



Review

# Biogenic Amines in Alcohol-Free Beverages

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Received: 20 December 2019; Accepted: 27 February 2020; Published: 9 March 2020



**Abstract:** Biogenic amines are ubiquitous bioactive compounds that are synthesized by living organisms and perform essential functions for their metabolism. In the human diet, their excessive intake can cause food poisoning. In food, especially in alcohol-free beverages, biogenic amines can be synthesized by enzymes, naturally present in raw materials, or by microorganisms, which may be naturally present in the matrix or be added during beverage transformation processes. For this reason, in alcohol-free beverages, biogenic amine amount can be considered, above a certain level, as undesired microorganism activity. Therefore, it is important to evaluate the biogenic amine profile of non-alcoholic beverages in order to monitor food quality and safety. Moreover, biogenic amines can be taken into account by industries in order to monitor production processes and products. This review article provides an overview on the biogenic amine profile of alcohol-free beverages (plant milk, nervine drinks, soft drinks, and fruit juices). Furthermore, the clinical and toxicological effects, the biogenic amines legislation, and biogenic amine synthesis have been evaluated in non-alcoholic beverages.

**Keywords:** biogenic amines; beverages; non-alcoholic drinks

## 1. Introduction

Biogenic amines (BAs) are ubiquitous bioactive compounds, which can be produced through decarboxylation of amino acids or through amination or transamination of aldehydes and ketones [1]. Microorganisms', plants', and animals' metabolism can synthesize BAs and degrade these compounds as a result of their normal metabolic activities [2]. BAs, characterized by low molecular weight, can be ranked in different classes on the basis of their chemical structure (aliphatic, aromatic, and heterocyclic amines). In addition to this grading, BAs can be classified by amino group number: monoamines (e.g., tyramine, serotonin, and phenylethylamine), diamines (e.g., putrescine, tryptamine, and cadaverine), and polyamines (e.g., spermine and spermidine) [3]. These compounds are divided into natural/endogenous and exogenous. Moreover, natural polyamines can be synthesized from normal plants' and animals' cellular metabolism, whereas exogenous polyamines originate from decarboxylation mediated by microorganisms [4].

Some biogenic amines such as spermine, spermidine, putrescine, and cadaverine are essential for cellular metabolism. These compounds are important for protein synthesis, nucleic acid regulation, and membrane stabilization [5]. Histamine, along with other biogenic amines such as tyramine, cadaverine, and putrescine, are implicated into inflammatory response, caused by ingestion of contaminated food and beverages [6]. This reaction is called "histamine poisoning", because the cause of disease is mainly related to histamine ingestion [7]. BA synthesis occurs with great incidence in high-protein food. The amines profile, in beverages, depend on matrix considered and on microorganisms or enzymes contained [8]. Moreover, BAs are generated by endogenous amino acid decarboxylase activity in raw food materials or by decarboxylase-positive microorganism growth.

In beverages, BAs can be synthesized by endogenous enzymes or microorganisms present in raw material or added during transformation processes [9]. Beverages are defined as “any liquid with a thirst quenching or refreshment function” and they involve different class of commodities [10]. They can be divided into the classes of alcoholic and alcohol-free drinks. Alcohol-free beverages include different types of drinks, for example, fruit juice, dairy beverages, and energy drinks. These drinks may come from animals (e.g., cow’s milk, goat’s milk) or fruits/vegetables (e.g., fruit juices, soymilk, oranges) [11,12]. In alcohol-free beverages, which are non-fermented drinks, biogenic amine content can be considered, above a certain level, as a microorganism activity unwanted signal. Indeed, in unfermented beverages, BA amount can be considered as a food quality and safety index [13]. Although there are reviews providing insight on BA chemistry and analysis [14,15], there is no study devoted to BA occurrence in non-alcoholic beverages.

The aim of this paper was to summarize the state of the art research concerning biogenic amines in alcohol-free beverages, focusing the attention on BAs’ clinical and toxicological aspects, their regulation, and their content in alcohol-free drinks.

## 2. Clinical and Toxicological Aspects

BA ingestion, through food and beverages, can have harmful effects on consumers’ health. Both BAs in food and those biosynthesized by cell metabolism have physiological functions [16]. BAs can be dangerous for human health in cases where they are ingested in considerable quantities or when their natural catabolism is inhibited [17]. One of the major problems that can be linked to BA contamination in food is scombroid syndrome (Scombrototoxin Fish Poisoning, SFP), also called “Histamine Poisoning” [7,17,18]. This poisoning is often associated with the ingestion of fish contaminated from histamine [19,20]. However, the other BAs are considered as enhancers of histamine effects. However, toxicity is such that it is not linked to any single BA’s action but to the synergistic effect of several BAs [21]. In lower concentration, histamine is physiologically present inside the human body and its normal function consists of allergic response initiation. Histamine is generally bound to heparin, and it is released into the bloodstream by cells after immune system stimulation [7]. Some polyamines, such as putrescine, spermine, spermidine, and cadaverine, are essential elements of cells, as they are fundamental in nucleic acid regulation and protein synthesis. These BAs also have the specific function of stabilizing cell membranes [22,23]. Polyamines can also act as secondary messengers in cellular metabolism, an essential process for cell growth and regeneration [24–26]. Even if some amines are physiologically present in the human body, their ingestion at a high concentration can be toxic and produce poisoning symptoms, involving a wide range of organs [27]. Below physiological conditions, BAs can be metabolized by three different enzymes, present in gastrointestinal tract, which have the ability to oxidize BA amino groups: diamino oxidase (DAO), monoamine oxidase (MAO), and histamine-N-methyltransferase (HMT). These enzyme systems are able to degrade concentration of amines that are normally taken in with food and beverages [17,28,29]. It is almost impossible to calculate the acceptable daily dose for the human organism, as individual BA toxicity is enhanced by a combination of several BAs [30]. The symptoms, caused by an excessive BAs intake, can occur not only in people with greater sensitivity to histamine and other BAs, but also in all subjects with a normal catabolic activity of BAs. The symptoms caused by BA ingestion are similar to food poisoning. Among the major effects, caused by BAs, are nausea, vomiting, respiratory distress, hot flushes, headache, dizziness, skin redness, itching, oral inflammation, hypotension, vasodilation, or vasoconstriction (Table 1) [20,31,32]. The duration of symptoms can diverge from 8 to 12 h, with more or less serious consequences for consumers [19]. In many cases, these symptoms disappear quickly if the intoxication is associated with the consumption of contaminated foods, and for this reason it is difficult to differentiate them from other diseases [33]. The EFSA (European Food Safety Authority) reported that dose–response at which symptoms occur, related to histamine ingestion, is 25–50 mg, and that poisoning occurs after consuming 75–300 mg of histamine [34].

**Table 1.** Physiological effect of biogenic amines.

Biogenic Amine	Physiological Effect	Reference
Histamine	Release of adrenaline and noradrenaline Allergic processes Stimulation of the smooth muscles of the uterus, intestine, and respiratory tract Stimulation of sensory and motor neurons Control of gastric secretion	[35–39]
Tyramine	Peripheral vascularization Increase in cardiac output Increased lacrimation and salivation Increased breathing Increased blood sugar levels Noradrenaline release of the sympathetic nervous system Migraine	[16,37,40,41]
Putrescine and cadaverine	Hypotension Bradycardia Lockjaw Extremity paralysis Enhancement of the toxicity of other amines	[37,42,43]
$\beta$ -Phenethylamine	Noradrenaline release of the sympathetic nervous Increase in blood pressure Migraine	[37]
Tryptamine	Increase in blood pressure	[38]
Spermine and spermidine	Hypotension Bradycardia Enhancement of the toxicity of other amines	[38–48]

### 3. Biogenic Amine Regulation

Food concentration limits have been defined as limiting food poisoning linked to BA intake. On the basis of recommendations reported in the Codex Alimentarius, the European Commission has given specific attention to food products with higher risk of biogenic amine contamination. These regulations are aimed at limiting BA amount in food, so as to protect consumers' health. At the European and extra-European level, maximum acceptable concentration limits have been defined only for histamine in fish and fish products, whereas in other foods there are only proposed and/or recommended limits. A limit for the other BAs or other food products do not occur in any national legislation [49]. Table 2 shows the European and extra-European regulations relating to histamine legal limits in fish. As for drinks, there is no uniform legislation. However, only some European countries, in an arbitrary way, have established the legal limits for histamine (2 mg/L for Germany, 6 mg/L in Belgium, 8 mg/L in France, 4 mg/L in the Netherlands, and 10 mg/L for Switzerland and Austria) only in two beverages: wine and beer [50].

**Table 2.** Concentration limit of histamine in different countries.

Regulation	Products	Limit (mg/kg)	Reference
Reg. (EC) no. 1019/2013 (modify Reg. (EC) no. 2073/2005)	Fish species ( <i>Scombridae</i> , <i>Clupeidae</i> , <i>Engraulidae</i> , <i>Coryphenidae</i> , <i>Pomatomidae</i> , and <i>Scomberesocidae</i> )	100–200	[51]
	Fish products that have undergone enzyme maturation treatment	200–400	
USFDA	Fish products	500	[52]
FSANZ	Sample composed of different species of fish	100	[53]
Codex Alimentarius	Fish and fish product ( <i>Clupeidae</i> , <i>Scrombridae</i> , <i>Scromberesocidae</i> , <i>Pomatomidae</i> , and <i>Coryphaenidae</i> families)	100	[54]

However, BAs are ubiquitous compounds, and the joint presence of several BAs in beverages can have harmful effects on health; therefore, it is of considerable importance to study and evaluate the BA profile of alcohol-free beverages.

#### 4. Beverage Classification

Beverages are consumed mainly for their nutritional value (e.g., milk, fruit juices), thirst-quenching qualities (e.g., non-alcoholic drinks), exciting nature (e.g., coffee, tea), and recreational value (e.g., alcoholic beverages). Drinks are a heterogeneous class of commodities. There is no uniform regulation that defines how beverages should be classified. However, beverage classification can be based on many aspects, including formulation, carbonization, alcohol content, and physiological effects, among others [55].

Figure 1 shows a scheme displaying general beverage classification. Primarily, beverages can be divided into natural and synthetic beverages. Natural drinks are all drinks that are obtained through natural ingredient use or through a natural transformation process (such as milk, fruit juice, and wine), whereas synthetic drinks are obtained through artificial compound mixing (such as aroma, coloring, and sugar syrup) [56]. Another type of beverage classification is based on the presence or absence of carbon dioxide. Carbon dioxide can be added to sugary beverages (such as cola drinks and carbonated water), or it can be produced naturally during fermentation processes, such as in wine and beer [57,58].

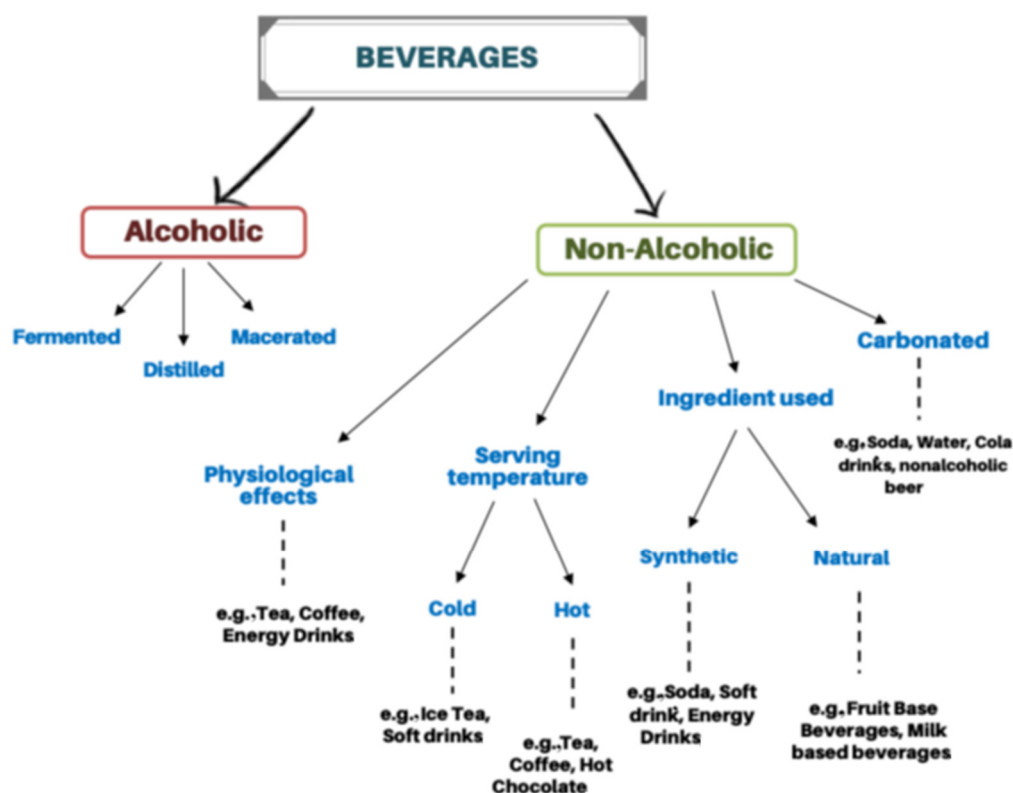


Figure 1. Beverage classification.

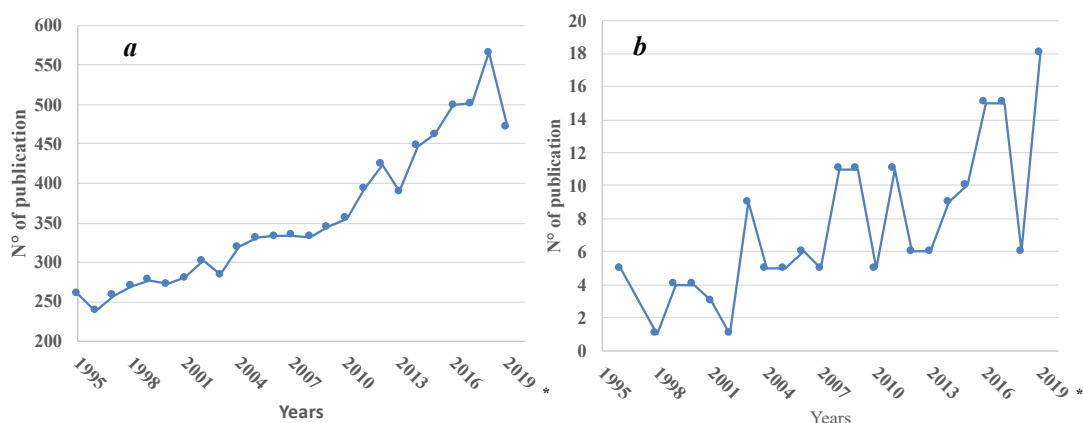
Another beverage classification is based on alcohol content. This classification is the most common form used to divide drinks. Alcoholic beverages are characterized by the presence of ethanol produced by fermentation processes. Generally, alcoholic beverages are furthermore divided into three classes: beer, wine, and spirits. Beer is the product obtained from fermentation of the soluble portion of barley (malt) or other cereal products such as rice and corn [59]. Wine is obtained from grape fermentation by yeasts (e.g., *Saccharomyces cerevisiae*) and lactic bacteria [60]. Lastly, spirits contain high amount of alcohol (>15%) [61]. Spirit drinks can be produced from different raw materials (e.g., herbs, fruits,

cereals) and they are subjected to distillation of fermented products or to raw material maceration in ethyl alcohol [61,62]. On the other hand, non-alcoholic beverages (such as fruit juices, milk, and soft drinks) do not contain ethanol. Another drink classification method is based on temperature in which they are served. For this reason, they are divided into cold and hot. Furthermore, beverages can be classified based on physiological effects (e.g., excitants) that can be expected from stimulant drinks (e.g., coffee and tea infusion), which are predicted by bioactive molecules (e.g., caffeine, theobromine, phenolic compounds, ethanol) that act on the nervous and circulatory systems. On the contrary, non-stimulating beverages have no physiological effects (e.g., fruit juices, soft drinks) [63,64].

Therefore, within this classification, alcohol-free beverages, depending on drink types considered, can be both natural or synthetic, carbonated or not, they can have physiological effects or not, and they can be served either hot or cold [56].

## 5. Biogenic Amines in Alcohol-Free and Non-Alcoholic Beverages

In beverages, BAs can be considered as an indicator of food quality, food safety, and product freshness, as BAs are related to their transformation processes (such as fermentation, maturing, and maturation) and also beverage conservation processes [13]. However, BA concentration and type in beverages can also be related to food matrix from which they originate [65]. The studies carried out on BA are about 16,300 (*Scopus* data). From 1995 to 2015, scientific article production on BAs remained almost similar, around 250 per year, whereas from 2016 onwards there has been a significant improvement of publication (Figure 2a). In the literature, there are few biogenic amine publications in alcohol-free beverages. Indeed, only 1% total articles treat BAs in beverages, approximately 181 publications in 25 years (Figure 2b).



**Figure 2.** Temporal evaluation of scientific publications on biogenic amines (BAs). Keywords: “Biogenic Amines” (a), “Biogenic Amines” + “Beverages” (b). (\* 2019 unfinished data).

### 5.1. Biogenic Amine Synthesis in Alcohol-Free and Non-Alcoholic Beverages

Although, BAs are ubiquitous compounds and they are found mainly in protein foods, they can also be present in many types of beverages [66]. In alcohol-free beverages, BA amount above certain levels is associated with undesirable microorganism activity. However, BA level in alcohol-free drinks is not necessarily linked with microorganism activity and/or food spoilage [13]. BAs are formed in beverages primarily by amino acid decarboxylation related to microbial metabolism or cellular enzymes. BAs are also formed by decarboxylating and proteolytic enzymes that are produced by microorganisms naturally present in native microbial flora of drinks, or microorganisms added as starter culture, or added through contamination [67].

In alcohol-free drinks, microorganisms can be added by contamination due to inadequate hygienic condition. For these reasons, BA content in drinks could be used as an indicator of food quality and

safety [1,13,68]. However, some BAs, especially endogenous polyamines, are naturally present in beverages, and they are synthesized during normal metabolic cell process [67].

BAs content depends on several aspects. First of all, BA concentration depends on type of alcohol-free beverage considered (i.e., animal or fruits/vegetables origin). Moreover, BA concentration and type mainly depend on microorganism presence with decarboxylase activity on decarboxylase enzyme types and on free amino acid amounts [69,70].

In alcohol-free beverages, derived from fruits/vegetables matrix, some BAs are representative of different plant species and can be used as a chemotaxonomic index [70]. Spermidine and spermine polyamines are ubiquitous in plants, along with polyamine precursor putrescine, and consequently they are mainly found in fruits/vegetables drinks. In plants, these polyamines play the role of organic nitrogen sources, and they are involved in embryogenesis, root growth, control of intracellular pH, flower and fruit development, secondary metabolite synthesis, and abiotic stress response (e.g., potassium deficiency, osmotic shock, pathogen infection) [71,72].

In drinks based on animal matrix, such as milk and chocolate milk, the polyamines (spermidine and spermine), which have important role in cell growth, and the others BAs are detected with great variability, depending upon animal species and thermal treatments [73].

In both types of alcohol-free beverages, either animal or fruits/vegetables origin, there are some factors that can influence BA synthesis that are associated with microorganism presence or enzymes with decarboxylase capacity [74]. BA synthesis is influenced by different factors, which depend upon raw material (such as pH, chemical composition, and free amino acid), microorganisms and enzymes with decarboxylase activity, and processing and storage condition (such as temperature, sugar, and additives). The issue that most affects biogenic amine development is microorganism activity, associated or not, with raw material, transformative drink process, and storage [75].

Although, all the conditions entertain BA synthesis directly or indirectly, namely, by acting on microorganisms or enzyme decarboxylase activity of beverage matrix. Firstly, biogenic amine amounts are influenced by free amino acid amount in drinks. The availability of free amino acids depends on proteolytic activity or beverage matrix concerned [76]. Instead, pH can regulate, directly or indirectly, decarboxylase enzymes by increasing or decreasing their activity. Enzyme activities increase when beverage pH is low. Furthermore, the upsurge in BA amount may be linked to the defense mechanism of microorganisms, which use biogenic amines to respond to acidity improvement [77]. Storage temperature is another critical factor, which act on microorganism and enzyme activities, affecting the biogenic amine synthesis [4].

Another important factor in BA synthesis are additives (e.g., sugar, soy lecithin, and sodium sulphide) contained in beverage matrixes. Additive addition can reduce microorganisms and enzyme activities; thus, BA synthesis is reduced indirectly [78].

In drinks, the additive of particular importance is sugar. Moreover, the absence of sugar influences the predisposition of the formation of biogenic amines during storage [79]. All these factors act in a combined manner, affecting the concentration of biogenic amines in food [80].

In Figure 3, a scheme is reported on biogenic amines synthesis and their precursor amino acids. BA synthesis occurs by removing the carboxyl group of amino acids by decarboxylase enzymes. The reaction mechanism also includes the presence of cofactors, such as vitamin B6, which acts as a link between enzyme active site and amino acids, leading to intermediate base formation, which are subsequently transformed into biogenic amines. Each BA is generated from an amino acid and a specific decarboxylase enzyme, with some exceptions (e.g., agmatine, spermine, and spermidine, which can all be synthesized, starting from arginine) [3,67,81].



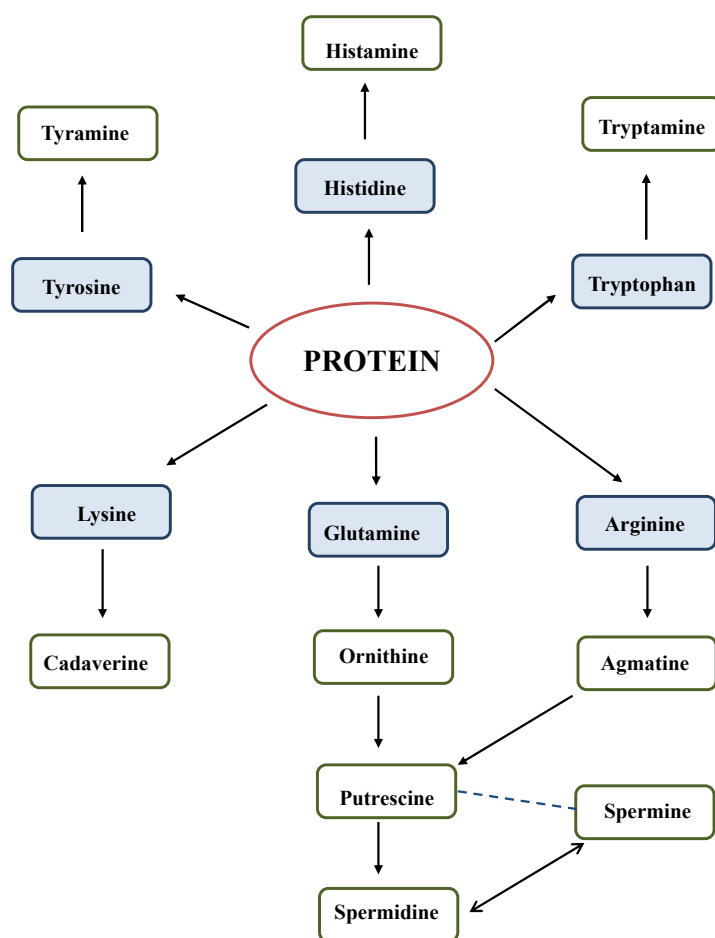


Figure 3. BA biosynthesis.

### 5.2. Biogenic Amine Amount in “Plant Milk”

Fruits/vegetables milks, which are also called “plant milk” or “non-dairy milk”, are beverages based on fruits/vegetables raw materials (e.g., barley, rice, wheat). The commodity definition of “milk” is specified by regulation (EC) no. 1308/2013, which defines it as “the normal mammary secretion obtained from one or more milking without either addition thereto or extraction therefrom” [82]. Although plant milks do not fall into this product category, indeed consumers have given it the name “milk” for its color likeness with animal milk [83]. Plant milks are produced through raw material milling and extraction, solid fraction separation, fruits/vegetables milk formulation and homogenization, ultra-high temperature (UHT) treatment, and packaging [84]. Fruits/vegetables milks can be classified according to the raw material used, such as cereal-based (e.g., oat milk, corn milk, rice milk), legume-based (e.g., soy milk, lupine milk), nut-based (e.g., coconut milk, walnut milk), seed-based (e.g., sesame milk, flax milk, hemp milk), and pseudo-cereal milk (e.g., quinoa milk, teff milk, amaranth milk) [85]. Table 3 reports literature data on BA contents in plant milks. In plant-based products, BAs may occur as a product of cellular metabolism and accumulated through enzymatic activity [83]. BA concentration and type detected in plant milks diverge according to milk classes taken into consideration. The analytical methods used for BA determination in plant milks are ultra-high-performance liquid chromatography (UHPLC)/fluorescence detection (FD) [4] and HPLC/FD [86]. The significant difference between the two methods concerns the analysis time; indeed, analysis time performed by UHPLC/FD is less than HPLC/FD [4,86]. The most detected BAs in plant milks are spermine, spermidine, histamine, cadaverine, and tyramine, whereas tryptamine has not been detected in any of the analyzed samples. The polyamines spermine and spermidine were found in higher quantities in soymilk, at concentrations of 8.37 mg/L and 2.04 mg/L, respectively [4].

Polyamines are present in all plant milks, although at different concentrations, and these can be linked to endogenous synthesis of polyamines or even to enzymatic activity of the matrix [86], although the highest concentration of histamine (8.37 mg/L), which is the only regulated amine, was observed in millet milk samples [87]. As for cadaverine, the highest concentration (5.36 mg/L) was found in barley milk, whereas tyramine concentration (not detected (nd)-0.66 mg/L), putrescine, and phenylethylamine appeared to be low in all samples analyzed in the literature [4,86,87].

**Table 3.** Biogenic amine amount for plant milk.

Beverages	No. *	Biogenic Amine Amount (mg/L)								Reference
		Phe	Put	Cad	His	Tyr	Try	Spd	Spm	
Soy milk-based product	1	ni *	ni	ni	ni	ni	ni	8.18	1.98	[4]
Soy milk UHT	6	ni	ni	ni	ni	ni	ni	7.77 ÷ 8.37	1.91 ÷ 2.04	
Spelt milk	3	nd*	nd	0.49 ÷ 1.17	5.26 ÷ 6.64	nd	ni	nd	nd	
Oat milk	3	nd	nd	0.64 ÷ 0.71	6.59 ÷ 8.06	nd	ni	0.46 ÷ 0.69	0.10 ÷ 0.17	
Millet milk	5	nd	nd	nd ÷ 0.23	6.14 ÷ 8.37	nd ÷ 0.25	ni	nd	nd	[87]
Barley milk	4	nd	nd	1.47 ÷ 5.36	nd ÷ 3.15	0.28 ÷ 0.62	ni	nd ÷ 0.31	nd ÷ 0.14	
Quinoa milk	4	nd	nd	0.52 ÷ 0.68	6.77 ÷ 7.69	0.10 ÷ 0.22	ni	nd	nd ÷ 0.14	
Rice milk	4	nd	nd	nd ÷ 0.25	3.10 ÷ 7.44	nd	ni	-	nd ÷ 0.17	

\* nd = not detected; ni = not investigated; No. = sample numbers.

### 5.3. Biogenic Amine Amount in Fruit Juice

Fruit juices are defined by the directive (EC) no. 2001/112, which can consist of 100% pure juice made from pulp of fresh fruit or from concentrate. Fruit juice does not contain flavorings, dyes, stabilizers, or any other added ingredients [88]. Different types of fruit juices can be distinguished: fruit concentrated juice and fruit nectars. In the former beverages, juice is obtained by reconstituting concentrated fruit juice with potable water. On the other hand, fruit nectars are obtained with the addition of water, with or without the addition of sugars, to the fruit juices, fruit purée, and/or concentrated fruit purée. The fruit content varies in a range from 25% to 99%, and minimum juice content varies according to the fruit in question [89]. The raw materials from which fruit juices originate are natural sources of BAs, especially polyamines [85]. In fact, polyamines are ubiquitous compounds in the plant kingdom, together with their putrescine precursor [79]. HPLC is the analytical method used for identification of BAs in fruit juice samples, supported by a UV or FD detector. The analysis time varies between 20 and 71 min, depending on the chromatographic method that is set, whereas the limit of detection (LOD) varies between 0.001 and 0.023 mg/L and the limit of quantification (LOQ) varies between 0.001 and 0.017 mg/L [79,90–93].

In all fruit juices analyzed in literature, the presence of putrescine (0.01–60.97 mg/L) was found; the highest amount was found in orange concentrated juice (60.97 mg/L) [90] and in orange juice (43.70 mg/L) (Table 4) [79]. Consequently, spermine (0.24–3.58 mg/L) and spermidine (nd-5.41 mg/L) were detected in all the samples analyzed in the literature [79,89–93]. Furthermore, the highest cadaverine concentration was detected in apricot juice (17.93 mg/L) and indeed in orange concentrated juice ethylamine (38.6 mg/L) [89]. The other BAs (phenylethylamine, tyramine, tryptamine, methylamine) have been identified only in some types of fruit juices: pear nectar, orange juice, apple juice, mango juice, pineapple, litchi juice, and grapefruit juice [89–93]. Histamine was only found in orange juice at a lower concentration (nd-0.26 mg/L) [91].



Table 4. Biogenic amine amount in fruit juice.

Beverages	No. *	Biogenic Amine Amount (mg/L)											Reference
		Phe	Put	Cad	His	Tyr	Try	Spd	Spm	Ser	Ety	Met	
Pear nectar	3	nd *	1.23 ÷ 3.10	5.88 ÷ 17.2	nd	nd	ni *	1.70 ÷ 2.17	1.47 ÷ 2.19	nd	nd	nd	
Apricot nectar	3	nd	1.10 ÷ 3.25	6.81 ÷ 11.25	nd	nd	ni	1.32 ÷ 2.95	2.21 ÷ 2.74	nd	nd ÷ 2.45	nd	[90]
Peach nectar	3	nd	1.88 ÷ 7.22	6.51 ÷ 13.03	nd	nd	ni	1.34 ÷ 1.96	1.58 ÷ 3.58	nd	nd	nd	
Orange juice	1	0.63	0.65	0.99	0.50	0.67	0.89	0.76	ni	ni	ni	ni	[91]
Orange juice	21	nd	22.6 ÷ 43.7	nd	0.03 ÷ 0.26	0.02 ÷ 0.67	nd	1.80 ÷ 4.20	0.08 ÷ 0.34	nd ÷ 0.48	ni	ni	[79]
Orange juice	1	0.01	0.01	ni	ni	nd	0.05	0.01	ni	ni	ni	0.06	
Apple juice	1	0.09	0.01	ni	ni	0.12	0.06	0.01	ni	ni	ni	0.78	
Grape juice	1	0.10	0.06	ni	ni	nd	0.35	nd	ni	ni	ni	nd	[92]
Mango juice	1	0.17	0.06	ni	ni	0.14	0.31	0.01	ni	ni	ni	nd	
Pineapple juice	1	0.03	0.12	ni	ni	0.01	0.01	nd	ni	ni	ni	0.84	
Litchi juice	1	0.01	0.01	ni	ni	0.02	nd	0.03	ni	ni	ni	0.07	
Apricot juice	7	nd	1.39 ÷ 7.10	3.96 ÷ 17.93	nd	nd	ni	1.97 ÷ 2.52	1.22 ÷ 2.51	nd	nd	nd	
Peach 50% juice	6	nd	1.41 ÷ 3.19	1.95 ÷ 10.06	nd	nd	ni	1.34 ÷ 2.01	1.22 ÷ 2.72	nd	nd	nd	
Peach 70% juice	6	nd	2.25 ÷ 3.59	4.11 ÷ 6.05	nd	nd	ni	2.02 ÷ 4.44	1.37 ÷ 1.89	nd	nd	nd	
Pear 50% juice	6	nd	1.11 ÷ 2.68	1.91 ÷ 8.31	nd	nd	ni	1.16 ÷ 1.77	1.17 ÷ 3.53	nd	nd	nd	
Pear 70% juice	6	nd	1.42 ÷ 4.44	3.75 ÷ 6.15	nd	nd	ni	1.90 ÷ 2.65	1.22 ÷ 1.41	nd	1.12 ÷ 1.23	nd	[89]
Apple concentrate juice	10	nd	0.59 ÷ 1.68	0.55 ÷ 4.27	nd	nd	ni	0.24 ÷ 0.67	0.24 ÷ 0.99	nd	nd ÷ 0.41	nd	
Pineapple concentrate juice	10	nd	1.53 ÷ 1.98	nd ÷ 3.14	nd	nd	ni	2.55 ÷ 5.41	1.53 ÷ 3.17	nd ÷ 4.61	0.22 ÷ 1.65	nd ÷ 1.88	
Grapefruit concentrate juice	11	nd	7.17 ÷ 20.8	0.38 ÷ 2.28	nd	nd	ni	1.03 ÷ 2.11	0.32 ÷ 0.50	nd ÷ 1.74	6.21 ÷ 12.98	nd ÷ 1.17	
Orange concentrate juice	12	nd	34.70 ÷ 60.97	nd	nd	nd	ni	2.04 ÷ 3.66	0.37 ÷ 1.37	nd ÷ 1.32	24.06 ÷ 38.64	nd ÷ 2.72	
Orange juice	5	nd	0.55 ÷ 2.21	ni	nd ÷ 0.04	nd ÷ 0.06	nd	0.08 ÷ 0.14	ni	ni	ni	ni	[91]

\* nd = not detected; ni = not investigated; No. = sample numbers.

#### 5.4. Biogenic Amine Content in Stimulating Beverages

Nervine drinks are all drinks that contain bioactive compounds that can lead to physiological effects on the central nervous system. Nervine drinks originate from nervine foods such as coffee, tea, and cocoa [94]. The brew process involves raw material extraction, by boiling water or steam, of water-soluble compounds contained in stimulating beverages. The main factors, which involve BA presence in nervine beverages, can be traced back to industrial treatments (fermentation, toasting, etc.) that the raw materials undergo before beverage preparation. In the literature there are few works that have investigated BAs in nervine drinks; instead, greater attention has been given to BA evaluation and determination in raw materials due to the fact that BAs can be taken as quality indicators of industrial processes [86]. Liquid chromatography is the method that has been applied for the identification and quantification of BA in stimulating beverages. The analysis time varied according to the method chosen between 20 and 80 min. In addition, the LOD and LOQ were 0.31–0.50 mg/L and 0.56–1.50 mg/L, respectively [95–100]. Table 5 shows BA concentrations in nervine beverages. Tea infusion is a drink made from the leaves of *Camellia sinensis* plant. Depending on the manufacturing process, different types of tea can be distinguished: black tea, green tea, white tea, oolong tea, yellow tea, and pu-er tea [96]. The literature studies investigated the BA profile of black and green tea infusions, reconstituted instant tea, and tea-based drinks [56]. Endogenous BAs were recovered in tea infusion, most notably, putrescine (nd-14.2 mg/L), spermidine (nd-10.4 mg/L), and spermine (nd-10.3 mg/L) [96,97]. Spizzirri et al. (2016) [97] found a higher content of BAs in samples analyzed compared to the study carried out by Bruckner et al. (2012) [95]. Furthermore, literature studies showed that black tea infusion, compared to green tea infusion, instant tea, and tea-based drinks, also contained histamine (nd-20.0 mg/L) and cadaverine (nd-14 mg/L), which are mostly involved in BA food poisoning [96,97]. Coffee drink is obtained from seeds of *Coffea* plants. Coffee brewing is performed by three different methods: decoctions, infusion, and pressure methods. There are only three studies in the literature that have evaluated the BA profile of coffee-based drinks [56]. In three types of coffee studied (Espresso coffee, Turkish coffee, and reconstituted instant coffee), the following BAs were found: phenylalanine, putrescine, cadaverine, histamine, tyramine, spermidine, spermine, and serotonin [98–100]. Instant coffee was found to have a higher content of serotonin (8.0–18.0 mg/L), compared to Espresso [98] and Turkish coffee [99]. Histamine, present in all the samples analyzed, was detected at low concentrations (nd-1.62 mg/L), which is lower than the European recommended concentration [98–100]. The BA profile shows how the coffee brew process influences the BAs amount set out in beverages [98]. The study by Restuccia et al. (2015) [97] showed that use of high temperatures and coffee beverage extraction pressures reduce BA amount in finished drinks, compared to Turkish coffee, obtained by coffee seed decoction [99,101], or reconstituted instant coffee [100].

**Table 5.** Biogenic amine amount in nervine beverages (mg/L).

Beverages	No. *	Biogenic Amine Amount										Reference
		Phe	Put	Cad	His	Tyr	Try	Agm	Spd	Spm	Ser	
Black tea	14	nd * ÷ 0.03	nd ÷ 0.02	nd	nd	nd ÷ 0.19	ni *	ni	nd ÷ 0.05	nd ÷ 0.34	ni	[96]
Green tea	6	nd	0.02 ÷ 0.08	nd	nd	nd ÷ 0.04	ni	ni	0.03 ÷ 0.09	0.06 ÷ 0.32	ni	
Instant tea	1	0.02	nd	nd	nd	0.03	ni	ni	0.05	ni	ni	
Black tea infusion	14	nd ÷ 2.00	8.4 ÷ 10.2	nd ÷ 14.0	nd ÷ 20.0	nd	ni	ni	6.5 ÷ 10.8	nd ÷ 0.3	nd ÷ 14.1	[97]
Green tea	7	nd	10.3 ÷ 14.6	nd	nd	nd	ni	ni	6.3 ÷ 10.4	nd	nd ÷ 11.5	
Tea drinks	3	nd	nd ÷ 6.9	nd	nd	nd	ni	ni	4.3 ÷ 6.7	nd	nd	
Espresso coffee	20	0.20 ÷ 1.21	0.60 ÷ 2.27	0.19 ÷ 1.84	0.22 ÷ 1.62	0.25 ÷ 1.89	ni	ni	0.45 ÷ 1.20	nd ÷ 1.95	nd ÷ 0.90	[98]
Turkish coffee	10	nd ÷ 4.99	0.5 ÷ 1.5	1.4 ÷ 9.0	nd	nd ÷ 19.7	ni	ni	nd	nd	3.70 ÷ 13.55	[99]
Instant coffee	16	0.4 ÷ 7.4	0.4 ÷ 5.3	0.4 ÷ 8.1	0.4 ÷ 1.4	0.4 ÷ 5.5	nd	0.4 ÷ 5.3	0.4 ÷ 7.7	0.5 ÷ 4.1	8.00 ÷ 18.0	[100]

\* nd = not detected; ni = not investigated; No. = sample numbers.

### 5.5. Biogenic Amine Amount in Soft Drinks

Among alcohol-free beverages there are soft drinks. These are a heterogeneous beverage class that includes carbonated beverages, milk-based drinks, non-alcoholic beers, and energy drinks [56]. In the literature there are few studies carried out to evaluate BA profile and amount in these drinks. The beverages analyzed in the literature are orange carbonated-based drink [79], non-alcoholic beer [102,103], dairy beverages [104], and milk chocolate [105]. The BA determination methods used in soft drinks is high performance liquid chromatography, supported by UV [79,102,104,105]; FD [102,103] detector. The analysis time for the reported literature methods varied between 35 and 71 min. In addition, the LOD and LOQ were 0.003–1.40 mg/L and 0.009–4.60 mg/L, respectively [79,102–105]. Vieira et al. (2007) evaluated BA amount in 35 samples of orange carbonated-based drinks. In these samples, putrescine (0.69–5.14 mg/L), histamine (0.01–0.03 mg/L), spermidine (0.01–0.32 mg/L), and spermine (0.01–0.04 mg/L) were found [79]. The BA concentration in orange carbonated-based drinks follows the amino profile of orange juices from which they are produced [106]. Non-alcoholic beer is another soft drink in which BAs have been studied. Non-alcoholic beer is a drink that contains less than 0.5% alcohol by volume [107]. The amines most present were putrescine (0.31–1.43 mg/L), cadaverine (0.11–0.56 mg/L), tryptamine (0.28–2.56 mg/L), spermine (0.13–0.72 mg/L), and spermidine (0.16–0.81 mg/L). Phenylethylamine, histamine, and tyramine were only found in some of the analyzed non-alcoholic beer samples (Table 6) [102,103]. Another type of soft drink considered is dairy beverages. These drinks are made from milk, both fruits/vegetables and animal, to which flavors and other ingredients are added [108]. In the literature, the BA profile of only one sample of dairy beverages [104] and eight samples of chocolate milk [105] was studied. The analyses showed that the BAs detected in the dairy beverages sample were putrescine (3.2 mg/L), histamine (1.9 mg/L), and agmatine (3.1 mg/L) [104]. In chocolate milk samples, the BAs detected were putrescine (nd-0.4 mg/L), tyramine (nd-0.03 mg/L), tryptamine (nd-0.1 mg/L), spermine (nd-2.0 mg/L), and spermidine (nd-2.0 mg/L) [105]. To date, there are limited data in the literature to fully and thoroughly assess the BA profile of soft drinks.

**Table 6.** Biogenic amine amount in soft drinks.

Beverages	No. *	Biogenic Amines amount (mg/L)										Reference
		Phe	Put	Cad	His	Tyr	Try	Agm	Spd	Spm	Ser	
Orange carbonated-based	35	nd *	0.69 ÷ 5.14	nd	0.01 ÷ 0.03	nd	nd	nd	0.01 ÷ 0.32	0.01 ÷ 0.04	nd	[79]
Non-alcoholic beer	10	nd ÷ 0.54	0.31 ÷ 1.43	0.11 ÷ 0.42	nd ÷ 0.62	nd ÷ 0.48	0.29 ÷ 2.56	nd	0.16 ÷ 0.81	0.13 ÷ 0.68	ni *	[102]
Non-alcoholic beer	5	nd ÷ 0.48	0.56 ÷ 1.30	0.28 ÷ 0.56	nd ÷ 0.37	nd ÷ 0.27	0.28 ÷ 1.30	nd	0.35 ÷ 0.73	0.24 ÷ 0.72	ni	[103]
Dairy beverages	1	nd	3.2	nd	1.90	nd	nd	3.10	nd	nd	ni	[104]
Milk chocolate	8	ni	nd ÷ 0.40	ni	nd	nd ÷ 0.03	nd ÷ 0.10	ni	nd ÷ 1.00	nd ÷ 2.00	ni	[105]

\* nd = not detected; ni = not investigated; No. = sample number.

## 6. Conclusions

To our best knowledge, this is the first review that deals with BAs in alcohol-free beverages. In recent years, alcohol-free beverage consumption has been growing strongly [109,110]. In soft drinks, it is essential to monitor BA amount as an important aspect related to BA control in the monitoring of the quality of drinks. Another aspect to pay attention to is histamine, cadaverine, and tyramine content in alcohol-free beverages, in order to promote consumer health defense, as BAs may be involved in food poisoning [30]. For these reasons, BAs can be taken into consideration as product and process indicators, which allow beverage quality and safety to be controlled along the entire production chain. To date, in the literature there are few studies regarding BAs in alcohol-free beverages. BA concentration and type content in alcohol-free beverages differ significantly on the basis of drink type taken into consideration, on transformation and production process, and on raw material from which they are originated. The most common BAs that have been detected at different amounts in all alcohol-free beverages, are the polyamines spermine and spermidine, and their precursor putrescine. This is related to the fact that these amines are specific for cellular metabolism, both animal and fruits/vegetables. Moreover, in all alcohol-free beverages, histamine (nd-20.0 mg/L)—the only BA that is currently regulated—has lower concentrations than the limit established by European law (100 mg/L) [51]. Following previous considerations, it should be useful to actively promote BA study profile in alcohol-free drinks, not only in order to protect human health but also to allow beverage industries to monitor food quality and food safety along the entire production chain.

**Funding:** This research received no external funding.

**Conflicts of Interest:** The authors declare no conflict of interest.

## Abbreviations

Him	Histamine
Tyr	Tyramine
Put	Putrescine
Cad	Cadaverine
Met	Methylamine
Agm	Agmatine
Phe	Phenylethylamine
Spm	Spermine
Spd	Spermidine
Try	Tryptamine
Ety	Ethylamine
Ser	Serotonin
nd	not detected
ni	not investigated
Bas	Biogenic amines
LOD	Limit of detection
LOQ	Limit of quantification

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