Supplementary Information

Table S1. Overview of group-specific primers used in DGGE and qPCR assays. Primer annealing temperatures were experimentally optimized to maximize group specificity of the amplification. Twenty-five µl reaction volumes, containing 1 µL sample DNA, 1X GoTaq® Flexi Buffer (Promega, Madison, WI), 2.0 mM $MgCl_2$, 0.2 mM dNTPs, 1.25U Taq Polymerase and 10 µmol of each primer, were subjected to 94°C for 5 min, 35 cycles of 94°C for 0.5 min, dedicated annealing temperature for 0.5 min and 72°C for 1 min, and final elongation at 72°C for 5 min. In bacteria-group specific PCR assays Eub338 or Eub518 were used in combination with group-specific primer, except for *Actinobacteria* where Eub1392 was used as reverse primer. Relative abundances of groups were obtained by dividing with the total bacteria number, resulting from PCRs with primers Eub338 and Eub518. qPCR standards were prepared by cloning PCR-amplified 16S rRNA genes of reference stains into the pGEM-T Easy plasmid vector (Promega, Madison, WI). PCR products amplified from plasmid vectors by using group-specific primers were used as real-time PCR standards [1, 2].

Target group	Primer sequence (5'-3')	Primer name	Reference strain	Gradient Range (%)	Annealing Temperature (°C)	Reference
Bacteria	ACTCCTACGGGAGGCAGCAG	Eub338[2](F)[3]		NA[4]	60	[3]
Bacteria	ATTACCGCGGCTGCTGG	Eub518(R)[3]		NA[4]	60	[3]
Bacteria	ACGGGCGGTGTGTACA	Eub1392(R)[3]		NA[4]	60	[4]
Alphaproteobacteria[1]	TCTACGRATTTCACCYCTAC	ALF685(R)[3]	*Paracoccus denitrificans*	40-55	57	[1]
Betaproteobacteria[1]	TCACTGCTACACGYG	Bet680(R)[3]	*Burkholderia plantarii*	40-55	57	[1]
Gammaproteobacteria[1]	GATTGAACGCTGGCGGCAGG	GAM-m[2](F)3	*Shewanella putrefaciens 200*	30-55	58	[5]
Actinobacteria	GGTACAGAGGGCTGCGATAC	Actino1175F[2](F)[3]	*Rhodococcus opacus*	40-65	58	[6]
Acidobacteria	GAT CCTGGCTCAGAATC	Acid31[2](F)[3]	*Acidobacterium capsulatum*	35-55	57	[1]
Firmicutes	GCAGTAGGGAATCTTCCG	Lgc353(F)[3]	*Bacillus subtilis*	35-65	60	[1]
Bacteroidetes	GTACTGAGACACGGACCA	Cfb319(F)[3]	*Bacteroides cellulosolvens*	35-55	58	[1]
Fungi	TCCGTAGGTGAACCTGCGG	ITS1f[2](F)[3]	*Cladosporium cladosporioides f. pisicola*	30-55	53	[1]
Fungi	CGCTGCGTTCTTCATCG	5.8s(R)[3]	*Cladosporium cladosporioides f. pisicola*	30-55	53	[1]

[1] This class is within the phylum Proteobacteria.
[2] Primer with GC-clamp of 5'-CGCCCGGGGCGCGCCCCGGGCGGGGCGGGGGCACGGGGGG-3' for DGGE
3(F) Forward primer, (R) reverse primer.
[4] NA: not applicable or data not available

Table S2. (**A**) Soil chemistry and (**B**) ANOVA analysis for the effect of original pH and copper arrangement on measured soil chemical parameters

A

original pH	original Cu(kg/ha)*	pH	Total Cu (mg/kg)	Cu_{CaCl2} (µg/kg)	DOC (mg/kg)
4	0	4.03± 0.08	30.0± 6.9	980.8± 256.2	100.9± 4.3
4.7	0	4.44± 0.21	32.8± 13.0	608.7± 549.8	84.5± 11.9
5.4	0	4.74± 0.22	40.4± 9.8	376.3± 57.8	86.0± 8.8
6.1	0	5.11± 0.11	42.2± 6.6	232.0± 36.1	78.8± 10.6
4	250	3.96± 0.07	48.0± 6.4	2153.5± 499.6	102.3± 12.7
4.7	250	4.49± 0.20	74.2± 12.5	1482.0± 552.6	89.9± 9.9
5.4	250	4.89± 0.21	63.2± 7.7	560.3± 147.4	84.5± 13.0
6.1	250	4.96± 0.25	53.5± 9.1	422.5± 172.9	80.8± 4.8
4	500	4.10± 0.32	69.5± 17.7	3190.3± 728.6	97.5± 8.8
4.7	500	4.42± 0.05	80.9± 7.3	1906.8± 662.2	90.1± 11.8
5.4	500	4.51± 0.04	81.4± 7.9	1546.3± 257.2	88.0± 3.0
6.1	500	5.08± 0.10	82.2± 8.2	585.3± 113.7	80.4± 18.2
4	750	4.12± 0.10	86.1± 8.2	4147.8± 1362.5	95.7± 18.2
4.7	750	4.41± 0.23	85.3± 13.5	2423.8± 631.0	85.9± 4.6
5.4	750	4.70± 0.04	96.4± 6.8	1461.8± 140.6	78.5± 17.2
6.1	750	5.15± 0.33	111.3± 14.6	895.5± 539.0	75.9± 6.9

*assuming a ploughing depth of 30 cm and soil density of 1.25 g/cm^3, the original Cu concentration of 750 kg/ha corresponds to about 200 mg/kg soil

B

	pH	Total Cu	Cu_{CaCl2}	DOC
original pH	●	**	●	●
original Cu	ns	●	●	ns
original pH* original Cu	ns	ns	ns	ns

ns: not significant; +: P≤0.10; *P≤0.05; **: P≤0.01; ●: P≤0.001

Table S3. Significant GO terms found between low and high copper concentration (copper) and low and high soil pH (pH).

GO term	Description (Biological Process)	Annotated [a]	Significant [b]	P-value [c]
Copper				
GO:0045045	secretory pathway	64	6	0.0050
GO:0032940	secretion by cell	68	6	0.0067
GO:0046903	secretion	76	6	0.0116
GO:0045055	regulated secretory pathway	22	3	0.0176
GO:0016192	vesicle-mediated transport	110	7	0.0198
GO:0006091	generation of precursor metabolites and energy	65	5	0.0236
GO:0006635	fatty acid beta-oxidation	10	2	0.0258
pH				
GO:0051260	protein homo-oligomerization	12	4	0.0045
GO:0006308	DNA catabolic process	8	3	0.0100
GO:0006085	acetyl-CoA biosynthetic process	3	2	0.0107
GO:0006297	nucleotide-excision repair, DNA gap filling	3	2	0.0107
GO:0015671	oxygen transport	3	2	0.0107
GO:0006044	N-acetylglucosamine metabolic process	11	3	0.0203
GO:0006200	ATP catabolic process	4	2	0.0206
GO:0006221	pyrimidine nucleotide biosynthetic process	4	2	0.0206
GO:0007004	telomere maintenance via telomerase	4	2	0.0206
GO:0000302	response to reactive oxygen species	18	4	0.0208
GO:0001737	establishment of imaginal disc-derived wing hair orientation	5	2	0.0329
GO:0022611	dormancy process	5	2	0.0329
GO:0042067	establishment of ommatidial polarity	5	2	0.0329
GO:0008406	gonad development	13	3	0.0407
GO:0002526	acute inflammatory response	7	3	0.0465
GO:0007173	Epidermal growth factor receptor signaling pathway	6	2	0.0474
GO:0016319	mushroom body development	6	2	0.0474
GO:0015986	ATP synthesis coupled proton transport	14	3	0.0496

[a]: the total number of genes present in this GO term. [b]:the number of genes found for this GO term in the significant gene list and [c]: P value calculated by the weighted Fisher exact test.

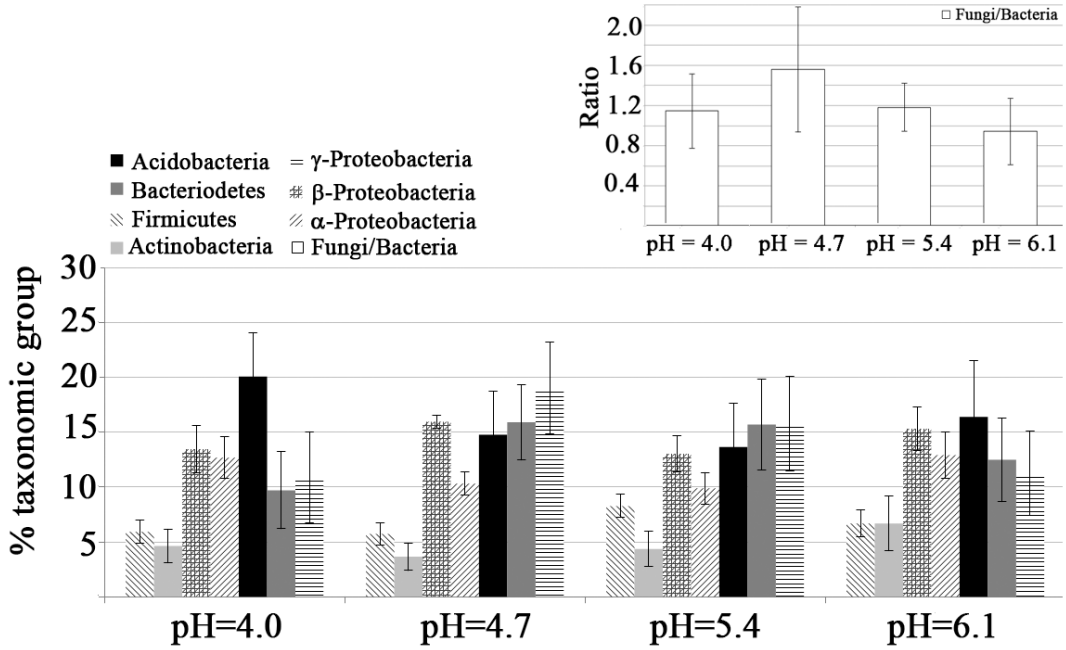
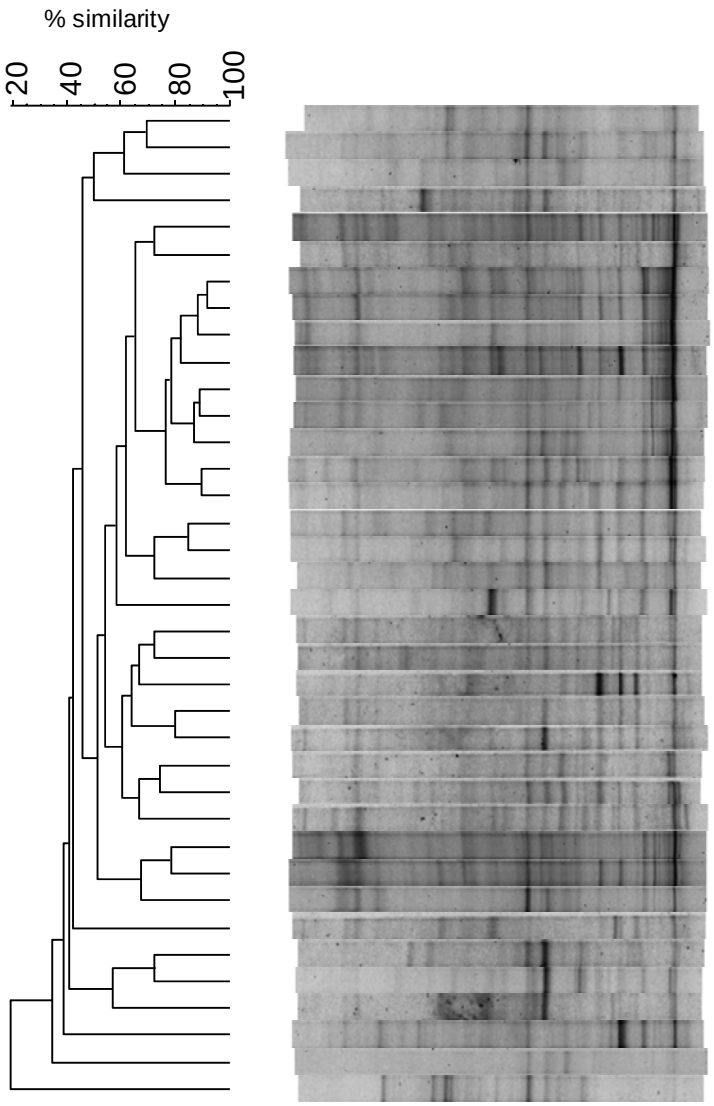


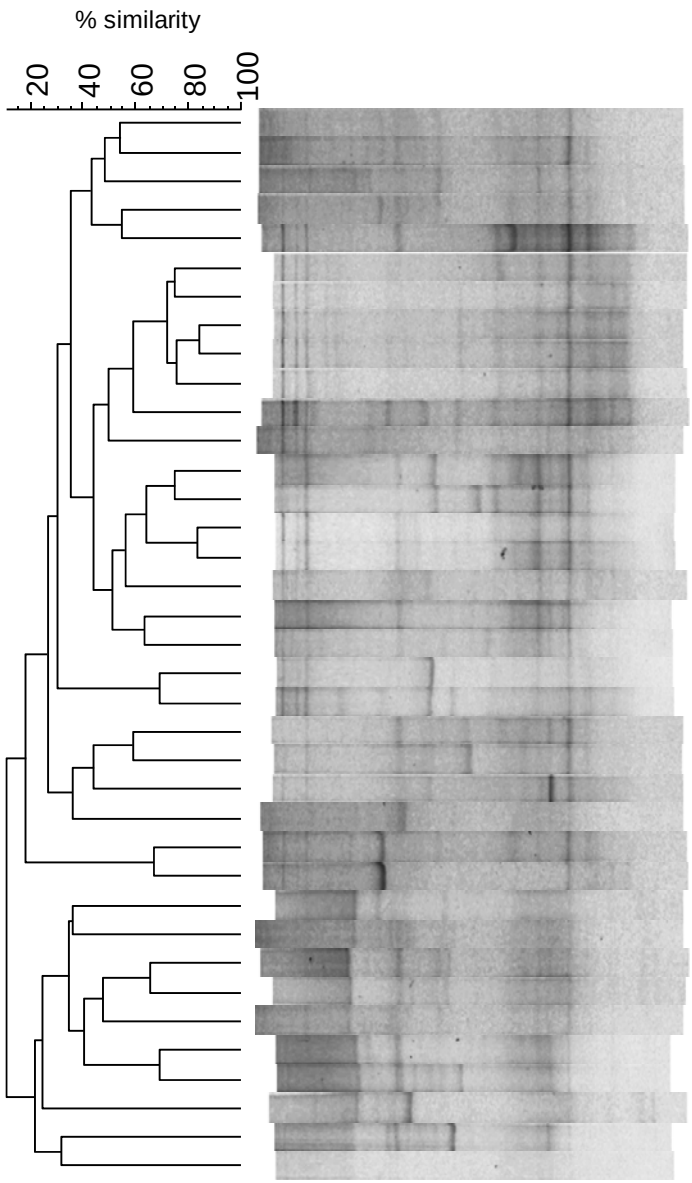
Figure S1. Changes in the percent distribution of different bacterial phyla and fungi in samples from different pH treatments. The error bars represent the standard deviation of triplicate measurements.

A. Fungi



Plot	pH	Cu (kg/ha)
116	6.1	0
23	4	500
62	6.1	250
59	6.1	750
25	4.7	0
91	6.1	750
14	4	0
22	4	0
58	4	750
35	4	0
61	6.1	0
99	6.1	0
51	5.4	0
4	4	250
114	6.1	250
69	5.4	0
78	4.7	500
125	5.4	250
77	4	500
3	5.4	750
18	5.4	750
29	4.7	750
20	6.1	750
90	4	750
6	4.7	750
54	4	500
21	4	750
63	4.7	0
81	4.7	0
5	5.4	0
32	6.1	0
103	5.4	750
112	4	750
52	4.7	750
27	6.1	250
83	5.4	0
108	6.1	500

B. Acidobacteria



Plot	pH	Cu (kg/ha)
29	4.7	750
52	4.7	750
18	5.4	750
3	5.4	750
103	5.4	750
125	5.4	250
62	6.1	250
116	6.1	0
68	5.4	500
114	6.1	250
59	6.1	750
32	6.1	0
5	5.4	0
51	5.4	0
61	6.1	0
83	5.4	0
27	6.1	250
63	4.7	0
81	4.7	0
58	4	750
99	6.1	0
112	4	750
78	4.7	500
69	5.4	0
6	4.7	750
20	6.1	750
91	6.1	750
4	4	250
21	4	750
54	4	500
77	4	500
90	4	750
25	4.7	0
35	4	0
23	4	500
14	4	0
22	4	0

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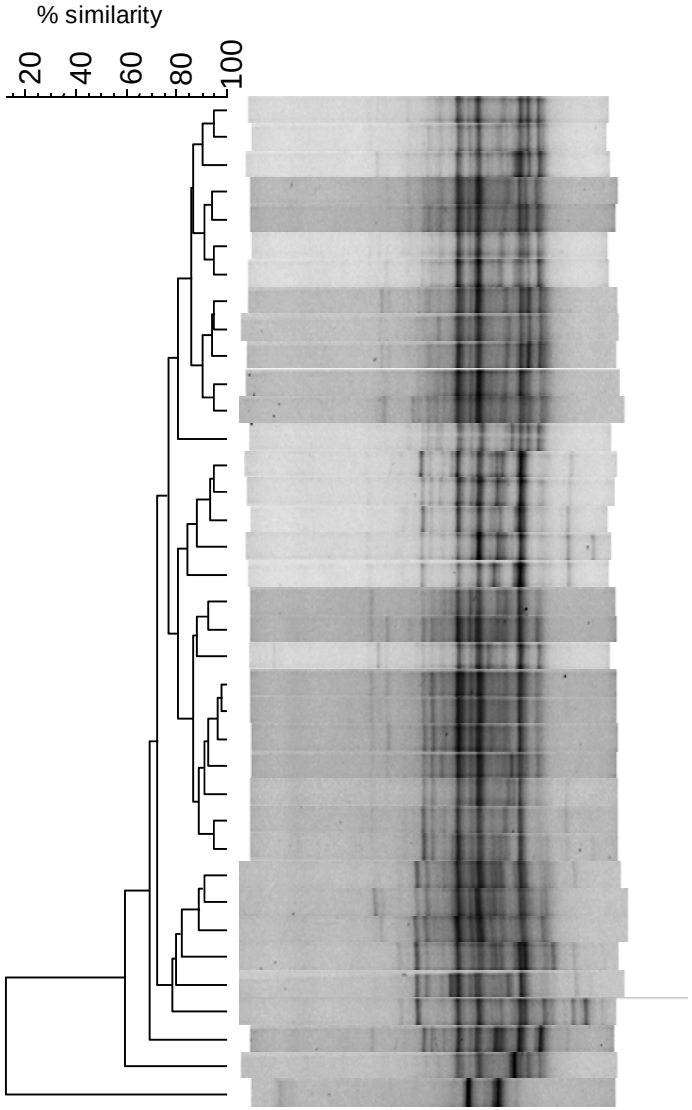
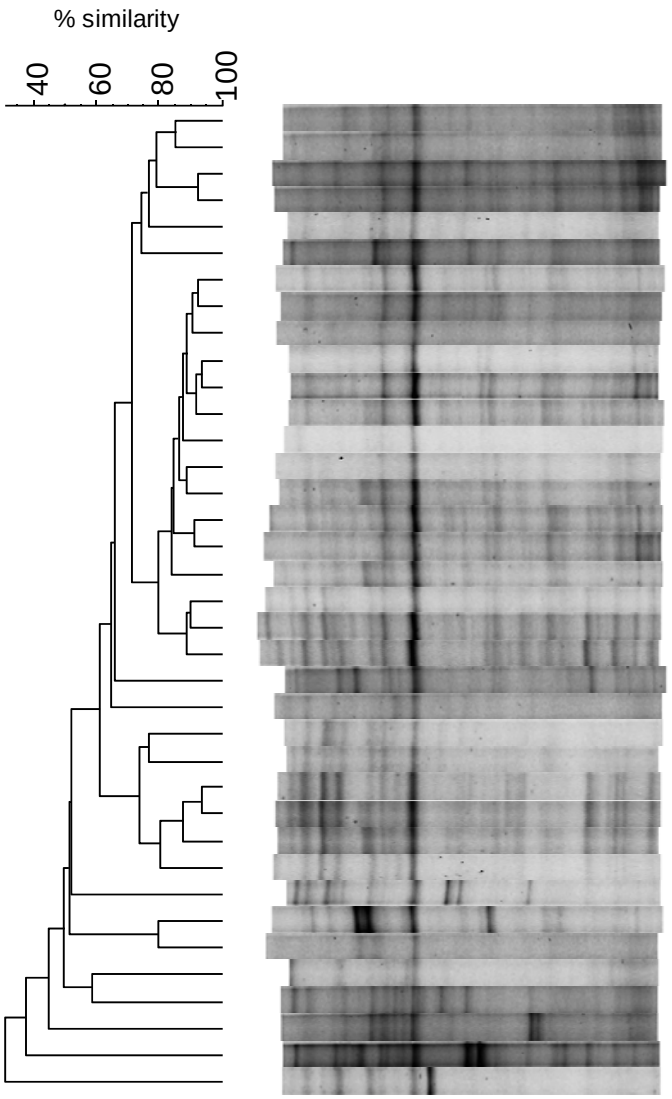
C. Actinobacteria



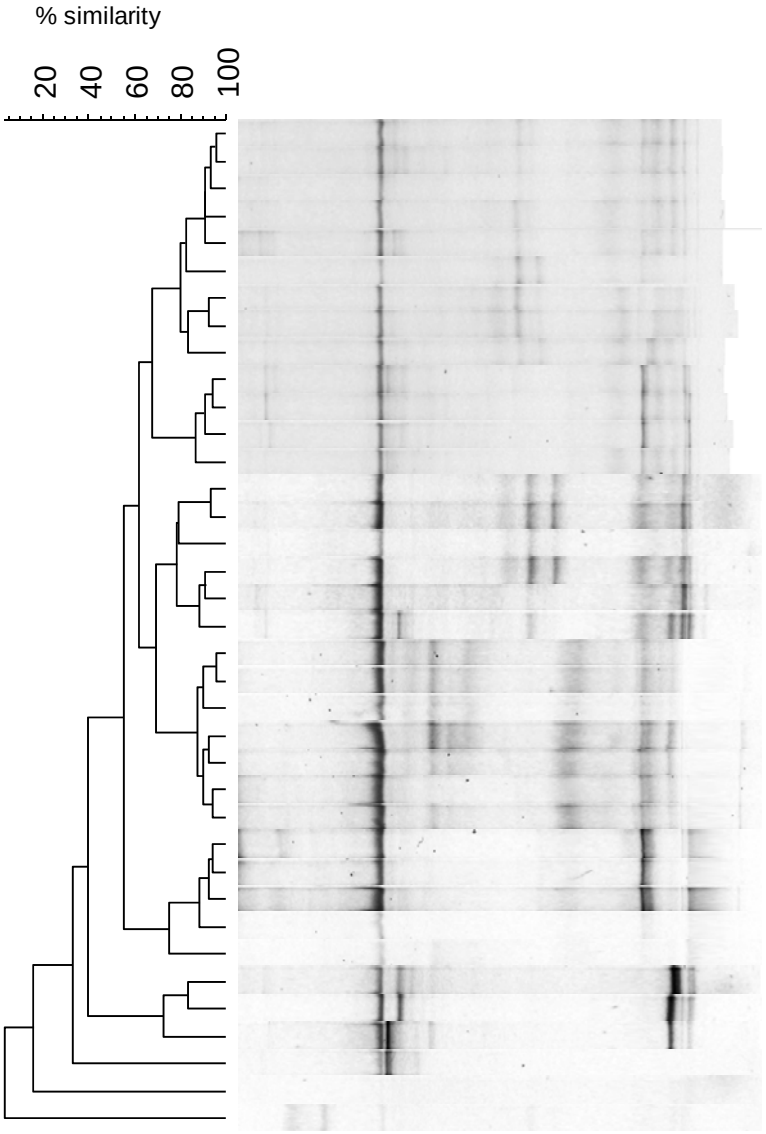
Plot	pH	Cu (kg/ha)
116	6.1	0
23	4	500
62	6.1	250
32	6.1	0
91	6.1	750
125	5.4	250
27	6.1	250
51	5.4	0
61	6.1	0
83	5.4	0
5	5.4	0
81	4.7	0
108	6.1	500
77	4	500
78	4.7	500
4	4	250
112	4	750
114	6.1	250
103	5.4	750
18	5.4	750
69	5.4	0
3	5.4	750
52	4.7	750
29	4.7	750
20	6.1	750
90	4	750
6	4.7	750
21	4	750
35	4	0
63	4.7	0
25	4.7	0
22	4	0
58	4	750
14	4	0
54	4	500
99	6.1	0
59	6.1	750

D. Bacteroidetes



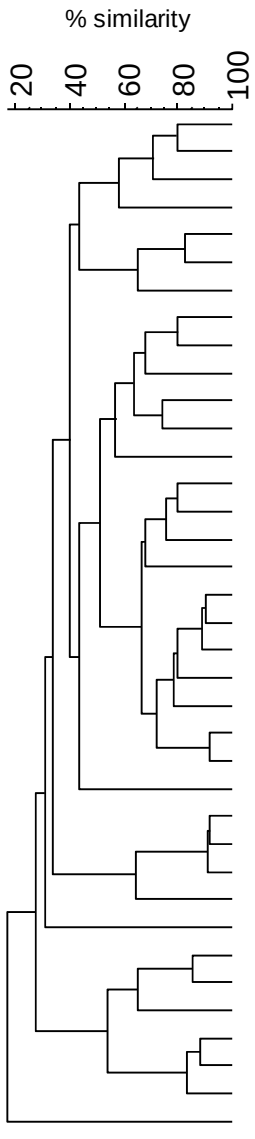
Plot	pH	Cu (kg/ha)
114	6.1	250
116	6.1	0
61	6.1	0
99	6.1	0
125	5.4	250
27	6.1	250
3	5.4	750
81	4.7	0
5	5.4	0
59	6.1	750
91	6.1	750
20	6.1	750
103	5.4	750
25	4.7	0
63	4.7	0
29	4.7	750
32	6.1	0
52	4.7	750
6	4.7	750
90	4	750
21	4	750
23	4	500
83	5.4	0
112	4	750
78	4.7	500
14	4	0
22	4	0
35	4	0
4	4	250
54	4	500
18	5.4	750
51	5.4	0
109	4	500
62	6.1	250
69	5.4	0
58	4	750
77	4	500

E. Betaproteobacteria



Plot	pH	Cu (kg/ha)
3	5.4	750
29	4.7	750
18	5.4	750
20	6.1	750
52	4.7	750
103	5.4	750
59	6.1	750
91	6.1	750
32	6.1	0
6	4.7	750
21	4	750
54	4	500
90	4	750
116	6.1	0
125	5.4	250
62	6.1	250
114	6.1	250
27	6.1	250
78	4.7	500
61	6.1	0
99	6.1	0
51	5.4	0
5	5.4	0
81	4.7	0
25	4.7	0
63	4.7	0
14	4	0
58	4	750
22	4	0
35	4	0
83	5.4	0
4	4	250
77	4	500
23	4	500
112	4	750
69	5.4	0
108	6.1	500

F. Gammaproteobacteria



Plot	pH	Cu (kg/ha)
125	5.4	250
23	4	500
116	6.1	0
62	6.1	250
32	6.1	0
91	6.1	750
59	6.1	750
61	6.1	0
99	6.1	0
63	4.7	0
18	5.4	750
69	5.4	0
27	6.1	250
112	4	750
90	4	750
25	4.7	0
78	4.7	500
29	4.7	750
52	4.7	750
3	5.4	750
103	5.4	750
20	6.1	750
5	5.4	0
81	4.7	0
83	5.4	0
14	4	0
22	4	0
35	4	0
58	4	750
51	5.4	0
21	4	750
54	4	500
6	4.7	750
4	4	250
114	6.1	250
77	4	500
108	6.1	500

Figure S2. Unweighted pair-group clustering analysis using arithmetic averages of group-specific DGGE profiles after Pearson product correlation. For each sample the plot number, the original pH and the original copper concentration are indicated. Groups were: **(A)** Fungi **(B)** *Acidobacteria*, , **(C)** *Actinobacteria*, **(D)** *Bacteriodetes*, **(E)** *Beta*proteobacteria and **(F)** *Gammaproteobacteria* targeted DGGE fingerprints.

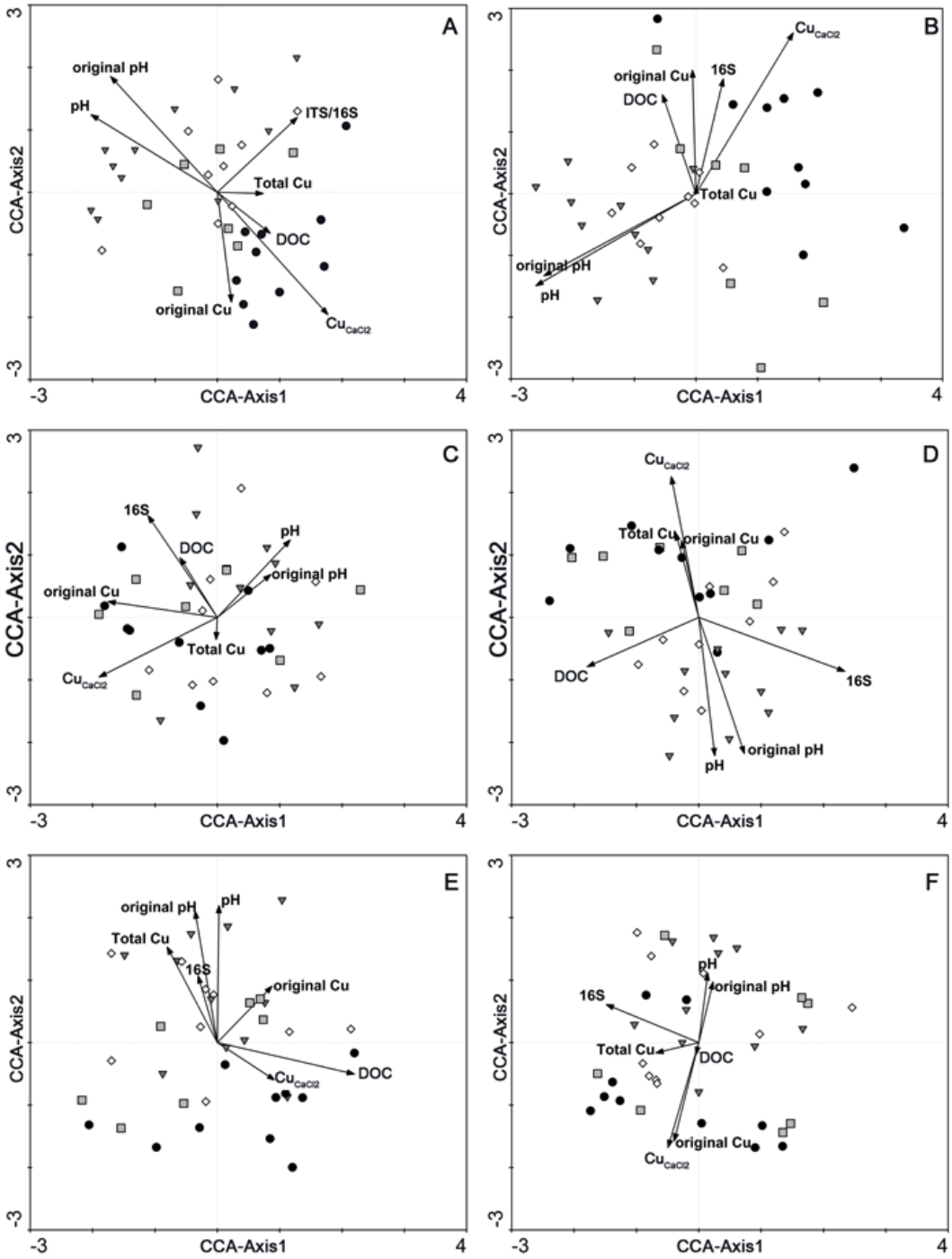


Figure S3. Ordination plots for CCA analysis. Samples are displayed by closed circles (●) for pH 4.0, squares (■) for pH 4.7, diamonds (◇) for pH 5.4 and down triangles (▼) for pH 6.1. Arrows represent environmental parameters. The plot can be interpreted qualitatively by following the direction of arrows for environmental parameters. The arrow length corresponds to variance that can be explained by the environmental variable. Band analysis from **(A)** Fungi **(B)** *Acidobacteria*, , **(C)** *Actinobacteria*, **(D)** *Bacteriodetes*, **(E)** *Beta*proteobacteria and **(F)** *Gammaproteobacteria* targeted DGGE fingerprints.

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