

AmpliCon Diversity Assay List

At RTL Genomics, we are able to use almost any amplicon assay for your project. Illumina projects should use amplicons with a length less than 550 bp. Longer assays may be used if desired, but will make our denoising and analysis process unavailable for your data. We may also be unable to perform taxonomic analysis for some functional genes.

Our recommended assays are listed below, but you can choose to combine forward and reverse primers in a different configuration to target the variable region of interest. Recommended assays have the best historical performance and are commonly used regions. All primers have different amplification bias for certain organisms, and that information is not always known for all primers. Primers marked as “universal” show amplification of multiple kingdoms with the same primer. Other primers preferentially amplify only their listed kingdom organism.

If the primer you are looking for is not listed, we still may have it, but if that is not the case it can easily be ordered. Please inquire and we will be happy to assist you.

Recommended assays:

Assay	Forward Primer	Reverse Primer	Notes
28F	28F - GAGTTGATCNTGGCTAG	519R - GTNTTACNGCGGCKGCTG	Default 16S, V1-V3
357wF	357wF - CCTACGGNGGCWGCAG	806R GGACTACHVGGGTWTCTAAT	V3-V4
515F	515F - GTGCCAGCMGCCGCGTAA	806R GGACTACHVGGGTWTCTAAT	Earth Microbiome, V4
515yF	515yF - GTGYCAGCMGCCGCGTAA	926pfR - CCGYCAATTYMTTRAGTT	Earth Microbiome, V4-5
Arch517F	Arch517F - GCYTAAGSRNCCGTAGC	Arch909R TTTCAGYCTTGCGRCCGTAC	Default Archaea 16S, V4-5
ITS1	ITS1F CTTGGTCATTTAGAGGAAGTAA	ITS2aR GCTGCCTTCTTCATCGATGC	Default Fungal ITS, ITS1

Bold are from Earth Microbiome Project, Italicized are default primers for RTLGenomics

** Bacterial & Archaeal “universal”

*** Bacterial & Archaeal & Eukaryotic “universal”

Bacterial Primers
16S

27F AGAGTTGATCCTGGCTCAG	338R GCTGCCTCCGTAGGAGT
	533R TTACCGCGGCTGCTGGCAC
27-YMF AGAGTTGATYMTGGCTCAG	534R TYACCGCGGCTGCTGG
27mF AGRGTTYGATYMTGGCTCAG	
28F-YM GAGTTGATYMTGGCTCAG	These primers are mixed at a 4:1:1:1 ratio (28F-YM is at 4 parts)
28F-Borrellia GAGTTGATCCTGGCTTAG	
28F-Chloroflex GAATTGATCTGGTTTAG	
28F-Bifdo GGGTCGATTCTGGCTCAG	
<i>28F GAGTTGATCNTGGCTCAG</i>	519R GTNTTACNGCGGCKGCTG
	388R TGCTGCCTCCGTAGGAGT
63F CAGGCCAACACATGCAAGTC	530aR GTATTACCGCGGCTGCTG
104F GGCGVACGGGTGAGTAA	530R CCGCNGCNGCTGGCAC
338F TCCTACGGGAGGCAGCAGT	797R GGACTACCAGGGTATCTAACCTGTT
338aF ACTCCTACGGGAGGCAGCAG	518R ATTACCGCGGCTGCTGG
338bF ACWCCTACGGGWGGCWGC	
Firm350F GGCAGCAGTRGGGAATCTTC	Firm814R ACACYTAGYACTCATCGTT
Cyan359F GGGGAATYTTCCGCAATGGG	Cyan781R GACTACWGGGTATCTAACCCWTT
AMX368F TTCGCAATGCCGAAAGG	AMX821R AAAACCCCTCTACTTAGTGCCC
357F (341F) CCTACGGGAGGCAGCAG *	907R CCGTCAATTCTAGTTGAGTT
	926wR CCGTCAATTYMTTTRAGTT

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357wF <i>CCTACGGGNGGCWGCAG</i>	785R <i>GACTACHVGGGTATCTAATCC</i>
515F <i>GTGCCAGCMGCCGCGTAA</i>	806R <i>GGACTACHVGGGTWTCTAAT**</i>
	806bR <i>GGACTACNVGGGTWTCTAAT</i>
	909R <i>CCCCGYCAATTCTTTTRAGT</i>
	926R <i>CCGTCAATTCTTTTRAGTTT</i>
515yF <i>GTGYCAGCMGCCGCGTAA</i>	926pfR <i>CCGYCAATTYMTTTRAGTTT</i>
515wF <i>GTGYCAGCMGCCGCGTA</i>	
530F <i>GTGCCAGCMGCNGCGG</i>	
926wF <i>AAACTYAAAKGAATTGRCGG</i>	1392R <i>ACGGGCGGTGTGTRC***</i>
939F <i>TTGACGGGGGCCGCACAAG</i>	1492R <i>TACCTTGTACGACTT</i>

Archaeal Assays

16S

Arch340wF <i>CCCTAYGGGGYGCASCAG</i>	Arch806R <i>GGACTACVSGGTATCTAAT</i>
<i>Arch517F GCYTAAAGSRNCCGTAGC</i>	<i>Arch909R TTTCAGYCTTGCGRCCGTAC</i>
Arch519wF <i>CAGCMGCCGCGTAA</i>	Arch1017R <i>GGCCATGCACCWCCTCTC</i>

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Eukaryote/Fungi Assays
18S

Euk1AF CTGGTTGATCCTGCCAG	Euk516R ACCAGATTGCCCTCC
EukA7F AACCTGGTTGATCCTGCCAGT	Euk570R GCTATTGGAGCTGGAATTAC
Euk300F AGGGTTCGATTCCGGAG	Euk555R GCTGCTGGCACCAAGACT
Euk515F GTGYCAGCMGCCGCGGTA	EukNSR951R TTGGYRAATGCTTCGC
	Euk1052R TGCATGGYYGYCGYCAGYTC
Euk516F GGAGGGCAAGTCTGGT	Euk1055R CGGCCATGCACCACC
Euk1560F TGGTGCATGGCCGTTCTTAGT	Euk2035R CATCTAAGGGCATCACAGACC
Euk1391F GTACACACCGCCCCGTC	EukBR TGATCCTCTGCAGGTTCACCTAC
<i>TAReukF CCAGCASCYCGGTAATTCC</i>	<i>TAReukR ACTTCGTTCTTGATYRA</i>
	EukBalR ACTTCGTTCTTGATYRR
	EukSto2R ACTTCGTTCTTGAT
	TAReukR3mR ACTTCGTTCTTGATYRATGA

ITS

ITS1F CTTGGTCATTTAGAGGAAGTAA	ITS2aR GCTCGTTCTTCATCGATGC
<i>ITS3F GCATCGATGAAGAACGCGAGC</i>	<i>ITS4R TCCTCCGCTTATTGATATGC</i>
ITS9F GAACGCAGCRAAIIGYGA	

23S

AlgaeF GGACAGAAAGACCCTATGAA	AlgaeR TCAGCCTGTTATCCCTAGAG
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Arthropod COI

mlCOlntF GGWACGGWTGAACWGTWTAYCCYCC

HCO2198R TAAACTCAGGGTGACCAAAAAATCA

Plant trnL

trnLcF CGAAATCGGTAGACGCTACG

trnLdR GGGGATAGAGGGACTTGAAC

trnLgF GGGCAATCCTGAGCAA

trnLhR CGCGCATGGTGGATTACAATCC

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