

## Oil and fatty acid profile of seeds of soybean cultivars and their relationship with biodiesel and feeding

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## **ABSTRACT**

The aim of this study was to evaluate the fatty acid profile in soybean cultivars from northeastern Brazil, materials developed primarily to achieve a minimum level of oil and protein. The purpose is to serve as to warn about the need to modify fatty acids profiles to enhance both the oil for human consumption and that used in biodiesel production. Results showed the predominance of linoleic acid (average 50.1%) followed by oleic acid (27.9%), palmitic acid (11.97%), linolenic acid (6.68%) and stearic acid (3.38%) in the composition of fatty acids. Such condition makes soybean oil less competitive for both human consumption (for its high content of saturated fatty acids) and the biodiesel industry (problems with oxidative stability and flow in the cold). Considerations are also made on conventional and modern techniques to overcome these drawbacks.

**Key words**: *Glycine max*, biodiesel, saturated fat, oleic acid.

Soybean oil consumed in the United States in 2016 reached 54% of total fat and edible oil (Soystats 2017). The attraction for this consumption of soy is determined mainly by price, among others, and by fatty acid composition, which traditionally contains 11% of palmitic acid (16:0), 4% of stearic acid (18:0), 23% of oleic acid (18:1), 54% of linoleic acid (18:2), and 8% of linolenic acid (18:3) (Guo and Petrovic 2005). Palmitic and stearic acids are so-called saturated acids in the sense that there is no double bond between the carbon atoms in their chemical makeup, causing the fatty acids to saturate with hydrogen. The other acids are called unsaturated: the monounsaturated oleic acid (18:1), and the polyunsaturated acids; linoleic acid (18:2) and linolenic acid (18:3).

The World Health Organization (WHO), in conjunction with the Centers for Disease Control and Prevention (CDC) and the FDA in The United States of America, advise people to limit their intake of saturated fat since it has been shown it has been linked to cardiovascular disease. A diet high in saturated fats can lead to high cholesterol, atherosclerosis, coronary artery disease and stroke. Consuming saturated fats increases low-density lipoprotein (LDL) – also known as the "bad" cholesterol. Unsaturated fat, on the other hand, increases high-density lipoproteins, resulting in a healthier cholesterol profile.

The consumption of oils with high of oleic acid is very desirable, since this monounsaturated fatty acid: a) improves the useful life of the oil; b) reduces the need for a process called hydrogenation, which increases the cost of oil and causes the unwanted trans fat acid that has been linked to many human health problems (myocardial infarction and arteriosclerosis). The harmful health effects of *trans* fats are even worse than those of saturated fats (Ascherio and Willet 1997; Danaei 2009). In addition, saturated fats increase LDL, while *trans* fats also increase the proportion of LDL in the blood.

The fatty acid profile of vegetable oils is also an important aspect when used as fuel by the new environment conservation policy and as substitute fossil fuels, supposedly involved in the greenhouse effect. In biodiesel production, there is a need for oils with high levels in oleic acid and low in saturated fatty acids to improve oxidative stability and increase flow in the cold. The oxidative stability of the biodiesel is related to the oxidation that occurs in all natural oils, causing quality degradation and affecting its viscosity, which can clog engine filters and cause serious damages. Due to the high oxidation rate, soybean biodiesel has the shortest shelf life compared to petroleum diesel (Dunn 2005). The flow properties in the cold refer to the loss of fluidity of the oil caused by the low temperature and the occurrence of crystallization.

Both the oxidative stability and the cold flow properties of the biodiesel are strongly influenced by the lipid composition of the soybean oil. The flow in the cold is affected by saturated fatty acids (stearic and palmitic acids). Oxidative stability depends on the number of carbon double bonds of fatty acids, decreasing the large number of carbon double bonds in the acid. Thus, oleic acids with a double bond (18: 1) has higher oxidative stability than linoleic acids (18: 2), and linolenic acids (18: 3) with three double bonds is the least stable (Clemente and Cahoon 2009). Lipid profile information of the oil of soybean cultivars in the country are scarce and nonexistent in the Northeast region. The objective of the present study was to evaluate the current state of the oil quality of some soybean cultivars in the Mid-North region and its relationship with health and biodiesel production.

Four conventional cultivars developed by EMBRAPA and grown under experimental field conditions as check varieties in final evaluation in Chapadinha-MA (Latitude 03° 44' 30" S) in Northeast region of Brazil, during the 2009/10 crop (BRS Tracajá, BRS Candeia, BRS 326 and BRS Carnauba) were evaluated. Ten seeds were randomly selected from each cultivar for fatty acid analysis. Each sample was placed inside a paper envelope, then, it was manually crushed with a hammer and put in separate test tubes for fatty acid extraction. Crushed seeds



were extracted in 5mL chloroform: hexane:methanol (8:5:2, v/v/v) overnight. Derivatization was done by transferring 100 mL of extract to vial and adding 75 mL of methylating reagent (0.25 M methanolic sodium methoxide:petroleum ether:ethyl ether, (1:5:2, v/v/v). Hexane was added to dilute samples to approximately 1 mL. An Agilent (Palo Alto, CA) series 6890 capillary gas chromatographfitted with a flame ionization detector (2758C) was used with an AT-Silar capillary column (Alltech Associates, Deerfield, IL). Standard fatty acid mixtures (Animal and Vegetable Oil Reference Mixture 6, AOACS) were used as calibration reference standards (Figure 1).



Figure 1. Chromatograph coupled to mass spectrometry Agilent.

Average oil content of the cultivars analyzed was 17%, ranging from 15.6 to 19.8% (Table 1). This average is lower than that reported by Peluzio et al., (2014) for cultivars from the State of Tocantins, averaging 19.6%. It was also lower than the averages published by Costa et al. (2001) for cultivars from the States of Paraná, Santa Catarina and Mato Grosso, which reached 19.1%. It is known that environmental effects on quantitative traits such as seed oil in soybean can significantly affect the performance of a given genotype when exposed to varying growing conditions (Hemingway et al., 2015).

**Table 1**. Percentage of oil contents (on dry basis) of seed oils from four soybean cultivars grown in the Northern Region of Brazil. Chapadina, MA, Brazil. 2011.

Cultivar	Oil (%)
BRS Tracajá	22.09
BRS Candeia	21.78
BRS 326	22.36
BRS Carnaúba	22.74
Mean	22.09

The oil content of soybean cultivars in the Northeast of Brazil (17%) is also lower than their Argentine peers, whose average is 23.0%, ranging from 22.0 to 24.1%, (Maestri et al., 1998). Similarly, North American soybean



cultivars have higher oil contents than their peers in the Brazilian Northeast, with an average of 18.6% (Hurburgh 1994). This comparisons has limitations since Brazilian data refer to a single site and a smaller number of genotypes.

Table 2 shows the analyzed soybeans cultivars. Fatty acids composition showed the predominance of linoleic acid (average of 50.1%), followed by oleic acid (27.9%), palmitic acid (12.0%), linolenic acid (6.7%) and stearic acid with 3.4%. This sequence of predominance of certain fatty acids is also reported (on average) for the twenty main Brazilian cultivars (Pereira et al., 1991) and those destined for human consumption (Vieira et al., 1999). All cultivars evaluated in this study followed this trend of medium profile, except BRS Tracajá, which has high content of oleic acid (42%) and low relative content of linoleic acid (38.7%) and linolenic acid (4.8 %). The differentiated profile of this cultivar cannot be attributed to the selection process, since neither fatty acid evaluations nor selection were performed. It is also worth mentioning that BRS Tracajá was the cultivar with the lowest oil content (15.6%).

Comparing the lipid profile of cultivars from the Brazilian Northeast with their Argentine peers (Maestri et al., 1998), and North American (Oliva et al., 2006) peers, with the exclusion of BRS Tracajá, the percentages of fatty acids are similar, except for stearic acid content that in the Brazilian genetic material appears with lower values (Table 2).

**Table 2**. Fatty acid profiles of seed oil contents (on dry basis) of seed oils from four soybean cultivars grown in the Northern Region of Brazil. Chapadina, MA, Brazil. 2011.

Cultivar	Palmitic	Estearic acid	Oleic	Linoleic	Linolenic
	acid (%)	(%)	acid (%)	acid (%)	acid (%)
BRS Tracajá	11.5	3.1	42.0	38.7	4.8
BRS Candeia	11.7	3.3	26.9	51.8	6.3
BRS 326	12.7	3.6	19.0	56.0	8.7
BRS Carnaúba	12.0	3.5	23.7	53.9	6.9
Mean	12.0	3.4	27.9	50.1	6.7

The modifying of the composition of soybean oil has been a goal of some breeding programs, especially in developed countries. As a result, new genotypes with different fatty acid profiles have been developed through genetic recombination, induced mutation and transgenic (Oliva et al., 2006; Haun et al., 2014).

A list of modified genotypes with altered fatty acid profiles to meet improvement demands to make the oil healthier and more suitable for the biodiesel industry is shown in Table 3. It showns that changes are oriented to increase oleic acid at the expense of linoleic acid and, to a lesser extent, linolenic and palmitic acids. The highest values obtained for oleic acid in the modified genotypes range from 43 to 53%. In this sense, the cultivar BRS Tracajá with 42% oleic acid is interesting because it reached this value without any selection procedure. However, this content is not very different from that found by Vieira et al. (1999), which considered the cultivar Embrapa-4 atypical with oleic acid content higher than the other cultivars (39.9%).

Efforts to change the fatty acid profile of soybeans have privileged the increase of oleic acid due to the importance of this component for the health and biodiesel industry. By means of the conventional genetic improvement, it has been possible to raise the content of this acid up to 70% (Scherder and Fehr 2008). Another way of modifying the fatty acid profile is with the molecular approach by combining mutant alleles of the FAD2-1A and FDA2-1B genes, which can synergistically increase oleic acid levels at the expense of linoleic acids. With this technique it was obtained lines containing the mentioned alleles, in homozygous condition, with 80 to 90% of oleic acid of the total fatty acids (Hoshino et al., 2010; Pham et al., 2010; Pham et al., 2011; Pham et al., 2011; Thapa et al., 2016; Sweeney et al., 2017).



**Table 3.** Profile of fatty acids from cultivar seeds (in bold in the table) and modified soybean genotypes in five environments in Missouri, North Caroline and Mississippi. 2004 (Oliva et al., 2006).

Genótipo	Palmitic	Estearic	Oleic acid	Linoleic	Linolenic
	acid (%)	acid (%)	(%)	acid (%)	acid (%)
IA 3017	11.00	4.20	27.60	56.00	1.10
IA 3018	10.50	3.70	24.20	59.30	2.30
S01-9370	10.40	3.70	20.90	61.30	3.70
C1943	4.00	3.70	34.20	51.20	6.70
S01-9267	4.40	3.00	26.40	58.70	7.30
C1727	15.20	3.60	19.50	53.60	8.10
M23	9.40	3.60	44.90	35.40	6.60
N 97-3393-4	8.50	4.10	49.80	34.80	2.70
N 98-4445A	9.10	4.20	53.30	30.70	2.70
Holl	9.20	3.60	43.50	39.70	4.00
CR03-529	10.90	4.50	38.30	42.80	3.50
MD 00-6605	4.10	3.90	30.80	57.50	3.60
MD 99-5458	4.00	3.90	25.20	62.80	4.10
DKB 38-52	11.20	3.70	23.50	54.90	6.70
MPV 457	10.20	4.40	25.50	52.70	7.20
AG 4902	10.40	4.10	23.70	54.10	7.70
Manokin	11.30	4.40	21.30	55.60	7.31
Cultivars means	10.78	4.15	23.50	54.33	7.22
Non cultivars means	8.52	3.82	33.75	49.54	4.35

## **ACKNOWLEDGEMENT**

The author would like to thank Embrapa and the University of North Carolina for undertaking this work as part of my postdoctoral fellowship. Also to Dr. Andrea Cardinal for being my postdoctoral counterpart and Bill Novinsky from the Laboratory of the Agricultural Research Service of Raleigh-NC for the generous help with the determination of oil and lipid profiles of soybean seeds by nuclear magnetic resonance.



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Received: November 19, 2018. Accepted: December 10, 2018. Published: December 28, 2018.