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Oil quality of passion fruit seeds subjected to a pulp-waste purification process

Qualidade do óleo das sementes do maracujá submetidas ao processo de purificação dos resíduos da polpa

Suelen Alvarenga Regis^I Eder Dutra de Resende^{I*} Rosemar Antoniassi^{II}

ABSTRACT

Passion fruit seeds must be clean and dry before the extraction processing to obtain high-quality oil for edible and cosmetic purposes. This research studies the viability of a cleaning process of seeds by evaluating the oil quality. The research examined 2 maturation stages of the fruit and one purification process of the seeds, compared to the control. The oil quality was evaluated by fatty acid composition, acidity, peroxide value and oxidative stability. The pulp waste suffered a thermal treatment in an alkaline water solution at 60°C for 10min and was further purified in an experimental decanter. In the control treatment, the pulp waste was processed using only water at ambient conditions. The passion fruit seeds were totally cleaned by the thermal/chemical treatment, allowing a faster drying (less than 50% of the drying time) of the seeds and a bit higher yield of oil extraction (proportionally around 7.7%), without changes in quality of the oil.

Key words: *Passiflora edulis*, industrial processing, seed by-products, pulp waste.

RESUMO

As sementes de maracujá devem ser limpas e secas antes do processo de extração para obtenção de um óleo de alta qualidade, para fins comestíveis ou para produtos cosméticos. Este trabalho investigou um processo de purificação das sementes e seu efeito na qualidade do óleo. A pesquisa contemplou dois estádios de maturação dos frutos e um processo de purificação das sementes, comparado com o controle. A qualidade do óleo foi avaliada através da composição de ácidos graxos, acidez do óleo, índice de peróxido e estabilidade oxidativa. O resíduo sofreu um tratamento térmico em solução alcalina, mantida a 60°C por 10min e, posteriormente, foi processado em um decantador experimental. No tratamento de controle, o resíduo da polpa sofreu tratamento em água à temperatura ambiente. Concluiu-se que as sementes foram totalmente limpas por meio do tratamento térmico/químico,

permitindo uma secagem mais rápida das sementes (menos de 50% do tempo de secagem) e havendo um maior rendimento de extração de óleo (proporcionalmente, cerca de 7,7%), sem alteração na sua qualidade.

Palavras-chave: *Passiflora edulis*, processamento industrial, co-produtos das sementes, resíduos da polpa.

INTRODUCTION

The potential value of passion fruit seeds for oil extraction is highlighted by considering the quantity of industrially processed fruit. The total production of passion fruit in Brazil in 2010 reached 920,158t. (IBGE, 2012). It is estimated that approximately 40% of Brazilian production is used for industrial processing (ROSSI, 2001). Thus, the generation of 15,569t. of seeds can be projected, under the assumption that 4.23% of the fruit mass corresponds to dry seeds extracted from the pulp (OLIVEIRA et al., 2011). It is thereby possible to obtain 3,736t. of oil, based on the average content of 24% in passion fruit seeds.

The seeds of passion fruit exhibit an oil content ranging from 18.5% to 29.4%. This oil is composed primarily of linoleic fatty acid (55-66%), oleic acid (18-20%) and palmitic acid (10-14%) and contains a low concentration of linolenic acid (~1.0%). The oil content and its chemical composition are influenced by climatic factors, environment,

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production location, growing conditions and variety (KOBORI & JORGE, 2005, LIU et al., 2008, NYANZI et al., 2005).

Laboratory tests have compared methods of cleaning passion fruit seeds, with a focus on the feasibility of germination studies. Cleaning methods include fermentation (MELETTI, 2000), treatment with acid and alkaline solutions (SILVA, 1998), and friction with sand in sifters (MARTINS et al., 2006).

To extract oil by pressing seeds, it is necessary to remove the arils, which form agglomerates with seeds and promote their fermentation and moisture absorption. One method of removing arils from the pulp residue is to wash the seeds, which must be followed by drying. The high seed moisture content favours the action of lipases, which are enzymes that hydrolyse triglycerides, thereby reducing oil quality. In Brazil, the limits of acidity and peroxide value for crude oil obtained by pressing are up to 4.0mg KOH g⁻¹ and 15meq kg⁻¹, respectively (BRASIL, 2005).

Small- and medium-sized industrial operations that process passion fruit in Brazil work with daily production ranging from 10t. to 100t. The pulp waste has high moisture content and undergoes immediate fermentation when kept in the disposal area. Its utilization requires the use of technologies that ensure the fast processing of large volumes of residue to enable the separation of arils and the purification of seeds, allowing the seeds' rapid drying and storage for oil extraction.

This study aimed to evaluate a process for cleaning the seeds from the pulp residue of passion fruit, focusing on the application of this method for processing large volumes of waste generated in industrial processing, with consideration given to preserving the quality of the seeds and their oil. This study covers the processing of fruits in different stages of maturation as they are normally found in the raw material used by the industry.

MATERIALS AND METHODS

A sample of approximately 135 passion fruits (*Passiflora edulis* Sims) was obtained from a commercial farm at Campos dos Goytacazes-RJ in November/2009. The fruits were harvested in two stages of maturation: an intermediate (well defined yellow spot) and fully ripe. The fruits were washed and sanitised with a solution of chlorinated water (100ppm active chlorine) for 15min; they were then stored at 12°C and 90% relative humidity (RH) prior to processing.

The fruits were analysed for standard skin colour using the Hunter colorimeter (MiniScan Spectrophotometer Colorimeter). The readings were taken at two equidistant points from the sun-exposed and non-exposed faces of the fruit. The results were expressed by the percentage of yellow peel colour based on the Hunter b parameter, as described by SILVA et al. (2008).

After the peel colour characterization the fruits were cut with stainless steel knife to remove the pulp. This pulp was processed in a 600mL beaker with mechanical stirring (Fisatom, model 715, Brazil) at 1,000rpm for 2min. Then, the material was filtered and pressed manually in a synthetic filter cloth with a 1-mm mesh. A 50g sample of this residue was used in each purification treatment, providing an amount of seeds for carrying out 4 replicates of the oil analysis.

Fifteen trials were performed for each cleaning treatment, using three fruits per trial. For the control treatment, 50g samples of the residue were subjected to mechanical agitation (Fisatom, model 715, Brazil) for 10min at 1,000rpm in a beaker containing distilled water with a solid/liquid ratio of 1:7, under ambient temperature. For the thermal/chemical treatment, a mixture of distilled water to which a 1mol L⁻¹ solution of NaOH had been added at a concentration of 4.86% (v/v) (REGIS, 2010); this suspension was then maintained in hydrothermal bath at 60°C under the same stirring conditions and solid/liquid ratio as the Control experiment. After agitation, the material was filtered through synthetic fabric and processed for 5min in an Experimental Decanting Apparatus, using a controlled flow of water to promote the entrainment of arils and the decanting of the seeds. The seeds were collected in a sieve and partially dried under a thin layer of synthetic fabric (1mm mesh) using paper towels, after which they were weighed using a semi-analytical balance (Gehaca, model AG200, Brazil) to identify the seed wet mass. The seeds were then dried in a tray dryer (Pardal, Brazil) at 60°C with forced air circulation. In the Control treatment, the seeds were dried for 2h due to the presence of arils. In the thermal/chemical treatment, the seeds were dried for a shorter period of time (1h). The samples were cooled in desiccators and placed in airtight plastic tubes for storage at -20°C and subsequent oil extraction.

For each trial, seed samples containing 15g were evaluated according to their physiological maturation by applying a standard

colour scale with 4 degrees, after REGIS (2010): 1 - totally white seeds; 2 - white seeds with small brown spots; 3 - light brownish seeds; 4 - dark brownish seeds. The results were expressed as the percentage of seeds exhibiting each pattern of the colour scale.

For the oil analysis, the passion fruit seeds were ground, and oil extraction was conducted with petroleum ether (30-60°C) in a Soxhlet apparatus for 16h. Moisture was removed by heating to 105°C. The acidity and peroxide value of oil were measured according to AOCS Official Methods (AOCS, 2009). Saponification and iodine value were calculated based on the fatty acid composition. Oil Stability Index (OSI) measurements were performed in a Rancimat 679 (Metrohm) at 110°C, using an air flow of 10L h⁻¹ and 5g of sample, according to AOCS (2009). Total carotenoid content analysis was carried out using UV-Visible spectrophotometry (Agilent 8453 Spectrophotometer), with absorbance measured at 452nm, as described by DAVIES (1972).

Methyl esters of fatty acids (FAME) were prepared according to HARTMAN & LAGO (1973). Gas chromatography was performed in a Agilent 6890 chromatograph fitted with a cyanopropylsiloxane capillary column (60m x 0.32mm x 0.25mm) applying the initial column temperature at 100°C and held for 3min; it was increased to 150°C at a rate of 50°C min⁻¹, further increased to 180°C at a rate of 1°C min⁻¹, and finally increased to 200°C at a rate of 25°C min⁻¹, where it was held for 10min. Hydrogen was used as a carrier gas at a flow rate of 2.5mL min⁻¹ (measured at 40°C). A 1.0mL quantity of a 2% dichloromethane solution of the sample was injected, using an injector operating at 250°C in split mode (1:50) and an FID detector held at 280°C. The results were expressed both as a weight percentage (area normalisation) and as a weight per unit weight of the dry material. The identification of FAME was based on a comparison of retention times with those of the NU CHEK standards 62, 79 and 87 (Elysian, MN).

The treatments consisted of one cleaning process applied to fruit residues obtained of fruits in ripe and intermediate stages, compared to the Control treatment applied to fruit residues in ripe stage. The experiment was conducted with 15 trials of cleaning process applied to treat the residues of two fruits that allowed getting one homogeneity sample of seeds necessary to perform all the chemical evaluations. The chemical

analyses were conducted with four replicates, so contemplating the use of 120 fruits per treatment. The results were compared by confidence intervals with 5% significance according to Student's t-test. A sampling dimension of infinite population was employed with a deviation of 10% around the data mean and 95% probability. Statistical analyses were performed using the computer program SAEG (SAEG, 2007).

RESULTS AND DISCUSSION

The physical aspects of the seeds processed in the control treatment and after the thermal/chemical treatment of pulp residues of fruits in the ripe or intermediate stages are illustrated in figures 1a, 1b and 1c, respectively.

The efficiency of the purification process of the seeds, as shown in figure 1, is noteworthy. The separation process in the decanter has taken approximately 10min. Thus, the complete process of separation and purification demanded an operating time of approximately 20min, making it highly advantageous for processing the large volumes that are generated daily in the industry.

The ripe fruits used in the Control treatment and thermal/chemical treatment showed similar colour measurements, with an average of 94.86% yellow peel colour (Table 1). The fruits in the intermediate stage were greener (33.61% yellow peel colour). CERQUEIRA et al. (2011) found that the passion fruit harvested in summer season with 40% yellow peel had not reached physiological maturity adequate to support the ripening process. Otherwise, COELHO et al. (2010) found that the passion fruit harvested in the winter have full physiological maturity when they reach approximately 30% yellow peel. Thus, the fruits in the intermediate stage used in this experiment did not reach full physiological maturity, as they had been harvested in the summer season.

The fruit mass, the seeds moisture and the oil content are presented in table 1. The same amount of crude residue was used in the different treatments; however, the control treatment had a higher average weight of wet seeds than the fruits in the intermediate or ripe stages, whose seed mass did not differ from each other. This was due to the presence of arils retained in the seeds.

The final moisture content of the seeds from the control treatment was 34.75% higher than the average final moisture of the seeds in different stages of maturation that had undergone the thermal/

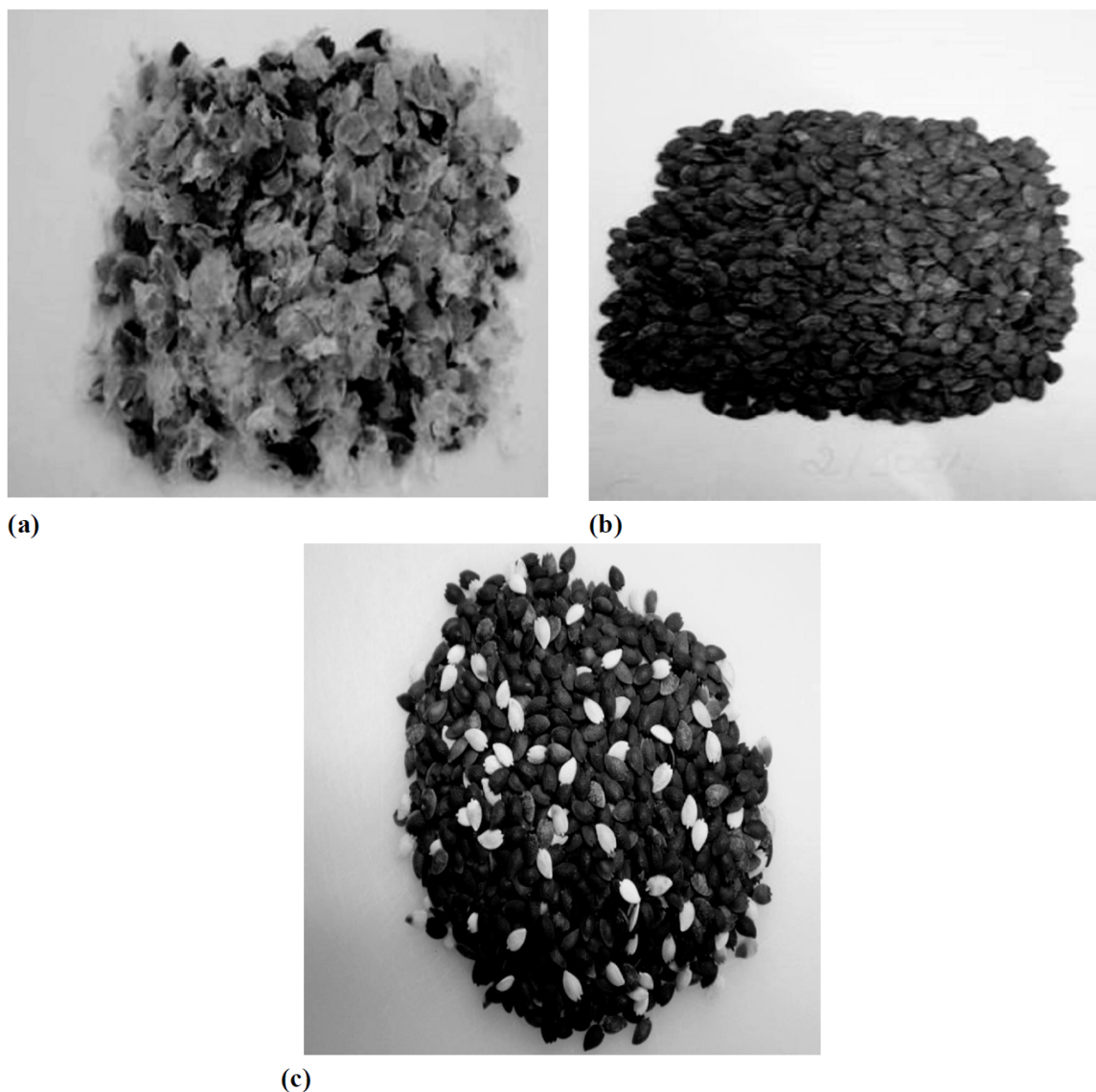


Figure 1 - Physical aspects of the seeds after the two treatments applied to the pulp residues: (a) control treatment in ripe fruit residues (b) thermal/chemical treatment in ripe fruit residues (c) thermal/chemical treatment in unripe fruit residues.

chemical treatment (Table 1). The moisture content of the seeds in the ripe stage was equal to that observed in the intermediate stage, which underwent the same drying conditions.

The seeds processed in the control treatment underwent a drying process two times longer than that employed in the seeds of other treatments, under the same drying conditions, thus emphasising the importance of the purification for accelerating the drying time.

In assessing the physiological status of seeds maturity, the data in table 1 show that fruits of the intermediate stage had higher quantities of seeds exhibiting patterns 3 and lower quantities exhibiting pattern 4, compared with the fruits in the ripe stage. This may have occurred because the fruits in the intermediate stage had not reached full physiological maturity.

The purification of the seeds using alkaline solution and the further entrainment of the arils

Table 1 - Physical and chemical characterisation of fruit, seeds and oil processed through thermal/chemical and control treatments applied to pulp residues of fruits at intermediate and ripe stages. Data represented with different letters in the same lines present statistical differences at $P=0.05$, according to Student's t-test.

| Measurements | Control Treat. (ripe stage) | Thermal/Chemical Treat. (ripe stage) | Thermal/Chemical Treat. (intermediate stage) | Variance Coefficient |
|---|--------------------------------|---|---|-------------------------|
| Fruit peel yellowish (%) | 95.88 ^a | 93.83 ^a | 33.61 ^b | 5.87 |
| Fruit mass (g) | 155.09 ^b | 152.22 ^b | 179.63 ^a | 3.80 |
| Wet seeds (g) | 25.60 ^a | 15.63 ^b | 15.45 ^b | 5.30 |
| Dry seeds (g) | 12.08 ^a | 11.25 ^{ab} | 10.88 ^b | 4.02 |
| Seeds moisture content (%) | 6.10 ^a | 3.98 ^b | 4.47 ^b | 9.77 |
| Seeds maturity (% scale 1*) | - | 0.10 ^a | 0.31 ^a | 125.89 |
| Seeds maturity (% scale 2*) | - | 0.07 ^a | 0.05 ^a | 226.77 |
| Seeds maturity (% scale 3*) | - | 0.58 ^b | 8.25 ^a | 25.57 |
| Seeds maturity (% scale 4*) | - | 99.24 ^a | 91.36 ^b | 1.17 |
| Oil content (% dry basis) | 26.92 ^b | 29.17 ^a | 26.87 ^b | 3.75 |
| Oil acidity (% oleic acid) | 0.75 ^a | 0.72 ^a | 0.74 ^a | 5.62 |
| Saponification index (mg KOH g ⁻¹) | 192.84 ^a | 192.83 ^a | 192.79 ^a | 0.049 |
| Iodine index (g I ₂ 100g ⁻¹) | 132.33 ^{ab} | 134.32 ^a | 130.56 ^b | 1.22 |
| Oxidative stability (h) | 6.25 ^a | 4.52 ^b | 6.05 ^a | 7.35 |
| Carotenes (mg kg ⁻¹) | 27.02 ^a | 3.91 ^b | 4.46 ^b | 27.99 |
| Peroxide index (meq kg ⁻¹) | 0 | 0 | 0 | - |

*1 - totally white seeds, 2 - white seeds with small brown spot, 3 - light brownish seeds, 4 - dark brownish seeds.

through water flow also promoted the removal of most of the immature seeds, especially those with white coloration. This seed entrainment may have contributed to the small difference in the number of immature seeds observed when evaluating the two stages of maturation. However, the process proved to be efficient in the purification of mature seeds that are suitable for the extraction of oil. The seeds of the ripe stage used in the control treatment showed lower oil content than those seeds processed by the thermal/chemical treatment. This indicates that the presence of arils retained in the seeds may have influenced the increase in the dry base weight of the samples, thereby contributing to the smaller proportion of oil in the dry matter (Table 1).

The seeds in the ripe stage did not show difference in oil content as compared to that of the intermediate stage (Table 1). It may be due to the purification process that removed most of the immature seeds in the intermediate stage, thus minimising the mass of immature seeds used in the analysis of oil content (Figure 1c).

KOBORI & GEORGE (2005) reported that passion fruit seeds contain a lipid percentage of approximately 25%. In a comparative study of oil extraction using different varieties of passion fruit, NYANZI et al. (2005) found oil content ranging from 18.5 to 28.3%, with the *Passiflora edulis* var.

flavicarpa having an oil content of 20.6%. However, VIEIRA (2006) found no difference in oil content between the species *Passiflora edulis* (25.62%) and *Passiflora nitida* (26.10%), but both differed significantly from the *Passiflora alata* species, which had lower oil content (20.12%).

For passion fruits produced in the Brazilian cerrado, LOPES et al. (2010) observed that the oil content in seeds of *Passiflora setacea* ranged from 31.2 to 33.5%, followed by *Passiflora nitida* (29.5%), *Passiflora edulis* (27.3%) and *Passiflora cincinnata* (15.3 to 19.3%). The lower oil content of *Passiflora cincinnata* may be related to a thick lignified integument in seeds of this species. The species *Passiflora setacea*, which had the highest oil content, has a thin integument.

The quality of the oil obtained after the purification processes is shown in Table 1. The oil acidity (free fatty acids - FFA) was not influenced by the thermal/chemical treatment for the seeds of fruits in both maturation stages, with an average value of 0.74%. Thus, the treatment with dilute alkaline solution did not cause the seeds to absorb this solution under the conditions of time and temperature to which they were subjected. The free acidity of the oil stems from the partial hydrolysis of glycerides is a variable closely related to the quality of the raw material,

its processing and storage conditions, as well as the purity of the oil (CORREIA, 2009).

High values of FFA for passion fruit seed oil were reported by KOBORI & JORGE (2005), with an average of 7.35%, while LIU et al. (2008) found 2.11%, and FERRARI et al. (2004) obtained 0.67%, approaching the results of this research. According to the Brazilian legislation (BRASIL, 2005) the maximum level of acidity is 4mg KOH g⁻¹, which corresponds to 2% FFA content.

The index of saponification was calculated from the fatty acid composition, showing no detectable differences between the seeds subjected to the thermal/chemical and Control treatments (Table 1). However, there was higher value of the iodine index in the oil of the seeds of ripe fruits compared with seeds in the intermediate stage, both subjected to the thermal/chemical treatment. This difference was due to a tendency towards a higher concentration of linoleic fatty acid (C18:2) (Table 2), once the fatty acid composition was used to calculate the iodine index.

The average iodine value of passion fruit seed oil assessed by FERRARI et al. (2004), 136.5g I₂ 100g⁻¹, was slightly higher than the average of the treatments of this research, 132.4g I₂ 100g⁻¹, possibly due to the slightly higher content of unsaturated fatty acids (87.54%). Measures of saponification indexes were equal for the three treatments, averaging 192.82mg KOH g⁻¹.

The oil of the seeds in ripe stage purified by the thermal/chemical treatment exhibited a 27.7% decrease in oxidative stability value compared to the oil of seeds purified by the control treatment (Table 1). This can be explained by the significant amount of carotenes in the oil of seeds obtained from the Control treatment, which may confer stronger antioxidative power and thereby retard the oxidation. The presence of a high concentration of carotene in the oil of the seeds from the Control treatment was due to arils attached to the seeds, which retain carotenes from the juice.

The oil of seeds in the intermediate stage showed higher oxidative stability than that seeds in the ripe stage, both subjected to the thermal/chemical treatment (Table 1). This can be explained by the lower iodine value and the trend of lower linoleic acid concentration in the seed oil from the fruit in the intermediate stage (Table 2).

It is also noteworthy that a peroxide value of 0 was detected in the oil from the seeds of the fruits in different stages of maturation, even after the thermal/chemical treatment to purify the seeds (Table 1). This shows that this process caused no oxidation effects in the crude oil. In contrast, the peroxide index measured by LIU et al. (2008) was 2.23mmol kg⁻¹, in comparison to the measurements obtained by FERRARI et al. (2004), which showed an average of 4.7meq kg⁻¹.

Table 2 - Fatty acid composition (%) of oil from the passion fruit seeds purified through thermal/chemical and control treatments applied to pulp residues of fruits at intermediate and ripe stages. Data represented with different letters in the same lines present statistical differences at P=0.05, according to Student's t-test.

| Composition | Control Treat (ripe stage) | Thermal/Chemical Treat. (ripe stage) | Thermal/Chemical Treat (intermediate stage) | Sunflower ** | Corn * |
|-------------|-------------------------------|---|--|--------------|--------|
| C14:0 | 0.08 ^a | 0.07 ^a | 0.09 ^a | 0.10 | 0.10 |
| C16:0 | 10.60 ^a | 10.50 ^a | 10.75 ^a | 6.69 | 25.00 |
| C16:0 cis | 0.18 ^a | 0.17 ^a | 0.17 ^a | - | - |
| C17:0 | 0.07 ^a | 0.06 ^a | 0.07 ^a | - | - |
| C18:0 | 3.09 ^a | 3.04 ^a | 3.13 ^a | 2.96 | 2.8 |
| C18:1 | 18.96 ^a | 17.11 ^a | 19.91 ^a | 13.96 | 17.1 |
| C18:2 | 66.27 ^a | 68.34 ^a | 65.12 ^a | 70.65 | 52.7 |
| C18:2 trans | Tr | tr | tr | - | - |
| C18:3 | 0.39 ^a | 0.38 ^a | 0.36 ^a | 0.10 | - |
| C20:0 | 0.16 ^a | 0.16 ^a | 0.16 ^a | 0.19 | - |
| C22:0 | Tr | tr | tr | - | - |
| C24:0 | 0.06 ^a | 0.06 ^a | 0.064 ^a | 0.19 | - |
| unsaturated | 85.80 ^a | 86.00 ^a | 85.57 ^a | - | - |
| saturated | 14.07 ^a | 13.90 ^a | 14.26 ^a | - | - |

Reference: * VIEIRA, 2006 ** PIGHINELLI et al., 2009.
tr: trace.

The fatty acid composition is presented in table 2. It appears that the overall composition of fatty acids was not affected by the thermal/chemical treatment of the crude residue for both maturation stages, compared to the Control treatment.

The main fatty acids present in the oil are as follows, in ascending order: stearic (C18:0), with overall average 3.08%; palmitic, (C16:0) with 10.62%; oleic (C18:1), with 18.66%; and linoleic, (C18:2) with 66.74%. It was noted only traces of trans fatty acids (Table 2). The average content of unsaturated fatty acids (85.79%) was higher than the saturated fatty acids (14.08%). This highlights the chemical and nutritional quality of this oil because it has 66.74% of polyunsaturated fatty acids, a low content of saturated fatty acids, and only traces of trans fatty acids.

In terms of nutrition, passion fruit seed oil is similar to sunflower oil (Table 2), which contains high levels of unsaturated fatty acids, particularly linoleic acid. Furthermore, passion fruit seed oil exceeds the proportion of essential fatty acids contained in major commercial varieties of soybean oil and corn oil.

NYANZI et al. (2005) compared the fatty acid composition of different varieties of passion fruit seed oil, ranging from 85.4 to 88.6% of unsaturated fatty acids. Of these, linoleic fatty acid (C18:2) showed the highest percentage (67.8 to 74.8%), followed by oleic (C18:1), with 13.6 to 16.9%, palmitic (C16:0), with 8.8 to 11%, and stearic (C18:0), with 2.2 to 3.0%, respectively.

According to VIEIRA (2006), the fatty acids that make up the passion fruit seed oil do not differ qualitatively among different species, but they do vary in their relative proportions (%). The relative proportions of the major fatty acids (i.e., linoleic, oleic and palmitic acids) were similar for the species *Passiflora edulis* (67.99, 14.54 and 15.30%) and *Passiflora alata* (63.16, 15.02 and 18.75%), but they were clearly divergent for *Passiflora nitida* (35.53, 28.35 and 28.97%).

CONCLUSION

The passion fruit seeds were totally cleaned by stirring the pulp residue with dilute alkaline solution at 60°C during 10min, allowing a faster drying of the seeds and a bit higher yield of oil extraction.

The fatty acid composition and the oil quality were not affected by the thermal/chemical treatment used for seed purification. However it

caused a bit lower oxidative stability of the oil and higher iodine content due to the cleaning of arils soaked with carotenes.

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