Supplementary Materials

Preferential elimination of Ba^{2+} through irreversible biogenic manganese oxide sequestration

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Table S1-S3
Figure S1-S6

Table S1 Data summary of sequestration experiments for Ba^{2+} by newly formed and heated BMOs in 20 mM HEPES (pH 7.0) with or without exogenous Mn^{2+}

Condition		Sequestration efficiency (%) (added ions; mM)				Two-step extraction					
		Ba^{2+}		Mn^{2+}		Total Ba^{2+} (mM)	Exchangeable (%)	Reducible (%)	Total Mn (mM)	Exchangeable (%)	Reducible (%)
Newly formed[d] Aerobic Only Mn^{2+}	1st	-		>99	(1.01±0.00)	-	-	-	2.10±0.01	15.8±0.1	84.2±0.1
	2nd	-		>99	(1.02±0.01)	-	-	-	3.00±0.02	12.5±0.1	87.5±0.1
	3rd	-		98.7±0.6	(1.00±0.00)	-	-	-	4.00±0.03	13.2±0.0	86.8±0.0
	Total	-		>99		-	-	-			
Newly formed[d] Aerobic Only Ba^{2+}	1st	17.7±0.7	(1.00±0.00)		-	0.18±0.00	93.7±0.2	6.3±0.2	1.02±0.00	7.5±0.1	92.5±0.1
	2nd	0.6±0.4	(1.01±0.00)		-	0.19±0.00	93.9±0.1	6.1±0.1	1.02±0.01	8.0±0.3	92.0±0.3
	3rd	0.7±0.3	(0.99±0.00)		-	0.20±0.00	93.2±0.9	6.8±0.9	1.04±0.00	7.3±0.2	92.6±0.2
	Total	6.4±0.2			-						
Newly formed[d] Aerobic Only Ba^{2+}	1st	n.a.	(10.4±0.5)		-	0.21±0.05	96.3±0.3	3.7±0.3	1.06±0.21	5.9±0.5	94.1±0.5
	2nd	n.a.	(10.5±0.3)		-	0.22±0.03	96.0±0.3	4.0±0.3	1.07±0.14	5.8±0.3	94.2±0.3
	3rd	n.a.	(10.7±0.5)		-	0.25±0.03	95.5±0.4	4.5±0.4	1.17±0.14	6.1±0.0	93.9±0.0
	Total	n.a.			-						
Newly formed[d] Aerobic Ba^{2+}/Mn^{2+}	1st	75.1±0.1	(0.14±0.00)	>99	(0.97±0.01)	0.11±0.00	61.9±0.1	39.9±0.1	1.98±0.01	11.3±0.0	88.8±0.0
	2nd	62.3±0.9	(0.15±0.00)	>99	(0.99±0.00)	0.23±0.00	55.2±0.0	44.8±0.0	3.01±0.01	8.7±0.0	91.3±0.0
	3rd	55.1±0.1	(0.15±0.00)	>99	(0.99±0.00)	0.31±0.01	41.7±1.2	58.3±1.2	4.20±0.04	7.2±0.0	92.8±0.0
	Total	64.0±0.3		>99							
Heated[d] Aerobic Ba^{2+}/Mn^{2+}	1st	37.7±0.4	(0.16±0.00)	27.2±0.5	(1.00±0.00)	0.04±0.00	79.8±0.2	20.2±0.2	1.14±0.00	15.9±0.1	84.1±0.1
	2nd	9.4±0.6	(0.16±0.00)	12.1±0.2	(1.01±0.00)	0.05±0.00	74.6±1.1	25.4±1.1	1.29±0.02	16.0±0.7	84.0±0.7
	3rd	8.2±1.5	(0.16±0.00)	9.9±2.1	(1.00±0.00)	0.06±0.00	76.3±0.5	23.7±0.5	1.35±0.00	16.1±0.1	83.9±0.1
	Total	18.4±0.6		16.4±0.9							
Newly formed[d] Aerobic Ba^{2+}/Mn^{2+}	1st	27.0±0.5	(0.93±0.01)	>99	(0.97±0.01)	0.27±0.00	78.8±0.2	21.2±0.2	1.95±0.02	6.6±0.1	93.4±0.1
	2nd	11.4±0.5	(0.91±0.00)	>99	(0.95±0.01)	0.39±0.00	65.2±0.5	34.8±0.5	2.95±0.03	5.3±0.5	94.7±0.5
	3rd	10.0±0.3	(0.93±0.01)	>99	(0.97±0.01)	0.52±0.00	59.3±0.1	40.7±0.1	3.97±0.01	5.4±0.1	94.6±0.1
	Total	16.1±0.0		>99							
Heated[d] Aerobic Ba^{2+}/Mn^{2+}	1st	6.0±0.4	(1.02±0.00)	12.3±1.3	(0.99±0.01)	0.10±0.00	90.3±0.9	9.7±0.9	1.25±0.02	14.0±0.1	86.0±0.1
	2nd	<1	(1.02±0.00)	4.9±1.3	(1.00±0.01)	0.10±0.00	87.4±0.6	12.6±0.6	1.21±0.00	14.0±0.1	86.0±0.1
	3rd	<1	(1.03±0.00)	5.4±1.8	(1.00±0.00)	0.13±0.00	83.6±0.0	16.4±0.0	1.36±0.00	16.8±0.2	83.2±0.2
	Total	~1.4		7.5±1.3							

Newly formed or heated BMOs (1 mM as Mn) were treated thrice with solutions of $Ba(NO_3)_2$ with or without 1 mM $Mn(NO_3)_2$ in 20 mM HEPES (pH 7.0). Bathing solutions were renewed thrice every 24 h

Table S1 Continued

Condition		Sequestration efficiency (%) (added ions; mM)				Two-step extraction					
		Ba^{2+}		Mn^{2+}		Total Ba^{2+} (mM)	Exchangeable (%)	Reducible (%)	Total Mn (mM)	Exchangeable (%)	Reducible (%)
Newly formed[d] Aerobic Ba^{2+}/Mn^{2+}	1st	10.7±0.9	(3.11±0.03)	>99	(1.01±0.01)	0.34±0.00	78.8±0.7	21.2±0.7	2.05±0.01	5.5±0.2	94.6±0.2
	2nd	4.2±0.9	(3.08±0.02)	>99	(1.00±0.01)	0.38±0.00	67.5±0.6	32.5±0.6	2.94±0.01	4.3±0.1	95.7±0.1
	3rd	5.7±1.0	(3.14±0.04)	>99	(1.01±0.01)	0.61±0.01	55.9±0.6	44.1±0.6	3.92±0.02	4.0±0.2	96.0±0.2
	Total	6.8±0.5		>99							
Heated[d] Aerobic Ba^{2+}/Mn^{2+}	1st	1.1±0.3	(3.36±0.01)	8.9±1.5	(1.06±0.01)	0.12±0.00	89.8±0.3	10.2±0.3	1.20±0.01	13.0±0.1	87.0±0.1
	2nd	1.5±0.2	(3.37±0.01)	5.5±3.2	(1.08±0.03)	0.16±0.00	85.4±1.1	14.6±1.1	1.33±0.03	13.6±0.2	86.4±0.2
	3rd	2.0±0.6	(3.39±0.01)	4.7±0.8	(1.06±0.01)	0.17±0.00	82.1±0.5	17.9±0.5	1.37±0.03	15.3±1.2	84.7±1.2
	Total	1.5±0.06		6.4±1.8							
Newly formed[d] Aerobic Ba^{2+}/Mn^{2+}	1st	n.a.	(11.0±1.1)	>99	(1.06±0.03)	0.41±0.02	80.1±1.1	19.9±1.1	2.21±0.08	5.7±0.2	94.3±0.2
	2nd	n.a.	(10.1±0.1)	>99	(1.07±0.03)	0.57±0.03	68.4±2.9	31.6±2.9	3.26±0.15	4.9±0.2	95.1±0.2
	3rd	n.a.	(10.8±0.2)	>99	(1.04±0.00)	0.71±0.01	55.6±1.8	44.4±1.8	4.13±0.07	4.6±0.2	95.4±0.2
	Total	n.a.		>99							
Heated[d] Aerobic Ba^{2+}/Mn^{2+}	1st	n.a.	(10.7±0.3)	6.3±0.8	(1.01±0.02)	0.15±0.01	92.7±0.4	7.3±0.4	1.27±0.07	14.5±0.2	85.5±0.2
	2nd	n.a.	(10.0±0.1)	8.5±5.6	(1.00±0.00)	0.17±0.06	90.0±1.9	10.0±1.9	1.28±0.44	14.5±0.3	85.5±0.3
	3rd	n.a.	(10.3±0.1)	10.5±1.0	(1.02±0.00)	0.20±0.01	82.5±0.4	17.5±0.4	1.61±0.09	14.6±0.2	85.4±0.2
	Total	n.a.		8.4±2.0							

Table S2 Data summary of sequestration experiments for Sr^{2+}, Ca^{2+}, or Mg^{2+} by newly formed BMOs with and without exogenous Mn^{2+} in 20 mM HEPES at pH7.0.

Condition		Sequestration efficiency (%) (added ions; mM)				Two-step extraction					
		$Sr^{2+}/Ca^{2+}/Mg^{2+}$		Mn^{2+}		Total $Sr^{2+}/Ca^{2+}/Mg^{2+}$ (mM)	Exchangeable (%)	Reducible (%)	Total Mn (mM)	Exchangeable (%)	Reducible (%)
Newly formed[d] Aerobic Sr^{2+}/Mn^{2+}	1st	n.a.	(11.3±0.4)	>99	(1.03±0.01)	0.31±0.01	>99	<1	2.19±0.06	8.1±0.3	91.9±0.3
	2nd	n.a.	(12.0±0.5)	>99	(1.04±0.01)	0.40±0.05	99.0±0.2	1.0±0.2	3.15±0.23	8.2±0.1	91.8±0.1
	3rd	n.a.	(11.4±0.5)	>99	(1.04±0.02)	0.48±0.05	98.1±0.3	1.9±0.3	3.98±0.01	9.4±0.4	90.6±0.4
	Total			>99							
Newly formed[d] Aerobic Ca^{2+}/Mn^{2+}	1st	n.a.	(11.8±2.5)	95.7±0.2	(1.15±0.06)	0.28±0.05	>99	<1	2.17±0.13	9.8±0.3	90.2±0.3
	2nd	n.a.	(11.0±0.2)	96.0±0.3	(1.17±0.02)	0.37±0.01	>99	<1	3.25±0.11	9.2±0.2	90.8±0.2
	3rd	n.a.	(11.1±0.3)	96.2±0.2	(1.16±0.04)	0.51±0.03	>99	<1	4.30±0.10	10.1±0.2	89.9±0.2
	Total			96.0±0.2							
Newly formed[d] aerobic Mg^{2+}/Mn^{2+}	1st	n.a.	(9.7±0.1)	98.5±0.1	(1.11±0.03)	0.18±0.03	98.4±1.6	1.6±1.6	2.21±0.21	10.7±0.1	89.3±0.1
	2nd	n.a.	(9.5±0.1)	98.5±0.1	(1.07±0.00)	0.37±0.02	82.2±6.9	17.8±6.9	3.46±0.27	10.9±0.2	89.1±0.2
	3rd	n.a.	(9.7±0.0)	98.6±0.0	(1.10±0.03)	0.41±0.01	84.1±0.7	15.9±0.7	4.39±0.14	10.9±0.4	89.1±0.4
	Total			98.5±0.1							

Newly formed BMOs (1 mM as Mn) were treated thrice with mixed solutions of 10 mM $Sr(NO_3)_2$, $Ca(NO_3)_2$, or $Mg(NO_3)_2$ with 1 mM $Mn(NO_3)_2$ in 20 mM HEPES (pH 7.0). Bathing solutions were renewed thrice every 24 h. Sequestration efficiency was not analyzed (n.a.) due to no measurable difference in concentration before and after sequestration experiments.

Table S3 Data summary of competitive sequestration experiments using newly formed BMOs (1 mM as Mn) in 10 mM HEPES buffer (pH 7.0) with and without exogenous Mn^{2+}.

Condition		Sequestration efficiency (%) (added ions; mM)			Two-step extraction								
		Ba^{2+}	$Sr^{2+}/Ca^{2+}/Mg^{2+}$	Mn^{2+}	Total Ba^{2+} (mM)	Exchangeable (%)	Reducible (%)	Total Ca^{2+}/Mg^{2+} (mM)	Exchangeable (%)	Reducible (%)	Total Mn (mM)	Exchangeable (%)	Reducible (%)
Newly formed[d] (mM) Aerobic $Ba^{2+}/Sr^{2+}/Mn^{2+}$	1st	23.4±0.5 (1.00±0.00)	5.5±0.4 (1.05±0.00)	>99 (1.00±0.00)	0.33±0.01	73.4±1.5	26.6±1.5	0.05±0.00	>99	<1	2.49±	9.6±0.2	90.4±0.2
	2nd	12.2±0.3 (0.99±0.00)	<1 (1.04±0.00)	>99 (1.02±0.00)	0.37±0.00	59.7±0.3	40.3±0.3	0.03±0.00	>99	<1	3.07±	6.2±0.0	93.8±0.0
	3rd	7.4±0.4 (0.99±0.00)	<1 (1.01±0.00)	>99 (0.98±0.01)	0.49±0.00	53.2±0.3	46.8±0.3	0.04±0.00	>99	<1	4.17±	6.0±0.1	94.0±0.1
	Total	14.4±0.3	~0.7	>99									
Newly formed[d] (mM) Aerobic Ba^{2+}/Sr^{2+}	1st	15.5±1.8 (1.04±0.00)	2.7±1.6 (1.01±0.01)	-	0.17±0.01	95.2±0.1	4.8±0.1	0.04±0.00	>99	<1	0.99±0.10	7.7±0.2	92.3±0.2
	2nd	0.2±2.9 (1.04±0.02)	-0.8±2.7 (1.00±0.02)	-	0.18±0.03	97.3±0.5	2.7±0.5	0.04±0.01	>99	<1	0.98±0.15	8.0±0.5	92.0±0.5
	3rd	-3.5±2.7 (1.04±0.04)	-2.4±3.2 (0.99±0.04)	-	0.17±0.01	94.4±0.1	5.6±0.1	0.03±0.00	>99	<1	0.99±0.06	6.1±0.1	93.9±0.1
	Total	~4.0	~0.2										
Newly formed[d] (mM) Aerobic $Ba^{2+}/Ca^{2+}/Mn^{2+}$	1st	22.8±2.8 (1.02±0.02)	2.7±0.2 (0.94±0.02)	>99 (1.02±0.02)	0.22±0.03	80.8±4.4	19.2±4.4	0.03±0.01	98.5±2.2	1.5±2.2	1.97±0.25	8.7±0.9	91.3±0.9
	2nd	10.6±1.2 (0.98±0.01)	n.d. (0.92±0.02)	>99 (1.00±0.01)	0.36±0.02	67.6±2.1	32.4±2.1	0.04±0.00	98.5±0.8	1.5±0.8	3.07±0.14	7.1±0.2	92.9±0.2
	3rd	8.2±2.3 (0.99±0.01)	n.d. (0.93±0.01)	>99 (1.02±0.00)	0.46±0.03	60.0±3.4	40.0±3.4	0.04±0.00	94.5±3.5	5.5±3.5	9.96±0.23	8.3±0.1	91.7±0.1
	Total	13.9±1.5	n.d.	>99									
Newly formed[d] (mM) Aerobic Ba^{2+}/Ca^{2+}	1st	16.7±2.6 (1.00±0.03)	n.d. (0.94±0.01)	-	0.17±0.03	96.0±0.7	4.0±0.7	0.04±0.01	>99	<1	1.10±0.15	7.8±0.2	92.2±0.2
	2nd	n.d. (0.99±0.01)	n.d. (0.92±0.00)	-	0.17±0.01	95.6±0.2	4.4±0.2	0.03±0.00	>99	<1	1.05±0.06	7.3±0.2	92.7±0.2
	3rd	n.d. (0.99±0.00)	n.d. (0.93±0.01)	-	0.19±0.00	96.1±0.2	3.9±0.2	0.04±0.00	97.1±2.6	2.9±2.6	1.11±0.02	10.4±0.2	89.6±0.2
	Total	n.d.	n.d.										
Newly formed[d] (mM) Aerobic $Ba^{2+}/Mg^{2+}/Mn^{2+}$	1st	25.3±1.6 (1.02±0.02)	n.d (1.07±0.01)	>99 (1.03±0.01)	0.25±0.01	77.4±1.0	22.6±1.0	0.02±0.00	>99	<1	2.10±0.07	7.6±0.2	92.4±0.2
	2nd	10.9±1.0 (0.98±0.01)	n.d. (1.05±0.01)	>99 (1.02±0.01)	0.38±0.02	67.3±3.6	32.7±3.6	0.02±0.00	>99	<1	2.99±0.20	7.6±0.4	92.4±0.4
	3rd	8.7±0.4 (0.99±0.01)	n.d. (1.05±0.00)	>99 (1.03±0.01)	0.48±0.01	58.5±1.7	41.5±1.7	0.02±0.00	>99	<1	4.10±0.11	6.9±0.3	93.1±0.3
	Total	14.9±0.6	n.d.	>99									
Newly formed[d] (mM) Aerobic Ba^{2+}/Mg^{2+}	1st	16.7±2.6 (1.00±0.03)	n.d. (1.07±0.01)	-	0.17±0.01	96.3±0.2	3.7±0.2	0.03±0.00	>99	<1	1.10±0.08	7.1±0.0	92.9±0.0
	2nd	n.d. (0.99±0.01)	n.d. (1.03±0.06)	-	0.16±0.02	96.8±0.2	3.2±0.2	0.03±0.00	>99	<1	0.97±0.08	8.4±0.3	91.6±0.3
	3rd	n.d. (0.99±0.00)	n.d. (1.06±0.01)	-	0.17±0.01	96.5±0.3	3.5±0.3	0.03±0.00	>99	<1	1.03±0.09	8.7±0.1	91.3±0.1
	Total	n.d.	n.d.										

Newly formed BMOs were treated trice with mixtures of 1 mM $Ba(NO_3)_2$ with 1 mM $Sr(NO_3)_2$, $Ca(NO_3)_2$, or $Mg(NO_3)_2$ in 20 mM HEPES (pH 7.0) with and without exogenous Mn^{2+}. Bathing solutions were renewed thrice every 24 h.

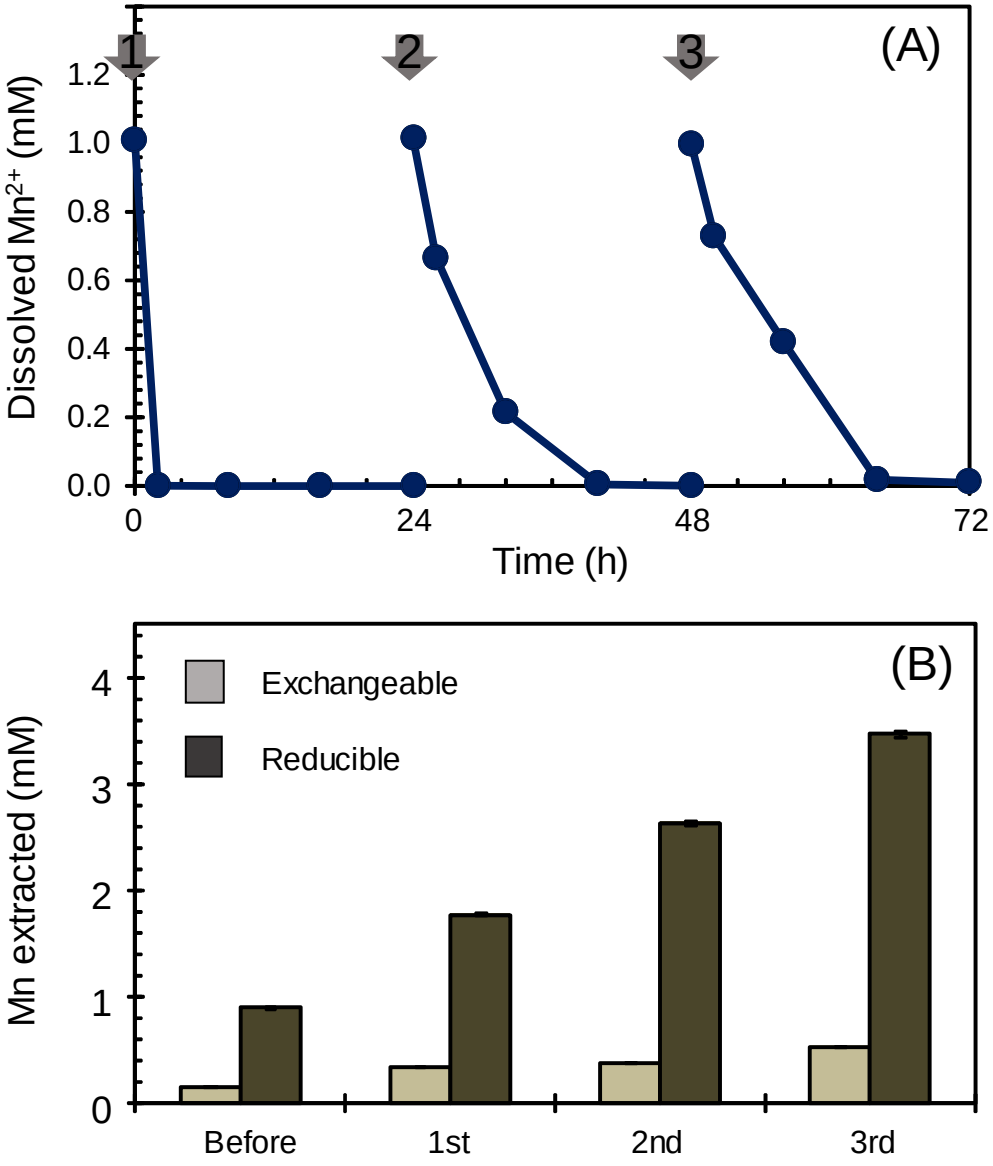

Figure S1 Mn^{2+} oxidation by newly formed biogenic manganese oxides (1 mM) in 1 mM $Mn(NO_3)_2$ in 20 mM HEPES (pH 7.0). (A) Dissolved Mn^{2+} and (B) exchangeable and reducible Mn in the solid phases assessed via two-step extraction. The bathing solutions were renewed every 24 h (indicated by arrows). Error bars represent standard deviations (n = 3).

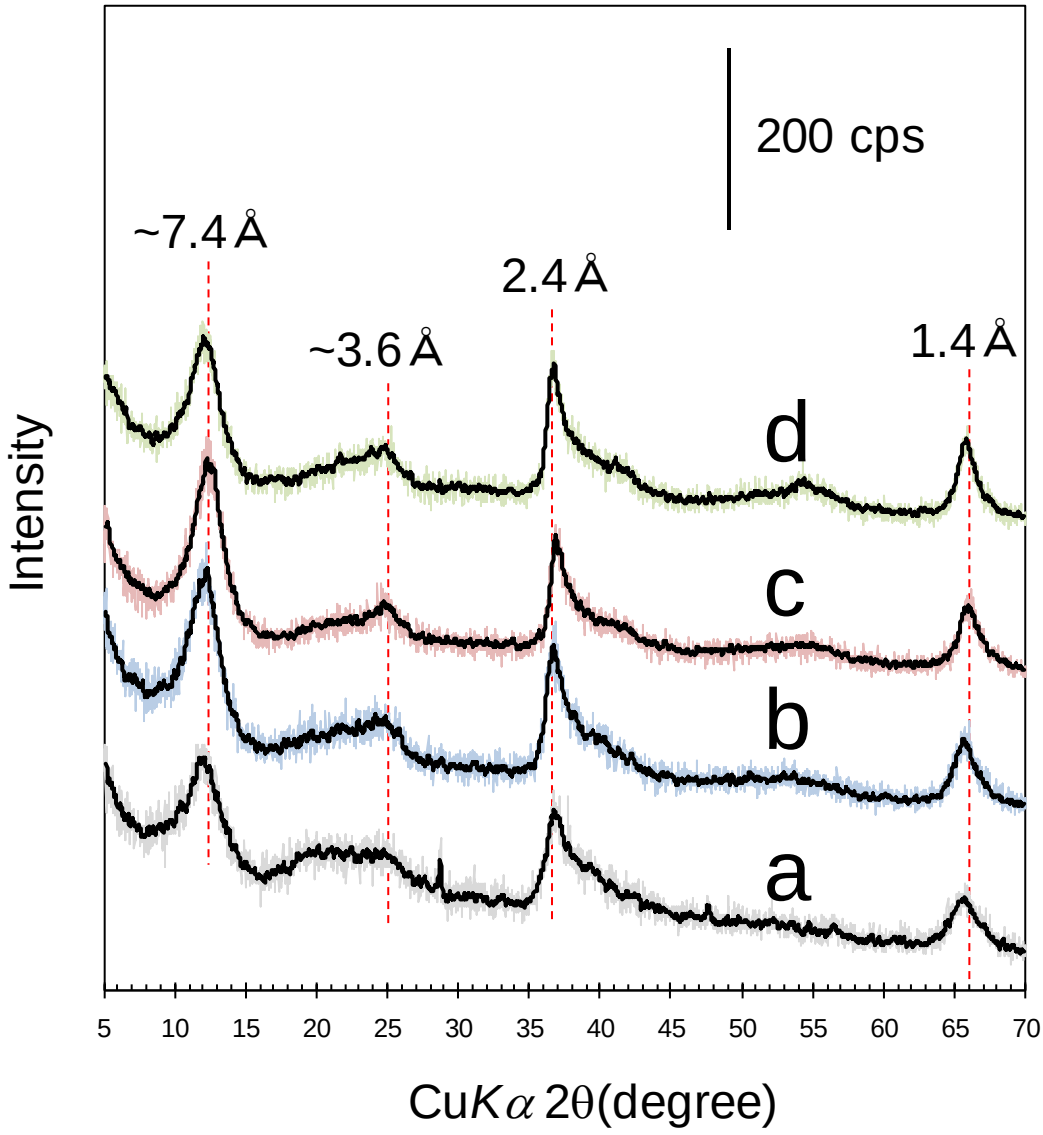

Figure S2 X-ray diffraction patterns of newly formed biogenic manganese oxides (1 mM as Mn) (a) not treated and treated with 1 mM $Mn(NO_3)_2$ in 20 mM HEPES (pH 7.0) (b) one, (c) two, and (d) three times, along with renewal of the bathing solutions every 24 h.

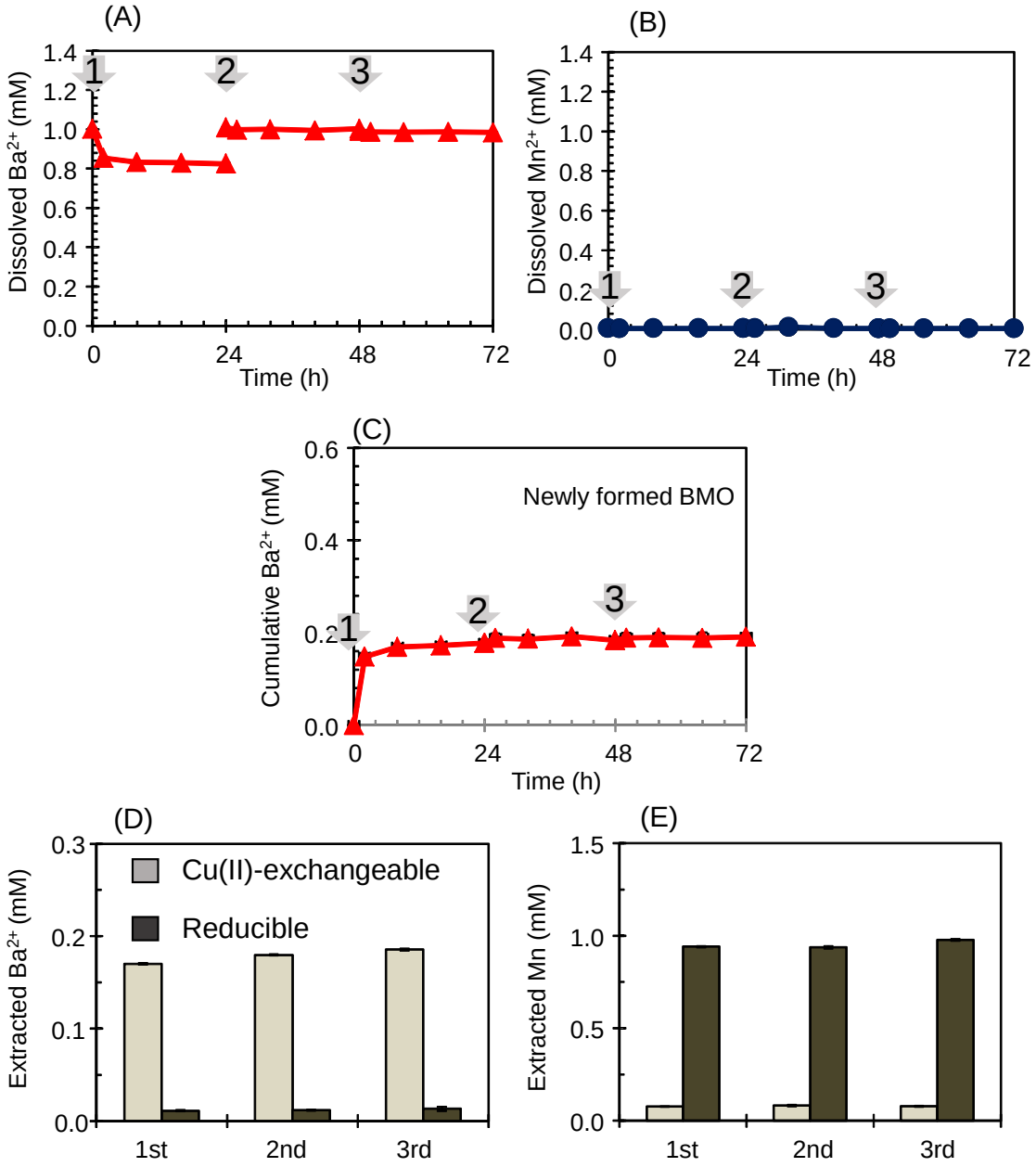

Figure S3 Repeated treatment of newly formed biogenic manganese oxides (1 mM) in 1 mM $Ba(NO_3)_2$ in 20 mM HEPES (pH 7.0). (A) Dissolved Ba^{2+}, (B) dissolved Mn^{2+}, and (C) cumulative Ba^{2+} sequestration. The bathing solutions were renewed every 24 h (indicated by arrows). Exchangeable and reducible (D) Ba and (E) Mn in the solid phases measured via two-step extraction. Error bars represent standard deviations (n = 3).

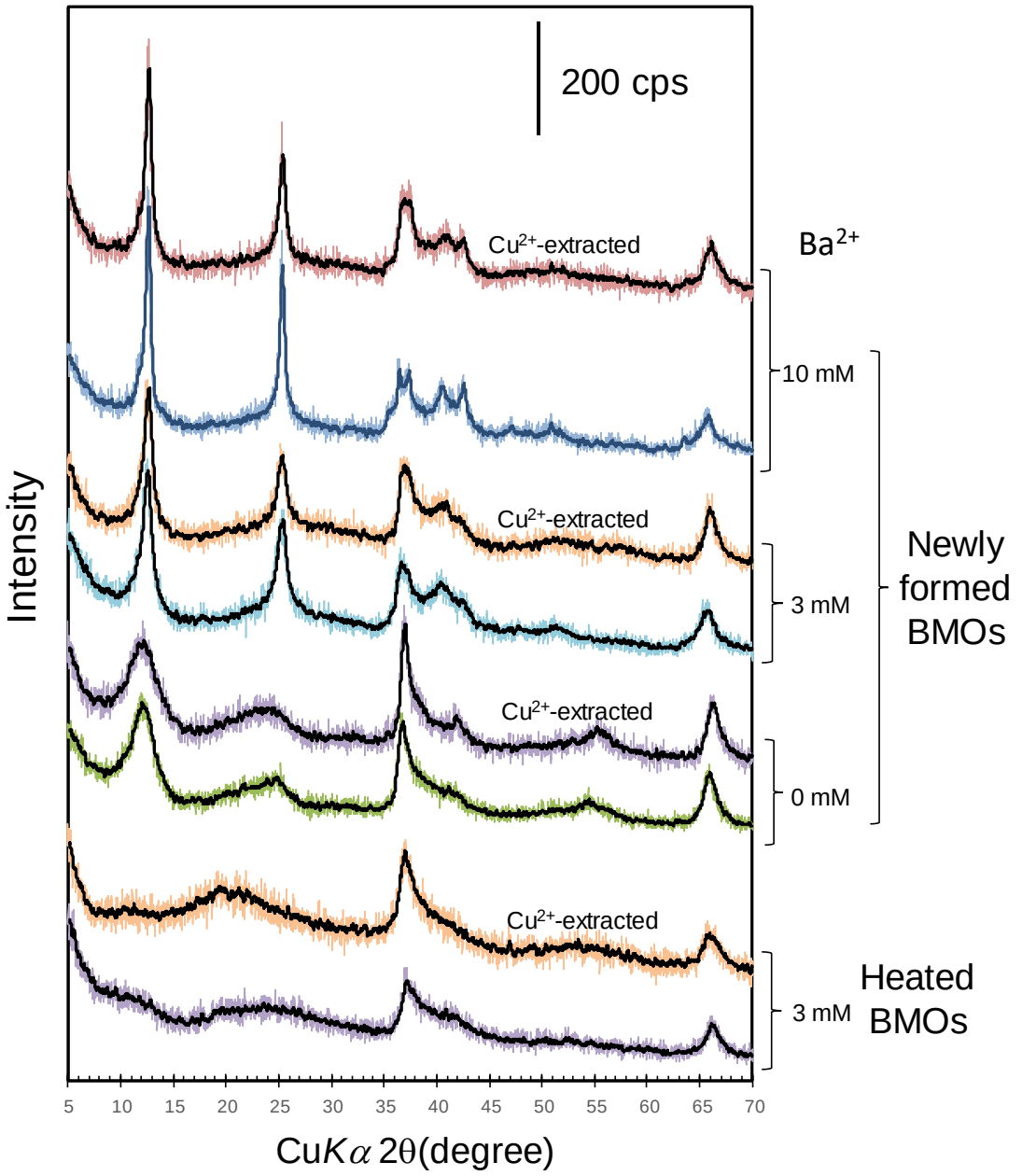

Figure S4 Effect of Cu^{2+}-extraction on X-ray diffraction (XRD) patterns. XRD analysis of newly formed and heated biogenic manganese oxides (1 mM as Mn) treated thrice with mixed solutions of $Ba(NO_3)_2$ (0–10 mM) with 1 mM $Mn(NO_3)_2$ in 20 mM HEPES (pH 7.0), along with the renewal of the bathing solutions every 24 h and subsequently extracted with 10 mM $Cu(NO_3)_2$ for 24 h.

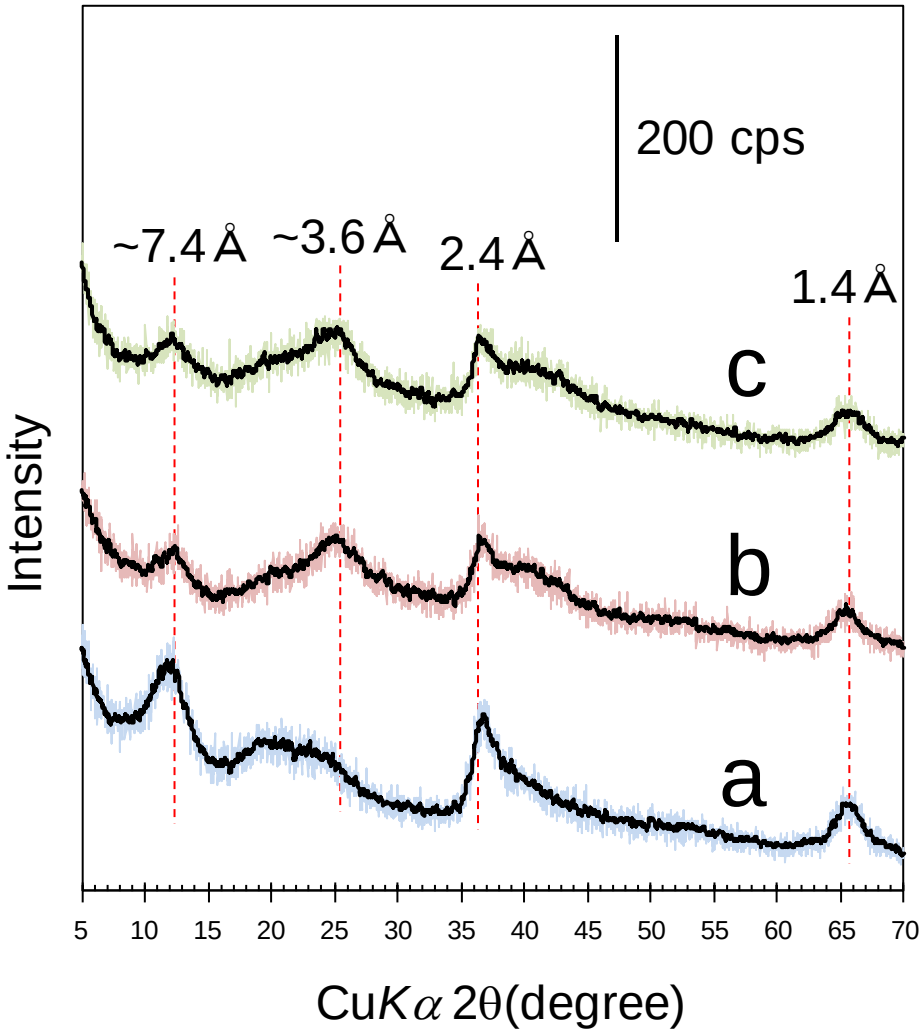

Figure S5 X-ray diffraction patterns of newly formed biogenic manganese oxides (1 mM as Mn) (a) not treated and treated thrice with (b) 1 mM and (c) 10 mM $Ba(NO_3)_2$ in 20 mM HEPES (pH 7.0). Bathing solutions were renewed every 24 h.

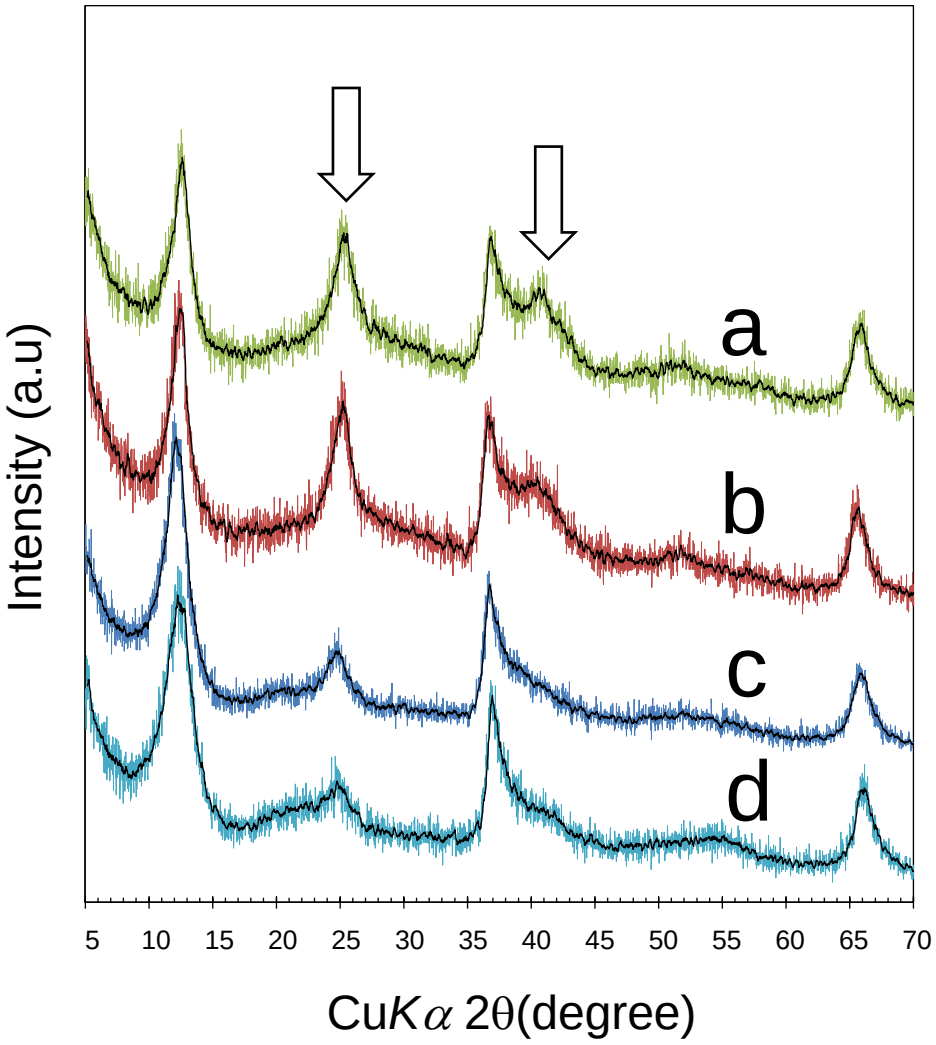

Figure S6 X-ray diffraction patterns of newly formed biogenic manganese oxides (1 mM as Mn) treated thrice in exogenous 1 mM $Mn(NO_3)_2$ with (a) 1 mM $Ba(NO_3)_2$, (b) 1 mM $Ba(NO_3)_2$ + 1 mM $Sr(NO_3)_2$ and (c) 1 mM $Sr(NO_3)_2$, and (d) without neither $Ba(NO_3)_2$ nor $Sr(NO_3)_2$ in 20 mM HEPES (pH 7.0). Bathing solutions were renewed every 24 h. XRD pattern for BMO treated with 1 mM $Ba(NO_3)_2$ + 1 mM $Sr(NO_3)_2$ is resemble to that with 1 mM $Ba(NO_3)_2$, but not with 1 mM $Sr(NO_3)_2$, especially in the parts suggested by the allows.