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Transformation of Zinc under Aerobic and Anaerobic Conditions in Some Soils of Kandhamal District, Odisha, India

Subhajit Dutta [a*], Abhiseka Dash [a] and Suchhanda Mondal [a]

[a] Department of Soil Science and Agricultural Chemistry, Institute of Agriculture, Palli Siksha Bhavana, Visva-Bharati, Sriniketan, Bolpur, 731236, West Bengal, India.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

An incubation study was carried out in the laboratories of the Department of Soil Science and Agricultural Chemistry, Visva-Bharati (WB), to study the transformation of native soil zinc fractions under aerobic and anaerobic conditions. Four surface soil samples of different available zinc content (0.408, 0.63, 1.016, 4.194 mg kg-1) were taken for this experiment. Soil samples were collected from farmers' fields in rice-growing areas of Kandhamal district of Odisha and analyzed for initial physicochemical properties. A five-gram portion of each soil sample was incubated in a 50 mL centrifuge tube at room temperature for a period of 0, 15, 30, 45, 60, 75, and 90 days under both aerobic and anaerobic conditions. Destructive sampling was done, and different zinc fractions were determined after every interval. Both aeration conditions and incubation period largely

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*Corresponding author: Email: Subhajit3110@gmail.com;

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influenced the transformation of native zinc fractions in soil. Irrespective of the incubation period, the anaerobic condition of the soil resulted in decrease in the content of WSEX-ZN, CRYOX-Zn, OC-Zn, MnOx-Zn, and increased the content of AMOX-Zn and RES-Zn fractions. Under anaerobic conditions, with progress in incubation period, a gradual decrease in WSEX-Zn and CRYOX-Zn fractions and an increase in AMOX-Zn and RES-Zn fractions were observed, whereas the OC-Zn and MnOx-Zn fractions were decreased after showing an increase during the initial period of incubation. Under aerobic conditions, WSEX-Zn, OC-Zn, CRYOX-Zn content decreased, and AMOX-Zn content increased; however, the RES-Zn and MnOx-Zn showed no remarkable changes with the incubation period. The transformation was more remarkable under anaerobic conditions than under aerobic conditions. Thus, anaerobic conditions over the 90-day incubation study helped to decrease the content of WSEX-Zn, OC-Zn, CRYOX-Zn, MnOx-Zn, and to increase the content of AMOX-Zn and RES-Zn. Among the entire zinc fractions studied, both the content and percent contribution of RES-Zn fraction to the total zinc remained highest throughout the incubation study, whereas WSEX-Zn content remained lowest. Change in relative preponderance order of different native zinc fractions was also observed after 90 days of incubation, both under aerobic and anaerobic conditions; however, it was more prominent in the case of the later.

Keywords: Zinc; fractions; anaerobic; aerobic.

1. INTRODUCTION

It was estimated that about 30% of the agricultural soils may be zinc deficient to the extent that normal growth and development of a wide range of agriculturally important crops are affected worldwide (Shukla et al., 2021). Work done by the All India Coordinated Scheme on micronutrients in soils and plants has shown that 47 per cent of Indian soils are deficient in zinc (Mahadevakumar et al., 2020), while Takkar (1996) indicated that among all the micronutrients, zinc deficiency is widespread in Indian soil. The problem is aggravated under anaerobic condition due to abiotic stress (Hacisalihoglu et al., 2003) in which rice is mostly grown in India. Jena et al. (2016) reported that 33.45 % of the soil of Kandhamal district (Odisha) is deficient in zinc. In soil the total zinc fraction exists in different chemical pools, *viz.* water soluble, easily exchangeable, adsorbed, chelated, or complexed, occluded by or co-precipitated with metal oxides, carbonates or phosphates and other secondary minerals and also held in primary minerals (Viets, 1962). But these discrete chemical forms are differing in their solubility and thus availability to plants. Organic amendments increase availability of certain zinc fractions, which can improve the nutrient uptake and finally yield (Raza et al., 2025). This increase in Zn availability is of great importance since only 5% or less than 5 % of total Zn present in soil is available to plants at any given time. The form, in which Zn is present in soil, plays a crucial role in determining its availability to plants. Water soluble plus exchangeable and organically complexed forms are considered to be available, amorphous sesquioxide bound form is potentially available and crystalline sesquioxide bound and residual zinc forms are unavailable to plants (Mandal et al*.,* 1992). The equilibrium among the different pools is influenced by soil electro chemical properties like pH & Eh, concentration of zinc and other cations, particularly Fe & Mn (Ramjan et al., 2014). The most important effect of submerging a soil in water is to reduce the oxygen supply. As a result, the entrained oxygen is quickly exhausted and the soil becomes almost or fully devoid of oxygen in a few days of submergence. The lack of free oxygen or the prevalence of anaerobic condition causes soil reduction and triggers a series of physical, chemical, and biological processes. Eh along with pH regulate several biochemical reactions in submerged soils that may control the distribution of different fractions of zinc and its availability under flooded condition (Sahrawat, 2015). It is known that much of the Zn remain in soils bound by oxides of iron. On submergence, these oxides undergo reduction due to anaerobiosis to the lower valent forms which are more soluble. The bound Zn is therefore released and may become available. Earlier it was observed that Zinc and its fractions gradually gets reduced in 35 days of incubation (Fogat et al., 2025). Therefore, the knowledge of the transformation of native soil zinc in different fractions under submerged (anaerobic) condition at different residence period is essential to predict the appropriate time for Zn fertilizer application and to know the importance of flooding or pre-flooding on soil Zn availability (Soltani et al., 2015).

2. MATERIALS AND METHODS

The four soil samples are from different village's *viz*. Dutipada, Balaskumpa, Bilabari & Bisipara. These villages are situated in Kandhamal, district of Odisha. The four soil samples (S1, S2, S3, S4), differing in their available Zn content (0.408, 0.63, 1.016, 4.194 mg kg^{-1}), were taken as four experimental material for the present study. Each sample was incubated for 0,15,30,45,60,75 and 90 days under both aerobic and anaerobic conditions. Each experimental unit (total 4 x 7x 2 x 3 replications =168) consisted of exactly 5 gram of respective soil in a 50 mL polyethylene centrifuge tube. Aerobic conditions were maintained inside the respective experimental units by maintaining the soil at nearly about field capacity moisture content and for anaerobic conditions 5±0.5 cm height of standing water (double distilled) was maintained above the soil in the experimental unit. Any loss of water through evaporation was compensated by periodic addition of double distilled water. The respective experimental units were incubated under laboratory conditions at room temperature. After completion of each incubation period destructive sampling was done and the soils in respective experimental units were subjected for determination of different zinc fractions (fertility fractions), which are as follows. Water soluble plus exchangeable zinc (WSEX-Zn), Organically complexed zinc (OC-Zn), Amorphous sesquioxide bound zinc (AMOX-Zn), Crystalline sesquioxide bound zinc (CRYOX-Zn), Easily reducible manganese oxide bound zinc (MnOx-Zn) and Residual zinc (RES-Zn).

The Zn fractionations (fertility fractions) were determined using a modified sequential extraction procedure of Murthy (1982). The Zn pools in soils were divided into six fractions (mentioned in section 3.6.4 of this chapter), including residual. Zn forms in soils, except residual, were extracted from soils by the addition of individual extracting solutions and shaken in a rotary shaker at room temperature. The supernatant solutions obtained from each successive stage of extraction were centrifuged at 3,500 rpm for 15 min, decanted, and filtered with Whatman No. 42 filter paper. Residual Zn was extracted by digesting the residue of 5th fraction with a mixture of 5mL of concentrated nitric acid, 10 mL of hydrofluoric acid and 10 mL of perchloric acid, the cooled solution was transferred to a 50 mL volumetric flask and subsequently diluted to volume with double distilled water and stored for analysis (Spalbar et al., 2017). The concentration of zinc in the extractant was determined by Atomic Absorption Spectrophotometer (Page *et al*., 1982).

3. RESULTS AND DISCUSSION

3.1 Transformation of Water-Soluble Plus Exchangeable Zinc (WSEX-Zn) Fraction

The transformation pattern of WSEX-Zn fraction under aerobic and anaerobic conditions at 0, 15, 30, 45, 60, 75, and 90 DAI is presented in Tables 1-4. It was observed that the aeration condition has a significant effect on WSEX-Zn content in all four soils. Irrespective of the incubation period, significantly lower WSEX-Zn content of 0.185, 0.148, 0.204, and 0.248 mg kg^{-1} was recorded under anaerobic conditions (M2) than 0.260, 0.209, 0.294, and 0.340 mg kg^{-1} recorded under aerobic conditions (M_1) in S1, S2, S3, and S4, respectively. A similar type of finding has also been observed by Wijebandara (2007). The incubation period (I) has also shown a significant effect on WSEX-Zn content, which significantly decreased along with the incubation period in all four soils. This finding is in line with the observation of Sharma (2002). Significantly highest WSEX-Zn amount of 0.323, 0.265, 0.336, and 0.459 mg kg^{-1} was recorded in S1, S2, S3, and S4, respectively, at 0 DAI, which is at par with the value of 15 DAI in case of S2 & S3. In all the four soils the WSEX-Zn has shown a significant decrease along with the progress of the incubation period (I) under both aerobic (M_1) and anerobic (M_2) conditions however, the decrease was more under the later one. Similar findings were observed by Wijebandara, 2007. Under aerobic condition (M_1) the highest WSEX-Zn value of 0.326, 0.264, 0.336, and 0.453 mg kg^{-1} were observed in S1, S2, S3 and S4 respectively at 0 DAI and this maximum value is at par with the values observed at 15 DAI & 30 DAI (in S1 & S2), at 15DAI, 30 DAI, & 45 DAI (in S3). Similarly, under anaerobic conditions (M2), s the highest WSEX-Zn values of 0.319, 0.266, 0.337, and 0.464 mg kg^{-1} were observed in S1, S2, S3, and S4, respectively, at 0 DAI, which is at par with 15 DAI only in the case of S3. After 0DAI, the value of WSEX-Zn under anaerobic conditions (M2) was always lower than that of aerobic conditions (M1), but it was found to be significant from 30 DAI onwards up to 90 DAI, except in S4, where a significant decrease was observed after 15 DAI. The decrease in WSEX – Zn fraction under anaerobic (M2) situation has been observed by many workers (Wijebandara *et al*., 2014; Kumar and Babel (2011) and Ramzan *et a*l., (2014) in their incubation studies on the transformation of native soil zinc. This decrease might be due to the increase in soil pH value under submergence which decreased the solubility of the native WSEX-Zn by precipitation of some amount of it as hydroxides and/or adsorption on the surface of freshly formed hydrated oxides of Fe & Mn (Wijebandara *et al*., 2014).

3.2 Transformation of Organically Complexed Zinc (OC-Zn) Fraction

From the Tables 1-4 it was observed that the aeration condition of soil had shown a significant effect on OC-Zn content and significantly higher values of 1.706, 1.767, 2.159 and 3.811 mg kg^{-1} were recorded under aerobic condition (M_1) than 1.582, 1.661, 2.024 and 3.712 mg kg^{-1} recorded under anaerobic condition (M_2) in S1, S2, S3, and S4, respectively. Such effect of aeration is in line with the findings of Wijebandara (2007) under field capacity and submergence moisture regime. The incubation period (I) also showed significant effect on OC-Zn content in all the four soils. In S1 & S4 maximum OC-Zn content of 1.907 & 4.821 mg kg^{-1} were observed at 15 DAI, respectively and it was 1.990 & 2.524 mg kg^{-1} at 30 DAI for S2 & S3, respectively. These maximum values are at par with the values observed at 0DAI & 30 DAI in S1 & S4 and with that at 0DAI & 15 DAI in S2 &S3. In all the four soils after approaching the maximum at 15 or 30 DAI, the values gradually decreased and reached to minimum content of 1.315, 1.377, 1.590, 2.571 mg kg^{-1} at 90 DAI in S1, S2, S3, & S4, respectively and these minimum values are at par with the values obtained at 75DAI in respective soils. Decreasing effect of incubation period was also observed by Wijebandara (2007) during his 30, 60 & 90 days of incubation study under field capacity and submergence moisture regime. It was also observed that in all the four soils, under anaerobic condition (M_2), the amount of OC-Zn showed an increase immediately after submergence, attained a peak value of 2.021, 2.128, 2.751 & 5.576 at 30 DAI in S1, S2, S3 & S4, respectively and thereafter showed a declining trend by significantly decreasing from the initial value at 45 DAI .The maximum value at 30 DAI is at par with that at 15 DAI & O DAI in all the soils except S4, where it is at par with only 15 DAI i.e., significantly increased from the initial value. Similar trend was also reported by Chatterjee et al. (1992) and Singh (2018). Singh (2018) explained that such an increase is possible due to the release of Zn bound by crystalline sesquioxides under submerged conditions and its subsequent chelation by metal-binding biochemical substances resulting from incomplete microbial decomposition of soil organic matter under anaerobic conditions. Further the declining trend towards the latter period of submergence might be due to the decrease in stability of Zn-organic complexes at lower Eh of the soil attained after prolonged submergence (Mandal et al*.,* 1992). Under aerobic conditions (M1) in all soils, there was a gradual decrement in OC-Zn content throughout the incubation period, with a significant decrease from the initial at 60DAI in S1 & S2, at 45DAI in S3, and at 15 DAI in S4. However, significantly lower amount of OC-Zn was observed in anaerobic condition (M_2) than aerobic condition (M_1) from 45DAI onwards in all soils except S2 where the decrease was observed after 60 DAI. But in initial periods, a significantly higher value was observed under anaerobic conditions (M2) than that of aerobic conditions (M1) at 15 DAI and 30 DAI in all the soils except S1, where it is significant only at 30 DAI. Similar findings of decrease of OC-Zn content under aerobic condition (M_1) was also reported by Alvarez and Gonzalez (2006).

3.3 Transformation of Amorphous Sesquioxide Bound Zinc (AMOX-Zn) Fraction

The AMOX-Zn content was significantly affected by the aeration condition (M) and significantly higher AMOX-Zn value of 2.445, 2.258, 3.906 and 11.821 mg kg^{-1} were found under anaerobic condition (M_2) than 1.938, 1.728, 3.124 and 10.510 mg kg^{-1} under aerobic condition (M_1) in S1, S2, S3, and S4, respectively. Similar finding of higher AMOX-Zn content under field capacity than submergence moisture regime was also obtained by Wijebandara (2007). In all the four soils the AMOX-Zn content significantly varied with the incubation period (I) and showed an increasing pattern of transformation with progress of incubation period. Significant increase in the value from initial was observed at 30 DAI in all the four soils. Maximum values of 2.775, 2.604, 4.460 & 13.433 mg kg^{-1} were observed at 90 DAI in S1, S2, S3, & S4, respectively and these values are at par with the values recorded at 75 DAI in S1, S2 & S3. Similar increase in AMOX-Zn with incubation time was also reported by Kamali et al. (2010). It can be interpreted that, under anaerobic condition (M_2), the AMOX-Zn fraction have shown an increasing trend with the progress of incubation period in all the four soils under study. A significant increase in the value was observed at 45 DAI in both S1 & S3 where as it is 15 DAI, & 30 DAI in S2 & S4, respectively. The significantly highest values of 3.265, 3.101, 5.247 & 15.115 mg kg^{-1} were recorded at 90 DAI in S1, S2, S3 and S4, respectively. The highest value recorded at 90 DAI is at par with the value at 75 DAI in all the soil except S4. Increasing in AMOX-Zn fraction with incubation period under submergence condition was also observed by Tehrani (2005); Bahera et al., (2008) and Safari et al., (2009). and Wijebandara et al*.* (2014). Under the reducing condition in an anaerobic soil, due to submergence, there was an increase in the formation of hydrated oxides of Fe^{2+} iron and Mn^{2+} manganese which, when freshly formed, have a high surface area and hence possess strong adsorptive capacity. The WSEX-Zn fraction already present in soil or that released from other forms might have been adsorbed on the surface of these freshly precipitated hydrated oxides, resulting in an increase in the AMOX-Zn fraction content in anaerobic condition of the soil under submergence. Under aerobic condition (M_1) the AMOX-Zn content also showed an increasing trend of transformation with the period of incubation, by significantly increasing from the initial at 45 DAI (in S1 & S3) and at 30 DAI (in S2 & S4). This increasing pattern, under aerobic conditions, is in line with the findings of Kamali et al. (2010) at 15, 45, and 90 days of incubation. Although more or less identical values of AMOX-Zn fraction, under both aeration conditions, were recorded up to 30 DAI, the value under anaerobic condition (M_2) significantly exceeded that under aerobic condition (M_1) from 45 DAI onwards up to 90 DAI. Such significantly higher values under anaerobic conditions (M2) than aerobic (M2) conditions are in line with the findings of Wijebandara (2007).

3.4 Transformation of Crystalline Sesquioxide-Bound Zinc (CRYOX-Zn) Fraction

The aeration condition (M) has a significant effect on the CRYOX-Zn fraction in all four soils. Significantly higher values of 19.25, 16.37, 15.36, and 17.97 mg kg^{-1} were found under aerobic conditions (M1) than 15, 12.77, 11.33, and 13.22 mg kg^{-1} under anaerobic conditions (M2) in S1, S2, S3, and S4, respectively. Wijebandara (2007) has also reported a higher value of CRYOX-Zn content under the field capacity moisture regime than that under submergence. The CRYOX-Zn fraction was significantly varied with the incubation period (I) in all the four soils. The values showed a decreasing pattern of transformation with progress of incubation period up to 90 DAI. Significantly higher values of 20.34, 17.19, 16.26 & 19.06 mg kg^{-1} were observed at 0 DAI in S1, S2, S3 and S4, respectively. This higher value at 0DAI was at par with that at 15 & 30 DAI in both S1 & S2, and only with 15 DAI in the case of S4. Its value significantly decreased from the initial at 45 DAI (in S1 & S2), at 15 DAI (in S3), at 30 DAI (in S4) and reached minimum at 90 DAI in all the four soils. Sharma (2002) has also found the significant increasing effect of incubation time on CRYOX-Zn fraction. Under aerobic condition (M1), the CRYOX-Zn content was significantly decreased with the progress of incubation period only in S3 whereas, the effect is not significant in S1, S2, & S4. Under anaerobic condition (M2), in S3 and S4, significant decrease in CRYOX-Zn fraction was observed with the increase in incubation period up to 90 DAI and the decrease from initial (O DAI) became significant at 15 DAI (in S3) and 30 DAI (in S4). Under anaerobic condition, significantly higher values of 16.26 & 19.06 mg kg^{-1} CRYOX-Zn content was observed at 0DAI in S3 & S4, respectively and this higher value is at par with that of 30 DAI in S4. In all the four soils, the CRYOX-Zn content is always less under anaerobic condition (M2) than under aerobic condition (M1) but this difference, which is prominent from 15 DAI up to 90 DAI, is significant only in case of S3 & S4. Singh (2018) and Wijebandara *et al*. (2014) have also observed the decreasing pattern of transformation of CRYOX-Zn fraction under flooded moisture conditions. This result also corroborates the findings of Mandal *et al*. (1992), who reported significantly less transformation of applied zinc into CRYOX-Zn fraction in 15 days pre-flooded soil than in 0 days pre-flooded soil. Under reducing conditions in anaerobic soil, due to submergence, some of the crystalline sesquioxides might have undergone transformation to an amorphous form, resulting in the release of a part of the Zn occluded by the former and the subsequent adsorption of this by the latter. This can explain the decreasing pattern of CRYOX-Zn fraction with the progress of the incubation period under anaerobic conditions.

Table 1. Content of zinc fractions (mg kg^{-1}) under aerobic (M_1) and anaerobic (M_2) conditions at different incubation periods (I_0 to I_{90}) in soil-1 (S1)

	WSEX-Zn			**OC-Zn**			**AMOX-Zn**			**CRYOX-Zn**			**MnOx-Zn**			**RES-Zn**		
	M_1	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**
I_0	0.326	0.319	0.323	1.900	1.906	1.903	1.600	1.594	1.597	20.35	20.34	20.34	3.325	3.315	3.320	41.91	41.93	41.92
I_{15}	0.299	0.254	0.276	1.850	1.965	1.907	1.627	1.738	1.682	19.81	18.81	19.31	3.391	3.957	3.674	42.43	42.67	42.55
I_{30}	0.286	0.212	0.249	1.770	2.021	1.895	1.757	1.802	1.780	19.55	15.74	17.64	3.454	4.019	3.736	42.60	45.61	44.10
I_{45}	0.243	0.162	0.203	1.723	1.498	1.611	1.909	2.728	2.319	19.30	13.43	16.37	3.457	3.651	3.554	42.76	47.92	45.34
I_{60}	0.242	0.122	0.182	1.634	1.421	1.528	2.138	2.776	2.457	18.96	12.51	15.73	3.514	2.681	3.097	42.92	49.89	46.40
I_{75}	0.218	0.116	0.167	1.546	1.157	1.352	2.249	3.213	2.731	18.69	12.12	15.40	3.584	2.484	3.034	43.12	50.32	46.72
I_{90}	0.205	0.111	0.158	1.520	1.110	1.315	2.286	3.265	2.775	18.12	12.05	15.09	3.703	2.422	3.062	43.56	50.45	47.00
Mean	0.260	0.185		1.706	1.582		1.938	2.445		19.25	15.00		3.490	3.218		42.76	46.97	
	SE (M)	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**
M	0.064	0.009	0.019	0.023	0.034	0.069	0.028	0.04	0.083	0.591	0.837	1.714	0.078	0.111	0.228	0.524	0.742	1.52
I	0.012	0.017	0.035	0.044	0.063	0.129	0.053	0.076	0.155	1.107	1.566	3.207	0.147	0.208	0.427	0.981	1.388	2.843
M × I	0.017	0.024	0.049	0.062	0.089	0.182	0.075	0.107	0.219	1.566	2.215	NS	0.208	0.295	0.604	1.388	1.963	4.021
CV (%)	13.260			6.618			5.973			15.837			10.766			5.360		

Table 2. Content of zinc fractions (mg kg^{-1}) under aerobic (M_1) and anaerobic (M_2) conditions at different incubation periods (I_0 to I_{90}) in soil-2 (S2)

	WSEX-Zn			**OC-Zn**			**AMOX-Zn**			**CRYOX-Zn**			**MnOx-Zn**			**RES-Zn**		
	M_1	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**
I_0	0.264	0.266	0.265	1.968	1.960	1.964	1.320	1.318	1.319	17.20	17.19	17.19	3.525	3.540	3.533	47.23	47.23	47.23
I_{15}	0.256	0.217	0.237	1.876	2.065	1.970	1.350	1.573	1.461	16.95	16.22	16.59	3.588	4.335	3.961	47.48	47.10	47.29
I_{30}	0.250	0.192	0.221	1.852	2.128	1.990	1.645	1.622	1.633	16.63	13.57	15.10	3.618	4.425	4.022	47.50	49.56	48.53
I_{45}	0.204	0.136	0.170	1.814	1.694	1.754	1.752	2.503	2.127	16.45	11.13	13.79	3.656	3.260	3.458	47.62	52.77	50.20
I_{60}	0.178	0.081	0.130	1.650	1.435	1.542	1.863	2.661	2.262	16.17	10.68	13.43	3.714	3.165	3.439	47.93	53.47	50.70
I_{75}	0.158	0.072	0.115	1.614	1.193	1.404	2.057	3.026	2.541	15.90	10.35	13.13	3.841	2.370	3.106	47.93	54.49	51.21
I_{90}	0.154	0.070	0.112	1.598	1.156	1.377	2.108	3.101	2.604	15.31	10.24	12.77	3.955	2.327	3.141	48.38	54.62	51.50
Mean	0.209	0.148		1.767	1.661		1.728	2.258		16.37	12.77		3.700	3.346		47.72	51.32	
	SE (M)	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**
M	0.054	0.008	0.016	0.023	0.033	0.068	0.027	0.039	0.08	0.505	0.714	1.462	0.090	0.127	0.261	0.554	0.784	1.605
I	0.010	0.014	0.029	0.044	0.062	0.127	0.051	0.073	0.15	0.944	1.336	2.735	0.169	0.238	0.488	1.037	1.466	3.003
M × I	0.014	0.02	0.041	0.062	0.088	0.18	0.073	0.104	0.212	1.336	1.889	NS	0.238	0.337	0.691	1.466	2.074	NS
CV (%)	13.804			6.283			6.368			15.877			11.723			5.129		

Table 3. Content of zinc fractions (mg kg^{-1}) under aerobic (M_1) and anaerobic (M_2) conditions at different incubation periods (I_0 to I_{90}) in soil-3 (S3)

	WSEX-Zn			**OC-Zn**			**AMOX-Zn**			**CRYOX-Zn**			**MnOx-Zn**			**RES-Zn**		
	M_1	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**
I_0	0.336	0.337	0.336	2.475	2.466	2.471	2.624	2.612	2.618	16.27	16.25	16.26	3.088	3.090	3.089	34.72	34.74	34.73
I_{15}	0.329	0.279	0.304	2.386	2.577	2.482	2.739	2.798	2.769	15.70	13.63	14.66	3.164	3.825	3.495	35.20	36.40	35.80
I_{30}	0.322	0.215	0.268	2.298	2.751	2.524	2.991	2.933	2.962	15.60	12.82	14.21	3.188	4.100	3.644	35.09	36.67	35.88
I_{45}	0.277	0.180	0.229	2.234	1.896	2.065	3.117	4.329	3.723	15.49	9.75	12.62	3.235	2.855	3.045	35.15	40.48	37.81
I_{60}	0.266	0.175	0.221	1.987	1.728	1.857	3.225	4.418	3.822	15.34	9.27	12.30	3.263	2.910	3.087	35.41	41.01	38.21
I_{75}	0.265	0.126	0.195	1.882	1.424	1.653	3.502	5.002	4.252	14.91	8.88	11.89	3.316	2.240	2.778	35.63	41.83	38.73
I_{90}	0.264	0.120	0.192	1.853	1.326	1.590	3.673	5.247	4.460	14.21	8.68	11.44	3.412	2.160	2.786	36.10	41.97	39.03
Mean	0.294	0.204		2.159	2.024		3.124	3.906		15.36	11.33		3.238	3.026		35.33	39.01	
	SE (M)	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**
M	0.008	0.012	0.024	0.0243	0.034	0.07	0.055	0.079	0.162	0.090	0.127	0.26	0.059	0.084	0.172	0.138	0.196	0.401
I	0.015	0.022	0.045	0.0454	0.064	0.131	0.104	0.148	0.303	0.168	0.238	0.487	0.111	0.157	0.321	0.259	0.366	0.75
M × I	0.021	0.031	0.063	0.0642	0.091	0.186	0.147	0.209	0.428	0.238	0.336	0.689	0.157	0.222	0.454	0.366	0.518	1.061
CV (%)	15.160			5.314			7.283			3.088			8.665			1.707		

Table 4. Content of zinc fractions (mg kg^{-1}) under aerobic (M_1) and anaerobic (M_2) conditions at different incubation periods (I_0 to I_{90}) in soil-4 (S4)

	WSEX-Zn			**OC-Zn**			**AMOX-Zn**			**CRYOX-Zn**			**MnOx-Zn**			**RES-Zn**		
	M_1	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**
I_0	0.453	0.464	0.459	4.800	4.790	4.795	9.330	9.310	9.320	19.07	19.06	19.06	5.695	5.690	5.693	69.86	69.89	69.87
I_{15}	0.396	0.336	0.366	4.457	5.185	4.821	9.444	9.622	9.533	18.42	17.57	17.99	6.181	5.835	6.008	70.31	70.65	70.48
I_{30}	0.381	0.282	0.331	4.004	5.576	4.790	10.237	10.277	10.257	18.07	13.67	15.87	5.857	6.760	6.309	70.65	72.64	71.64
I_{45}	0.348	0.232	0.290	3.821	3.290	3.556	10.276	11.485	10.881	18.14	11.38	14.76	5.927	4.510	5.219	70.68	78.30	74.49
I_{60}	0.299	0.151	0.225	3.401	2.766	3.084	11.017	12.566	11.791	17.72	10.59	14.16	6.020	4.405	5.213	70.74	78.73	74.74
I_{75}	0.256	0.136	0.196	3.168	2.265	2.716	11.517	14.377	12.947	17.32	10.25	13.78	6.096	3.325	4.710	70.84	78.86	74.85
I_{90}	0.250	0.135	0.192	3.027	2.115	2.571	11.751	15.115	13.433	17.04	10.02	13.53	6.254	3.263	4.758	70.87	78.57	74.72
Mean	0.340	0.248		3.811	3.712		10.510	11.821		17.97	13.22		6.004	4.827		70.57	75.38	
	SE (M)	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**	**SE (M)**	**SE (D)**	**$CD_{0.05}$**
M	0.007	0.01	0.02	0.033	0.047	0.095	0.067	0.095	0.195	0.294	0.416	0.853	0.071	0.101	0.206	0.344	0.486	0.996
I	0.013	0.018	0.038	0.061	0.087	0.179	0.125	0.178	0.364	0.551	0.779	1.595	0.133	0.188	0.386	0.643	0.91	1.863
M × I	0.019	0.026	0.054	0.087	0.123	0.253	0.177	0.251	0.515	0.779	1.102	2.256	0.188	0.266	0.545	0.910	1.287	2.635
CV (%)		10.876			4.015			2.757			8.653			6.022			2.160	

3.5 Transformation of Manganese Oxide Bound Zinc (MnOx-Zn) Fraction

It was observed that aeration condition of soil showed significant effect on MnOx-Zn content in all the four soils and significantly lower values of 3.218, 3.346, 3.026 and 4.827 mg kg^{-1} were recorded under anaerobic condition (M_2) than 3.490, 3.700, 3.238 and 6.004 mg kg^{-1} under aerobic condition (M_1) in S1, S2, S3, and S4, respectively. Incubation period (I) has also shown a significant effect on MnOx-Zn content in all four soils. In all the soils, during the initial periods of incubation, the value gradually increased and reached a maximum at 30 DAI, which significantly differed from the initial value in S3 & S4 and at par in S1 & S2. After attaining the maximum at 30 DAI, it was decreased from the initial 45 DAI (in S2 & S4) or 60 DAI (in S1 & S3) and became minimum at 90 DAI. However, the decrease from the initial was significant only in S4. Under anaerobic conditions (M2), the MnOx-Zn fraction was gradually increased and reached maximum values of 4.019, 4.425, 4.100, and 6.760 mg kg^{-1} at 30 DAI in all the soils. The increment from the initial is significant in all the soil except S4, where it was at par with the value at 0 DAI. After reaching the maximum at 30 DAI (in all soils), it gradually started to decrease, became significantly lower from the initial value at 60 DAI (in S1 & S4) or 75 DAI (in S2 & S3) and reached to a minimum value of 2.422, 2.327, 2.160 & 3.263 mg kg^{-1} at 90 DAI in S1, S2, S3 & S4, respectively. A similar pattern of transformation of MnOx-Zn, under submerged conditions, has also been observed by Singh (2018) and Chatterjee et al. (1992). Singh (2018) also explained the reason of this pattern of transformation as follows. Under anaerobic conditions, some amount of higher oxides of manganese are converted to the lower valent compounds (Chatterjee et al., 1992), which on diffusion to the oxic region get reoxidized with subsequent formation of manganic hydroxides having a large specific surface area and hence high adsorption capacity. The zinc released from other bound forms might have been held by the manganic hydroxides resulting in an increase in the content of MnOx-Zn fraction. But during the latter period of incubation, the redox potential of soil is decreased, which causes the reduction of the freshly formed manganic hydroxide, resulting in the release of the adsorbed zinc and hence the decrease in the MnOx-Zn fraction. In all the four soils, under aerobic condition (M_1), the value of MnOx-Zn was gradually increased up to 90 DAI but the increase is not significant except in S4, where the maximum value, recorded at 90 DAI only, was significantly higher than the value recorded at 0 DAI. During the initial period, up to 30 DAI, the MnOx-Zn fraction under anaerobic conditions (M2) was higher than that under aerobic conditions (M_1). However, this increasing difference was prominent at both 15 & 30 DAI (in S2 & S3) or only at 30 DAI (in S4) and insignificant in case of S1. Significantly lower value of MnOx-Zn content was also observed from 60 DAI onwards (in S1), 75 DAI onwards (in S2 & S3) and 45 DAI onwards (in S4).

3.6 Transformation of Residual Zinc (RES-Zn) Fraction

Aeration condition (M) has shown significant effect on the RES-Zn fraction in all the four soils. Significantly higher values of 46.97, 51.32, 39.01 & 75.38 mg kg^{-1} were recorded under anaerobic condition (M_1) than 42.76, 47.72, 35.33 & 70.567 mg kg^{-1} under aerobic condition (M_1) in S1, S2, S3 and S4, respectively. It was observed that the Incubation period (I) has also shown a significant increasing effect on the content of RES-Zn fraction in all four soils. As the incubation period progressed, the value increased and significantly differed from the initial at 45 DAI (in S1 & S4) or 60 DAI (in S2) or 15 DAI (in S3), and attained maximum values of 47.00, 51.50, 39.03 mg kg^{-1} at 90 DAI in S1, S2, & S3, respectively. In S4, the maximum value of 74.85 mg kg^{-1} was observed at 75 DAI, which is at par with those values at 45, 60 & 90 DAI. Under aerobic condition (M_1), the RES-Zn fraction recorded an increasing trend along with the progress of incubation period (I) but, the increment was not significantly different from the initial value in all the soils except in S4, where it significantly differed only at 90 DAI. Under anaerobic conditions (M2), the value of RES-Zn also showed an increasing pattern with the incubation period (I). The RES-Zn fraction content was significantly increased from the initial value at 45 DAI (in S1), 15 DAI (in S3), and 30 DAI (in S4) and recorded significantly highest value of 50.45, 41.97 mg kg^{-1} at 90 DAI in S1 and S3 but in S4, it is 78.86 mg kg^{-1} at 75 DAI, which is at par with 90 & 60DAI. Similar progressive increase in RES-Zn fraction has also been observed by Wijebandara et al. (2014) in his 30, 60 & 90 days of incubation study under submerged condition. This progressive increase in RES-Zn fraction with advancement of incubation period indicated considerable mobilization of Zn to residual fraction from other fractions (Wijebandara et al*.,* 2014). However, significantly higher values of RES-Zn fraction were observed under M_2 than M_1 from 45 DAI onwards in S1 & S4 but in S3, it was observed from 30 DAI onwards. Enoch et al., 2017, Singh, (2011) and Kamali et al., (2010) also reported significantly higher value of RES-Zn fraction under submergence moisture regime than field capacity moisture regime.

3.7 Contribution of Different Native Soil Zinc Fractions to Total Zinc under Aerobic and Anaerobic Conditions before and after 90 days of Incubation

3.7.1 Distribution of native soil zinc fractions in Soil-1

From Table 1, it was observed that initially (at 0 DAI) the content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & and RES-Zn fractions was 0.326, 1.9, 1.6, 20.349, 3.325, & 41.91 mg kg^{-1} and it constituted 0.47, 2.738, 2.305, 29.319, 4.791 & and 60.378 % of total Zn, respectively with the following order of preponderance: WSEX-Zn < AMOX-Zn < OC-Zn < MnOx-Zn < <CRYOX-Zn < RES-Zn. However, a change in initial content of zinc fractions and their order of preponderance was observed at 90DAI in both aerobic (M_1) and anaerobic (M_2) conditions. Under aerobic conditions (M1) at 90 DAI, an alteration in order between OC-Zn and AMOX-Zn was recorded. The contents of WSEX-Zn, OC-Zn, AMO-Zn, CRYOX-Zn, MnOx-Zn & and RES-Zn fractions were 0.205, 1.520, 2.286, 18.123, 3.703 & 43.56 mg kg^{-1}, which constituted 0.296, 2.190, 3.294, 26.116, 5.336 & and 62.768 percent of total Zn, respectively, with the following order of preponderance: WSEX-Zn < OC-Zn < AMOX-Zn < MnOx-Zn < <CRYOX-Zn < RES-Zn. Whereas, under anaerobic conditions (M2) at 90 DAI, a change in order among the OC-Zn, AMOX-Zn, and MnOx-Zn was observed. The content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was 0.111, 1.110, 3.265, 12.050, 2.442 & 50.45 mg kg^{-1} which constitute 0.160, 1.599, 4.704, 17.361, 3.489 & 72.687 percent of total Zn, respectively with the following order of preponderance: WSEX-Zn < OC-Zn < MnOx-Zn < AMOX-Zn < <CRYOX-Zn < RES-Zn.

3.7.2 Distribution of native soil zinc fractions in Soil-2

From the Table 2 it was observed that, initially (at 0 DAI) the content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was found to be 0.264, 1.968, 1.320, 17.2, 3.525 & 47.23 mg kg^{-1} and constituted 0.369, 2.752, 1.846, 24.052, 4.929 & 66.051 % of total Zn, respectively with the following order of preponderance: WSEX-Zn < AMOX-Zn < OC-Zn < MnOx-Zn < <CRYOX-Zn < RES-Zn. However, a change in initial content of zinc fractions and their order of preponderance was observed at 90DAI in both aerobic (M_1) and anaerobic (M_2) conditions. Under aerobic conditions (M1) at 90 DAI, an alteration in order between OC-Zn and AMOX-Zn was recorded. The content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was 0.154, 1.598, 2.108, 15.312, 3.955 & 48.38 mg kg^{-1} and constituted 0.215, 2.235, 2.948, 21.412, 5.531 & 67.658 percent of total Zn, respectively, with the following order of preponderance: WSEX-Zn < OC-Zn < AMOX-Zn < MnOx-Zn < <CRYOX-Zn < RES-Zn. Whereas, under anaerobic conditions (M2) at 90 DAI, a change in order among the OC-Zn, AMOX-Zn, and MnOx-Zn was observed. The content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was 0.070, 1.156, 3.101, 10.237, 2.327, & 54.62 mg kg^{-1}, which constitute 0.098, 1.617, 4.336, 14.317, 3.254 & 76.378 percent of total Zn, respectively, with the following order of preponderance: WSEX-Zn < OC-Zn < MnOx-Zn < AMOx-Zn < CRYOX-Zn < RES-Zn.

3.7.3 Distribution of native soil zinc fractions in Soil-3

From the Table 3, it was observed that initially (at 0 DAI) the content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was found to be 0.336, 2.475, 2.624, 16.269, 3.088, 34.72 mg kg^{-1}, which constituted 0.565, 4.160, 4.409, 27.340, 5.190 & 58.337 % of total Zn, respectively with the following order of preponderance : WSEX-Zn < OC-Zn < AMOX-Zn < MnOx-Zn < <CRYOX-Zn < RES-Zn. However, a change in initial content of zinc fractions and their order of preponderance was observed at 90DAI in both aerobic (M_1) and anaerobic (M_2) conditions. Under aerobic condition (M_1) at 90 DAI, an alteration in order between AMOX-Zn & MnOx-Zn was recorded. The content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was 0.264, 1.853, 3.673, 14.207, 3.412 & 36.01 mg kg^{-1}, which constitute 0.444, 3.114, 6.172, 23.875, 5.734 & 60.661 percent of total Zn, respectively with the following order of preponderance: WSEX-Zn < OC-Zn< MnOx-Zn < AMOX-Zn <CRYOX-Zn < RES-Zn. Also, under anaerobic condition (M_2) at 90 DAI, a change in order between the AMOX-Zn & MnOx-Zn was observed. The contents of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was 0.120, 1.326, 5.247, 8.681, 2.160 & 41.973 mg kg^{-1}, which constituted 0.202, 2.229, 8.818, 14.587, 3.630 & 70.535 percent of total Zn, respectively with the following order of preponderance: WSEX-Zn < OC-Zn < MnOx-Zn < AMOx-Zn < <CRYOX-Zn << RES-Zn.

3.7.4 Distribution of native soil zinc fractions in Soil-4

From the Table 4, it was observed that initially (at 0 DAI) the content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was found to be 0.453, 4.800, 9.330, 19.069, 5.695, & 69.86 mg kg^{-1} and constituted 0.415, 4.395. 8.543, 17.460, 5.215 & 63.971 % of total Zn, respectively with the following order of preponderance: WSEX-Zn < OC-Zn < MnOx-Zn < AMOX-Zn < <CRYOX-Zn < RES-Zn. However, a change in initial content of zinc fractions and their order of preponderance was observed at 90DAI only under anaerobic (M_2) condition. Under aerobic condition (M_1) at 90 DAI, the content of WSEX-Zn, OC-Zn, AMO-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was 0.250, 3.027, 11.751, 17.039, 6.254 & 70.87 mg kg^{-1}, which constitute 0.229, 2.772, 10.762, 15.605, 5.727 & 64.905 percent of total Zn, respectively and no change in initial distribution order was observed. Whereas, under anaerobic condition (M_2) at 90 DAI, a change in order between the AMOX-Zn & CRYOX-Zn was observed. The content of WSEX-Zn, OC-Zn, AMOX-Zn, CRYOX-Zn, MnOx-Zn & RES-Zn fraction was found to be 0.135, 2.115, 15.115, 10.019, 3.263 & 78.566 mg kg^{-1} and constituted 0.124, 1.937, 13.840, 9.173, 2.988 & 71.939 percent of total Zn, respectively, with the following order of preponderance: WSEX-Zn < OC-Zn < MnOx-Zn < <CRYOX-Zn< AMOX-Zn << RES-Zn. So, in S1, S2, and S3, a change in the initial content of different Zn fractions and their order of distribution was observed both under aerobic (M_1) and anaerobic (M_2) conditions at 90 DAI. Whereas, in S4, the changes in distribution order was observed only under anaerobic condition (M_1). However, the changes are more remarkable under anaerobic conditions. Change in distribution order of different zinc fractions between 30 & 60 DAI has also been reported by Soltani et al. (2015).

4. CONCLUSION

In a nutshell, it can be said that over the 90 days of incubation study, a more remarkable decrease under anaerobic conditions was noticed than that under aerobic conditions in all the native zinc fractions except AMOX-Zn & RES-Zn, where a remarkable increase was observed. Irrespective of the incubation period, significantly lower amounts of WSEX-Zn, OC-Zn, CRYOX-Zn, and MnOx-Zn were observed under anaerobic conditions (M2), whereas AMOX-Zn & RES-Zn content was high under anaerobic conditions (M2). Under anaerobic conditions, with an incubation period, gradual decreases in WSEX-Zn and CRYOX-Zn fractions and an increase in AMOX-Zn and RES-Zn fractions were observed. The OC-Zn & MnOx-Zn fractions also indicated a decrease after showing an increase during the initial period of incubation under anaerobic conditions (M2). Under aerobic conditions, WSEX-Zn, OC-Zn, CRYOX-Zn, and AMOX-Zn decreased; however, RES-Zn and MnOx-Zn showed no remarkable changes with the incubation period. Thus, the information derived from the present investigation may be helpful in deciding appropriate water management practices and the time of zinc fertilizer application to soil for enhancing zinc nutrient use efficiency and its availability to crop plants to achieve better growth and yield.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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