

## PHYSICO-CHEMICAL AND MICROBIOLOGICAL CHARACTERISTICS OF DEAD BIRD AND LITTER COMPOST DURING WINTER SEASON

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### ABSTRACT

An exploratory study was conducted to evaluate the physico-chemical and microbiological characteristics of dead poultry and litter compost during different stages of composting in winter season. The present study was conducted under completely randomized design (CRD) by repeating the experimental trial for three times. For this purpose, three composting bins were loaded with organic waste comprising dead poultry and litter by following the internationally accepted standard method of bin filling. Sampling was done at the end of each phase to record the data for physico-chemical, mineral and microbiological parameters. The data were analyzed by using ANOVA technique under CRD. The results indicated higher maximum temperature, maximum moisture, and crude protein in primary phase, average temperature in secondary phase, dry matter, ash, calcium, phosphorus, and potassium content in curing phase of composting. However, nitrogen content, C:N ratio and bacterial count were significantly ( $P \leq 0.05$ ) reduced during curing phase of composting. It may be stated that composting technology is an environmentally safe method of dead birds' disposal that can be used to convert organic waste into useful organic fertilizer.

**Key words:** physico-chemical characteristic, microbiology, compost, winter season

### INTRODUCTION

Poultry is rapidly growing industry of the world particularly in tropical and sub-tropical zones (Daghir, 2009) and is classified among one of the best animal protein sources existing worldwide (Kocaman *et al.*, 2006). Pakistan is an agro livestock based economy and the poultry industry shares a major part in the economy of Pakistan (Abidin, 2011). The total share of agriculture in the national GDP is 21% while, the livestock and poultry share is 55% of total agricultural GDP. The poultry industry has an annual growth rate of 8-10% (Economic Survey, 2015-16).

In Pakistan, high prevalence of diseases due to improper biosecurity measures lead to high mortality rates in poultry. These dead birds are the potential threat of disease spread; hence their disposal is a big challenge. Dumping of stock is a biosecurity hazard, which not only threats to water quality but also a source of concern for environment protection agencies (Bonhotal *et al.*, 2002). Several methods are being applied for proper disposal of poultry carcasses, including burial, incineration, composting and rendering with some advantages and disadvantage. Burying of dead birds is a big source of soil and water contamination (Wood *et al.*, 2010). Incineration is a biological safe method of dead birds' disposal, but it is slow and also expensive even when highly efficient incinerators are used (Blake and Donald,

2002). Composting is a cost effective and environmental friendly method for disposal of dead poultry (Bonhotal *et al.*, 2002). It takes place under aerobic, thermophilic natural biological decomposition process (Sivakumar, 2006). It is a natural process which is speeded by participation of organic waste through prescribed manner for optimum microbial growth (Keener *et al.*, 2000).

For poultry rearing, wood shavings and rice husk are widely used as bedding material. Almost 25 ton poultry litter is removed which contains several tons of poultry manure and other wastes from a 30,000 capacity poultry house at the end of poultry flock, which is mostly used as fertilizer in agriculture lands and also in brick factories as a fuel (Economic Survey, 2015-16). Moreover, poultry litter (rice husk, wood shavings and wheat straw) is a good quality organic fertilizer due to its high contents of nitrogen, phosphorus and potassium. It improves organic matter in soil, enhances nutrient and water retention capacity, and stabilizes soil structure, aeration and water infiltration (Deksissa *et al.*, 2007). However, usage of raw poultry litter disseminates pathogens, e.g., *E. coli*, causing endocrine disorders when N and P contents elevates in soil and water bodies (Deksissa *et al.*, 2007). Improper disposal of litter results in contamination of land and water with nitrate, causing respiratory illnesses, cancer and fetal anomalies (Sims *et al.*, 2000). In composting process, the biological risks associated with dead birds can be managed as well

change in physical and chemical characteristics of the original substrate.

Composting technology has widely been used and studied worldwide, but so far no work has, been conducted in this area in Pakistan. The present study was, therefore, undertaken to evaluate physico-chemical, mineral and microbiological characteristics of dead bird and litter compost during different phases of composting in winter season.

## MATERIALS AND METHODS

The present study was executed at Compost Unit, Department of Poultry production, Ravi Campus, University of Veterinary and Animal Sciences, Lahore. The trial was executed in triplicate under Randomized Complete design (CRD). For this purpose, three composting bins were filled with organic waste comprising dead poultry and litter by following the internationally accepted standard method of bin filling. The dimension of each bin was 6 ft L × 7 ft W × 5 ft H. Litter was obtained from big bird breeder unit (Litter was arranged into 6 layers, in each layer 360 kg litter was used), of the university and dead birds (72) were collected from a commercial broiler farm. Chemical reagents were obtained from Sigma Aldrich, USA. The duration of the proposed study was 3 months (December, January and February). Moisture was maintained in the range of 50 to 55%. Composting process was carried out as per the procedure followed by USDA (1998) and NRCS (2004).

**Analysis of Litter and Dead Broilers:** Before the start of composting, sample of 500g litter and 10 dead birds were randomly collected and used for bacterial count. After the completion of each phase, five random samples of composting material were collected and analyzed for above said analysis.

**Physical characteristics:** The study comprised three phases i.e. primary, secondary and curing phase. Temperature was monitored daily by using digital compost thermometer. Temperature was recorded from five different locations and average was calculated. Temperature reached thermophilic stage (130 °F to 150 °F) in primary bin within 5 to 7 days after filling, and then it started to decline. When it reached down to 129°F, the material was turned to secondary bin (secondary phase). The purpose of turning was just to aerate and homogenize the material. As a result of aeration, temperature again started to rise and reached thermophilic stage (130 °F to 150 °F) within five days after turning. Secondary phase was complete when temperature was again decreased to 129 °F. After that material (compost) was shifted to a storage area and was kept there until the temperature was declined to the room temperature (80 °F to 90 °F). External temperature and

humidity were recorded three times daily. The change in color of the compost material was also recorded at all the stages.

**Chemical analysis:** Five samples were collected from five different locations at the end of each phase of composting. The total carbon was determined from total organic matter value by using the conventional “Van Bemelem Factor” of 1.724. The weight loss on ignition was divided by 1.724 to determine the percentage of total carbon (Navarro *et al.*, 1993; Lawson and Keeling, 1999). Total Kjeldahl nitrogen was calculated by digesting the compost sample in concentrated sulphuric acid and by distillation and trapping the ammonia in 0.1N sulphuric acid titrated against standard 0.1N sodium hydroxide solution (AOAC, 2000). C:N ratio was calculated based on the total carbon and total Kjeldahl nitrogen concentration (Zhu *et al.*, 2004). The compost samples were analyzed for calcium and potassium using Flame photometer (Jackson, 1973). The concentration of phosphorous was estimated by using spectrophotometer (Vanado- molybdate yellow colour method) as described by Jackson (1973). Total ash content was determined by assessing loss on ignition in a muffle furnace at 550°C for 5 hours (Allison, 1965). An aqueous solution of compost sample mix was prepared (1:10 W/V compost-water extract) and pH was measured (Tiquia and Tam, 2002) by using a digital pH meter (Systronic make MKVI). Dry matter, crude protein, ether extract were determined using official methods of AOAC (2000).

**Bacterial Count:** Total plate count was carried out by using method of Cunningham *et al.* (2011). *Coliform*, *Salmonella* and *Shigella* count was performed by the method of Cappuccino and Sherma (2007).

**Statistical Analysis:** The data thus obtained were analyzed through ANOVA technique (Steel *et al.*, 1997) using PROC GLM. Means were compared through Duncan’s (1955) Multiple Range (DMR) test with the help of SAS 9.1.

## RESULTS AND DISCUSSION

### Physical characteristics

**Temperature and Moisture:** In primary phase or first heating cycle of composting, maximum temperature was 164.3 °F, while, minimum temperature was 65 °F. However, average temperature was 126.83 °F. Maximum and minimum moisture percentages were 54 and 45%, respectively, whereas, average moisture percentage was 48.85%. Similarly, in the secondary phase of composting, maximum and minimum temperatures were 147.8°F and 123.6 °F, respectively. However, overall average temperature in the secondary phase was 136.33°F. Maximum and minimum moisture percentages in the

secondary phase were 49 and 45, while, average moisture percentage was 47.04. In curing phase, maximum, minimum, and average temperatures were 135.58°F, 123.3°F and 131.81°F, respectively. Maximum, minimum and average moisture percentage were 44, 38, and 41, respectively.

The principal stages for microorganisms in composting includes; mesophilic (50°F to 110°F) and thermophilic (110 to 160°F) (Koberstein, 2002). Thermophilic stage is an active stage of composting where the rate of decomposition is much quicker than a mesophilic stage (Langston *et al.*, 2002). Temperature above 131 °F is sufficient for the elimination of most of the pathogenic microorganisms (Joshua *et al.*, 1998; Kube, 2002). Similarly, parasites, fecal and plant pathogens within compost are destroyed when its temperature reaches above 131 °F. Moisture is kept around 50% during the composting process (Salma *et al.*, 2006). More than 60% of moisture excludes oxygen from the pores of the compost heap and stops its aerobic activity. Additionally, moisture above 60% produces odor and stops temperature to rise as well (Murphy and Carr, 1991; Looper, 2002).

#### Proximate Analysis and pH

**Dry Matter:** Means of dry matter revealed significant ( $P \leq 0.05$ ) difference among different phases of composting. Dry matter was found to be the highest in curing phase followed by secondary and primary phases. The highest dry matter in finished compost may be attributed to the higher moisture loss in maturation or curing phase. Adeleye and Kitts (1983) and Muller (1982), similarly, reported increased dry matter content in finished compost. Due to microbial degradation of organic matter and subsequent heat generation, there was reduction in moisture content; weight and volume of compost pile thus, increasing dry matter at the end of composting (Gajalakshmi and Abbasi, 2008).

**Ash:** Different phases of composting demonstrated variations in ash percent. Higher ash percentage was observed in curing phase than primary and secondary phases of composting. Similarly, total ash content progressively increased and total organic content decreased as composting process proceeded, indicating mineralization and degradation of organic matter (Chefetz *et al.*, 1996). A study on composting, likewise, reported the increased ash value due to prolongation of composting period (Flachowsky and Hennig, 1990). Similar results were also documented by Jacob *et al.* (1997) indicating that composting reduced the organic matter that ultimately enhanced the ash content in the final product.

**Ether extract:** In the present study, means of ether extract indicated similar pattern among different phases of composting. However, Tiquia and Tam (2000)

reported decrease in ether extract percentage as the compost matured, indicating inverse relationship between ether extract and the time of composting process (Sivakumar, 2006).

**Crude protein:** Means of crude protein showed significant difference among different phases of composting. In the present study, value of crude protein decreased as the composting process proceeded; resulting in reduced crude protein in curing phase followed by secondary and primary phase of composting. This may be due to microbial degradation of organic matter during composting process. It was also confirmed by Tiquia and Tam (2000) indicating reduction in crude protein content as the compost matured. Sivakumar (2006) also attributed the decrease in crude protein content to the microbial degradation of organic matter during composting process.

**pH:** Means of pH revealed non-significant difference among different phases of compost. In the present study, from primary to curing phase of composting, pH remained in the range of 8.81 to 8.86. Kumar *et al.* (2007), likewise, reported a pH range of 7.27 and 8.53 in the finished dead bird compost, indicating stabilized end product, compost (Tiquia and Tam, 2002). Similarly, finished compost of dead birds had slightly alkaline pH, suggesting stabilization and was in line with earlier reports with pH ranging from 6.82 to 8.9 (Ahmed *et al.*, 2012).

#### Bacterial Count

**Total plate count:** Different phases of composting demonstrated variations in total plate count. Significant decline in total plate count was observed in secondary and curing phase of composting compared to primary phase. The rise and fall of microbial activities is directly related with the rise and fall of temperature (Tiquia and Tam, 2002). Microbial activity weakened rapidly when temperature reaches above 66 °C and gradually reaches low values as temperature of compost reaches above 71 °C (Keener *et al.*, 2000). Most pathogenic bacteria and parasites are killed and most viruses are inactivated at the temperature of 55°C for 3 consecutive days (Berge *et al.*, 2009).

**Salmonella and Shegella Count:** Means of *salmonella* and *shegella* count revealed significant difference among different phases of composting. *Salmonella* and *Shegella* count reduced significantly in secondary and curing phase than primary phase of composting (Table 2). In the current study composting was completed by subjecting the organic waste into two heating cycles, which might have resulted reduced pathogenic bacterial count in secondary and curing phase of composting. It is quite possible that only one heating cycle during composting process might not have effectively destroyed pathogenic organisms as it is reported that the long composting time

can effectively eradicate *Salmonella and Coliform* bacteria (Bicùdo and Goyal, 2003). Similar to these findings, Das *et al.* (2002) observed 99.9% and 100% reduction in *E. coli* and salmonella, respectively. The finished compost, likewise, had undetectable level of salmonella (Vinodkumar, 2014).

**Coliform count:** Means of *coliform* count exhibited significant difference among different phases of composting. Significantly ( $P \leq 0.05$ ) reduced *Coliform* count was observed during secondary and curing phase than primary phase of composting (Table 3), which might be due to the long composting time or two heating cycles. *Coliforms* can grow in adverse environments characterized by low pH and low temperatures. Fecal *Coliforms* can survive for long periods in water and soil (Bicùdo and Goyal, 2003). However, the long time and high temperature during composting is reported to kill pathogens and help control disease outbreaks (Bonhotal *et al.*, 2002). Bicùdo and Goyal (2003), similarly, reported that the long composting time can effectively eradicate *Coliform* bacteria. Imbeah (1998), likewise, stated that composting reduces the pathogenic organisms due to the high heat produced during the process of composting. All microbial flora are inactivated within 24 hour as the temperature reaches 50 °C during an aerobic thermophilic phase (Bicùdo and Goyal, 2003).

#### Mineral analysis

**Calcium:** Means of calcium showed significant difference among the phases of composting. Highest calcium content was recorded in curing phase of composting followed by secondary and primary phase of composting. The total Ca content indicated an increasing trend from the primary to curing phase of composting. The loss of organic matter during composting process may be the plausible reason for the increase in the Ca content in curing phase or finished compost. Sakthivadivu *et al.* (2015), likewise, reported an increasing trend in Ca content from the primary to secondary stage of composting. Kumar *et al.* (2007) also reported similar findings. Chefetz *et al.* (1996) reported similar progressive increase in total Ca content as composting proceeded.

**Phosphorus:** Different phases of composting indicated marked difference in phosphorous content. Curing phase showed the highest phosphorous content followed by secondary and primary phase of composting that may be due to the mineralization and degradation of organic matter. Similarly, Chefetz *et al.* (1996) observed progressive increase in total P content as composting proceeded, indicating an increasing trend in phosphorus content from primary to the secondary phase of

composting (Sakthivadivu *et al.*, 2015). Similar range or values (18 to 34.8 g/kg) of total P content in dead bird compost have previously been reported (Murphy and Carr, 1991; McCaskey, 1994; Lawson and Keeling, 1999; and Kumar *et al.*, 2007).

**Potassium:** Means of potassium displayed variation among different phases of composting. The highest potassium content was recorded in curing phase followed by secondary and primary phase of composting. This increasing trend in K content may be due to the mineralization and degradation of organic matter as it is reported that the loss of organic matter during composting process results in increased potassium content (Chefetz *et al.*, 1996). Similar increasing trend in K content has already been reported (Sakthivadivu *et al.*, 2015). Likewise, Murphy and Carr (1991), Lawson and Keeling (1999), McCaskey (1994) and Kumar *et al.* (2007) observed similar values of potassium in dead difference in nitrogen bird compost (18 to 38 g/kg).

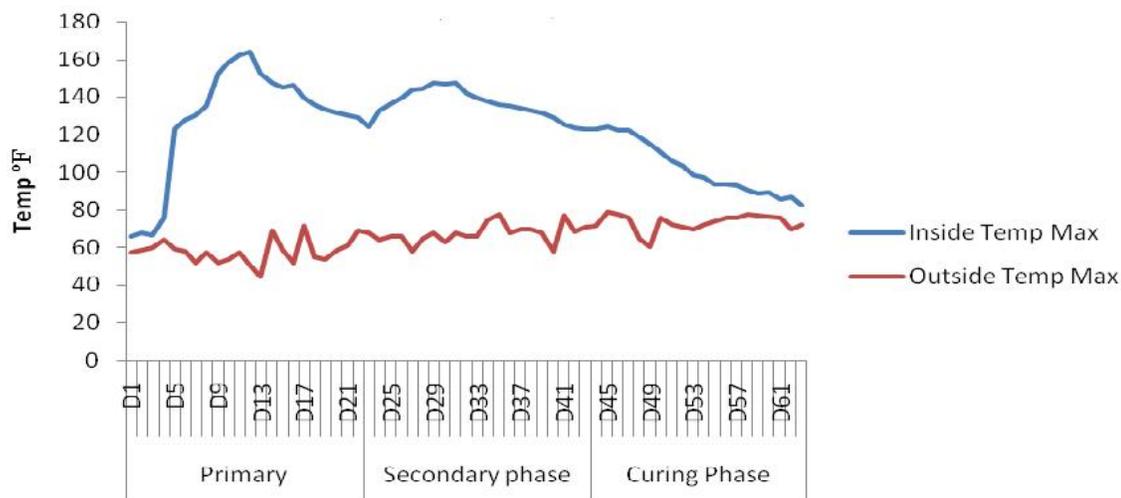
**Nitrogen:** Different phases of composting indicated significant free extract. Secondary and curing phase indicated significantly reduced nitrogen content compared to primary or initial phase of composting that may be due to the ammonia volatilization. Similar reduction in nitrogen during composting of dead birds has already been reported (Bertoldi *et al.*, 1983) and confirmed by Sivakumar *et al.* (2007), concluding that nitrogen content decreases in composting of poultry manure (Hansen *et al.*, 1989), Mahimairaja *et al.*, 1994) and hatchery waste (Das *et al.*, 2002). Similarly, heavy loss of N (up to 33 %) during composting of poultry manure has also been reported (Hansen *et al.*, 1989).

**C:N ratio:** Significant difference was observed among different phases of composting with respect to C: N ratio. Curing phase showed significant reduction in C: N ratio followed by secondary and primary phase of composting. The reduction in C:N ratio in curing phase may be due to the degradation of organic waste. In the current study, C:N ratio gradually reduced from the primary to curing phase (Sakthivadivu *et al.*, 2015), indicating maturity of the finished compost, as it is reported that a C:N ratio below 20:1 is considered satisfactory for compost maturity (Hirai *et al.*, 1983). Similarly, C:N ratio in windrows with dead birds reduced below 20:1, indicating the maturity (Gajalakshmi and Abbasi, 2008). C/N values in stabilized composts vary between 5 and 20, depending on the type of raw material (Tiquia *et al.*, 1996). Similar to the current findings, higher rate of reduction (from 23:1 to 7:1) in C:N ratio was reported by Cummins *et al.* (1993), Lawson and Keeling (1999) and Sivakumar *et al.* (2007) in dead bird compost.

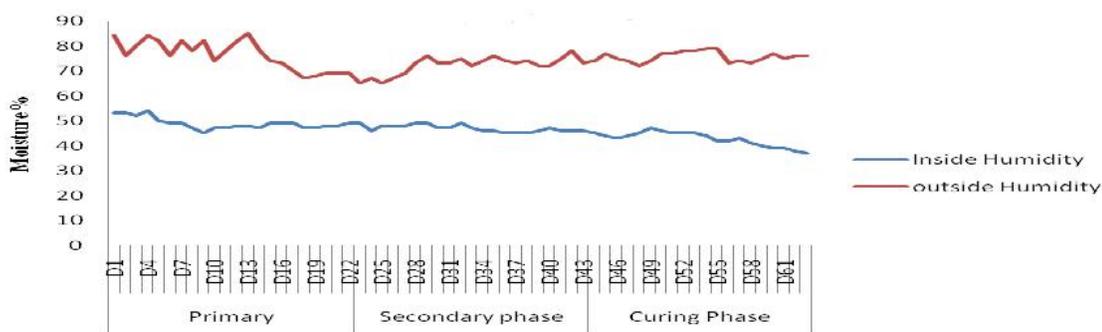
**Table 1. Bacterial count of Meat and Litter before composting.**

Parameters	Samples Meat		Parameters	Samples Meat/litter Litter	
	Total plate count (cfu/g)	64800 × 10 <sup>3</sup>		DM %	24.985 × 10 <sup>3</sup>
Salmonella and shegella count(cfu/g)	46000 × 10 <sup>3</sup>		ASH %	4.450 × 10 <sup>3</sup>	18.88
Coliform count (cfu/g)	417000 × 10 <sup>3</sup>		BE %	7.253750	1.18
			CP %	63.94	12.79
			Moist %	73.98	20.63

**Table 2. Proximate Analysis of Meat and Litter before composting.**



**Figure 1. Trend of daily temperature of different phases of compost**



**Figure 2. Trend of daily moisture of different phases of compost**

**Table 3. Effect of temperature and Humidity on different phases of compost during winter.**

Parameters	Phases	Primary Phase	Secondary Phase	Curing Phase
Maximum Temp(°F)		164.3	147.8	135.5
Minimum Temp (°F)		65	123.6	123.3
Average Temp(°F)		126.83	136.33	131.81
Maximum Moisture (%)		54	49	44
Minimum Moisture (%)		45	45	38
Average Moisture (%)		48.85	47.04	41

**Table 4. Comparative proximate analysis of different phases of compost during winter.**

Parameters	Primary Phase	Secondary Phase	Curing Phase
DM %	47.00±0.41 <sup>c</sup>	51.75±0.48 <sup>b</sup>	61.75±0.48 <sup>a</sup>
ASH %	39.49±1.36 <sup>b</sup>	39.87±0.82 <sup>b</sup>	42.89±0.71 <sup>a</sup>
EE %	3.64±0.79	2.32±0.27	2.57±0.16
CP%	19.71±0.82 <sup>a</sup>	16.52±0.50 <sup>b</sup>	14.48±0.27 <sup>c</sup>
pH	8.81±0.035	8.68±0.13	8.86±0.02

Different alphabets on means within rows show significant difference ( $P \leq 0.05$ )

**Table 5. Effect of winter season on bacterial count (000) during different phases of compost.**

Parameters	Phases		
	Primary Phase	Secondary Phase	Curing Phase
TPC	2142 × 10 <sup>3</sup> ±36249.14 <sup>a</sup>	55200±1714.64 <sup>b</sup>	952±19.34 <sup>b</sup>
Coliform	263.4 × 10 <sup>3</sup> ±8022.47 <sup>a</sup>	500±7.07 <sup>b</sup>	0±0 <sup>b</sup>
Salmonella and Shigella	2.06 × 10 <sup>3</sup> ±146.97 <sup>a</sup>	164±16.31 <sup>b</sup>	0±0 <sup>b</sup>

Note: Different alphabets on means within rows show significant difference ( $P \leq 0.05$ )

**Table 6. Mineral analysis during different phases of Compost.**

Phases Minerals	Primary	Secondary	Curing
Calcium (g/Kg)	32.57± 0.74 <sup>c</sup>	56.56±0.60 <sup>b</sup>	67.15±0.52 <sup>a</sup>
Phosphorous (g/Kg)	16.32±0.34 <sup>c</sup>	24.52±0.41 <sup>b</sup>	32.08±0.46 <sup>a</sup>
Potassium (g/Kg)	16.24±0.31 <sup>c</sup>	23.78±0.50 <sup>b</sup>	32.11±0.60 <sup>a</sup>
Nitrogen (g/Kg)	19.24±0.75 <sup>a</sup>	15.78±0.35 <sup>b</sup>	14.32±0.50 <sup>b</sup>
C:N_ratio	17.57±0.35 <sup>a</sup>	15.33±0.19 <sup>b</sup>	14.22±0.17 <sup>c</sup>

Note: Different alphabets on means within rows show significant difference ( $P \leq 0.05$ )

**Conclusions:** From the present findings, an inference can be drawn that composting can be utilized for hygienic disposal of dead birds and their wastes with recycling nutrients in the form of compost that can be used as fertilizer. It is more environmentally safe disposal option than traditionally used in Pakistan.

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