. In a biological system, the extraction of organic compounds is also very important because several metabolites can be extracted, including those found in sludge [16]. Furthermore, the processes can concentrate other compounds, as proteins [16].

Based on this context, one alternative is to use a conventional organic solvent (n-Hexane, for example) and  $\mathrm{CO}_2$  under supercritical conditions to evaluate the extraction of canola oil. One of the main factors that determine the nutritional quality of canola oil is based on the composition of fatty acids, especially oleic acid [1]. This monounsaturated fatty acid is known for its reducing effect on blood sugar levels and heart protection [17]. The amount of fatty acids can be different depending on the hybrids and sowing dates [18].

An effort of processing canola seeds on the technological viewpoint should be accompanied by an economic approach. The typical responses of economic evaluation are the cost of manufacturing (COM) and the percent contribution of the main itemized costs (e. g., raw materials, fixed capital investment, operational labor, and utilities) [19]. Therefore, the objective of this study was to assess the grain yields, protein contents, oil yields, oil composition, and COM from three different canola hybrids submitted to three different sowing dates (early autumn, mid-autumn, and late autumn).

# 2. Materials and methods

#### 2.1. Sample

Canola seeds were acquired from Embrapa Trigo (Passo Fundo, RS, Brazil), which were previously treated and tested. They presented

germination power above 95% for all times and hybrids used. The cultivation was carried out in the experimental area of the State University of Rio Grande do Sul (UERGS), located at 29° 53′ 00″ S latitude and 53° 00′ 00″ W longitude, in the *Três Vendas* district of Cachoeira do Sul - Brazil. The average altitude of the region is 125 m.

The experimental design for cultivation consisted of randomized blocks,  $3 \times 3$  factorial, with three replications. The factors were composed of three different canola hybrids (Hyola 433, Hyola 571, and Hyola 61) and three sowing dates in 2018 (April 21, May 20, and June 24). Each experimental plot consisted of a total area of 3.4 m<sup>2</sup> (1.7 m  $\times$  2.0 m), with a plot useful area of 1.53 m<sup>2</sup> (1.02 m  $\times$  1.50 m).

# 2.2. Grain yield

After canola was harvested, the grain yield was obtained by the mass of grains harvested in the plot area, discounting the moisture content of the grains and extrapolating the values obtained to  $10,000\,\mathrm{m}^2$  (1 ha). The data were analyzed with the Snedecor F test (*p-value* < 0.05) and the means were compared using Tukey's test (*p-value* < 0.05). The statistical analyses were performed using the computational program SISVAR\* [20].

# 2.3. Soxhlet extraction

Oil extraction by a conventional method was performed using 1 g of each sample of harvested canola grains (based on hybrid/sowing date) and 150 mL of n-Hexane for 240 min using a Soxhlet apparatus (Marconi, Model MA491/6, Brazil). The samples (for each hybrid and sowing date) were placed in a filter paper cartridge and the cartridge was placed into the main chamber of the Soxhlet extractor. The n-Hexane was loaded in a distillation flask heated by a band heater at the bottom of the apparatus. The solvent evaporated in the flask and condensed successively in the main chamber, thus extracting the oil. At the end of each experimental run (240 min), the n-Hexane was totally evaporated from the flask and the mass of oil was quantified by the gravimetric method. The experimental assays were completely randomized and performed in triplicate. The responses were expressed as the mean  $\pm$  standard deviation.

Table 1
Base cost for equipment composing the supercritical (SFE-CO<sub>2</sub>) and Soxhlet plants.

Item	M <sup>a</sup>	Unit base cost (US\$) b,c	Quantity (un.)	Total pilot cost (US\$) d
SFE-CO <sub>2</sub>				_
Jacketed extraction vessel	0.82	200.00	2	431,733.00
CO <sub>2</sub> pump	0.55	1225.00	1	132,608.00
Cooler	0.59	320.00	1	48,701.00
Heater	0.59	210.00	1	31,960.00
Pressure gauge	0.00	70.00	6	420.00
Blocking valve	0.60	10.00	6	9943.00
Micrometering valve	0.60	40.00	2	13,258.00
Safety valve	0.60	20.00	1	3314.00
Temperature controller	0.60	15.00	2	4971.00
Piping, connectors, mixers, splitters, and crossheads e	0.40	100.00	_	3017.00
Structural material for supporting the equipment <sup>e</sup>	0.40	60.00	_	1810.00
Total cost of SFE-CO <sub>2</sub> plant <sup>d</sup>				681,735.00

Item	M <sup>a</sup>	Unit base cost (US\$) f,c	Quantity (un.)	Total pilot cost (US\$) <sup>g</sup>
Soxhlet Soxhlet apparatus Rotary evaporator	0.49 0.59	1500.00 1700.00	1 -	44,268.00 100,103.00
Total cost of Soxhlet plant <sup>g</sup>				144,371.00

<sup>&</sup>lt;sup>a</sup>M constant depending on equipment type, based on Green and Perry (2007), Peters and Timmerhaus (1991), Smith (2005) and Turton et al. (2012); <sup>b</sup> Based on an operating plant with two extraction vessels of 20 mL (laboratory scale); <sup>c</sup> Direct quotation for reference year of 2018; <sup>d</sup> Operating plant with two extraction vessels of 100 L (calculated by power law); <sup>e</sup> Total cost; <sup>f</sup> Based on an operating plant with a Soxhlet apparatus of 1 L (laboratory scale); <sup>g</sup> Operating plant with a Soxhlet apparatus of 1000 L (calculated by power law).

# 2.4. Supercritical CO2 extraction

The experimental assays were performed at a laboratory scale equipment (Fig. 1) composed mainly by: (i) a jacketed 20 mL extraction vessel (316 stainless steel) with internal diameter of 2.5 cm and 19.5 cm of height, supporting up to 50 MPa; (ii) a piston pump (Jasco, Japan); (iii) an ultrathersmostatic cooling bath (Solab, Brazil) for controlling the temperatures of  $\rm CO_2$  at the piston pump and in the serpentine; (iv) an ultrathersmostatic heating bath (Solab, Brazil) with thermocouples for pre-heating the  $\rm CO_2$ ; (v) a heating electric jacket (1500 W) to control the temperature inside the extraction vessel; (vi) blocking and micrometering valves (HIP 15-11AF2 316SS, Erie, USA); and (vii) tubing of stainless steel of 3.175 mm of internal diameter (HIP, Erie, USA).

For the extraction procedures using each sample, 18 g of canola grains were loaded into the extraction vessel, which the apparent bed density was  $0.9 \,\mathrm{g}\,\mathrm{cm}^{-3}$ . In the sequence, the solvent was pumped in the bed and the condition of pressure (40 °C) and temperature (35 MPa) was established based on a scientific work [21]. The system was maintained at a static time for 20 min and, thereafter, the dynamic extraction was started with a CO2 flow rate of 4 g min-1. The total extraction time (180 min) was fixed after evaluating the extraction kinetic curves obtained by preliminary tests. This time corresponds to a solvent to feed mass ratio of  $40\,\mathrm{g}\;\mathrm{CO_2}\,\mathrm{g}^{-1}$  of canola seeds. The oil was collected in vials and stored under refrigeration (2 °C) until the chromatographic analyses. The remaining solid was also collected at the end of the extractions and stored under refrigeration (2 °C) for further analysis. The oil yield (Eq. (1)) was calculated based on the oil recovered from the fresh raw material. The Tukey's test was applied to determine the significant differences among the yields, at a 5% uncertainty level, using STATISTICA 8.0® (Statsoft Inc., USA). The experimental assays were completely randomized and performed in duplicate. The responses were expressed as mean ± standard deviation.

$$\textit{Yield (wt. \%)} = \frac{\textit{oil extracted (g)}}{\textit{initial dry mass of canola (g)}} \cdot 100 \tag{1}$$

#### 2.5. Determination of protein

The protein content in the extracted canola grains (coproducts) was determined following the standard analytical procedures of *Association of Official Analytical Chemists* [22]. The total percentage of nitrogen in the coproducts was analyzed by the Micro Kjeldahl method. The analyses were performed in duplicate.

# 2.6. Determination of fatty acids

The content of fatty acids was analyzed in each sample of oil. For this determination,  $10\,\mu L$  of each sample of oil was solubilized in 1 mL of n-Hexane and 250  $\mu L$  of a 4 mg mL $^{-1}$  solution of methyl tridecanoate (C23:0 Me) in isooctane. The solvents were evaporated at 40 °C under vacuum. The fatty acids methyl esters (FAME) derivatizations were performed according to the method described by Visantainer [23]. The FAME was analyzed in a GC system (Shimadzu, GCMS-QP2010 Ultra, Japan) by injecting 1  $\mu L$  into a capillary column Rtx-5MS (Restek-USA) (30 m  $\times$  0.25 mm  $\times$  0.25  $\mu m$ ). The carrier gas (Helium, purity > 99%, White Martins, Brazil) flowed at a constant pressure of 103.4 kPa. The following column temperature gradient was used: 3 °C min $^{-1}$  from 50 °C to 140 °C (10 min). The injector and detector were maintained at 140 °C and the split ratio was 60:1. The compositions were expressed as milligram of each fatty acid per gram of oil (mg g $^{-1}$ ).

# 2.7. Cost of manufacturing of canola oil

The cost of manufacturing (COM) of canola oil was simulated in the SuperPro Designer 9.0° software (Intelligen Inc., Scotch Plains, NJ, USA) for a pilot/industrial scale. The COM was determined for each sample obtained for each condition of hybrids, sowing dates, and extraction methods. The processes were designed to operate for 7920 h per year, which corresponds to 3 daily shifts for 330 days per year. The yearly remaining time was considered for cleaning and equipment maintenance. The capacity considered in the study was that one able to process 180 kg of canola seeds per batch (SFE-CO<sub>2</sub>: two extraction

Table 2 Input economic parameters used for simulating the COM of canola oil and protein obtained by supercritical fluid extraction (SFE) with  ${\rm CO_2}$  and Soxhlet extraction with n-Hexane.

Economic parameter	Value	Dimension
Fixed capital investment (FCI)		
Total cost of SFE plant	681,735.00	(US\$)
Total cost of Soxhlet plant	100,103.00	(US\$)
Annual depreciation rate	10	(%)
Annual maintenance rate	6	(%)
Project lifetime	25	(years)
Annual time worked	7920	(h year <sup>-1</sup> )
Cost of raw material (CRM)		
Canola seeds	0.30	$(US$ kg^{-1})$
Transport and pre-processing of canola seeds <sup>a</sup>	40.00	(US\$ ton-1)
N-Hexane <sup>b</sup>	4.50	$(US$ kg^{-1})$
Industrial CO <sub>2</sub> b	2.85	(US\$ kg <sup>-1</sup> )
Cost of operational labor (COL)		
Wage (with benefits and administration) c	14.00	(US $$h^{-1}$ worker $^{-1}$ )
Number of workers per shift	2	(Worker shift <sup>-1</sup> )
Cost of utilities (CUT)		
Water (for cooling and cleaning) b	1.00	(US\$ ton-1)
Steam <sup>b</sup>	12.00	(US\$ ton-1)
Glycol solution b	5.00	(US\$ ton-1)
Electricity <sup>b</sup>	0.25	$(US  kW^{-1} h^{-1})$

<sup>&</sup>lt;sup>a</sup> The pre-processing steps include drying (when needed) and storing the samples until further use; <sup>b</sup> Direct quotation for the reference year of 2018; <sup>c</sup> Bureau of Labor Statistics, http://www.bls.gov/fls/country/brazil.htm, USA, accessed on September 24<sup>th</sup>, 2018.

vessels of 100 L; Soxhlet: an apparatus with a total capacity of 1000 L). The behavior of canola extraction yields and oil composition was assumed to have the same performance at larger scales as the results obtained at laboratory scale.

Scaling up the equipment cost to the required capacity was done through the power law (Eq. (2)) [24–26], where M is a constant depending on the equipment type,  $C_1$  is the equipment cost with a capacity  $Q_1$ , and  $C_2$  is the known base cost for equipment with a capacity  $Q_2$ . Values of M were obtained from scientific works reported in the literature [24–27] because the cost of a specific item is a function of size, design pressure, materials of construction, and design temperature. The base costs acquired in 2018 (local quotation, including import fees for the items that are not produced in Brazil) are presented in Table 1 for calculating the costs at the pilot/industrial scale. Other input data, as the cost of raw materials, wage, and utilities are presented in Table 2. The recycle of solvents was considered in the study.

$$C_1 = C_2 \left(\frac{Q_1}{Q_2}\right)^M \tag{2}$$

For costs estimation, some important parameters are used. The COM of canola oil depends on three main categories. The first one refers to fixed manufacturing costs (e. g., equipment, instrumentation, and building). These costs exist even though the plant is not processing, which can include insurance and depreciation of equipment. They are the first expenses when the investors start a project. In the scenarios evaluated in this work, it was considered that the investors have all capital resources to start the projects. Otherwise, if bank financing is needed for paying the fixed manufacturing costs, all fees and charges should be taken into account. The second one refers to direct manufacturing costs (e. g., purchasing costs of grains, solvents, and electricity), which are related to the amount of canola processed in the plant. They include the operators that operate directly the processing equipment. The waste generated after performing the process is included in this category. The third one refers to general expenses (e. g., management costs, engineering, and research, innovation &

**Table 3**Test of means for grain yields (kg ha<sup>-1</sup>) of canola according to the sowing date, hybrid, and interaction between these factors.

Sowing date	Hybrid			
	Hyola 433	Hyola 571	Hyola 61	Means
April 21 May 20 June 24 Means	$\begin{array}{c} 2371  \pm  139^{\ aA} \\ 2386  \pm  223^{\ aAB} \\ 1057  \pm  21^{\ bA} \\ 1938  ^{A} \end{array}$	1441 ± 292 bB 2088 ± 235 aB 1252 ± 189 bA 1594 B	2726 ± 167 <sup>aA</sup> 2807 ± 247 <sup>aA</sup> 731 ± 521 <sup>bA</sup> 2088 <sup>A</sup>	2180 <sup>a</sup> 2427 <sup>a</sup> 1013 <sup>b</sup> -

Averages followed by the same letter, lowercase in the column and uppercase in the row, do not differ from each other by the Tukey's test (p-value < 0.05).

development). These expenses are costs spent on running the business. Therefore, the COM was estimated by the SuperPro Designer 9.0° software taking into account such categories of parameters. Overall, the percent contribution of the itemized costs on the COM was evaluated based on the fixed capital investment (FCI), cost of raw materials (CRM), cost of operational labor (COL), and cost of utilities (CUT).

#### 3. Results and discussion

#### 3.1. Grain yield

The sowing date, canola hybrids and the interaction between these factors of variation caused significant differences in the grain yield of canola. These results are presented in Table 3.

The highest grain yields were obtained when the sowing was performed on April 21 and May 20, reaching 2180 and 2427 kg ha $^{-1}$ , respectively. Amongst the hybrids studied, Hyola 433 and Hyola 61 were the most productive (1938 and 2088 kg ha $^{-1}$ , respectively). The combination of sowing date and hybrids of canola resulted in higher grain yields for the Hyola 61 hybrid, sown on April 21 or May 20 (no significant difference of means). Overall, all hybrids presented grain yield above the national (Brazil) average for at least one sowing date, which is estimated at 1550 kg ha $^{-1}$  [9].

These findings demonstrate that the use of canola cultivation into a crop rotation system is promising. However, for a better decision for its suitable cultivation, not only the grain yield should be taken into account, but the properties of the oil and protein contained in the canola grains. Therefore, in the next sections, the technological and economic aspects for canola processing are presented and discussed.

**Table 4**Oil yields of canola grains obtained by supercritical fluid extraction (SFE) with CO<sub>2</sub> and Soxhlet extraction with n-Hexane.

Sample	Oil yields (wt.%)	
Hybrid – Sowing date	SFE - CO <sub>2</sub>	Soxhlet – n-Hexane
Hyola 61 – April 21 Hyola 61 – May 20 Hyola 61 – June 24 Hyola 433 – April 21 Hyola 433 – May 20 Hyola 433 – June 24 Hyola 571 – April 21 Hyola 571 – May 20 Hyola 571 – June 24	18.7 ± 0.7 abcCDE 18.2 ± 0.5 abcCDE 19.5 ± 0.4 abcBCD 15.3 ± 1.2 cE 20.4 ± 0.8 abcBCD 17.6 ± 2.8 bcCDE 16.9 ± 0.5 bcDE 21.3 ± 2.2 abBC 23.1 ± 0.7 aB	$31.4 \pm 0.1^{\text{ eA}}$ $32.7 \pm < 0.1^{\text{ cA}}$ $33.9 \pm < 0.1^{\text{ dA}}$ $34.7 \pm < 0.1^{\text{ aA}}$ $34.6 \pm 0.2^{\text{ aA}}$ $34.2 \pm 0.1^{\text{ aA}}$ $31.9 \pm < 0.1^{\text{ bA}}$ $32.4 \pm < 0.1^{\text{ bCA}}$ $34.5 \pm 0.1^{\text{ aA}}$

Averages followed by the same letter, lowercase in the column and uppercase in the row, do not differ from each other by the Tukey's test (p-value < 0.05).

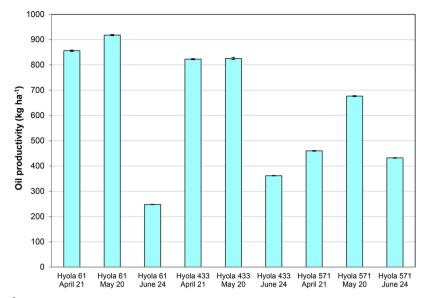


Fig. 2. Productivity of oil (kg ha<sup>-1</sup>) for the different hybrids and sowing dates; oil extraction by Soxhlet method with n-Hexane; the bars refer to standard deviation.

#### 3.2. Oil content

The oil yield was statistically different for the canola hybrids, sowing dates, and extractive methods (Table 4). When analyzing the total oil yield amongst all samples, the Soxhlet extraction presented the highest values, reaching up to 34 wt.%. As expected, the Soxhlet extraction generally provides the highest total yields because it is the traditional chemical method for extracting lipids from oleaginous samples. In some other studies reported elsewhere, it is shown higher oil yields when using Soxhlet extraction with n-Hexane from different vegetal raw materials [28,29]). Notwithstanding, the purpose of using/evaluating supercritical CO<sub>2</sub> extraction stands for the likely extraction of specific compounds at a larger concentration in the oil, as polyunsaturated fatty acids (this is presented and discussed in section 3.4).

Considering that the main objective of this work was to evaluate the extraction of canola oil as a function of hybrids and sowing date, the statistical analysis is also presented in Table 4. When using the Soxhlet method, no significant differences (p-value < 0.05) were seen for the samples Hyola 433 – April 21, Hyola 433 – May 20, Hyola 433 – June 24, and Hyola 571 – June 24. For these samples, the oil yields were approximately 34–35 wt.%, which are slightly higher than that one reported for Hyola 401 (33.1 wt.%) using Soxhlet extraction with n-Hexane [30]. In a study performed with supercritical CO<sub>2</sub> at 40 °C and 25 MPa, the yield was 19.5 wt.% (Pederssetti et al., 2011), which is similar to this study.

When performing an integrated evaluation of grain yields and oil yields, the total oil productivity per hectare is different amongst the samples (Fig. 2). Overall, the canola sown on April 21 or May 20 presented higher productivities of oil. Likewise, the hybrids Hyola 61 and Hyola 433 presented higher productivities of oil for these sowing dates. The highest oil productivity was found for the sample Hyola 61 sown on May 20 (918 kg ha<sup>-1</sup>) and lowest oil productivity was found for the sample Hyola 61 sown on June 24 (248 kg ha<sup>-1</sup>). We infer that sowing of canola for this hybrid in the late autumn delayed flowering and reduced reflection of radiation during flowering. Therefore, according to Fig. 2, the early sowing is preferable for reaching the highest oil productivities.

#### 3.3. Protein content

The main coproduct of canola processing is the cake or press-cake, which has a relatively high protein content, making it an attractive potential raw material for producing protein-based products. Due to

Table 5 Protein content (wt.%) in the coproducts (solid biomass) after performing supercritical fluid extraction (SFE) with  ${\rm CO_2}$  and Soxhlet extraction with n-Hexane.

Sample	Protein content (wt.%; i	nitial mass)
Hybrid – Sowing date	SFE - CO <sub>2</sub>	Soxhlet – n-Hexane
Hyola 61 – April 21 Hyola 61 – May 20 Hyola 61 – June 24 Hyola 433 – April 21 Hyola 433 – May 20 Hyola 433 – June 24 Hyola 571 – April 21 Hyola 571 – May 20	34.9 ± 0.1 bcdeAD 34.8 ± 0.1 cdeBD 35.7 ± 0.1 abcdAD 29.1 ± 0.1 gC 31.7 ± 0.2 gCE 32.0 ± 0.2 efgBE 32.8 ± 0.1 defBDE 31.7 ± 0.2 fgCE	32.4 ± 0.2 efD 37.9 ± 0.1 abB 38.2 ± 0.1 aB 33.8 ± 0.2 defCD 33.0 ± 0.2 defCD 37.7 ± 0.1 abcAB 35.0 ± 0.1 bcdeACD 34.6 ± 0.1 defCD
Hyola 571 – June 24	$37.7 \pm 0.1^{\text{ abcA}}$	$35.5 \pm 0.1$ abcdABC

Averages followed by the same letter, lowercase in the column and uppercase in the row, do not differ from each other by the Tukey's test (p-value < 0.05).

their biodegradable character and good techno-functional properties, canola proteins have been extensively studied in recent years [31].

The protein contents were different amongst some samples (Table 5), where the results ranged from 29.1 wt.% to 38.2 wt.%. The coproducts processed by supercritical  $CO_2$  in the condition Hyola 571 (June 24) and by Soxhlet with n-Hexane in the condition Hyola 61 (June 24) presented the highest protein contents for each extraction method. These responses are confronting the responses of oil yield. Indeed, more oil commonly leads to less protein and vice versa. Consequently, an integrated analysis should be performed for selecting the best hybrid and sowing date based on the main responses as the grain yields, oil yields, protein yields, and costs of oil and protein production (this last one is presented and discussed in Section 3.5).

The results of protein content presented in this study are similar to those reported elsewhere [32,33], where a range of 34–38 wt.% of protein is commonly found for canola bran cultivated and processed in Brazil. For example, the solid biomass of canola grains processed by supercritical CO<sub>2</sub> and Soxhlet with n-Hexane presented a maximum of 39.9 wt.% and 36.7 wt.%, respectively. One advantage of using supercritical CO<sub>2</sub> for the extraction of canola oil is the elimination of the roasting stage and, therefore, the proteins remain intact [34]. However, the protein content does not depend only on the sowing date and oil extraction method but on the hybrid as well. For example, the protein content in the Hyola 401 hybrid did not exceed 29 wt.% [30]. The

 Table 6

 Fatty acids composition (mg  $g^{-1}$ ) in the samples of canola oil obtained by supercritical fluid extraction (SFE) with CO<sub>2</sub> and Soxhlet extraction with n-Hexane.

Color   Colo		SFE - CO <sub>2</sub> Hyola 61 April 21	May 20	June 24	Hyola 433 April 21	May 20	June 24	Hyola 571 April 21	May 20	June 24
28.69 ± 13.3  29.75 ± 6.98  27.74 ± 6.02  27.8 ± 6.02  15.8 ± 6.03  21.8 ± 6.04  20.04 ± 6.04  27.8 ± 6.05  15.8 ± 6.04  20.04 ± 6.04  27.8 ± 6.05  15.8 ± 6.04  20.04 ± 6.04  27.8 ± 6.05  10.2 ± 6.04 ± 6.04  27.8 ± 6.05  10.2 ± 6.04 ± 6.04  27.8 ± 6.05 ± 6.04 ± 6.04  27.8 ± 6.05 ± 6.04 ± 6.04 ± 6.05 ± 6.04 ± 6.04 ± 6.05 ± 6.04 ±		$0.58 \pm 0.02$	$0.59 \pm 0.02$	$0.62 \pm 0.01$ $0.15 + 0.01$	$0.47 \pm 0.04$	$0.49 \pm 0.02$	$0.49 \pm 0.08$	$0.46 \pm 0.01$	$0.43 \pm 0.04$	$0.47 \pm 0.01$
13 ± 0.06         2.79 ± 0.06         1.55 ± 0.02         2.08 ± 0.06         1.99 ± 0.04         1.99 ± 0.03		36.89 ± 1.33	37.57 ± 0.98	$37.74 \pm 0.72$	$31.58 \pm 2.40$	33.56 ± 1.28	$34.70 \pm 5.59$	$33.39 \pm 1.07$	$31.90 \pm 3.29$	$34.26 \pm 0.99$
1.05 ± 0.001 0.34 ± 0.00 0.32 ± 0.001 0.25 ± 0.001 0.27 ± 0.004 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.001 0.32 ± 0.003 0.30 ± 0.002 0.30 ±		$2.16 \pm 0.08$	$2.13 \pm 0.06$	$2.79 \pm 0.05$	$1.56 \pm 0.12$	$2.08 \pm 0.08$	$1.99 \pm 0.32$	$2.04 \pm 0.07$	$1.81 \pm 0.19$	$2.06 \pm 0.06$
1.55 ± 0.05 1.39 ± 0.04 1.39 ± 0.03 1.15 ± 0.04 1.15 ± 0.04 1.15 ± 0.04 1.24 ± 0.05 1.15 ± 0.04 1.24 ± 0.05 1.15 ± 0.04 1.24 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ± 0.04 1.25 ± 0.05 1.15 ±		$0.32 \pm 0.01$	$0.34 \pm 0.01$	$0.32 \pm 0.01$	$0.30 \pm 0.02$	$0.29 \pm 0.01$	$0.27 \pm 0.04$	$0.79 \pm 0.03$	$0.67 \pm 0.07$	$0.69 \pm 0.02$
6.83 ± 0.25 = 0.29		$1.26 \pm 0.05$	$1.39 \pm 0.04$	$1.39 \pm 0.03$	$1.12 \pm 0.09$	$1.15 \pm 0.04$	$1.24 \pm 0.20$	$2.06 \pm 0.07$	$1.71 \pm 0.18$	$1.81 \pm 0.05$
6 5772 ± 119.70 5772 ± 14.23 52590 ± 10.77 ± 0.01		$6.83 \pm 0.25$	$2.31 \pm 0.06$	$4.23 \pm 0.08$	$4.91 \pm 0.37$	$3.42 \pm 0.13$	$2.01 \pm 0.32$	$3.48 \pm 0.11$	$2.34 \pm 0.24$	$2.09 \pm 0.06$
c clistation of the control of the	n9c	$547.23 \pm 19.70$	$547.25 \pm 14.23$	$529.90 \pm 10.07$	$540.30 \pm 41.06$	$565.09 \pm 21.47$	$535.00 \pm 86.14$	$499.84 \pm 15.99$	$497.58 \pm 51.25$	$495.03 \pm 14.36$
Color = 1991 = 242         1292 = 328 = 338         14881 = 248         1164 0 = 633         142 0 = 633         142 0 = 633         142 0 = 633         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         143 0 = 644         144 0 = 647         144 0 = 648         144 0 = 647         144 0 = 648         144 0 = 647         144 0 = 648         144 0 = 647         144 0 = 648         144 0 = 6	1 <u>6</u> 1	$0.15 \pm 0.01$	$0.19 \pm 0.01$	$0.21 \pm 0.01$	$0.15 \pm 0.01$	$0.15 \pm 0.01$	$0.18 \pm 0.03$	$0.17 \pm 0.01$	$0.16 \pm 0.02$	$0.17 \pm 0.01$
0.019 ± 0.011 0.020 ± 0.011 0.020 ± 0.020 ± 0.020 0.015 ± 0.011 0.010 0.020 ± 0.020 0.019 ± 0.011 0.020 ± 0.020 0.	nec	$119.91 \pm 4.32$	$129.83 \pm 3.38$	$140.81 \pm 2.68$	$116.49 \pm 8.85$	$118.35 \pm 4.50$	$142.33 \pm 22.92$	$139.90 \pm 4.48$	$121.12 \pm 12.48$	$144.41 \pm 4.19$
6.713 ± 2.42 7.244 ± 1.88	16t	$0.19 \pm 0.01$	$0.20 \pm 0.01$	$0.17 \pm 0.01$	$0.20 \pm 0.02$	$0.18 \pm 0.01$	$0.16 \pm 0.03$	$0.14 \pm 0.01$	$0.14 \pm 0.01$	$0.16 \pm 0.01$
6.44 ± 0.023 5.77 ± 0.15 6.13 ± 0.012 7.46 ± 0.57 7.38 ± 0.38 ± 0.39 ± 0.018 8.77 ± 0.018 6.42 ± 0.03 99 ± 0.017 8.72 ± 0.68 ± 0.03 6.42 ± 0.03 8.70 ± 0.038 ± 0.039 ± 0.017 8.72 ± 0.68 ± 0.01 0.14 ± 0.01 0.15 ± 0.01 0.15 ± 0.01 0.15 ± 0.01 0.15 ± 0.01 0.13 ± 0.01 0.13 ± 0.01 0.13 ± 0.01 0.14 ± 0.02 0.14 ± 0.02 0.14 ± 0.01 0.14 ± 0.02 0.14 ± 0.02 0.14 ± 0.01 0.14 ± 0.02 0.14 ± 0.02 0.14 ± 0.01 0.14 ± 0.02 0.14 ± 0.01 0.14 ± 0.02 0.14 ± 0.01 0.14 ± 0.02 0.14 ± 0.01 0.14 ± 0.02 0.14 ± 0.01 0.14 ±	13	$67.13 \pm 2.42$	$72.48 \pm 1.88$	$81.14 \pm 1.54$	$64.39 \pm 4.89$	$61.31 \pm 2.33$	$76.31 \pm 12.29$	$68.14 \pm 2.18$	$57.53 \pm 5.93$	$70.35 \pm 2.04$
9.08 ± 0.0.3 9 49.2 ± 0.0.4 8.99 ± 0.0.17 8.57 ± 0.65 88 8 ± 0.0.4 8.0 ± 0.135 8.70 ± 0.28   0.041 ± 0.01 0.04 ± 0.01 0.05 ± 0.01 0.03 ± 0.01 0.03 ± 0.01 0.03 ± 0.01 0.04 ± 0.01 0.04 ± 0.01 0.010 ± 0.01 0.03 ± 0.01 0.01 0.01 0.01 0.01 0.01 0.01 0.		$6.48 \pm 0.23$	$5.77 \pm 0.15$	$6.13 \pm 0.12$	$7.46 \pm 0.57$	$7.38 \pm 0.28$	$6.42 \pm 1.03$	$5.62 \pm 0.18$	$5.61 \pm 0.58$	$5.44 \pm 0.16$
Souther - In-Hexame         Avoid = 0.01         0.13 ± 0.01         0.13 ± 0.01         0.13 ± 0.01         0.13 ± 0.01         0.14 ± 0.01         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.01         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.02         0.14 ± 0.01         0.14 ± 0.02         0.14 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01         0.12 ± 0.01 </td <td>6</td> <td><math>9.08 \pm 0.33</math></td> <td><math>9.42 \pm 0.24</math></td> <td><math>8.99 \pm 0.17</math></td> <td><math>8.57 \pm 0.65</math></td> <td><math>8.88 \pm 0.34</math></td> <td><math>8.40 \pm 1.35</math></td> <td><math>8.76 \pm 0.28</math></td> <td><math>7.57 \pm 0.78</math></td> <td><math>8.48 \pm 0.25</math></td>	6	$9.08 \pm 0.33$	$9.42 \pm 0.24$	$8.99 \pm 0.17$	$8.57 \pm 0.65$	$8.88 \pm 0.34$	$8.40 \pm 1.35$	$8.76 \pm 0.28$	$7.57 \pm 0.78$	$8.48 \pm 0.25$
Sowhlet - n-Hexane         Hyola 433         May 20         June 24         Hyola 433         Hyola 433         Hyola 433         Hyola 571         Hyola 571         Hyola 433         Hyola 433         Hyola 571         Hyola 571         Hyola 433         Hyola 433         Hyola 571         Hyola 433         Hyola 433         Hyola 433         Hyola 571         Hyola 433         Hyola 433         Hyola 571         Hyola 433         Hyola 571         Hyola 571         Hyola 433         Hyola 571		$0.41 \pm 0.01$	$0.40 \pm 0.01$	$0.39 \pm 0.01$	$0.00 \pm 0.01$	$0.36 \pm 0.01$	$0.36 \pm 0.06$	+1	$0.32 \pm 0.03$	$0.41 \pm 0.01$
Sowhlet – n-Hexane         Hyola 433         May 20         June 24         April 21         Hyola 571           April 21         April 21         April 21         May 20         June 24         April 21         April 21           April 21         April 21         April 21         April 21         April 21         April 21           0.58 ± 0.01         0.57 ± 0.01         0.55 ± 0.01         0.55 ± 0.01         0.74 ± 0.01         0.14 ± 0.01         0.14 ± 0.01         0.14 ± 0.01         0.14 ± 0.01         0.14 ± 0.01         0.14 ± 0.01         0.14 ± 0.01         0.11 ± 0.01		$0.14 \pm 0.01$	$0.15 \pm 0.01$	$0.16 \pm 0.01$	$0.13 \pm 0.01$	$0.13 \pm 0.01$	$0.14 \pm 0.02$	$0.14 \pm 0.01$	$0.12 \pm 0.01$	$0.13 \pm 0.01$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		Soxhlet – n-Hexane Hyola 61	3		Hyola 433	;	,	Hyola 571	3	
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		April 21	May 20	June 24	April 21	May 20	June 24	April 21	May 20	June 24
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		$0.58 \pm 0.01$	$0.57 \pm 0.01$	$0.55 \pm 0.01$	$0.44 \pm 0.01$	$0.45 \pm 0.01$	$0.47 \pm 0.01$	$0.47 \pm 0.01$	$0.50 \pm 0.01$	$0.52 \pm 0.01$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		$0.14 \pm 0.01$	$0.12 \pm 0.01$	$0.12 \pm 0.01$	$0.12 \pm 0.01$	$0.10 \pm 0.01$	$0.12 \pm 0.01$	$0.11 \pm 0.01$	$0.11 \pm 0.01$	$0.13 \pm 0.01$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		$34.60 \pm 0.28$	$33.53 \pm 0.17$	$33.91 \pm 0.14$	$35.13 \pm 0.07$	$31.90 \pm 0.19$	$35.05 \pm 0.14$	$35.42 \pm 0.18$	$33.62 \pm 0.24$	$36.49 \pm 0.15$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		$1.84 \pm 0.01$	$1.81 \pm 0.01$	1.90 ± 0.01	$2.03 \pm 0.01$	$1.51 \pm 0.01$	$2.20 \pm 0.01$	$2.02 \pm 0.01$	$1.87 \pm 0.01$	$2.21 \pm 0.01$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		$0.27 \pm 0.01$	$0.32 \pm 0.01$	1.05 ± 0.01	$0.28 \pm 0.01$	0.26 ± 0.01	$0.27 \pm 0.01$	10.73 ± 0.01	100 ± 0.01	10.74 ± 0.01
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		2.01 + 0.02	3.48 + 0.02	3.08 + 0.01	$1.14 \pm 0.01$ $4.47 + 0.01$	3.50 + 0.01	$2.87 \pm 0.01$	$1.81 \pm 0.01$ $1.23 + 0.01$	1.99 ± 0.01 2.96 + 0.02	$1.91 \pm 0.01$ $1.46 + 0.01$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	n9c	$517.97 \pm 4.14$	$512.05 \pm 2.56$	464.38 ± 1.86	$574.82 \pm 1.15$	550.47 ± 3.30	$534.32 \pm 2.14$	$549.41 \pm 2.75$	496.52 ± 3.48	$513.89 \pm 2.06$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	19t	$0.17 \pm 0.01$	$0.14 \pm 0.01$	$0.16 \pm 0.01$	$0.16 \pm 0.01$	$0.13 \pm 0.01$	$0.14 \pm 0.01$	$0.17 \pm 0.01$	$0.19 \pm 0.01$	$0.19 \pm 0.01$
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	n6c	$118.80 \pm 0.95$	$106.36 \pm 0.53$	$123.60 \pm 0.49$	$124.31 \pm 0.25$	$115.09 \pm 0.69$	$139.89 \pm 0.56$	$133.54 \pm 0.67$	$134.97 \pm 0.94$	$151.61 \pm 0.61$
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	19t	$0.19 \pm 0.01$	$0.20 \pm 0.01$	$0.16 \pm 0.01$	$0.17 \pm 0.01$	$0.12 \pm 0.01$	$0.18 \pm 0.01$	$0.16 \pm 0.01$	$0.16 \pm 0.01$	$0.16 \pm 0.01$
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	23	$67.47 \pm 0.54$	$61.37 \pm 0.31$	$69.46 \pm 0.28$	$64.07 \pm 0.13$	$64.21 \pm 0.39$	$72.83 \pm 0.29$	$62.62 \pm 0.31$	$66.11 \pm 0.46$	$72.69 \pm 0.29$
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$		$5.46 \pm 0.04$	$6.13 \pm 0.03$	$5.56 \pm 0.02$	$7.11 \pm 0.01$	$6.80 \pm 0.04$	$6.81 \pm 0.03$	$6.41 \pm 0.03$	$5.66 \pm 0.04$	$5.97 \pm 0.02$
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	_	$8.78 \pm 0.07$	$8.47 \pm 0.04$	$7.95 \pm 0.03$	+1	$8.59 \pm 0.05$	$8.79 \pm 0.04$	+I	+1	8.68 ± 0.03
SFE - CO <sub>2</sub> Hyola 61  April 21  0.07 $\pm$ 0.01  0.13 $\pm$ 0.01  Hyola 571  April 21  April 21  0.07 $\pm$ 0.01  0.06 $\pm$ 0.01  0.06 $\pm$ 0.01  0.07 $\pm$ 0.01  0.01 $\pm$ 0.02  0.01 $\pm$ 0.01		$0.38 \pm 0.01$	$0.38 \pm 0.01$	$0.38 \pm 0.01$	+1	$0.26 \pm 0.01$	+1	+1	+1	$0.43 \pm 0.01$
FFE - CO2         Hyola 61         Hyola 433         Hyola 433         Hyola 433         Hyola 571         Hyola 571 <td></td> <td><math>0.13 \pm 0.01</math></td> <td><math>0.11 \pm 0.01</math></td> <td><math>0.13 \pm 0.01</math></td> <td>+1</td> <td><math>0.13 \pm 0.01</math></td> <td>+1</td> <td>+1</td> <td><math>0.14 \pm 0.01</math></td> <td><math>0.15 \pm 0.01</math></td>		$0.13 \pm 0.01$	$0.11 \pm 0.01$	$0.13 \pm 0.01$	+1	$0.13 \pm 0.01$	+1	+1	$0.14 \pm 0.01$	$0.15 \pm 0.01$
April 21 May 20 June 24 April 21 May 20 June 24 April 21 May 20 June 24 April 21 April 21 May 20 June 24 April 21 May 20 June 24 April 21 April 21 June 24 Apr		SFE - CO <sub>2</sub> Hyola 61	;	,	Hyola 433	;	;	Hyola 571	;	,
$\begin{array}{cccccccccccccccccccccccccccccccccccc$		April 21	May 20	June 24	April 21	May 20	June 24	April 21	May 20	June 24
$0.14 \pm 0.01$ $0.17 \pm 0.01$ $0.11 \pm 0.01$ $0.00$ $0.10 \pm 0.02$ $0.47 \pm 0.02$	9u	$0.07 \pm 0.01$ $3.33 \pm 0.12$	$0.06 \pm 0.01$ $3.15 \pm 0.08$	$0.10 \pm 0.01 \\ 3.24 \pm 0.06$	$0.06 \pm 0.01$ $0.01 \pm 0.01$	$0.06 \pm 0.01$ $0.01 \pm 0.01$	$0.06 \pm 0.01$ $0.01 \pm 0.01$	$\begin{array}{c} 0.07  \pm  0.01 \\ 0.01  \pm  0.01 \end{array}$	$\begin{array}{c} 0.05  \pm  0.01 \\ 0.01  \pm  0.01 \end{array}$	$0.06 \pm 0.01$ $0.02 \pm 0.01$
		$0.16 \pm 0.01$	$0.14 \pm 0.01$	$0.17 \pm 0.01$	$0.11 \pm 0.01$	0.00	$0.10 \pm 0.02$	$0.47 \pm 0.02$		$0.14 \pm 0.01$ (continued on next page)

Table 6 (continued)

	$\begin{array}{l} {\rm SFE \cdot CO_2} \\ {\rm Hyola \ 61} \\ {\rm April \ 21} \end{array}$	May 20	June 24	Hyola 433 April 21	May 20	June 24	Hyola 571 April 21	May 20	June 24
C22:6 n3 C23	$1.09 \pm 0.04$ $4.41 \pm 0.16$	$1.18 \pm 0.03$ $4.75 \pm 0.12$	$1.07 \pm 0.02 \\ 4.32 \pm 0.08$	$1.14 \pm 0.09 \\ 4.72 \pm 0.36$	$1.14 \pm 0.04$ $4.46 \pm 0.17$	$1.01 \pm 0.16 \\ 4.16 \pm 0.67$	$1.22 \pm 0.04$ $4.19 \pm 0.13$	$1.03 \pm 0.11$ $4.22 \pm 0.43$	$1.18 \pm 0.03 \\ 4.32 \pm 0.13$
C24	$2.03 \pm 0.07$	$1.86 \pm 0.05$	$1.96 \pm 0.04$	$1.93 \pm 0.15$	$2.05 \pm 0.08$	$1.92 \pm 0.31$	$1.94 \pm 0.06$	$1.85 \pm 0.19$	$1.88 \pm 0.05$
	Soxhlet – n-Hexane			007			1		
	Hyola 61 April 21	May 20	June 24	Hyola 433 April 21	May 20	June 24	Hyola 5/1 April 21	May 20	June 24
C20:4 n6	$0.07 \pm 0.01$	$0.05 \pm 0.01$	$0.07 \pm 0.01$	$0.06 \pm 0.01$	$0.06 \pm 0.01$	$0.06 \pm 0.01$	$0.06 \pm 0.01$	$0.08 \pm 0.01$	$0.07 \pm 0.01$
C22	$2.97 \pm 0.02$	$3.13 \pm 0.02$	$3.02 \pm 0.01$	$0.01 \pm 0.01$	$0.01 \pm 0.01$	$0.01 \pm 0.01$	$0.01 \pm 0.01$	$0.01 \pm 0.01$	$0.01 \pm 0.01$
C22:1	$0.14 \pm 0.01$	$0.13 \pm 0.01$	$0.13 \pm 0.01$	$0.16 \pm 0.01$	$0.12 \pm 0.01$	$0.12 \pm 0.01$	$0.11 \pm 0.01$	$0.45 \pm 0.01$	$0.13 \pm 0.01$
C22:6 n3	$1.08 \pm 0.01$	$1.08 \pm 0.01$	$1.05 \pm 0.01$	$1.21 \pm 0.01$	$1.15 \pm 0.01$	$1.18 \pm 0.01$	$0.00 \pm 0.01$	$1.36 \pm 0.01$	$1.34 \pm 0.01$
C23	$4.48 \pm 0.04$	$4.38 \pm 0.02$	$4.70 \pm 0.02$	$4.52 \pm 0.01$	$4.27 \pm 0.03$	$3.91 \pm 0.02$	$4.73 \pm 0.02$	$4.77 \pm 0.03$	$4.62 \pm 0.02$
C24	$1.84 \pm 0.01$	$2.00 \pm 0.01$	$1.87 \pm 0.01$	$2.08 \pm 0.01$	$2.10 \pm 0.01$	$2.00 \pm 0.01$	$2.25 \pm 0.01$	$2.05 \pm 0.01$	$2.20 \pm 0.01$

C14 (myristic acid); C15 (pentadecylic acid); C16 (palmitic acid); C16-1 (palmitoleic acid); C18-10 (margicaric acid); C17 (margicaric acid); C17 (margicaric acid); C18 (estearic acid); C18:109 (oleic acid); C18:206 (linoleic acid); C18:3n3 (α-linolenic acid); C20 (arachidic acid); C20:1 (gadoleic acid); C20:2 (11,14-eicosadienoic acid); C20:4 (arachidonic acid); C21 (henecosanoic acid); C22:1 (erucic acid); C22:6 (docosahexaenoic acid); C23 (tricosanoic acid); C24 (lignoceric acid); c: cis; t: trans.

protein content is also dependent on the region where the canola is cultivated. In Argentina, Sánchez et al. [35] reported 20.3% of protein. In Germany, Rathke et al. [36] obtained the highest protein content in their study as 22.5%, while in Pakistan, Davut [37] obtained 21.5%. Otherwise, up to 39.5 wt.% of protein was recovered from defatted dry matter of canola grains cultivated in France [38]. It is important to determine the percentage of protein after oil extraction because it is used as an important source for animal feed [39].

# 3.4. Fatty acids in the oil

Twenty-nine fatty acids were identified and quantified in the samples of canola oil obtained from different hybrids and sowing dates by SFE-CO<sub>2</sub> and n-Hexane (Table 6). The main fatty acids identified were: palmitic acid (C16), oleic acid (C18:1 n9; cis and trans isomers), linoleic acid (C18:2 n6; cis isomer), and  $\alpha$ -linolenic acid (C18:3 n3).

The emphasis is put on oleic and linoleic acids, which reached up to  $565\,\mathrm{mg\,g^{-1}}$  of oil and  $130\,\mathrm{mg\,g^{-1}}$  of oil, respectively. Fard et al. [40] emphasize the importance of the presence of oleic and linoleic acids because they play a significant role in increasing the nutritional quality of canola oil. Another important factor to be analyzed is the presence of erucic acid (C22: 1). A low content (<  $0.2\,\mathrm{mg\,g^{-1}}$  of oil) of erucic acid in canola oil is a positive outcome. Unfortunately, a high concentration of erucic acid limits the use of oil for human and animal consumption, as it is a fatty acid that can cause heart damage. This favorable characteristic was developed after the genetic enhancement of rapeseed to canola because the reduction of erucic acid substantially increases the fatty acid levels [6].

Furthermore, differences in the concentration of fatty acids in the oil as a function of the extraction method and the hybrids and sowing dates can be observed (Table 7). Considering the major fatty acids, the use of supercritical  $CO_2$  slightly favored the extraction of  $\alpha$ -linolenic, cis-linoleic, oleic and palmitic acids. For instance, for the condition that presented the highest grain yields (Hyola 61 – May 20), the concentrations of  $\alpha$ -linolenic, cis-linoleic, oleic and palmitic acids were 6.9%, 22.0%, 18.1%, and 12.2% higher, respectively, when applying supercritical  $CO_2$ . However, these levels of improved concentration are low. In fact, we were expecting more concentrated oil in fatty acids based on the scientific literature [41,42]. Considering that oleic acid is an indicator of the nutritional quality of canola oil, all hybrids presented satisfactory results in terms of nutritional quality. With respect to the sowing date, there is a trend of higher concentration of oleic acid when the early autumn (in Brazil) is selected for the sowing (April 21).

The fatty acids presented in this study are corroborated by the findings reported by Pederssetti et al. [34], which the main fatty acids in the oil were oleic acid ( $680\,\mathrm{mg\,g^{-1}}$ ) and linoleic acid ( $150\,\mathrm{mg\,g^{-1}}$ ). After analyzing the effect of microwaves for canola oil extraction, Sánchez et al. [35] reported  $622.7\,\mathrm{mg\,g^{-1}}$  of oleic acid and  $189.4\,\mathrm{mg\,g^{-1}}$  of linoleic acid. After using the technique of ultrasound for canola oil bleaching, Icyer and Durak [43] presented  $479.2\,\mathrm{mg\,g^{-1}}$  of oleic acid and  $218.6\,\mathrm{mg\,g^{-1}}$  of linoleic acid. A satisfactory concentration of these fatty acids in canola oil was also reported by Fard et al. [40]. The authors tested the cultivars Sarigol (mid-season), Delgan (early season), Jacomo (late season), Jeromeh (late season) and Hyola 401 (early season). The best result was obtained with the cultivar Delgan:  $642.5\,\mathrm{mg\,g^{-1}}$  and  $223.5\,\mathrm{mg\,g^{-1}}$  of oleic and linoleic acids, respectively. Therefore, the trend of higher concentration of unsaturated fatty acids in the oil is a common response in other studies as well [40,43].

Farahmandfar et al. [44] obtained canola oil composed by (mass basis) oleic (65.39%), linoleic (16.32%),  $\alpha$ -linolenic (7.54%) and palmitic acids (4.29%). Similar results were found by Shahbazi and Shavisi [45], which obtained canola oil mainly composed by (mass basis) oleic acid (65.01%), linoleic acid (19.56%), linolenic acid (8.11%) and palmitic acid (4.48%).

In the fatty acid profiles, it is possible to observe that canola oil is

Test of means for the four major fatty acids (mg g<sup>-1</sup>) in the samples of canola oil obtained by supercritical fluid extraction (SFE) with CO<sub>2</sub> and Soxhlet extraction with n-Hexane.

-	$495.0 \pm 14.4$ DEF	$144.4 \pm 4.2^{B}$ 70.4 $\pm 2.0^{BC}$	$34.3 \pm 1.0$ CDE		$513.9 \pm 2.1^{E}$ $151.6 \pm 0.6^{A}$ $72.7 \pm 0.3^{B}$ $36.5 \pm 0.2^{B}$
June 24	495.0	144.4 = 70.4 ±	34.3 ±	June 24	$513.9 \pm 2.1^{1}$ $151.6 \pm 0.6^{\prime}$ $72.7 \pm 0.3^{1}$ $36.5 \pm 0.2^{1}$
May 20	497.6 ± 51.3 ABCDEFG	$121.1 \pm 12.5$ CDEF $57.5 \pm 5.9$ DE	$31.9 \pm 3.3$ DE	May 20	$496.5 \pm 3.5^{\mathrm{F}}$ $135.0 \pm 1.0^{\mathrm{C}}$ $66.1 \pm 0.5^{\mathrm{C}}$ $33.6 \pm 0.2^{\mathrm{E}}$
Hyola 571 April 21	499.8 ± 116.0 ABCDEFG	$139.9 \pm 4.5^{BC}$ $68.1 \pm 2.2^{BC}$	$33.4 \pm 1.1$ DE	Hyola 571 April 21	$549.4 \pm 2.8 ^{BC}$ $133.5 \pm 0.7 ^{C}$ $62.6 \pm 0.3 ^{DE}$ $35.4 \pm 0.2 ^{C}$
June 24	535.0 ± 86.1 ABCDEFG	$142.3 \pm 22.9^{\text{ABCDE}}$ $76.3 \pm 12.3^{\text{ABCD}}$	$34.7 \pm 5.6$ ABCDE	June 24	$534.3 \pm 2.1^{\mathrm{D}}$ $139.9 \pm 0.6^{\mathrm{B}}$ $72.8 \pm 0.3^{\mathrm{B}}$ $35.1 \pm 0.1^{\mathrm{CD}}$
May 20	$565.1 \pm 21.5$ ABC	$118.4 \pm 4.5^{D}$ $61.3 \pm 2.3^{D}$	$33.6\pm1.3^{\mathrm{DE}}$	May 20	550.5 $\pm$ 3.3 $^{\rm B}$ 115.1 $\pm$ 0.7 $^{\rm E}$ 64.2 $\pm$ 0.4 $^{\rm D}$ 31.9 $\pm$ 0.2 $^{\rm E}$
Hyola 433 April 21	$540.3 \pm 41.1$ ABCDEF	$116.5 \pm 8.9^{\text{ DEF}}$ $64.4 \pm 4.9^{\text{ CD}}$	$31.6~\pm~2.4~^{\rm E}$	Hyola 433 April 21	$574.8 \pm 1.2^{\text{ A}}$ $124.3 \pm 0.3^{\text{ D}}$ $64.1 \pm 0.1^{\text{ D}}$ $35.1 \pm 0.1^{\text{ C}}$
June 24	529.9 $\pm$ 10.1 <sup>D</sup>	$140.8 \pm 2.7^{ B}$ $81.1 \pm 1.5^{ A}$	$37.7 \pm 0.7^{\text{ A}}$	June 24	$464.4 \pm 1.9 \text{ G}$ $123.6 \pm 0.5 \text{ D}$ $69.5 \pm 0.3 \text{ BC}$ $33.9 \pm 0.1 \text{ E}$
May 20	$547.3 \pm 14.2$ <sup>CD</sup>	$129.8 \pm 3.4^{\circ}$ $72.5 \pm 1.9^{\circ}$	$37.6 \pm 1.0^{\text{ AB}}$	May 20	$512.1 \pm 2.6^{E}$ $106.4 \pm 0.5^{F}$ $61.4 \pm 0.3^{E}$ $33.5 \pm 0.2^{E}$
SFE - $CO_2$ Hyola 61 April 21	$547.2 \pm 19.7$ <sup>CD</sup>	$119.9 \pm 4.3^{\text{ DE}}$ $67.1 \pm 2.4^{\text{ C}}$	$36.9 \pm 1.3^{\text{ABC}}$	Soxhlet – n-Hexane Hyola 61 April 21	$518.0 \pm 4.1^{E}$ $118.8 \pm 1.0^{E}$ $67.5 \pm 0.5^{C}$ $34.6 \pm 0.3^{DE}$
	C18:1 n9*	C18:2 n6c C18:3 n3	C16		C18:1 n9° C18:2 n6c C18:3 n3 C16

\* Means the sum of the cis and trans isomers.

C18:1n9 (oleic acid); C18:2n6c (cis-linoleic acid); C18:3n3 ( $\alpha$ -linolenic acid); C16 (palmitic acid).

A-G analysis performed for each row (each fatty acid); different letters indicate differences among the samples of oil obtained from different hybrids, sowing dates, and solvents.

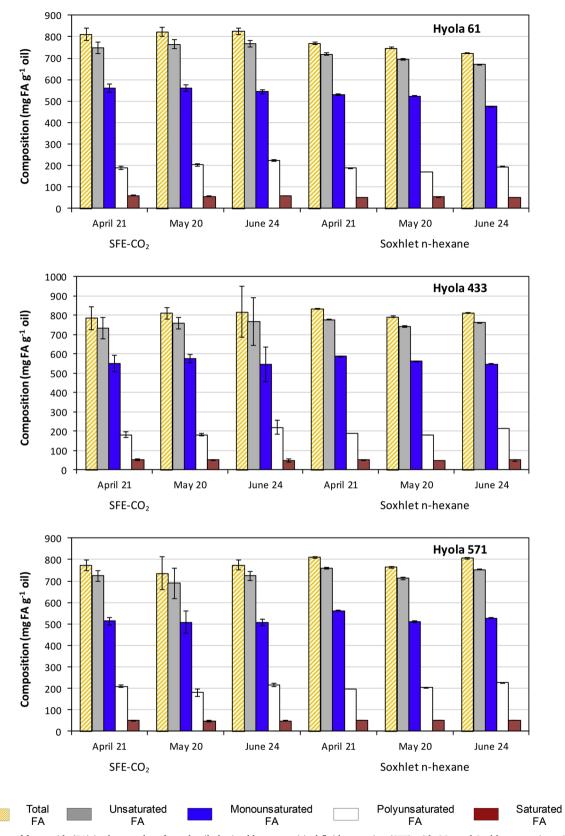


Fig. 3. Composition of fatty acids (FA) in the samples of canola oil obtained by supercritical fluid extraction (SFE) with CO<sub>2</sub> and Soxhlet extraction with n-Hexane; the bars refer to standard deviation.

very nutritional because it has high levels of monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA), as linoleic and linolenic acids (Fig. 3). According to Eskin (2015), this combination makes canola oil extremely healthy due to these cardioprotective

substances. According to Barbosa et al. [46], a low amount of saturated fatty acids (SFA) is an important characteristic because their excessive consumption is also harmful to human health. MUFA and PUFA are essential fatty acids for reducing total cholesterol, thus contributing to

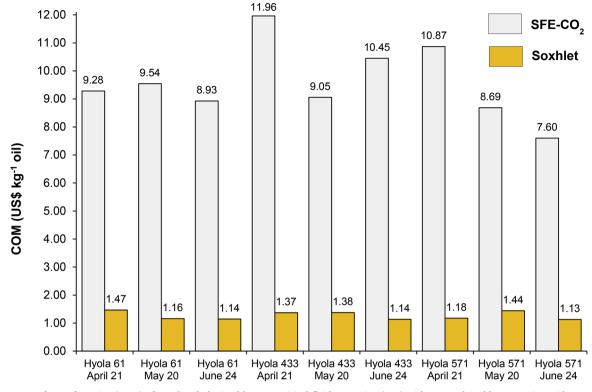


Fig. 4. Cost of manufacturing (COM) of canola oil obtained by supercritical fluid extraction (SFE) with CO2 and Soxhlet extraction with n-Hexane.

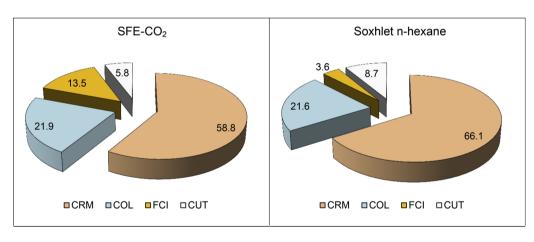


Fig. 5. Contribution of each component (CRM, CUT, COL, and FCI) on the COM of canola oil obtained by supercritical fluid extraction (SFE) with CO<sub>2</sub> and Soxhlet extraction with n-Hexane; the scenarios were considered for the highest oil yields for SFE-CO<sub>2</sub> (Hyola 571 – June 24) and for Soxhlet with n-Hexane (Hyola 433 – April 21).

cardiovascular health.

# 3.5. Cost of manufacturing of canola oil

The COM of canola oil can provide valuable insights even at earlier stages of research and projects when decisions regarding canola cultivation and operational conditions can be evaluated within the research background. The results can confirm the technological potential of such processing routes and can encourage further advances toward commercial application. Therefore, the COM of canola oil is presented for different hybrids, sowing dates, and extraction methods (Fig. 4). For the simulation of COM of oil in the SuperPro Designer 9.0°, two scenarios were considered: one with a selling price of canola cake as US\$ 0.48 kg $^{-1}$  when the cake presented more than 35 wt.% protein and the other with a selling price of canola cake as US\$ 0.35 kg $^{-1}$  when the cake presented protein in the range 29–35 wt.%.

For SFE-CO<sub>2</sub>, the COM of oil ranged from US\$  $7.60\,\mathrm{kg}^{-1}$  to US\$  $11.96\,\mathrm{kg}^{-1}$ . For Soxhlet with n-Hexane, the COM of oil ranged from US\$  $1.13\,\mathrm{kg}^{-1}$  to US\$  $1.47\,\mathrm{kg}^{-1}$ . The capacity considered in the study for

both extraction methods was  $180\,\mathrm{kg}$  of canola seeds per batch. The values of COM were lower for Soxhlet extraction because this method was able to recover more oil than SFE-CO<sub>2</sub> did (Table 4), yielding larger oil productivity. We could not find other studies that reported an economic approach of canola processing for comparing our results. Overall, when more oil is recovered, there is a trend of reducing the values of COM [47,48]. However, if a smaller amount of grains is processed per batch, there is a trend of increasing the COM (US\$/kg oil) as a consequence of the influence of other costs, especially the fixed ones [49,50].

Even though the Soxhlet extraction needs a further processing step (solvent evaporation) for obtaining pure canola oil, the increase in the consumption of energy did not increase the final value of COM, but the CUT (Fig. 5). In fact, besides the higher oil yields, the values of COM are lower for Soxhlet extraction because the cost of the equipment is lower than that one operating by supercritical technology (Table 1). For SFE-CO<sub>2</sub>, the percent contribution of COL is 13.5% while for Soxhlet with n-Hexane its contribution is only 3.6% (Fig. 5). Overall, the main influence on COM is the CRM, especially the cost of canola seeds.

Considering a same selling price of canola seeds (US\$  $0.30 \, \mathrm{kg}^{-1}$ ) for all hybrids and sowing dates tested in this study, it is possible to indicate Hyola 61 (May 20 or June 24), Hyola 433 (June 24) or Hyola 571 (April 21 or June 24) as the best conditions that present values of COM lower than US\$  $1.20 \, \mathrm{kg}^{-1}$ . When making an integrated evaluation for the responses of grain yields, oil yields, protein contents, oil composition, and values of COM, we infer the Hyola 61 sown on May 20 (autumn in Brazil) as the best combination of hybrid and sowing date.

#### 4. Conclusion

This research investigated the grain yields, oil yields, oil composition, protein contents, and cost of manufacturing of oil from three hybrids of canola (Hyola 61, 433 and 571) and three sowing dates (April 21, May 20 and June 24) of canola cultivation in Brazil. As the main general conclusions, higher grain yields are obtained when canola is sown on early autumn (April 21) or mid-autumn (May 20) for all hybrids. However, in the late season (seeds sown on June 24), most of the samples contained slightly more oil and more protein contents. Otherwise, in terms of fatty acids composition, there was not a clear trend as a function of hybrids or sowing dates. Even so, high concentrations of unsaturated fatty acids were obtained in most of the samples of oil. Furthermore, the Soxhlet extraction method with n-Hexane provided higher oil yields for all samples than supercritical CO2 extraction method did. Consequently, the values of cost of manufacturing of canola oil were lower for the conventional method, which did not exceed US\$ 1.47 kg-1 oil. Therefore, based on the main responses evaluated in this work, the hybrid Hyola 61 sown on May 20 seems to be the most promising condition for being cultivated by farmers and for being processed by food-related industries.

#### Conflict of interest

The authors declare that they have no conflict of interest.

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# References

- V.J. Barthet, Canola: Overview, Encyclopedia of Food Grains, second ed., Elsevier, Cambridge, 2015.
- [2] D.F. Figueiredo, A.E. Murakami, M.A. Dos Santos Pereira, A.C. Furlan, F.L.B. Toral, Desempenho e morfometria da mucosa de duodeno de frangos de corte alimentados com farelo de canola, durante o período inicial, Rev. Bras. Zootec. 32 (2003) 1321–1329.
- [3] G. Yang, X. Zhou, C. Li, Y. Nie, X. Zhang, Cotton stubble mulching helps in the yield improvement of subsequent winter canola (Brassica napus L.) crop, Ind. Crops Prod. 50 (2013) 190–196.
- [4] A.D.S. Vieira, A. Doyle, R.Q. Aucélio, N.A. Rey, A study of canola degradation mediated by CuO, J. Environ. Chem. Eng. 5 (2017) 3766–3772.
- [5] H.B. Vieira, C.C.D.A. Buschinelli, N.P. Ramos, G.O. Tomm, Gestão ambiental em propriedade rural dedicado a produção de canola para geração de biodiesel na região de Passo Fundo (RS). Annals of International Congress of Bioenergy, Curitiba, (2010).
- [6] N.A.M. Eskin, Rapeseed Oil/Canola, Encyclopedia of Food and Health, first ed., Elsevier, Cambridge, 2015.
- [7] H.S. Rostagno, L.F.T. Albino, J.L. Donzele, P.C. Gomes, R.F. Oliveira, D.C. Lopes, A.S. Ferreira, S.L.T. Barreto, Tabela Brasileira para Aves e Suínos, Compos. Aliment. Exig. Nutr. 2 (2005) 185.
- [8] G. Chen, R.R. Weil, R.L. Hill, Effects of compaction and cover crops on soil least limiting water range and air permeability, Soil Tillage Res. 136 (2014) 61–69.
- [9] CONAB, Companhia Nacional de Abastecimento. Acompanhamento da safra brasileira, (2017).

- [10] M. Khanali, S.A. Mousavi, M. Sharifi, F. Keyhani Nasab, K. Chau, Life cycle assessment of canola edible oil production in Iran: a case study in Isfahan province, J. Clean. Prod. 196 (2018) 714–725.
- [11] W.F. Schillinger, S.J. Werner, Horned lark damage to pre-emerged canola seedlings, Ind. Crops Prod. 89 (2016) 465–467.
- [12] V. Najdanovic-Visak, F.Y.-L. Lee, M.T. Tavares, A. Armstrong, Kinetics of extraction and in situ transesterification of oils from spent coffee grounds, J. Environ. Chem. Eng. 5 (2017) 2611–2616.
- [13] M. Herrero, A. Cifuentes, E. Ibañez, Sub- and supercritical fluid extraction of functional ingredients from different natural sources: plants, food-by-products, algae and microalgae - a review, Food Chem. 98 (2006) 136–148.
- [14] B. Suryawanshi, B. Mohanty, Modeling and optimization: supercritical CO2 extraction of Pongamia pinnata (L.) seed oil, J. Environ. Chem. Eng 6 (2018)
- [15] K. Chhouk, Wahyudiono, H. Kanda, M. Goto, Efficacy of supercritical carbon dioxide integrated hydrothermal extraction of Khmer medicinal plants with potential pharmaceutical activity, J. Environ. Chem. Eng. 6 (2018) 2944–2956.
- [16] A. Sepehri, M.-H. Sarrafzadeh, Effect of nitrifiers community on fouling mitigation and nitrification efficiency in a membrane bioreactor, Chem. Eng. Process. Process Intensif. 128 (2018) 10–18.
- [17] Q. Zhang, Y. Zheng, X. Hu, W. Lv, D. Lv, J. Chen, M. Wu, Q. Song, J. Shentu, Ethnopharmacological uses, phytochemistry, biological activities, and therapeutic applications of *Alpinia oxyphylla* Miquel: a review, J. Ethnopharmacol. 224 (2018) 149–168.
- [18] N. Nasr, M. Khayami, R. Heidari, R. Jamei, Genetic diversity among selected varieties of Brassica napus (Cruciferae) based on the biochemical composition of seeds, J. Sci. (Univ. Tehran) (JSUT) 32 (2006) 37–40.
- [19] G.L. Zabot, M.N. Moraes, P.I.N. Carvalho, M.A.A. Meireles, New proposal for extracting rosemary compounds: process intensification and economic evaluation, Ind. Crops Prod. 77 (2015) 758–771.
- [20] D.F. Ferreira, Sisvar: a computer statistical analysis system, Ciência e Agrotecnologia 35 (2011) 1039–1042.
- [21] M. Koubaa, H. Mhemdi, E. Vorobiev, Influence of canola seed dehulling on the oil recovery by cold pressing and supercritical CO<sub>2</sub> extraction, J. Food Eng. 182 (2016) 18–25.
- [22] AOAC, Official Methods of Analysis. AOAC International, sixteenth ed., AOAC International, Arlington, Virginia, 1995.
- [23] J.V. Visentainer, Analytical aspects of the flame ionization detector response of fatty acid esters in biodiesels and foods. Ouim. Nova 35 (2012) 274–279.
- [24] D. Green, R. Perry, Perry's Chemical Engineers' Handbook, eighth ed., McGraw-Hill Education. New York. 2007.
- [25] R. Smith, Chemical Process Design and Integration, Wiley, Chichester, 2005.
- [26] R. Turton, R.C. Bailie, W.B. Whiting, J.A. Shaeiwitz, D. Bhattacharyya, Analysis, Synthesis and Design of Chemical Processes, fourth ed., Prentice Hall, Upper Saddle River, 2012.
- [27] M.S. Peters, K.D. Timmerhaus, Plant Design and Economics for Chemical Engineers, fourth ed., McGraw-Hill, Auckland, 2012.
- [28] S. Stamenković, I.G. Djalovićb, M.D. Kostić, P.M. Mitrović, V.B. Veljković, Optimization and kinetic modeling of oil extraction from white mustard (Sinapis alba L.) seeds, Ind. Crops Prod. 121 (2018) 132–141.
- [29] S. Ben-Youssef, J. Fakhfakh, C. Breil, M. Abert-Vian, F. Chemat, N. Allouche, Green extraction procedures of lipids from Tunisian date palm seeds, Ind. Crops Prod. 108 (2017) 520–525.
- [30] F.T. Lucas, E.L.M. Coutinho, J.M.V. Paes, J.C. Barbosa, Produtividade e qualidade de grãos de canola em função da adubação nitrogenada e sulfatada, Semin. Ciências Agrárias 34 (2013) 3205.
- [31] M. Das Purkayastha, A.K. Borah, S. Saha, A.K. Manhar, M. Mandal, C.L. Mahanta, Effect of maleylation on physicochemical and functional properties of rapeseed protein isolate, J. Food Sci. Technol. 53 (2016) 1784–1797.
- [32] G.O. Tomm, S. Wiethölter, G.A. Dalmago, H.P. Santos, Tecnologia para produção de canola no Rio Grande do Sul, Embrapa trigo 88 (2009) 1–39.
- [33] A.M. De Castro, L. Castilho, R. dos, D.M.G. Freire, Characterization of babassu, canola, castor seed and sunflower residual cakes for use as raw materials for fermentation processes, Ind. Crops Prod. 83 (2016) 140–148.
- [34] M.M. Pederssetti, F. Palú, E.A. Da Silva, J.H. Rohling, L. Cardozo-Filho, C. Dariva, Extraction of canola seed (*Brassica napus*) oil using compressed propane and supercritical carbon dioxide, J. Food Eng. 102 (2011) 189–196.
- [35] R.J. Sánchez, C.M. Mateo, M.B. Fernández, S.M. Nolasco, Bidimensional modeling applied to oil extraction kinetics of microwave-pretreated canola seeds, J. Food Eng. 192 (2017) 28–35.
- [36] G.W. Rathke, T. Behrens, W. Diepenbrock, Integrated nitrogen management strategies to improve seed yield, oil content and nitrogen efficiency of winter oilseed rape (Brassica napus L.): A review, Agric. Ecosyst. Environ. 117 (2006) 80–108.
- [37] K. Davut, The effect of different nitrogen doses on seed yield, oil, protein and nutrient contents of spring rape, Pakistan J. Bot. 40 (2008) 807–813.
- [38] O. Laguna, A. Barakat, H. Alhamada, E. Durand, B. Baréa, F. Fine, P. Villeneuve, M. Citeau, S. Dauguet, J. Lecomte, Production of proteins and phenolic compounds enriched fractions from rapeseed and sunflower meals by dry fractionation processes, Ind. Crops Prod. 118 (2018) 160–172.
- [39] C. Wang, J. Wu, G.M. Bernard, Preparation and characterization of canola protein isolate-poly(glycidyl methacrylate) conjugates: a bio-based adhesive, Ind. Crops Prod. 57 (2014) 124–131.
- [40] N.S. Fard, H.H.S. Abad, A.H.S. Rad, E.M. Heravan, J. Daneshian, Effect of drought stress on qualitative characteristics of canola cultivars in winter cultivation, Ind. Crops Prod. 114 (2018) 87–92.
- [41] E.R. Abaide, G.L. Zabot, M.V. Tres, R.F. Martins, J.L. Fagundez, L.F. Nunes

- S. Druzian, J.F. Soares, V.D. Prá, J.R.F. Silva, R.C. Kuhn, M.A. Mazutti, Yield, composition, and antioxidant activity of avocado pulp oil extracted by pressurized fluids, Food Bioprod. Process. 102 (2017) 289–298.
- [42] J.F. Soares, G.L. Zabot, M.V. Tres, F.C. Lunelli, V.M. Rodrigues, M.T. Friedrich, C.A. Pazinatto, D. Bilibio, M.A. Mazutti, N. Carniel, W.L. Priamo, Supercritical CO2 Extraction of black poplar (Populus nigra L.) extract: experimental data and fitting of kinetic parameters, J. Supercrit. Fluids 117 (2016) 270–278.
- [43] N.C. Icyer, M.Z. Durak, Ultrasound-assisted bleaching of canola oil: improve the bleaching process by central composite design, LWT 97 (2018) 640–647.
- [44] R. Farahmandfar, M. Asnaashari, R. Sayyad, Comparison antioxidant activity of Tarom Mahali rice bran extracted from different extraction methods and its effect on canola oil stabilization, J. Food Sci. Technol. 52 (2015) 6385–6394.
- [45] Y. Shahbazi, N. Shavisi, Effect of methanolic *Prosopis farcta* extract on storage stabilization of canola oil, J. Food Sci. Technol. (2018), https://doi.org/10.1007/ s13197-018-3503-5.
- [46] K.M. Barbosa, L.P. Cardoso, A.P.B. Ribeiro, T.G. Kieckbusch, M.H.M. Buscato, Crystallization of low saturated lipid blends of palm and canola oils with sorbitan

- monostearate and fully hydrogenated palm oil, J. Food Sci. Technol. 55 (2018) 1104–1115.
- [47] J.C.F. Johner, T. Hatami, G.L. Zabot, M.A.A. Meireles, Kinetic behavior and economic evaluation of supercritical fluid extraction of oil from pequi (*Caryocar brasiliense*) for various grinding times and solvent flow rates, J. Supercrit. Fluids 140 (2018) 188–195.
- [48] G.L. Zabot, M.N. Moraes, M.A.A. Meireles, Process integration for producing tocotrienols-rich oil and bixin-rich extract from annatto seeds: a techno-economic approach, Food Bioprod. Process. 109 (2018) 122–138.
- [49] T. Hatami, J.C.F. Johner, G.L. Zabot, M.A.A. Meireles, Supercritical fluid extraction assisted by cold pressing from clove buds: extraction performance, volatile oil composition, and economic evaluation, J. Supercrit. Fluids 144 (2019) 39–47.
- [50] J. Viganó, G.L. Zabot, J. Martínez, Supercritical fluid and pressurized liquid extractions of phytonutrients from passion fruit by-products: economic evaluation of sequential multi-stage and single-stage processes, J. Supercrit. Fluids 122 (2017) 22 (2017)