

Supplementary Material

1 Supplementary tables

Supplementary Table S1. Soybean genotypes included in the field experiments conducted in Plains, GA in 2018 and 2019. All genotypes had a sample size of nine, included bulked soil, rhizosphere, and root samples from three replications.

Genotype	Maturity Group (MG)	Type of Resistance
PI 548402	IV	Peking-type resistance
PI 90763	IV	Peking-type resistance
PI 89772	IV	Peking-type resistance
PI 437654	III	Peking-type resistance
G93-9009	VI	Peking-type resistance
PI 88788	III	PI 88788-type resistance
PI 209332	IV	PI 88788-type resistance
PI 548316	III	PI 88788-type resistance
G93-9106	VII	PI 88788-type resistance
Bossier	VIII	Susceptible
CNS	VII	Susceptible
Hutcheson	V	Susceptible
Lee	VI	Susceptible
Lee 74	VI	Susceptible

Supplementary Table S2. HG Type 0 female index of soybean genotypes included in a greenhouse experiment in 2020 at 28 days after inoculation. All genotypes had 27 samples included bulked soil, rhizosphere, and root samples from three replicates at each of three time points: 10, 28, and 42 days after inoculation.

Soybean Genotypes	Maturity Group (MG)	Type of SCN Resistance	Number of cysts ^a	FI (%)^a	Phenotype
Woodruff	VIII	Peking-type resistant	0.7	0.5	Resistant
G00-3880	VII	PI 88788-type resistant	1.9	1.3	Resistant
Lee 74	VI	Susceptible	142.8	100	Susceptible

^a Number of cysts: average number of cysts on the roots of each soybean genotype.

^b Female index (FI) = (average number of cysts) x 100/ (average number of cysts on 'Lee 74').

Supplementary Table S3. Barcodes for 16S sequencing

No.	Forward barcode ^a	Reverse barcode
1	AACCAACC	GTGAGACA
2	CCAACCAA	TACGTTGG
3	GGTTGGTT	ATGCTACG
4	TTGGTTGG	TTCGTTCG
5	AGTCGACT	ACACAGTC
6	CCATCCTA	GAGTCAGA
7	GTCAAGAG	CGATGGTT
8	TAGGTTGC	ATCGTTGG
9	AAGCAAGC	TAGCAACC
10	CGTTCGTT	ACGTCATG
11	GCAAGCAA	GATGTCGT
12	TTCGTTCG	GTGACTCA
13	AGGTGAAC	TCACAGAC
14	CTACAGCA	AGTGTCTG
15	GACACTGT	CACTGAGT
16	TCTGTCTC	GTGTGTGT
17	TCTGTGTC	AACGAACG

^a Barcode sequences were based on Caporaso et al. (2011)

Supplementary Table S4. List of enriched genera in field condition with a linear discriminant analysis (LDA) score of 4.0 among three sample types at two years.

Phylum	Class	Family	Genus	Adjusted P-value	LDA Score	Direction
2018						
<i>Firmicutes</i>	<i>Bacilli</i>	<i>Bacillaceae</i>	<i>Bacillus</i>	< 0.001	4.85	Bulked soil
<i>Actinobacteria</i>	<i>Thermoleophilia</i>	<i>Solirubrobacteraceae</i>	<i>Solirubrobacter</i>	< 0.001	5.01	Bulked soil
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Caulobacteraceae</i>	<i>Phenylobacterium</i>	< 0.001	5.31	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Sphingomonadaceae</i>	<i>Sphingomonas</i>	< 0.001	5.57	Rhizosphere
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Streptomycetaceae</i>	<i>Streptomyces</i>	< 0.001	5.88	Root
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Sphingomonadaceae</i>	<i>Novosphingobium</i>	< 0.001	5.80	Root
2019						
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Micrococcaceae</i>	<i>Pseudarthrobacter</i>	< 0.001	4.94	Bulked soil
<i>Actinobacteria</i>	<i>Thermoleophilia</i>	<i>Solirubrobacteraceae</i>	<i>Solirubrobacter</i>	< 0.001	5.02	Bulked soil
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Caulobacteraceae</i>	<i>Phenylobacterium</i>	< 0.001	5.26	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Sphingomonadaceae</i>	<i>Sphingomonas</i>	< 0.001	5.66	Rhizosphere
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Streptomycetaceae</i>	<i>Streptomyces</i>	< 0.001	5.96	Root
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Sphingomonadaceae</i>	<i>Novosphingobium</i>	< 0.001	5.97	Root

Supplementary Table S5. List of enriched genera in two treatments with a linear discriminant analysis (LDA) score of 4.0 among three sample types at 10, 28 and 42 days after inoculation (DAI).

Phylum	Class	Family	Genus	Adjusted P-value	LDA Score	Direction
Water treatment						
At 10 DAI						
<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Chthoniobacteraceae</i>	<i>Candidatus</i>	0.001	5.50	Bulked soil
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Pseudoduganella</i>	0.002	5.69	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Bradyrhizobium</i>	0.001	6.02	Root
At 28 DAI						
<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Chthoniobacteraceae</i>	<i>Candidatus</i>	< 0.001	5.48	Bulked soil
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Ralstonia</i>	< 0.001	5.78	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Bradyrhizobium</i>	< 0.001	6.44	Root
At 42 DAI						
<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Chthoniobacteraceae</i>	<i>Candidatus</i>	< 0.001	5.50	Bulked soil
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Burkholderia</i>	0.0012	5.66	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Bradyrhizobium</i>	< 0.001	6.23	Root
HG0 Treatment						
At 10 DAI						

<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Chthoniobacteraceae</i>	<i>Candidatus</i>	< 0.001	5.48	Bulked soil
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Pseudoduganella</i>	0.0011	5.9	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Bradyrhizobium</i>	< 0.001	6	Root

At 28 DAI

<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Chthoniobacteraceae</i>	<i>Candidatus</i>	< 0.001	5.5	Bulked soil
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Burkholderia</i>	< 0.001	5.78	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Bradyrhizobium</i>	< 0.001	6.41	Root

At 42 DAI

<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Chthoniobacteraceae</i>	<i>Candidatus</i>	0.0003	5.47	Bulked soil
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Rhizobiaceae</i>	<i>Allorhizobium</i>	0.0027	5.65	Rhizosphere
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Bradyrhizobium</i>	0.0005	6.11	Root

Supplementary Table S6. List of enriched genera in three sample types with a linear discriminant analysis (LDA) score of 3.5 between two treatments at 10, 28 and 42 days after inoculation (DAI).

Phylum	Class	Family	Genus	P-value	LDA Score	Direction
Bulked soil at 10 DAI						
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Pseudoduganella</i>	0.01	4.89	HG0
<i>Acidobacteria</i>	<i>Blastocatellia</i>	<i>Pyrinomonadales</i>	<i>RB41</i>	0.00	4.56	HG0
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Nitrosomonadaceae</i>	<i>MND1</i>	0.01	4.32	HG0
<i>Planctomycetes</i>	<i>Phycisphaerae</i>	<i>WD2101</i>	<i>WD2101</i>	0.02	4.24	HG0
<i>Chloroflexi</i>	<i>Anaerolineae</i>	<i>Anaerolineaceae</i>	<i>UTCFX1</i>	0.03	3.97	HG0
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Dongiaceae</i>	<i>Dongia</i>	0.00	3.97	HG0
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Hyphomicrobiaceae</i>	<i>Pedomicrobium</i>	0.04	3.92	HG0
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Micromonosporaceae</i>	<i>Planosporangium</i>	0.01	3.8	HG0
<i>Planctomycetes</i>	<i>Phycisphaerae</i>	<i>Phycisphaeraceae</i>	<i>AKYG587</i>	0.00	3.6	HG0
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Rhodoplanes</i>	0.05	3.54	HG0
<i>Acidobacteria</i>	<i>Holophagae</i>	<i>Subgroup_7_fa</i>	<i>Subgroup_7_ge</i>	0.01	4.53	Water
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Micrococcaceae</i>	<i>Pseudarthrobacter</i>	0.00	4.21	Water
<i>Proteobacteria</i>	<i>Deltaproteobacteria</i>	<i>0319-6G20</i>	<i>0319-6G20</i>	0.05	3.99	Water
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Chitinophagaceae</i>	<i>Segetibacter</i>	0.00	3.88	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Diplorickettsiaceae</i>	<i>Aquicella</i>	0.02	3.82	Water
<i>GAL15</i>	<i>GAL15_cl</i>	<i>GAL15_fa</i>	<i>GAL15_ge</i>	0.04	3.79	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Legionellaceae</i>	<i>Legionella</i>	0.01	3.64	Water
<i>Chloroflexi</i>	<i>Dehalococcoidia</i>	<i>S085_fa</i>	<i>S085_ge</i>	0.04	3.59	Water
Rhizosphere at 10 DAI						
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Pseudoduganella</i>	0.00	5.58	HG0

<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Chromobacteriaceae</i>	<i>Crenobacter</i>	0.00	4	HG0
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Chromobacteriaceae</i>	<i>Pseudogulbenkiania</i>	0.00	3.94	HG0
<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Pedospaeraceae</i>	<i>ADurb.Bin063-1</i>	0.01	3.7	HG0
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Microscillaceae</i>	<i>Ohtaekwangia</i>	0.02	3.6	HG0
<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Opitutaceae</i>	<i>Opitutus</i>	0.02	4.61	Water
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Rhizobiaceae</i>	<i>Mesorhizobium</i>	0.00	4.6	Water
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Chitinophagaceae</i>	<i>Niastella</i>	0.05	4.59	Water
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Caulobacteraceae</i>	<i>Asticcacaulis</i>	0.02	4.42	Water
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Bradyrhizobium</i>	0.00	4.42	Water
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Chitinophagaceae</i>	<i>Flavisolibacter</i>	0.01	3.93	Water
<i>Proteobacteria</i>	<i>Deltaproteobacteria</i>	<i>Polyangiaceae</i>	<i>Aetherobacter</i>	0.01	3.85	Water
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Chitinophagaceae</i>	<i>Vibrionimonas</i>	0.00	3.73	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Methylophilaceae</i>	<i>Methylobacillus</i>	0.00	3.71	Water
<i>Armatimonadetes</i>	<i>Armatimonadia</i>	<i>Armatimonadales_fa</i>	<i>Armatimonadales_ge</i>	0.02	3.7	Water
<i>Acidobacteria</i>	<i>Acidobacteriia</i>	<i>Acidobacteriaceae</i>	<i>Terriglobus</i>	0.02	3.69	Water
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Micromonosporaceae</i>	<i>Actinoplanes</i>	0.02	3.6	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Solimonadaceae</i>	<i>Alkanibacter</i>	0.00	3.59	Water
<i>Verrucomicrobia</i>	<i>Verrucomicrobiae</i>	<i>Chthoniobacteraceae</i>	<i>Chthoniobacter</i>	0.01	3.56	Water
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Labraceae</i>	<i>Labrys</i>	0.02	3.51	Water
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Chitinophagaceae</i>	<i>Segetibacter</i>	0.00	3.5	Water
Root at 10 DAI						
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Microscillaceae</i>	<i>Ohtaekwangia</i>	0.05	4	HG0
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Dongiaceae</i>	<i>Dongia</i>	0.03	3.95	HG0
<i>Proteobacteria</i>	<i>Deltaproteobacteria</i>	<i>Polyangiaceae</i>	<i>Aetherobacter</i>	0.04	3.87	Water

<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Devosiaceae</i>	<i>Devosia</i>	0.04	3.6	Water
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Rhizobiales</i>	<i>Alsobacter</i>	0.03	3.59	Water
Bulked soil at 28 DAI						
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Caulobacteraceae</i>	<i>Phenyllobacterium</i>	0.03	4.61	HG0
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Chromobacteriaceae</i>	<i>Crenobacter</i>	0.02	3.57	HG0
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Xanthomonadaceae</i>	<i>Thermomonas</i>	0.02	3.56	HG0
<i>Nitrospirae</i>	<i>Nitrospira</i>	<i>Nitrospiraceae</i>	<i>Nitrospira</i>	0.01	4.79	Water
<i>Chloroflexi</i>	<i>TK10_cl</i>	<i>TK10_fa</i>	<i>TK10_ge</i>	0.04	4.28	Water
<i>Gemmatimonadetes</i>	<i>Gemmatimonadetes</i>	<i>Gemmatimonadaceae</i>	<i>Gemmatimonas</i>	0.05	4.18	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Nitrosomonadaceae</i>	<i>Nitrosospira</i>	0.02	4.17	Water
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Chitinophagaceae</i>	<i>Flavisolibacter</i>	0.05	4	Water
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Reyranellaceae</i>	<i>Reyranella</i>	0.04	3.85	Water
<i>Acidobacteria</i>	<i>Holophagae</i>	<i>Holophagaceae</i>	<i>Holophaga</i>	0.02	3.82	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>TRA3-20_fa</i>	<i>TRA3-20_ge</i>	0.01	3.74	Water
Rhizosphere at 28 DAI						
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Rhizobiaceae</i>	<i>Rhizobium</i>	0.01	5.22	HG0
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Hymenobacteraceae</i>	<i>Hymenobacter</i>	0.02	4.17	HG0
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Xanthomonadaceae</i>	<i>Lysobacter</i>	0.04	4.07	HG0
<i>Deinococcota</i>	<i>Deinococci</i>	<i>Deinococcaceae</i>	<i>Deinococcus</i>	0.04	3.53	HG0
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Diplorickettsiaceae</i>	<i>Aquicella</i>	0.04	3.64	Water
<i>Acidobacteria</i>	<i>Holophagae</i>	<i>Holophagaceae</i>	<i>Holophaga</i>	0.00	3.55	Water
Root at 28 DAI						
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Micrococcaceae</i>	<i>Pseudarthrobacter</i>	0.01	4.01	HG0
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Nocardiodaceae</i>	<i>Nocardiodides</i>	0.00	3.85	HG0

<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Sphingomonadaceae</i>	<i>Qipengyuania</i>	0.04	3.8	HG0
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Microbacteriaceae</i>	<i>Curtobacterium</i>	0.01	3.69	HG0
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Microbacteriaceae</i>	<i>Leifsonia</i>	0.03	3.54	HG0
Bulked soil at 42 DAI						
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Rhizobiaceae</i>	<i>Rhizobium</i>	0.02	4.84	HG0
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Pseudonocardiaceae</i>	<i>Actinophytocola</i>	0.01	3.96	HG0
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Beijerinckiaceae</i>	<i>Bosea</i>	0.02	3.93	HG0
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Dongiaceae</i>	<i>Dongia</i>	0.01	3.77	HG0
<i>Nitrospirae</i>	<i>Nitrospira</i>	<i>Nitrospiraceae</i>	<i>Nitrospira</i>	0.01	4.5	Water
<i>Gemmatimonadetes</i>	<i>Gemmatimonadetes</i>	<i>Gemmatimonadaceae</i>	<i>Gemmatimonas</i>	0.02	4.31	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Nitrosomonadaceae</i>	<i>Ellin6067</i>	0.02	3.96	Water
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Diplorickettsiaceae</i>	<i>Aquicella</i>	0.02	3.96	Water
<i>Proteobacteria</i>	<i>Deltaproteobacteria</i>	<i>BIrii41_fa</i>	<i>BIrii41_ge</i>	0.02	3.84	Water
<i>Patescibacteria</i>	<i>Saccharimonadia</i>	<i>Saccharimonadales_fa</i>	<i>Saccharimonadales_ge</i>	0.00	3.76	Water
<i>Armatimonadetes</i>	<i>Fimbriimonadia</i>	<i>Fimbriimonadaceae</i>	<i>Fimbriimonadaceae</i>	0.02	3.67	Water
Rhizosphere at 42 DAI						
<i>Armatimonadetes</i>	<i>Armatimonadia</i>	<i>Armatimonadales_fa</i>	<i>Armatimonadales_ge</i>	0.01	3.73	HG0
<i>Actinobacteria</i>	<i>Actinobacteria</i>	<i>Pseudonocardiaceae</i>	<i>Pseudonocardia</i>	0.00	3.65	HG0
<i>Proteobacteria</i>	<i>Alphaproteobacteria</i>	<i>Xanthobacteraceae</i>	<i>Rhodoplanes</i>	0.02	3.55	HG0
<i>Acidobacteria</i>	<i>Acidobacteriia</i>	<i>Acidobacteriaceae</i>	<i>Acidobacteriaceae</i>	0.01	4.03	Water
Root at 42 DAI						
<i>Proteobacteria</i>	<i>Gammaproteobacteria</i>	<i>Burkholderiaceae</i>	<i>Massilia</i>	0.02	5.25	HG0
<i>Bacteroidetes</i>	<i>Bacteroidia</i>	<i>Chitinophagaceae</i>	<i>Heliimonas</i>	0.04	3.5	HG0

2 Supplementary Figures

Figure S1 (A)

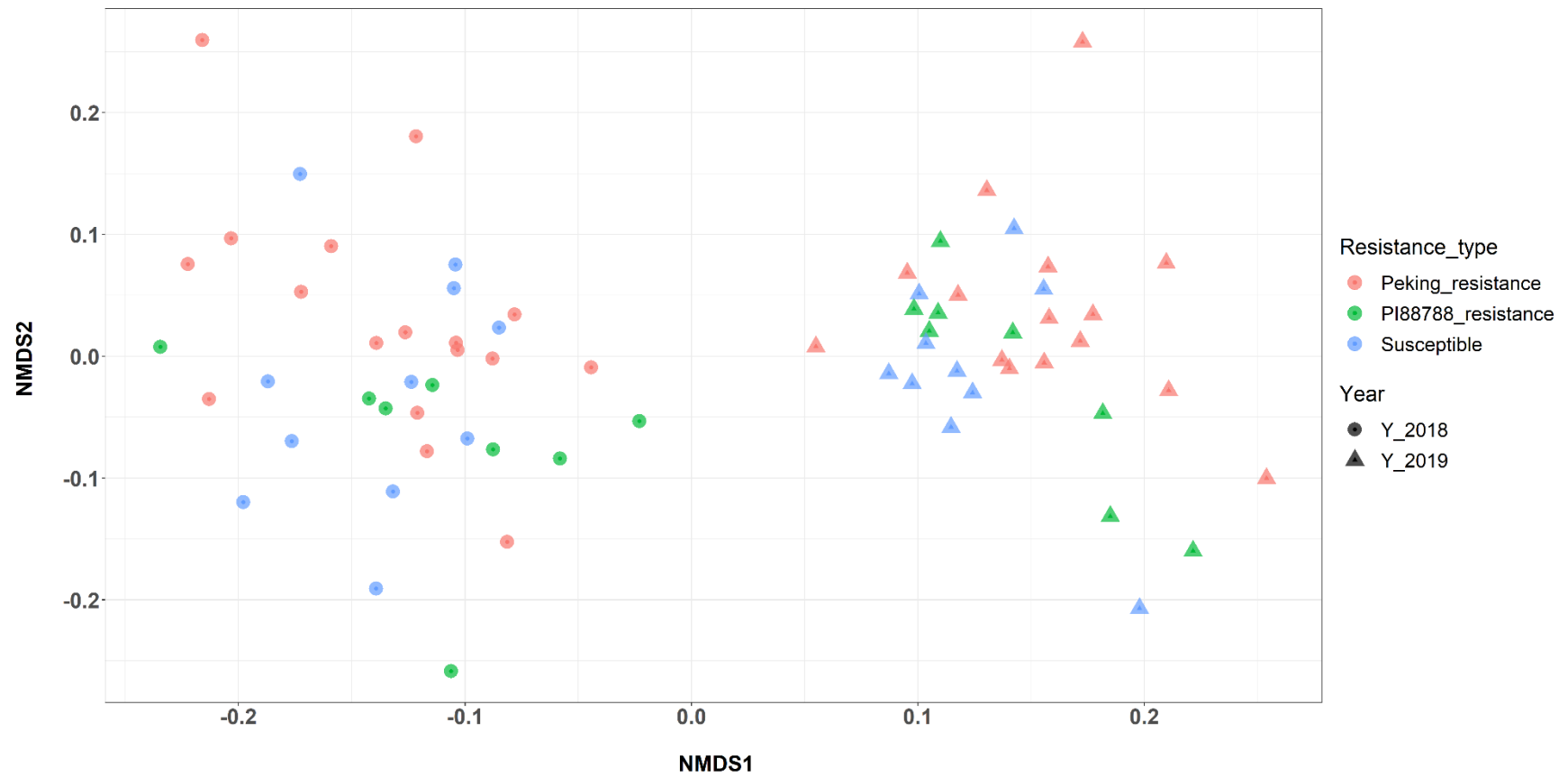


Figure S1 (B)

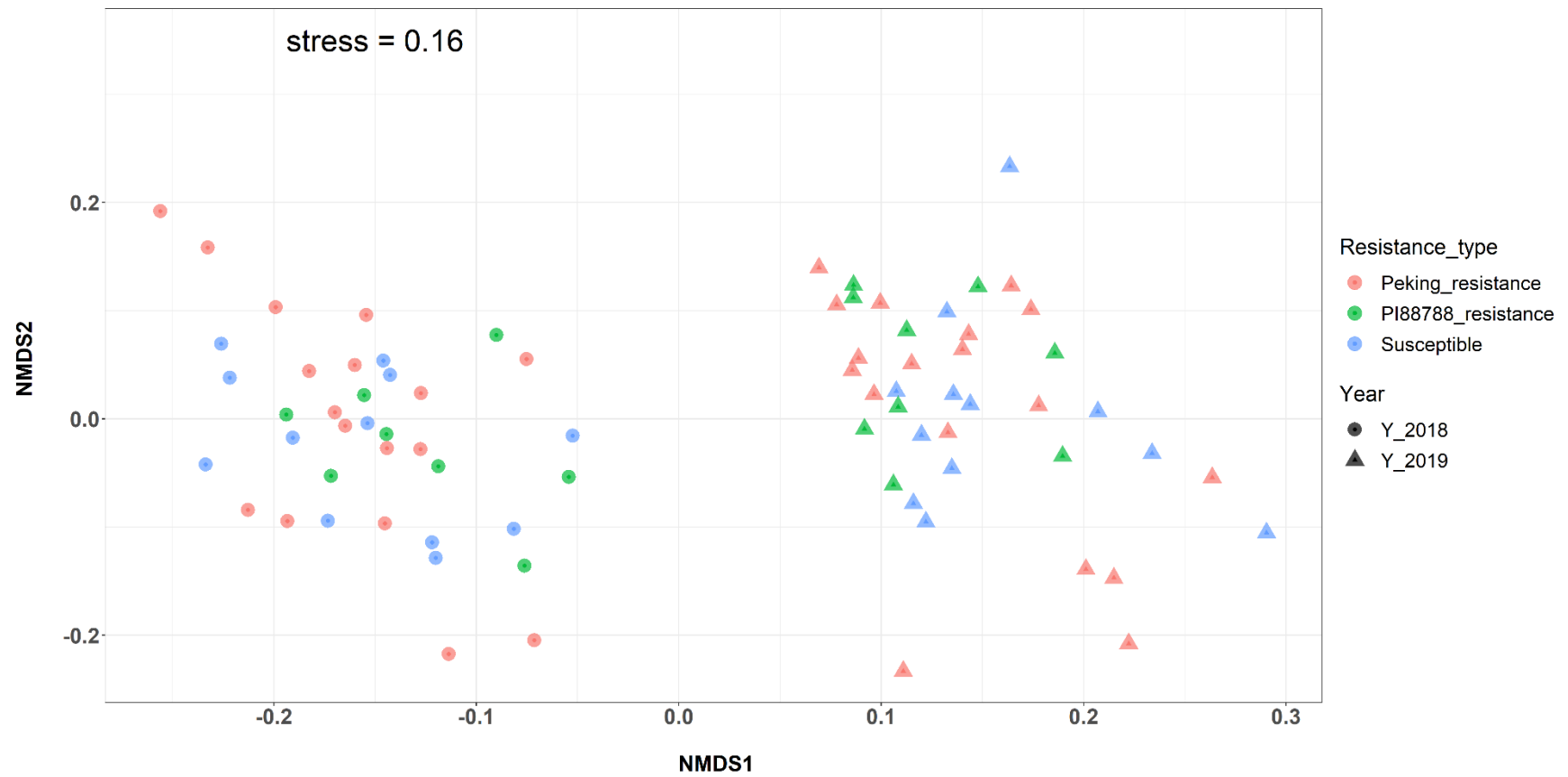
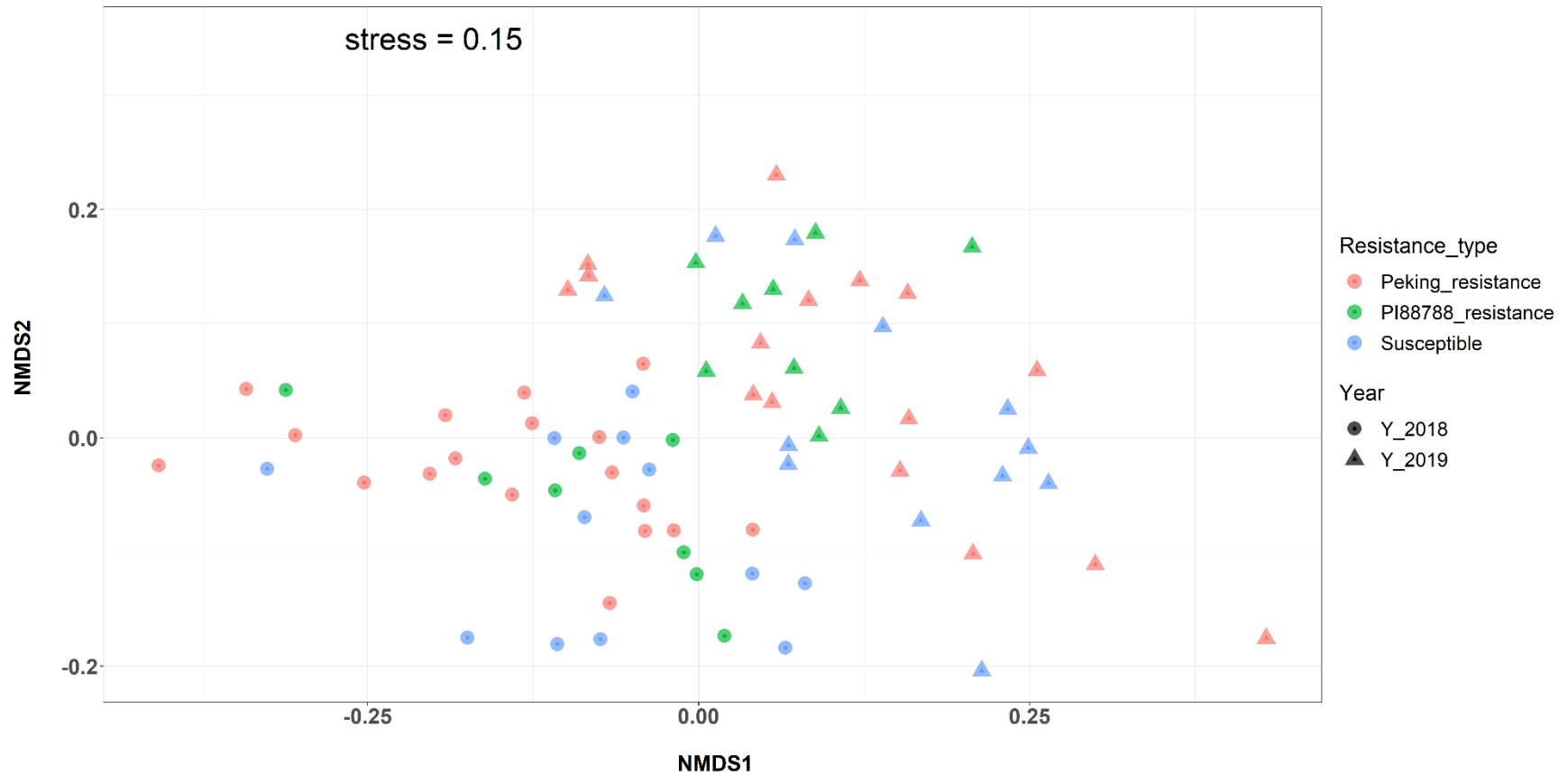


Figure S1 (C)



Supplementary Figure S1. Non-metric Multidimensional Scaling (NMDS) plot based on the weighted Unifrac distance comparing the β diversity among two phenotypes: SCN resistance and susceptible in three sample types. A) Bulk soil; B) Rhizosphere; C) Root.

Figure S2 (A)

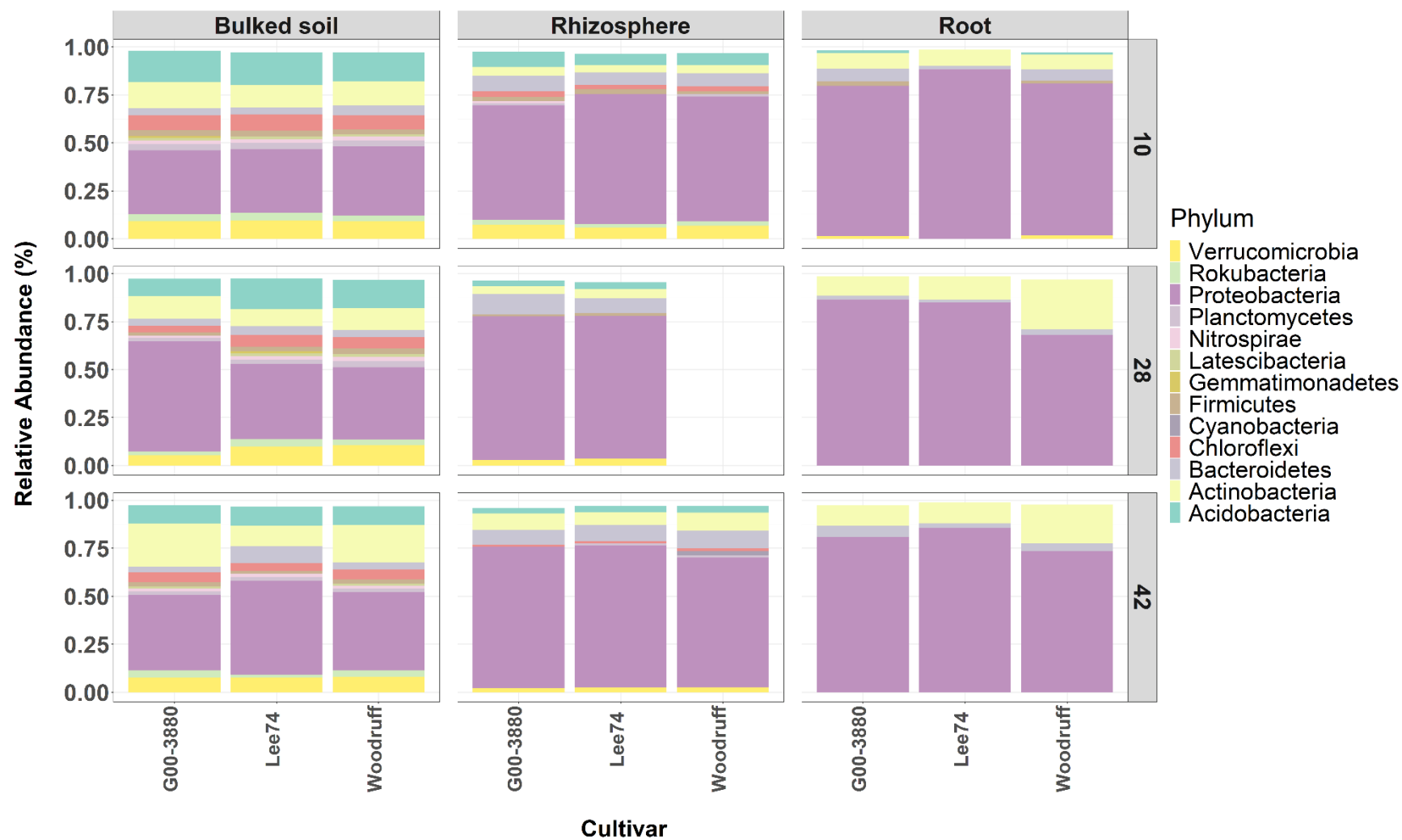
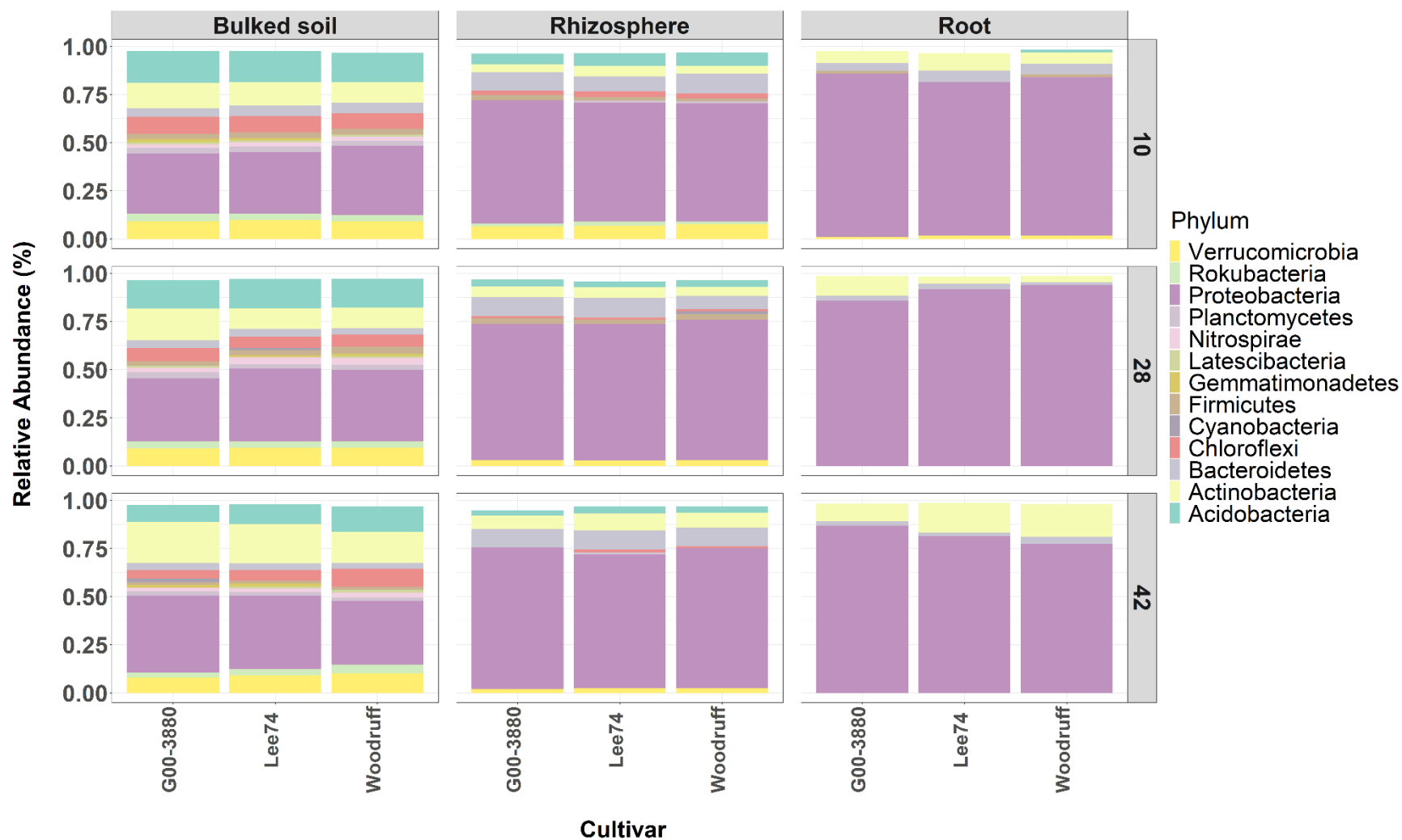


Figure S2 (B)



Supplementary Figure S2. The distribution of core phyla of three types of samples: bulked soil, rhizosphere, and root from two treatments: A) HG Type 0 (SCN race 3) and B) water in a greenhouse test in 2020.