Bioactive amines in non-conventional edible plants from Brazil: health benefits and concerns

Bruno Martins Dala-Paula
bruno.paula@unifal-mg.edu.br

Federal University of Alfenas

Angélica Pereira Todescato
Federal University of Alfenas

José Eduardo Gonçalves
Universidade Federal de Minas Gerais

Maria Beatriz Abreu Glória
Universidade Federal de Minas Gerais

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Abstract

Some bioactive amines in foods are associated with beneficial health effects, but also with food poisoning and intolerance. This study aimed to investigate the levels of nine bioactive amines in ten fruits and vegetables from Brazil using HPLC-FL. Total amine levels ranged from 3.24-58.83mg/kg in serralha and seriguela, respectively. Tryptamine was not detected in any sample. The highest spermidine level was found in seriguela (13.41mg/kg). Putrescine was the prevalent amine in custard apple, acerola, and ora-pro-nobis; spermidine, in jabuticaba, starfruit, and kale; tyramine, in seriguela and marolo. Histamine was detected only in ora-pro-nobis, serotonin, in starfruit, and high agmatine levels were found in acerola, starfruit and serralha. Based on amine levels, seriguela, marolo, custard apple, acerola, jabuticaba, and kale could be dietary sources of polyamines, and starfruit a source of serotonin. However, individuals in use of monoamine-oxidase inhibitors may consider limiting the consumption of seriguela and marolo to avoid adverse effects.

Introduction

Presently, humanity confronts a global syndemic distinguished by the concurrent emergence of epidemics related to obesity, malnutrition, and climate change. This necessitates the implementation of multi-faceted measures across diverse domains [1]. Among them, the adoption of a sustainable and healthy diet, with a reduction in the consumption of red meats and processed foods high in sugar, along with an increase in the consumption of fruit, vegetables, and plant-based protein sources, has been widely encouraged [2]. Another trend is to diversify diets [3]. The Food Guide for the Brazilian Population highlights that a healthy diet should come from environmentally, economically, and socially sustainable food systems, thus encouraging local, organic, and agroecological food consumption [4].

Encouraging the consumption of local food production and non-conventional edible plants (NCEP) is a measure aimed at diversifying the diet, enriching it with bioactive compounds and promoting sustainability, reducing the consumption of ultra-processed foods and those derived from intensive agriculture [1, 3]. The term NCEP is commonly used to describe plants that are native, exotic, spontaneous, wild, or cultivated, which are not part of agricultural commodities and are traditionally consumed or used therapeutically in certain regions or cultures, and they have significant economic and nutritional potential [5]. This definition also comprises food plants with unusual processing methods and generally have no market value or are only sold on a small scale [6]. Brazil encompasses/has a vast biodiversity of NCES, which still lacks studies regarding their bioactive compound composition, such as bioactive amines (BA) [5].

BA are characterized by low molecular mass aliphatic, cyclic, or heterocyclic organic bases that present high biological activity and are essential for human health [7, 8]. Spermidine (SPD) is a polyamine that performs numerous physiological and biochemical functions, including autophagy and apoptosis induction, macromolecule stabilization, such as DNA, RNA, and proteins, transcription as well as antioxidant and anti-inflammatory activity [9, 10]. Furthermore, population studies have associated diets
rich in polyamines, especially SPD, with a reduction in all-cause mortality and an increase in human longevity [11, 12]. SPD may also be related to skin rejuvenation by increasing collagen and elastin expression as well as lipid synthesis in aged skins [13]. Agmatine (AGM) has been reported to be a biological marker of autistic spectrum disorders, and it also shows neuroprotective, anticonvulsant, anxiolytic, and antidepressant effects [14]. Putrescine (PUT) is a mandatory intermediate for SPD formation, and its exogenous administration in rats has been associated with a reduction in seizure duration [15].

Histamine (HIM) is involved in mediating inflammatory processes and immune cell differentiation, vasodilation, vascular permeability, gastric acid production, circadian cycle regulation, congestion, bronchospasm, and respiratory tract secretion [16]. Tyramine (TYM) has vasoconstrictor properties and anti-inflammatory effects by inhibiting chemokine production [17]. Its urinary levels have been inversely related to inflammatory markers and cardiometabolic risk factors [18]. Additionally, it stimulates lipolysis by activating glucose transporters in human adipocytes [19]. However, increased intake of HIM and TYM can lead to adverse effects due to food intoxication or in individuals predisposed to intolerance [16]. According to the European Food Safety Authority, no adverse effect level (NOAEL) was established as 600 mg TYM per person per meal (healthy individuals). However, if an individual is taking third generation monoamine oxidase inhibitor (MAOI) or classical MAOI drugs, NOAEL decreases to 50 and 6 mg/meal, respectively [20]. Healthy individuals exposed to 25 to 50 mg of HIM per meal haven’t shown adverse health effects. However, for individuals with HIM intolerance, even trace amounts of ingested HIM can trigger adverse effects. Therefore, only levels below detectable limits can be deemed safe [20].

Cadaverine (CAD) is a diamine found in only a few foods, such as certain types of mushrooms, some plants, and primarily in legumes [21]. CAD and PUT can enhance the toxic effect of HIM by significantly reducing its degradation rate by diamine oxidase (DAO) [22]. 2-Phenylethylamine (PHM) induces pleasurable sensations, improving mood and protecting against neurodegenerative diseases and depression [23]. Tryptamine (TRM) acts as a neurotransmitter and exhibits antioxidant properties [24] and anti-inflammatory effects by suppressing lipopolysaccharide-induced inflammation in a murine macrophage model [25].

Considering the effects of the nine presented BA on human health and longevity, as well as the implications of a diverse diet in locally produced fruit and vegetables for both human health and planetary sustainability, this study aimed to determine the levels of nine BA (PUT, CAD, AGM, SPD, HIM, TYM, PHM, TRM, SRT) in fruit and vegetables, most of which are classified as NCEP in Brazil.

**Materials And Methods**

2.1 Samples

Fruit and vegetables (Table 1) were purchased from a traditional open-air market in Alfenas, MG, Brazil (−21.42467, −45.94997 coordinates), considered an intangible heritage of the municipality, which takes place every Sunday morning. They were randomly gathered (2 kg fruits and 600 g leafy vegetables),
consisting of three lots each. The samples consisted of fully developed and ripe fruits and vegetables, with size, flavor, and color characteristic of consumption standards.

Table 1 near here

All fruit and vegetables were washed thoroughly with potable running water for 3 minutes. *Marolo*, custard apple, and *jabuticaba* had their peels removed, and the pulp was manually extracted with the assistance of a stainless-steel knife from the seeds. The pulps of *seriguela*, *acerola*, and starfruit were obtained along with their skins, after removing the seeds. All the pulps and the leaves of the vegetables (*ora-pro-nobis*, kale, *almeirão-roxo*, and *serralha*) were ground separately in a food processor (Walita Master Plus, Walita, São Paulo, SP, Brazil) and thoroughly homogenized.

2.2 Reagents

BA standards were purchased from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA), including spermidine trihydrochloride, putrescine dihydrochloride, agmatine sulfate, cadaverine dihydrochloride, 5-hydroxytryptamine (SRT), histamine dihydrochloride, tyramine hydrochloride, 2-phenylethylamine hydrochloride and tryptamine. And *o*-phthalaldehyde was also purchased from Sigma-Aldrich Chemical Co. All reagents were of analytical grade, except for HPLC solvents, which were of chromatographic grade. Ultrapure water was obtained from a Milli-Q System (Millipore Corp., Milford, MA, USA). The mobile phases were filtered through HAWP and HVWP membranes (47 mm diameter and 0.45 µm pore size, Millipore Corp., Milford, MA, USA), used for aqueous and organic solvents, respectively.

2.3 Determination of bioactive amines

Amines were extracted from 5 g samples with 7 mL 5% (w/v) trichloroacetic acid (TCA). After vortexing for 5 minutes, the slurry was centrifuged at 10,000 x g for 20 minutes at 4 °C in a centrifuge (Hitachi-Himac CR21, Kioto, Japan), and the supernatant was collected. The solid residue was extracted twice more with 7 and 6 mL of 5% TCA. The supernatants were combined, and the total volume was registered and filtered through a 0.45 µm pore size membrane [8].

Amines were separated by ion-pair reverse phase HPLC and quantified by fluorescence after post-column derivatization with *o*-phthalaldehyde [8]. Liquid chromatography was performed with an LC-10 AD system connected to an RF-551 spectrofluorimetric detector at 340 and 445 nm of excitation and emission, respectively, and to a CBM-10 AD controller (Shimadzu, Kyoto, Japan). A reversed-phase µBondapak C18 column (300 x 3.9 mm i.d., 10 µm) was used with a µBondapak C18 guard-pak insert (Waters, Milford, MA). The mobile phases were (A) solution of 0.2 M sodium acetate and 10 mM 1-octanesulfonic acid sodium salt adjusted to pH 4.9 with acetic acid and (B) acetonitrile. The flow rate was 0.8 mL/min, and the gradient was: 13 minutes at 11% B; 19 minutes at 30% B; 24 minutes at 11% B; and 45 minutes at 11% B. The post-column derivatization reagent was delivered at 0.4 mL/min. It consisted of 1.5 mL Brij-35, 1.5 mL mercaptoethanol and 0.32 g *o*-phthalaldehyde dissolved in 500 mL solution of 25 g of boric acid and 22 g of KOH (pH adjusted to 10.5 with 30 g/L KOH). The column and
the post-column reaction apparatus were kept at 23 ± 1 °C. Amines were identified by comparison of retention times of the amines in the samples to those of the amines in standard solutions and by adding the suspected amine to the sample. The method was evaluated using performance metrics to confirm its linearity, limit of detection, and limit of quantification. To assess linearity, three analytical curves were prepared with concentrations of 0.1, 0.2, 0.4, 0.8, 1.0, 2.0, 4.0, 8.0, and 12 µg/mL. All other parameters were determined by Vale and Gloria [26]. Quantification was accomplished by direct interpolation in external standard curves, and levels were expressed in mg/100 g.

2.4 Statistical analysis

All results were submitted to the Ryan-Joiner normality test. The data that followed a parametric distribution were submitted to one-way analysis of variance (ANOVA), and the means were compared by Tukey’s test at 95% significance and those that followed a non-parametric distribution were submitted to Kruskall-Wallis’, and the median were compared by Dunn’s test. All statistical analysis were performed by Prism 5 Version 5.03 (GraphPad Software, Inc.).

Two multivariate exploratory techniques, principal component analysis (PCA) and hierarchical cluster analysis (HCA), were used for fruit and vegetable characterization regarding BA content. In PCA, the means of all detectable amines were used as variables for the principal components, with the matrix type set as covariance. The dendrogram for HCA was obtained by clustering the variables (the same ones used in PCA). McQuitty linkage was applied to the distance matrix, and the Euclidean method was used to calculate the distance between observations (Minitab® 16.2.3).

Results And Discussion

3.1 Occurrence of bioactive amines in conventional and non-conventional edible plants

This study presents, for the first time, the profile of BA in some NCEP (seriguela, marolo, custard apple, jabuticaba, ora-pro-nobis, almeirão-roxo, and serralha) and also in tropical plants (starfruit, acerola and kale). The linearity of the analytical curve, as well as the limits of detection and quantification obtained by the analytical method employed for the quantification of nine bioactive amines in this study, were adequate and are presented in Table 2.

Of the nine investigated BA, six were found in most of the fruit and vegetables (SPD, AGM, PUT, CAD, TYM, and PHM), whereas SRT was exclusively detected in one fruit (starfruit), and HIM in one vegetable (ora-pro-nobis). TRM was < LOD (0.05 mg/kg) in all samples (Figure 1). SPD and PUT were detected in all samples, followed by PHM and CAD, which were detected in 60% and 43.3% of the samples, respectively. AGM and TYM were detected in 30% of the samples, whereas HIM was detected in one vegetable sample (10%), and SRT, in one fruit sample (10%). These results suggest that SPD and its biosynthetic pathway precursor PUT are the most widely found amines in these fruit and vegetables.
PUT is a diamine that can be synthesized through two pathways. The most common pathway in eukaryotes involves the decarboxylation of ornithine, while the other pathway, found in bacteria and some plants, is characterized by the decarboxylation of arginine, generating AGM, followed by the removal of one ammonia molecule from N-carbamoyl putrescine [27]. The high occurrence of SPD and PUT in vegetables is widely described in the literature [28-31]. In general, cereals, legumes, and soy derivatives have the highest SPD levels [31]; however, citrus fruits [32, 33], and banana, pumpkin, tomato [33], and papaya [29] contain higher PUT levels.

![Figure 1 near here](image1)

Starfruit had the most diversified amine profile among fruit, with six amines, while *ora-pro-nobis* and *serralha* presented five amines, making them the vegetables with the most diversified profile. SPD and PUT were the prevalent amines (both in similar levels between them) in starfruit and *serralha*, and PUT, in *ora-pro-nobis*. On the other hand, custard apple, *jabuticaba*, and kale were the products with the least diverse amine profile. SPD was the prevalent amine in *jabuticaba* and kale, and PUT, in custard apple. TYM was detected in *seriguela*, *marolo* and *ora-pro-nobis*, and HIM, only in *ora-pro-nobis*. The presence of HIM was described in a few vegetables, including eggplant, tomato, spinach, chard, asparagus [34], *jiló* (*Solanum gilo* Raddi), green onion, and bean sprouts [8]. *Seriguela* and *marolo* had TYM as the predominant amine.

### 3.2. Bioactive amine levels in conventional and non-conventional edible plants

The total BA levels in fruit and vegetables (Figure 2) varied from 6.82 up to 58.83 mg/kg in fruit and from 3.24 to 9.63 mg/kg in vegetables. In general, fruit showed higher total amine levels compared to vegetables, with the highest levels in *seriguela* (58.83 mg/kg), followed by *marolo* (~46 mg/kg), whereas *almeirão-roxo* and *serralha* had the lowest. Kale and *ora-pro-nobis* were the vegetables with the highest total amine levels (~8.8-9.6 mg/kg), and the lowest total levels were found in *serralha* and *almeirão-roxo* (~3.2-3.8 mg/kg).

![Figure 2 near here](image2)

High total amine levels (>20.0 mg/kg) have been reported in some other tropical fruits, such as passion fruit (75.3 mg/kg), pineapple (21.0 mg/kg) [29], and banana (65.14 mg/kg) [33]. The total amine levels found in the fruit and vegetables included in this study were below 10 mg/kg, which is similar to literature values for most vegetables [28, 30, 35], except for parsley (32.5 mg/kg), spinach (31.6 mg/kg) and green onion (14.9 mg/kg), which had higher amine levels [8].

*Seriguela, marolo, kale, jabuticaba* and custard apple were the samples with the highest SPD levels, whereas kale was the highest in vegetable. Lettuce (44.0 mg/kg), passion fruit (30.5 mg/kg), mango (30.0 mg/kg), broccoli (23.9-41.3 mg/kg), parsley (22.0 mg/kg) and spinach (13.1 mg/kg) are also considered high sources of SPD [28, 29, 31, 35, 36]. The lowest SPD level in fruit was found in starfruit. *Serralha* was the vegetable with the lower SPD level (Table 3). AGM is a less investigated amine in
foods, and it was reported to have lower occurrence in fruit and vegetables [8, 29]. AGM has been found in pineapple, papaya, passion fruit (≤1.2 mg/kg), and in broccoli, cauliflower, jiló, green onion, and spinach (nd-8.4 mg/kg). The presence of AGM in acerola, starfruit, and serralha indicates that the formation of this amine is possible in these samples, allowing us to believe that PUT synthesis pathway could be through arginine. Based on the new evidence of the role of AGM in health [14, 37], efforts should be made to expand research on AGM levels in other fruit and vegetables.

<Table 3 near here>

The highest PUT levels were found in custard apple and marolo (~15 mg/kg), whereas the lowest levels were found in kale (0.66 mg/kg). High contents of this diamine were also reported in the literature for orange (110.0 mg/kg), mango (>80.0 mg/kg), lime (41.0 mg/kg), pear (24.0 mg/kg) [35], bean sprouts (31.9 mg/kg) [8] and passion fruit (9.9-84.1 mg/kg) [36]. Similar levels compared to custard apple and marolo were reported for banana (12.3-15.3 mg/kg) and tomato (10.6-14.9 mg/kg) [8, 35], and lower levels, for papaya (10.1 mg/kg), pumpkin (6.6 mg/kg) and spinach (4.48 mg/kg) [29, 33, 35]. Recent studies have shown the effects of exogenous PUT administration in different vegetables to mitigate the effects of abiotic stressors, such as drought [38], high temperatures [39], and salinity [40]. According to Simon-Sarkadi et al. [41], storing certain leafy vegetables such as Chinese cabbage, endive, iceberg lettuce, and radicchio at a refrigeration temperature (5 °C) for more than 5 days resulted in an increase in PUT levels by 3 to 8 times. Considering the high solubility of PUT in water, its levels in vegetables subjected to cooking can be reduced due to leaching [31].

CAD was detected in 50% of the products, e.g., in fruits, such as acerola and starfruit, and in vegetables, such as ora-pro-nobis, almeirão-roxo, and serralha, with levels ranging from 0.15 to 0.94 mg/kg. High levels were reported in bean sprouts (40.6 mg/kg) [8] and cabbage (6.03 mg/kg). CAD is less predominant in plant compared to SPD and PUT [35]. HIM was only found at low level in ora-pro-nobis (1.25 mg/kg). High TYM levels were detected in seriguela and marolo. TYM was found in seriguela at 41.56 mg/kg, which is higher compared to levels reported in some alcoholic beverages, including beer and wine (6.31 and 2.42 mg/kg, respectively). In general, HIM and TYM are not widespread in fruit and vegetables but can be a characteristic of certain members of the Solanaceae family (e.g., eggplant, jiló, and tomato, with HIM ranging from 1.1 to 83.2 mg/kg and TYM ranging from 1.8 to 2.7 mg/kg) and spinach, both at low levels, below 10 mg/kg [8, 34].

Of the NCEP analyzed in this study, seriguela and marolo can contribute significantly to TYM intake and lead to adverse health effects. The adverse effects of TYM intoxication include palpitations, pain, headaches, and migraine attacks, which can occur between 30 minutes up to a couple of hours after consuming a meal containing TYM [16]. NOAEL can be reached and exceeded if moderate portions of such fruit were eaten raw or even as dessert or juice after a meal containing fermented and ripened food products such as cheese, sausage, alcoholic beverages, salami, and sausage [16]. Sensitive individuals should avoid or limit seriguela and marolo intake to prevent TYM toxicity. The approximate consumption of 144 g of seriguela or 329 g of marolo in natura would be sufficient to reach NOAEL for TYM for an
individual using classical MAOI. One must also consider that these fruits are also used in the preparation of juice, jams, and sweets. Healthcare professionals, especially doctors and nutritionists, should be aware of these risk situations to properly advise the population. TYM intoxication in a healthy individual would be unlikely, considering the large quantity of fruit needed to reach 600 mg of this amine. Therefore, seriguela and marolo consumption should not be discouraged, especially considering that these unconventional fruits are important sources of SPD, a polyamine associated with reduced mortality from cardiovascular diseases and all causes [12]. However, sensitive individuals and those taking MAOI drugs may prefer consumption of TYM-free vegetables.

PHM was detected in four fruits and two vegetables at low levels, ranging from 0.16 to 0.59 mg/kg. The highest level was found in seriguela, and lower levels were found in almeirão-roxo, serralha and starfruit. Custard apple, jabuticaba, kale and ora-pro-nobis did not have this neuroactive amine. Seriguela had a level twice as high as marolo, acerola and starfruit, while the levels quantified in vegetables were similar (~0.16 mg/kg). PHM are mostly found in fermented foods and beverage because of microbial decarboxylase activity, such as salami, wine, fermented foods, soy food products and yogurt [42, 43]. However, few non-fermented foods also contain this neuroactive amine [44]. Moret et al. [28] reported PHM in peeled tomato, lentil, red beet (2.0 mg/kg) and bean (1.0 mg/kg), which are higher compared to the products investigated in this study. PHM act as catecholamines-releasing agent, a family of hormones and neurotransmitters derived from tyrosine, an amino acid, regulating the neuroendocrine-immune response to stress [44]. PHM is also a stimulator of the hypothalamus, inducing pleasurable sensations and enhancing mood lifting and sexual drive [8, 45].

SRT at level of 1.01 mg/kg was only detected in ora-pro-nobis. Some fruit and vegetables can be sources of SRT. Adão and Glória [46] found that SRT was the predominant amine in bananas, with level around 17.5 mg/kg, higher than SPD and PUT levels. Some other fruits and vegetables have been described as sources of SRT, including tomatoes, spinach (>34.0 mg/kg), kiwi, pineapple, avocado, green onion (<10 mg/kg and >5.0 mg/kg), strawberries, oranges, passion fruit (<5.0 mg/kg and >1.0 mg/kg), sweet cherries, peaches, grapes, apples, pears, and watermelon (<1.0 mg/kg) [45-50].

It is important to highlight that unconventional fruit and vegetables are less studied compared to agricultural crops considered commodities. However, unconventional fruit and vegetables are widely consumed both in diets and in traditional medicine by populations who cultivate them in their backyard gardens, urban vertical gardens, and rural farms [5]. Unconventional fruit and vegetables have great potential to diversify diets and contribute as sources of bioactive compounds, such as polyamines and other BA.

3.3 Distribution of bioactive amines by type of vegetable

Marolo and custard apple are from the same botanic family (Annonaceae), but different species and the same edible part was analyzed, the pulp. Similar SPD and PUT levels were present in both fruits; however, TYM and PHM were detected only in marolo, which presented total amine levels ~ twice as high as marolo. TYM was the most prevalent amine in marolo, and PUT, in custard apple. Similarly, almeirão-
roxo and serralha are members of the Asteraceae family and they contain similar profile and total amine level. However, AGM was found in serralha, but it was absent in almeirão-roxo (Figure 1 and Table 3).

Custard apple, jabuticaba and kale were the fruits and vegetable with the least diversified profile of amines; however, these samples had a significant total BA content. Kale had the highest mean total amine levels among leafy vegetables, which are 3 times higher than serralha and 2.5 times than almeirão-roxo. Seriguela was the fruit with the highest total amine levels, which are 8.6 times higher than starfruit, 5.2 times higher than jabuticaba, 3.5 times higher than acerola and 2.5 times higher than custard apple(Figure 2 and Table 3).

TYM was the most prevalent amine in seriguela and marolo, PUT, in custard apple, acerola, starfruit, ora-pro-nobis, almeirão-roxo and serralha, and SPD, in jabuticaba and kale. SPD levels in almeirão-roxo and serralha were very similar to PUT levels in same vegetables. Among the 16 different vegetables (lettuce, arugula, spinach, parsley, broad bean, potato, carrot, onion, fennel, pepper, zucchini, broccoli, savoy cabbage, cauliflower, cucumber and tomato) analyzed by Moret et al. [27], the highest TYM level was 12.0 mg/kg in arugula. Sánchez-Pérez et al. [34] reported TYM levels of 27 different vegetables and its products and 23 fruits and its products. They observed the highest levels in (green) beans (2.46 mg/kg) and in plum (4.02 mg/kg). Comparing the reported literature levels for fruit and vegetables, TYM levels for seriguela and marolo were considerably higher. Seriguela, marolo, jabuticaba and custard apple had higher SPD levels compared to several fruits, such as peach, banana, kiwi, papaya, fig, lime, pineapple, pear, peach, apple, mandarin, strawberry, and can be considered important sources of this polyamine for a diverse diet [29, 30, 35]. Kale had the highest SPD level among the leafy NCEP in this study; therefore, it could be an important product for a polyamine-rich diverse diet. However, SPD level in kale is still lower than those reported for lettuce, arugula, spinach, parsley, celery, and green onion [28, 35].

According to Madeo et al. [9], an SPD-rich diet can protect from a range of age-associated pathologies, thereby contributing to a longer life span and healthier aging. A diet rich in SPD also exerts beneficial effects on cognitive performance in animal (flies, mice) and humans, due to this polyamine's effects on functional autophagy and mitophagy, allowing elevated mitochondrial function in neuronal tissue [51]. Therefore, all the fruit and vegetables analyzed, with less significance of ora-pro-nobis, almeirão-roxo and serralha, are relatively good sources of this polyamine, and their consumption should be encouraged. However, seriguela and marolo are NCEP sources of TYM, and their consumption, especially of seriguela, which have high TYM content, can lead to adverse health effects, particularly for sensitive individuals taking MAOI drugs. They should, therefore, be avoided by these individuals.

No information on BA was available in the literature for conventional and NCEP presented in this research; therefore, this information fills a gap regarding these products, typical of the Brazilian diet. Moreover, most of the studies investigated only a few amines, whereas nine BA were simultaneous investigated in this study.

3.4. Differentiation of fruit and vegetables based on multivariate analysis
The HCA dendrogram (Figure 3.A) differentiated three groups (green connectors: starfruit, *ora-pro-nobis*, *almeirão-roxo* and *serralha*; purple connectors: *jabuticaba* and kale; red connectors: custard apple and *acerola*). In addition, the dendrogram indicated that, even in the same group, some vegetables had higher similarities among themselves, such as in the first group (green connectors). *Almeirão-roxo* and *serralha* were similar, and they are also from the same botanical family: *Asteraceae*. Similarly, starfruit and *ora-pro-nobis* are more similar between themselves. Even though *marolo* and custard apple are members of the same botanical family (*Annonaceae*), they were not clustered together regarding BA levels.

Multivariate analysis of automatically scaled data indicated that a two-principal component (PC) model explained 64.7% of the variance and confirmed previous dendrogram findings (Figure 3.B). PCA separated *seriguela* and *marolo* from the other conventional and NCEP studied (Figure 3.A). According to PC1 loadings (Figure 3.C), TYM, SPD, PHM and total amines were the components with the highest positive impact, whereas CAD and HIM were the ones with the negative highest impact. PC2 explained 21.5% of the variance. SRT and AGM were the ones with the highest positive impact, and CAD and HIM, with the highest negative impact. Leafy vegetables, *ora-pro-nobis*, *almeirão-roxo*, *serralha*, and a non-conventional fruit, starfruit, were in the same cluster (Figure 3.B - green rectangle), characterized by low SPD, total amine and TYM levels. Even though kale is a leafy vegetable, and *jabuticaba*, a non-conventional fruit, they were clustered together with respect to the profile of only two amines: SPD and PUT (Figure 3.B – purple circle). The last cluster was constituted by *acerola* and custard apple (Figure 3.B – red rectangle), characterizing intermediate SPD, PUT and total amine content levels.

<Figure 3 near here>

**Conclusion**

Eight out of the nine amines investigated were found in some conventional and NCEP from Brazil. SPD and PUT were the most frequently found amine (100% of samples), followed by PHM and CAD. TYM and AGM were found only in 30% of the samples, and HIM and SRT were only present in *ora-pro-nobis* and starfruit, respectively.

PUT was the predominant amine in custard apple, *acerola*, starfruit and *ora-pro-nobis*, SPD, in *jabuticaba* and kale, and TYM, in *seriguela* and *marolo*. *Almeirão-roxo* and *serralha* had similar levels of PUT and SPD. However, the highest SPD levels were found in *seriguela*, *marolo*, *jabuticaba* and kale compared to other fruits and vegetables. The conventional and NCEP studied can contribute to a diverse diet rich in polyamines and other BA.

High TYM levels were found in *seriguela* and *marolo*, and their consumption could be worrisome for sensitive individuals, in special those taking MAOI drugs. Similarly, HIM was detected in *ora-pro-nobis* and, therefore, should be avoided by HIM-intolerant individuals.
Abbreviations

AGM  agmatine
ANOVA  analysis of variance
BA  bioactive amines
CAD  cadaverine
DAOI  diamine oxidase inhibitor
FL  fluorescence detector
HCA  hierarchical cluster analysis
HIM  histamine
HPLC  high performance liquid chromatography
MAOI  monoamine oxidase inhibitor
NCEP  non-conventional edible plants
NOAEL  no adverse effect level
PCA  principal component analysis
PHM  2-phenylethylamine
PUT  putrescine
SPD  spermidine
SPM  spermine
SRT  5-hydroxytryptamine
TCA  trichloroacetic acid
TRM  tryptamine
TYM  tyramine

Declarations

Statements and Declarations
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**CRediT authorship contribution statement - BMDP**: conceptualization, methodology, investigation, data curation, formal analysis, original draft preparation, funding acquisition, writing-reviewing and editing, supervision, project administration. **APT**: investigation, data curation. **JEG**: investigation, methodology, data curation, writing-reviewing and editing. **MBAG**: conceptualization, methodology, investigation, writing-reviewing and editing, funding acquisition, supervision, project administration.

**Chemical compounds studied in this article:**


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**Tables**

Tables 1-3 is available in the Supplementary Files section.

**Figures**
Figure 1

Average profile (in percentage) of bioactive amines identified in fruits and vegetables. TYM: tyramine; PUT: putrescine; CAD: cadaverine; HIM: histamine; SRT: serotonin; AGM: agmatine; SPD: spermidine; PHM: phenylethylamine.
Figure 2

Total content of bioactive amines in fruits and vegetable samples. n = 3 for each sample. Bars followed by the same letter are not significantly different (Tukey’s Test, p ≤ 0.05).
Figure 3

(a) Dendrogram obtained by clustering individual amines and total amines contents/ (b) Loadings (PC1 and PC2); (c) Scatter plot (PC1 and PC2: 64.7%); Leg.: (3. A) 1: seriguela; 2: marolo; 3: fruta-do-conde; 4: acerola; 5: jabuticaba; 6: starfruit; 7: kale; 8: ora-pro-nobis; 9: almeirão-roxo; 10: serralha. (3. B): TYM: tyramine; PUT: putrescine; CAD: cadaverine; HIM: histamine; SRT: serotonin; AGM: agmatine; SPD: spermidine; PHM: phenylethylamine.

Supplementary Files

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- Tables.docx
- Graphicalabstractfinal.png