

Risks of salmonellosis linked to the consumption of lettuce grown on the outskirts and in unexploited intra-urban areas of the city of Bouaké(Central-Côte d'Ivoire)

ABSTRACT

Lettuce is one of the most popular and consumed vegetables in the world. It could be contaminated with *Salmonella* spp. which would lead to food poisoning. The objective of this study was to quantitatively assess the risk of salmonellosis linked to the consumption of lettuce produced in Bouaké (Central-Côte d'Ivoire). A survey of 138 urban lettuce producers and 384 salad lettuce consumers was first conducted. Then, microbiological analyzes were carried out on forty-five lettuce samples collected in the city of Bouaké. A probabilistic approach was followed to quantitatively assess the risks associated with the consumption of lettuce. Finally, the risk of ingestion of the infectious dose which is 10^5 cells of *Salmonella* spp. was estimated by a Monte Carlo type simulation. Microbiological analyzes showed that *Salmonella* spp. loads ranged from 0 ± 0 CFU/g to $7.73 \cdot 10^2 \pm 2.33 \cdot 10^1$ CFU/g. Furthermore, the prevalence of *Salmonella* spp. in lettuce was 77.78%. The risk of salmonellosis exists and is 6.36% to 6.56%; i.e. 6,360 to 6,560 cases per 100,000 inhabitants. This study showed that lettuces were contaminated with *Salmonella* spp. There is a risk of developing typhoid or paratyphoid salmonellosis following their consumption.

Key words: *Lactuca sativa*, *Salmonella* spp, risk assessment, Bouaké, Côte d'Ivoire.

1. INTRODUCTION

In Côte d'Ivoire, the urbanization rate was around 52% in 2017. The trend of urbanization and migration towards cities is increasing. In addition, urban population growth increases the need for food crops and especially market gardening. Fruits and vegetables are increasingly consumed by populations in developing countries. They are renowned for their organoleptic and nutritional qualities. In Ivory Coast, market gardening is characterized by an abundance of products (Bancal & Tano, 2019). Market gardening is practiced in lowlands and by vulnerable social groups. It is an agricultural activity linked to the production of vegetables in general, including lettuce (Napo, 2013). It generates income for producers. It is also a source of empowerment and improvement in the living conditions of market gardeners (Koné *et al.*, 2018; Kouakou, 2019). Furthermore, the availability of very limited reliable agricultural statistics in Côte d'Ivoire (Ducroquet *et al.*, 2017) constitutes an insufficiency for this sector

despite its strong contribution to food security. Although fruits and vegetables of urban origin are renowned for their nutritional qualities, the same is not true for their health qualities. In Bouaké, a large city in the center of the country, urban market gardening faces enormous difficulties. This concerns, among other things, the proximity of certain production sites to places where pipes loaded with domestic and industrial effluents lead (Kanda *et al.*, 2013). We also note the use of wastewater for irrigation (Dan-Badjo *et al.*, 2013). This could lead to microbial contamination of vegetables by *Salmonella* spp. with a negative impact on the health of the consumer. Many studies have mentioned the contamination of vegetables and mainly lettuce by *Salmonella* spp (Kroupitski *et al.*, 2009; Alio *et al.*, 2017). Consumption of food contaminated with *Salmonella* spp could lead to collective foodborne illness (TIAC). It very often causes typhoid fever which can even lead to death (Harizi, 2009). Would the consumption of vegetables, particularly lettuce, present a risk for the consumer? This study was carried out with the aim of providing elements of response to this concern. Its objective is to quantitatively assess the risk of salmonellosis linked to the consumption of lettuce produced in Bouaké. More specifically, it was a question of determining the lettuce consumption pattern and characterizing the associated risk.

2. MATERIALS AND METHODS

2.1. Study areas

The study area is the city of Bouaké located in the center of Côte d'Ivoire. The department of Bouaké is part of the Gkêkê region. It is limited to the North by the department of Katiola, to the South by the department of Tiébissou, to the east by that of M' Bahiakro and to the west by the departments of Béoumi, Sakassou and Botro. The department has 5 sub-prefectures (Bouaké, Brobo, Mamini, Bounda and Djébonoua) and 3 communes (Bouaké, Djébonoua, and Brobo) (Kouamé *et al.*, 2017).

2.2. Collection of data related to the production and consumption of lettuce

To conduct surveys at producer level, sheets including questionnaires were developed. They made it possible to collect information on urban lettuce production in the city of Bouaké. The investigation for the characterization and production of lettuce consisted of two essential parts. The first part provides information on the profile of producers (sex, age, nationality, level of education) and the second part concerns the characteristics of urban production (source of irrigation water, type of fertilizer and use of pesticides). In total, the survey covered one hundred and thirty-eight (138) lettuce producers. As for the diagnostic

investigation, emphasis was placed on the quantities and frequency of lettuce consumption and the symptoms encountered. A total number of three hundred and eighty-four (384) salad lettuce consumers were interviewed. The people surveyed are of both sexes, of all levels of study and from all social strata.

2.3. Sampling

Three sites located in the center of the city of Bouaké were selected. On each site, three different boards constituted a study block. When the crop was ripe, three lettuce plants (a sample) weighing 500 g were harvested and then packaged in a stomacher bag. These sachets have been carefully labeled with an individual identification number. They were subsequently transported in coolers containing cold accumulators to the Agrovalorization laboratory for microbiological analyses. In total, 45 lettuce samples, 5 samples per study site, were taken.

2.4. Testing for *Salmonella* spp

The search for *Salmonella* spp was carried out in 4 stages according to standard NF / ISO 6579: 2002 Amd 1: 2007. These are pre-enrichment, enrichment, isolation and finally reading and identification.

2.5. Assessment of microbiological risk linked to the *Salmonella* genus

The risk assessment linked to the *Salmonella* genus in lettuce (*Lactuca sativa* L.) was carried out following the method described by the Codex Alimentarius Commission (FAO/WHO, 1999). It took place in 4 stages which are: identification of the danger, characterization of the danger, assessment of exposure and characterization of the risk. The exposure assessment step provides a mathematical link (model) between the quantity of microorganisms ingested and the fraction of a consuming population that would become ill. As for that of risk characterization, it establishes the risk of the occurrence of the disease for a given population (Assidjo *et al.*, 2013; Coulibaly, 2015; Akmel *et al.*, 2017). For this study, the infectious dose was 10^5 cells (Dromigny, 2012).

2.6. Data analysis

The various data collected are processed using SPSS 22 software. For the microbiological risk assessment, the data were processed using MATLAB R 2016b software. The results of the treatments are presented in the form of graphs, pie charts and tables.

3. RESULTS

3.1. Profile of producers, inputs used and conditions linked to the consumption of lettuce produced in Bouaké

3.1.1. Profile of urban lettuce producers

The profile of urban lettuce producers is summarized in Table 1 below. Lettuce is produced by both genera in the town of Bouaké. It is dominated by the male gender with a rate of 53.62% compared to 46.38% for the female gender. The age of the producers varies between 30 and 60 years. Ivoirians are more numerous in this sector of activity with a rate of 45.65%, followed by Burkinabés and Malians with respective rates of 36.96% and 17.39%. The majority of lettuce producers surveyed in the city are illiterate with a rate of 56.52%. However, there are also primary and secondary level producers at rates of 33.33% and 10.15% respectively.

Table 1: Profile of urban lettuce producers in Bouaké

		Frequency	Percentage (%)
Age	< 30 years	21	15,21
	30-60 years	109	79
	> 60years	8	5,79
Genre	Male	74	53,62
	Female	64	46,38
Nationalité	Ivoirian	63	45,65
	Burkinabé	51	36,96
	Malian	24	17,39
Level of study	illiterate	78	56,52
	Primary	46	33,33
	Secondary	14	10,15

3.1.2. Main inputs used in lettuce production

The main inputs used in lettuce production are recorded in Table 2. 37.68% of the producers surveyed use water from unprotected wells for watering. On the other hand, 62.32% use surface water for watering lettuce. Poultry droppings and chemical fertilizers are used by respondents in 73.91% of cases as fertilizer in the city of Bouaké. Lettuce producers, 26.09%, use a mixture of poultry droppings, cow dung and chemical fertilizer to fertilize the beds. Furthermore, almost all producers use pesticides to combat lettuce pests.

Table 2: Main inputs used in urban lettuce production in Bouaké

Inputs	Fréquence	Pourcentages (%)
Water from unprotected wells	52	37,68
Surface water	86	62,32
Poultry droppings and chemical fertilizers	102	73,91
Poultry droppings, cow dung and chemical fertilizers	36	26,09
Pesticides	138	100

3.1.3. Conditions linked to the consumption of lettuce produced in the city of Bouaké

No consumers were sick during the surveys. But previous potential symptoms associated with a food infection related to lettuce consumption were reported by 74.23% of consumers. The most commonly mentioned conditions were abdominal pain, nausea, vomiting, diarrhea and high fevers. Figure 1 illustrates the conditions linked to the consumption of lettuce in Bouaké.

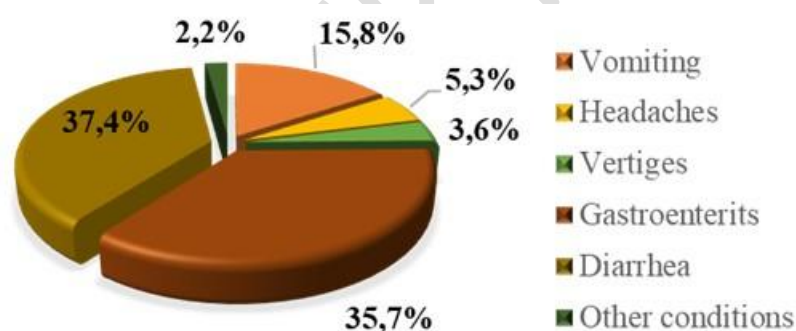


Figure 1: Conditions linked to lettuce consumption in Bouaké

3.2. Risk of salmonellosis linked to the consumption of lettuce produced in the city of Bouaké

3.2.1. Daily quantities of lettuce consumed per person and per meal

The distribution of quantities of lettuce consumed obtained from the consumer survey is presented in Figure 2. The daily quantities of lettuce vary from 91.28 ± 32.42 g to 155.38 ± 29.69 g.

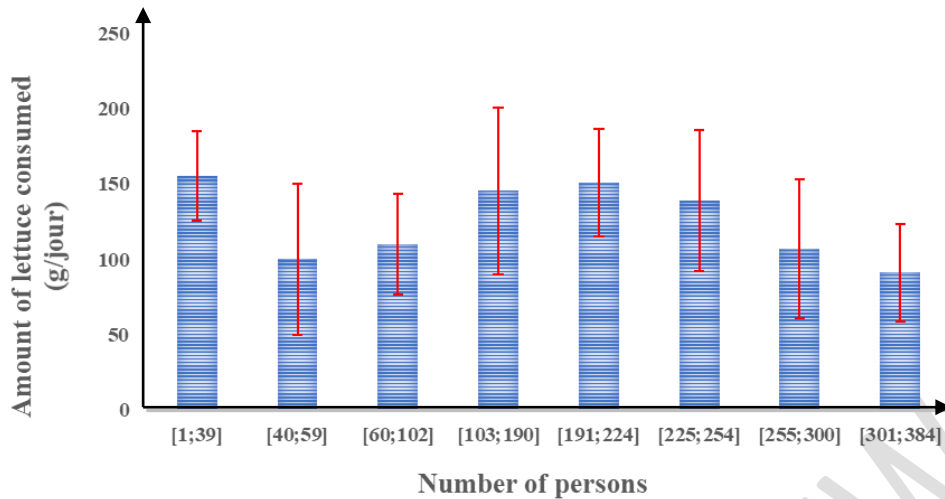


Figure 2: Daily quantities of lettuce consumed per person in Bouaké

3.2.2. Distribution of the *Salmonella* spp pathogen in lettuce

The distribution of the pathogenic germ studied in lettuce is shown in Figure 3. The analyzes show that the pathogenic germ sought was found in 35 lettuce samples from the city of Bouaké. However, microbial loads range from 0 ± 0 CFU/g to $7.73.102 \pm 2.33.101$ CFU/g. Furthermore, the prevalence of *Salmonella* spp in lettuce was 77.78%.

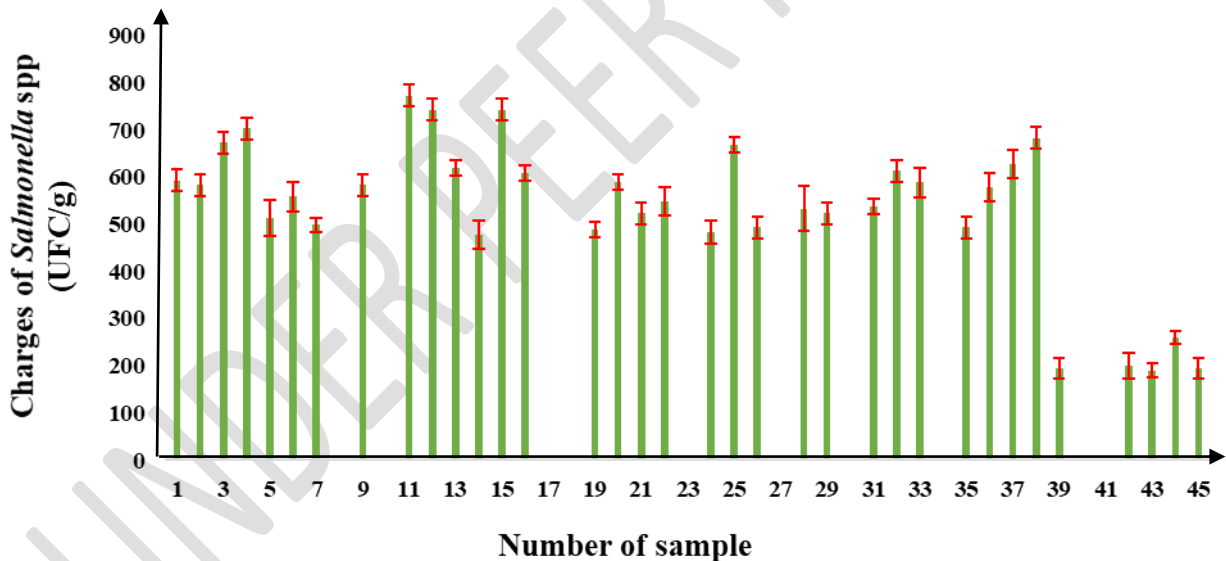


Figure 3: Distribution of *Salmonella* spp in lettuce from Bouaké

3.2.3. Risk of *Salmonella* spp in lettuce and exposure assessment

Figure 4 is the result of the simulation using the Monte Carlo method of the distribution of the microbial load (*Salmonella* spp) and that of the daily consumption of lettuce. It represents the distribution of ingestion of the contaminant (*Salmonella* spp) by consumers. It also provides information on the risk of contracting typhoid or paratyphoid fever and non-typhoid salmonellosis linked to the consumption of lettuce. As a result, the risk of salmonellosis

linked to the consumption of lettuce in the city of Bouaké is of the order of 6.36% to 6.56%; i.e. 6,360 to 6,560 cases per 100,000 inhabitants.

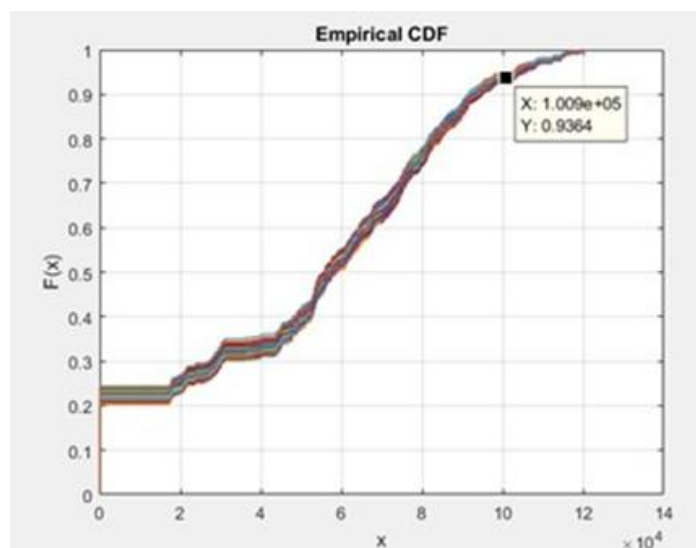


Figure 4: Risk of salmonellosis linked to the consumption of lettuce produced in Bouaké according to Monte Carlo type simulation.

4. DISCUSSION

Urban agriculture, and particularly lettuce production, is experiencing significant development in the city of Bouaké. It is an activity practiced by both genders. It is dominated by the male gender with a rate of 53.62% compared to 46.38% for the female gender. These data differ from those reported by Kouassi *et al.* (2019). Indeed, in a similar study in Daloa (Côte d'Ivoire), the authors showed that urban lettuce production is dominated by women in 58.1% of cases compared to 41.9% for men. The results obtained in this study are similar to those of Bâ *et al.* (2018), who showed in a recent study in Dakar, Senegal that market gardening activity was dominated by men in 95% of cases. Furthermore, according to Alio *et al.* (2017), urban lettuce production is exclusively reserved for men in the majority of cases (100%). The strong involvement of men in this activity could be explained by the unavailability of land. The below-average rate of women could be linked to the arduousness of the work. Added to this is the fact that women are generally specialized in the marketing of agricultural crops (N'diaye *et al.*, 2021). But also by the lack of financing for market gardening activities. The omnipresence of seniors, even retirees, seeking additional income (Bâ *et al.*, 2018) and certainly the arduousness of the work (transplanting and irrigation) mean that the age of the producers surveyed is between 30 and 60 years old. This result is inconsistent with those of N'diaye *et al.* (2021). Indeed, according to a study carried out in Casamance in Senegal, the age of urban producers is between 20 and 80 years old. Similarly, the age of producers in a study carried out on the Kollo site (Niamey) ranges from 17 to 70

years (Boukary *et al.*, 2023). Market gardening in Bouaké is mainly practiced by Ivorians in 45.65% of cases on the sites investigated. Unlike the studies carried out by Ganacadja *et al.* (2022), the activity is mainly carried out by non-nationals with a rate of 87.87%. This result comes from a study carried out on the analysis of some characteristics of the market gardening sector in three provinces of Gabon. The strong involvement of Ivorians in market gardening in Bouaké could be due to the fact that they are engaging more and more in this activity. The percentage of illiterates (56.52%) obtained from this study is higher than the 34% of illiterates reported by Sandrine *et al.* (2017). This illiteracy rate could be explained by the fact that market gardening does not necessarily require a fairly high level of education. However, the level of education impacts the mastery of Good Agricultural Practices (GAP). The producers surveyed (37.68%) use water from unprotected wells and surface water (62.32% of producers) for watering lettuce. These risky practices were reported by Kouadio *et al.* (2023) who showed in a study that urban producers use well water for irrigation. Likewise, Kouassi *et al.* (2019) showed that lettuce growers use surface water for watering. Poultry droppings and chemical fertilizers are used by respondents (73.91%) as fertilizer in the city of Bouaké. This result corroborates with those found by Kouadio *et al.* (2023), De Bon *et al.* (2019) and Yeo *et al.* (2022) in their various works. Nearly 26.09% of urban lettuce producers use a mixture of poultry droppings, cow dung and chemical fertilizer for fertilizing the beds. This goes hand in hand with the results of Boukary *et al.* (2023) stating that urban producers use organic manure combined with two types of chemical fertilizer (NPK and Urea). The use of these fertilizers proves that cultivable soils are increasingly poor in nutrients. All producers (100%) use pesticides. This result is contrary to that of Boukary *et al.* (2023) which showed that 74% of producers use chemicals. The use of pesticides could be explained by the high presence of diseases and crop pests.

Furthermore, the results showed that 74.23% of consumers developed illnesses following the consumption of lettuce. The symptoms detected are markers of typhoid or paratyphoid fever and non-typhoid salmonellosis. This could be explained by the non-rational use of poultry droppings and cow dung as fertilizer. Some producers seem to have less control over fertilizer application doses (FAO, 2021). Added to this is the irrigation of the plants with well water in watering cans. In fact, these methods lead to microbial contamination of the products. Therefore, a complex epidemiology of *Salmonella* spp. (Alio *et al.*, 2017). The results also reveal that the daily quantities of lettuce consumed by the people surveyed vary from 91.28 ± 32.42 g and 155.38 ± 29.69 g. In contrast, Yao *et al.* (2016) showed in a study that the total quantity of lettuce consumed in one day by all households surveyed in Port-Bouët is 4,180 g.

This high consumption of lettuce by populations could be explained by the fact that this vegetable is very popular. Furthermore, lettuce is an important nutritional source.

The results obtained showed that *Salmonella* spp. was found in 35 lettuce samples analyzed. Therefore, the prevalence of *Salmonella* spp in lettuce is 77.78%. Microbial loads range from 0 ± 0 CFU/g to $7.73.102 \pm 2.33.101$ CFU/g. This prevalence is opposed to that of Kouassi *et al.* (2019) found in their study on lettuce. Conversely, Traoré *et al.* (2015) found a prevalence of 50% in a study carried out in Burkina Faso. This very high prevalence rate is explained by the illiterate rate (56.52%) present on the study sites. This high presence of *Salmonella* spp. in lettuce would increase the risk of salmonellosis. It would therefore be important to characterize this risk.

The study carried out highlighted the risk of salmonellosis linked to the consumption of lettuce produced in Bouaké. This risk is of the order of 6.36% to 6.56%; i.e. 6,360 to 6,560 cases of salmonellosis per 100,000 inhabitants. The high risk of salmonellosis is surely due to the high presence of *Salmonella* spp. in lettuce. It could worsen with the presence of other germs. It is important for consumers to wash lettuce well and disinfect it with sodium hypochlorite, respecting the recommended doses.

5. CONCLUSION

This study focused on the assessment of the risks of salmonellosis linked to the consumption of lettuce produced in the town of Bouaké (Central Côte d'Ivoire). It revealed that lettuce is produced by illiterate people at a rate of 56.52% in difficult conditions. Lettuce is very popular among the population of this city with a daily consumption of 91.28 ± 32.42 g to 155.38 ± 29.69 g per capita. The microbiological analyzes carried out gave a prevalence of 77.78% of *Salmonella* spp. in the lettuce samples. Which means that these samples are not suitable for consumption. The conditions recorded during this study are characteristic of *Salmonella*. Furthermore, the risk assessment showed that the risk associated with the presence of *Salmonella* spp. for the lettuce consumer in Bouaké exists. Taking into account the infectious dose of 10^5 cells, the probability of developing salmonellosis when consuming lettuce is 6.36% to 6.56%; i.e. 6,360 to 6,560 cases per 100,000 inhabitants. This reveals a real public health problem. Given the high consumption of lettuce by populations, these results should attract the attention of the competent authorities who could organize awareness campaigns on BPA and microbiological risks of food origin.

1. REFERENCES

- Agossou J, Adédéméy DJ, Noudamandjo A, Ahohoui, Afouda L, Tovihoudji P, Tido NC, Mazou B, Fayomi B, Koumakpai, Ayivi B, Akpona S, Sylvain E. Risks of typhoid and paratyphoid fevers linked to the use of water waste in urban and peri-urban agriculture: case of market gardening in the town of Parakou (Benin). *Approx. Laug. & Santé*, 2014, 13 (5): 405-416.
- Akmel DC, Aw S, Montet D, Assidjo N, Degni ML, Akaki D, Moretti C, Elleingand E, Brabet C, Baud G, Mens F, Yao B, Michel T, Durand N, Assin H, Berthiot L, Tapé T Quantitative assessment of the microbiological risk associated with the consumption of attieke in Cote d'Ivoire. *F. Control*, 2017, 81: 65-73.
- Alio SA, Inoussa MM, Bakasso Y, Samna SO. Diversity and dynamics of *Salmonella* isolated from lettuce (*Lactuca sativa* L.) in market garden crops in Niger (West Africa). *J. of Appl. Bios.* 2017, 119: 11917-11928.
- Assidjo E, Aw S, Akmel C, Akaki D, Elleingand E, Yao B. (). Risk analysis: Innovative tool for improving food safety. Dakar: African Review of animal health and production. *African Journal of Animal Health and Production*, 2013:11(S):3-13.
- Ba A. Cantoreggi NL. Urban and peri-urban agriculture (AUP) and agri-urban household economy in Dakar (Senegal). *Int. J. Approx. Agric. Biotechnology*. 2018, 3:195–207.
- Bancal V. & Tano K. Study of Modalities for Reducing Post-Harvest Losses in Market Gardening Crops in Ivory Coast. *EXPERTISE REPORT*, 2019, 91 p.
- Boukary H, Rabe MM, Bori H., Soumaila Abdoulaye A. Yahaya BZ. Peasant Practices for the Production of Onion Bulbs (*Allium cepa* L.) in a Peri-urban Area of Niamey: Case of the Kollo Market Gardening Site. *ESI Preprints*. 2023, 10:257-273
- Coulibaly KR. Assessment of the risk of salmonellosis linked to the consumption of curdled milk (artisanal fermented milk) produced in Yamoussoukro. Doctoral thesis from Nangui Abrogoua University, Abidjan, Ivory Coast, 2015, 39 p.
- Dan-badjo TA, Guéro Y, Dan Lamso N, Baragé M, Balla A. Sterckeman T. Evaluation of metal trace contamination of lettuce and cabbage in the Gounti valley, Niamey *J. of Appl. Bios.* 2013, 67: 5326-5335.
- De Bon H., Fondio L. & Dugué P. Study of identification and analysis of constraints to market gardening production according to the major agro-climatic zones of Côte d'Ivoire. Expert report. Montpellier: CIRAD, 2019, 140 p
- Dromigny E. Microbiological criteria. Regulations Microbiological agents Self-control. 1st ed., Technique and Documentation Lavoisier, Paris, France, 2012, 231 p.

- Ducroquet H, Tillie P, Louhichi K. Gomez Y.P.S. Agriculture in Côte d'Ivoire under the microscope. State of play of plant and animal production sectors and review of agricultural policies. EUR 28754 FR, Publications Office of the European Union, Luxembourg, 2017, 137 p.
- FAO. AgrInvest Project - Food Systems, Study on the onion value chain in Niger. 2021, 50 p.
- FAO/WHO. Principles and guidelines for the conduct of microbiological risk assessment. CAC/GL-30. Rome, Italy, 1999, 6 p.
- Ganacadja C, Mavoungou JF, Mouketou A, Biroungou C. Nzengue E. Analysis of Some Characteristics of the Market Gardening Sector in Three Provinces of Gabon. ESI Preprints. 2022, 55 p.
- Harizi K.. Research and identification of pathogenic bacteria *Salmonella* and *Listeria* in foods. Master's thesis in quality control and safety of animal and plant products, University of Gabés, Gabés, Tunisia, 2009, 60 p.
- Kanda M., Djaneye-Boundjou G, Wala K, Gnandi K, Batawila K, Sanni A. Akpagana K. Assessment of pesticide residues and trace element contamination in market gardens of Togo. African J. Approx. Sci. and Tech., 2013, 6:380-390.
- Koné M, Anoh ML. Beda EEL. Study on promising sectors and value chains in the regions of Gbêkê, Poro, Tonkpi and the district of Abidjan, Ivory Coast, Final report, 2018, 137 p.
- Kouakou PAK. Economic Determinants and Social Impact of the market gardening sector in the Municipality of Boundali. Ag. Af. 2019, 103–113.
- Kouamé AR, Vei KN. Yao NF. Peri-urban market gardening production in Bouaké: dynamism of actors and functions. Revue Ivoirienne de Géographie des Savanes, 2017, I: 66-77.
- Kouassi KC, Kouassi KA, Yao KM., Kouassi AG, Koffi NR. Assessment of the Risk of Microbial contamination of an Urban Crop in the City of Daloa (Côte d'Ivoire): Case of Lettuce (*Lactuca sativa* L.). J. of F. Res. 2019, 8: 122-132.
- Kroupitski Y, Golberg D, Belausov E, Pinto R, Swartzberg D, David GD. SInternalization of *Salmonellaenterica* in leaves is induced by light and involves chemotaxis and penetration through open stomata. Appl. Approx. Microbiol. 2009, 75:6076–6086.
- Lalatiana O RR. Contribution to the study of the microbiological quality of a street food in the town of Talatan'ny Volonondry (Madagascar): case of Koba Ravina. Thesis from the Faculty of Medicine, Pharmacy and Odonto-Stomatology, Cheikh Anta Dilop University, Dakar, Senegal, 2006.100 p.

- NAPO H. Diagnostic study of onion (*Allium cepa* L.) production techniques in the Yatenga province. Master thesis, agricultural extension option, Polytechnic University of Bobo-Dioulasso, Burkina Faso, 2013, 83 p.
- Ndiaye O, Diatta U, Abeudje A, Dramé M, Ndiaye S, Tidiane Ba C. Characterization of market gardening perimeters established by women's groups as a resilience strategy in post-conflict zones (Casamance, Senegal). *Euro. Sci. J.* 2021; 17(13): 118-135
- Sandrine MBL, Auguste NN, Frédéric FC. Peri-urban market gardening in Libreville and Owendo (Gabon): cultural practices and sustainability *Cah. Agric.* 2017, 26, 45002 © DOI: 10.1051/cagri/2017026.
- Kouadio RO, Kouakou RN, Florence F Dimi TD. Intra-urban market gardening is challenged by the growing demand of city dwellers for vegetables: misuse of pesticides and health risks in Bouaké (Côte d'Ivoire) *Approx. R. & Santé*, 2023, 22 (3): 230-242
- Traoré O, Nyholm O, Siitonen A, Bonkougou OJI, Traoré SA, Barro N, Haukka K. Prevalence and diversity of *Salmonella enterica* in water, fish and lettuce in Ouagadougou, Burkina Faso *BMC Microbiol.* 2015 ; 15:151.
- Yao BL, Kpan KGK, Messoum FG, Dembélé A, Traoré KS. Assessment of the phytosanitary risk linked to the consumption of lettuce (*Lactuca sativa* L.) grown in the commune of Port-Bouët (Abidjan). *Rev. Mar. Sci. Agron. Vet.* 2016; 4 (3): 23-30.
- Yeo KT, Fondio L, Kouakou KL. Characterization and diversity of market gardening production systems in the center (Bouaké) of Ivory Coast with a view to an agroecological transition. *J Anim Plant Sci* 2022; 52:9538–9551.