Nutritional Composition and Chemical Safety of Wagashi Gassirè Cheese Sold in the Southern Benin Markets


Abstract: In this study, the nutritional composition and the chemical safety of Wagashi Gassirè (WG) cheese sold in southern Benin markets were assessed. For this purpose, 15 WG were analysed for fatty acids, essential minerals, and chemical hazards (dioxins, aflatoxin M1 (AFM1), biogenic amines, metals, antibiotic and pesticide residues). The risks related to arsenic, lead, aluminium, AFM1, histamine, and tyramine were calculated using the methods recommended by the European Food Safety Authority. Oleic, palmitic and stearic acids, calcium, and phosphorus were the main fatty acids and minerals detected. Lead (0.08 ± 0.06 mg/kg) and AFM1 (0.3 ± 0.0 µg/kg) were detected in all samples and exceeded the maximum level set by the international standard. Cadaverine and tyramine were the main biogenic amines found. No pesticide residues were detected using a multi-residue method targeting compounds. Residues of quinolones, tetracyclines, and colistin antibiotics were also detected. The calculated chronic exposure indicated no public health concern for the chemical contaminants targeted. Moreover, the average cancer risk related to AFM1 intake was $3 \times 10^{-4}$ cases/$10^5$ persons/year for the Benin population through WG consumption. This study contributes to the nutritional characterization of WG and identifies lead and AFM1 as the most relevant chemical hazards of this product.

Keywords: West Africa; dairy products; Wagashi Gassirè cheese; nutritional properties; chemical contaminants; human risk assessment
1. Introduction

World cheese production is projected to grow at 1.2% over the next decade (2020–2029) [1]. The United States is the world’s largest producer of cheese, followed by Germany, France, and Italy. Europe is the largest producing region followed by America and Asia [2]. In Africa, Egypt, South Africa, South Sudan, Tanzania, Zambia, and Ethiopia are the countries with the largest volume of cheese production, while Niger and Nigeria are the largest producers in West Africa [3]. The most widespread and consumed local cheese in Nigeria is Wagashi Gassirè (WG), which is produced by the Peuhl sociocultural group [4]. The traditional technology of WG was developed in Benin before spreading to Nigeria, Togo, Burkina Faso, and Niger [5].

WG is a soft, non-ripened cheese whose processing is based on the hot coagulation (65 to 70 °C) of fresh, whole cow’s milk induced by Calotropis procera (commonly known as the apple of Sodom) extract that contains an enzyme known as calotropain [4–6]. To obtain the extract, crushed leaves and stems are triturated in a separate small quantity of milk or water. The resulting suspension is filtered, and the filtrate is added to the heated milk for coagulation. The milk becomes solid after 25–30 min. The resulting coagulum is cooked at 90 °C for 30 min, moulded, and drained to obtain a white, soft, non-salted, and non-ripened cheese. The obtained white WG is sometimes stained to obtain red WG using various dyes, such as cobs and panicles from sorghum and teak (Tectona grandis) leaves [7].

WG is produced throughout Benin, and the departments of Borgou-Alibori, Zou-Collines, Mono-Couffo, and Atocora-Donga are the main production areas [8,9]. However, WG markets are located in urban areas (Abomey-Calavi, Cotonou, Parakou, Bohicon) where there is high demand [9].

WG is an important source of protein (23%), fat (18%), and minerals (1.3–1.8%) [10,11]. Its consumption could help the population to meet the adult recommended daily intakes of, for example, 0.83 g protein/kg body weight (bw), 1000 mg calcium, and 11 mg iron [12]. Currently, the available data are related to the pH and proximal composition (dry matter, titratable acidity, fat, ash, and total sugars) of WG produced in Benin [8,13]. To date, and to our knowledge, no data are yet available on the nutritional values such as the essential mineral and fatty acid contents of WG produced in Benin.

The production and marketing conditions of WG in Benin are potential sources of chemical contamination [7]. The most relevant dairy product contaminants include natural toxins, polychlorinated biphenyls (PCBs), dioxins, and metals such as aluminium, arsenic, cadmium, lead, or mercury. Moreover, microbiological contamination of dairy products could result in the formation of biogenic amines, produced from free amino acids subjected to decarboxylase enzymes coming from spoilage bacteria. Food intoxication due to biogenic amines is mostly related to the presence of histamine, tyramine, and cadaverine [14]. High histamine intake is associated with vomiting and diarrhea, headache, hypotension, heart palpitations, asthma attacks, urticaria, and other rashes typical of allergies [15]. Typical signs of tyramine intoxication are migraine, headaches, and increased blood pressure [16]. Among metals, aluminium is a neurotoxic agent that could increase the probability of developing Alzheimer’s disease [17]. Lead is well known to have detrimental effects on human health, including effects on the nervous, renal, cardiovascular, reproductive, immune, and hematologic systems. In children, lead is associated with poor child neurodevelopment [18]. Lead exposure from piped water and the consumption of animals killed by ammunition has been shown to present a public health concern in Benin, as the blood lead levels of pregnant women and children were shown to be above the 50 µg/L threshold set by the US Centers for Disease Control and Prevention [19,20]. Currently, few data are available on the chemical contaminants in WG produced in Benin. Some data are available regarding its aflatoxin M1 (AFM1) content [13,21].

The main objective of this work is to determine the nutritional properties and chemical hazard contents of WG sold in the southern Benin markets and to assess potential chemical risks associated with its consumption. As WG is the most widespread and consumed local
cheese in Benin, the data from this study will be useful for consumption recommendations and prevention programs for reducing and eliminating chemical hazards in WG.

2. Materials and Methods

2.1. Sampling

Fifteen samples of WG were purchased in Abomey-Calavi. The samples included the two forms of WG (white and red). Regarding the variety of WG processing methods, the selection retained WG (white and red) that was produced according to the method described by Dossou et al. [5], and this determination was based on the declaration made by the sellers. The WG samples were collected three days after production in three street markets: three red WG (RWG) in Godomey, six RWG in Dekougbe, and six white WG (WWG) in Calavi-Tokpa. All the collected samples of WG were stored at −80 °C before they were subjected to analysis to determine their pH, proximal and nutritional composition, and chemical hazard content. Description of sample preparation, conditions of instrumental analyses, and quality assurance of the analytical results are presented in the Supplementary Materials.

2.2. pH, Proximal and Nutritional Components Analysis

The pH was determined using a Knick pH meter 765 Calimatic (electrode Mettler-Toledo 406-M6-DXX-S7/25, Columbus, OH, USA). The moisture was determined by the difference in mass of the samples before and after freeze-drying for 48 h (Lyoquest, Telstar, Barcelona, Spain). The crude protein content was determined by the Kjeldahl method according to ISO 8968-1:2014 [22]. The fat was extracted by the Folch method [23]. After saponification and methylation, twenty-seven fatty acid methyl esters were analysed by gas chromatography (GC) (Thermo Fisher Scientific, Waltham, MA, USA) using a CPSil88 fatty acid column (100 m × 0.25 mm; 0.2 µm) (Varian, Agilent Technologies, Santa Clara, CA, USA) coupled to a PolarisQ ion trap mass spectrometer (MS) (Thermo Fisher Scientific, USA) [24]. Thirty-one minerals were determined by inductively coupled plasma mass spectrometry (ICP-MS) [25] and classified into essential minerals (eleven) and metallic trace elements (twenty) [12].

2.3. Chemical Contaminants Analysis

Dioxins and dioxin-like PCB compounds (hereafter referred to by the general term “dioxins”) were determined by DR-CALUX (Dioxin Responsive-Chemically Activated Luciferase Gene Expression) cell-based assay [26]. AFM1 was determined by the ELISA (Enzyme-Linked Immunosorbent Assay) method using the RIDASCREEN kit of R-Biopharm (Darmstadt, Germany). UPLC-FLD (Ultra-Performance Liquid Chromatography combined with Fluorescence Detection) was used to quantify ten biogenic amines in WG samples [27]. The antibiotic residues were screened by BeadyPlex using kit187 from Unisensor (Seraing, Belgium). The BeadyPlex is a flow-cytometry-based method able to detect 80 residues belonging to 10 antibiotic families. Two multi-residue chromatographic methods were used to analyse 103 pesticide residues in the WG samples: gas chromatography coupled to an electron capture detector (GC/ECD, limit of quantification (LOQ): 0.01 mg/kg wet weight) for chlorpyrifos, endosulfan, and the remaining organochlorines (19 residues), and liquid chromatography coupled to a tandem mass spectrometry detection (LC-MS/MS, LOQ: 0.001 mg/kg wet weight) for other components (84 residues) [28].

2.4. Exposure and Risk Assessment

To calculate the exposure, the WG consumption and body weight data used were those described by Ingenbleek et al. [29]. This study determined the average daily consumption and the 95th percentile (i.e., “high-level consumption”) of the Beninese population, as well as the average daily consumption of fresh/fermented milk, concentrated/dehydrated milk, and other dairy products in the departments of Littoral in the south and Borgou in the north of Benin. As WG is the main local dairy product consumed in Benin, the WG
consumption data used in this study correspond to the “other dairy products” consumption. The mean and P95 consumption for the whole of Benin were 2.4 and 47.4 g per adult of 60 kg bw per day, while the mean consumption for Littoral and Borgou were 1.8 and 8.2 g per adult of 60 kg bw per day [29]. A deterministic approach was used to calculate the dietary intake of chemical contaminants. In this approach, the exposure was obtained by multiplying the WG consumption by the WG contamination expressed as the lower bound of the mean chemical contaminant content of the 15 WG (the lower bound of the mean for the 15 samples was calculated by replacing 0 with the results “<LOQ”).

The exposure and the risk were assessed for some biogenic amines (histamine and tyramine), some metals (aluminium, lead, and arsenic), and for AFM1. For carcinogenic and genotoxic substances such as lead, arsenic, and AFM1, the Margin of Exposure (MOE) approach was used, as recommended by the European Food Safety Authority (EFSA) [30,31]. The MOE is the ratio between a Benchmark Dose (BMD10 or BMDL10) and the level of exposure [30,31]. When using a BMDL based on human studies (which is the case for lead and arsenic), an MOE of 1 or above is considered to be of low public health concern [30,31]. For lead, the BMDL10 is 0.63 µg/kg.bw/day for nephrotoxicity in adults [32]. BMDL10 ranging from 0.3 to 8 µg/kg.bw/day should be used regarding the adverse effects (lung cancer, skin cancer, bladder cancer, skin lesions) of arsenic [33]. When using a BMDL based on animal studies (which is the case for AFM1), an MOE of 10,000 or above is considered to be of low public health concern [30,31]. AFM1 induces liver cancer with a potency one-tenth that of aflatoxin B1 (for AFB1, BMDL10 was 0.4 µg/kg.bw/day); therefore, a potency factor of 0.1 for the AFM1 risk assessment was used in this study [34]. In addition to the quantitative MOE approach used for AFM1, the qualitative approach to liver cancer risk estimation proposed by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) was also used [35]. In this approach, the population cancer risk (cancers per year per 100,000 population) was obtained using the following equation [35]:

\[
\text{Cancer risk} = [\text{PHBV}^+ \times (\text{AF exposure}) \times \text{HBV}^+] + [\text{PHBV}^- \times (\text{AF exposure}) \times (1 - \text{HBV}^+)]
\]

where PHBV+ is the potency estimate P for the HBV+ fraction of the population (mean of 0.269 cancers per 100,000 person-years per ng aflatoxin/kg bw per day for HBsAg-positive), PHBV− is the potency estimate P for the HBV− fraction of the population (mean of 0.017 cancers per 100,000 person-years per ng aflatoxin/kg bw per day for HBsAg-negative), HBV+ is the population fraction of chronic HBV cases (6% in Benin adult population) [36], and AF exposure is the AFM1 exposure.

For substances without genotoxic properties (histamine, tyramine, and aluminium) the exposure was compared either to a TWI (tolerable weekly intake) for aluminium or to a NOAEL (No Observed Adverse Effect Level) for histamine and tyramine. The TWI was 1000 µg/kg.bw/week for neurotoxicity and embryotoxicity of aluminium [17] while the NOAELs were 50 mg for histamine and 600 mg for tyramine per meal for a healthy person [14].

2.5. Statistical Analysis

MS Excel 2013 was used for recording the data, and the descriptive analysis was performed using the SAS System 9.4 software. For nutritional parameters, Student’s t-test was performed to compare the means of RWG and WWG. Means were calculated considering the samples for which the analysed nutritional parameter was detected. For chemical hazards, the lower bound means were calculated and used for Student’s t-test. The statistical significance was set at 5%.

3. Results

3.1. pH, Proximal and Nutritional Composition of Commercial Wagashi Gassirè

Figures 1 and 2 present the pH and the proximal composition of WG. The average pH was 5.4 ± 0.3, and the moisture, protein, and fat contents (per wet weight) were 59.1 ± 2.9%,
13.5 ± 1.2%, and 21.0 ± 2.3%, respectively. The pH of RWG (5.6 ± 0.2) was significantly higher (p < 0.01) than that of WWG (5.1 ± 0.1) (Figure 1), while the proximal composition was similar in RWG and WWG (Figure 2). The data regarding the pH, moisture, fat, and protein contents of the 15 WG samples are presented in Table S1.

Figure 1. pH of Wagashi Gassirè sold in southern Benin markets. RWG: Red Wagashi Gassirè, WWG: White Wagashi Gassirè, WG: Wagashi Gassirè, n: number of samples, **: p < 0.01.

Figure 2. Moisture, protein, and fat content of Wagashi Gassirè sold in southern Benin markets. RWG: Red Wagashi Gassirè, WWG: White Wagashi Gassirè, WG: Wagashi Gassirè, w.w: wet weight, n: number of samples, ns (non-significant): p > 0.05.

The fatty acid content of the 15 WG is shown in Table S2. From the twenty-seven fatty acids subjected to the analysis, seven were found in all samples (lauric, myristic, palmitic, stearic, palmitoleic, oleic, and linoleic acids), capric acid was detected in 13 samples, and tetracosanoic and cis-10-heptadecenoic acids were detected in 4 and 3 samples, respectively (Table 1). Saturated (SFA), mono (MUFA), and polyunsaturated (PUFA) fatty acids represented 62.3 ± 3.3, 35.1 ± 3.4, and 2.5 ± 0.3% of the total fatty acids, respectively. Palmitic, stearic, and oleic acids were the most important fatty acids, with proportions of 31.7 ± 2.0, 17.9 ± 1.3, and 31.9 ± 3.4% of total fatty acids, respectively. Among the two essential fatty acids, i.e., linoleic acid (LA) and alpha linolenic acid (ALA), only LA was detected with a proportion of 2.5 ± 0.3%. No significant difference was observed between fatty acid proportions in RWG and WWG.
Table 1. Fatty acid composition of Wagashi Gassirè sold in the southern Benin markets (g/100 g total fatty acids).

<table>
<thead>
<tr>
<th>Fatty Acids</th>
<th>WG (n = 15)</th>
<th>RWG (n = 9)</th>
<th>WWG (n = 6)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± SD</td>
<td>Mean ± SD</td>
<td>N1 Mean ± SD</td>
</tr>
<tr>
<td>Capric acid C10:0</td>
<td>2.3 ± 0.5</td>
<td>2.3 ± 0.4</td>
<td>8 2.4 ± 0.7</td>
</tr>
<tr>
<td>Lauric acid C12:0</td>
<td>1.9 ± 0.3</td>
<td>1.8 ± 0.2</td>
<td>9 2.0 ± 0.4</td>
</tr>
<tr>
<td>Myristic acid C14:0</td>
<td>8.4 ± 0.7</td>
<td>8.2 ± 0.7</td>
<td>9 8.7 ± 0.6</td>
</tr>
<tr>
<td>Palmitic acid C16:0</td>
<td>31.7 ± 2.0</td>
<td>31.4 ± 2.2</td>
<td>9 32.1 ± 1.7</td>
</tr>
<tr>
<td>Stearic acid C18:0</td>
<td>17.9 ± 1.3</td>
<td>17.6 ± 1.2</td>
<td>9 18.4 ± 1.4</td>
</tr>
<tr>
<td>Lignoceric acid C24:0</td>
<td>0.9 ± 0.1</td>
<td>0.9 ± 0.2</td>
<td>9 1.0*</td>
</tr>
<tr>
<td>Palmitoleic acid C16:1</td>
<td>2.1 ± 0.9</td>
<td>2.4 ± 1.0</td>
<td>9 1.6 ± 0.5</td>
</tr>
<tr>
<td>cis-10-heptadecenoic acid</td>
<td>5.4 ± 7.8</td>
<td>5.4 ± 7.8</td>
<td>3 &lt;LOQ</td>
</tr>
<tr>
<td>Oleic acid C18:1,9c</td>
<td>31.9 ± 1.3</td>
<td>31.7 ± 3.8</td>
<td>9 32.1 ± 1.4</td>
</tr>
<tr>
<td>Linoleic acid C18:2,9c,12c</td>
<td>2.5 ± 0.3</td>
<td>2.5 ± 0.3</td>
<td>9 2.4 ± 0.1</td>
</tr>
</tbody>
</table>


Table 2. Essential mineral composition of Wagashi Gassirè sold in the southern Benin markets (mg/kg wet weight).

<table>
<thead>
<tr>
<th>Essential Minerals</th>
<th>WG (n = 15)</th>
<th>RWG (n = 9)</th>
<th>WWG (n = 6)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± SD</td>
<td>Mean ± SD</td>
<td>N1 Mean ± SD</td>
</tr>
<tr>
<td>Sodium Na</td>
<td>53 ± 46</td>
<td>35 ± 11</td>
<td>9 81 ± 64</td>
</tr>
<tr>
<td>Magnesium Mg</td>
<td>81 ± 58</td>
<td>48 ± 16</td>
<td>9 131 ± 65</td>
</tr>
<tr>
<td>Phosphorus P</td>
<td>1913 ± 577</td>
<td>1601 ± 389</td>
<td>9 2381 ± 503</td>
</tr>
<tr>
<td>Potassium K</td>
<td>62 ± 81</td>
<td>17 ± 9</td>
<td>9 129 ± 98</td>
</tr>
<tr>
<td>Calcium Ca</td>
<td>2152 ± 1122</td>
<td>1526 ± 680</td>
<td>9 3092 ± 1010</td>
</tr>
<tr>
<td>Iron Fe</td>
<td>4.9 ± 2.6</td>
<td>5.4 ± 2.2</td>
<td>9 4.3 ± 3.4</td>
</tr>
<tr>
<td>Manganese Mn</td>
<td>0.5 ± 0.3</td>
<td>0.7 ± 0.3</td>
<td>9 0.3 ± 0.1</td>
</tr>
<tr>
<td>Copper Cu</td>
<td>0.7 ± 0.2</td>
<td>0.7 ± 0.2</td>
<td>9 0.7 ± 0.2</td>
</tr>
<tr>
<td>Zinc Zn</td>
<td>20 ± 3</td>
<td>21 ± 4</td>
<td>9 19 ± 3</td>
</tr>
<tr>
<td>Selenium Se</td>
<td>0.1*</td>
<td>0.1*</td>
<td>1 &lt;LOQ</td>
</tr>
<tr>
<td>Molybdenum Mo</td>
<td>0.020 ± 0.008</td>
<td>0.021 ± 0.008</td>
<td>9 0.022 ± 0.009</td>
</tr>
</tbody>
</table>

RWG: Red Wagashi Gassirè, WWG: White Wagashi Gassirè, WG: Wagashi Gassirè, different superscript letters in the same row indicate a significant difference (p < 0.05) between RWG and WWG, LOQ (see Table S3), N1 and N2: number of samples in which the mineral was detected and quantified in RWG and WWG respectively, n: number of analysed samples, SD: standard deviation, * individual value.

3.2. Chemical Contaminants Found in Commercial Wagashi Gassirè

Among the twenty metallic trace elements analysed (Table S4), fourteen were detected, including lithium, aluminium, chromium, nickel, arsenic, rubidium, strontium, tin, barium, thallium, lead, bismuth, uranium, and vanadium (Table 3), in generally very low concentrations (below 1 mg/kg, except for aluminium, strontium, and barium). Significant differences were found for some metallic trace elements. The lead and rubidium contents were significantly higher in WWG (0.12 ± 0.08 and 0.58 ± 0.52 mg/kg, respectively) than in RWG (0.06 ± 0.03 and 0.05 ± 0.01 mg/kg, respectively). The vanadium content was...
significantly lower in WWG (0.004 ± 0.005 mg/kg) than in RWG (0.041 ± 0.021 mg/kg) (Table 3). Aluminium was detected in 8/9 RWG samples and in only one WWG sample. Aluminium concentrations varied a lot between RWG samples, ranging from not-detected to 573 mg/kg (Table S4).

**Table 3.** Metallic trace element composition of Wagashi Gassirè sold in the southern Benin markets (mg/kg wet weight).

<table>
<thead>
<tr>
<th>Metallic Trace Elements</th>
<th>WG (n = 15)</th>
<th>RWG (n = 9)</th>
<th>WWG (n = 6)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>LB Mean ± SD</td>
<td>LB Mean ± SD</td>
<td>N1</td>
</tr>
<tr>
<td>Lithium Li</td>
<td>0.009 *</td>
<td>&lt;LOQ</td>
<td>0</td>
</tr>
<tr>
<td>Aluminium Al</td>
<td>74.6 ± 150.4</td>
<td>123.4 ± 181.4</td>
<td>8</td>
</tr>
<tr>
<td>Chromium Cr</td>
<td>0.01 ± 0.02</td>
<td>0.02 ± 0.02</td>
<td>6</td>
</tr>
<tr>
<td>Nickel Ni</td>
<td>0.004 ± 0.013</td>
<td>0.02 *</td>
<td>1</td>
</tr>
<tr>
<td>Arsenic As</td>
<td>0.022 ± 0.008</td>
<td>0.020 ± 0.009</td>
<td>8</td>
</tr>
<tr>
<td>Rubidium Rb</td>
<td>0.26 ± 0.41</td>
<td>0.05 ± 0.01 a</td>
<td>9</td>
</tr>
<tr>
<td>Strontium Sr</td>
<td>2.5 ± 1.2</td>
<td>2.5 ± 1.6</td>
<td>9</td>
</tr>
<tr>
<td>Tin Sn</td>
<td>0.007 ± 0.018</td>
<td>0.055 *</td>
<td>1</td>
</tr>
<tr>
<td>Barium Ba</td>
<td>2.3 ± 1.3</td>
<td>2.4 ± 1.7</td>
<td>9</td>
</tr>
<tr>
<td>Thallium TI</td>
<td>0.00019 ± 0.00035</td>
<td>0.0027 ± 0.00041</td>
<td>4</td>
</tr>
<tr>
<td>Lead Pb</td>
<td>0.08 ± 0.06</td>
<td>0.06 ± 0.03 a</td>
<td>9</td>
</tr>
<tr>
<td>Bismuth Bi</td>
<td>0.005 ± 0.005</td>
<td>0.007 ± 0.004</td>
<td>9</td>
</tr>
<tr>
<td>Uranium U</td>
<td>0.26 ± 0.024</td>
<td>0.041 ± 0.021 a</td>
<td>9</td>
</tr>
</tbody>
</table>

Mean levels of dioxins (pg/g fat), AFM1 (µg/kg wet weight), and biogenic amines found in WG are presented in Table 4, while individual data for the 15 samples of WG are shown in Tables S5–S7. Dioxins were not detected in six WG samples, including four RWG and two WWG, and ranged, in contaminated samples, from 1.2 to 4.3 pg/g fat (Table S5) with a lower bound mean of 1.5 ± 1.5 pg/g fat (Table 4). AFM1 was detected in all WG samples, with a minimum and maximum content of 0.1 µg/kg and 0.4 µg/kg (Table S6), respectively, and a lower bound mean of 0.3 ± 0.0 µg/kg (Table 4). AFM1 concentration was significantly higher in WWG than in RWG samples.

**Table 4.** Dioxins and PCBs, aflatoxin M1, and biogenic amine contents of Wagashi Gassirè sold in the southern Benin markets.

<table>
<thead>
<tr>
<th>Chemical Contaminants</th>
<th>WG (n = 15)</th>
<th>RWG (n = 9)</th>
<th>WWG (n = 6)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>LB Mean ± SD</td>
<td>LB Mean ± SD</td>
<td>N1</td>
</tr>
<tr>
<td>Dioxins and PCBs (pg BEQ/g fat)</td>
<td>1.5 ± 1.5</td>
<td>1.0 ± 0.8</td>
<td>5</td>
</tr>
<tr>
<td>Afatoxin M1 (µg/kg wet weight)</td>
<td>0.3 ± 0.0</td>
<td>0.1 ± 0.0 a</td>
<td>9</td>
</tr>
<tr>
<td>Biogenic amines (mg/kg wet weight)</td>
<td>2-phenylethylamine 0.4 ± 0.8</td>
<td>0.07 ± 0.004</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>Putrescine 10.0 ± 13.0</td>
<td>15.5 ± 14.2 a</td>
<td>6</td>
</tr>
<tr>
<td></td>
<td>Cadaverine 19.1 ± 35.1</td>
<td>30.1 ± 42.6</td>
<td>6</td>
</tr>
<tr>
<td></td>
<td>Histamine 10.5 ± 18.1</td>
<td>15.7 ± 21.7</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>Tyramine 13.9 ± 18.5</td>
<td>20.1 ± 21.9</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>Spermidine 0.4 ± 0.6</td>
<td>0.3 ± 0.6</td>
<td>3</td>
</tr>
</tbody>
</table>

RWG: Red Wagashi Gassirè, WWG: White Wagashi Gassirè, WG: Wagashi Gassirè, different superscript letters in the same row indicate a significant difference (p < 0.05) between RWG and WWG, LOQ (see Table S4), N1 and N2: number of samples in which the mineral was detected and quantified in RWG and WWG respectively, n: number of analysed samples, SD: standard deviation, LB: Lower Bound, * individual value.
Among the 10 biogenic amines determined, methylvamine, tryptamine, serotonin, and spermine were not detected in WG samples (Table S7). Putrescine, cadaverine, histamine, tyramine, and spermidine were detected in both WWG and RWG samples (but not in all samples), while 2-phenylethylamine was detected in RWG only (Table 4). Moreover, no significant difference was observed among the two types of WG regarding the biogenic amine content except for putrescine, for which higher concentrations appeared in RWG than in WWG samples (15.5 ± 14.2 versus 1.8 ± 3.7 mg/kg), but it was detected in only two WWG samples. The main biogenic amines detected in WG samples were cadaverine (19.1 ± 35.1 mg/kg) and tyramine (13.9 ± 18.5 mg/kg). The mean histamine content was 10.5 ± 18.1 mg/kg (Table 4). The total biogenic amine content of WG samples ranged from 1 to 231 mg/kg (Table S7).

For antibiotic residues, six WG samples tested positive using the BeadyPlex (Unisensor, Seraing, Belgium) (Table S8). The BeadyPlex screening test showed that the detected antibiotic residues belonged to the families of quinolones, tetracyclines, and colistin. Quinolone and tetracycline residues were found in RWG samples while colistin was found in WWG samples.

One hundred and three pesticide residues were screened (Table S9). No pesticide residues were detected by the two methods used (LC/MSMS and GC/ECD).

### 3.3. Risk Assessment for Trace Elements, Biogenic Amines, and Aflatoxin M1 in Commercial Wagashi Gassire

Dietary exposure to aluminium, lead, arsenic, and AFM1 from WG consumption in various scenarios (mean/Benin, P95/Benin, Littoral, and Borgou) and the corresponding associated risks are shown in Table 5. For all chemical hazards, the exposure in Borgou was higher than in Littoral. The dietary exposure to aluminium for high-level consumers (P95) represented 41% of the tolerable weekly intake (TWI) of 1000 µg/kg.bw/week. For the other population groups, the aluminium intake did not exceed 7% of the TWI, showing no health concern regarding aluminium exposure through WG consumption (Table 5).

<table>
<thead>
<tr>
<th>Contaminants</th>
<th>Exposure * and MOE</th>
<th>Mean/Benin</th>
<th>P95/Benin</th>
<th>Littoral</th>
<th>Borgou</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aluminium</td>
<td>Weekly Intake (µg/kg.bw/week)</td>
<td>20</td>
<td>412</td>
<td>15</td>
<td>71</td>
</tr>
<tr>
<td></td>
<td>% TWI</td>
<td>2</td>
<td>41</td>
<td>1</td>
<td>7</td>
</tr>
<tr>
<td>Lead</td>
<td>Daily Intake (µg/kg.bw/day)</td>
<td>0.003</td>
<td>0.069</td>
<td>0.002</td>
<td>0.011</td>
</tr>
<tr>
<td></td>
<td>MOE (BMDL = 0.63 µg/kg.bw/day)</td>
<td>179</td>
<td>9</td>
<td>239</td>
<td>53</td>
</tr>
<tr>
<td>Arsenic</td>
<td>Daily Intake (µg/kg.bw/day)</td>
<td>0.0009</td>
<td>0.0178</td>
<td>0.0006</td>
<td>0.0030</td>
</tr>
<tr>
<td></td>
<td>MOE (BMDL = 0.3 µg/kg.bw/day)</td>
<td>333</td>
<td>17</td>
<td>443</td>
<td>97</td>
</tr>
<tr>
<td>Aflatoxin M1</td>
<td>Daily Intake (µg/kg.bw/day)</td>
<td>0.000010</td>
<td>0.000199</td>
<td>0.000007</td>
<td>0.000034</td>
</tr>
<tr>
<td>Histamine</td>
<td>Daily Intake (mg/adult/day)</td>
<td>0.02</td>
<td>0.5</td>
<td>0.01</td>
<td>0.08</td>
</tr>
<tr>
<td>Tyramine</td>
<td>Daily Intake (mg/adult/day)</td>
<td>0.03</td>
<td>0.6</td>
<td>0.02</td>
<td>0.1</td>
</tr>
</tbody>
</table>

MOE: Margin of Exposure, TWI (aluminium): Tolerable Weekly Intake of 1000 µg/kg.bw/week (EFSA, 2008); BMDL (arsenic) = 0.3 µg/kg.bw/day for adverse effects (lung cancer, skin cancer, bladder cancer, skin lesions) of arsenic in adults (EFSA, 2009); BMDL (lead) = 0.63 µg/kg.bw/day for nephrotoxicity of lead in adults (EFSA, 2010), BMDL (aflatoxin M1) = 4 µg/kg.bw/day for liver carcinogenicity of aflatoxin M1 in animals (obtained with the BMDL of 0.4 µg/kg.bw/day for AFB1 and the potency factor of 0.1) (EFSA, 2020), * exposure calculated using lower bound mean of chemical contaminant.

For lead and arsenic, the estimated dietary exposures were below the BMDL values, with the MOEs ranging from 17 to 443 and from 9 to 239, respectively. These MOEs above 1 indicate an absence of concern regarding the lead and arsenic exposure of the Beninese population through WG consumption (Table 5). This conclusion is the same for AFM1 exposure, as the obtained MOE ranged from 20,042 to 527,768, far above 10,000 (Table 5). Moreover, the calculated cancer risks were 0.0003, 0.006, 0.0002, and 0.001 cases/100,000 persons/year, respectively, for mean, P95, Littoral, and Borgou WG consumers.
The lower bound mean of histamine and tyramine in WG were 10.5 and 13.9 mg/kg wet weight, respectively, corresponding to an exposure ranging from 0.01 to 0.5 and 0.02 to 0.6 mg/adult/day, respectively. These intakes are far lower than the NOAEL of 50 mg of histamine and 600 mg of tyramine per meal for a healthy person. Thus, exposure to histamine and tyramine through WG consumption is of low health concern.

4. Discussion

4.1. Physicochemical Parameters and Proximal Composition of Wagashi Gassirè

pH is the first physicochemical parameter used to assess product quality. The average pH (5.4) of WG samples of this study was found to be lower compared to the literature, which reported an average pH of 6.2 to 6.8 [13,37]. These reported pHs were close to the pH of the milk. Indeed, the pH of cow milk is about 6.5 [38,39]. As WG is non-salted and non-ripened cheese sold unpacked, the lower pH reported in this study could indicate fermentative activity related to the development of the microflora naturally present in the milk before processing or to post-production contamination during storage, transport, and marketing [40]. This fermentative activity would be more important with time, as the WG samples of this study were analysed three days after production. The growth and activity of spoilage microflora are mostly a function of substrate base and chemical and physical parameters such as temperature, pH, water activity, and atmosphere [40]. Abomey-Calavi is characterized by a sub-equatorial climate with an average temperature between 27 and 29 °C and rainfall ranging from 1100 to 1300 mm/year. In this city, most of the markets are spaces where food products, often unpacked such as WG, are exposed to ambient air. In such hot and humid environments, unpacked foodstuffs, like WG, with high water content (59.1% in the case of WG samples) may be subjected to the rapid growth of microflora. This growth may be more rapid in WWG than RWG, as the pH of WWG was significantly lower than that of RWG. Based on the declarations made by the WG sellers in this study, RWG was obtained by boiling WWG in the coloured water obtained by soaking and triturating sorghum cobs [5]. This hot colouring would destroy some lactic bacteria, as they are generally sensitive to high temperatures; this would explain the lower pH of WWG compared to RWG [40].

In Nigeria, protein content of 23% and fat content of 18% have been reported for WG [11]. The concentration of protein (13.5%) in the WG samples in this study was lower than expected. However, lower fat content (6–11%) was reported in comparison to the fat content (21.0%) of the WG of this study [41]. These contradictory data are related to the moisture content of WG samples from the studies mentioned. WG production involves the use of calotropain, a protease for milk coagulation [6]. This enzymatic coagulation is the step where the casein component of the milk protein system forms a gel network that entraps fat. Thus, the protein and fat contents of the cheese depend on the milk used and the coagulation process. Fat and protein contents in milk from Girolando, Borgou, Borgou-Gir, and Lagunaire cattle breeds raised in Benin were 3.8 and 3% respectively [38,39]. During the processing of the milk into WG, the cheese yield varied from 20 to 33% [5,7]. This would correspond to 8–12% of protein and 12–18% of fat in WG, based on the cows’ milk composition, the common WG yield, and assuming that 75% of the total protein and 95% of the total fat in milk were recovered in rennet-coagulated cheeses [42]. These expected protein and fat concentrations are lower than the WG samples analysed in this study (13.5% of proteins and 21.5% of fat). This could be due to the fact that the milk used by the WG processors would contain more protein and fat than the values reported in the literature.

4.2. Nutritional Values of Wagashi Gassirè

Fat content is essential in cheese for a wide range of functional properties. The fat content and the fatty acid profile of cheeses were directly related to those of the milk used [42]. Cow milk fatty acids include saturated (SFA), monounsaturated (MUFA), and polyunsaturated fatty acids (PUFA), which represent about 69, 27, and 4% of the total fat, respectively [43]. Cow milk has also a high content of palmitic, stearic, and oleic acids, and
within SFA, the major ones are palmitic (30%), myristic (11%), and stearic (12%) acids [42,43]. These fatty acid profiles of cow milk fat are similar to those found in this study for the WG. Milk, cheese, and other dairy products suffer from an adverse nutritional image largely due to a perceived association of some SFA, particularly lauric, myristic, and palmitic acids, with an increase in cardiovascular risk, obesity, and some cancers [44,45]. However, up-to-date research does not support an association between biomarkers of dairy fat intake and the risk of diabetes mellitus or cardiovascular diseases. Furthermore, the observational evidence does not endorse the hypothesis that high-fat dairy foods contribute to metabolic syndrome or cardiovascular risk and even indicate that dairy fat consumption within typical dietary patterns is inversely associated with this risk [45]. For high consumers of dairy products (P95/Benin) [29], the daily consumption of WG would correspond to an intake of 6.2 g SFA/day based on the levels of SFA in the WG samples of the present study. SFA daily intake is recommended to be as low as possible [12]. Unlike SFA, PUFA has attracted increasing attention regarding its health-promoting biological properties [45]. Due to a lack of appropriate enzymes, mammals may not synthesize de novo two PUFAs: α-linolenic acid and linoleic acid, which are considered essential fatty acids. These fatty acids play multiple roles (cell membrane structure, cell signalling, etc.) [43,44]. Only linoleic acid was found in the WG samples in this study, in an average proportion of 2.5% of total fatty acids. Daily consumption of WG (P95/Benin) of 47.4 g (according to Ingenbleek et al. [29]) represents 1.6% of the adequate daily intake of linoleic acid (4% of energy intake, i.e., 14.8 g) set for very active male adults (18–29 years) according to EFSA [12].

A large portion of worldwide milk production is used to make cheese, which contains a wide range of mineral elements. The mineral composition of cheeses is mainly associated with raw milk and the conditions of milk coagulation [42,45]. Calcium and phosphorus are the minerals present in the highest concentration in dairy products such as the WG [43]. In human nutrition, adequate calcium and phosphorus intake is essential. Obtaining enough calcium and phosphorus in the diet gives healthy bones and teeth, and calcium may also help prevent hypertension, decrease the odds of developing colon or breast cancer, improve weight control, and reduce the risk of developing kidney stones [44]. The adequate daily intake of phosphorus is 550 mg/day for adults (≥18 years), and the population reference intake of calcium is 1000 mg/day for male adults (18–24 years) [12]. Daily consumption of WG would cover these recommendations at 16.4% for phosphorus and 10.2% for calcium for heavy consumers [29]. Therefore, consumption of WG could increase the intake of calcium and phosphorus and thus contribute to the solution of deficiency of essential minerals. Significant differences were found between WWG and RWG regarding essential mineral (sodium, magnesium, phosphorus, potassium, and calcium) contents. Indeed, WWG was richer in minerals (essentials and metals) than RWG. Mineral losses were more than 50% for calcium, potassium, magnesium, sodium and rubidium, but less for phosphorus. These differences are linked to the colouring process. Boiling produced the highest loss of minerals, followed by heating with steam, stewing, and frying, due to the water solubility of minerals, which increases with the temperature [46]. Then the minerals leached from WWG into the coloured water during the colouring process. However, this conclusion needs to be confirmed by further studies that determine the mineral contents before and after colouring.

4.3. Risk Assessment
4.3.1. Risk Assessment of Arsenic and Lead in Wagashi Gassirè Cheese

Arsenic and lead are environmental contaminants that can be found in foodstuffs and that have carcinogenic and genotoxic properties [32,33]. As explained above, the collection point of WG samples of this study may be susceptible to environmental contamination. WG is sold unpacked, and dust could be a source of lead contamination because it deposits on WG during production and selling. Furthermore, the trace metal contamination may be related to the raw milk and materials used for WG production. In Benin, sources of lead exposure are varied; key risk factors for human exposure include water sources
(piped water), hunting (animals killed with lead-contaminated bullets), food (fish, meat, rice), occupation, and soil [19,20]. The average lead contamination of WG observed in the present study was 0.08 mg/kg wet weight, exceeding the maximum level set by Beninese regulations [47], the European Commission [48], and the CODEX [49], i.e., 0.02 mg/kg in milk and dairy products. This lead content in WG samples was higher than that reported (0.002 mg/kg) by Jitaru et al. [50] in milk and dairy products from Borgou and Littoral during their Sub-Saharan Africa Total Diet Study. Arsenic occurs naturally in the earth’s crust, and it is a constituent of more than 200 mineral species, especially those including sulphide. Anthropogenic sources of arsenic release to the environment include industrial emissions, mainly from non-ferrous mining and smelting, metal-using industry, and the production of energy from fossil fuels [33]. The average arsenic content in WG samples (0.02 mg/kg wet weight) was lower than that found in milk and dairy products (0.003 mg/kg) in Benin [50]. The lead content was significantly higher in WWG than in RWG, probably due to the colouring process as explained above for minerals [46]. Lead is well known to have detrimental effects on human health, including on the nervous, renal, cardiovascular, reproductive, immune, and hematologic systems. In children, lead is associated with poor child neurodevelopment [18]. The main adverse effects reported to be associated with long-term ingestion of inorganic arsenic in humans are skin lesions, cancer, developmental toxicity, neurotoxicity, cardiovascular diseases, abnormal glucose metabolism, and diabetes [33]. The calculated risk for lead and arsenic based on the nephrotoxicity of lead and the adverse effects of arsenic (lung cancer, skin cancer, bladder cancer, skin lesions) showed an MOE greater than 1 in all cases. These MOEs above 1 indicate an absence of concern regarding the lead and arsenic intake from WG consumption [32,33]. However, these estimates need to be extended to all dietary sources of lead and arsenic. Lead presents a public health concern in Benin. In Sô-Ava (Abomey-Calavi, Benin), 31.3% of blood lead levels of pregnant women were above the 50 µg/L threshold set by the US Centers for Disease Control and Prevention [19]. In Allada (Benin), the prevalence of blood lead levels higher than the 50 µg/L threshold in children was 54.8% at 1 year of age and 59.5% at 6 years of age, showing an increase in lead exposition during life [20]. An increase in blood lead levels of 12 µg/L in the child may decrease the intelligence quotient score by one point [32]. Lead exposure continues to be a major public health concern as no level of lead exposure can be considered safe. Due to the chronic exposure of consumers and the potential long-term health effects of lead and arsenic exposure, a monitoring program should be designed to reduce their prevalence in foodstuffs.

4.3.2. Risk Assessment of Aluminium in Wagashi Gassirè Cheese

Aluminium has been extensively used in the industry, and it is currently added to a vast number of products available to everyone. However, food and water represent the most common forms of human exposure to this metal [51]. As with lead, aluminium content in WG samples (74.6 mg/kg wet weight) was found to be higher than that reported (0.37 and 0.09 mg/kg wet weight in Littoral and Borgou, respectively) by Jitaru et al. [50]. In developing countries, inexpensive aluminium cookware made from mixed scrap metal, including engine and electronic appliance parts, cans, and other aluminium scraps, is widely used. Aluminium, lead, and other metals leach from cookware during cooking, increasing metal exposure through food consumption [17,50]. Jitaru et al. [50] compared concentrations of foods prepared in stainless steel and artisanal cookware. Mean concentrations of aluminium and lead were 6.7 and 26 times higher, respectively, when tomatoes were cooked in artisanal aluminium cookware compared to stainless steel. In Benin, the production equipment of WG was generally rudimentary: sieves with iron filters, plastic colanders, aluminium cooking pots, and plastic material for storage before and during selling [7]. The use of aluminium cooking pots may explain the highest concentration found for aluminium and lead in the WG samples in this study. Ingenbleek et al. [52] reported mean aluminium dietary exposures of 457 and 2358 µg/kg.bw/week, in Littoral and Borgou, respectively. In this study, aluminium exposures of 15 and 71 µg/kg.bw/week
were found in Littoral and Borgou, respectively. The worst case showed an exposure of 412 µg/kg.bw/week, representing 41% of the TWI of 1000 µg/kg.bw/week [17]. This means an absence of concern about the aluminium intake from WG consumption. However, actions should be taken to reduce this exposure, as aluminium is considered to produce neurotoxicity and embryotoxicity, as well as to affect the male reproductive system. In addition, aluminium could increase the probability of developing Alzheimer’s disease [51].

4.3.3. Risk Assessment of Histamine and Tyramine in Wagashi Gassirè Cheese

Synthesis of biogenic amines is possible by the conjugation of several conditions: the availability of the substrate amino acids, the presence of microorganisms with the appropriate catabolic pathway activated, and environmental conditions favourable to the decarboxylation activity [53,54]. These conditions depend on several factors, such as the milk treatment, use of starter cultures, NaCl concentration, time, the temperature of ripening and preservation, pH, temperature, and post-ripening technological processes [54,55]. The main biogenic amines generally found in cheeses are putrescine, cadaverine, histamine, tyramine, and tryptamine [14]. This pattern was observed in WG samples in this study as well, except for tryptamine. Spermidine is converted by microorganisms to spermine, and its concentration is generally lower than that of spermine [33]. The non-detection of spermine in the WG samples indicates low microbial activity. Intake of low amounts of biogenic amines normally does not have harmful effects on human health. However, when the amount in food is too high and the detoxification ability is inhibited or disturbed, biogenic amines could cause adverse health effects [56]. In some non-ripened cheeses, 82 to 477 mg of biogenic amines have been detected per kilogram of cheese [53,56]. In this study, the biogenic amine content of WG samples was between 1 and 231 mg/kg wet weight, similar to the values reported by the literature. According to the European Food Safety Authority, a threshold level of 50 mg/meal for histamine and 600 mg/meal for tyramine in healthy persons should be considered [14]. These established NOAEL are higher than the exposure levels calculated in this study, indicating an absence of concern about the histamine and tyramine intake from WG consumption.

4.3.4. Risk Assessment of Aflatoxin M1 in Wagashi Gassirè Cheese

The prevalence of AFM1 in WG samples is due to the contamination of feed eaten by lactating cows with AFB1 and its subsequent biotransformation into AFM1 excreted in the milk. Nearly 0.3–6.2% of the AFB1 ingested by lactating cows is converted into AFM1 in the milk, and the level of AFM1 is approximately 3–5 times greater in cheese than in the original milk due to the concentration effect of the coagulation process [57]. The AFM1 content of the milk used to produce the WG analysed in this study may range from 0.1 to 0.06 µg/kg wet weight, based on the lower bound mean AFM1 of the WG samples. This is lower than or just above the maximum level set by the Beninese regulations [47] and the European Commission [48], i.e., 0.05 µg/kg, and that of the CODEX [49], i.e., 0.5 µg/kg, in raw milk. A few reports have been published on AFM1 in milk [38] and WG produced in The Republic of Benin [13,21]. The difference between the results reported by these authors (no detection of AFM1 in WG) and the ones in this study may be related to the different levels of AFM1 in the milk used to produce WG or to the analytical method used. The different levels of AFM1 in the milk used to produce WG could also be explained by the significant difference regarding AFM1 concentration between WWG and RWG in this study. Indeed, AFM1 concentration was significantly higher in WWG than in RWG samples, suggesting that the milk used to produce WWG would be more contaminated than that used for RWG production [57]. The calculated MOE for liver cancer due to AFM1 intake were far above 10,000, indicating an absence of concern about the AFM1 intake from WG consumption [34]. Kortei et al. [58] reported a health concern about AFM1 intake, as MOEs lower than 10,000, through raw cow milk consumption, were obtained in different regions of Ghana. These authors also reported a cancer risk
of $1.94 \times 10^{-3}$–0.07 cases/100,000 person/year for AFM1 intake, which is lower than that obtained in this study ($0.2 \times 10^{-3}$–10$^{-3}$ cases/100,000 person/year).

4.4. Dioxins Content, Antibiotics, and Pesticides Residues of Wagashi Gassirè

Dioxins are persistent organic pollutants (POPs) found everywhere in the environment, including in plants, animals, and human beings. In this study, dioxins were found in some WG samples below the maximum level set by the Beninese regulations [47], i.e., 6 pg/g, and the European Commission [48], i.e., 4 pg/g fat for the sum of dioxins and dioxin-like PCBs in milk and dairy products. Many human activities, such as road traffic, directly or indirectly generate dioxins. Emissions from the combustion of plastic wastes in household furnaces contribute to significant air pollution containing dioxins and PCBs [59]. The collection point of the WG samples in this study may be susceptible to environmental contamination. WG is sold unpacked, and dioxins and PCBs could deposit on WG during selling since atmospheric deposition is generally the dominant pathway of PCBs into agricultural food chains [59]. Furthermore, the dioxin and PCB contamination may be related to the raw milk used for WG production.

The misuse of antibiotics by dairy farmers and the lack of respect given to the withdrawal period until milking are the main reasons for the antibiotic residue presence in milk and dairy products [60]. Tetracyclines, fluoroquinolones, and colistin were the antibiotic residues detected in the WG samples in this study. In West Africa, the most commonly used antibiotics by dairy farmers and detected in milk and dairy products are tetracyclines, beta-lactams, aminoglycosides, penicillin, sulphonamides, and macrolides [60].

Most of the selected pesticide residues tested in this study were those commonly used in agriculture in Benin [61]. Therefore, finding them in WG is expected. However, no pesticide residues were detected from the 103 pesticides tested. This indicates that the milk used for WG production was not contaminated or that the WG preservation processes caused a degradation of the pesticide residues tested below their detection limit. Indeed, Gafour [62] showed that pasteurization, sterilization, fermentation, or coagulation cause a net degradation of malathion: 82.2% for pasteurization in 20 min versus 91.2% for sterilization in 15 min. The most used pesticides in northern Benin are endosulfan (a forbidden insecticide) and glyphosate (an herbicide) [28,61]. Endosulfan was tested in this study but glyphosate was not. Cypermethrin (lower bound mean of 0.5 and upper bound mean of 0.7 µg/kg wet weight) and chlorpyrifos (lower bound mean of 0.0 and upper bound mean of 0.3 µg/kg wet weight) are some pesticide residues found in some dairy products consumed in Benin [63].

5. Conclusions

This study contributed to the physicochemical and nutritional characterization of WG and the assessment of some chemical risks associated with its consumption. The detection of linoleic acid, an essential fatty acid, and the essential minerals and protein contents argue for the good nutritional properties of WG. Colouring WG was associated with important essential mineral losses. The observed concentrations of AFM1 and lead indicate they are the most relevant chemical hazards of WG. The dietary exposure to arsenic, lead, aluminium, AFM1, histamine, and tyramine indicated no public health concern related to these chemical contaminants through WG consumption. Regarding the sample size in this study, it would be appropriate to conduct a larger study to confirm the influence of colouring on WG quality. Moreover, studies should be conducted to identify the main sources of chemical contamination of WG, from production to consumption, to propose strategies for reducing chemical contamination of WG. Finally, risk assessment of aluminium, arsenic, lead, AFM1, histamine, and tyramine should be extended to all dietary sources of these chemical contaminants.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/dairy5020022/s1, Analytical methods and Tables S1–S9.

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