

# SAMPLE SUBMISSION GUIDE

## Microbiome Profiling

Using amplicon sequencing on Illumina MiSeq

Sending samples according to the requirements below provides you best possible data quality and results!

## Sample Requirements:

For questions and special samples, please do not hesitate to contact us at ngsalliance-eu@eurofins.com.

## Samples for DNA extraction

Please ship the raw material samples in appropriate safe lock tubes. If not stated otherwise you may ship your samples at ambient temperature.

#### Fermented products (cheese, yoghurt, etc.):

Ideally, please send 5 -10 g of sample or a whole packing in a sealed container.

#### Enrichment cultures / starter cultures:

Please send a representative sample or an overnight culture freshly frozen or cooled on ice packs.

#### **Buccal swabs**

Please send 2 swabs per sample frozen or chilled.

## Human or animal faeces

Please send minimal 2 x 250 mg of faeces frozen.

#### Water, waste water & other liquids

- Please filter 500 ml water sample through a 0.22 µm pore size filter. Filter should be filled to full capacity to harvest 10<sup>9</sup> bacterial cells. Filter should be free and removed from the holder (i.e. not attached to a column). Before sending the filter should be dried and wrapped in sterile aluminium foil.
- If you do not have the possibility to filter, please contact us.

#### Soil & other sediment samples

Please send minimal 2 x 250 mg of frozen samples.



## **Extracted DNA samples**

Please provide your DNA samples in Tris-buffer / buffer from commercial DNA extraction kits with minimum volume and concentration as given in the table below. If available, please send more material than requested.

Samples need to be shipped in <u>96-well plates</u> at ambient temperature. Please seal the plates using cap stripes.

Sample type	Purified microbial DNA	DNA from host and bacteria / fungi
Volume	1 target: 20 µl 2 targets: 30 µl 3 targets: 40 µl 4 targets: 50 µl	1 target: 20 µl 2 targets: 30 µl 3 targets: 40 µl 4 targets: 50 µl
Concentration	10 - 50 ng/μl	50 ng/μl

## **Amplicons for index PCR option**

Please provide amplicons generated with NGS primers (consisting of the target specific sequences defined by the customer and the adapter sequences defined below).

Illumina adapter sequence that has to be added on the 5'- end of the forward primer: 5'-TCGTCGGCAGCGTCAGATGTGTATAAGAGACAG-forward primer target sequence-3'

Illumina adapter sequence that has to be added on the 5'- end of the reverse primer: 5'- GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAG-reverse primer target sequence-3'

Please note that the complete sequence of your primers is necessary for sorting of reads and the optional bioinformatics analysis. Sequences are to be given in our sample submission form.

Amplicons size (including the adaptors you introduced with your target specific PCR) needs to be between 400 – 900 bp in order to be accepted.

Quality check needs to be performed by agarose gel electrophoresis (= clear visible band(s) of expected size using 5  $\mu$ I PCR product). The gel picture needs to be handed out to Eurofins Genomics. No purification step should be performed at customer's lab. The total required volume/amplicon is 25  $\mu$ I. Please specify the agarose gel including volume loaded, the size of the amplicons and the type of marker (incl. manufacturer).

Amplicons need to be shipped in <u>96-well PCR plates</u> with 200 µl well volume at ambient temperature (no deep well plates).



## Sample Submission:

#### General instructions

- Prepare your samples. Please use water-resistant markers to label your samples or plate(s) directly and avoid removable stickers. Sample name or plate name needs to be unique and identical to the sample description in the sample submission form (Excel). Correct sample labelling is in the responsibility of the customer.
- Download the appropriate sample submission form (Excel sheets) for either <u>samples for DNA extraction</u>, <u>extracted DNA</u> or <u>amplicons ready for index PCR</u>, and fill in all necessary information. (www.eurofinsgenomics.eu/ngs-guide)
- 3. Please send the sample submission form electronically prior to start of the project to the following Email-address: MicrobiomeProfiling\_Submission@eurofins.com.
- 4. Prepare your parcel for shipping. Please make sure to provide samples in sealed containers in your transport box. It is mandatory that you send a copy of your order confirmation from the online shop and from the sample submission form on top of the sealed container. Raw Material with potential S2 organisms: Samples need to be packed in sealed & thick plastic bags before placing in the transport box.

## Sample Shipment

Please send your samples to:

APG - Laboratory Eurofins Genomics Anzinger Str. 7a 85560 Ebersberg Germany

## Important customs information

**EU:** Original samples or samples in lysis buffer may be provided by the customer.

**Non-EU countries:** Samples need to be sent to Eurofins Genomics either as extracted DNA or in lysis buffer. Please contact us regarding your choice of lysis buffer or containers prior to sample collection.

In case you plan to send your sample in lysis buffer, please contact us to select an appropriate lysis buffer.