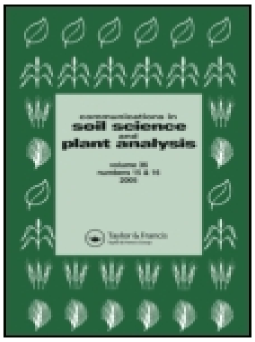

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Walkley-Black Recovery Factor to Reassess Soil Organic Matter: Indo-Gangetic Plains and Black Soil Region of India Case Studies

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Maintenance of soil organic carbon (SOC) stock is important for monitoring soil health, which appears to be fragile in view of the reported climatic changes due to global warming in tropical countries such as India. This requires accurate and reproducible measurement of SOC. The wet-digestion technique following the Walkley-Black (WB) method to determine SOC has been used throughout the world in soil science, agronomy, and environmental science laboratories. WB suggested a universal correction factor to convert organic carbon to calculate exact quantity of organic carbon present in soil assuming 77% recovery of SOC. We understand that such a blanket recommendation may not hold well in different bioclimatic systems and for soils representing various depths. We present corrected Walkley-Black recovery factors ($WBRF_c$) for different bioclimates and soil depths in two food-growing zones in India.

Keywords Bioclimatic system, climate change, corrected Walkley-Black recovery factor, soil depth, Walkley-Black method

Introduction

Organic matter improves the physical and chemical environment of the soil by holding moisture and thus supporting plant growth. It supplies plant nutrients, the release of which depends upon microbial activity. The term *soil organic carbon (SOC)* encompasses the nonmineral fractions of soil, and any vegetation or animal matter forming part of the sample analyzed is included during its determination in the laboratory. In practice, macro-

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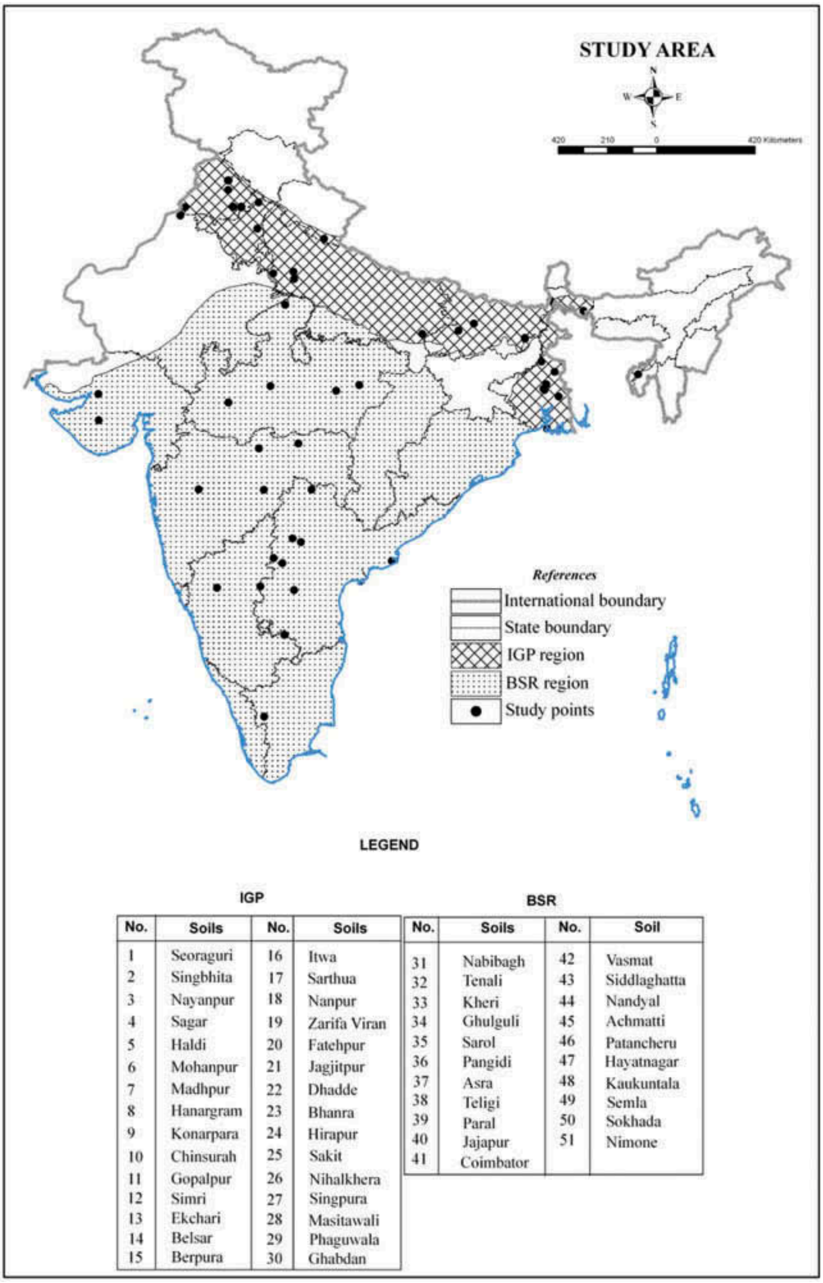
organic matter is largely excluded by fine grinding and sieving of the soil sample. The analytical results, therefore, depend upon the mean size of sieve used in the preparatory stage. Total soil organic matter is estimated by measuring organic carbon content in soils using special techniques. Humus forms most of such organic materials and has been defined as a relatively stable, dark-colored substance. The variable chemical composition of the soil organic matter further complicates its assessment (Hesse 1971). The variation in organic-matter composition is influenced by climate and other parameters. The climatic parameters (viz. temperature and rainfall) exert significant influence on the organic-matter content. Diurnal variation in temperature influences the type and quality of vegetation in a bioclimatic zone. Generally, soils in bioclimates with greater rainfall will contain more organic matter due to enhanced net primary production followed by enhanced microbial activity.

Various methods are available to determine SOC in the soil, which includes the wet-oxidation method. In this method the soil organic matter, which contains about 48–58% of organic carbon, is oxidized by chromic acid utilizing the heat of reaction as well as dilution of sulfuric acid (Walkley and Black 1934; Jackson 1973). The method claims to estimate organic carbon to an extent of 77%; as to make it 100%, it is multiplied with the correction factor of 1.29. This factor is known as the Walkley-Black recovery factor (WBRF) and this varies among the soils, their management regimes, and even soil horizons (Tabatabai 1996). Differences in the type of management systems could also result in the variations in recovery of organic carbon (C) with the Walkley-Black (WB) method. Diaz-Zorita (1999) recovered 15% less organic C under pasture than agricultural systems due to the greater amounts of chemically stable organic compounds in pasture residues than in crop residues. Correction factors of 1.69 for brown forest soils (Typic Hapludolls) in Graminae pasture systems and 1.35 for agriculture management soils were accordingly proposed. Another method is the dry-combustion technique (Yeomans and Bremner 1991) in which the sample is heated to elevated temperature and the carbon dioxide (CO_2) gas evolved is measured with the instrument carbon/nitrogen (C/N analyzer) which helps to find out the total N and total C. A variety of studies over the past 50 years considered the justification of percent of recovery of SOC in the WB method to the extent of 77%. Because this recovery is influenced by vegetation cover, climatic factors, organic-matter composition, depth in the profile, amount of organic matter, and degree of decomposition, the WBRF ceases to a single and universal constant. Keeping this in view, the present study was undertaken to use horizon-wise soils from different bioclimatic systems representing the Indo-Gangetic Plains (IGP) and Black Soil Region (BSR) to arrive at an acceptable WBRF—the corrected WBRF ($WBRF_c$)—that could be used by various soil laboratories and others for soils of similar bioclimate and depth ranges. This will not only help to determine SOC by the WB method correctly but also help reassess SOC stock at regional and national levels.

Materials and Methods

Study Area

The study was conducted in different bioclimatic zones. A total of 526 soil samples (303 of IGP and 223 of BSR) were selected from the IGP and BSR (Figure 1). The IGP ranks as one of the most extensive fluvial plains of the world. The deposit of this tract represents the last chapter of the earth's history. It came into existence due to the collision of the Indian and Chinese Plates during Middle Miocene (Prakash and Kumar 1991). The IGP covers about 52.01 mha and represents eight agroecological regions (AERs), fourteen

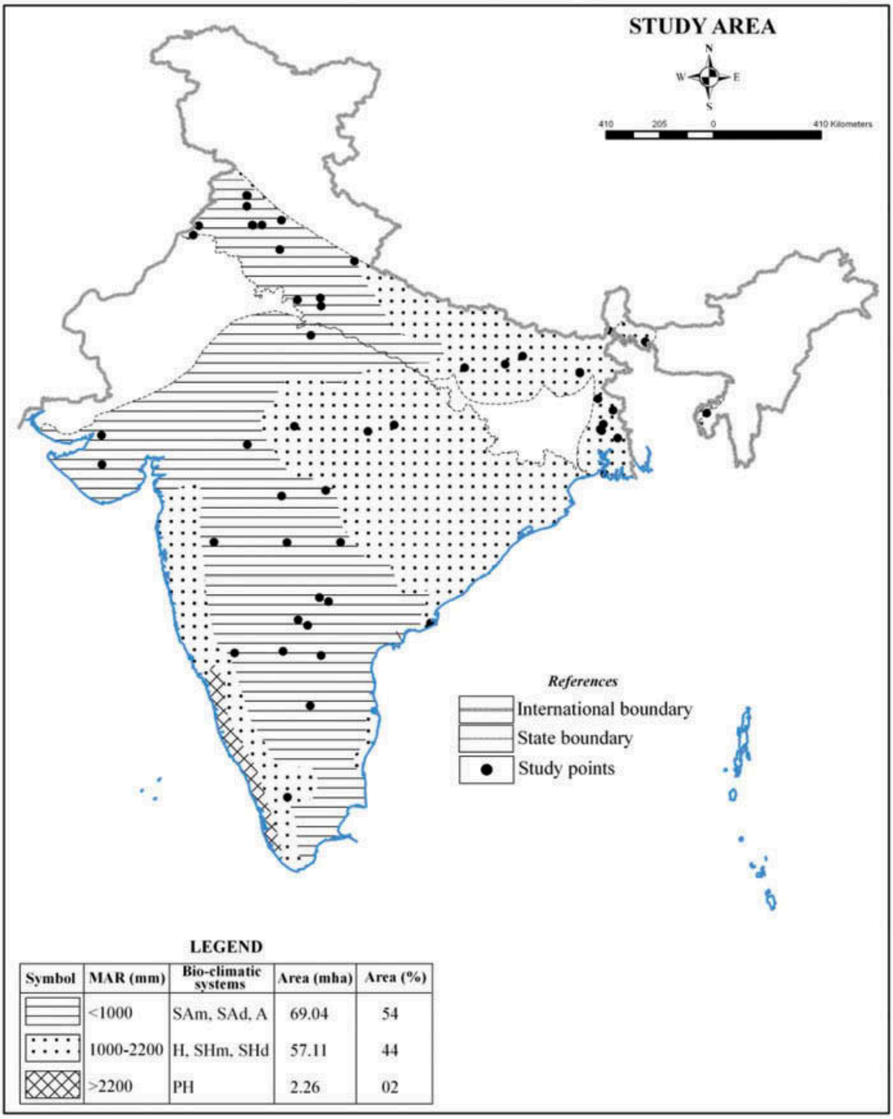


No.	Soils	No.	Soils
1	Seoraguri	16	Itwa
2	Singbhita	17	Sarthua
3	Nayanpur	18	Nanpur
4	Sagar	19	Zarifa Viran
5	Haldi	20	Fatehpur
6	Mohanpur	21	Jagjitpur
7	Madhpur	22	Dhadde
8	Hanargram	23	Bhanra
9	Konarpara	24	Hirapur
10	Chinsurah	25	Sakit
11	Gopalpur	26	Nihalkhera
12	Simri	27	Singpura
13	Ekchari	28	Masitawali
14	Belsar	29	Phaguwala
15	Berpura	30	Ghabdan

No.	Soils	No.	Soil
31	Nabibagh	42	Vasmat
32	Tenali	43	Siddlaghatta
33	Kheri	44	Nandyal
34	Ghulguli	45	Achmatti
35	Sarol	46	Patancheru
36	Pangidi	47	Hayatnagar
37	Asra	48	Kaukuntala
38	Teligi	49	Semla
39	Paral	50	Sokhada
40	Jajapur	51	Nimone
41	Coimbator		

Figure 1. Study area showing different benchmark spots in Indo-Gangetic plains (IGP) and Black Soil Regions (BSR).

agroecological sub regions (AESRs) (Mandal et al. 2014), and seven bioclimatic systems (Figure 2). The nature and properties of the alluvium vary in texture from sandy to clayey, calcareous to noncalcareous, and acidic to alkaline. Though the overall topographic situation remains fairly uniform with elevations of 150 m above mean sea level in Bengal basin and 300 m above mean sea level in the Punjab plain, local geomorphic



Symbol	MAR (mm)	Bio-climatic systems	Area (mha)	Area (%)
▤	<1000	SAm, SAd, A	69.04	54
⁙	1000-2200	H, SHm, SHd	57.11	44
▦	>2200	PH	2.26	02

Figure 2. Study area showing distribution of bioclimatic systems in the IGP and BSR. (Total area is 128.41 including 52.01 mha for IGP and 76.4 mha for BSR.)

variations are significant (Shankaranarayana 1982). In the IGP, the rice-wheat cropping system is dominant, followed by cotton-wheat, bajra-wheat, and maize-wheat.

Black soils are common in the semi-arid tropics (SAT) in India, although their presence has been reported in the humid and arid bioclimates also (Bhattacharyya, Pal, and Deshpande 1993; Bhattacharyya et al. 2008a, b). These soils are spatially associated with red soils and form a major soil group of India. They occur on various parent materials and climates. These soils occur in various physiographic positions such as red soils in the hills and black soils in the valleys in Maharashtra and Madhya Pradesh (Bhattacharyya, Pal, and Deshpande 1993; Bhattacharyya and Pal 1998; Bhuse, Ray, and Bhattacharyya 2002). Interestingly these soils have also been reported in juxtaposition in the states of Tamil Nadu, Maharashtra, and Andhra

Pradesh (Pal and Deshpande 1987; Pal 1988). Exactly the opposite situation was found in Tamil Nadu where red soils are in the valleys and black soils on the hills (Bhuse 2000). The black soils (Vertisols and their intergrades) represent a wide area, named the Black Soil Region (BSR) of India. These soils are potentially huge crop production zones in the country. These are extensively spread in the states of Uttar Pradesh, Madhya Pradesh, Gujarat, Rajasthan, Maharashtra, Andhra Pradesh, Tamil Nadu and Karnataka. Reports of Vertisols and their intergrades occur in many other states; their total acreage is 76.4 mha covering thirty agro-eco-subregions (Mandal et al. 2014; Velayutham et al. 1999; Bhattacharyya et al. 2007) and five bioclimatic systems (Figure 2). A total of 223 samples were selected for the present study. The dominant crops in the BSR are cotton and soybean, followed by sorghum and wheat.

Methods

The SOC was determined by the WB method (Walkley and Black 1934). Calcium carbonate equivalent in soils was determined following the standard methods (Jackson 1973). Soil inorganic carbon (SIC) constitutes 12% of calcium carbonate ($CaCO_3$). Prior to analysis, the soil samples were air dried and ground to 100 mesh. The Walkley and Black method was developed in 1934 (Walkley and Black 1934), based on the Schollenberger method and further refined (Walkley 1947). Essentially, concentrated sulfuric acid (H_2SO_4) was added to a mixture of soil and aqueous potassium dichromate ($K_2Cr_2O_7$). The heat of dilution raises the temperature sufficiently to induce a substantial but not complete oxidation by the acidified dichromate. Residual dichromate was titrated using ferrous sulfate solution. The difference between the sample titrated by ferrous ammonium sulfate [$Fe\ (NH_4)_2(SO_4)_2 \cdot 6H_2O$] (FAS) and that of the blank titration determined the amount of easily oxidizable organic carbon.

The percentage Walkley-Black carbon (WBC) is given by the formula

$$\text{WBC}\ (\%) = V\ (B - T) / B \times 0.3 / W \times (100 + m) / 100 \times \text{WBRF}$$

where V is the volume of 1 N $K_2Cr_2O_7$ solution, B is the volume of FAS reagent for blank, T is the volume of FAS reagent for soil sample, m is the air dry moisture percentage of the soil sample used, and WBRF is the Walkley-Black recovery factor of 1/0.77.

The correction factor (CF) is a compensation for the incomplete oxidation and is the inverse of recovery. This CF was set to 1.29 (recovery of 77%) (Walkley and Black 1934) for determining SOC using the dry combustion method by C/N analyzer, where total carbon (TC) is obtained. The SOC was the difference between TC and soil inorganic carbon (SIC). For TC analysis, the sample was heated to 900 °C, and the CO_2 gas evolved was detected by infrared (IR) radiator by a C/N analyzer. All the samples were analyzed in triplicate, allowing us to develop a dataset of more than 1500 samples.

Determination of $CaCO_3$ to calculate the SIC was done by treating the soil with a known volume of acid to neutralize all carbonates and the excess acid was back titrated with a standard alkali solution with the help of bromothymol blue or phenolphthalein as an indicator. The corrected Walkley-Black recovery factor ($WBRF_c$) was estimated using wet (Walkley-Black method) and dry combustion (C/N analyzer) methods using the following equations:

$$\text{TC}_m = \text{Walkley Black} + \text{SIC}_l \tag{1}$$

$$\text{C/N} = \text{TC}_m - \text{SIC}_l \tag{2}$$

$$\mathrm{SOC_w} = (\text{Walkley Black})^{*}77/100 \text{ (assuming 77\% recovery)} \tag{3}$$

$$\mathrm{WBRF_c} = \mathrm{C/N/SOC_w} \tag{4}$$

where TC_m is total carbon obtained by C/N analyzer, Walkley Black is soil organic carbon obtained by the Walkley and Black method, SIC_l is soil inorganic carbon obtained by laboratory method (Bhattacharyya et al. 2008b), C/N is soil organic carbon obtained by the C/N analyzer [from Eq. (2)], and SOC_w is soil organic carbon without using WBRF.

RESULTS AND DISCUSSION

Indo-Gangetic Plains (IGP)

The SOC values determined by dry combustion techniques (C/N analyzer, henceforth mentioned as C/N) mostly differ from those of the WB method (henceforth mentioned as Walkley-Black) when averaged over sampled layers (Table 1). The values of soil inorganic carbon (SIC) is provided for comparing the difference between C/N and Walkley-Black method and the laboratory-drawn SIC values. Comparison of SOC determined by the WB method (Walkley-Black) and C/N analyzer (C/N) for various depths show that SOC decreased with depth in all the profiles (Figures 3–6). The recovery percentage and the WBRF (i.e., 77% and 1.29, respectively) in all the bioclimatic zones are not similar. The recovery percentage and WBRF obtained for different bioclimatic zones of the IGP show that the recovery percentage decreases with depth in all the bioclimatic zones.

WBRFc in Different Bioclimatic Systems

The culturable microbial community composition in soils varies within each representative soils. Soil heterotrophic aerobic microbial communities decline with increasing soil depth. The maximum microbial communities are restricted within the first 30 cm of soil. With every increase in 30 cm in depth, an approximately 10% decrease in Shannon's diversity index was observed (Lupwayi, Arshad, and Rice 2001). Moreover, nearly 90% decline in microbial diversity was observed from surface to subsurface soils (Srivastava et al. 2014). These authors reported significant differences in microbial population among different bioclimates of the IGP. Greater bacterial and fungal populations in subhumid to humid climates were also reported by them. Rainfall is an important physical parameter that directly influences the microbial population through soil water precipitation and moisture retention. All these facts indicate that the distribution of rainfall in different bioclimatic systems and the depth of soil controlling microbial population in the IGP influence decomposition of organic matter, and this fact is reflected in greater recovery (percentage) in the surface soils of wetter climate as 87% in perhumid to 78% in semi-arid and arid systems (Table 2). Because recovery (%) varies with the bioclimateic systems it brings variation in the recovery factors (Table 2). This demands necessary corrections in the laboratory manuals. The WBRFc of 1.15, 1.23, and 1.28 for three different bioclimatic systems such as perhumid, humid, and subhumid systems, and semi-arid and arid systems, respectively, for 0- to 30-cm soil depth can thus be recommended as an acceptable correction factor to revise the organic carbon

Table 1
Soil organic and inorganic carbon in different rainfall zones of the Indo-Gangetic plains

				Walkley-Black[d]				C/N[e]				SIC[f]			
Soils	AESR[a]	Bioclimate[b]	MAR[c] (mm)	0–30	30–50	50–100	100–150	0–30	30–50	50–100	100–150	0–30	30–50	50–100	100–150
Mean annual rainfall > 2200 mm															
Seoraguri	15.3	PH	3292	0.68*******	0.36	0.50	0.37	0.59	0.36	0.46	0.39	0.09	0.09	0.10	0.11
Singbhita	15.3	PH	3390	1.30	0.78	0.44	0.38	1.23	0.80	0.45	0.45	0.12	0.12	0.13	0.11
Nayanpur	15.3	PH	2178	0.56	0.54	0.56	0.50	0.32	0.44	0.51	0.55	0.22	0.25	0.23	0.22
Mean annual rainfall 1000–2200 mm															
Sagar	18.5	H	1783	0.58	0.27	0.24	0.26	0.51	0.31	0.34	0.45	0.18	0.22	0.16	0.19
Haldi	13.2	H	1700	0.90	0.57	0.35	0.21)	0.76	0.48	0.31	0.39	0.15	0.16	0.14	0.26
Mohanpur	15.1	H	1620	0.84	0.50	0.39	0.21	0.65	0.40	0.33	0.18	0.11	0.09	0.12	0.14
Hangram	15.1	SHm	1400	0.93	0.35	0.33	0.26	0.74	0.34	0.35	0.27	0.16	0.19	0.16	0.12
Konarpara	15.1	SHm	1400	0.71	0.34	0.42	0.27	0.51	0.43	0.41	0.24	0.08	0.09	0.09	0.11
Gopalpur	15.1	SHm	1350	0.59	0.17	0.10	0.07	0.51	0.19	0.17	0.14	0.21	0.28	0.28	0.29
Madhpur	15.1	SHm	1338	0.44	0.14	0.09	0.09	0.38	0.16	0.11	0.13	0.12	0.28	0.50	0.83
Chinsurah	15.1	SHm	1338	1.07	0.54	0.52	0.47	0.84	0.63	0.62	0.59	0.17	0.22	0.26	0.28
Berpura	9.1	SHd	1125	0.53	0.20	0.20	0.25	0.50	0.24	0.28	0.45	0.13	0.15	0.13	0.14
Ekchari	13.1	SHd	1105	0.41	0.16	0.10	0.06	0.34	0.18	0.11	0.12	0.30	0.29	0.28	0.22
Belsar	13.1	SHd	1105	0.26	0.12	0.06	0.02	0.24	0.13	0.07	0.06	0.19	0.19	0.20	0.20
Itwa	9.2	SHd	1005	0.37	0.15	0.11	0.07	0.39	0.19	0.15	0.13	0.13	0.15	0.17	0.17
Simri	9.2	SHd	1003	0.52	0.32	0.29	0.26	0.41	0.28	0.25	0.21	0.09	0.15	0.16	0.16
Sarthua	13.1	SHd	1003	0.55	0.36	0.29	0.25	0.62	0.48	0.40	0.42	0.10	0.22	0.25	0.24
Mean annual rainfall < 1000 mm															
Nanpur	13.1	Sam	870	0.63	0.33	0.27	0.24	0.67	0.43	0.39	0.39	1.27	1.18	1.91	1.87
Hirapur	10.2	SAd	782	0.30	0.15	0.18	0.18	0.17	0.07	0.07	0.38	0.31	0.26	1.22	1.61
Sakit	4.3	SAd	782	0.33	0.06	0.06	0.03	0.29	0.09	0.09	0.05	0.22	0.21	0.24	0.26

Bhanra	4.1	SAd	750	0.64	0.17	0.11	0.10	0.90	0.27	0.22	0.20	0.34	0.23	0.19	0.20
Fatehpur	4.1	SAd	734	0.43	0.21	0.21	0.21	0.52	0.23	0.22	0.25	0.12	0.10	0.15	0.26
Jagjitpur	9.1	SAd	734	0.59	0.38	0.29	0.25	0.46	0.30	0.27	0.22	0.19	0.14	0.10	0.18
Singhpura	4.4	SAd	725	0.37	0.26	0.21	0.18	0.34	0.26	0.22	0.33	0.19	0.20	0.39	0.84
Zarifaviran	4.1	SAd	705	0.49	0.19	0.14	0.14	0.26	0.21	0.17	0.17	0.41	0.39	0.29	0.51
Dhadde	9.1	SAd	681	0.91	0.48	0.13	0.11	0.45	0.18	0.11	0.09	0.15	0.18	0.24	0.25
Nihalkhera	2.3	A	550	0.39	0.27	0.26	0.22	0.38	0.48	0.45	0.28	0.60	0.70	1.12	1.28
Phaguwala	4.1	A	435	0.60	0.19	0.20	0.07	0.49	0.20	0.29	0.44	0.42	0.72	1.23	1.22
Ghabdan	4.1	A	435	0.47	0.23	0.22	0.33	0.65	0.41	0.42	0.40	0.38	0.36	0.25	0.21
Masitawali	2.3	A	221	0.63	0.30	0.20	0.20	0.47	0.29	0.22	0.29	0.24	0.28	0.54	0.89

[a] Agroecological subregions: PH, perhumid; H, humid; SHm, subhumid moist; SHd, subhumid dry; SAm, semi-arid moist; SAd, semi-arid dry; A, arid.

[b] Bioclimate, MAR ranges: perhumid, >2200 mm; humid, 1600–2200 mm; subhumid moist, 1200–1600 mm; subhumid dry, 1000–1200 mm; semi-arid moist, 850–1000 mm, semi-arid dry, 550–850 mm; arid, <550 mm.

[c] Mean annual rainfall.

[d] Soil organic carbon obtained by Walkley and Black method.

[e] Soil organic carbon obtained by (C/N analyzer).

[f] Soil inorganic carbon in the form of calcium carbonate.

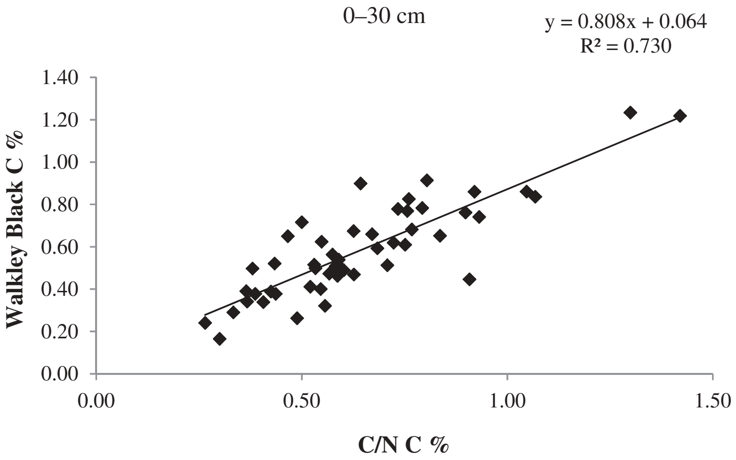


Figure 3. Comparison of SOC determined by the Walkley-Black method and C/N analyzer in the Indo-Gangetic Plains (IGP) and Black Soil Region (BSR) (0–30 cm deep).

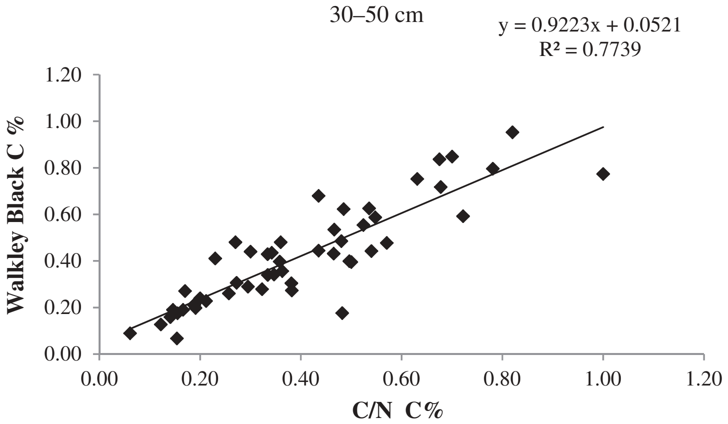


Figure 4. Comparison of SOC determined by the Walkley-Black method and C/N analyzer in the Indo-Gangetic Plains (IGP) and Black Soil Region (BSR) (30–50 cm deep).

determined traditionally in the laboratory. This will portray a true picture of actual organic carbon content in the soils of the IGP.

WBRFc in Different Soil Depths

As mentioned in the previous section, microbial population controlling organic-matter decomposition reduces with increase in soil depth (Srivastava et al. 2014). Therefore, the recovery (%) of organic matter will vary with soil depth resulting in different $WBRF_c$ in the same bioclimatic systems (Table 2). As expected in the IGP, the $WBRF_c$ varied from 1.15 to 2.09 for different bioclimatic zones in various depths. The $WBRF_c$ are 1.15, 1.25, 1.31, and 1.72 in soil depths of 0–30, 30–50, 50–100, and 100–150 cm respectively where the mean annual rainfall (MAR) is >2200 mm. In the region where the MAR is

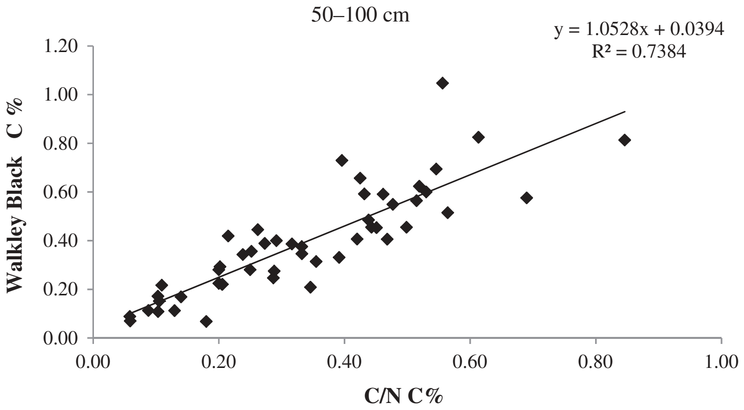


Figure 5. Comparison of SOC determined by the Walkley-Black method and C/N analyzer in the Indo-Gangetic Plains (IGP) and Black Soil Region (BSR) (50–100 deep).

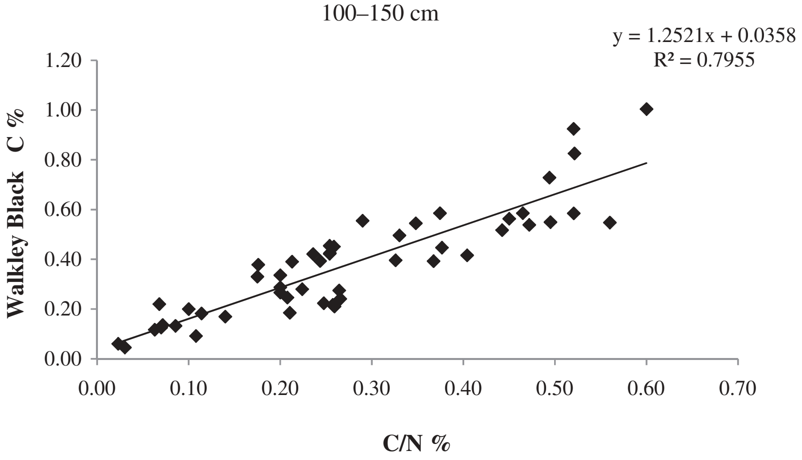


Figure 6. Comparison of SOC determined by the Walkley-Black method and C/N analyzer in the Indo-Gangetic Plains (IGP) and Black Soil Region (BSR) (100–150 cm deep).

1000–2200 mm, WBRF$_c$ values are 1.23, 1.43, 1.57, and 2.03 in depths of 0–30, 30–50, 50–100, and 100–150 cm respectively. In the drier part of IGP (MAR < 1000 mm) the WBRF$_c$ values are 1.28, 1.49, 1.65, and 2.09 in depths of 0–30, 30–50, 50–100, and 100–150 cm respectively.

The recovery of organic matter in the IGP varies from 58 to 87% in the regions where MAR is >2200 mm. In areas where MAR is 1000–2200 mm the recovery of organic matter varies from 49 to 81% and in MAR < 1000 mm the recovery varies from 48 to 78% (also see Figures 3–6).

Black Soil Regions

The SOC values determined by dry combustion techniques (C/N analyzer) mostly differ from those of the WB method in each zones (Table 3). The values of soil inorganic carbon

Table 2
Recovery and corrected Walkley-Black recovery factor ($WBRF_c$) for soils of the Indo-Gangetic Plains and Black Soil Regions

			Recovery (%) in soil depth (cm)				$WBRF_c$ in soil depth (cm)			
No.	Bioclimatic zones	MAR (mm)	0–30	30–50	50–100	100–150	0–30	30–50	50–100	100–150
Indo-Gangetic Plains										
1.	Perhumid	>2200 mm	87	80	76	58	1.15	1.25	1.31	1.72
2.	Humid and Subhumid	1000–2200 mm	81	70	64	49	1.23	1.43	1.57	2.03
3	Semi-arid and Arid	<1000 mm	78	67	61	48	1.28	1.49	1.65	2.09
Black Soil Region										
4.	Subhumid	1000–2200 mm	78	70	63	54	1.29	1.42	1.58	1.86
5.	Semi-arid and Arid	<1000 mm	76	69	63	51	1.31	1.44	1.60	1.97

Table 3
Soil organic and inorganic carbon in different rainfall zones of black soil regions

				Walkley-Black[d]				C/N[e]				SIC[f]			
Soil	AESR[a]	Bioclimate[b]	MAR[c] (mm)	0–30	30–50	50–100	100–150	0–30	30–50	50–100	100–150	0–30	30–50	50–100	100–150
Mean annual rainfall 1000–2200 mm															
Kheri	10.1	SHm	1440	0.59	0.44	0.45	0.47	0.54	0.44	0.45	0.54	0.43	0.51	0.46	0.41
Tenali	7.3	SHm	1250	0.79	0.48	0.43	0.35	0.78	0.62	0.59	0.54	0.46	0.45	0.47	0.43
Nabibagh	10.1	SHm	1209	0.63	0.37	0.31	0.29	0.89	0.58	0.49	0.46	0.08	0.07	0.06	0.02
Ghulguli	10.3	SHd	1100	0.76	0.63	0.43	0.37	0.83	0.75	0.66	0.59	0.16	0.16	0.14	0.14
Sarol	5.2	SHd	1100	0.55	0.38	0.35	0.26	0.40	0.27	0.21	0.22	0.56	0.60	0.59	0.31
Pangidi	6.2	SHd	1071	1.05	1.00	0.85	0.60	0.86	0.77	0.81	1.00	0.76	0.77	0.68	0.67
Mean annual rainfall <1000 mm															
Asra	6.3	SAm	975	0.80	0.68	0.61	0.52	0.91	0.84	0.82	0.83	0.81	0.90	0.93	0.95
Paral	6.3	SAd	794	0.57	0.52	0.51	0.52	0.56	0.55	0.56	0.58	0.75	0.73	0.66	0.64
Jajapur	7.2	SAd	792	0.38	0.30	0.25	0.20	0.50	0.44	0.36	0.34	0.53	0.62	0.90	1.35
Vasmat	6.2	SAd	789	0.77	0.48	0.44	0.52	0.68	0.48	0.48	0.92	1.25	1.20	1.15	1.53
Patancheru	7.2	SAd	764	1.42	0.82	0.55	0.24	1.22	0.95	0.69	0.42	0.06	0.08	0.08	0.11
Hayatnagar	7.2	SAd	764	0.92	0.68	0.53	0.45	0.86	0.72	0.60	0.56	0.05	0.07	0.12	0.08
Kaukuntala	7.2	SAd	674	0.72	0.47	0.33	0.20	0.62	0.43	0.38	0.27	0.11	0.07	0.09	0.12
Siddlaghatta	8.2	SAd	661	0.53	0.36	0.25	0.11	0.52	0.40	0.28	0.18	0.26	0.32	0.35	0.45
Nandyal	7.1	SAd	650	0.42	0.33	0.32	0.33	0.39	0.34	0.39	0.50	0.75	0.75	0.77	0.85
Achmati	6.4	SAd	638	0.75	0.72	0.69	0.56	0.61	0.59	0.58	0.55	0.57	0.57	0.62	0.66
Semla	5.1	SAd	635	0.76	0.70	0.56	0.49	0.77	0.85	1.05	0.73	1.83	1.91	1.79	1.97
Teligi	3.1	SAd	632	0.67	0.47	0.46	0.44	0.66	0.53	0.59	0.52	0.82	0.83	0.84	1.14
Coimbatore	8.1	SAd	612	0.57	0.50	0.47	0.40	0.47	0.40	0.41	0.42	0.86	0.85	0.81	0.45
Sokhada	5.3	A	533	0.50	0.44	0.40	0.29	0.72	0.68	0.73	0.56	1.91	1.92	1.90	1.94
Nimone	6.1	A	520	0.73	0.55	0.48	0.25	0.78	0.59	0.55	0.42	1.50	1.53	1.65	1.68

[a] Agroecological subregions: PH, perhumid; H, humid; SHm, subhumid moist; SHd, subhumid dry; SAm, semi-arid moist; SAd, semi-arid dry; A, arid.

[b] Bioclimate, MAR ranges: perhumid, >2200 mm; humid, 1600–2200 mm; subhumid moist, 1200–1600 mm; subhumid dry, 1000–1200 mm; semi-arid moist, 850–1000 mm, semi-arid dry, 550–850 mm; arid, <550 mm.

[c] Mean annual rainfall.

[d] Soil organic carbon obtained by Walkley and Black method.

[e] Soil organic carbon obtained by (C/N analyzer).

[f] Soil inorganic carbon in the form of calcium carbonate.

(SIC) are provided for comparing the difference between C/N and Walkley-Black method and the laboratory-drawn SIC values. The recovery percentage and recovery factor obtained for different bioclimatic zones of BSR are shown in Table 2.

WBRFc in Different Bioclimatic Systems

As reported for the soils in the IGP, soils of BSR also indicate influence of bioclimate and soil depth on culturable microbial population (Velmourougane et al. 2014). In the surface horizon, soils in the subhumid moist system recorded greater culturable microbial population, whereas those in the arid regions showed the least. The variations in microbial populations among the bioclimatic systems were attributed to the differences in soil physical and chemical properties. Besides bioclimatic system, depth of soils is also an important factor for the variation in microbial population. Surface soil horizon recorded maximum population and almost 50% of the microbial population is restricted to the 0- to 30-cm depth only (Velmourougane et al. 2014).

These studies, carried out on the basis of nearly 200 samples, support our observation that indicates marginally high recovery percentage in a subhumid bioclimate in black soils than in semi-arid and arid systems (Table 2). Our study suggests $\mathrm{WBRF_c}$ values of 1.29 for subhumid and 1.31 for both semi-arid and arid bioclimatic systems to be recommended for calculating exact SOC using the WB method. On a comparative note, under similar rainfall conditions, IGP soils permit more recovery of organic matter than the soils of BSR by the WB method, possibly due to the fact that the IGP soils have greater labile pool compared to the BSR soils (Chivane and Bhattacharyya 2010).

WBRFc in Different Soil Depths

In the BSR, $\mathrm{WBRF_c}$ varied from 1.29 to 1.97 for the different bioclimatic zones in various depths (Figures 3–6). The $\mathrm{WBRF_c}$ values were 1.29, 1.42, 1.58, and 1.86 in soil depths of 0–30, 30–50, 50–100 and 100–150 cm respectively where the MAR is 1000–2200 mm. In the drier ecosystems (MAR < 1000 mm), these factors were 1.31, 1.44, 1.60, and 1.97 in depths of 0–30, 30–50, 50–100, and 100–150 cm respectively. Consequently, the recovery of organic matter varies from 54 to 78% and 51 to 76% in the regions where the MAR is 1000–2200 mm and <1000 mm respectively. The recovery percentage significantly varied in all the bioclimatic regions. The relation between mean annual rainfall and recovery factor in various depths in the IGP and BSR is shown in Figures 7–10.

The recovery of soil organic matter appears to be less than the standard value of 77% in soils of both the IGP and BSR for a common depth of 100–150 cm, assuming 1.29 as the widely accepted WBRF, considering 77% recovery of organic matter during the wet combustion method. It seems that in the semi-arid and arid tracts of the IGP (<1000 mm MAR), the $\mathrm{WBRF_c}$ is close to the value proposed by Walkley and Black (1934) (Table 2). It is understood that due to increase in rainfall and with the proliferation of microbial activities the rate of decomposition of organic matter increases, bringing more humus (highly decomposed organic material) in soils. This results in an increased oxidizable organic matter, which results in a decline of WBRFc. Our observations of relatively low recovery factors in humid and subhumid to perhumid bioclimatic zones cutting across different depths of soil support this connotation (Table 2). For the BSR, we made similar observations. It is a well-known fact that down the depth, activity of microbes decreases, and this results in low organic-matter accumulation as well as lesser degrees of humification. This is the reason why the quantity of oxidizable organic matter in the subsoils is less as compared to the surface horizons. Cutting across different bioclimatic zones in both the

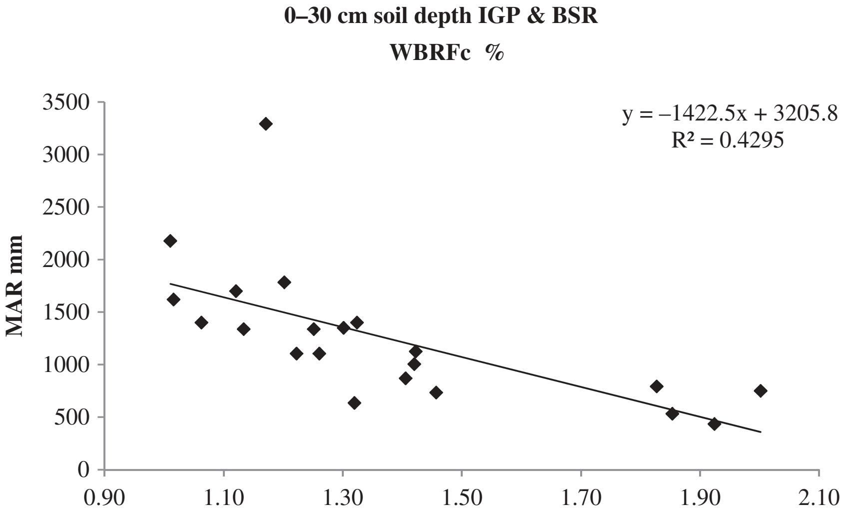


Figure 7. Relation between corrected Walkley-Black recovery factor ($WBRF_c$) and mean annual rainfall (MAR) in the Indo-Gangetic Plains (IGP) and Black Soil Regions (BSR) (0–30 cm deep).

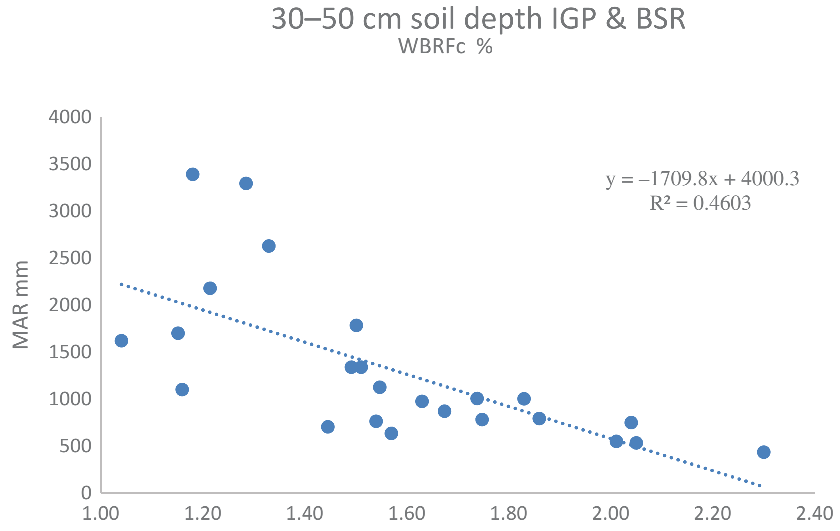


Figure 8. Relation between the corrected Walkley-Black recovery factor ($WBRF_c$) and mean annual rainfall (MAR) in the Indo-Gangetic Plains (IGP) and Black Soil Regions (BSR) (in the x-axis) (30–50 cm deep).

IGP and BSR, WBRFc increases down the depth, with its concomitant decrease in percentage recovery (Table 2).

In the dry areas of the IGP and BSR in surface soils (0–30 cm), the commonly used $WBRF_c$ is acceptable because it is within the range of 1.28–1.31 and is near to the value proposed by Walkley and Black (1934). In high rainfall regions for the subsurface soils the $WBRF_c$ used in the soil-testing laboratory may not work; therefore the present study recommends a revised value of the $WBRF_c$ for both the IGP and BSR as shown in

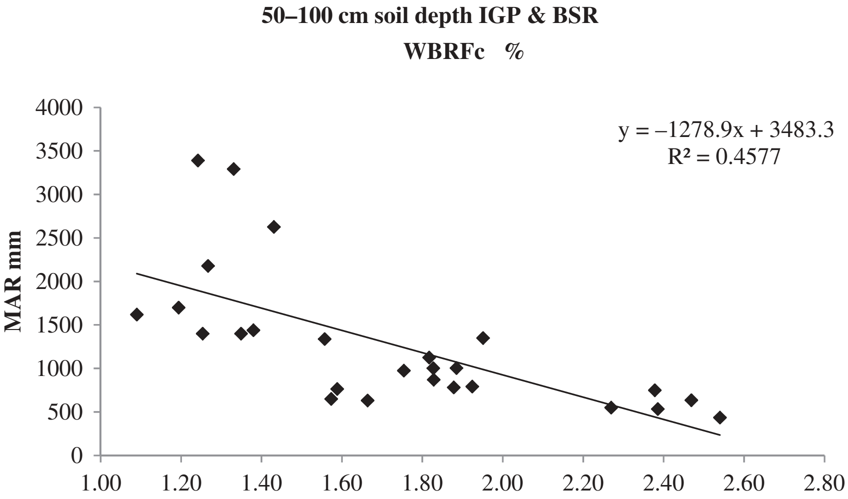


Figure 9. Relation between the corrected Walkley-Black recovery factor ($WBRF_c$) and mean annual rainfall (MAR) in the Indo-Gangetic Plains (IGP) and Black Soil Regions (BSR) (in the x-axis) (50–100 cm deep).

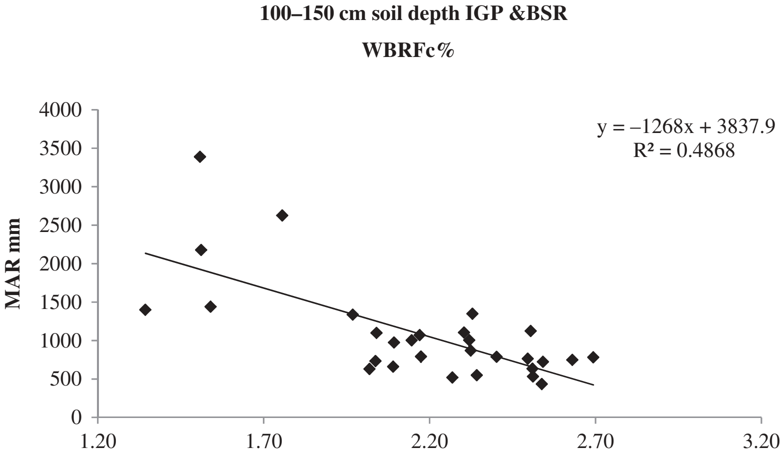


Figure 10. Relation between the corrected Walkley-Black recovery factor ($WBRF_c$) and mean annual rainfall (MAR) in the Indo-Gangetic Plains (IGP) and Black Soil Regions (BSR) (in the x-axis) (100–150 cm deep).

Table 2. The recovery appears to be more than the standard value. The study shows an apparent relationship between increasing depth, decreasing carbon content, and increasing conversion factor. Perhaps the most serious drawback of WB method is that the final estimation of organic carbon requires a correction factor because oxidation of organic matter is incomplete and highly variable. Recovery can change from 27 to 100%, with 77% being a typical value (Nelson and Sommers 1982). Average correction factor for group of soils vary from 1.03 to 1.86 but an average value applied to any given individual soil may result in a substantial error. Incomplete oxidation of carbon makes wet combustion a poor choice for use on soils with high organic-matter content, those containing more than 5% carbon (Soon and Abboud 1991; Collins and Kuehl 2001). These limitations mean that results obtained from

Table 4
Various Walkley-Black recovery factors reported in the literature for surface soils

	Walkely-Black recovery			
No.	Percent	Factor	Reference	Remarks
1.	91.7–44.05	1.09–2.27	Nelson and Sommers (1996)	Dichromate method that uses heat of dilution or minimal heating does not give complete oxidation of organic compounds in soil although the most active form of organic C is converted to CO_2. The average correction factor appropriate for a group of soil varied from 1.19–1.33. $Cr_2O_7^{2-}H_2SO_4$ methods involving minimal heating give variable recovery and require a small correction factor (e.g., 1.15) to account for unreacted organic C.
2.	59–94	1.69–1.06	Allison (1960)	The effectiveness of the wet-combustion procedure described was determined on samples of noncalcareous Illinois soils.
3.	63	1.58	De Vos et al. (2007)	Total organic carbon of 542 forest soil samples were measured with TOC analyzer equipped with a solid sample module and the other with Walkely-Black procedures. For each laboratory and type of soil, it is recommended that specific recovery factors are determined in order to standardize the result.
4.	59	1.69	Diaz-Zorita (1999)	Samples from Graminean perennial pastures.
5.	61	1.63	Mikhailova, Noble, and Post (2003)	For a fine-silty, mixed, frigid Pachic Hapludoll of Russia under cultivated and pasture management regimes.
6.	97	1.03	Olayinka, Adebayo, and Amusan (1998)	For acidic sandy soils in Nigeria.
7.	100	1.00	Rhodes, Kamara, and Sutton (1981) and Chacon et al. (2002)	For tropical soils.

(*Continued*)

Table 4
(*Continued*)

	Walkely-Black recovery			
No.	Percent	Factor	Reference	Remarks
8.	71	1.40	Soon and Abbound (1991)	For colder climate soils.
9.	79	1.26	Navarro et al. (1993)	For calcareous soils.
10.	70–74	1.41–1.35	Hussain and Olson (2000)	For total and mineral organic C analysis.
11.	87	1.15, 1.25, 1.31, 1.72	This paper	0–30, 30–50,50–100, 100–150 cm deep in >2200 mm MAR in the IGP, India[a]
12.	81	1.23, 1.43, 1.57, 2.03	This paper	0–30, 30–50, 50–100, 100–150 cm deep in 1000–2200 mm MAR in the IGP, India[a]
13.	78	1.28, 1.49, 1.65, 2.09	This paper	0–30, 30–50, 50–100, 100–150 cm deep in <1000 mm MAR in the IGP, India[a]
14	78	1.29, 1.42, 1.58, 1.86	This paper	0–30, 30–50, 50–100, 100–150 cm soil deep in 1000–2200 mm MAR in the BSR, India[a]
15.	76	1.31, 1.44, 1.60, 1.97	This paper	0–30, 30–50, 50–100, 100–150 cm soil depth in <1000 mm MAR in the BSR, India[a]

[a] For details please see Tables 1, Table 2, and Table 3.

WB should be considered only an estimate of the amount of an easily oxidizable organic carbon in soils (Nelson and Sommers 1996; Jolivet, Arrouays, and Bernoux 1998). Through some reference it is well established that about 60–86% of SOC is only oxidized, and therefore a standard correction factor of 1.29 (considering recovery of 77%) is used to obtain the corrected SOC value. However, the actual recoveries vary depending upon the site and method used (Allison 1960). The difference in correction factors obtained for the soils of different study areas points out that in addition to the limitation of the WB method itself, the site characteristics including physiography, parent material, soil and climatic conditions and presence of stable organic carbon compounds in the mineral fraction (De Vos et al. 2007) exercise important roles in causing the variability in the underestimation of SOC. Variability in any estimated factor comes from two sources in the method used to estimate organic carbon and the natural variability in the composition of soil. The manual approach adopted in the WB method would have also introduced some errors in SOC estimation. Thus there is a strong need to develop an appropriate correction factor for Indian soils to improve the SOC

Table 5

Variations of Walkley-Black recovery factor in various soil horizons

No.	Depth	Recovery (%)	Factor (mean)	Remark
Soils of the Indo-Gangetic Plains[a]				
1.	0–30	87	1.15	>2200 mm MAR; perhumid bioclimatic system represented by (1) Seoraguri (*Typic Endoaqualfs*). The other soils are Singbhita (*Umbric Endoaqualfs*) and Nayanpur (Typic *Endoaqualfs*).
	30–50	80	1.25	
	50–100	76	1.31	
	100–150	58	1.72	
2.	0–30	81	1.23	1000–2200 mm MAR; humid and subhumid bioclimatic system represented by (2) Madhpur (*Chromic Vertic Endoaqualfs*). The other soils are Haldi (*Typic Haplustalfs*), Hangram (*Vertic Endoaqualfs*), Konarpara (*Vertic Endoaqualfs*), Mohanpur (*Vertic Endoaqualfs*), Sagar (*Vertic Endoaquepts*), Gopalpur *(Chromic Endoaquerts*), Chinsurah (*Typic Endoaqert*), Simri (*Typic Haplustepts*), Ekchari (*Vertic Endoaqualfs*), Belsar (*Aeric Endoaqualfs*), Berpura (*Oxyaquic Haplustalfs*), Itwa (*Typic Endoaqualfs*), and Sarthua (*Vertic Endoaqualfs*).
	30–50	70	1.43	
	50–100	64	1.57	
	100–150	49	2.03	
3.	0–30	78	1.28	<1000 mm MAR; semi-arid and arid bioclimatic system represented by (3) Nanpur (*Fluventic Endoaquepts*). The other soils are Zarifa Viran (*Typic Natrustalfs*), Nihalkhera (*Aridic Haplustalfs*), Singhpura (*Vertic Haplustalfs*), Sakit (*Oxyaquic Vertic Haplustalfs*), Fathepur (*Inceptic Haplustalfs*), Jagjitpur (*Oxyaquic Vertic Haplustalfs*), Dhadde (*Oxyaquic Vertic Haplustalfs*), Bhanra (*Typic Ustipsamments*), Masitawali (*Torrifluventic Haplustepts*), Ghabdan (*Haplargidic Natrustalfs*), Phaguwala (*Oxyaquic Haplustalfs*), and Hirapur (*Vertic Natrustalfs*).
	30–50	67	1.49	
	50–100	61	1.65	
	100–150	48	2.09	
Soils of the Black Soil Regions				
1.	0–30	78	1.29	1000–2200 mm MAR; subhumid bioclimatic system represented by Nabibagh (*Typic Haplusterts*). The other soils are Tenali (*Typic Haplusterts*), Kheri (Typic *Haplusterts*), Ghulguli (*Typic Haplusterts*), Sarol (*Typic Haplusterts*), and Pangidi (*Typic Haplusterts*).
	30–50	70	1.42	
	50–100	63	1.58	
	100–150	54	1.86	
2.	0–30	76	1.31	<1000 mm MAR; semi-arid and arid bioclimatic system represented by Telgi (*Sodic Haplusterts*). The other soils are Asra (*Sodic Haplusterts*), Semla (*Aridic Haplusterts*), Sokhada (*Calcic Haplusterts*), Paral (*Sodic Haplusterts*), Jajapur (*Sodic Haplusterts*), Coimbatore (*Typic Haplusterts*), Vasmat (*Typic Haplusterts*), Siddlaghatta (*Vertic Haplustepts*), Nandyal (*Sodic Haplusterts*), Achmati (*Sodic Haplusterts*), Patancheru (*Typic Rhodustalfs*), Hayatnagar (*Typic Rhodustalfs*), Kaukuntala (*Vertic Haplustalfs*), and Nimone (Sodic Haplusterts).
	30–50	69	1.44	
	50–100	63	1.60	
	100–150	51	1.97	

[a] Source: Bhattacharyya et al. 2008a,b; 2013.

estimates obtained using the WB technique (Table 4). Agroclimatic zones and variability in the land use and land cover in addition to soil type could be considered as important factors for quantifying the variation of SOC by the WB method and for finding an appropriate correction factor for the Indian soils (Table 5).

Conclusions

Studies over the past 50 years showed that the fundamental assumption, upon which the recovery percentage of about 77% by the WB method to determine SOC was based, is arbitrary. This value actually varies significantly for Indian soils and should not be considered as a universal constant. The WBRF may be influenced by vegetative cover, climate, quality and quantity of organic matter, soil depth, quantity and quality of microbial population, and degree of organic-matter decomposition, all of which may reflect in the changes in the carbon content. This study demonstrates that there should be different recovery factors for different climatic zones as well as for various depth intervals of soil. These values should be used in different laboratories for near accurate estimation of SOC for various soils of India and may serve as a state-of-the-art information at this point of time. In many soil-testing laboratories, organic C values are used for assessing soil N status for recommending doses of N fertilizers. This study recommends adoption of these WBRF in these laboratories to suggest appropriate recommendations of fertilizers to the farmers.

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