ARTICLE INFO

Keywords:
Calotropis procera
Milk
Sanitary quality
Chemical contaminants
Traditional cheese

ABSTRACT

Purpose: Wagashi Gassirè (WG) is a traditional cheese produced from cow milk following local processing methods in Benin. The aim of this study was to describe the milk processing methods and the preservation practices with the objective of improving WG production and sanitary quality.

Methods: A survey was carried out among 390 actors (84 dairy farmers, 165 producers, 53 traders, and 88 consumers) from two municipalities (Dassa and Nikki) in Benin.

Results: WG is highly preferred by consumers for its whiteness (63.0%), softness (24.7%), smoothness (19.2%), and firmness (13.7%). WG production is based on the coagulation of milk using Calotropis procera extracts as coagulant. Six milk processing methods, including three new WG production methods were identified, depending on how the C. procera extracts were pre-treated and used during WG production. Boiling (67%) was the most widely used as WG preservation method. The use of aluminium cooking pots (100% of WG producers), WG open-air production (66.7% of producers) and antibiotic misuse (59.3% of dairy farmers) may lead to the chemical or microbiological contamination of WG.

Conclusions: Six WG production and six preservation methods were identified after the survey among WG producers and traders. Future studies should assess the sanitary and physico-chemical quality of WG from the identified processing and preservation methods. The next step of research should be also focused on the development of specific standards to produce a better quality of WG.

1. Introduction

Around the world, a diversity of cheeses (1400 according to some estimates) is available (McSweeney et al., 2017). Cheese processing is one of the most significant methods of preserving the major milk components such as proteins, minerals, and fat. The most important step during cheese processing is milk coagulation, achieved through the addition of enzymes to the milk (Mazorra-Manzano et al., 2018). During coagulation, the casein component of the milk protein system forms a gel network that entraps fat (Mazorra-Manzano et al., 2018; Baba-Moussa et al., 2007). Four types of enzymes are used in cheese factories, which are animal rennet, genetically engineered chymosin, coagulants of microbial origin, and plant-derived clotting enzymes (Jacob et al., 2011). The plant-derived clotting enzymes are an alternative to animal rennet and have the advantage of being accessible and easy to use, even on a small scale (Rayanatou 2017). Their use is a common practice in Mediterranean, West African, and Southern European countries. In Portugal and Spain, the extract of Cynara cardunculus flowers is used to produce

* Corresponding author.
E-mail address: dossoual6@gmail.com (D.W. Alphonse).

https://doi.org/10.1016/j.heliyon.2022.e10605
Received 18 March 2022; Received in revised form 1 July 2022; Accepted 7 September 2022
2405-8440/© 2022 The Authors. Published by Elsevier Ltd. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).
traditional cheeses such as Serra da Estrella, Castelo Branco, Torta Del Casar, La Serena or Los Ibores (Roseiro et al., 2003). In Mexico, extracts from Solanum elaeagnifolium have been used for production of pasta flata cheese type, known as “Asadero” while the traditional cheese “Gybnay baya”, widely consumed in Sudan, is produced using the extract of the fruits of Solanum dubium (Mazorra-Manzano et al., 2018; Rayanatou 2017; Kheir et al., 2011).

In Benin, fresh cow milk is processed, generally by the sociocultural group Peulh, into a traditional local cheese called Wagashi Gassir (WG) which remains in the country, the most consumed dairy product (Mattiello et al., 2017; Dossou et al., 2006). WG is a soft cheese obtained by hot (65–70 °C) coagulation of fresh whole milk using Calotropis procera extract that contains a plant enzyme known as calotropain (Mattiello et al., 2017; Baba-Moussa et al., 2007; Dossou et al., 2006). In Benin, WG is mainly produced in the northern part of the country where dairy farming is more developed and widespread (MAEP 2014; Kora 2005).

WG is an important source of proteins (23–36%), fats (18–27%), and minerals (1.3–1.8%) (Adyeye 2016; Alalade and Adeneye 2006; Uzeh et al., 2006; Appiah et al., 1998). It could help to meet the daily nutritional requirements of proteins (0.8 g), calcium (2,500 mg), and other nutrients for adults (CSS 2016). WG is highly valued and consumed daily by local consumers and also by people from neighbouring countries where it is used as a meat and fish substitute in various dishes (Baba-Moussa et al., 2007). WG and other dairy products contribute to more than 50% of the annual Peulh household income (Sessou et al., 2013). Income from the marketing of dairy products provides remuneration and work motivation to Peulh women. This almost regular income allows them to provide for other household needs and gives them decision-making power and autonomy (Chabi-Toko et al., 2015). As WG also requires a vast distribution network, it is an important source of income also for other actors of this value chain besides milk processors.

Several studies have shown the presence of dioxins and polychlorinated biphenyl (0.048 pg TEQ/g wet weight) and some pesticides, such as cypermethrin (0.7 μg/kg wet weight) and chlorpyriphos-ethyl (120 μg/kg wet weight), in milk and dairy products from Benin (Vaccher et al., 2020; Ingenbleek et al., 2019; Dossou et al., 2016). However, it is not well known whether and at which step(s) of the production and distribution chain (from dairy farmers to consumers) WG may be chemically contaminated. Moreover, to our knowledge, the main factors associated with WG chemical contamination during its storage have not been precisely documented. Therefore, it is necessary to identify and describe the traditional milk processing methods to produce WG, preservation practices, and conditions of sale to propose appropriate alternatives in order to decrease the risk of chemical contamination.

2. Material and methods

The aim of this study was to identify and describe the traditional milk processing methods, WG preservation practices, and conditions of sale to develop and propose appropriate alternatives to improve WG production, storage, and sale.

2.1. Characteristics of the study area

The study was carried out in Dassa and Nikki, two municipalities of the agricultural development pole identified in Benin Republic for diversification and intensive cattle production (Figure 1). These municipalities are part of the main cattle and WG production areas in Benin. Nikki is a WG production area with a dairy processing unit. Dassa is a WG production area and a crossroad and important WG commercial and transaction area (MAEP 2014).

Figure 1. Geographical localization of the surveyed actors.
2.2. Survey sample size and selection of respondents

This survey was carried out using a questionnaire to collect information on WG production/sale. The snowball method was used to identify the included people: dairy farmers (who had at least 10 cows), active processors (still active in the last year and with at least 10 years of experience in WG production), traders (still active in the last year and with at least 10 years of experience in WG sale), and WG consumers (Johnston and Sabin 2010). Data saturation threshold was used to determine the sample size. Consumers were randomly selected without any specific criterion.

2.3. Collected data

A semi-structured questionnaire was used for the one-to-one interviews with the different actors identified. The data collected were on: WG production (milk processing and WG preservation methods used by processors and traders); potential sources of contamination during processing, preservation, and trading; quality criteria according to processors and consumers; dairy farmers’ practices that could affect the quality of the milk used as raw material to produce WG. Data were collected in the field using the Epicollect5 system through web and mobile applications (Aanensen et al., 2009).

2.4. Production monitoring

Based on the survey results, the three (3) most used methods were monitored to determine WG production yields. A total of 24 trials of productions (8 trials per processing methods) were carried out. For each unit operation (UO), time (from start to end of the UO), temperature (at the UO end), and the ingredient quantity were measured. The production yields were determined by dividing the quantity of cheese obtained by the quantity of milk used for the production.

2.5. Statistical analysis

Data from the semi-structured questionnaire were translated in a structured form and loaded in MS Excel 2013 to perform the descriptive analysis using the SAS System 9.4 software. The confidence intervals (CI) at 95% were determined for different variables before frequency comparison with the Chi-square test. The 95% CIs were calculated using the following formula: $CI = \left[ \hat{p} - 1.96 \sqrt{\frac{\hat{p}(1-\hat{p})}{n}} \right] \text{ and } \left[ \hat{p} + 1.96 \sqrt{\frac{\hat{p}(1-\hat{p})}{n}} \right]$ where $\hat{p}$ is the proportion and n the number of respondents for the variable. The Z-test was used to compare percentages in pairs for variables with more than two classes.

For the data on production monitoring, means were compared with a simple linear regression analysis (ANOVA). Statistical significance was set at 5%.

2.6. Ethical approval

The study protocol was approved by the Faculty of Agronomic Sciences of the University of Abomey-Calavi, Benin Republic. The survey and the production monitoring were conducted according to ethical guideline of the ethics committee of the University of Abomey-Calavi. All the interviews have been carried out with informed written consent of the respondents.

3. Results

3.1. Sociocultural status of the different actors

For this study, 390 people were interviewed using a semi-structured questionnaire in the municipalities of Dassa and Nikki (Table 1). Table 2 presents the sociocultural profile of the respondents. Dairy farmers (n = 84) were mostly men (94.0%), married (92.4%), and illiterate (92.8%). The percentage of dairy farmers younger than 30 and older than 60 years (16.7% and 25.0%, respectively) was significantly lower ($P < 0.001$) than that of 30–60-year-old dairy farmers (58.3%). Dairy farmers belonged to the Peulh (86.9%) and Gando (13.1%) sociocultural groups. The Gando group is found exclusively in Nikki, whereas Peulh farmers are in both municipalities.

WG producers (n = 165) were only women who belonged to different sociocultural groups: Peulh (61.8%), Gando (35.1%), Idaatcha (1.8%), and Fon (1.2%). These women were mainly married (95.1%) between 30 and 60 years of age (75.6%), and illiterate (92.7%).

Traders (n = 53) belonged to many sociocultural groups: Peulh (32.0%), Baatonou (20.7%), Fon (13.2%), and Idaatcha (11.3%). WG trading was carried out almost exclusively by women (98.1%), generally married (92.4%). Approximately 44% of traders were literate.

### Table 1. Typology of the respondents in the two municipalities.

| Typology of respondents | Number of respondents per municipality | Sub-total |
|--------------------------|----------------------------------------|----------|
| Dairy farmers            |                                        |          |
| Dassa                    | 54                                     | 30       |
| Nikki                    | 30                                     | 84       |
| Processors               |                                        |          |
| Dassa                    | 90                                     | 75       |
| Nikki                    |                                         | 165      |
| Traders                  |                                        |          |
| Dassa                    | 35                                     | 18       |
| Nikki                    |                                         | 53       |
| Consumers                |                                        |          |
| Dassa                    | 56                                     | 32       |
| Nikki                    |                                         | 88       |
| Total                    | 235                                     | 155      |
|                           |                                         | 390      |

### Table 2. Sociocultural profile of the different actors surveyed (% ± CI).

| Sociocultural characteristics | Dairy farmers (n = 84) | Processors (n = 165) | Traders (n = 53) | Consumers (n = 88) |
|------------------------------|------------------------|----------------------|------------------|---------------------|
| Sex                          |                        |                      |                  |                     |
| Men                          | 94.0 ± 5.0a            | 0.0 ± 0.0b           | 1.9 ± 3.8b       | 51.7 ± 10.4c        |
| Women                        | 5.9 ± 5.0b             | 100.0 ± 0.0b         | 98.1 ± 3.8b      | 48.3 ± 10.4c        |
| Age (in years)               |                        |                      |                  |                     |
| <30                          | 16.7 ± 9.4a            | 12.2 ± 7.1a          | 40.0 ± 16.2b     | 50.8 ± 12.4c        |
| ≥30 and ≤60                  | 58.3 ± 12.5b           | 75.6 ± 9.3b          | 54.3 ± 16.5b     | 41.8 ± 12.2c        |
| >60                          | 25.0 ± 10.9a           | 12.2 ± 7.1a          | 5.7 ± 7.9b       | 7.9 ± 6.7b          |
| Marital status               |                        |                      |                  |                     |
| Married                      | 96.4 ± 3.9a            | 95.1 ± 3.3a          | 92.4 ± 7.1a      | 70.8 ± 9.4a         |
| Single                       | 3.6 ± 3.9b             | 4.9 ± 3.7b           | 7.6 ± 7.1b       | 28.2 ± 9.4b         |
| Matrimonial status           |                        |                      |                  |                     |
| Monogamy                     | 45.7 ± 10.8a           | 43.9 ± 7.7b          | 48.9 ± 14.0b     | 79.4 ± 9.9b         |
| Polygamy                     | 54.3 ± 10.8a           | 56.1 ± 7.7a          | 51.0 ± 14.0b     | 20.6 ± 9.9b         |
| Educational level            |                        |                      |                  |                     |
| None                         | 92.8 ± 5.3a            | 92.7 ± 3.9a          | 56.6 ± 13.3b     | 16.8 ± 7.8b         |
| Primary                      | 5.9 ± 5.0b             | 5.4 ± 3.4b           | 24.5 ± 11.6b     | 24.7 ± 8.9b         |
| Secondary                    | 1.2 ± 2.3c             | 1.8 ± 2.0c           | 15.1 ± 9.6b      | 43.8 ± 10.3c        |
| University                   | 0.0 ± 0.0f             | 0.0 ± 0.0f           | 3.8 ± 5.1c       | 14.6 ± 7.3c         |
| Sociocultural group          |                        |                      |                  |                     |
| Peulh                        | 86.9 ± 7.2c            | 61.8 ± 7.4c          | 32.0 ± 12.6d     | 7.8 ± 5.6b          |
| Gando                        | 13.1 ± 7.2b            | 35.1 ± 7.3c          | 2.2 ± 3.1c       |                     |
| Fon                          | 1.2 ± 1.8c             | 13.2 ± 9.1bc         | 16.8 ± 7.8bc     |                     |
| Idaatcha                     | 1.8 ± 2.0c             | 11.3 ± 8.5bc         | 24.7 ± 8.9b      |                     |
| Mahi                         | 3.8 ± 5.1b             | 2.2 ± 3.1c           | 2.2 ± 3.1c       |                     |
| Dendi                        | 9.4 ± 7.9bc            | 2.2 ± 3.1c           |                     |                     |
| Minan                        | 5.7 ± 6.2c             |                     |                     |                     |
| Baatonou                     | 20.7 ± 10.9c           | 14.6 ± 7.3c          |                     |                     |
| Yoruba                       | 3.8 ± 5.1b             | 5.6 ± 4.8b           |                     |                     |
| Adjia                        | 3.4 ± 3.8b             |                     |                     |                     |
| Others                       | 20.2 ± 8.3c            |                     |                     |                     |

The letters a, b, c, and d notify significant difference ($P < 0.05$) between percentages for the same variable, CI: confidence interval at 95%.
Consumers (n = 88) were mostly younger than 60 years of age, and belonged to the same sociocultural groups as the processors and also to other sociocultural groups (Dendi, Mina, Mahi, Yoruba, and Adja). They were mostly literate (83.2%) and 14.6% had a university diploma.

3.2. Traditional milk processing methods for Wagashi Gassiré production

3.2.1. Different steps of the traditional processing methods to prepare Wagashi Gassiré

Two WG types were produced: white and coloured WG. On the basis of the processors’ responses, white WG (WWG) production was performed following ten unit operations (UO) grouped into four steps (Figure 2):

Step 1: Milk preparation for processing. In the first UO (UO1), milk was filtered. This UO was performed by 66.1% of processors. Then, filtered milk was heated (UO2: 59 °C for 20 min). Heating was done by 82.7% of processors.

Step 2: Extract preparation. WG is produced using *C. procera* extracts. The combination of *C. procera* leaves and stem was used by 66.1%, stem alone by 32.1%, and sap by 9.1% of processors. *C. procera* leaves alone (1.8%) and *Carica papaya* leaves (1.8%) were rarely used. Fermented corn supernatant (1.2%) and salt (3.6%) also were used, but always in combination with *C. procera* extract. Usually, for the coagulant preparation, *C. procera* leaves or stems were washed using well, river or tap water (UO3: 13.5%) and crushed (UO4: 95.5%). Crushed *C. procera* was triturated (UO5: 98.7%) with heated milk (80.8%) or water (18.0%). The resulting mixture of milk/water and *C. procera* was filtered (UO6: 71.2%) and used as coagulant. Some processors (UO7: 5.5%) used all the crushed extract as coagulant and then skimmed off the *C. procera* rubble.

Step 3: Milk coagulation. The coagulant was added (UO8: 100%) to the heated milk. After the coagulant addition, the milk became solid and the coagulum obtained was cooked (91 °C for 34 min, UO9: 100%).

Step 4: Mould filling/drainage. The cooked coagulum was put into a mould (UO10: 100%): oval vegetable or plastic colanders of different sizes that allow the simultaneous draining.

The obtained WG is white and soft. These four steps were common to all the milk processing method variants.

3.2.2. Processing method variants

Milk processing variants to prepare WG depended on the used *C. procera* parts (sap, stem, leaves; alone or in combination) for milk coagulation, and on their pre-treatment (trituration or not in water or milk) before coagulation. In total, six WG production variants were identified on the basis of the method used to prepare the coagulant (Figure 3 and Table 3):

- **Method 1 (M1).** *C. procera* sap was added to the heated milk for coagulation. This method was used by 9.1% of processors.
- **Method 2 (M2).** *C. procera* crushed leaves and stems were added to the heated milk for coagulation. Then, *C. procera* rubble was skimmed. This method was used by 5.5% of processors.
- **Method 3 (M3).** *C. procera* crushed leaves and stems were triturated in a separate small quantity of milk. The resulting solution was filtered and the filtrate was added to the heated milk for coagulation. This method was used by 49.7% of processors.
- **Method 4 (M4).** *C. procera* crushed leaves and stems were triturated in a separate small quantity of water. The resulting solution was filtered and the filtrate was added to the heated milk for coagulation. This method was used by 9.1% of processors.
- **Method 5 (M5).** *C. procera* crushed stem was triturated in a separate small quantity of milk. The resulting solution was filtered and the filtrate was added to the heated milk for coagulation. This method was used by 26.1% of processors.
- **Method 6 (M6).** *C. procera* crushed stem was triturated in a separate small quantity of water. The resulting solution was filtered and the filtrate was added to the heated milk for coagulation. This method was used by 7.3% of processors.

![Figure 2. Fresh milk processing into Wagashi Gassiré.](image-url)
The choice of processing variant varied among sociocultural groups (Table 3). The M2 variant was used only by the Peulh group (5.5% of all processors). Conversely, the M3 variant was the most widespread (49.7%) method and the only method used by the Fon and Idaatcha sociocultural groups. The other four methods (M1, M4, M5 and M6) were used by the Peulh and Gando sociocultural groups (45.9% of all processors).

Then, for three most used processing methods (M1, M3, and M5), according to the survey results, the production conditions (heating time/temperature, cooking time/temperature, coagulant concentration for M3 and M5) and WG yield were monitored (Table 4). The ratio of milk to C. procera was 7.9 for M3 (i.e. 7.9 g of milk was used per gram of C. procera extract) and 19 for M5 (i.e. 19.0 g of milk per gram of C. procera extract) (P < 0.001). The cheese yields varied from 27.0 (M1) to 33.0% (M5), but were not significantly different among methods. These yields were obtained using coagulation (i.e. heating) temperatures that ranged from 56 to 64 °C.

3.3. Wagashi Gassir preservation practices by processors and traders

Analysis of the semi-structured interviews of processors and traders identified six WG preservation practices: boiling, sun-drying, colouring, immersion, smoking, and frying (Table 5).

Specifically, WG was boiled in water (69.4%), in salty water (17.9%), or in water with shea leaves (1.9%). Sometimes, WG was packed in a plastic bag (20.4%) before boiling. Boiling in water (62.2%) and in plastic bags (97.6%) were more used in Dassa than in Nikki (37.8% and 2.4%; P < 0.01).

Sun-drying (23.3%) was more common in Nikki than in Dassa (85.4% and 14.6%; P < 0.0001), and more frequently done by processors than traders (85.4% and 14.6%; P < 0.05).

WWG was coloured to obtain red WG using various dyes, such as sorghum cobs, sorghum panicles and teak (Tectona grandis) leaves. Sorghum cobs and panicles were soaked and then triturated or boiled with potash, bicarbonate, or salt. WWG was then immersed in the obtained coloured water. Teak leaves were only boiled to obtain coloured water for WWG colouring. Colouring was only used by processors and more in Nikki than Dassa (80.0% and 20.0%; P < 0.01).

Immersion in whey (4.6%), well or tap water (3.9%) was used for short-term preservation, before selling during the same day, and only by processors.

Smoking (2.4%) and frying in oil or fat (3.8%) were less frequently used by processors and also traders. They represented less than 7.0% of all WG preservation methods. Frying was only used by traders from Nikki.

Boiling, the main preservation practice, was used by processors and traders belonging to all identified sociocultural groups. Conversely, colouring and immersion in whey were specific to the Peulh and Gando groups. Frying was not reported by Peulh participants.

---

**Figure 3.** Different Wagashi Gassir production methods in function of the coagulant used. M1: sap, M2: leaves and stem without trituration or filtration followed by skimming; M3: trituration in milk of leaves and stem; M4: trituration in water of leaves and stem; M5: trituration of stem in milk; M6: trituration of stem in water.

---

**Table 3.** Utilization rate of each processing method by the different sociocultural groups.

| Processing method | Number of respondents (n = 165) | Percentage | Sociocultural groups |
|-------------------|---------------------------------|------------|----------------------|
| M1                | 15                              | 9.1        | Peulh, Gando         |
| M2                | 9                               | 5.5        | Peulh                |
| M3                | 82                              | 49.7       | Peulh, Gando, Fon, Idaatcha |
| M4                | 15                              | 9.1        | Peulh, Gando         |
| M5                | 43                              | 26.1       | Peulh, Gando         |
| M6                | 12                              | 7.3        | Peulh, Gando         |

M1: sap, M2: leaves and stem without trituration or filtration followed by skimming; M3: trituration in milk of leaves and stem; M4: trituration in water of leaves and stem; M5: trituration of stem in milk; M6: trituration of stem in water.

---

**Table 4.** WG production conditions and yields.

| Processing methods (mean ± SD) | M1 | M3 | M5 |
|-------------------------------|----|----|----|
| Heating time (min)            | 20 ± 12a | 23 ± 8a | 21 ± 6a |
| Heating temperature (°C)      | 56 ± 9a  | 58 ± 10a | 64 ± 10a  |
| Ratio milk/C. procera extract (g/g) | 7.9 ± 3.7a | 19.0 ± 6.0b |
| Cooking time (min)            | 35 ± 7a  | 26 ± 12a | 30 ± 11a  |
| Cooking temperature (°C)      | 92 ± 3a  | 90 ± 7a  | 90 ± 5a  |
| Mould filling/drainage time (min) | 11 ± 3a  | 16 ± 3a  | 12 ± 7a  |
| Cheese yield (%)              | 27 ± 6a  | 31 ± 8a  | 32 ± 9a  |

a and b show significant differences (P < 0.05) between processing methods. SD: standard deviation.


3.4. Dairy farmers’ practices and Wagashi Gassiré production conditions

The dairy farmers reported that the quantities of antibiotics administered to animals did not respect the recommended doses (59.3% of interviewed farmers) and that the treated animals may be milked on treatment day (54.6%). This milk was then consumed (85.7%) or processed (83.3%). Dairy farmers said that they used herbicides (2,4-D amine salt, atrazine, prometryn, fluorometuron and glyphosate) in cotton, maize, cassava, yam, and other crop fields (79.5%).

Moreover, 66.7% of processors did not have a dedicated production room and 43.6% of the existing rooms were not protected against rain, dust, wind, and pests. The production equipment was generally rudimentary: sieves with iron filters (49.5%), plastic colanders (95.0%), aluminium cooking pots (100.0%), and plastic material for WG storage before and during selling.

According to the traders, WG sale was ambulant (28.9%) and along roads (62.2%). Containers were often uncovered (85.7%). In the various sale locations, 97.1% of traders were exposed to dust, rain, and wind.

3.5. Wagashi Gassiré quality criteria

According to the processors, WG quality mostly depended on the quality of the milk used as raw material and on the main UO of the processing methods, such as coagulation (72.4%) and cooking (14.7%). They also said that the cleanliness of the production materials, the absence of contact between milk and salt/water and the use of fresh C. procera leaves or stems were important measures. The milk used as raw material should have been milked on the day of production (30.1%). Milk from recently calved cows (colostrum) should not be used.

According to the consumers, the most important criteria to assess WG quality were: whiteness (63.0%), softness (24.7%), smoothness (19.2%), and firmness (13.7%). Other criteria were also used, such as hardness (8.2%) and the absence of oil on the product surface (1.4%). The same criteria were used also by processors, except for firmness and absence of oil on the product surface. However, the importance of these criteria varied in function of the actors. Three additional criteria were used by processors: cooked taste and aroma (6.9%), absence of rubble (2.8%), and milk aroma (0.7%). Whiteness was the most important criterion to differentiate between processors (77.8%) and consumers (63.0%) (P <0.05).

4. Discussion

4.1. Importance of peulh women in the Wagashi Gassiré value chain

The sociocultural group Peulh is the main actor of the WG value chain. Indeed, dairy farmers are mostly Peulh who own 95.0% of the national cattle herd (Dahouda et al., 2019; Youssao et al., 2013). In Peulh culture, milking is carried out by men and women, but milk is exclusively the property of married women who are in charge of its management (Chabi-Toko et al., 2015). In agreement, in this study, women represented less than 5.0% of dairy farmers, but more than 95.0% of processors and traders, showing their importance in this value chain. Women processed milk mainly into WG. In Benin, traditional food production actors (particularly in agri-food crafts) are mostly 30 to 50-year-old women (Assogba et al., 2019; Dah et al., 2018; Badoussi et al., 2014; Saroumi and Kpenavoun 2014). The proportion of literate participants was higher among traders than processors. Indeed, trading require skills related to travelling and accounting that are more accessible to literate people younger than 60 years of age. This may explain the higher proportion of literate and the lower proportion of older people in this group. As the main actors of WG production were illiterate women, intervention in this sector should be targeted mainly to this group.

4.2. The efficiency of milk processing and Wagashi Gassiré preservation methods

Cow milk is the main raw material used for WG production (Dossou et al., 2006). More than a third of the surveyed processors reported difficulties in obtaining milk. Milk availability depends mainly on the season. During the rainy season, milk production increases due to the abundance of fodder resources (Dah et al., 2018; Gagara et al., 2017). However, in the dry season, their scarcity leads to transhumance of dairy farmers to other areas where fodder resources are available. The transport of milk to the municipalities of origin then depends on the transport infrastructure quality (Corniaux et al., 2005).

The main operations identified for milk processing into WG were filtration, heating, coagulation, cooking, and moulding/draining, as previously reported (Anihouvi et al., 2019; Gagara et al., 2019; Dah et al., 2018; Dossou et al., 2006; Belewu et al., 2005). The main coagulant used for WG production was C. procera extract (Baba-Moussa et al., 2007). Six different methods to produce WG could be identified in function of the C. procera part used and extract pre-treatment. The utilization of these methods varied in the different sociocultural groups. The Peulh sociocultural group used all the identified methods, showing that they are the main actors of WG production. Conversely, Gando and Fon used five methods, and Idaatcha only one. Generally, the sociocultural groups living in the same geographical area used the same methods. They exchange knowledge through friendships, kinship, and intercultural marriages (Badoussi et al., 2014). Thus, the Gando, Fon, and Idaatcha might have learnt about WG production from the Peulh, who still keep some of the production secrets. Traditionally, the Gando are servants of the Peulh and would have learnt WG production while
working for them (Hahonou 2011). The Idatcha are the largest pop-
ulation and the most important sociocultural group in the Dassa mu-
nicipality. Their know-how in WG production might be linked to
intercultural relations with the Peuhl during their transhumance
through Dassa.

The coagulant activity of *C. procera* extracts is due to the presence of
calotropain, a cysteine protease (Baba-Moussa et al., 2007; Dossou et al.,
2006). The main plant-derived clotting enzymes with coagulant activity
on milk are cysteine proteases, serine proteases, and acid proteases.
Several plants with similar enzymes including calotropain from *Carica
papaya*, and bromelain from *Ananas comosus* are used for milk coagu-
lotion (Rayanatou 2017; Kheir et al., 2011; Roseiro et al., 2003). How-
ever, the cheese yields obtained with papain and bromelain are lower than
those obtained with *C. procera* and the cheese flavour is bitter
(Adetundji and Salawu 2008; Elolo and Courdjio 2005). Therefore,*
*C. procera* extracts are considered as the best coagulant for WG pro-
duction (Adetundji and Salawu 2008). Only *C. procera* leaves are used in
Burkina Faso (Dah et al., 2018), whereas leaves, stem, fruits, and sap are
used in Nigeria and Benin (Dossou et al., 2006; Belewu et al., 2005).
Stem and leaves are crushed, mixed with milk and then filtered. The
filtrate constitutes the coagulant (Dah et al., 2018). Our survey high-
lighted other traditional methods: the use of water for coagulant prepa-
ration (M4 and M6), and coagulant skimming without prior crushing or
filtering (M2). These methods are new and are reported for the first time.
Generally, plant-derived clotting enzymes are extracted by aqueous
maceration (Jacoby et al., 2011; Kheir et al., 2011). The plant organs
(fresh or air-dried) are crushed and soaked in water at room temperature
(28–30 °C). Spain and Portugal have the largest variety and production
of cheeses using extracts of *Cynara* sp. obtained by aqueous maceration
(Roseiro et al., 2003). But in our study, aqueous maceration is less than
maceration in milk. The M3 method for coagulant preparation
described in this study was the most used in Benin representing about
50.0% of all WG production processes. However, *C. procera* sap (M1)
displays the best milk clotting activity and thus should give the best
cheese yield (Baba-Moussa et al., 2007). Yet, cheese yields were similar
when using methods M1, M3, and M5. Additional investigations should
be done starting from a milk of standard composition because cheese
yields also depend on the milk quality (Rayanatou 2017; Jeantet et al.,
2014). Cheese yield may vary from 20.0 to 33.0% when *C. procera*
extracts are used (Dossou et al., 2006; Belewu et al., 2005). Furthermore,
coaugulant skimming instead of titration and filtering could lead to the
presence of more *C. procera* rubble in WG. Some processors argued that
the coagulant prepared according to the M5 method would allow
obtaining WG of the same colour as the milk with a good cheese yield.
Similarly, the coagulant prepared according to the M6 method should
minimize coagulation failure and help to obtain the whitest WG (Dossou
et al., 2006). The WG production methods using leaves and stems are
ancestral and allow using less *C. procera* plants that are becoming rare.
The identified production processes should be evaluated to identify the
method that best meets the consumers’ preferences, and provides the
best cheese yield with good nutritional properties.

WG preservation by boiling is the main method used in Benin. In
Burkina Faso, cold preservation also has been reported (Dah et al., 2018;
Sessou et al., 2013; Dossou et al., 2006). Sun-drying would allow longer
preservation because it considerably reduces the cheese water content,
thus limiting microbial development. Conversely, frying with oil favours
earlier rancidity (Sessou et al., 2013; Lahlali et al., 2008; Dossou et al.,
2006). Moreover, the shelf life of WG at room temperature is limited due to
the different sources of chemical and microbiological contamination as
well as the product characteristics (Aw, pH) and environmental condi-
tions that promote microbial growth.

### 4.3 Potential sources of Wagashi Gassiré contamination

Cow milk is the main raw material used in WG production (Dossou
et al., 2006). As the production is traditional, all operations are done
manually, sometimes without respecting good hygiene practices, such as
hand and material washing (Gagar et al., 2019). This could lead to a
probable microbial contamination and reducing of the shelf life of WG.
Therefore, it is necessary to increase the actors’ awareness about good
manufacturing and good hygiene practices to obtain products of good
sanitary quality for consumers.

The misuse of antibiotics by dairy farmers and the no respect of the
withdrawal period after animal treatment are the main reasons of
antibiotic residue presence in milk and dairy products (Mensah et al.,
2014a). These two behaviours were reported by the majority of the
dairy farmers surveyed. In Benin and in West Africa, several studies
showed that milk usually contains antibiotic residues, particularly
tetracyclines, beta-lactams, aminoglycosides, penicillin, sulphona-
mides, and macrolides (Madougou et al., 2019; Mensah et al. 2014b,
2019; Oluwafemi et al., 2018; Olatoye et al., 2016; Bagre et al., 2015;
Addo et al., 2011). Therefore, dairy farmers should be trained to the
use of good hygiene and husbandry practices according to the legis-
lation recommendation. In addition, the surveyed dairy farmers re-
ported using herbicides in crop fields. The most used pesticides in
northern Benin are endosulfan (a forbidden insecticide) and glypho-
sate (a herbicide) (Dognou et al., 2018; Gouda et al., 2019; Adeckian
et al., 2015; Aghohessi et al., 2011). As some pesticide residues have
been found in food consumed in Benin and West Africa, they might be
present also in WG (Douny et al., 2021; Ingenbleek et al., 2019;
Dossou et al., 2016).

The production and marketing conditions and the materials used also
are potential sources of chemical and microbial contamination. Dust
could be a source of lead contamination because it deposits on WG during
production and selling (Mama et al., 2013; Ekaneme 1998). Moreover,
some of the materials used for WG production and storage contain
aluminium that could migrate to WG (EFSA 2008).

#### 4.4. Impact of the Wagashi Gassiré production method on its quality

Cheese quality depends on the conditions of milk coagulation and
cooking, particularly the dose of coagulant used, the temperature, and
the milk quality (pH, protein, fat, and calcium content) (Jeanet et al.,
2014). In cheese factories, the milk quality is standardized through
specific operations, such as physical purification by filtration or
centrifugation, fat content standardization by adding cream or skim
milk, protein content standardization by adding casein or by ultrafil-
tration, pH adjustment by adding lactic leavening, and calcium content
adjustment by adding calcium chloride (Rayanatou 2017). These op-
erations allow the production of cheese of regular composition and
constant quality. Since the production of WG cheese is traditional, no
standardization of the milk is done. This could lead to products of
variable quality. Moreover, the milk-clotting enzymatic mechanism of
calotropin, similar to chymosin, is in two steps: the hydrolysis of the
*Phe*105-*Met*106 peptide bond in the casein micelle-protective protein
(enzymatic phase) and the release of the hydrophobic portion known as
glycomacropeptide (f106-169) conducing to casein aggregation and clot
formation (nonenzymatic phase) (Mazzorra-Manzano et al., 2018). The
enzymatic phase is temperature-dependent and affects the cheese yield
and the cheese properties (Rayanatou 2017; Jacob et al., 2011). The
coagulation of the milk must therefore be performed at the optimal
temperature of the clotting enzyme. For WG production, calotropin
activity is optimal at 70 °C (Baba-Moussa et al., 2007; Dossou et al.,
2006). This temperature was not always easy to reach by processors, as
shown by the monitoring performed (59 °C), because the production is
traditional. Plant extracts, with enough quantity of proteolytic en-
zymes, have the capacity to clot milk under optimum enzymatic activity
conditions and affect the cheese yield (Mazzorra-Manzano et al., 2018).
For instance, the peptidases from *C. procera* exhibited dose-dependent
milk-clotting activity up to 50–60 μg to 2 mL of milk and the doses
higher than 60 μg were not effective to improve substantially the
milk-clotting activity (Freitas et al., 2016). For WG processing, a
significant difference was observed in the quantity of crude extract of *C. procera* used for the processing methods M3 and M5. The similar milk-clotting activity (Mazorra-Manzano et al., 2018). According to WG producers, the milk to be used must have been milked on the day of production because a long interval between milking and cheese production is associated with natural milk fermentation that decreases WG yield. This needs to be confirmed by comparative studies.

In Burkina Faso, consumers choose WG for its nutritional value, aroma, and preservation properties. However, most of them find WG less attractive than foreign cheeses and less available (Dah et al., 2018). For Beninese consumers, whiteness is the most important criterion. However, the hygiene conditions during production as well as WG aroma and presentation (packaging) should be improved to interest consumers in urban centres accustomed to European cheese.

5. Conclusion

In Benin, there are various methods to produce WG that use different *C. procera* parts (sap, stem, leaves; alone or in combination) and different plant extract pre-treatment (trituration or not in water or milk) before milk coagulation. WG production process, storage and sale/distribution conditions, as well as husbandry practices, and seasonality of the milk used as raw material are all factors that could affect WG sanitary quality. More research is needed to assess the sanitary quality of WG obtained using these different processing methods and preservation practices.

Declarations

Author contribution statement

Dossou Wanignon Alphonse: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data. Wrote the paper.

Seko Orou Baké Marie Thérèse: Performed the experiments; Contributed reagents, materials, analysis tools or data.

Sessou Philippe, Youssou Abdou Karim Issiaka, Farougou Souaiou, Hounhouigan Djidjoho Joseph, Mahillon Jacques, Mongbo Rock, Poncelet Marc, Madode Yann Eméric, Douny Caroline, Scippo Marie-Louise, Clingurat Antoine, Azokpota Paulin: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data.

Sessou Philippe, Youssou Abdou Karim Issiaka, Farougou Souaiou, Hounhouigan Djidjoho Joseph, Mahillon Jacques, Mongbo Rock, Poncelet Marc, Madode Yann Eméric, Douny Caroline, Scippo Marie-Louise, Clingurat Antoine, Azokpota Paulin: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data.

Funding statement

This work was supported by Académie de recherche et d’enseignement supérieur [PROJET WALAC].

Data availability statement

Data will be made available on request.

Declaration of interests statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

Acknowledgements

We thank all survey respondents and the processors during the production monitoring for their full cooperation.

References

Addo, K.K., Mensah, G.I., Aning, K.G., Addo, K.K., Mensah, G.I., Aning, K.G., Nartey, N., Nipah, G.K., Benou, C., Akery, M.L., Nartey, N., 2011. Microbiological quality and antibiotic residues in informally marketed raw cow milk within the coastal savannah zone of Ghana. Trop. Med. Int. Health 16 (2), 227–232.

Aannensen, D.M., Huntley, D.M., Feil, E.J., al-Owen, F., Spratt, B.G., 2009. Epicollect: linking smart phones to web applications for epidemiology, ecology and community data collection. PLoS One 4 (9), 6968.

Adechian, S.A., Nasser-Baco, M., Akponkpe, I., Imorou-Toko, I., Egah, J., Anihouvi, V.B., 2015. Les pratiques paysannes de gestion des pesticides sur le maïs et le coton dans le bassin de l’oued d’A憬ouadouga, Vertigo 15 (2), 16-534.

Adetundji, V.O., Salawu, O.T., 2008. Western soft cheese ‘warra’ processed with Coloprotoc procera and Curica papaya: a comparative assessment of nutritional values. Afr. J. Biotechnol. 7 (18), 3360–3362.

Adechian, S.A.O., 2016. A preliminary study on the safety and quality of street-vended warankasi (a Nigerian soft white cheese) from Ibadan, Oyo state, Nigeria. Br. Food J. 119 (3), 322–330.

Agbohessi, F.T., Imorou-Toko, I., Yabi, J.A., Dassoundo-Assogba, J.F.C., Kestemont, P., 2011. Caractèrisation des pesticides chimiques utilisés en production cotonnière et impact sur les indicateurs économiques dans la Commune de Banikoara au nord du Bénin. Int. J. Biol. Chem. Sci 5 (5), 1828–1841.

Ahlade, O.A., Adeneye, J.A., 2006. The effect of storage period on the chemical composition and coliform microflora of wara cheese. Int. J. Dairy Sci 1 (2), 126–130.

Anihouvi, E.L., Anihouvi, V.B., 2019. Processing methods, preservation practices, and quality attributes of smoked and smoked-dried fishs consumed in Benin. Cogent Food Agric 5 (1), 1641255.

Baba-Moussa, F., Baba-Moussa, L., Ahissou, H., Bokossa, I., Capo-Chichi, B., Toukourou, F., Sanni, A., 2015. Le lait et produits laitiers de vache consommés dans les ménages Peul du Nord-Est du Bénin. Int. J. Biol. Chem. Sci 9 (6), 2716–2726.

Badoussi, E., Azokpota, P., Madode, Y.E., Kayode, F.A.P., Dossou, A., Soumanous, M., Hounhouigan, D.J., Anihouvi, V.B., 2014. Variations in the traditional processing methods of *Penicillus histrauce* butter in northern Benin. Food Chain 4 (3), 261–274.

Bagre, T.S., Samandououlosou, S., Traore, M., Ely, D., Thabesa, G.B., Barro-Ibara, H., Bouda, S.C., Traore, A.S., Barro, N., 2015. Détection biologique des résidus d’antibiotiques dans le lait et produits laitiers de vache consommés à Ouagadougou, Burkina Faso. J. Appl. Biosci. 87, 8105–8112.

Bagre, M.A., Belewu, K.Y., Nkwoonzou, C.C., 2005. Effect of biological and chemical preservatives on the shelf life of West African soft cheese. Afr. J. Biotechnol. 4 (10), 1076–1079.

Chabi-Toko, R., Adegbidi, A., Leballay, P., 2015. Valorisation des produits laitiers dans les ménages du Nord-Est du Bénin. Int J Biol Chem Sci 9 (6), 2716–2726.

Corniaux, C., Dautreux, G., Dieye, P.N., Poccard-Chapuis, R., 2005. Les minilaiteries comme modèle d’organisation des filières laitieres en Afrique de l’Ouest: succès et limites. Rev. Elev. Med. Vet. Pays Trop. 58 (4), 237–243.

CSS (Conseil Supérieur de la Santé), 2016. Recommandations nutritionnelles pour la belle. Avise n 9285.

Dah, P.A., Guira, F., Tankoano, A., Traore, K., Sawadogo-Lingani, H., Sawadogo, A., 2017. La fabrication du fromage peulh au Bénin. J. Appl. Sci. Technol. 29 (5), 1–12.

Dahouda, M., Boubacar, A.M.Y., Dossa, L.H., Dotse, O.I., Ahououa, S.G., Kipi, S.P., Youssou, I.A.K., 2019. Stratégies d’alimentation et gestion des ressources alimentaires dans les élevages bovins des Communes de Niki, Kalâdé et N’Dali au Nord Est Benin. Rev. Int. Sci. Appl. 2 (2), 48–70.

Dognon, S.R., Dognon, H.R., Youssou, A.K.A., Scippo, M.L., Youssou, A.K.I., 2018. The use of pesticides in agriculture in North-East Benin. Int. J. Agron. Agric. Res. 12 (6), 81–88.

Dossou, J., Ako, D., Dabade, D.S., Azokpota, P., Mombo, J., 2018. Production et transformation du lait frais en fromage peulh au Bénin. Rapport de guide de bonnes pratiques. Université d’Abomey-Calavi, Abomey-Calavi, Bénin.

Douny, C., Zoumouou, Y.M.B.G., Alna, M., Toku, I.L., Igout, A., Guedegba, L., Chabi, S.K., Kestemont, P., Scippo, M.L., 2021. Contamination of water, sediment and fish with residues of pesticides used in cotton production in Northern Benin. Arch. Environ. Contam. Toxicol. 81, 367–385.

EFSA (European Food Safety Authority), 2008. Safety of aluminium from dietary intake. EFSA J 754, 1–34.

Ekamene, O., 1998. The street food trade in Africa: safety and socio-environmental issues. Food Control 9 (14), 211–215.

Ello, G.O., Coudjio, R.L., 2005. Propriétés physico-chimiques et réactivité comparées des agents coagulants végétaux dans la fabrication du fromage frais wagashi. J. Rec. Sci. Univ. Lomé. 7 (1), 159–164.

Freitas, C.D.T., Leite, H.B., Oliveira, J.P.B., Amaral, J.L., Egito, A.S., Vairo-Cavalli, S., Lobo, M.D.P., MonteiroMoreira, A.C.O., Ramos, M.V., 2016. Insights into milk-clotting activity of latex peptidases from *Acanthopanax scleroxylon*. Food Res. Int. 87, 50–59.

Gagrag, M.H., Dossou, P., Dossa, F., Ahououa, S., Azokpota, P., Youssou, L., Goseo, A.S., Farougou, S., 2019. A study on the indigenous methods of processing milk in Niger. Curr. Agric. Res. J. 7 (2), 213–223.
