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# Characterization of Organic Manures for Polysaccharides, Microbial Biomass and Humic Substances

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## Authors' contributions

This work was carried out in collaboration between all authors. Authors SA and SSI designed the study, wrote the protocol and author KB wrote the first draft of the manuscript. Author SSI reviewed the experimental design and all drafts of the manuscript. Author KB managed the analyses of the study and performed the statistical analysis. All authors read and approved the final manuscript.

#### Article Information

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# ABSTRACT

Soil organic matter (SOM) is an important indicator of soil quality as it determines many soil characteristics such as nutrient mineralization, structural stability and water holding capacity. An understanding of different bio-chemical processes undergoing inside the conventional sources of organic matter, which are used for farm management regarding improvement of soil structure which rely on organic carbon input as organic carbon is the main binding force between soil particles. Farm yard manure (FYM), poultry litter (PL) and municipal solid waste compost (MSWC) were characterized in an incubation experiment for six months in a completely randomized design (CRD). This study was carried out in 2011 at Department of Soil Science and SWC, PMAS Arid Agriculture University, Rawalpindi. Organic matter fractionation, total organic carbon (TOC), total polysaccharide content (TPC) and microbial biomass carbon (MBC) were determined. Results showed that PL has significantly highest values for all of the parameters except humic acid as compared to other manures. Results showed that TOC, MBC and humic acid decreased, fulvic acid remained unchanged and TPC increased significantly with incubation period. Interaction of manures and days showed that PL at 180 days had significantly highest contents of TOC and TPC. MBC

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content and humic acid were significantly highest at 0 and 30 days, respectively. Fulvic acid was highest at 30, 120, 150 and 180 days in PL. Correlation analysis showed that TPC was positively and significantly correlated with fulvic acid (0.732), TOC was negatively correlated with humic acid (-0.295) and positively correlated with MBC (0.668). These results suggested that the PL was better among other manures under study regarding TOC, TPC and MBC.

Keywords: Humic acid; Microbial biomass carbon; Organic matter fractionation; Polysaccharides; Manures; Soil structure.

# **1. INTRODUCTION**

Soil organic matter is a major component of earth's carbon cycle and contains more carbon than entire terrestrial vegetation and earth's atmosphere. Within soil, the organic matter has a profound effect on a wide range of key soil properties such as: Cation exchange capacity, soil structure, nutrient cycling and others [1]. Therefore, SOM has recently gained immense attention in scientific literature. In its totality, it includes soil organisms (e.g. microbial biomass), simple organic compounds (e.g. polysaccharides), large and complex humic substances, as well as relatively fresh residue at various stages of decomposition [2]. Humic substances account for the largest and most decomposed proportion of SOM with chemically complex and ill-defined structure. For simplification of their study, humic substances are fractionated into fulvic acid, humic acid and humin [1]. The humic substances are well known to stimulate plant growth chiefly by the formation of metal-chelates [3]. The soil polysaccharides and microorganisms, though very modest in quantities, constitute important part of SOM. The polysaccharides influence cation exchange capacity, carbon metabolism, complexing of metals, and more importantly stability of soil aggregates [1]. The soil microorganisms exert a controlling influence on the dynamics of SOM and nutrients and are capable of producing plant growth hormones which might act as plant growth regulators [4].

A number of organic matter sources (such as farm yard manure (FYM), poultry litter (PL) and composts are used to enhance SOM content, especially in soils where crop residues are not returned due to alternative uses. Being partially decomposed before their application to soil, these organic amendments are very efficient in building SOM [5]. The potential of different organic matter sources to improve soil quality and biological activity is well documented [6,7]. They are also known to differ greatly in their nature and composition of humic and non humic substances [8.9]. However, information regarding the transformations in different humic substances non humic substances during and the decomposition process of different organic matter sources is limited. A purposeful understanding of these changes is important in order to select sources most suitable for soil application, particularly in organic farming systems [10]. This study was, therefore, designed to compare and observe the dynamics the contents of humic substances, in polysaccharides and microbial biomass carbon during decomposition of the most widely used organic matter sources such as FYM, PL and municipal solid waste compost (MSWC).

## 2. MATERIALS AND METHODS

The incubation study was carried out for a period of 6 months using three organic matter sources *viz.* municipal solid waste compost (MSWC), poultry litter (PL) and farm yard manure (FYM). One kg of each organic matter source, replicated three times, were added to air tight containers and kept in an incubator at 30 °C. The samples for various analyses were collected at 30 days interval i.e. 0, 30, 60, 90, 120, 150 and 180 days.

Total organic carbon (TOC) in samples was measured by wet digestion method [11] with a little modification of reducing the sample weight to 0.25 g instead of 1 g, due to high amount of carbon present in the manure samples. Total polysaccharide contents (TPC) were extracted from a 1g dried and ground samples by adding 20 mL distilled water at 80 °C for 24 hours. The supernatant solution was collected after centrifugation at ambient temperature for 25 minutes and the total polysaccharide contents were measured colorimetrically [12]. Microbial biomass carbon (MBC) was measured by the fumigation extraction method using a 0.025 M solution of K<sub>2</sub>SO<sub>4</sub> to extract relatively labile organic C from the fumigated and non-fumigated samples [13] and then carbon in the samples was analyzed by titrating them against acidified ferrous ammonium sulphate. Organic matter

fractionation was carried out by extracting humic acid and fulvic acid using NaOH and HCI [1]. The concentration of these fractions was determined colorimetrically against carbon standards. Carbon recovery was calculated by subtracting humic substances (FA + HA) from TOC. The FA/HA ratio was also calculated.

Data collected from different analyses was subjected to two-way ANOVA (Analysis of variance) using completely randomized design (CRD). The means were separated by LSD (Least significant difference) test at 5 % level of significance [14]. MSTATC and IBM SPSS software were used for statistical analysis.

#### 3. RESULTS AND DISCUSSION

#### 3.1 Total Organic Carbon (TOC)

Total organic carbon (TOC) was significantly higher in PL than the other OM sources throughout the six month incubation period (Fig 1). The TOC content was higher in FYM as compared to MSWC for initial 60 days but after it the difference between them was nonsignificant throughout the remaining period. TOC content in PL did not decrease during the course of incubation except on day 120. In contrast, the TOC contents of FYM and MSWC decreased gradually with incubation period and reached a minimum after 90 days of incubation.

Results showed that equivalent quantity of PL had higher TOC content than FYM and MSWC. Active SOM is composed of the fresh plant or

animal material which is food for microbes and is composed of easily digested sugars and proteins. With the decomposition of organic matter, the content of total carbon decreases from 46–48% of the raw materials to 30–34% at the end [15]. Due to the presence of sawdust in PL, which is resistant to decomposition, its TOC is preserved efficiently, thats why returned into higher TOC contents [16].

### 3.2 Non Humic substances

#### 3.2.1 Total Polysaccharides Content (TPC)

In all the sources of organic matter, TPC contents increased significantly from day 120 onward till the end of 180 days incubation (Fig. 2). Differences among OM sources for TPC contents were generally non-significant throughout the incubation period, except at day 120 where PL had higher TPC content than FYM or MSWC.

The increase of TPC is probably due to decomposition of organic carbon which was taking place as the time of maturity was increasing. Another factor of continuous increase in TPC was the temperature of incubation (30°C), because its the optimal temperature for polysaccharides production by the microbes [17]. Manures serve as source of energy and nutrients for soil microbes and plant roots that produce extracellular polysaccharides which are responsible for flocculation of soil and mineral particles into aggregates [18].

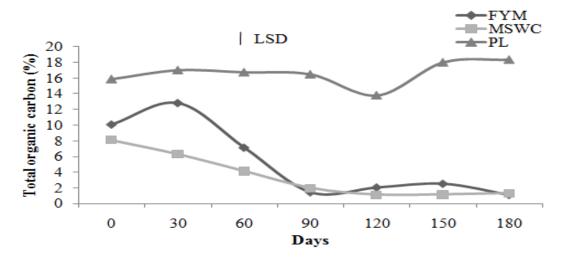


Fig. 1. Dynamics of TOC in different organic matter sources during 180 days of incubation

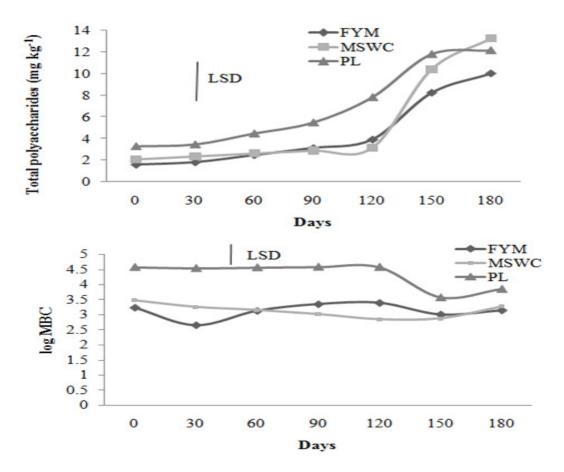


Fig. 2. Dynamics of non humic carbon fractions (TPC and MBC) in different organic matter sources during 180 days of incubation

#### 3.2.2 Microbial biomass carbon (MBC)

The microbial biomass carbon (MBC) in PL was higher than FYM and MSWC throughout the incubation except at day 120 (Fig. 2). The MBC contents of FYM and MSWC were statistically similar to each other throughout the incubation period. The MBC in FYM and MSWC remained similar to their initial levels, while in PL it decreased significantly after day 120.

Because of very high MBC values of PL, log of the MBC was plotted (Fig. 2). The results showed that MBC still in PL was significantly higher than in other manures throughout the incubation period except at 150 and 180 days. It indicates that PL will have higher microbial activity as compared to other manures. Looking at the trend in all the manures during incubation, MBC content was increasing at a somehow constant rate during 30 to 120 days except MSWC. After 120 days there was a sudden decrease in microbial activity in both PL and FYM, but overall it showed that MBC content decreased with time as compared to the start of the experiment. The decline in MBC should be associated with decrease in TOC, which also govern the microbial activities in the soil. Correlation of MBC and TOC (Table 1) showed that they are significantly and positively associated with each other (r = 0.668), so the reason behind the trend of MBC is the TOC and TPC of PL, which was also remained higher throughout the period as compared to the other two manures. The decrease in MBC of PL can be ascribed by the significant and positive correlation of soil organic carbon (SOC) and microbial biomass carbon concentration [19]. Temporal variability of MBC across the whole period was generally, not significantly related to temporal variability values of TPC.

Because when TPC was decreasing at higher rate, microbial biomass carbon levels were increasing at a higher rate, which suggests that if microbial activity reduces it will lead to higher polysaccharide content.

#### 3.3 Humic Substances

The dynamics of fulvic acid (FA) in three different OM sources is given in Fig. 3. The PL had higher fulvic acid (FA) content than FYM and MSWC throughout the incubation period. The FYM and MSWC had similar FA contents throughout the incubation period. Regardless of the incubation period, the FA contents in all the three OM sources remained statistically similar to their initial FA contents.

Humic Acid (HA) contents at the start of the incubation were similar and higher in MSWC or PL than in FYM (Fig. 3). In FYM and MSWC the HA contents remained similar to their initial levels throughout the incubation period. The MSWC had higher HA content than FYM throughout the incubation period. At day 30, HA content was highest in PL, but after wards it started decreasing gradually and at day 120, it became similar to FYM.

The carbon recovery as well as FA:HA ratio were highest in PL (Fig. 4), closely followed by the C recovery in FYM. The MSWC had the lowest carbon recovery and FA:HA ratio, which shows that PL had higher degree of humification and that it is easily decomposable as compared to FYM and MSWC. The MSWC particularly has lowest FA: HA ratio was less than 1.

Results of humic and fulvic acid (Fig. 3) showed that MSWC and PL had overall higher contents, respectively as compared to FYM. In PL at 30 days time, the humic acid content was significantly higher as compared to other days, meanwhile at 30, 120 and 180 days the fulvic acid content was highest as compared to other days. In case of fulvic acid, all the manures were increasing at almost constant rate but during 90 to 120 days PL has lower content of fulvic acid. And it continued decreasing trend from 90 to 120 days but then it started improving in the last month. In case of MSWC the humic acid was significantly higher from 90 to 180 days. Overall humic acid has significant negative correlation (Table 1) with TOC content of the manures (r = -0.295), and fulvic acid has a very strong positive correlation with TPC of manures (r=0.732). During the 30 to 60 days of incubation there is a decreasing trend of humic acid in all the manures but then it started to increase slightly as the organic carbon is being decomposed into complex substances like humic and fulvic acids. Humic substances are the most stable bio-chemical resulting from compounds decomposition of organic matter [20].

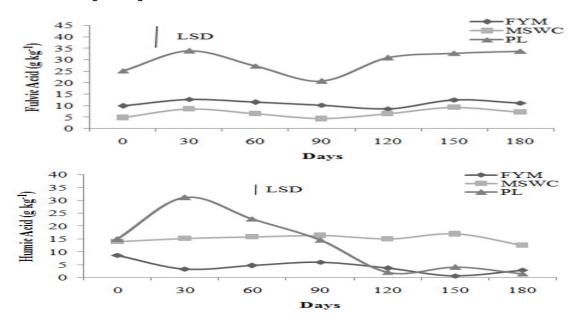


Fig. 3. Dynamics of humic carbon fractions (Humic acid and Fulvic acid) in different organic matter sources during 180 days of incubation

The lesser value of MSWC was due to its compost character, it has higher humic acid contents throughout the period which shows that it is not easily decomposable as compared to other manures. In this study organic manures having higher percentage of carbon like PL, can be a good remedy for structurally degraded soils. Because PL has shown that as the

decomposition process preceded the fulvic acid and polysaccharides increased which can be good source for aggregation of soil particles. Lower FA: HA ratio also showed that it is still decomposing after 6 months of incubation so it will lead to higher concentration of humic substances which are good source of resistant carbon for soils.

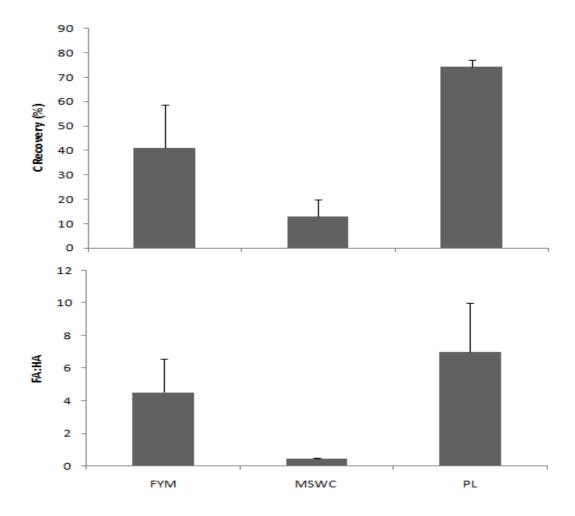


Fig. 4. Carbon recovery and FA: HA ratio in different organic matter sources averaged over 180 days

Carbon fractions	Total polysaccharides	Total organic carbon	Micrboial biomass carbon	Humic acid	Fulvic acid
Total polysaccharides	1	0.024	-0.040	0.123	0.732
Total organic carbon		1	0.668	-0.295	-0.040
Micrboial biomass carbon			1	-0.063	-0.115
Humic acid				1	0.049
Fulvic acid					1

## 3.4 Relationship between Humic and Nonhumic Substances in Different Manures

Humic substances formation is always related to the non-humic fractions as they are the building blocks of stable soil organic matter which is resistant to decomposition. Organic materials are always ready to decompose when they are applied to the soil either for fertility or remedial purposes. The results of correlations between different carbon fractions (Table 1) showed the overall trends between the different parameters.

Correlation of MBC and TOC (Table 1) showed that they are significantly and positively associated with each other (r=0.66), so the reason behind the trend of MBC is the TOC of poultry litter, which was also remained higher throughout the period as compared to the other two manures. Significant and positive correlation of soil organic carbon (SOC) and microbial biomass carbon concentration was also documented [19].

In all the manures, TPC was positively correlated with humic acid (0.122 < r < 0.212) and fulvic acid (0.583 < r < 0.863). In case of FYM and PL, MBC has a positive correlation with humic acid (0.10 < r < 0.122) but negative correlation with fulvic acid (-0.592 < r < -0.239), the trend was different in MSWC, negative correlation was observed with humic acid (r = -0.612) and positive with fulvic acid (r = 0.054). All of the manures exhibited different trends in TOC, it had a positive correlation with humic acid (r = 0.413)but negative correlation with fulvic acid (r = -0.419) in FYM. In MSWC, TOC had a negative correlation with both humic acid (r = -0.593) and fulvic acid (r = -0.332). On the other hand, in PL it had a negative correlation with humic acid (r = -0.289) but positive correlation with fulvic acid (r =0.192).

# 4. CONCLUSION

The usage of organic manures is the integral part of all the integrated management practices for structural and fertility improvement in agricultural soils. This study clearly indicates that different organic sources differ in their bio-chemical properties. By comparing the results of our study, it is elucidated that PL is the best source regarding the TOC, TPC and MBC, so it could be a better source to sequester the carbon in soil environment for agricultural point of view and also as a remedy for structurally degraded soils. Because PL has remarkably shown an increase in fulvic acid and polysaccharide contents and these materials can be good source for aggregation of soil particles.

# **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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