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# **Integrated humification of poultry waste**

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#### Abstract

In this study effects of adding chemical and microbial agents in the humification process of poultry wastes were carried out. Chemicals and microbes play a vital role to accelerate the humification process. *Salmonella* and lime were applied individually to observe the variation in maturity and stability parameters like degree of polymerization (DP), C/N, humification rate (HR), humification index (HI), cation exchange capacity (CEC), and nitrate nitrogen concentration of the humified poultry wastes. Thus it was acclaimed that the microorganisms *Salmonella* encourage the compost formation where as humification with lime proceeded relatively with a lesser time than *Salmonella*. Furthermore, the decline in carbon and nitrogen ratio and increase in CEC, DP, HR, HI, potassium and phosphorous were recognized in a higher scale in chemically treated wastes compared to that of microbial one. It was concluded that use of chemicals reduce the quantum of waste in minimum time.

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## Introduction

The soil physicochemical properties can be changed by induction of soil microorganisms and micronutrients and can increase the decomposition rate of poultry wastes (Gul et al., 2015). Under such condition acidification and ammonium volatilization process in soil increase. Various chemicals like alum, NaHSO<sub>4</sub>,H<sub>2</sub>SO<sub>4</sub>and aluminum phosphate decrease the water soluble phosphorus and surface adsorption complexes, but increased the carbon and nitrogen mineralization rate (Hunger et al., 2004). Lime is also used to raise the pH of immature compost because many acids are produced during composting process which later on stabilize. The production of acids play an important role in killing the dangerous pathogens and lime adjust the pH of compost up to 6.5 thereby enabling a condition ideal for plant growth (Iqbal et al., 2015).

The *Salmonella* was isolated from poultry waste, an eminent pathogen indicator used in composting. The *Salmonella* spp. is very useful in stabilization of compost material. It is related to human pathogen and very important in order to evaluate

the quality of stable compost to limit the health risk (Briancesco, 2008). In composting, inactivation of pathogen during the composting period can be affected by a number of mechanisms (Wilkinson, 2008). Temperature of compost is an important factor beause it effects the activity of microorganisms such as microbial metabolic rate and population structure (Iqbal *et al.*, 2010). In composting process the microbial activity begins at ambient temperature which later on elevate the temperature of compost material in thermophilic phase. In it many non-thermos tolerant organisms are activated and it gradually decrease the microbial activity, turns cooling and results maturation of the composting mass (Steger *et al.*, 2007).

In the process of decomposition, cation exchange capacity (CEC), humification rate (HR), degree of polymerization (DP), humification index (HI), concentration of fulvic- (FA) and humic acid (HA) increases due to conversion of lingo cellulose into humus by microbes (Ming *et al.*, 2015; Taiwo *et al.*, 2016).

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When it occurs, the ratio of carbon to nitrogen gets below 20 and this is considered as indicator of compost maturity. Through microbial activity metabolism of protein increases NH<sub>2</sub>, volatilization of which may cause the decrease in NH<sup>+4</sup>-N due to the insignificant alkaline pH of the compost. The large microbial population could increase degradation of biochar and diversity of microbes in the composting heaps. Fungi partially degrade biochar during the process of composting. Losses of nitrogen may occur during the composting of poultry manure due to the initial elevation of NH, and the incidence of easily mineral able compounds of N, like uric acid can be emitted during the early phases of composting. Decrease of NH, volatilization is notified due to low concentration of N in soil amended with biochar and high carbon and nitrogen ratio. This favors the microbial immobilization of soils containing comparatively small amounts of inorganic N (Lehmam and Rondon, 2005). The aim of this study was to compare the humification of poultry wastes by applying chemicals and microbes and to evaluate the decomposition time also.

## Materials and methods

## Isolation of bacteria from Poultry wastes

Freshly collected samples of poultry waste were streaked on potato dextrose agar (PDA) plates. These plates were incubated for 48 - 72 h at 38-42°C without aeration. Colonies of different bacterial strains were recognized on these plates. Different species of *Salmonella* were separated from this culture of bacteria on agar plate.

## Isolation of Salmonella

Colonies typical (pale or colorless with or without black centers) to *Salmonella* on the agar plates on desoxycholate Citrate Agar (DCA) and a bright red color on propylene glycol deoxycholate agar (PGDA) were cultured on the agar plates having triple sugar iron agar slants, urea agar and lysine broth and incubated at 38°C for 48h. By sub-culturing onto fresh (PGDA) plates, the purity of suspected *Salmonella* colonies were identified that were hydrogen sulfide positive on triple sugar iron (TSI) agar, urease negative and lysine positive. The purified *Salmonella* colonies were moved to agar slants by streaking and further biochemical tests were performed which included sugar, decarboxylase test and glycerol fermentation.

Salmonella spp. cell culture

## aMicrobial method

Poultry waste was collected from different poultry farms and mixed with wheat straw of 0.5 mm in size. All apparatus used

for composting process were sterilized. Experimental flasks were washed thoroughly with double distilled water (DDW) and sterilized. After adding poultry waste and wheat straw in it, the mouth of the flasks were closed by putting cotton plug in it and covered with aluminum foil. The flasks thus made ready with poultry waste, were autoclaved at 121°C and at 15 psi for 15 minutes in order to kill all the microbes present in the poultry wastes. Three flasks were taken for control and three for experimental compost separately. In the experiment, initially 100 g of each sterilized poultry waste mixture (poultry waste: 70% and 30% wheat straw) was used. The sterilized sample mixed with 10% of broth inoculums of each Salmonella isolates, separately. Similarly sterilized raw material were mixed with different chemicals (N, P, K, alum, ammonium, nitrate) to analyze the humification of poultry waste. During composting the inoculated flasks were incubated at 38°C.

#### Chemical Method

Poultry waste and wheat straw were melded in a mechanical composter of 10 L capacity. The composter drum (300 mm long and 250 mm in dia) made up of 3 mm thick sheet of stainless steel (SS) and was fixed on an iron stand. Inside the drum, steel angles were welded horizontally to provide appropriate mixing of wastes. Simultaneously, two holes (200 and 100 mm) were made both at the upper and the lower portion of the drum, respectively. With the help of SS containers, the mixed poultry waste and wheat straw were loaded into the composter and filled up to 50% of the total volume. To ensure proper mixing and aeration, the composter was run regularly 5 h daily at 500 rpm by electrical gearbox. A thermocouple was fixed to monitor the regular changes in temperature after every five hours. Alum (0.3%) was used as an inorganic additive, which was spreaded on the poultry waste mixture and homogenized in a mechanical composter in each experimental batch. Samples without additives and microbes were run and observed for the same parameters for all chemicals and microbes. Controls were done in triplicate. All the results were reported as an average. The 300 g samples were collected from each runs after five days intervals regularly up to sixty five days. Samples were dried at 75°C and passed through 2 mm sieve for the analysis of ash, carbon nitrogen ratio (C/N), humification index(HI), degree of polymerization (DP) and humification rate (HR).

## Analytical method

The chemical properties (C/N,HR, FA, DP, HA, HI, CEC and ash) of the compost were verified after Iqbal *et al.* (2010). The micronutrient (Zn, Mn, Fe, Cu, and Cd) concentration of the compost were analyzed by using the Atomic Absorption Spectrophotometer (Analytik Jena, (novAA:800). The chemical properties of the compost

samples were anal yzed by using standard methods (Peech *et al.*, 1962; Nelson and Sommers, 1996). The analyses of total N and total C from the experimental samples were carried out by catalytic tube combustion using the Vario Macro Elementar Analysensysteme Gmb HCHNS analyzer (S.N: 11046079) The carbon and nitrogen ratio was calculated as the quotient of total carbon over total nitrogen.

## Results and discussion

Fig.1a.showed the comparison of C/N ratio between experimental, control and chemical decomposition. As C/N ratio was decreasing continuously in the experimental samples of control, microbial and chemical studies, chemicals were applied to poultry wastes for the purpose of decomposition. Results show further decrease in C/N as compared to microbial values. With the raise in composting time, there was a decline in carbon and nitrogen ratio for both the composts namely, microbial and chemical (Igbal et al., 2014). At 25th day of decomposition C/N starts to decrease largely as compared to microbial ratio. Chemical decomposition gives 17.86% C/N where as microbial decomposition gives 22.89% C/N. Maximum difference noted on the 30<sup>th</sup> day of decomposition when microbial gave 22.06% C/N and chemical decomposition gave 16.04 % C/N. In poultry waste low carbon to nitrogen ratio caused the loss of N through NH<sub>3</sub> volatilization (Tiquia and Tam 2002). According to (Golueke *et al.*, 1981) carbon and nitrogen ratio less than 20 show the acceptable maturity and ratio of 15 or less is expedients. One of the most vital factors that affect the quality of compost is carbon to nitrogen ratio. In composting, a range of C/N ratio from 25 - 30 considered ideal (Kumar *et al.*, 2010). In composting, lower initial carbon to nitrogen ratio is helpful in increasing the amount of treated manure which also helps in increasing the loss of N as NH<sub>3</sub> gas. Compost stability showed the low carbon to nitrogen ratio, while reduction in carbon showed the maturity of compost. Microbes use organic compound for their nutrition that is the main reason in reduction of C/N ratio (Waqas *et al.*, 2017).

The qualitative differences in microbial and chemical decompositions have been depicted in Fig.1b. The decomposition rate was high in chemical method than microbes, because microbes used the organic carbon for their energy purpose. Moreover, both treatment results showed the significant difference. During observation on 25<sup>th</sup> day the nitrate concentration was 198.65 mg/kg and 143.06 mg/kg by chemical and microbial method respectively. This remarkable change was also observed on 50<sup>th</sup> day of composting between these two method results i.e. microbial decomposition gives 307.68 mg/kg value of NO<sub>3</sub><sup>-</sup> while chemical decomposition of NO<sub>3</sub><sup>-</sup> gives 352.54 mg/kg (Fig.1b). Nitrate is a salt and it dehydrates the surrounding

Table I. Chemical characteristics of poultry waste, experimental, control compost and wheat straw

Parameters	Poultry Waste	Wheat Straw	Control Compost	Chemical Compost	Microbial Compost
Moisture (%)	55.78	11.59	38.65	29.18	29.16
pН	7.8	6.54	8.02	8.87	8.42
Ash (%)	42.45	35.49	40.09	54.32	47.65
Organic Matter (%)	57.55	64.51	59.91	45.68	52.35
Total Nitrogen (%)	3.85	0.1	1.6	1.86	1.6
Total Carbon (%)	31.44	35.25	32.73	24.96	28.60
C/N ratio	8.16	352.5	20.45	13.41	17.87
Total P (%)	1.6	0.17	1.18	2.37	2.19
Total K (%)	1.12	0.06	2.33	2.21	2.11
Total Sulphur (%)	0.27	0.39	0.21	0.24	0.17
Total Hydrogen (%)	6.8	1.7	4.3	3.34	3.28
Zn (mg/kg)	21.43	0.07	11.46	16.53	16.47
Pb (mg/kg)	0.53	0.08	0.18	1.34	0.28
Mn (mg/kg)	1.15	0.09	0.12	1.64	1.51
Cu (mg/kg)	0.54	0.15	0.37	0.56	0.40

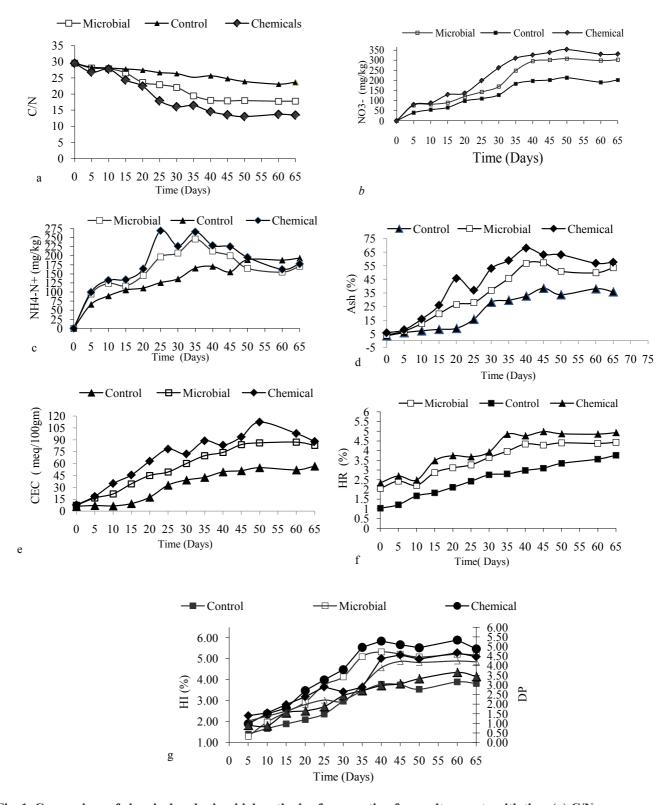


Fig. 1. Comparison of chemical and microbial methods of composting for poultry waste with time (a) C/N (b) Nitrate (c) ammonia (d) ash (e) CEC (f) HR (g) HI & DP

soil, as well as it burns up the OM in soil because of being very strong oxidizer (Zhen et al., 2014). NH, volatilization may cause decline in NH4+-N that indicates the increase in NO<sub>3</sub>-N. The temperature rise in compost may cause the loss in NH, +-N and rise of NO, --N may help to plant for uptake of nitrogen for plant growth. In sewage sludge the ammonia concentration decrease with time due to decomposition of nitrogenous waste (Yuan et al., 2015). Increased content of NO<sub>3</sub> than NH4+-N shows the process of composting has been performed under appropriate aeration (Maria Gomez-Brandon et al., 2008). The application of chemicals and Salmonella to decompose the poultry waste showed maximum achievements in chemical method. On 10<sup>th</sup>day, the ammonia released was 134.34 mg/kg where as 117.09 mg/kg were released for microbial method. It was 13 percent less than chemical method. This pattern did prevail throughout the composting but at the end of the process, there no significant difference was observed (Fig.1c). The decrease of ammonia was due to the volatilization at high pH and temperature (Guo et al., 2012).

Fig. 1d depicted the comparison of control and microbial and chemical treatments on the same day of composting samples. At 5<sup>th</sup> day of decomposition, no significant difference was seen among the samples. But as the process of composting proceeded the degree of decomposition increased gradually (Fig.1d). After 40th day, the percentage of decomposition of ash increases in chemical as compared to microbial samples. Ash and organic matter related inversely to each other. High organic matter content results low ash (Waqas *et al.*, 2017). Higher microbial activities give highest rate of decomposition. Furthermore, the ash content is an additional sign of decomposition process that showed a high degree of decomposition.

On the 25th day of decomposition, CEC values in control, microbial and chemically treated samples were 32.76, 49.32 and 78.36 meq/100 g, respectively (Fig.1e). The values were increasing with composting time. Increase in CEC during composting occurs because of aggregation of negatively charged materials such as phenolic hydroxyl and carboxyl groups (Harada and Inoko, 1980; Gao *et al.*, 2010). Microbial alteration of lingo cellulose into humus can also cause increase in CEC (Iqbal *et al.*, 2010).

The decomposition of poultry waste is a humification process indeed, where the production of fulvic acid initially increased compared to humic acid. The concentration of the latter increased as the composting process move forward whereas the fulvic acid decreased. The humification rate depends upon the nature of waste, moisture, chemical reaction and polymerizing

agent. As the humification rate (HR) increases the humification index (HI), the degree of polymerization (DP) is also increased. They are directly proportional to each other. The microbial activity results are not significant than chemical. Chemicals enhanced the decomposition rate than microbes because it raised the pH and temperature of the process. On 5th day of decomposition, HR value of control, microbial and chemically treated samples showed 1.21, 2.43 and 2.69%, respectively. Significant difference can be seen on the 35th day of decomposition when the HR values increased to 4.84, 3.97 and 2.80% respectively in control, microbial and chemically treated samples (Fig-1f). Moreover, the same observation was observed in HI and DP (Fig-1g). The results of the present study are in agreement with Iqbal *et al.* (2015).

## Conclusion

The addition of wheat straw reduced the moisture of poultry waste which enhanced the humification process of the waste. The addition of alum increased the humification rate than *Salmonella* and reduces the time for decomposition. All the humification parameters were correlated with each other and the present findings can be applicable on other wastes also.

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