

Stabilization and Sanitation of Chicken Litter by Heap Composting

R. Aboutayeb^{1,*}, M. Elgharous², Z. Abail², M. Elhari³, Y. Koulali¹

¹Department of biology, health and Environment, Faculty of Sciences and Techniques, University Hassan I, Settat, Morocco.

²Regional Center of Agricultural Research, Settat, Morocco

³Departement of Bacteriology, Regional Laboratory of Analysis and Research, Marrakech, Morocco

Abstract

The Region Chaouia-ouardigha produces approximately 200.000 tons of chicken litter each year. These wastes present environmental and health risks because of their high content of nitrogen and high density of pathogens.

The objective of this study is to evaluate the effect of composting to stabilise and sanitise the chicken litter.

After five months of composting, both physicochemical and microbiological parameters showed a significant change (at significance level $p < 0.05$) of final compost properties: a self heating of the heap was sufficient to destroy or reduce density of 7 types of pathogens: total aerobic mesophilic Flora, fecal coliforms, Enterobacteriaceae, Escherichia coli), reducing-sulfite Anaerobic bacteria, Staphylococcus aureus, and yeasts and molds. As for Salmonella, it was absent in all the samples analyzed. Also, the final compost recorded a Slight acidification to reach neutral value; a decrease in content of nitrogen, carbon and organic matter; an Increase of electrical conductivity and nitrates content.

Keywords: poultry manure, composting, assessment, pathogen, hygienization.

1. Introduction

The region Chaouia-Ouardigha is located North West of Morocco in an area of 16510 km². The region is the first producer of poultry in Morocco. Censuses for 2011 indicate the presence of 777 units producing broilers in area with a production capacity of 9,7 millions broilers per breeding cycle [1].

Approximately, 200.000 tons of chicken litter was produced in 2011 in Chaouia-Ouardigha. This waste can generate environmental negative impacts, including

bad odours, pathogens dissemination, nitrate leachate and groundwater-pollutant contamination [2,3].

These environmental problems, due to the spreading of poultry manure, are mitigated by composting [4]. This technique is increasingly considered a good way for recycling the manure as a stabilized and sanitized fertilizer for agriculture [5]. Thus, it permits to avoid a negative impact on the environment caused by these wastes.

The intensity and concentrated activity of the poultry livestock generate vast amounts of biodegradable wastes, which must be managed under appropriate disposal practices to avoid a negative impact on the environment such as odour, gaseous emissions, soils and water pollution [5,6].

Composting in heap gives several advantages: the compost is produced at a low cost, brings positive effects on the physicochemical and biological properties of soil application [7]. Therefore, it is important to assess the composting process using physicochemical and biological parameters.

The aim of this study is to assess the effect of composting in heap on the chemical and microbiological properties of chicken manure.

2. Materials and Methods

2.1. Collecting manure

The chicken litter used in the present study was collected from two poultry farms located in Settat (a province of Chaouia-Ouardigha region). The litter was transported to the Center of Agricultural Qualification Ouled Moumen (CAQ), the site of the experiment, where mixed homogeneously. We have taken five composite samples: three samples cooled and kept for microbiological analysis and two samples air-dried for physicochemical analysis.

2.2. Operation of Composting

The litter was composted aerobically in a heap for 5 months. During composting process, moisture content was adjusted to around 50%. The heap has been manually turned once a week during bio-oxidative phase and two to three times per month through the maturation phase. Turning schedule permits rapid decomposition at thermophilic temperatures [8]. Heap temperatures were monitored using alcohol thermometer at 3 locations: the top, the middle and the bottom. The ambient temperature was collected by the weather station to the CAQ. At the end of composting, 8 samples were collected in sterile plastic-bags to serve for microbiological and chemical analysis.

2.3. Physicochemical analysis:

All the samples were analyzed for the following parameters according to the manual of analysis methods [9]. Dry matter content was assessed by drying at 70°C for 48 hours. pH and electrical conductivity (1:10 w/v Sample-water extract) were measured using a pH meter electrode and a conductivimeter respectively. Organic carbon (OC) was determined by titration using potassium dichromate. Organic matter (OM) was calculated according to the equation ($OM = 1,724 OC$). The total nitrogen determined by Kjeldahl method. Nitrates are determined by complexation with chromotropic acid and measuring the absorbance in a spectrophotometer at 410 nm [10]. Ammonium was determined colorimetrically at 636 nm. Phosphorus was determined by colorimetry at 882 nm [11] and potassium by extraction with ammonium acetate and determination using a flame photometer.

2.4. Microbiological Analysis

Microbiological analyses have focused on enumeration of 8 groups of microorganisms by determination of the number of colony forming units (CFU/g). The enumeration of total aerobic mesophilic flora was determined using the Petri dishes containing a medium of agar glucose and yeast extract; inoculated and incubated at 30 °C for 72 hours [12]. Enterobacteriaceae was inoculated in VRBG medium (Violet Red Bile Glucose) at 37 °C for 24h [13]. Fecal coliform was inoculated in VRBL agar (Violet Red Bile Lactose) at 44 °C for 24 hours [14]. Escherichia coli at 44 °C for 48 hours according to Mackenzie test [15]. The population of Staphylococcus aureus was determined in the Baird-Parker medium at 37 °C for 48 hours [16]. Sulfite-reducing anaerobic bacteria was inoculated on TSN agar (tryptone, sulfite, neomycin) at 46 °C for 24 hours [17]. Yeasts and molds inoculated on Sabouraud agar at 25 °C for 3 days [18]. Salmonella was determined

in 25g sample on SS agar (salmonella-shigella) at 37°C for 48 hours [19].

2.5. Statistical Analysis

The results were statistically analyzed using SPSS statistics 17.0 software for analysis of variance (ANOVA) at a significance level of 5% ($P < 0.05$). The analyses were performed in two repetitions.

3. Results and discussion

3.1. Temperature Monitoring:

Temperature has been widely recognized as one of the most important parameters in the composting process [4]. Figure 1 shows the evolution of the ambient temperature and that of compost throughout composting. The peak temperature that occurred during thermophilic phase in the middle (63 °C) part were higher than those recorded in the bottom (60 °C) and surface (53°C) locations of the heap (Figure 1).

The rise in temperature indicates an intense microbial activity; it should reach 55°C to destroy pathogenic microorganisms [20]. The optimum temperature range for composting is between 40 and 65 °C [21].

The development of the temperature profile indicates the different phases of the process. In general, the composting process can be divided into two main phases: the biooxidative phase and the maturing phase also called the curing phase [20,22]. A graph shows two distinct phases: a bio-oxidative phase divided into three sub phases (mesophilic, thermophilic and cooling); and maturation phase.

During the maturation phase, temperatures at different locations of the heap reach ambient level almost at the same time despite their differences in thermophilic temperatures (Figure 1), indicating that the chicken litter is becoming stable.

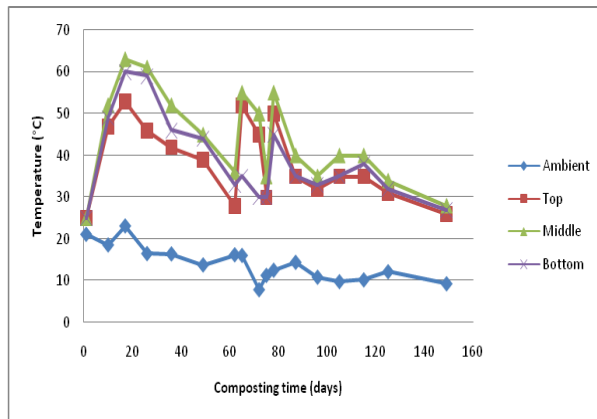


Figure 1. Air and heap temperature changes during composting of chicken litter.

3.2. Physicochemical analysis:

Table 1 shows the average values obtained in the analysis of samples taken before and at the end of composting. During the process, the heap recorded a slight acidification (The pH decrease from 7,73 to 7,29). This decrease can be explained by the production of organic acids. All composts showed a neutral or slightly alkaline pH [23]. Neutral pH is an indicator of stabilized Organic matter [4]

Table 1. Chemical properties at the beginning and the end of composting (Dry matter)

	Initial	Final
	Mean \pm SD	Mean \pm SD
pH	7,73 \pm 0,11	7,29 \pm 0,13
EC (dS/m)	5.00 \pm 0,00	6.60 \pm 0,55
OM(%)	57.60 \pm 5,00	40.38 \pm 4.28
NH ₄ ⁺ /NO ₃ ⁻	7.24 \pm 0,90	0.59 \pm 0,09
P (g/kg)	5,98 \pm 0,85	4,51 \pm 0,57
K (g/kg)	1,46 \pm 0,07	1,10 \pm 0,29

SD : Standard deviation; EC : Electrical conductivity; OM : Organic matter; NO₃⁻: Nitrates; NH₄⁺ : Ammonium; P : phosphorus; K : potassium.

Electrical conductivity (EC) reflects the degree of salinity in the heap before and after composting. The EC values were relatively high (from 5.0 to 6.6 dS/m). This change may be explained by the ions concentrations, due to weight loss. Most of compost showed an increase of EC during composting [24].

Moisture content decreased during composting to 26.9% in the final compost. This loss was explained by the evaporation of large quantity of water occurred due to the temperature rise during the active phase and frequency of turning. However, the reduction in moisture content was limited by the composting period characterized by

high relative humidity (81% on average) and low ambient temperature (14°C on average).

During composting, the content of organic matter decreased significantly (from 57.6% to 40.3%dm). This change due to the mineralization of OM was similar to the results reported by [4,23]. The degradation rate of the OM decreases gradually as composting progresses because of the reduction in available carbon sources and other nutrients.

The total nitrogen (Figure 2) content recorded a significant decrease which can be explained by volatilization of nitrogen as ammonia [25-28], turning operations [29], leaching or consumption by flora Microbial. These results are consistent with those found in [24] where each chicken can produce 0.15 to 0.20 kg of waste per day, containing between 1-6% (dm). These high levels of nitrogen limit the spreading of poultry manure.

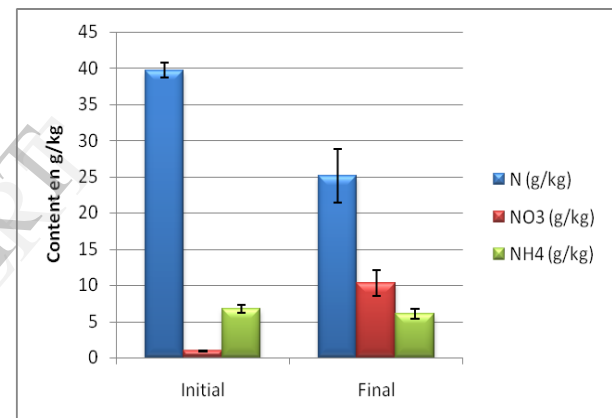


Figure 2: Content of Total nitrogen (N), nitrates (NO₃) and ammonium (NH₄) at the beginning and the end of composting

The C/N ratio is particularly important; a report of 20 to 40 would be acceptable with an optimum between 25 and 30 to start composting. The C/N ratio decreased fewer than 10 (C/N =9.5) in the final compost. This could be due to a high consumption of carbon content used as energy source of microbial flora. According to [5], a C/N ratio less than 12 is an indicator of compost maturity.

At the beginning of composting, manure had a nitrate content of 0.94 g / kg (Figure 2). At the end of the operation, this content has reached a level of 10.36 g/kg. This significant increase is due to the action of nitrification led by nitrifying bacteria. The final compost showed 0,6%(dm) of NH₄⁺ content. According to [30], recommended NH₄⁺ content should not exceed 0.4%. However, some composts show more maturity in despite of considerable levels of NH₄⁺ [8].

Thus, The $\text{NH}_4^+/\text{NO}_3^-$ ratio recorded a significant drop during composting process from 7.24 to 0.59 (Table 1). The latter value (less than 3.0) is an indicator of compost maturation according to the California Compost Quality Council [31].

Phosphorus levels do not show significant difference. This stabilization may be due, according to [8], to a consumer by microorganisms, which offsets the resulting concentration of the reduced weight of the pile. The total potassium content decreased during composting, from 1.47 to 1.1 g / kg. Nevertheless, the loss of potassium was recorded because of consumption by the microbial flora.

3.3. Microbiological analysis:

Microbiological hazards due to the presence of high pathogens densities are one of the problems posed by the direct use of poultry manure in agriculture. These health risks could be minimized by composting. Reducing the survival of these pathogens especially during the thermophilic phase is one of the main roles of composting for decreasing the risk of contamination.

Initially, the heap showed a high density of microorganisms. These analyses involved 8 microflora. The total aerobic mesophilic flora (Figure 3) was significantly reduced by 1.7 Log units. This decrease was probably due to the high temperature and unfavorable conditions established during the thermophilic phase [32].

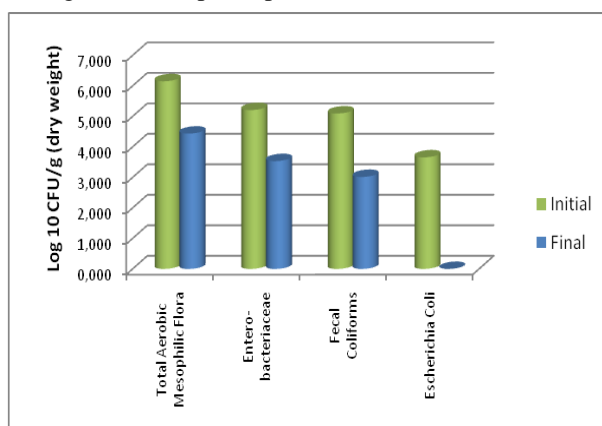


Figure 3. Populations of total aerobic mesophilic flora, enterobacteriaceae, fecal coliforms and Escherichia coli at the beginning and the end of composting

The reducing in concentration of microorganisms used as an indicator of fecal contamination was also significant (figure 3): Populations of Enterobacteriaceae, Fecal coliforms (FC) and Escherichia coli were significantly Log reduced by 1.67, 2.08 and 3.65 respectively in the final compost.

Thermotolerant coliforms concentration is used as environmental sanitation indicator [33,34].

In our experiment, the concentration of thermotolerants (FC) is below the recommended limit (10^3 CFU/g) indicating the composting efficiency [3]. The decrease was presumably the result of the high temperature and unfavorable conditions established during the thermophilic phase [32].

The total destruction of Escherichia-coli population was certainly due to the rise of temperature and aerobic conditions [35].

Furthermore, the population of staphylococcus exceeded 10^6 CFU/g. It was significantly reduced by 1.93 Log during composting (figure 4). The remaining density might be due to a quality of the pathogen as ubiquitous bacteria. Despite successful composting, the health risk relative to a potential pathogen growth is still present [36,37]. This risk is greater in the peripheral parts [38].

The human pathogen Staphylococcus aureus is transmitted with food consumption and its presence in compost used for vegetable production can cause serious epidemiological problems [39-41].

Further, population of sulfite-reducing anaerobic bacteria was reduced by 1.61 log; it may be due to high temperature in thermophilic phase, the aerobic conditions and the presence of nitrates (figure 4).

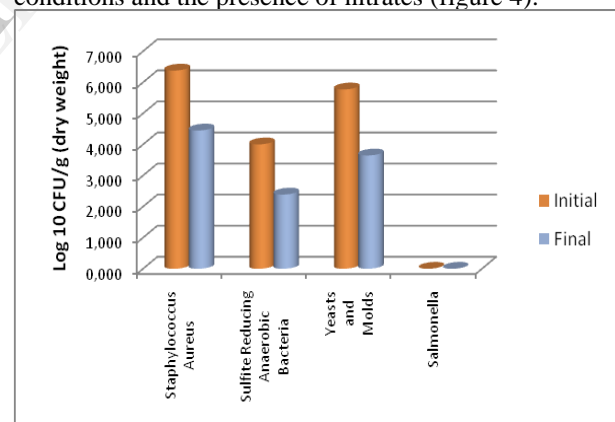


Figure 4. Populations of Staphylococcus aureus, sulfite-reducing anaerobic bacteria, salmonella, and yeasts and molds at the beginning and the end of composting

Salmonella was not detected in all samples before and after composting (figure 4). This can be explained by compliance with regulatory requirements for poultry farming and the absence of post-contamination factors that may contaminate the pile during composting.

Finally, Yeasts and molds were log reduced by about 2.12 units (figure 4). The mushrooms are mostly mesophilic. Therefore the temperature rise over 50°C had certainly a lethal effect. The rest of the fungal population is concentrated mainly on the periphery of the pile where the temperature is lower [42]. Another explanation which could

justify the presence of fungal microflora is the presence of favorable conditions in maturation phase: A low water activity and the prevalence of complex substrates (lignin and cellulose). These conditions can promote the growth of fungi and actinomycetes [4,21].

Our results indicate that the composting process in the heap was capable to ensure the organic matter stabilization and destroying populations of several pathogens.

4. Conclusion

The environmental problems associated with chicken litter application could be mitigated by stabilizing its nutrient and organic matter (OM) contents by composting before application to agricultural soils.

As a consequence, the assessment of both chemical and microbiological parameters has been demonstrated that heap composting of 5 months can be sufficient to produce stabilized and sanitized compost from chicken litter. At the high temperatures reached during thermophilic phase of composting, pathogens are reduced or totally destroyed making the chicken litter compost safer for agricultural use. The maturation phase was accompanied by a decline of compost temperatures to ambient level, relative increases in nitrates and electrical conductivity and decreases in Carbon, Organic matter, pH and nitrogen.

It can be concluded that chicken litter composting in heap is a suitable way to stabilize organic matter, produce heat and thus leading to hygienization. This process may be considered as an effective and efficient way to produce stabilized and sanitized compost.

Acknowledgement

The authors thank the staff and students of the center for Agricultural Qualification Ouled Moumen, the staff of the Regional Center for Agricultural Research (Settat) and of the regional laboratory of analysis and research (Marrakech), Mr. J. Kogboma and Miss S. Rida for their contribution in testing and implementation of laboratory analysis.

References

[1] R. Aboutayeb, Y. Koulali, A. Madar, A. Sbia, "Cartographie et analyse de la distribution des élevages avicoles en utilisant le système d'information géographique cas de la région Chaouia Ouardigha au Maroc" *ScienceLib Editions Mersenne*, France. Volume 5, N° 130518, 2013.

[2] H. Heinonen-Tanski, M. Mohaibes, P. Karinen, J. Koivunen, "Methods to reduce pathogen microorganisms in manure," *Livestock Sci.* 102 pp 248–255, 2006.

[3] M. Ros, C. Garcia, T. Hernandez, A full-scale study of treatment of pig slurry by composting: kinetic changes in chemical and microbiological properties, *Waste Manage.* 26 (2006) 1108–1118

[4] S. Tiquia, N. Tam, "Characterization and composting of poultry litter in forced aeration piles." *Process Biochem.* 37, 869–880, 2002.

[5] M.P. Bernal, J.A. Alburquerque, R. Moral, "Composting of animal manures and chemical criteria for compost maturity assessment. A review" *Bioresource Technology* 100 pp 5444–5453, 2009.

[6] Burton, H., Turner, C., 2003. *Manure management, second ed.* Treatment Strategies for Sustainable Agriculture Silsoe Research Institute, Lister and Durling Printers, Flitwick, Bedford, UK.

[7] V. Cala, A. Cases, and I. Walter, "Biomass production and heavy metal content of *Rosmarinus officinalis* grown on organic waste-amended soil." *J. Arid Environ.* 62 (3) pp. 401–412, 2005.

[8] S. Kalamdhad Ajay, A.A. Kazmi, "Effects of turning frequency on compost stability and some chemical characteristics in a rotary drum composter". *Chemosphere* 74, pp.1327–1334, 2009.

[9] M. Elgharous, M. Elamrani, K. Elmjahed, INRA, Institut National de la recherche agronomique, *manuel des méthodes d'analyses de sol et de plantes*. 1ère édition, CRRA de Settat, Maroc, 1995.

[10] D.G. Hadjidemetriou, Comparative study of the determination of nitrates in calcareous soils by the ion-selective electrode, chromotropic acid and phenodisulphonic acid methods. *Analyst* 107, pp 25-29. 1982.

[11] S.R. Olsen, C.V. Cole, W.S. Watanabe, L.A. Dean, Estimation of available phosphorus in soil by extraction with sodium bicarbonate. *USDA Circular* 939, U.S. Government Printing Office, Washington, DC. 1954.

[12] AFNOR (Association Française de Normalisation), Norme française NF V 08-051, dénombrement des microorganismes par comptage des colonies obtenues à 30°C. AFNOR, Paris, 1999.

[13] AFNOR (Association Française de Normalisation), Norme française NF V 08-054, dénombrement des entérobactéries par comptage des colonies obtenues à 30°C, AFNOR, Paris, 1999.

[14] AFNOR (Association Française de Normalisation), NF V 08-060, Norme française, dénombrement des coliformes thermotolérants par comptage des colonies obtenues à 44°C, AFNOR, Paris, 1996.

[15] AFNOR (Association Française de Normalisation), NF V 08-053, Norme française, dénombrement des *Escherichia coli* par comptage des colonies à 44°C, AFNOR, Paris, 1993.

[16] AFNOR (Association Française de Normalisation), Norme française NF V 08-057-2, méthode de routine pour le dénombrement des staphylocoques à coagulase positive par comptage des colonies obtenues à 37°C, AFNOR, Paris, 1994.

[17] AFNOR (Association Française de Normalisation), Normalisation française XP V 08-061, dénombrement en anaérobiose des bactéries sulfitoréductrices par comptage des colonies, AFNOR, Paris, 1996.

[18] AFNOR (Association Française de Normalisation), Norme française XP V 08-059, dénombrement des levures et moisissures par comptage des colonies

- obtenues à 25°C, méthode de routine. AFNOR, Paris, 1995.
- [19] AFNOR (Association Française de Normalisation), Norme française NF V 08-052, , recherche des salmonella, méthode de routine. AFNOR, Paris, 1997.
- [20] M.P. Bernal, A.F. Navarro, A. Roig, J. Cegarra, D. García, 1996. Carbon and nitrogen transformation during composting of sweet sorghum bagasse. *Biol. Fert. Soils* 22, 141–148.
- [21] M. De Bertoldi, G. Vallini and A. Pera, “The biology of composting: a review.” *Waste Manage. Res.* 1, pp. 157–176, 1983.
- [22] Y. Chen, Y. Inbar. Chemical and spectroscopical analyses of organic matter transformation during composting in relation to compost maturity. In: Hoitink, H.A.J., Keener, H.M. (Eds.), *Science and Engineering of Composting: Design, Environmental, Microbiological and Utilization Aspects. Renaissance Publications*, Ohio, pp. 551–600, 1993.
- [23] K. Lasaridi, I. Protopapa, M. Kotsou, G. Pilidis, T. Manios, A. Kyriacou, “Quality assessment of composts in the Greek market: The need for standards and quality assurance.” *Journal of Environmental Management* 80 pp. 58–65, 2006.
- [24] F. Sellami, R. Jarboui, S. Hachicha, K. Medhioub, E. Ammar, “Co-composting of oil exhausted olive-cake, poultry manure and industrial residues of agro-food activity for soil amendment” *Bioresource Technology* 99 pp. 1177–1188, 2008.
- [25] B. Eghball, J.F. Power, J.E. Gilley, J.W. Doran, Nutrient, carbon, and mass loss of beef cattle feedlot manure during composting. *J. Environ. Qual.* 26, pp 189–193, 1997.
- [26] O. Martins, T. Dewes, Loss of nitrogenous compounds during composting of animal wastes. *Bioresour. Technol.* 42, pp 103–111, 1992.
- [27] J.M. Paillat, P. Robin, M. Hassouna, P. Leterme, Predicting ammonia and carbon dioxide emissions from carbon and nitrogen biodegradability during animal waste composting. *Atmos. Environ.* 39, pp 6833–6842, 2005.
- [28] R. Parkinson, P. Gibbs, S. Burchett, T. Misselbrook, . Effect of turning regime and seasonal weather conditions on nitrogen and phosphorus losses during aerobic composting of cattle manure. *Bioresour. Technol.* 91, pp 171–178, 2004.
- [29] M.L. Solano, F. Iriarte, P. Ciria, M.J. Negro, “Performance characteristics of three aeration systems in the composting of sheep manure and straw.” *J. Agric. Eng. Res.* 79, pp. 317–329, 2001.
- [30] F. Zucconi, M. de Bertoldi, “Compost specifications for the production and characterization of compost from municipal solid waste.” In: de Bertoldi, M., Ferranti, M.P., L’Hermite, P., Zucconi, F. (Eds.), *Compost: Production, Quality and Use. Elsevier*, Barking, pp. 30–50. 1987.
- [31] TMECC, Test Methods for the Examination of Composting and Compost. US Composting Council, Bethesda, MD. 2002.
- [32] A. Hassen, K. Belguith, N. Jedidi, A. Cherif, M. Cherif and A. Boudabous, “Microbial characterization during composting of municipal solid waste” *Bioresource Technol.* 80, pp. 217–225. 2001.
- [33] Lemunier, C. Francou, S. Rousseaux, S. Houot, P. Dantigny, P. Piveteau, J. Guzzo, Long-term survival of pathogenic and sanitation indicator bacteria in experimental biowaste composts, *Appl. Environ. Microbiol.* 71 (2005) 5779–5786.
- [34] S. Hachicha, F. Sellami, J. Cegarra, R. Hachicha, N. Drira, K. Medhioub, E. Ammar, “Biological activity during co-composting of sludge issued from the OMW evaporation ponds with poultry manure: Physico-chemical characterization of the processed organic matter” *Journal of Hazardous Materials*, 162 pp. 402–409, 2009.
- [35] A.V. Semenov, L.V. Overbeek, A.J. Termoshuizen, A.H.C. Van Bruggen, “Influence of aerobic and anaerobic conditions on survival of *Escherichia coli* O157:H7 and salmonella enteric serovar typhimurium in luria-Bertani broth, farm-yard manure and slurry.” *Journal of environmental management* 92, pp. 780-787, 2011.
- [36] R. Albrecht, Co-compostage de boues de station d’épuration et de déchets verts : Nouvelle méthodologie du suivi des transformations de la matière organique, Thèse de doctorat, université Paul Cezanne Aix Marseille III, France, 2007.
- [37] J. Sidhu, R. A. Gibbs, G. E. Ho, I. Unkovich, “The role of indigenous microorganisms in suppression of *Salmonella* regrowth in composted biosolids”. *Water Research* 35, pp. 913-920, 2001.
- [38] J. T. Pereiraneto, E. I. Stentiford, D. V. Smith, “Survival of Fecal Indicator Microorganisms in Refuse-Sludge Composting Using the Aerated Static Pile System.” *Waste Management & Research* 4, pp. 397-406, 1986.
- [39] L.R. Beuchat, . Surface Decontamination of Fruits and Vegetables Eaten Raw: A Review. Food Safety Unit, WHO/FSF/FOS/98.2, World Health Organization, 1998.
- [40] H. Michino, K. Otsuki, 2000. Risk factors in causing outbreaks of food-borne illness originating in school lunch facilities in Japan. *Journal of Veterinary Medical Science* 62 (5), pp 557–560.
- [41] K. Nguz, J. Shindano, S. Samapundo, A. Huyghebaert, , 2005. Microbiological evaluation of fresh-cut organic vegetables produced in Zambia. *Food Control* 16, pp 623–628.
- [42] A. Jouraiphy, “Compostage des boues actives-déchets verts : analyses physico chimiques, devenir des micropolluants, bilan humique et valorisation agronomique du compost.” Thèse de doctorat, Université Cadi Ayyad, faculté des sciences Semlalia, Marrakech, Maroc. 2007.