**Table S1:** Reported error rates in original articles (=primary data) and reviews for next generation sequencing platforms

| Company / Platform | Error rate [%] | Primary data | Reference |
| --- | --- | --- | --- |
| Roche 454 | 1 | No | Glenn, 2011 |
|  | 1.1 | Yes | Gilles et al., 2011 |
|  | 4  | Yes | Margulies et al., 2005 |
|  | < 1 | No | Thompson and Milos, 2011 |
|  | 0.25 | Yes | Huse et al., 2007 |
|  | 0.4 | Yes | Quinlan et al., 2008 |
|  | 1.1 | Yes | Lind et al., 2010 |
|  | 0.4 – 0.5 | Yes | Niu et al., 2010 |
|  | 0.4 | Yes | Quince et al., 2011 |
|  | 0.11 – 0.34 | Yes | Vandenbroucke et al., 2011 |
|  | approx. 0.4 | Yes | Loman et al., 2012 |
|  | 0.46 | Yes | Jünemann et al., 2013 |
| Illumina | 0.5 | No | Mardis, 2013 |
|  | <0.1 for >85% of reads | No | Glenn, 2011 |
|  | <2 | No | Liu et al., 2012 |
|  | 1 – 1.5 | No | Shendure and Ji, 2008 |
|  | < 1 | No | Thompson and Milos, 2011 |
|  | 0.6 - 1 | Yes | Dohm et al., 2008 |
|  | 1.3 | Yes | Hillier et al., 2008 |
|  | 0.26 - 0.80 | Yes | Quail et al., 2012 |
|  | < 0.8 | Yes | Quail et al., 2008 |
|  | 2.5 – 7.3 | Yes | Minoche et al., 2011 |
|  | 5.2 – 6.0 | Yes | Nguyen et al., 2011 |
|  | approx. 0.1 | Yes | Loman et al., 2012 |
|  | 0.09 | Yes | Jünemann et al., 2013 |
| Ion PGM | 1 | No | Mardis, 2013 |
|  | 0.46 – 2.4 | No | Glenn, 2011 |
|  | 0.5 | Yes | Merriman et al., 2012 |
|  | approx. 1.5 | Yes | Loman et al., 2012 |
|  | 1.71 | Yes | Quail et al., 2012 |
|  | 1.68 – 4.84 | Yes | Bragg et al., 2013 |
|  | 0.42 – 0.79 | Yes | Jünemann et al., 2013 |

**Table S2:** Amplicon sequencing studies targeting the phyllosphere microbiota based on NGS technology

| Sequencing technology | Target group | Target gene | Sequencing adapter addition | Sequencing statistics | Plant species and type of sample  | Major findings | Reference |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Illumina, MiSeq, 250 bp paired end | Bacteria and fungi | 16S rRNA, V4; ITS1 | Barcoded primers, adapter addition by sequence provider | * 5,120,803 filtered bacterial reads and 3,241,736 filtered fungal reads
 | 273 grape must samples from eight wineries representing four major grape growing regions of California | * regional, site-specific, climatic and grape variety factors shaped the fungal and bacterial microbiota inhabiting wine-grape surfaces
 | Bokulich et al., 2014 |
| Roche 454 | Bacteria | 16S rRNA, V4 to V6 | Fusion primers | * 505,082 raw reads
* 346,866 filtered reads
 | Rosettes of greenhouse grown *Arabidopsis thaliana* plants | * successional dynamics over a period 73 days: phyllosphere communities initially mirrored airborne communities, they subsequently converged to a distinct community composition
 | Maignien et al., 2014 |
| Roche 454 | Bacteria and fungi | 16S rRNA, V5-V9;ITS | Fusion primers | * 30,515 filtered bacterial reads and 134,172 fungal reads
* 40 – 3,294 bacterial and 1,353 – 9,303 fungal reads per sample
* mean length of 750 bp for bacterial and 650 bp for fungal reads
 | Leaf samples of grapevine (*Vitis vinifera*) treated with chemical fungicide or biological control agent , taken from three vineyards in Italy | * bacterial and fungal communities were only minimally affected by chemical and biological treatments,
* they mainly differed according to sampling site location
 | Perazzolli et al., 2014 |
| Roche 454 | Bacteria and fungi | 16S rRNA, V6;ITS2 and D2 | Fusion primers | * 142,096 raw reads
* 139,034 filtered reads; 79,398 for fungi (ITS2 plus D2) and 59,636 for bacteria
* 2,070 – 9,462 reads per sample
 | Leaves of grapevine (*Vitis vinifera*) from a vineyard in Portugal collected over the growing season | * microbial communities were shown to be highly structured and dynamic
* the major abundant microorganisms were the yeast-like fungus *Aureobasidium* and the prokaryotic Enterobacteriaceae
 | Pinto et al., 2014 |
| Roche 454 | Bacteria | 16S rRNA, V4  | Addition of barcodes and adapters in a 2nd PCR | * 35,965 filtered reads
 | Leaves and roots of tomato (*Solanum lycopersicum*) grown in productive greenhouses | * leaf endophytic communities strongly differed from those of the rhizosphere
 | Romero et al., 2014 |
| Roche 454 | Fungi | ITS1 | Addition of barcodes in a 2nd PCR (6 cycles); adapter addition by sequence provider | * 204,052 raw reads
* 51,596 filtered reads
* 1,388– 3,057 reads per sample
 | Garden grown balsam poplar *(Populus balsamifera)* genotypes | * host genotype shaped the foliar fungal microbiome
 | Bálint et al., 2013 |
| Roche 454 | Bacteria | 16S rRNA, V5 to V7 | Fusion primers | * mean read number 8,090 per sample
* minimum of 4,329 sequences per sample
 | Rosettes and roots of naturally grown *Arabidopsis thaliana* plants from four sites | * bacterial communities differ between leaves and roots
* different epi- and endophytic communities
* leaf and root endophytic communities do not differ in richness, diversity and evenness, but in community composition
* taxa with highest relative abundance are related to culturable species
 | Bodenhausen et al., 2013 |
| Ion PGM,316 Chip | Fungi | ITS1 | Fusion primers | * 2,394,051 raw reads
* liberal filtering: 461,596 reads
* stringent filtering: 86,556 reads
* mean read length 137 bp
 | Leaf, petiole, twig,and trunk samples from *Eucalyptus grandis*, South Africa | * highly diverse families from across the Kingdom Fungi co-occurred within a few samples of plant tissue
* communities were dominated by Ascomycota
 | Kemler et al., 2013 |
| Roche 454 | Bacteria | 16S rRNA  | Adapter addition by sequence provider | * 50,299 filtered reads
* 2,515 reads per sample
* read length >200 bp
 | Organic and conventionally grown leafy salad vegetables (baby spinach, romaine lettuce,red leaf lettuce, iceberg lettuce and green leaf lettuce) | * no significant differences in bacterial community composition on leaves of organic versus conventionally grown vegetables
* the dominant taxa from sequence data were also detected by culture-dependent methods
 | Jackson et al., 2013 |
| Roche 454 | Bacteria | 16S rRNA, V4 | Fusion primers | * between 17,341 and 35,503 raw reads per sample
 | Spermosphere and phyllosphere of spinach (*Spinacia oleracea*) seedlings and plants | * bacterial richness higher on seedlings than on seeds and cotyledons
 | Lopez-Velasco et al., 2013 |
| Roche 454 | Bacteria | 16S rRNA, V3 to V5 | Fusion primers | * 193,289 raw reads
* 90,815 filtered reads
* 2,343 - 10,010 reads per sample
 | *Arabidopsis thaliana* leaf wax mutants grown outdoor | * altered bacterial phyllosphere community composition on cuticular wax mutants
 | Reisberg et al., 2013 |
| Roche 454 | Bacteria | 16S rRNA, V5 and V6 | Unknown | * 225,243 raw reads
* 171,996 filtered reads
* 50,865 reads after removing singletons
* 1,838 – 10,776 reads per sample
* Mean read number 5,733 per sample
* Rarified to 1,838 reads per sample
* Mean read length 335 bp
 | Flowers from six apple trees (*Malus domestica*)collected at five time points; streptomycin application to half of the trees when flowers opened | * bacterial communities dominated by members of TM7 and *Deinococcus-Thermus*
* observation of a successional pattern and less abundant transient taxa
* communities on trees sprayed with streptomycin had slightly lower phylogenetic diversity, but no differences in structure or succession
 | Shade et al., 2013 |
| Roche 454 | Bacteria and fungi | ITS;16S rRNA | Fusion primers | * 50,835 ITS raw reads and 19,560 16S rRNA raw reads
* 34,045 filtered ITS reads and 8,425 16S rRNA reads
* 172 – 1,890 reads per sample
 | Strawberry (*Fragaria* x *ananassa*) treated with biological control agents against *Botrytis cinerea* | * altered fungal community composition at class level upon treatment with one of the tested biological control agents
* no effects on bacterial community composition
 | Sylla et al., 2013 |
| Roche 454 | Bacteria | 16S rRNA, V5 to V9 | Fusion primers | * mean read number 5,116 per sample
 | Field grown Romaine lettuce from different plantings, inoculated with attenuated strain of *Escherichia coli* O157:H7 | * bacterial diversity differed in dependence of irrigation method, season, plant age, and in presence or absence of *E. coli*
 | Williams et al., 2013 |
| Roche 454 | Bacteria | 16S rRNA  | Barcoded primers, adapter addition by sequence provider | * from only few sequences to 4,000 per sample
* rarefied to 200 reads per sample
 | Conventional and organic analogs of eleven store-bought fruits and vegetables (sprouts, spinach, lettuce, tomatoes, peppers, strawberries, apples, peaches, grapes, and mushrooms) | * certain products shared more similar communities (high relative abundances of Enterobacteriaceae) compared to others (dominated by Actinobacteria, Bacteroidetes, Firmicutes, and Proteobacteria)
* differences in community composition between conventional and organic analogs within product types; relative abundances of Enterobacteriaceae taxa less abundant in organically-grown products
 | Leff and Fierer, 2013 |
| Roche 454 | Bacteria and fungi | 16S rRNA, V2;18S rRNA | Fusion primers | * 92,695 filtered 16S rRNA reads and 194,260 18S rRNA reads
* mean read number per sample 3862 for 16S rRNA and 8094 for 18S rRNA amplicons
 | Leaves, stems, roots, flowers and fruits of outdoor grown tomato (*Solanum lycopersicum*) | * distinct microbial communities on different tomato plant organs
* differences related to distance of the plant part from the soil
 | Ottesen et al., 2013 |
| Roche 454 | Fungi | ITS1 | Barcoded primers, adapter addition by sequence provider | * 1,285,911 raw reads
* 953,385 filtered sequences
* mean read number 28,040 per sample
 | Leaf and stem samples of pine tree *Pinus sylvestris* and its parasite *Viscum**album* | * interannual changes in endophytic community composition are smaller than seasonal changes
* composition depends on host species and organ type, locality has a minor impact
 | Peršoh, 2013 |
| Roche 454 | Fungi | ITS1 | Fusion primers | * see Cordier et al 2012a
 | Beech (*Fagus sylvatica*) phyllosphere; data from the study of Cordier et al 2012a were used in this study | * underrepresentation of phytopathogenic and -parasitic ascomycetes, lichenized fungi and some basidiomycete taxa in cultivation-dependent approaches
 | Unterseher et al., 2013 |
| Roche 454 | Fungi | ITS1 and ITS2; 16S rRNA, V5 and V8  | Adapter addition by ligation | * 294,102 raw reads
* 209,544 filtered reads
* 1237 - 8046 reads per sample
* mean read length 318 bp
* sequencing of libraries from both ends
 | *Avicennia marina* and *Rhizophora stylosa* trees in a mangrove in New Caledonia | * host-specific highly diverse fungal communities on the two mangrove tree species in aerial and intertidal parts
 | Arfi et al., 2012 |
| Roche 454 | Fungi | ITS1 | Fusion primers | * mean read number of 2,400 and 6,195 for two datasets
* 75 – 4,033 and 946 – 20,493 reads per sample for the two datasets
 | European beech (*Fagus sylvatica*) at four sites over a gradient of 1000 m of elevation in the French Pyrénées Mountains | * fungal community composition varied at sites of different elevation
 | Cordier et al., 2012b |
| Roche 454 | Fungi | ITS1 | Fusion primers | * 96,130 filtered reads
* 123,163 raw reads
* mean of 3,560 reads per sample
* 386 – 6920 reads per sample
 | Samples from European beech *Fagus sylvatica* of four different spatial scales: tree, branch, group of leaves and individual leaf | * major part of the variability was at the smallest spatial scale, i. e. between individual leaves
* at larger scale, genetic variation of trees affected phyllosphere fungal assemblages
 | Cordier et al., 2012a |
| Roche 454 | Fungi | ITS2 | Nested PCR approach with fusion primers in 2nd PCR | * 491,280 raw reads
* 327,273 filtered reads
* mean of 1,031 reads per sample
* 502 – 2,423 reads per sample
 | Seasonal sampling of three bryophyte species in a Norwegian forest: *Hylocomium splendens*, *Pleuroium schreberi* and *Polytrichum commune* | * seasonal dynamics and host-specific patterns were observed besides effects of locality and tissue
 | Davey et al., 2013 |
| Roche 454 | Bacteria | 16 S rRNA, V4-V6 | Fusion primers | * 163,895 filtered reads
* rarified to 8,700 reads per sample
* mean read length 481 bp
 | Leaves of *Tamarix aphylla* trees, collected from a transect (500 km) in the Soronan desert | * diverse bacterial communities with four dominant phyla
* geographical distance was the most important parameter that affected community composition
 | Finkel et al., 2012 |
| Roche 454 | Bacteria | 16S rRNA, V1–V3 | Fusion primers | * 21,930 filtered reads
* mean of 1,327 reads per sample
* 553 – 2,281 reads per sample
* mean read length 431 bp
 | Leaves of six species of tropical treesat a rainforest arboretum in Malaysia (*Arytera littoralis, Schizostachyum brachycladum, Dillenia excela, Dyera costulata, Gnetum sp. shorea maxima)* | * each tree species had a distinctive bacterial phyllosphere community
* more similar communities on more closely related hosts
* Acidobacteria were one of the most abundant phyla across all samples
 | Kim et al., 2012 |
| Roche 454 | Bacteria | 16S rRNA  | Fusion primers | * 165,259 filtered reads
* mean of 9,486 reads per sample
* 5,852 – 12,836 reads per sample
 | Leaves of tomato (*Solanum lycopersicum*) from three different locations in Florida | * enrichments of *Salmonella* changes the taxonomic profile of a sample but does not necessarily increase detectability of *Salmonella*
 | Pettengill et al., 2012 |
| Roche 454 | Bacteria | 16S rRNA, V5 - V7 | Fusion primers | * 818,013 filtered reads
* mean of 9,296 reads per sample
* rarified to 2,836 reads per sample
 | Field-grown Romaine lettuce from California and Arizona | * differences in bacterial communities were related to season and field location, but not to plant cultivar
 | Rastogi et al., 2012 |
| Roche 454 | Fungi | ITS1 | Barcoded primers, adapter addition by sequence provider | * 665,155 filtered reads
* Mean of 5,117 reads per sample
* mean trimmed read length 157 bp
 | Endophytic fungi in leaves of *Metrosideros polymorpha* across environmental gradients but short geographic distance on Hawaii | * high diversity within sites and even higher diversity between sites
* variation between sites correlated with temperature and rainfall
 | Zimmerman and Vitousek, 2012 |
| Roche 454 | Bacteria | 16S rRNA, V1-V2,  | Fusion primers | * minimum of 1,300 reds per sample
 | Leaves of four *Weinmannia* tree species from a montane elevational gradient in eastern Peru | * tree associated phyllosphere bacteria exhibited no significant elevational gradient in diversity (in contrast to the plants)
 | Fierer et al., 2011 |
| Roche 454 | Bacteria, Archaea and fungi | 16S rRNA, V6;18S rRNA, V9  | Fusion primers | * 158,980 filtered bacterial reads, 48,673 fungal reads, and 27,388 archaeal reads
* mean of 11,355 bacterial reads per sample
* read length of approx. 60 bp
 | Leaves from three *Tamarix* tree species grown in Mediterranean andDead Sea regionand two locations in the USA along ecological gradients | * microbial communities on different *Tamarix* species grown in the same location were highly similar, while trees of the same species growing in different climatic regions hosted distinct microbial communities
 | Finkel et al., 2011 |
| Roche 454 | Bacteria | 16S rRNA, V5 and V6 | Fusion primers | * 37,474 filtered reads
* mean read number of 3,737 per sample
* 1,726 – 6,343 reads per sample
* rarified to 1,600 reads per sample
* mean read length 465 bp
 | Grape leaves and berries collected from a vineyard | * bacterial communities differed on the surface of leaves and berries
 | Leveau and Tech, 2011 |
| Roche 454 | Bacteria | 16S rRNA, V4 | Fusion primers | * 14,686 – 24,294 raw reads per sample
* mean read length of 200 bp
 | Freshly harvested and stored spinach (*Spinacia* *oleracea*) | * refrigerated conditions decreased species richness, diversity and evenness.
* growth inhibition of Escherichia spp. Was achieved at 4°C, but not at 10°C
 | Lopez-Velasco et al., 2011 |
| Roche 454 | Bacteria and fungi | 16S rRNA, V1-V2; ITS | Fusion primers | * 24,445 filtered bacterial reads
* mean read length 475 bp for bacterial reads
* no data for fungal reads
 | *Atriplex canescens* and *Atriplex torreyi* callus cultures | * micropropagated cultures were colonized by different bacterial and fungal taxa
 | Lucero et al., 2011 |
| Roche 454 | Bacteria | 16S rRNA, V1-V3  | Fusion primers | * 27,757 filtered reads
* mean of 1,734 reads per sample
 | Fruit surface of field grown tomatoes (*Solanum**lycopersicum*) irrigated with water from different sources | * bacterial communities on tomato fruit surfaces did not differ in dependence of the irrigation water source
 | Telias et al., 2011 |
| Roche 454 | Fungi | ITS2  | Fusion primers | * 105,838 raw reads
* 84,956 filtered reads
* mean of 1,259 reads per sample
* mean read length 268 bp
 | Leaves of oak (*Quercus macrocarpa*) from six sites in urban and nonurban environments collected over a growing season  | * fungal communities were temporally dynamic
* communities were diverse and differed between the urban and nonurban stands, albeit not consistently across the growing season
 | Jumpponen and Jones, 2010 |
| Roche 454 | Bacteria | 16S rRNA, V1 and V2 | Fusion primers | * 31,373 filtered reads
* mean read length 215 bp, ranging from 150 to 316 bp
 | Pitcher plant fluid from *Sarracenia alata* | * high bacterial richness
* increase in diversity and abundance with time
* bacterial community composition changes over time
 | Koopman et al., 2010 |
| Roche 454 | Bacteria | 16S rRNA, V5 and V6 | Fusion primers | * 115,394 filtered reads
* 600 – 1500 reads per sample
* minimum of 600 reads per sample
* mean read length of 240 bp
 | Leaves of 56 tree species in Colorado, and needles of *Pinus ponderosa* from various locations of the world | * higher variability in community composition between different tree species than within
* correspondence between tree phylogeny and bacterial community phylogeny
* minimal geographic differentiation across continents
 | Redford et al., 2010 |
| Roche 454 | Fungi | ITS1 | Fusion primers | * 18,020 filtered reads
* 9,168 and 8,852 reads per sample
* mean read length of 265 bp
 | Leaves of oak (*Quercus macrocarpa*) located within and outside a small urban center | * fungal phyllosphere communities were extremely diverse and strongly dominated by ascomycetes
* fungal communities on plants from the urban center showed lower richness and diversity
 | Jumpponen and Jones, 2009 |

**List of rhizosphere papers that apply amplicon sequencing using NGS technologies**

Anderson, C.R., Condron, L.M., Clough, T.J., Fiers, M., Stewart, A., Hill, R.A., and Sherlock, R.R. (2011). Biochar induced soil microbial community change: implications for biogeochemical cycling of carbon, nitrogen and phosphorus. *Pedobiologia* 54**,** 309-320.

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Becklin, K.M., Hertweck, K.L., and Jumpponen, A. (2012). Host identity impacts rhizosphere fungal communities associated with three alpine plant species. *Microb Ecol* 63**,** 682-693.

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Blaalid, R., Carlsen, T., Kumar, S., Halvorsen, R., Ugland, K.I., Fontana, G., and Kauserud, H. (2012). Changes in the root-associated fungal communities along a primary succession gradient analysed by 454 pyrosequencing. *Mol Ecol* 21**,** 1897-1908.

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