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# **Farm-Scale Mapping of Soil Microbiological Indicators Using Geostatistical Technique**

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#### *Authors' contribution*

*This work was carried out in collaboration among all authors. Author TG analysed soil properties, performed the statistical analysis and wrote the first draft of the manuscript. Author NA designed the study, supervised soil analysis and wrote the protocol. Author RNS helped in geospatial analysis. All authors read and approved the final manuscript.*

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# **ABSTRACT**

Soil microbiological properties viz. soil microbial biomass carbon (MBC) and dehydrogenase activity (DHA) are sensitive soil quality indicators. Spatial modeling and prediction map of soil MBC and DHA were generated for a semiarid agricultural farm, New Delhi, India from 288 geo-referenced grid samples spaced 100 m  $\times$  100 m distance using geospatial techniques and geo-statistics. Soil microbial biomass carbon (MBC) ranged from 19.7 to 519.7 µg g<sup>-1</sup> with standard deviation of 84.1 and soil DHA varied from 1.2 to 17.2 µg TPF g<sup>-1</sup> dry soil hr<sup>-1</sup> with sample variance of 10.89. Soil MBC and DHA had high data viability with coefficient of variation (CV) of 42.5 % and 53.2%, respectively. The best fit semivariogram for both soil MBC and DHA was exponential model and had practical spatial range of 1500 m and 1473 m respectively. Environmental disturbances or extrinsic factors dominantly influenced the spatial variability of soil MBC, expressing its weak spatial

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dependency. Besides, both soil structural/internal factors and extrinsic factors controlled soil DHA variability with moderate level of spatial dependency. Spatial variability map of soil MBC and DHA, prepared with good accuracy through ordinary kriging in GIS software, showed that major area of the farm had soil MBC ranging from 150 to 250 mg kg<sup>-1</sup> and had DHA from 1.2 to 10 µg TPF g<sup>-1</sup> dry soil hr-1 .

*Keywords: Spatial variability mapping; Microbial biomass carbon; Dehydrogenase activity; semivariogram; ordinary kriging.*

# **1. INTRODUCTION**

Human and animal health is closely linked to environmental quality and soil productivity. The soil quality indicators influences sustainable soil productivity and high soil quality is thought of as being biologically active. Soil microorganisms play important functions viz decomposition of soil organic matter & pollutants, nutrient transformation, energy flow etc, having significant implications on agro-ecosystem and environment**.** Soil microbial biomass was used to estimate the biological balance or status of soil ecosystem [1] and dehydrogenase activity was used to assess microbial activity [2]. Soil microbial biomass and activity are critical to soil ecosystem function and sensitive indicators of soil ecological perturbation and stress or restoration [3,4]. Soil microbial biomass, being faster turnover rate than total soil organic matter [5], is a early and sensitive indicator of soil quality [6]. Dehydrogenase activity, occurring in all<br>microorganisms for microbial respiratory microorganisms for microbial respiratory processes [7,8], is considered an accurate measure or indicator of microbial oxidative activity of soil [9]. Soil microbiological and enzyme activities are influenced by several factors viz micro-environmental factors (slope, soil depth and microsite) [10,11], environmental factors viz temperature and moisture, land use pattern and cropping system, soil eco-system, soil physical & chemical properties, soil fertility parameters, soil organic carbon content [12], soil amendments & fertilization, pesticides, pollution, soil management practices viz crop residue management, tillage/compaction, irrigation etc.

Geospatial techniques viz remote sensing, geographic information systems and global positioning system are being extensively used for natural resource mapping. Geostatistical tools, based on theory of regionalized variables, provides the extent of spatial structure or measurement of spatial scale for variability of soil attributes by semivariogram analysis and creation of surface prediction map by kriging interpolation through prediction of attribute values at unsampled locations [13,14]. Spatial variability of

soil properties as well as quality indicators are being widely assessed and mapped using geospatial tools now-a-days [15,16]. Even spatiotemporal assessment and monitoring of soil microbial properties was also reported using geospatial tools [17–19]. Spatial structure of soil microbiological properties and enzyme activities using geostatistical techniques was globally described at field scale [20–23], landscape scale [24], regional scale [25] etc. Soil abiotic parameters viz. soil organic carbon and nitrogen etc showed spatial variability at larger scale, whereas the soil biotic properties viz. microbial biomass and enzyme activity showed significant differences at short-scale [26]. A review work [27] showed that the short-scale and farm-scale spatial variability of soil biological properties using geostatistical techniques was rarely reported in Indian soils. Hence, present investigation was conducted to identify the nature of spatial structure & spatial range of soil biological properties at farm-scale alongwith generation of its spatial distribution map.

# **2. MATERIALS AND METHODS**

# **2.1. Study Location and Soil Sampling**

The assessment and mapping of soil biological quality parameters was conducted at the experimental farm (cultivated area 278 ha) of ICAR- Indian Agricultural Research Institute (ICAR-IARI), New Delhi having longitude of<br>77°8'40.5"-77°10'28.1" East, latitude of 77<sup>º</sup>8′40.5″-77<sup>º</sup>10′28.1″ East, latitude of 28<sup>º</sup>37′22.0″-28<sup>º</sup>38′58.7″ North and elevation of 217-241 m amsl. The location have semi-arid climate with hot summer and cold winter; June is the hottest month and January the coldest month. Annual normal rainfall in last five year (*i.e.* 2006- 10) was 729 mm, of which 612 mm (84%) was received during rainy season (June to September) and rest during winter months (November to March). Soils of IARI farm belongs to mixed, hyperthermic, Typic Haplustepts.

The farm has administrative blocks (Fig. 1) for efficient utilization and operation of farm and natural resources. Main Block (MB), Middle Block

(MID), Genetic Block (Gen), New Area (NA), Shadipur orchard & block, Sewage irrigated area (SA) are located in Northern part of the farm and Top Block (TB), Todapur (TDPR), Water Technology (WTC) Block, Paddock field, National Burreau of Plant Genetic Resources (NBPGR) Block, Precision Farming and Development Centre (PFDC) area, Green house area and forest area in Southern part of the farm. Besides, there exists several sub-blocks within administrative blocks with diversified cropping system. Off-season crops under protected agriculture structures and seed production for farmers as well as irrigation with sewage water were also practiced in the farm. There were fruit orchards of ber (*Ziziphus mauritiana*), mango (*Mangifera indica*), C*itrus*, aonla (*Emblica officinalis*), guava (*Psidium*), jamun (*Syzygium cumini*), grape (*Vitis vinifera*) *etc* at Shadipur block, Todapur block and NBPGR block in the farm. Block plantation of *Jatropha curcas* and *Eucalyptus* in Genetic block and natural forest in south east corner of IARI farm had also been observed.

Soil samples within 0-15 cm depth were collected from farm using grid intersection points on Google Earth image of ICAR-IARI farm with superimposing 100 m  $\times$  100 m grid. Total 288 geo-referenced soil samples collection were collected during fallow period (April-May, 2011) after harvest of *Rabi* crops.

#### **2.2 Soil Analysis**

Soil samples were air dried, grinded with mortar and pestle, sieved with 2 mm sieve for analysis of soil physical and chemical properties. Mechanical composition of the soil was determined by the hydrometer method [28]. Soil organic carbon was determined by wet oxidation method [29] and soil free carbonate was assessed by pressure calcimeter method [30]. Soil pH and electrical conductivity (EC) in 1:2.5 soil and water suspension was measured in a digital pH meter and EC meter respectively. Soil cation exchange capacity (CEC) was measured with sodium acetate replacement and centrifugation method [31]. Soil exchangeable sodium (Na) and potassium was determined by standard method in flame photometer instrument [32]. Exchangeable calcium and magnesium were extracted with KCl-triethanol amine solution, pH 8.2 [31] and determined in atomic adsorption spectrometer (AAS). Percent base saturation (PBS) was calculated from exchangeable base cations and quantity of CEC. Soil MBC & DHA were analysed from freshly collected dry or moist soils and soil samples were stored at refrigerator at 4 ºC. Soil moisture content of collected soil samples from field was estimated by gravimetric method using hot air oven. Soil microbial biomass carbon content was determined by fumigation extraction method [33]. Soil biological activity was determined by monitoring soil dehydrogenase enzyme activity [34] method.



**Fig. 1. Location, major block and collected sample points at ICAR-IARI farm, New Delhi**

#### **2.3 Statistical and Geostatistical Analysis**

The statistical parameters such as descriptive statistics and correlation matrix were conducted in MS-excel & SPSS 16.0. Geostatistical analysis viz semivariogram [35] and generation of surface maps of soil attributes using ordinary kriging [36] were conducted in ArcGIS software version 10.4.1.

#### **3. RESULTS AND DISCUSSION**

#### **3.1 Descriptive Statistics**

Descriptive statistics of soil biological properties and its histogram were presented in Table 1 and Fig. 2 respectively. Soil microbial biomass carbon (MBC) ranged from 19.7 to 519.7  $\mu$ g g<sup>-1</sup>. The mean and standard deviation of MBC was 197.7 and 84.1  $\mu$ g g<sup>-1</sup> respectively. The raw data of soil dehydrogenase activity (DHA) displayed a high variability ranging from 1.2 to 17.2 ug TPF g<sup>-1</sup>dry soil hr<sup>-1</sup> with SD and coefficient of variation (CV) of 3.3 and 53%, respectively. The ranges of MBC & DHA were consistent with several other workers [37–39]. Dehydrogenase activity and MBC showed high data variability due to its CV value greater than 35% [40]. Microbiological properties displayed a higher variability than soil physical and physicochemical properties as described by the coefficient of variation (CV%), which was in agreement with several studies carried out at field scale on arable soils [41,42]. Coefficient of skewness ( $\gamma_1$ ) and kurtosis ( $\gamma_2$ ) for MBC were 0.91 & 1.36 respectively and that for DHA data were 1.17 &1.12 respectively. For

normal distribution of datasets, (γ1, γ2) should be (0, 0). As per test of normality with Kolmogorov-Smirnov statistics, both MBC & DHA were found to be not normally distributed.

Soil physical and chemical properties influenced the soil microbial biomass carbon content and dehydrogenase enzyme activities (Table 2). Microbial biomass carbon was positively and significantly correlated at 1% level with SOC concentration ( $r = 0.37$ ), clay content ( $r = 0.21$ ), silt content ( $r = 0.18$ ), CEC ( $r = 0.19$ ), soil moisture content (0.14) and negatively & significantly correlated with sand content  $(r = -$ 0.24). Dehydrogenase activity was positively and significantly correlated with SOC concentration (r  $= 0.92$ ), MBC concentration ( $r = 0.33$ ), clay content ( $r = 0.33$ ), CEC ( $r = 0.43$ ), soil moisture content (0.21) and negatively & significantly correlated with soil  $pH$  ( $r = -0.31$ ). Soil microbial biomass and enzyme activities are closely related to the organic matter content as it provides substrate for microbial growth and activity [43– 45]**.** Again, dehydrogenase enzyme activity was also correlated with microbial biomass carbon indicating DHA as general indicator of potential microbial activity [12]. In coarse textured soil, with increase of clay content improves soil physical quality, water and nutrient holding capacity as well as microbial growth and activity. Hence, there was positive correlation found between clay content with MBC and DHA. Higher soil pH in alkaline and calcareous soil controlled the dehydrogenase activity indicating soil pH as good predictor of dehydrogenase activity as also reported by other authors [46].



**Fig. 2. Histogram of microbial biomass carbon and dehydrogenase enzyme activity at ICAR-IARI farm, New Delhi**



# **Table 1. Descriptive statistics of soil physical, chemical and biological properties at ICAR-IARI farm, New Delhi**

**Table 2. Pearson correlation matrix of soil physical, chemical and biological properties at ICAR-IARI farm, New Delhi**

	Clay	Silt	Sand	<b>SOC</b>	CaCO <sub>3</sub>	рH	EC	<b>Moisture</b>	<b>CEC</b>	<b>PBS</b>	<b>MBC</b>	<b>DHA</b>
Silt	$0.30^{**}$											
Sand	$-0.78$ **	$-0.83$ "										
<b>SOC</b>	0.39	$0.15^*$	$-0.33$									
CaCO <sub>3</sub>	$-0.02$	$-0.17$	$0.12^*$	$-0.02$								
pH	0.01	$0.13^{*}$	$-0.09$	$-0.29$ <sup>*</sup>	$0.13^{\degree}$							
EC	0.05	$0.21$ "	$-0.16$	$0.15^{\degree}$	0.01	.00						
Moisture	$0.25**$	$-0.08$	$-0.09$	$0.21***$	0.07	0.04	$-0.07$					
<b>CEC</b>	0.73	0.07	$-0.47$	$0.46$ **	0.03	$-0.14$	0.02	$0.30**$				
<b>PBS</b>	$-0.28$ **	$-0.11$	$0.24$ <sup>**</sup>	$-0.25$ <sup>**</sup>	0.04	$0.22$ <sup>**</sup>	0.08	$-0.13*$	$-0.55$ **			
<b>MBC</b>	$0.21***$	$0.18***$	$-0.24**$	$0.37***$	0.04	0.05	$0.17***$	$0.14*$	$0.19**$	$-0.01$		
<b>DHA</b>	$0.33**$	0.06	$-0.23$	$0.92**$	$-0.01$	$-0.31**$	$0.12*$	$0.21***$	$0.43**$	$-0.20**$	$0.33**$	

\*\* and \* Correlation coefficient is significant at the 0.01 and 0.05 level respectively (2-tailed).

### **3.2 Semivariogram of Soil Biological Indicators**

Geostatistical analysis such as semivariogram is pre-requisite for determining spatial dependency of soil attributes and kriging interpolation for thematic map generation. A plot of semivariance  $\hat{y}(h)$  on Y-axis versus lag distance (h) *i.e.* separation distance on X-axis is commonly known as the experimental semivariogram. The isotropic semi-variograms of soil MBC and dehydrogenase enzyme activity are shown in Fig. 3.

Among the theoretical semivariogram model viz linear, linear-to-sill, spherical, exponential and Gaussian model, the best fitted experimental model was selected based on least residual sum square (RSS). In the present study, the optimal experimental model of soil MBC and DHA was exponential model and the key parameters of the semi-variograms are given in Table 3. The parameter range of soil MBC and DHA were 500 m and 491 m respectively. The 'range' of the semi-variogram was the distance (h) at which semivariance value becomes constant (the sill) or attain a plateau. But exponential semivariogram model never appear a plateau and the graph is only asymptotic to the horizontal line corresponding to the sill. The "practical" or "effective" range for exponential model is the distance at which the variogram value is 95% of the sill. In an exponential variogram, the practical range (h) is  $h = 3a$  (where a =<br>Parameter range) [36]. The practical Parameter range) [36]. The practical spatial range of soil MBC and DHA at ICAR- IARI farm were 1500 m and 1473 m

respectively. The range is considered as the diameter of the zone of influence *i.e.* the average maximum distance within which the soil property of two samples was spatially auto-correlated or spatially dependent. It indicated that Soil MBC and DHA of two samples became similar with decreasing separation distance between the two points within spatial range.

Soil biological indicators such as MBC and DHA showed positive nugget, which may be explained by sampling error, short range variability, random and inherent variability. The nugget/sill ratio of <0.25, 0.25-0.75, and > 0.75 range are used to describe the degree of the spatial structure that showed strong, moderate and weak spatial autocorrelation, respectively and this proportion of spatial structure in semivariogram was induced by random factors [47]. The nugget/sill ratio of 0.76 for soil MBC indicated its weak spatial dependency, indicating high spatial heterogeneity due to environmental disturbances or extrinsic factors dominated over the structural factors. It indicated that agri-horticultural practices and long-term soil management altered the spatial structure and dependence of soil microbial biomass and activity, also supported by Katsalirou et al., [12]. The nugget to sill ratio of soil DHA was 0.56 showing moderate spatial autocorrelations and both structural factors viz climate, terrain, parent material, mineralogy, soil texture, inherent soil properties & other natural factors and random or stochastic factors viz such as cropping systems, fertilization, crop management factors and other human activities induced the spatial variability of soil dehydrogenase activities [48,49].



**Fig. 3. Experimental semivariogram and fitted models for (i) MBC & (ii) DHA at ICAR-IARI farm, New Delhi**



# **Table 3. Parameters for best fitted semivariogram model of soil microbial biomass carbon (MBC) & (DHA) at ICAR-IARI farm, New Delhi and other reported locations**

Spatial range of soil biological attributes during analysis of its spatial variability varies on scale of survey like field or plot scale, farm or landscape scale, regional scale etc (Table 3). Liu et al. [24] reported effective spatial range of 157 m with moderate spatial dependency using best fitted Matern semivariogram for soil MBC in a hilly red soil landscape in subtropical China. At farm-scale study of Shahbazi et al. [45], the minimum effective range for MBC & DHA as estimated by best fitted isotropic spherical semivariogram model in Mirabad area, North West of Iran across different land uses are 219 m with moderate spatial dependency & 190 m with strong spatial dependency respectively. As per report of Peigné et al. [51], spatial range of SMBC in an experimental field near Lyon (south-east of France) was 4898 m with exponential semivariogram model using classical ordinary kriging (COK) and was 56.3 m with spherical semivariogram model using robust ordinary kriging (ROK), that removes extreme outlier values. In a study of Piotrowska et al. [54], spatial range of DHA with strong spatial variability at the Sępopolska Plain, near the Budniki Village, Warmia region, northern Poland was reported as 84.3 m with best fitted spherical semivariogram model. In a study of Piotrowska & Długosz [53] on typical Luvisols at arable field scale from northwest Poland**,**  the best fitted semivariogram model for dehydrogenase activity was linear or mixed (spherical/linear) models with nugget effect &

spatial range of 14.9 m, having strong spatial dependency.

#### **3.3 Spatial Variability Maps of Soil Biological Indicators**

Spatial distribution of soil MBC & DHA at ICAR-IARI farm, New Delhi generated through ordinary kriging were displayed in Fig. 4. Majority of the farm area had soil microbial biomass carbon (MBC) ranging from 150 to 250 mg kg-1 . Entomology and Pathology block with fallow land and Main biomass carbon in the farm, indicative of good soil health. Todapur block and area under green house experiment had low soil MBC of 20-150 mg kg-1 . Soils of eastern corner (fallow area of Ento-Patho block) & northern corner of the farm had dehydrogenase activity (DHA) of 10 - 17.2  $\mu$ g TPF g<sup>-1</sup> hr<sup>-1</sup>, indicative of best soil health in the farm with respect to physiologically active soil microorganisms. Soils of western fringe (sewage irrigated area), northern area of the farm (Main block 16-17 and Shadipur orchard), eastern fringe and forest area had DHA of  $7.5$  - 10 µg TPF  $g^{-1}$  hr<sup>-1</sup>. Beside, the soils of northern part of main block and middle block; Genetic block, sewage irrigated area, Top block, PFDC area, Todapur orchard, NBPGR and Paddock field had dehydrogenase activity of 5-10 µg TPF g-1 hr-1 . Dehydrogenase activity in soils of rest of the farm area (Southern part of main block and middle block and Todapur area) ranged from 1.2 to 5.0  $\mu$ g TPF g<sup>-1</sup> hr<sup>-1</sup>.

<b>Soil attributes</b>	Mean prediction error	Root mean square error	Average standard error	Mean Standardized Error	Root Mean square <b>Standardized</b> error
MBC	$-0.3686$	81.4519	77.8693	$-0.0043$	1.0442
Dehydrogenase Activity	$-0.0358$	2.8335	2.7769	$-0.0105$	1.0174

**Table 4. Evaluation performance of kriged map for MBC & DHA through cross validation**





The prediction map of soil MBC and DHA at ICAR-IARI farm, New Delhi through ordinary kriging interpolation technique was evaluated (Table 4) through cross validation approach by several error measurements [56]. Estimation of soil MBC within the farm showed unbiased as mean standardized error (MSE= -0.0043) is near to zero and root mean square standardized error (RMSSE = 1.0442) is closer to one. Prediction of soil DHA through ordinary kriging interpolation was unbiased as MSE (-0.0105) are closer to zero and has good accuracy as indicated by RMSEE value (1.0174) closer to one. Average standard errors (ASE) of prediction for soil MBC and DHA are closer to its root mean square error (RMSE) value.

Farm scale variability of soil microbial properties and processes would be significantly controlled by soil properties such as organic matter content, pH values or texture [42,57]. Besides, the local changes of soil moisture [58], temperature, concentration of soil nutrients [59,60], substrate availability, root biomass, composition and activity of soil microorganisms are the most important factors affecting the soil biological properties. This can partially explain the differences in the spatial variability of properties in this study area. The external anthropogenic factors like different crop management practices, tillage operation, application of fertilizers/amendments etc, created in more variability of sensitive biological variables like MBC & DHA than in physicochemical ones [41,61] as the farm is being cultivated with diversified land use system, crop rotation, orchard, natural plantation etc. Besides, local differences of soil compactness, changed by tillage practices, may affect air-water conditions, which in turn affected soil microbial biomass and activity [53].

# **4. CONCLUSION**

Soil MBC at the farm ranged from 19.7 to 519.7  $\mu$ g g<sup>-1</sup> with average value of 197.7  $\mu$ g g<sup>-1</sup>. Soil dehydrogenase activity (DHA) had mean value of 6.2  $\mu$ g TPF g<sup>-1</sup> hr<sup>-1</sup> with ranging between 1.2 to 17.2  $\mu$ g TPF g<sup>-1</sup> hr<sup>-1</sup>. Microbial biomass carbon and dehydrogenase activity at the farm showed high spatial data variability. Soil organic carbon content, clay content, soil moisture content and other soil property significantly influenced soil microbial biomass carbon content and dehydrogenase activity. Both soil MBC & DHA showed exponential spatial structure at the farm and practical spatial range of soil MBC & DHA

were 1500 m and 1473 m respectively with weak & moderate spatial dependency respectively. In bulk of the IARI farm soils had SMBC within 150- 250 ug  $q^{-1}$  and DHA within 1.2 to 10 ug TPF  $q^{-1}$ dry soil hr<sup>-1</sup>.

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# **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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