Supplementary materials for

Mycoextraction: Rapid Cadmium Removal by Macrofungi-Based Technology from Alkaline Soil

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Table S1. Quality control statistics of RNAseq by the software Sickle and SeqPrep .

Sample	Clean reads	Clean bases	Error rate (%)	Q20 (%)	Q30 (%)	GC content (%)
Cd0_1	50284816	7541807350	0.0243	98.34	94.78	53.52
Cd0_2	52633654	7899403838	0.0238	98.56	95.36	53.6
Cd0_3	51435936	7716921498	0.0238	98.54	95.3	53.6
Cd10_1	43930516	6591829794	0.0236	98.61	95.49	53.58
Cd10_2	57697230	8653297790	0.0239	98.52	95.23	53.64
Cd10_3	51056862	7659025117	0.0241	98.42	94.97	53.64
Cd150_1	44800136	6714851871	0.0241	98.45	95.04	53.47
Cd150_2	44809140	6721379404	0.0242	98.39	94.93	53.43
Cd150_3	58542646	8781481369	0.0238	98.54	95.31	53.54

Table S2. The typical differentially expressed genes (DEGs) among the three treatments for heatmap.

Gene_name	Cd0_1-count	Cd0_2-count	Cd0_3-count	Cd10_1-count	Cd10_2-count	Cd10_3-count	Cd150_1-count	Cd150_2-count	Cd150_3-count	Function
PLEOSDRAFT_1099226	0	0	0	0	0	0	948.18	2164.16	1560.92	Hypothetical protein
PLEOSDRAFT_1099171	0	0	0	0	0	0	311.5	1136.41	731.2	Proline metabolic
PLEOSDRAFT_153320	0	0	0	0	0	0	130.97	365.29	266.88	Methane metabolism
PLEOSDRAFT_1073785	0	0.69	0	0	0	0	68.43	310.06	298.83	Tryptophan biosynthetic
PLEOSDRAFT_1038303	0	0	0	0	0	0	45.93	110.91	123.52	Trancriptional regulation
PLEOSDRAFT_1030898	0	0	0	0	0	0	57.26	60.02	67.59	Cell wall remodeling
PLEOSDRAFT_1073153	0	0	0	0	0	0	45.71	70.2	231.22	DNA repair
PLEOSDRAFT_1023504	0	0	0	22.72	29.61	11.16	43.2	38.72	59.72	Metal efflux
PLEOSDRAFT_1101316	5.71	17.24	4.16	3.73	9.94	19.48	166.34	368.14	324.95	Hypothetical protein
PLEOSDRAFT_1099120	0	0	0	0	0	0	144.71	12.68	224.63	Metal efflux
PLEOSDRAFT_1048814	0	2.91	0	0	0	30.33	689.85	1529.69	270.29	Tryptophan biosynthetic
PLEOSDRAFT_1056091	8.33	19.95	17.36	0	16.7	40.01	161.09	249.52	196.11	ROS response
PLEOSDRAFT_1102750	15	11	15	12	20	9	159	612	64	Hypothetical protein

PLEOSDRAFT_1091079	305	364	315	163	333	245	3377	5040	1368	Hypothetical protein
PLEOSDRAFT_162256	0	4.53	5.39	0	7.57	3.78	5.51	288.66	206.98	ROS response
PLEOSDRAFT_1114221	92	91	81	19.02	138.27	35.53	2218.6	5750.51	288.14	Hypothetical protein
PLEOSDRAFT_1114672	834.44	822.11	863.98	1916.7	1686.9	2708.54	7162.29	9262.7	3887.08	Hypothetical protein
Hydph16	107	108	77	132	151	116	1215	2028	190	Hydrophobin
PLEOSDRAFT_1119533	506.85	490.86	531.91	493.75	264.59	467.02	3193.62	3122.33	4468.35	Cell wall remodeling
PLEOSDRAFT_166408	4.08	1.1	0	1.55	8.53	12.75	84.68	32.92	15.32	Hypothetical protein
PLEOSDRAFT_1103449	3052.83	3369.64	3151.36	3319.72	5810.84	3697.7	0	0	0	Hypothetical protein
PLEOSDRAFT_152955	1231.46	1346.23	1302.22	1502.91	2110.18	1450.86	0	0	0	ROS induction
PLEOSDRAFT_1059138	1227.23	1277.16	1203.15	1279.14	1868.29	1333.05	0	0	0	Cell cycle
PLEOSDRAFT_165944	369.28	214.31	307.3	250.8	446.01	225.81	0	0	0	Hypothetical protein
PLEOSDRAFT_1044780	287.23	224.31	266.54	268.51	423.51	237.6	0	0	0	Cell wall remodeling
PLEOSDRAFT_1090102	94.02	123.76	118.68	0	65.08	48.57	0	0	0	Hypothetical protein
PLEOSDRAFT_1047466	93.63	97.22	104.31	114.47	240.82	221.11	0	0	0	Peroxisomes proliferate
PLEOSDRAFT_1101314	69.65	163.71	94	0	119.11	132.52	0	0	0	DNA repair
PLEOSDRAFT_1114501	140.38	68.29	92.03	62.17	159.56	6.27	0	0	0	Glucose transporters
PLEOSDRA	51.	60.	79.	59.6	150.8	207.5	0	0	0	Hypothetical

FT_1070098	35	34	88			8				protein
PLEOSDRAFT_31793	76.15	78.53	84.72	38.86	95.13	0	0	0	6.18	protein folding
PLEOSDRAFT_1102088	81.45	85.27	138.8	112.74	8.74	81.4	2.51	0	7.48	hypothetical protein
PLEOSDRAFT_1051043	56.34	12.84	39.86	53.31	71.73	119.52	0	0	0	Transcriptional repressor of defense genes
PLEOSDRAFT_1092804	3285.39	3295.41	3238.28	3384.46	4952.39	3212.49	85.33	7.27	15.77	ROS induction
PLEOSDRAFT_1046185	17.98	46.39	12.92	0	79.91	0	0	0	0	TCA
PLEOSDRAFT_1093790	5142	5290	5069	110	2275	7027	15	250	48	Hypothetical protein
PLEOSDRAFT_29565	4936.57	4949.35	4891.17	5425.67	8536.4	7681.14	591.71	302.99	607.78	Glyoxylate cycle
PLEOSDRAFT_1102787	171	172	173	124	205	110	14	9	29	Hypothetical protein
PLEOSDRAFT_1104249	219	232	209	165	296	168	15	3	36	Hypothetical protein
PLEOSDRAFT_158698	745.08	843.16	777.36	651.24	1173.44	929.17	29.87	4.23	1.8	Hypothetical protein
PLEOSDRAFT_1042152	0	0	0	1133.51	1701.57	1407.89	366.71	44.4	15	Phosphate-transporting related
PLEOSDRAFT_1032757	0	0	0	1174.86	1267	946.1	0	0	0	Microtubule motor
PLEOSDRAFT_40476	0	0	0	66.08	93.6	94.24	0	0	0	Hypothetical protein
PLEOSDRAFT_1108534	0	0	0	62.86	52.34	62.68	0	0	0	Hypothetical protein
PLEOSDRAFT_158699	0	4.02	0	245.67	445.55	699.32	284.34	5.97	0	Hypothetical protein
PLEOSDRA	194	227	182	771	1818	2719	420	142	790	Osmoregulation

FT_1077179										protein
PLEOSDRAFT_1091180	29	55	29	148	271	231	74	1407	80	Cysteine synthesis
PLEOSDRAFT_1100834	180.05	277.78	215.31	890.04	1281.34	1003.28	114.89	8.12	113	Hypothetical protein
PLEOSDRAFT_1087194	360.12	549.63	384.83	1225.22	2137.39	2251.15	2063.71	2344.24	620.71	Oxidoreductase
PLEOSDRAFT_1114116	3208.14	3474.26	3359.99	12992.77	11737.28	14251.25	19820.29	17231.59	25014.42	Hypothetical protein
PLEOSDRAFT_1113522	1097	1149	1201	2633	4979	6284	2248	1520	6575	Metal efflux
PLEOSDRAFT_1023504	0	0	0	22.72	29.61	11.16	43.2	38.72	59.72	Metal efflux
PLEOSDRAFT_1048474	2002.08	1950	1926.62	4920.99	6799.76	9495.16	4133.08	366.64	6469.22	ROS response
PLEOSDRAFT_1025748	3.44	8.26	12.95	34.44	55.54	56.66	25.35	0	11.77	Protein Kinases
PLEOSDRAFT_1077497	50	39	54	119	194	250	296	91	42	Hypothetical protein
PLEOSDRAFT_1082375	21.43	20.15	16.52	75.67	66.38	96.78	161.4	112.97	85.14	Hypothetical protein
PLEOSDRAFT_48690	321.29	486.29	596.36	1719.46	1224.15	2950.1	1848	479.77	664.72	Inorganic pyrophosphatase (PolyP)
PLEOSDRAFT_1097837	198	189	186	415	498	1034	990	418	1767	Hypothetical protein
CorA3	59.32	61.14	134.27	197.12	309.7	397.91	51.8	10.33	0	Metal efflux
PLEOSDRAFT_1105181	242.68	277.4	221.15	630.31	564.24	1199.4	354.78	93.46	572.18	Hypothetical protein
PLEOSDRA	131	153	135	0	0	0	3545.6	7270.0	4601.6	Hypothetical

FT_1051773	9.59	1.46	0.34				3	4	5	protein
PLEOSDRAFT_1093665	685.09	879.51	683.43	0	0	0	843.2	701.26	1390.19	Hypothetical protein
PLEOSDRAFT_1063163	638.13	674	797.68	0	0	0	679.87	1153.06	2528.31	Cell cycle
PLEOSDRAFT_1111600	162.8	208.31	174.11	0	0	0	0	0	256.95	Peroxisomal membrane protein
PLEOSDRAFT_61209	288.61	163.2	188.41	0	0	0	424.33	1166.5	1037.99	Ribosome biogenesis
PLEOSDRAFT_1113759	4907	5043	4951	234	220	137	7848	7497	1024	Heme-binding protein
PLEOSDRAFT_1088548	18934.21	20242.97	20354.08	818.77	1256.13	602.87	4097.7	11898.9	14143.97	Endopeptidase
PLEOSDRAFT_1090711	859.41	917.95	841.62	54.88	28.73	28.1	196.1	998.64	1958.59	DNA damage response
PLEOSDRAFT_1108098	2152	2262	2122	114	250	148	457	1458	2310	Hypothetical protein
PLEOSDRAFT_1060503	817.74	873.88	860.53	56.3	69.16	91.2	489.59	606.28	534.93	Phospholipid synthesis
PLEOSDRAFT_1062997	77.58	54.35	71.73	1.93	0	0	24.96	144.17	11.4	Hypothetical protein
PLEOSDRAFT_1068800	114.57	105.64	41.33	1.15	0	0	688.29	863.49	512.11	Hypothetical protein
PLEOSDRAFT_1034502	6384.91	6632.37	6512.08	110.19	453.17	310.25	608.39	19890.9	13860.25	Cell wall
PLEOSDRAFT_1061253	9126	10007	10129	722	1447	726	590	6166	4248	Aldo keto reductase
PLEOSDRAFT_1090123	791.62	908.93	867.23	67.59	75.13	109.64	24.46	59.02	663.72	Hypothetical protein
PLEOSDRA	127	134	136	129	170	149	720	2431	1533	MAPK

FT_1096954	8	6	1						
PLEOSDRAFT_1111581	88.23	89.49	58.28	3.06	5.04	0	4.04	85.89	199.91	Hypothetical protein
PLEOSDRAFT_34588	3343	3767	3480	376	553	300	1020	2723	2679	Aldo keto reductase
PLEOSDRAFT_1114418	875.65	873.75	770.11	64.02	150.49	61.61	224.16	419.52	1386.15	Cell cycle
PLEOSDRAFT_1108164	1366	1503	1479	75	178	228	29	480	1462	Hypothetical protein

Table S3. COG classification of differentially expressed genes (DEGs) in the Cd treatments.

Type	Functional Categories	A_vs._C	A_vs._B	B_vs._C
INFORMATION STORAGE AND PROCESSING	[A] RNA processing and modification	1	0	0
INFORMATION STORAGE AND PROCESSING	[B] Chromatin structure and dynamics	8	2	15
INFORMATION STORAGE AND PROCESSING	[J] Translation, ribosomal structure and biogenesis	8	10	51
INFORMATION STORAGE AND PROCESSING	[K] Transcription	3	2	10
INFORMATION STORAGE AND PROCESSING	[L] Replication, recombination and repair	21	1	53
CELLULAR PROCESSES AND SIGNALING	[D] Cell cycle control, cell division, chromosome partitioning	9	2	19
CELLULAR PROCESSES AND SIGNALING	[M] Cell wall/membrane/envelope biogenesis	7	4	17
CELLULAR PROCESSES AND SIGNALING	[O] Posttranslational modification, protein turnover, chaperones	15	12	52
CELLULAR PROCESSES AND SIGNALING	[T] Signal transduction mechanisms	12	6	26
CELLULAR PROCESSES AND SIGNALING	[U] Intracellular trafficking, secretion, and vesicular transport	2	0	2
CELLULAR PROCESSES AND SIGNALING	[V] Defense mechanisms	3	0	6
CELLULAR PROCESSES AND SIGNALING	[Z] Cytoskeleton	4	1	9
METABOLISM	[C] Energy production and conversion	16	28	54
METABOLISM	[E] Amino acid transport and metabolism	19	19	53
METABOLISM	[F] Nucleotide transport and metabolism	5	11	25
METABOLISM	[G] Carbohydrate transport and metabolism	33	31	88
METABOLISM	[H] Coenzyme transport and metabolism	4	2	18
METABOLISM	[I] Lipid transport and metabolism	25	13	41
METABOLISM	[P] Inorganic ion transport and metabolism	7	6	17
METABOLISM	[Q] Secondary metabolites biosynthesis, transport and catabolism	14	16	33
POORLY CHARACTERIZED	[R] General function prediction only	26	31	82
POORLY CHARACTERIZED	[S] Function unknown	14	4	27

Table S4. Typical upregulated genes of the 150mg/kg Cd treatment.

Description	Gene name	log2fc	Padjust	Annotation
Major facilitator Superfamily	PLEOSDRAFT_1099120	4.009	1.41E-08	ABCB-BAC
	PLEOSDRAFT_22795	1.429	3.94E-10	Major facilitator Superfamily
	PLEOSDRAFT_1112409	1.363	0.006909	Major facilitator Superfamily
	PLEOSDRAFT_1113522	1.317	0.004382	Major facilitator Superfamily
	PLEOSDRAFT_19893	1.222	0.000857	MFS DHA1 amino acid exporter
	PLEOSDRAFT_1065674	1.141	2.41E-13	Major facilitator Superfamily
	PLEOSDRAFT_1113350	1.002	2.01E-07	Major facilitator Superfamily
Multidrug and toxic compound extrusion(MATE)	PLEOSDRAFT_1105315	1.816	0.004615	Mate efflux family protein
Metal-phosphate complexes efflux	PLEOSDRAFT_1103984	1.127	0.02195	Putative inorganic phosphate transporter
Ca(2+)/H(+) exchanger	PLEOSDRAFT_161300	1.028	0.002793	Vacuolar Ca(2+)/H(+) exchanger
Na(+) transporter	PLEOSDRAFT_1101283	1.26	0.02286	Na(+)/H(+) antiporter C3A11.09
	PLEOSDRAFT_1063536	1.001	6.69E-08	Sodium transport atpase 5
K(+) transporter	PLEOSDRAFT_1050754	1.123	1.89E-11	Potassium voltage-gated channel
Endocytosis	PLEOSDRAFT_1023504	4.532	1.55E-12	Endocytosis
Copper resistance protein CRF1	PLEOSDRAFT_1060467	1.775	5.41E-06	Copper resistance protein CRF1
Cell wall remodeling	PLEOSDRAFT_1119533	2.734	8.38E-78	Carbohydrate-binding module family 13 protein
	VMH3	2.023	0.009956	Structural constituent of cell wall
	PLEOSDRAFT_1078467	1.874	2.33E-06	Carbohydrate esterase family 4 protein
	PLEOSDRAFT_47325	1.84	0.01823	Structural constituent of cell wall
	PLEOSDRAFT_1079778	1.784	1.41E-08	Glycoside hydrolase family 18 protein

	PLEOSDRAFT_1076572	1.768	0.000797	Glycoside hydrolase family 18 protein
	PLEOSDRAFT_1043732	1.67	3.05E-09	Glycoside hydrolase family 18 protein
	PLEOSDRAFT_1089150	1.619	4.35E-06	Scp-like extracellular
	PLEOSDRAFT_1060842	1.491	7.20E-11	Glycosyltransferase family 4 protein
	PLEOSDRAFT_1067159	1.432	1.04E-12	Carbohydrate esterase family 4 protein
	PLEOSDRAFT_186160	1.356	1.14E-07	Hypothetical protein PLEOSDRAFT_186160
	PLEOSDRAFT_1057207	1.107	0.03833	Carbohydrate esterase family 4 protein
	PLEOSDRAFT_1089767	1.039	0.02753	
Cysteine/iron-sulfur centers)/ thiol groups	PLEOSDRAFT_1111638	1.903	0.01139	Cytosolic Fe-S cluster-deficient protein 1
	PLEOSDRAFT_1071644	1.842	0.005331	Small cysteine-rich protein
	PLEOSDRAFT_1060726	1.648	3.93E-06	SH3 and cysteine rich domain 3
	HTP1	1.569	0.000506	Heme-thiolate peroxidase
Antioxidant	PLEOSDRAFT_1056091	3.408	1.59E-18	
	DyP3	1.202	0.02341	Dyp-type peroxidase
	HTP1	1.569	0.000506	Heme-thiolate peroxidase
Autophagy	PLEOSDRAFT_1097387	2.497	2.63E-16	Autophagy related protein
	PLEOSDRAFT_1040572	2.055	0.008622	Autophagy-related E2-like conjugation enzyme ATG3
	PLEOSDRAFT_24680	1.331	1.38E-11	Deubiquitinating enzyme 2
Laccase	PLEOSDRAFT_1097387	1.879	0.000606	Lacc3
	PLEOSDRAFT_1040572	1.521	0.02739	Lacc6
	PLEOSDRAFT_24680	1.054	0.004489	Lacc9
Tryptophan synthesis	PLEOSDRAFT_1073785	5.597	2.28E-20	Tryptophan biosynthesis
	PLEOSDRAFT_1048814	3.44	2.80E-06	Tryptophan biosynthesis
Proline synthesis	PLEOSDRAFT_1099171	7.325	3.66E-39	Argininosuccinate lyase
	PLEOSDRAFT_1065795	1.793	0.002247	Agmatinase
Cytochrome P450	PLEOSDRAFT_1111004	2.248	8.20E-06	Cytochrome P450 98A1

	PLEOSDRAFT_1033557	1.389	0.02708	Cytochrome P450 98A2
Heat shock protein (HSP)	PLEOSDRAFT_1053318	1.647	4.75E-07	Hsp20
Hydrophobin (hyd)	Hydph16	2.787	6.04E-06	Hydph16
	Hydph20	1.888	8.68E-05	Hydph20
Stress-inducible protein (SF)	PLEOSDRAFT_1090687	1.066	6.16E-07	Environmental stress-inducible protein
	PLEOSDRAFT_166368	2.238	6.11E-08	Stress factor 1

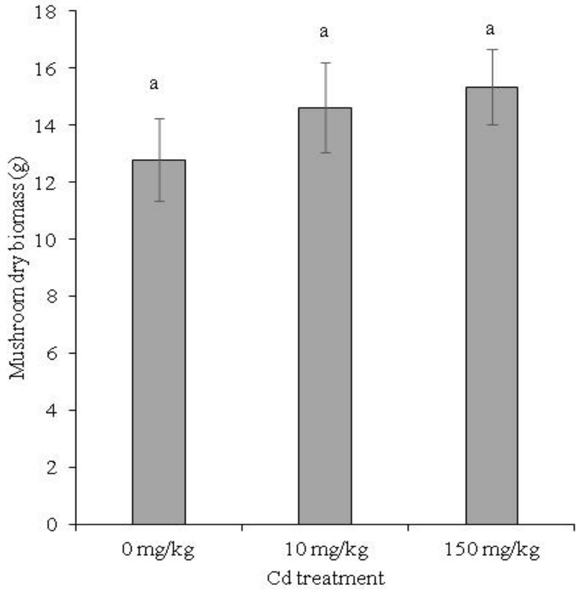
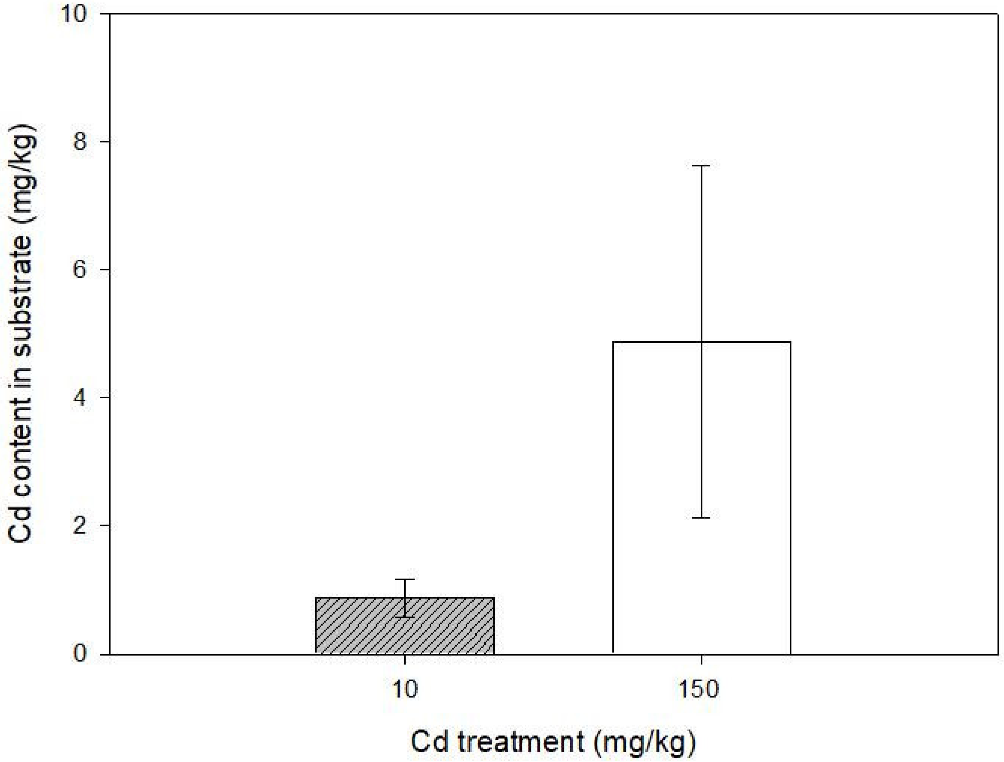
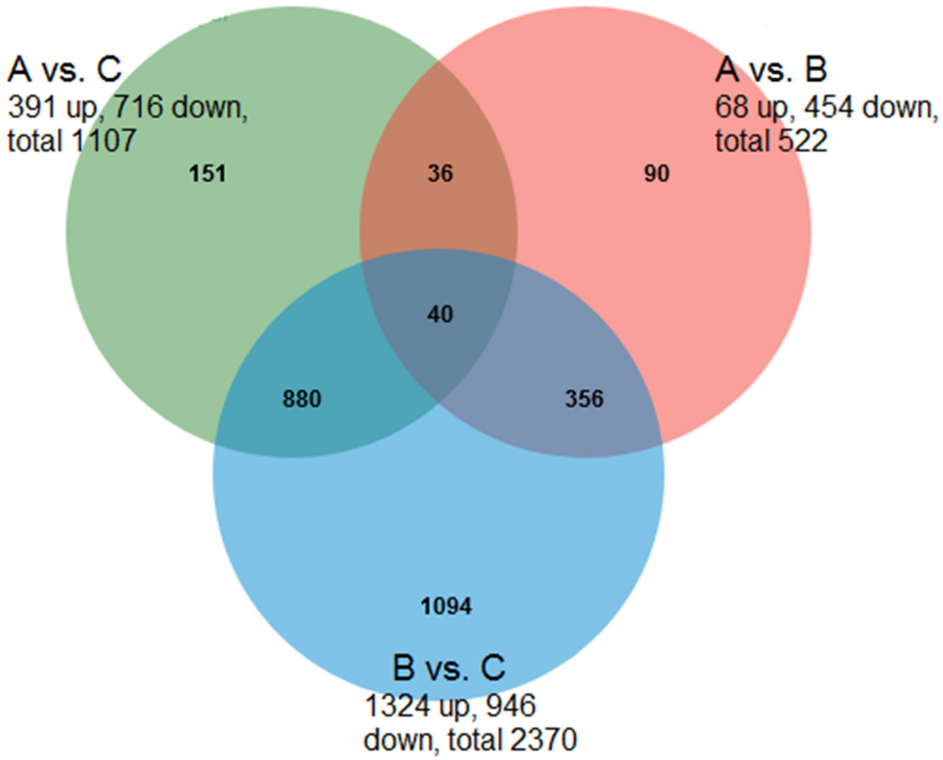


Figure S1. Total dry biomass of the fruiting body in the Cd treatments and control in this study ($n = 3$). Bars with different letters are significantly different at $p \leq 0.05$ (one-way ANOVA).



Suppl. Fig. 2 Cd content in the substratum of the two Cd treatments after 35-d mushroom cultivation.



Suppl. Fig. 3 A Venn diagram showing DEGs overlap of the fruiting body from the Cd treatments and control. A,B,C represent 0, 10 and 150 mg Cd/kg soil treatment, respectively. A minimum of twofold expressional difference (i.e., log2 ratio ≤ −1.0 or ≥1.0) in the paired libraries was used as a standard to judge each DEG at corrected p value (p adjust) of 0.05 or less.