



AMPLICON BASED  
**METAGENOME,**  
WHOLE META GENOME &  
**META TRANSCRIPTOME**  
SEQUENCING

A Lifesciences Division of

*Unipath*  
SPECIALTY LABORATORY Ltd.



[www.unigenome.in](http://www.unigenome.in)

# Amplicon based Metagenome, Whole MetaGenome and Meta Trascriptome Sequencing

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Microbiomes are ubiquitous and are found in the environment like ocean, the soil, and in/on other living organisms. Microbes are responsible for the health of the environmental niche in which they reside. In order to learn more about these communities, different approaches based on data from multiple omics have been pursued. While amplicon based metagenomics produces a taxonomical profile of the sample, metagenomics and metatranscriptomics helps us to obtain a functional profile, and metabolomics completes the picture by determining which byproducts are being released into the environment. Smaller version of metagenomics also includes amplicon based studies like 16s, 18s or ITS based on their target microbiome. One of the most common approaches to studying a microbiome is analyzing its constituent microbial genomes through meta-genomics. More recently, this definition has evolved to include not only the microbes and their genomes but also the aggregate of environmental and host factors. The inclusion of the host environment as part of the microbiome significantly expands its implications, with the interactions between the host and its associated microbial community now relevant to understanding the dynamics of the microbiome.

Metagenomics is the study of the genomes of microbial community as a whole and constitutes the first step to studying the microbiome. Main objective of the study is to infer the taxonomic profile of a microbial community. Metagenomics provides a partial glimpse into the functional profile of a microbial community which is better inferred using metatranscriptomics, which involves sequencing the complete (meta) transcriptome of the microbial community. Metatranscriptomics gives information of the genes that are expressed by the community as a whole. With the use of functional annotations of expressed genes, functional profile of a community under specific conditions can be identified, which are usually dependent on the status of the host. Basic difference between the two is, metagenomics helps address the question “what is the composition of a microbial community under different conditions?”, and metatranscriptomics helps answer the question “what genes are collectively expressed under different conditions?”

## Service types

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- Amplicon based metagenome analysis using 16S/18S/ITS genes
- Whole metagenome analysis
- Whole metatranscriptome analysis

## Sample Requirements

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Metagenomic DNA for Amplicon based and whole metagenome sequencing

- Total 4-5µg of purified high molecular weight intact double stranded gDNA, free from RNA contamination is desirable.
- Metagenomic DNA should have an absorbance ratio (A260/280) of ~1.8 to 2.0 with minimum 300-500 ng/µl concentration.

## Metatranscriptomics RNA

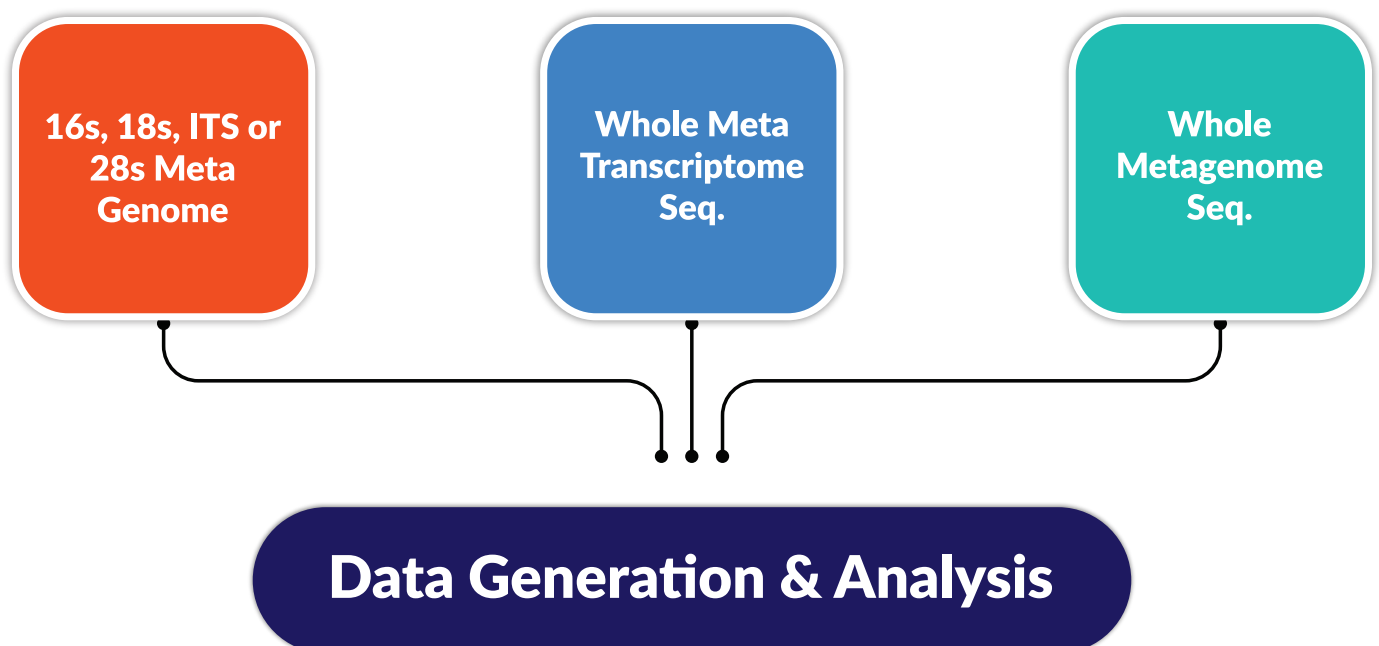
- 8-10 µg total RNA should be provided with RNA Integrity Number (RIN)>6.
- RNA must not be degraded and should be free from DNA contamination
- Inclusion of low RIN value of the samples will be processed upon customer's confirmation. Samples should be transported in cool packs to Unigenome, Ahmedabad, Gujarat, India.

## Quality control of Metagenomic DNA/Metatranscriptomics RNA

- Metagenomic DNA samples will be subjected to both qualification and quantification by 1% agarose gel electrophoresis & Qubit/Nanodrop respectively.
- Metatranscriptomics RNA samples will be subjected to both qualification and quantification by 1% denaturing agarose gel electrophoresis & Qubit/Nanodrop respectively. However, degraded RNA of the samples will be processed upon customer's confirmation and their risk.

## Work flow of Metagenome Sequencing

### Soil, Water, Rumen, Stool Any other sample



# Deliverables

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## Whole Metagenome Sequencing & Analysis on Illumina Platform

- Clean reads after filtration of adapter and low quality bases
- De novo assembly of reads to generate scaffolds
- Coding gene prediction from scaffolds
- Taxonomic assignment of predicted genes (Phylum to species level)
- Functional annotation of predicted genes against KEGG (including EC number and its abundance), GO, PFAM, COG and FIG databases
- Graphical representation for taxonomy assigned (bar chart, Krona graph) Comprehensive and compiled report and data deliverables

## Whole Meta transcriptome Sequencing and Analysis on Illumina Platform

- Clean reads after filtration of adapter and low quality bases
- De novo assembly of reads to generate scaffolds
- Coding gene prediction from scaffolds
- Taxonomic assignment of predicted genes (Phylum to species level)
- Graphical representation for taxonomy assigned (bar chart, Krona graph)
- Functional annotation of predicted genes against KEGG (including EC number and its abundance), GO, PFAM, COG and FIG databases
- Expression profiling of predicted coding genes
- Differential expression analysis of predicted genes (if more than 1 sample) Comprehensive and compiled report and data deliverables

## Amplicon based Metagenome for 16S, 18S or ITS

- Clean reads after filtration of adapter and low quality bases
- Trimming of primers
- Joining paired-end reads
- Denoising of joined reads
- Representative sequence set generation

- Taxonomy classification against Silva/Unite database
- Taxonomy barplot
- Rarefaction curve
- Heatmap for most abundant taxa
- Alpha diversity analysis
- Beta diversity Analysis and emperor plot (if more than 2 samples)
- Comprehensive and compiled report and data deliverables

**Note:**

- Custom amplicon based metagenome sequencing and analysis for COI, CytB, 12S etc. is also provided
- Customized analysis as per client's need is provided for any of the above mentioned services with additional charges



A Lifesciences Division of



**UNIGENOME**

Unipath House Behind Sahajanand College,  
Opposite Kamdhenu Complex, Panjarapole,  
Ambawadi, Ahmedabad-380015



[www.unigenome.in](http://www.unigenome.in)



[info@unigenome.in](mailto:info@unigenome.in)