

# Nutritional composition of tamarind (*Tamarindus indica* L.) from the Cerrado of Minas Gerais, Brazil

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## Nutritional composition of tamarind (*Tamarindus indica* L.) from the cerrado of Minas Gerais, Brazil.

**Abstract – Introduction.** The Cerrado is a Brazilian biome that has a large plant heterogeneity. Among the fruit species of the Cerrado, the tamarind stands out due to its economic potential and use in human feeding. Our study evaluated the physical and physicochemical characteristics, and occurrence and content of vitamin C, carotenoids, vitamin E and folates in tamarind (*Tamarindus indica* L.) from the Cerrado of Minas Gerais, Brazil. **Materials and methods.** The length, diameter, mass and fruit yield of tamarind were evaluated. Titratable acidity was determined by volumetric neutralization, pH by potentiometry, soluble solids by refractometry, moisture using an oven, ash using a muffle furnace, protein by the micro-Kjeldhal method, total dietary fiber by the gravimetric non-enzymatic method, and lipids with a Soxhlet extractor. Vitamin C and carotenoids were analyzed by HPLC-DAD, and vitamin E and folates by HPLC with fluorescence detection. **Results and discussion.** Tamarind pulp is composed mainly of carbohydrates ( $50.07 \text{ g}\cdot 100 \text{ g}^{-1}$ ) and moisture ( $35.29 \text{ g}\cdot 100 \text{ g}^{-1}$ ); it can be considered a good source of dietary fiber ( $4.13 \text{ g}\cdot 100 \text{ g}^{-1}$ ). The pH, titratable acidity and soluble solids are 2.95, 18.52 g tartaric acid $\cdot 100 \text{ g}^{-1}$  and 44.00 °Brix, respectively. Contents of vitamin C ( $4.79 \text{ mg}\cdot 100 \text{ g}^{-1}$ ) and folates ( $59.35 \text{ }\mu\text{g}\cdot 100 \text{ g}^{-1}$ ) in the tamarind are higher than those observed in traditional fruits. However, the fruit presents low vitamin E content ( $108.78 \text{ }\mu\text{g}\cdot 100 \text{ g}^{-1}$ ). **Conclusion.** The tamarind stands out due to its nutritional value, being a good source of vitamin C and dietary fiber, and an excellent source of folates.

## Brazil / Minas Gerais / *Tamarindus indica* / fruits / physicochemical properties / carotenoids / vitamin content / energy value

## Composition nutritionnelle du tamarin (*Tamarindus indica* L.) du Cerrado brésilien (Minas Gerais, Brésil).

**Résumé – Introduction.** Le Cerrado est un biome brésilien qui héberge une large diversité végétale. Parmi les espèces fruitières du Cerrado, le tamarin se distingue du fait de son potentiel économique et de son usage dans l'alimentation humaine. Notre étude a évalué les caractéristiques physiques et physico-chimiques, la présence et la teneur en vitamine C, caroténoïdes, vitamine E et folates dans le tamarin (*Tamarindus indica* L.) du Cerrado de l'état de Minas Gerais, au Brésil. **Matériel et méthodes.** La longueur, le diamètre, le poids et le rendement en fruits de tamarin ont été évalués. L'acidité titrable été déterminée par neutralisation volumétrique ; le pH par potentiométrie ; les solides solubles par réfractométrie ; l'humidité à l'aide d'un four ; les cendres à l'aide d'un four à moufle ; les protéines par le procédé micro-Kjeldhal ; les fibres alimentaires totales par un procédé de gravimétrie non enzymatique ; les lipides avec un extracteur Soxhlet. La vitamine C et les caroténoïdes ont été analysés par HPLC-DAD, et la vitamine E et les folates par HPLC avec détection par fluorescence. **Résultats et discussion.** La pulpe de tamarin est composée principalement de glucides ( $50,07 \text{ g}\cdot 100 \text{ g}^{-1}$ ) et d'humidité ( $35,29 \text{ g}\cdot 100 \text{ g}^{-1}$ ) ; elle peut être considérée comme une bonne source de fibres alimentaires ( $4,13 \text{ g}\cdot 100 \text{ g}^{-1}$ ). Le pH, l'acidité titrable et les solides solubles sont de 2,95, 18,52 g d'acide tartrique $\cdot 100 \text{ g}^{-1}$  et 44,00 °Brix, respectivement. Les contenus en vitamine C ( $4,79 \text{ mg}\cdot 100 \text{ g}^{-1}$ ) et folates ( $59,35 \text{ }\mu\text{g}\cdot 100 \text{ g}^{-1}$ ) dans le tamarin sont plus élevés que ceux observés dans les fruits traditionnels. Cependant, le fruit présente une faible teneur en vitamine E ( $108,78 \text{ }\mu\text{g}\cdot 100 \text{ g}^{-1}$ ). **Conclusion.** Le tamarin se démarque des autres production fruitière du Cerrado en raison de sa valeur nutritionnelle ; c'est une bonne source de vitamine C et de fibres alimentaires, et une excellente source de folates.

## Brésil / Minas Gerais / *Tamarindus indica* / fruits / propriété physicochimique / caroténoïde / teneur en vitamines / valeur énergétique

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## 1. Introduction

Brazil, due to its continental dimensions, has a variety of biomes including the Cerrado. Considered the second largest Brazilian biome, the Cerrado occupies a large and continuous area in the states of Goiás, Bahia, Minas Gerais and Mato Grosso, as well as some peninsulas and unconnected areas in other states [1, 2]. The Brazilian Cerrado has a large vegetable heterogeneity that is less known. Native or grown fruit species in the Cerrado play an important role due to their economic potential and utilization as foods. Fruits of the Cerrado have unique sensory characteristics and high nutritional value due to the contents of dietary fiber, vitamins and minerals, as well as other nutrients [3].

Among the fruit species found in the Brazilian Cerrado, the tamarind stands out (*Tamarindus indica* L.). This fruit belongs to the family Leguminosae and is native to tropical Africa; however, it is cultivated throughout the world, especially in tropical countries such as Brazil. In Brazil, the tamarind is found in tropical and subtropical regions; it is an ideal crop for semi-arid regions, especially in areas with prolonged drought such as the Cerrado [4, 5]. The tamarind fruit has many industrial and commercial applications. In folk medicine it is used as a laxative, expectorant, anti-inflammatory and antioxidant. Its pulp, which has a sour and refreshing taste, can be consumed fresh or processed in the form of candy, ice cream, liquor, soft drinks, concentrated juices and seasonings [6].

Studies have indicated that the tamarind contains elevated titratable acidity, is rich in pectin, B-complex vitamins and minerals, and contains carotenoids and vitamin C [7, 8]. Thus, it can be considered an important food in traditional diets [9]. However, these data refer to fruits grown in Africa.

Although residents of the Cerrado consume this fruit, little data is available on the nutritional composition of tamarind from the state of Minas Gerais, Brazil, especially regarding the contents of carotenoids and vitamins obtained using reliable analytical

methods such as high-performance liquid chromatography (HPLC).

Thus, our study evaluated the physical and physicochemical characteristics, the contents of carotenoids and vitamins, and the nutritional value of the tamarind from the Cerrado of Minas Gerais, Brazil.

## 2. Materials and methods

### 2.1. Raw material collection and sample preparation

Tamarind fruits (*Tamarindus indica* L.) were collected during the harvest season (October to March 2010) after dropping naturally from the trees, in a region of the Cerrado located in the municipality of Curvelo, Minas Gerais, Brazil (lat. 18°45' S, long. 44°25' W). To obtain five repetitions, the collection area was divided into sub-areas, and approximately 1.0 kg of fruit was collected from each sub-area.

In the laboratory, the fruits were selected according to the degree of maturation and absence of injuries. Ripe fruits were considered as those with dark brown shells and brittle and reddish pulp. Subsequently, the pulp of the fruits was manually removed using a stainless steel knife. The pulp obtained was blended using a domestic food processor (Faet Multipratic, MC5, Brazil), packed in polyethylene bags and stored at  $(-18 \pm 1)$  °C.

### 2.2. Physical characterization

Individual measurements of length and transverse and longitudinal diameter were carried out on 30 tamarind fruits using a digital caliper rule (Mitutoyo, Brazil). The mass of the whole fruit (MF), pulp (MP), bark (MB) and seeds (MS) was obtained by individual direct weighing on a semi-analytical balance (Gehaka, BG 2000, Brazil). The pulp yield was calculated using the equation  $[(MP / MF) \times 100]$ .

### 2.3. Physicochemical analyses

The physicochemical analyses were performed in three repetitions. Titratable acidity (TA), soluble solids (SS) and pH were determined according to the methodologies proposed by the Instituto Adolfo Lutz [10]; moisture, ash, protein, lipids and total dietary fiber were determined according to the methods of the Association of Official Analytical Chemistry [11]. Carbohydrate concentrations were estimated by the equation:  $[100 - (\% \text{ moisture} + \% \text{ fat} + \% \text{ protein} + \% \text{ total dietary fiber} + \% \text{ ash})]$ . The total energy was estimated considering the conversion factors of  $4 \text{ kcal}\cdot\text{g}^{-1}$  for protein and carbohydrate, and  $9 \text{ kcal}\cdot\text{g}^{-1}$  for lipids [11].

### 2.4. Extraction and analyses of carotenoids and vitamins

The analyses of carotenoids and vitamins were performed in five repetitions. During extraction and analysis, the samples and extracts were protected from both sunlight and artificial light with the use of amber glass bottles, aluminum foil and blackout curtains; they were also protected from oxygen by using lids and environments with nitrogen gas in glass bottles.

#### 2.4.1. Carotenoids

The occurrence and content of the major carotenoids ( $\alpha$ -carotene,  $\beta$ -carotene,  $\beta$ -cryptoxanthin and lycopene) were investigated in the tamarind pulp. Extraction was performed using the method proposed by Rodriguez-Amaya [12], with modifications. About 10.0 g of pulp were weighed in a mortar; then 5.0 g of Celite<sup>®</sup> and 60.0 mL of cooled acetone (divided into three 20.0-mL volumes) were added. After the addition of acetone, the suspension was homogenized manually with the aid of a pistil for approximately 5 min and vacuum-filtered in a Buchner funnel with filter paper. Then, the filtrate was transferred in three fractions to a separatory funnel containing 50.0 mL of cooled petroleum ether. Each fraction was washed with distilled water for complete removal of acetone. Anhydrous sodium sulfate was added to the ether extract to

remove any residual water. Subsequently, the extract was concentrated using a rotary evaporator (Tecnal, TE-211, Brazil) at  $(35 \pm 1) ^\circ\text{C}$ , transferred to a volumetric flask and the volume completed to 25.0 mL with petroleum ether. This extract was stored in a hermetically sealed amber glass bottle and stored at  $(-18 \pm 1) ^\circ\text{C}$ .

For analysis, ten-mL aliquots of the extract were evaporated under a flow of nitrogen gas, and the dry residue redissolved in 2.0 mL of HPLC-grade acetone (Tedia, Brazil). This extract was filtered through HV Millex filter units made of polyethylene, with  $0.45 \mu\text{m}$  of porosity (Millipore, Brazil), and 50.0  $\mu\text{L}$  was injected into the HPLC for analysis. Carotenoid analysis was carried out by HPLC using the chromatographic conditions developed by Pinheiro-Sant'Ana *et al.* [13], with modifications: HPLC system (Shimadzu, SCL 10AT VP, Japan) comprised of a high-pressure pump (Shimadzu, LC-10AT VP, Japan), autosampler with 50  $\mu\text{L}$  loop (Shimadzu SIL-10AF, Japan) and diode array detector (DAD) (Shimadzu SPD-M10A, Japan); chromatographic column, Phenomenex Gemini RP-18 (250 mm  $\times$  4.6 mm, 5 mm), fitted with a guard column, RP-18 Phenomenex ODS column (4 mm  $\times$  3 mm), the mobile phase consisting of (methanol:ethyl acetate:acetonitrile) (70:20:10, v/v) with a flow of  $2.0 \text{ mL}\cdot\text{min}^{-1}$  and a run time of 15 min. Chromatograms were obtained at 450 nm.

#### 2.4.2. Vitamin C

Extraction and analysis of ascorbic acid (AA), and conversion of dehydroascorbic acid (DHA) into AA were performed according to the methods proposed by Campos *et al.* [14], with modifications. In the extraction, about 5.0 g of pulp were homogenized for about 3.0 min in 15.0 mL of the extraction solution composed of ultrapure water added to metaphosphoric acid, 8% acetic acid,  $\text{H}_2\text{SO}_4$  0.3 N and 1 mM EDTA. The suspension obtained was centrifuged (Fanem, Excelsa Baby II - 206R, Brazil) at 4.000 rpm (1.789 g) for 15.0 min, vacuum-filtered in a Buchner funnel with filter paper, and diluted to 25.0 mL in a volumetric flask with ultrapure water. Subsequently, the suspension was centrifuged at 14.000 rpm (21.913 g) for

5.0 min, and the supernatant stored under refrigeration [ $(5 \pm 1) ^\circ\text{C}$ ] until analysis.

For conversion of dehydroascorbic acid into ascorbic acid, a 2.0-mL aliquot of the extract obtained in the previous stage was pipetted into an amber glass bottle and supplemented with 0.8 mL of a 1.2 M Trizma buffer solution (pH 9.0) containing 40 mM DTT (Sigma-Aldrich, Germany) to raise the pH to near neutrality (pH 6.0). For conversion of dehydroascorbic acid into ascorbic acid, the extract was kept at rest for 10.0 min at room temperature and away from light. Subsequently, the pH of the extract was reduced to 2.0 by adding 0.9 mL of 0.4 mM  $\text{H}_2\text{SO}_4$ .

The ascorbic acid analyses were performed by injection of 50.0  $\mu\text{L}$  of the extract previously filtered in filter units with a porosity of 0.45  $\mu\text{m}$ . Analyses of vitamin C were performed using the same HPLC system used for analysis of carotenoids and the chromatographic conditions used were: HPLC-DAD system, chromatographic column: RP-18 Lichrospher 100 (250 mm  $\times$  4 mm, 5  $\mu\text{m}$ ), the mobile phase consisting of ultrapure water with 1 mM  $\text{NaH}_2\text{PO}_4$ , 1 mM EDTA and pH adjusted to 3.0 with  $\text{H}_3\text{PO}_4$ , and a mobile phase flow rate of 1.0  $\text{mL}\cdot\text{min}^{-1}$ . Chromatograms were obtained at 245 nm [14]. The dehydroascorbic acid content was calculated by the equation: [DHA content = AA content after conversion – AA content before conversion].

### 2.4.3. Vitamin E

Extraction of the eight components of vitamin E ( $\alpha$ ,  $\beta$ ,  $\gamma$  and  $\delta$ -tocopherols and tocotrienols) was performed according to Pinheiro-Sant'Ana *et al.* [15]. Roughly 10.0 g of pulp were weighed and supplemented with 4.0 mL of heated ultrapure water [about  $(80 \pm 1) ^\circ\text{C}$ ], 10.0 mL of isopropyl alcohol, 1.0 mL of hexane (Tedia, Brazil) containing 0.05% of butylhydroxytoluene, and 5.0 g of anhydrous sodium sulfate. Gradually, 25 mL of the extraction solvent (hexane:ethyl acetate, 85:15, v/v) was added to the mixture. After these procedures, the suspension was homogenized using a micro-crusher at average speed for 1.0 min. Once homogenized, the samples were vacuum-filtered through

a Buchner funnel fitted with filter paper; the residue was maintained in the extraction tube. The extraction step was repeated, adding 5.0 mL of isopropyl alcohol and 30.0 mL of the solvent mixture, with subsequent homogenization and vacuum filtration. Then the extract was concentrated on a rotary evaporator at  $(70 \pm 1) ^\circ\text{C}$  for about 2 min, transferred to a volumetric flask and the volume completed to 25.0 mL with the solvent mixture.

After extraction, 5.0 mL aliquots of the extracts were dried in nitrogen gas, redissolved in 2.0 mL of HPLC-grade hexane and filtered through filter units with porosity of 0.45  $\mu\text{m}$ . Analyses of the different components of vitamin E were performed by HPLC with injection of 50.0  $\mu\text{L}$  of the extract in a HPLC system (Shimadzu SCL 10AD VP, Japan), comprising a high-pressure pump (Shimadzu LC-10AD VP, Japan), autosampler with a 50.0  $\mu\text{L}$  loop (Shimadzu SIL-10AF, Japan) and fluorescence detector (Shimadzu, RF10AXL, Japan). The chromatographic conditions used for the analysis included: HPLC system; fluorescence detection (excitation at 290 nm and emission at 330 nm); Luna chromatography column (Phenomenex Si60, 250 mm  $\times$  4 mm, 5  $\mu\text{m}$ ), fitted with a guard column (Phenomenex Si60, 4 mm  $\times$  3 mm); a mobile phase composed of (hexane:isopropanol:acetic acid), in the proportions (98.9:0.6: 0.5) and a mobile phase flow rate of 1.0  $\text{mL}\cdot\text{min}^{-1}$ . The total content of vitamin E was calculated by sum of the components of vitamin E identified in the samples.

### 2.4.4. Folates

The occurrence and content of three folate forms (THF, 5-MTHF and 5-FTHF) in the tamarind pulp were investigated according to Della Lucia *et al.* [16], with some modifications. Approximately 5.0 g of the pulp were homogenized in 20.0 mL of 0.1 M phosphate buffer, pH 6.0, containing 1.0% of ascorbic acid and 0.1% of 2-mercaptoethanol. The suspension was centrifuged at 4,000 rpm (1.789 g) for 15.0 min, vacuum-filtered in a Buchner funnel with filter paper, and diluted to 25.0 mL in a volumetric flask with ultrapure water. Then, the extract was heated for about 12.0 min in a water bath

at  $(100 \pm 1)$  °C and cooled in an ice bath until reaching a temperature below  $(37 \pm 1)$  °C. The cooled extract was centrifuged again at 4.000 rpm (1.789 g) for 15 min and submitted to deconjugation of the polyglutamates into monoglutamates. For deconjugation, one hundred  $\mu$ L of rat plasma containing conjugase ( $\gamma$ -glutamyl carboxypeptidase) were added to 3.0 mL of the previously obtained supernatant, and the extract was incubated in a water bath at  $(37 \pm 1)$  °C for 3 hours. Then, the extracts were heated in boiling water for 5.0 min to inactivate the enzymes.

Extract purification was performed using an ion-exchange column with a stationary phase of Q-Sepharose Fast Flow (Pharmacia, USA). The column was pre-conditioned with methanol (Tedia, Brazil) and water (1:1) at a flow rate of 2.0 drops per second. The extract was applied to the column at a flow rate of 2.0 drops per second. Then, retained folates were eluted in 1.5 mL of sodium acetate (0.1 M) containing 10% NaCl, 1% ascorbic acid and 0.1% 2-mercaptoethanol. Analyses were performed by injection of 50  $\mu$ L of the extracts previously filtered through filter units with a porosity of 0.45  $\mu$ m in the same system used for analysis of vitamin E, complemented with a mobile phase degassing system utilizing helium (Shimadzu DGU-2, Japan). The chromatographic conditions used for analysis included: HPLC system; ShimPack 100 RP18 chromatography column (150 mm  $\times$  4.6 mm, 4.6  $\mu$ m) (Merck, Germany), and a mobile phase composed of phosphate buffer (30 mM  $\text{NaH}_2\text{PO}_4$ , pH adjusted to 2.3 with  $\text{H}_3\text{PO}_4$ ) as eluent A and acetonitrile as eluent B. The binary gradient utilized was as follows: from 0 min to 5 min, 94% of eluent A + 6% of eluent B; from 5 min to 25 min, linear gradient to 75% of eluent A + 25% of eluent B; from 25 min to 33 min, 75% of eluent A + 25% of eluent B; from 33 min to 35 min, return to initial conditions followed by a stabilization period of up to 50 min. The mobile phase flow rate was 0.7 mL $\cdot$ min<sup>-1</sup> and fluorescence detection (excitation at 290 nm and emission at 360 nm). The mobile phase was degassed with helium gas for 15 min at 100 kPa

before initiating the analyses and at 50 kPa during the runs [16].

## 2.5. Identification and quantification of carotenoids and vitamins

Identification of the compounds was performed by comparing the retention times obtained for standards and samples analyzed under the same conditions. In addition, carotenoids and ascorbic acid were identified by comparison of the absorption spectra of the standards and samples using the DAD, and the folates and vitamin E by co-chromatography.

Quantification of the compounds was carried out using an external standardization curve. Appropriate dilutions were made from the standard solutions in order to achieve concentrations comparable with those observed in the tamarind pulp. For this, solutions of each compound present in the pulp (ascorbic acid,  $\alpha$ -tocopherol,  $\beta$ -tocopherol,  $\gamma$ -tocopherol, THF, 5-MTHF and 5-FTHF) were prepared.

Construction of the standard curves was performed by injection, in duplicate, of six increasing concentrations of the standard solutions in the range from (0.0589 to 5.8800)  $\mu$ g for ascorbic acid, (0.0010 to 0.1042)  $\mu$ g for  $\alpha$ -tocopherol, (0.0037 to 0.1120)  $\mu$ g for  $\beta$ -tocopherol, (0.0035 to 0.1040)  $\mu$ g for  $\gamma$ -tocopherol, (0.00004 to 0.04622)  $\mu$ g for THF, (0.00001 to 0.01077)  $\mu$ g for 5-MTHF, and (0.00003 to 0.03312)  $\mu$ g for 5-FTHF. Thus, there was a linear correlation between the peak areas and the concentrations of each compound injected.

The compounds presented in the tamarind pulp were quantified based on the analytical curves and regression equations for ascorbic acid ( $y = 1394501.207x - 117382.946$ ,  $R^2 = 0.998$ ),  $\alpha$ -tocopherol ( $y = 93284137.0896x + 47566,8009$ ,  $R^2 = 0.999$ ),  $\beta$ -tocopherol ( $y = 69128704.3544x - 12630.4206$ ,  $R^2 = 0.997$ ),  $\gamma$ -tocopherol ( $y = 234829959.333x + 731230.429$ ,  $R^2 = 0.995$ ), THF ( $y = 942240050.58x - 162371.44$ ;  $R^2 = 0.996$ ), 5-MTHF ( $y = 1237294689.67x - 259476.97$ ,  $R^2 = 0.998$ ) and 5-FTHF ( $y = 710036264.81x - 1088694.36$ ,  $R^2 = 0.996$ ). The real concentration was determined by calculations based on the dilutions utilized.

## 2.6. Quality control of the analytical methods

Recovery tests, linearity, repeatability, limit of detection (LOD) and limit of quantification (LOQ) were performed for quality control of the analytical methods used in the analysis of vitamins.

Recovery tests were carried out by addition of standards (ascorbic acid,  $\alpha$ -tocopherol,  $\beta$ -tocopherol,  $\gamma$ -tocopherol, THF, 5-MTHF and 5-FTHF) to the samples. The quantity of standard added varied between 50% and 100% of the initial content observed in tamarind pulp. The recovery percentage was calculated using the equation: % recovery = [(final content of compound – content of standard added) / (initial content of compound)]  $\times$  100. All procedures were performed in triplicate.

The linearity range of compounds was determined by injection, in duplicate, of six standard solutions with different concentrations using the same chromatographic conditions employed for extract analysis. Data obtained for the peak areas were used for linear regression analysis. The coefficient of determination ( $R^2$ ) obtained in each case was used to evaluate the linearity [17].

Repeatability tests were performed by extraction and analysis, in quintuplicate, of the same repetition containing the compounds identified. The repeatability was evaluated by calculating the relative standard deviation (RSD) of the peak areas and retention times of the analyzed components [17].

Evaluation of the LOD was performed by successive dilutions of standards followed by determination of the smallest detectable amount. The LOD was established as 3 times the amplitude of the baseline noise and the LOQ as 10 times the LOD [18].

## 2.7. Categorization of tamarind as a source of vitamins

The categorization of 100 g of tamarind pulp as a source of vitamins for children (aged 4–8 years), pregnant women and adult men (aged 19–30 years) was performed using

the criteria proposed by Philippi [19], which classifies food as a source of nutrients when supplying 5–10% of the Dietary Reference Intake (DRI); good source when supplying 10–20% of the DRI, and excellent source when supplying more than 20% of the DRI.

## 2.8. Experimental design and statistical analysis

A completely randomized design with five repetitions (different lots of fruit) was used for analyses of carotenoids and vitamins, with three repetitions for physicochemical analyses. Data was stored in spreadsheets using the Microsoft Office Excel software system, version 2007. Calculation of descriptive statistics (means, standard deviations and range of parameters) was performed for each parameter. To assess the linearity range of analytical standards, the data obtained for the peak areas were used for linear regression analysis and to calculate the coefficient of determination ( $R^2$ ). Statistical analysis was performed using SAS software (Statistical Analysis System), version 9.2 (2008), licensed to UFV.

## 3. Results and discussion

### 3.1. Physical characterization

The tamarind presented elongated pods, with a brown, woody and brittle shell. Inside the fruit was a reddish and viscous pulp, containing between two and six seeds (*figure 1*).

The longitudinal and transversal diameter of the fruits varied from (1.30 to 1.70) cm and (2.50 to 3.00) cm, respectively. Length varied between (3.70 and 11.10) cm. This range was higher than that observed by Sousa [20] in tamarind from the Cerrado of the state of Paraíba, Brazil [(7.37 to 9.22) cm]. Tamarinds from Minas Gerais presented masses of the whole fruit, pulp, shell and seeds of (9.30, 2.49, 1.91 and 4.85) g, respectively. The pulp yield observed in this study (51.25%) was similar to that observed

by Kumar and Bhattacharya [21] (55%) in tamarind grown in India.

### 3.2. Physicochemical characterization

Tartaric acid is the main acid in tamarind pulp. The presence of this acid is uncommon in fruits and its metabolic origin is unknown [22]. Thus, the titratable acidity (TA) of the tamarind pulp was expressed in grams of tartaric acid per 100 g of pulp. Tamarinds from Minas Gerais state have similar TA to that observed by Gurjão [4] in tamarinds from the Brazilian state of Paraíba (17.2 g of tartaric acid·100 g<sup>-1</sup>) (table I).

The soluble solids (SS) content of tamarinds from Minas Gerais state was lower than that observed in tamarinds grown in Florida, USA (54 °Brix to 70 °Brix) [23], and in northeastern Brazil (63 °Brix to 70 °Brix) [24, 25]. In contrast, this content was higher than that observed by Canuto *et al.* [26] (24 °Brix) in fruits from the Brazilian Amazon. The contents of SS correlate with the sugar and organic acid concentrations, which are parameters of interest for the marketing of fresh fruit due to the preference of consumers for sweet fruits [23]. Thus, this high SS content cannot be attributed only to the high sugar content, but also to the high content of titratable organic acids of the fruit.

The [SS/TA] ratio is a parameter that correlates with tamarind quality in terms of ripeness and flavor; it indicates the balance between sugars and organic acids [27]. The [SS/TA] ratio found in tamarinds from the Cerrado of Minas Gerais was low, but similar to that observed by Gurjão [4] (2.2) in tamarinds from the state of Paraíba.

Fruits evaluated in our study showed a slightly higher pH than those observed by Canuto *et al.* [26] (2.5). This pH was lower than that observed in citrus fruits such as oranges (3.66) and ponkan tangerine (4.05) [28].

### 3.3. Chemical characterization

Data related to macronutrient contents of the tamarind pulp grown in the Cerrado of



**Figure 1.** Photographic representation of tamarind fruits (*Tamarindus indica* L.).

Minas Gerais are scarce. The moisture and lipid contents observed in the tamarind pulp were higher than those of fruits collected from other Brazilian states (22.00 g·100 g<sup>-1</sup> and 0.5 g·100 g<sup>-1</sup>, respectively) [29], and in the United States (31.4 g·100 g<sup>-1</sup> and 0.6 g·100 g<sup>-1</sup>, respectively) [30]. These contents were lower than those of fruits from Nigeria [(14.81 and 1.03) g·100 g<sup>-1</sup>, respectively] [31].

Protein content of the tamarind from Minas Gerais was higher than that reported in fruits from other regions of Brazil and the United States (3.2 g·100 g<sup>-1</sup> and 2.8 g·100 g<sup>-1</sup>, respectively) [29, 30], and higher than those of fruits from Africa [(3.5 and 7.64) g·100 g<sup>-1</sup>] [31]. The differences in chemical compositions of the fruits evaluated in this study and fruits from the United States, Africa and other Brazilian states can be attributed to differences in the climate and soil of the sampling sites, which can affect the composition of a food.

The ash content observed in our present studies was higher than that observed in fruits collected in Africa and Brazil [(1.69 and 1.9) g·100 g<sup>-1</sup>, respectively] [31, 29]. The carbohydrate content was lower than that of fruits collected in Africa (56.00 g·100 g<sup>-1</sup>) [31], Brazil (72.5 g·100 g<sup>-1</sup>) [30] and the United States (62.5 g·100 g<sup>-1</sup>) [29].

The content of total dietary fiber of tamarind grown in Minas Gerais was lower than

**Table 1.** Physicochemical characteristics and total energy of the pulp of tamarind (*Tamarindus indica* L.) from the Cerrado (Curvelo, Minas Gerais, Brazil). Values are expressed in fresh matter (mean of three repetitions  $\pm$  standard deviation).

| Soluble solids<br>(°Brix) | Titratable acidity<br>(g tartaric acid·100 g <sup>-1</sup> ) | pH                 | Soluble solids /<br>titratable acidity | Moisture            | Ash                | Proteins           | Total dietary fiber<br>(g·100 g <sup>-1</sup> ) | Lipids             | Carbohydrates       | Total energy value<br>(kcal·100 g <sup>-1</sup> ) |
|---------------------------|--|--------------------|--|---------------------|--------------------|--------------------|---|--------------------|---------------------|---|
| 44.00<br>$\pm$ 4.25       | 18.52<br>$\pm$ 0.77  | 2.95<br>$\pm$ 0.10 | 2.38<br>$\pm$ 0.33                     | 35.29<br>$\pm$ 1.41 | 2.37<br>$\pm$ 0.06 | 6.09<br>$\pm$ 0.40 | 4.13<br>$\pm$ 0.01                              | 5.04<br>$\pm$ 0.13 | 50.07<br>$\pm$ 1.60 | 270.00<br>$\pm$ 6.21                              |



that reported in the Brazilian Table of Food Composition [29] ( $72.5 \text{ g}\cdot 100 \text{ g}^{-1}$ ) and verified by Amoo and Atasié ( $18.83 \text{ g}\cdot 100 \text{ g}^{-1}$ ) [31]. However, the content we found was higher than that observed by Murugan *et al.* ( $1.82 \text{ g}\cdot 100 \text{ g}^{-1}$ ) in fruits collected in Africa [32]. This content was also lower than that observed in other legumes, such as bean, soybean, lentil and pea [ $5.1$  to  $30.3 \text{ g}\cdot 100 \text{ g}^{-1}$ ] [33]. On the other hand, the fiber content of tamarind was similar to that observed in orange ( $4.0 \text{ g}\cdot 100 \text{ g}^{-1}$ ) and higher than that observed in tangerine ( $3.1 \text{ g}\cdot 100 \text{ g}^{-1}$ ), fruits traditionally known as high-fiber [29]. Tamarind may be considered a good source of dietary fiber, since consumption of 100 g of tamarind pulp can supply 10% to 20% ( $13.76 \text{ g}$  per day) of the fiber recommendation for adult men [33].

### 3.4. Carotenoids and vitamins

#### 3.4.1. Quality of analytical methods

Tests for quality control demonstrated that the analysis conditions were reliable, which reduced the possibility of vitamin losses during extraction and analysis, and allowed the detection of small concentrations of the compounds analyzed (table II).

In the repeatability test, the compounds presented a relative standard deviation (RSD) of the peak areas and retention times ranging between 0.00% and 3.00%. The detection limit and the quantification limit of the compounds analyzed by fluorescence ( $\alpha$ -tocopherol,  $\beta$ -tocopherol,  $\gamma$ -tocopherol, THF, 5-MTHF and 5-FTHF) ranged from ( $0.002$  to  $0.054$ )  $\mu\text{g}\cdot\text{mL}^{-1}$  and ( $0.021$  to  $0.530$ )  $\mu\text{g}\cdot\text{mL}^{-1}$ , respectively. The linearity range for each compound analyzed was large and presented coefficients of determination ( $R^2$ ) greater than 0.995. The recovery percent of the vitamin standards ranged from 90.1% to 96.4%.

#### 3.4.2. Qualitative composition

Typical chromatograms of the analyses of vitamins in tamarind from the Cerrado of Minas Gerais showed that, in the pulp, ascorbic acid (retention time - RT: 3.19 min),  $\alpha$ -tocopherol (RT: 5.76 min),  $\beta$ -tocopherol

(RT: 8.77 min),  $\gamma$ -tocopherol (RT: 9.98 min), tetrahydrofolate (RT: 8.08 min), 5-methyltetrahydrofolate (RT: 10.51 min) and 5-formyltetrahydrofolate (RT: 20.03 min) were detected. The carotenoids, dehydroascorbic acid,  $\delta$ -tocopherol, and  $\alpha$ -,  $\beta$ -,  $\gamma$ -, and  $\delta$ -tocotrienol were not identified in the pulp.

#### 3.4.3. Vitamin content

Studies on the contents of vitamin C, vitamin E and folates in fruits of the Cerrado are scarce in the literature. Furthermore, these data were not obtained by reliable analytical methods such as HPLC.

The unique component of vitamin C observed in tamarind was ascorbic acid (table III). The tamarinds from Minas Gerais showed higher vitamin C content than that of fruits grown in the Brazilian Amazon ( $0.1 \text{ mg}\cdot 100 \text{ g}^{-1}$ ) [26]. This difference can be attributed to differences in the climate and soil of the sampling sites, as well as the more reliable analytical method used in this study. The ascorbic acid content of the tamarind was also higher than that found in traditional fruits such as apple ( $1.50 \text{ mg}\cdot 100 \text{ g}^{-1}$ ) and grape ( $1.90 \text{ mg}\cdot 100 \text{ g}^{-1}$ ) [30], and lower than that observed in fruits from the Cerrado of Minas Gerais, such as jatobá (*Hymenaea stigonocarpa*) ( $8.91 \text{ mg}\cdot 100 \text{ g}^{-1}$ ) [34] and araticum (*Annona crassiflora* Mart.) ( $5.2 \text{ mg}\cdot 100 \text{ g}^{-1}$ ) [35].

The tamarind pulp showed low total vitamin E content, which mainly consists of  $\alpha$ -tocopherol (85.6%). This vitamin E content was lower than that found in fruits commonly consumed by the population such as strawberry ( $410.00 \mu\text{g}\cdot 100 \text{ g}^{-1}$ ), banana ( $150.00 \mu\text{g}\cdot 100 \text{ g}^{-1}$ ) and grape ( $540.00 \mu\text{g}\cdot 100 \text{ g}^{-1}$ ) [36], and in fruits of the Cerrado including araticum ( $494.04 \mu\text{g}\cdot 100 \text{ g}^{-1}$ ) [35], mangaba (*Hancornia speciosa*) ( $2732.47 \mu\text{g}\cdot 100 \text{ g}^{-1}$ ) [37], jatoba (*Hymenaea stigonocarpa*) ( $495.54 \mu\text{g}\cdot 100 \text{ g}^{-1}$ ) [30] and pequi (*Caryocar brasiliense*) ( $170.81 \mu\text{g}\cdot 100 \text{ g}^{-1}$ ) [38].

The main folate observed in the tamarind pulp was 5-methyltetrahydrofolate, which, according to Jastrebova *et al.* [39], is the major component in fruits and vegetables. Folate content in tamarind pulp was four times higher than that reported in the USDA

**Table II.** Repeatability, limit of detection and quantification, linearity range and recovery of vitamins in the pulp of tamarind (*Tamarindus indica* L.) from the Cerrado (Curvelo, Minas Gerais, Brazil).

| Compound                 | Repeatability                            |   | Detection limit<br>( $\mu\text{g}\cdot\text{mL}^{-1}$ ) | Quantification limit<br>( $\mu\text{g}\cdot\text{mL}^{-1}$ ) | Linearity range<br>( $\mu\text{g}$ ) | Recovery<br>(%) |
|--------------------------|--|---|---|--|--------------------------------------|-----------------|
|                          | Peak area<br>Relative standard deviation | Retention time<br>Relative standard deviation |   |  |                                      |                 |
| Ascorbic acid            | 2.47                                     | 1.27  | 12.321  | 123.214  | 0.0589 – 5.8800                      | 91.3            |
| $\alpha$ -tocopherol     | 2.19                                     | 2.76  | 0.025   | 0.251  | 0.0010 – 0.1042                      | 95.5            |
| $\beta$ -tocopherol      | 3.00                                     | 1.41  | 0.054   | 0.530  | 0.0037 – 0.1120                      | 94.9            |
| $\gamma$ -tocopherol     | 3.22                                     | 0.93  | 0.025   | 0.252  | 0.0035 – 0.1040                      | 93.5            |
| Tetrahydrofolate         | 1.58                                     | 0.70  | 0.003   | 0.031  | 0.00004 – 0.04622                    | 90.1            |
| 5-methyltetrahydrofolate | 1.10                                     | 0.00  | 0.002   | 0.024  | 0.00001 – 0.01077                    | 95.7            |
| 5-formyltetrahydrofolate | 1.21                                     | 0.05  | 0.002   | 0.021  | 0.00003 – 0.03312                    | 96.4            |

table [30] for fruits of the same species collected in the United States ( $14.00 \mu\text{g}\cdot 100 \text{g}^{-1}$ ), and higher than in other fruits of the Cerrado such as cagaita ( $25.74 \mu\text{g}\cdot 100 \text{g}^{-1}$ ) [40], araticum ( $27.36 \mu\text{g}\cdot 100 \text{g}^{-1}$ ) [35] and pequi ( $5.16 \mu\text{g}\cdot 100 \text{g}^{-1}$ ) [38]. The folate content of tamarind was similar to that observed in beans ( $59.00 \mu\text{g}\cdot 100 \text{g}^{-1}$ ), and lower than that observed in the soybean ( $375.00 \mu\text{g}\cdot 100 \text{g}^{-1}$ ) and lentil ( $433.00 \mu\text{g}\cdot 100 \text{g}^{-1}$ ) [30].

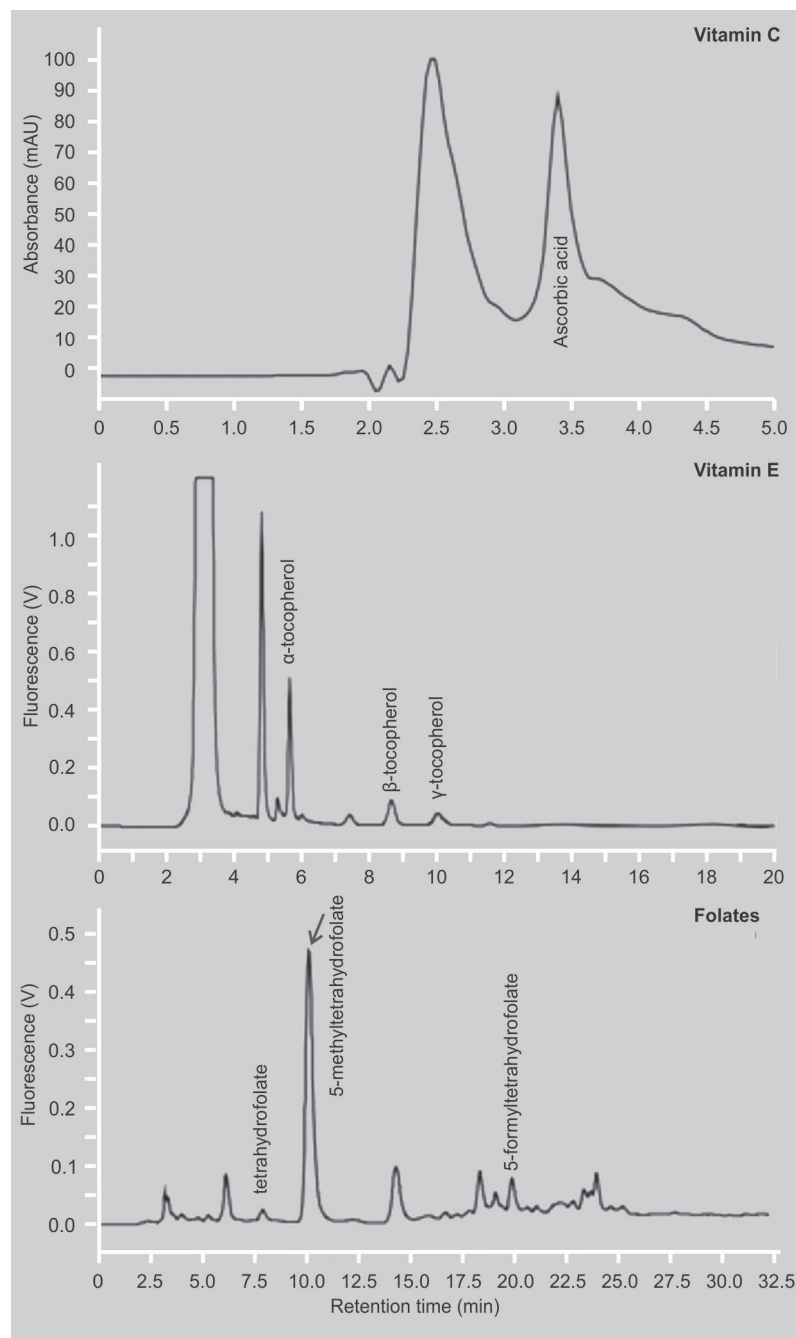
#### 3.4.4. Nutritional value of tamarind pulp as a source of vitamins

According to the criteria for classification of foods as a source, good source and excellent source of a nutrient [19], it was verified that the consumption of 100 g of tamarind pulp can supply 5% to 19% of the recommended daily allowance (RDA) of vitamin C for the different age groups (table IV). Thus, it can be considered a good source of vitamin C for children and adult men, and a source for pregnant women. The tamarind proved to be an excellent source of folates for children, good source for adult men, and excellent source for pregnant women. Due to the low content of vitamin E, the tamarind contributes little to supplying the recommendations of this vitamin (table IV).

## 4. Conclusion

Tamarind pulp studied in Minas Gerais, Brazil, showed a high content of soluble solids and titratable acidity, and low pH. It can also be considered a good source of total dietary fiber. This fruit was shown to be a good source of vitamin C for children and a source for adult men. The vitamin E content was lower than that observed in some fruits traditionally consumed by the Brazilian population.

Tamarind pulp proved to be a source of fiber for adults and a good or excellent source of folates for different age groups. Due to its nutritional value, the tamarind can contribute to supplying the nutritional needs of fiber, vitamin C and folates; this justifies its consumption *in natura* or processed, especially by families living in the Cerrado.



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## Figure 2.

HPLC analysis of vitamin C, vitamin E and folates in tamarind pulp (*Tamarindus indica* L.) from the Cerrado (Curvelo, Minas Gerais, Brazil). Chromatographic conditions are shown in the *Materials and methods* section.

**Table III.**

Content of carotenoids and vitamins in the pulp of tamarind (*Tamarindus indica* L.) from the Cerrado (Curvelo, Minas Gerais, Brazil). Values are expressed as fresh weight.

| Compound                  | mean of five repetitions $\pm$ standard deviation |                             | %      |
|---------------------------|---|-----------------------------|--------|
|                           | mg·100 g <sup>-1</sup>                            | $\mu$ g·100 g <sup>-1</sup> |        |
| Carotenoids               | Not detected                                      | –                           | –      |
| Vitamin C (ascorbic acid) | 4.79 $\pm$ 0.96                                   | –                           | 100.00 |
| Vitamin E                 | –   | 108.78 $\pm$ 18.25          | 100.00 |
| $\alpha$ -tocopherol      | –   | 93.16 $\pm$ 16.19           | 85.60  |
| $\beta$ -tocopherol       | –   | 10.89 $\pm$ 2.76            | 10.00  |
| $\gamma$ -tocopherol      | –   | 4.73 $\pm$ 1.20             | 4.30   |
| Folates                   | –   | 59.35 $\pm$ 9.86            | 100.00 |
| 5-methyltetrahydrofolate  | –   | 41.87 $\pm$ 5.92            | 70.50  |
| Tetrahydrofolate          | –   | 12.21 $\pm$ 2.13            | 20.60  |
| 5-formyltetrahydrofolate  | –   | 5.27 $\pm$ 1.31             | 8.90   |

**Table IV.**

Contribution of 100 g of the pulp of tamarind (*Tamarindus indica* L.) to supplying the daily recommendations of vitamins for children, pregnant women and adult men.

## a) Content per serving (100 g)

| Vitamin C<br>(mg) | Vitamin E<br>( $\mu$ g of $\alpha$ -tocopherol) | Folates<br>( $\mu$ g) |
|-------------------|---|-----------------------|
| 4.79              | 93.16   | 59.35                 |

## b) Percentage of intake adequacy

| Age group      | Vitamin C | Vitamin E | Folates |
|----------------|-----------|-----------|---------|
| Children       | 19.16     | 1.32      | 29.68   |
| Adult men      | 5.32      | 0.61      | 14.84   |
| Pregnant women | 5.63      | 0.73      | 9.89    |

Calculation based on the Recommended Dietary Allowance (RDA) of the Dietary Reference Intakes (DRIs) for the respective age groups and nutrients [31–43].

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### Composición nutricional del tamarindo (*Tamarindus indica* L.) del Cerrado brasileño (Minas Gerais, Brasil).

**Resumen – Introducción.** El Cerrado es un bioma brasileño que alberga una gran diversidad vegetal. Entre las especies frutales del Cerrado, el tamarindo se distingue por su potencial económico y su uso en la alimentación humana. Nuestro estudio evaluó las características físicas y físico-químicas, la presencia y el contenido en vitamina C, carotenoides, vitamina E y folatos en el tamarindo (*Tamarindus indica* L.) del Cerrado del estado de Minas Gerais, en Brasil. **Material y métodos.** Se evaluó la longitud, el diámetro, el peso y la producción de frutos del tamarindo. Se determinó la acidez valorable mediante neutralización volumétrica; el pH mediante potenciometría; los sólidos solubles por refractometría; la humedad con un horno; las cenizas con un horno de panadero; las proteínas por el procedimiento de micro-Kjeldhal; las fibras alimentarias totales por un procedimiento de gravimetría no enzimática; y los lípidos con un extractor Soxhlet. La vitamina C y los carotenoides se analizaron mediante HPLC-DAD, y la vitamina E y los folatos mediante HPLC con detección por fluorescencia. **Resultados y discusión.** La pulpa del tamarindo está compuesta principalmente de glúcidos ( $50,07 \text{ g} \cdot 100 \text{ g}^{-1}$ ) y de humedad ( $35,29 \text{ g} \cdot 100 \text{ g}^{-1}$ ), y puede considerarse como una buena fuente de fibras alimentarias ( $4,13 \text{ g} \cdot 100 \text{ g}^{-1}$ ). El pH, la acidez valorable y los sólidos solubles son de 2,95, 18,52 g ácido tartárico  $\cdot 100 \text{ g}^{-1}$  y 44,00 °Brix, respectivamente. El contenido en vitamina C ( $4,79 \text{ mg} \cdot 100 \text{ g}^{-1}$ ) y folatos ( $59,35 \text{ } \mu\text{g} \cdot 100 \text{ g}^{-1}$ ) del tamarindo son superiores al observado en frutas tradicionales. Sin embargo, la fruta presenta un bajo contenido en vitamina E ( $108,78 \text{ } \mu\text{g} \cdot 100 \text{ g}^{-1}$ ). **Conclusión.** El tamarindo se desmarca de otras producciones frutales del Cerrado debido a su valor nutricional; es una buena fuente de vitamina C y de fibras alimentarias, y una excelente fuente de folatos.

**Brasil / Minas Gerais / *Tamarindus indica* / frutas / propiedades fisicoquímicas / carotinoides / contenido vitamínico / valor energético**

