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info@starseq.com



Illumina MiSeq Sample Submission Form and Offer Acceptance

Fill in the order form completely! Don't forget your signature (otherwise no processing)! It is mandatory to completely fill in and sign this submission form. A printed and signed copy must be sent together with the samples. The project will not commence until the filled acceptance sheet has been returned.

| Customer Information | | | | |
|---|---|------------------------|--|--|
| Name*: | | | | |
| Company/Institution*: | | | | |
| Institute/Department: | | | | |
| Address*: | | | | |
| VAT-ID*: | | | | |
| Phone: | | | | |
| Email*: | | | | |
| StarSEQ Quotation No*: | | | | |
| Purchase Order No*: | | | | |
| | | | | |
| | voicing Information (if different from above) | | | |
| Name*: | | | | |
| Company/Institution*: | | | | |
| Institute/Department: | | | | |
| Address*: | | | | |
| VAT-ID*: | | | | |
| Phone: | | | | |
| | | | | |
| Email*: | | | | |
| *Required For EU countries: Please provide | ne VAT-ID No. of your company. If this certificate is not prese ountries: Send us an official confirmation of your entreprene | | | |
| *Required For EU countries: Please provide | ountries: Send us an official confirmation of your entreprene | | | |
| *Required For EU countries: Please provide | | | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU | ountries: Send us an official confirmation of your entreprener Sample Type | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA | Sample Type ChIP-Seq | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA | Sample Type ChIP-Seq Genomic DNA (Exome Enrichme | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA mRNA | Sample Type ChIP-Seq Genomic DNA (Exome Enrichmen Small RNA | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA mRNA cDNA | Sample Type ChIP-Seq Genomic DNA (Exome Enrichmeter) Small RNA Plasmid DNA | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA mRNA cDNA 16S/ITS | Sample Type ChIP-Seq Genomic DNA (Exome Enrichme) Small RNA Plasmid DNA Amplicon | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA mRNA cDNA 16S/ITS Amplicon (FFPE) | Sample Type ChIP-Seq Genomic DNA (Exome Enrichment Small RNA Plasmid DNA Amplicon Ready to load library Stool/Swab/Blood/Saliva | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA mRNA cDNA 16S/ITS Amplicon (FFPE) Tissue/Soil | Sample Type ChIP-Seq Genomic DNA (Exome Enrichme) Small RNA Plasmid DNA Amplicon Ready to load library Stool/Swab/Blood/Saliva Sample information | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA mRNA cDNA 16S/ITS Amplicon (FFPE) Tissue/Soil Concentration: | Sample Type ChIP-Seq Genomic DNA (Exome Enrichme) Small RNA Plasmid DNA Amplicon Ready to load library Stool/Swab/Blood/Saliva Sample information Number of samples: | urial activity please. | | |
| *Required For EU countries: Please provide 19% will be charged. For non-EU Genomic DNA Total RNA mRNA cDNA 16S/ITS Amplicon (FFPE) Tissue/Soil | Sample Type ChIP-Seq Genomic DNA (Exome Enrichme) Small RNA Plasmid DNA Amplicon Ready to load library Stool/Swab/Blood/Saliva Sample information Number of samples: | urial activity please. | | |

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| Requested Application | | | |
|-----------------------|-----------------|--|--|
| Library Prep □ | gDNA/RNA Prep 🛚 | Sequencing □ | |
| Library Prep Type: | | Sequencing Type: MiSeq | |
| gDNA | ☐ PCR-free ☐ | Single Read (only 50 (V2) and 150 bp (V3) □ | |
| mRNA | ☐ Stranded ☐ | Paired End Read □ | |
| small RNA | | 50 nt □ | |
| Mate Pair | | 75 nt (V3) □ | |
| ChIP-Seq/FFPE | | 150 nt (V3) □ | |
| Exome Enrichment | | 150 nt Micro □ | |
| Microsatellite | | 150 nt Nano □ | |
| gDNA Nextera XT/Ag | gilent QXT 🔲 | 250 nt □ | |
| Total RNA (incl. rRNA | A depletion) □ | 250 nt Nano □ | |
| Exome enrichment | | 300 nt (V3) □ | |
| Amplicon: 16S□ I | TS ☐ Region: | Cutom primer (must be provided by customer) \Box | |
| Size selection | | | |

| Sample Information | | | | | | | |
|--------------------|--------|---------|---------------|--------------------|--------|--------------|---------|
| | Sample | Species | Concentration | Size (if providing | Volume | Index (ready | Total |
| | Name | | (ng/μl) | a library or | (µI) | to load | DNA/RNA |
| | | | | ChIP-Seq/FFPE | | libraries | (µg) |
| | | | | samples) | | only) | |
| Sample 1 | | | | | | | |
| Sample 2 | | | | | | | |
| Sample 3 | | | | | | | |
| Sample 4 | | | | | | | |
| Sample 5 | | | | | | | |
| Sample 6 | | | | | | | |
| Sample 7 | | | | | | | |
| Sample 8 | | | | | | | |
| Sample 9 | | | | | | | |
| Sample 10 | | | | | | | |
| Sample 11 | | | | | | | |
| Sample 12 | | | | | | | |
| Sample 13 | | | | | | | |
| Sample 14 | | | | | | | |
| Sample 15 | | | | | | | |
| Sample 16 | | | | | | | |
| Sample 17 | | | | | | | |
| Sample 18 | | | | | | | |
| Sample 19 | | | | | | | |
| Sample 20 | | | | | | | |

For more samples please send a spread sheet by email. Sample names may only contain letters, numbers, underscores and minus signs.

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| Additional Comments/Information: | | | | | |
|----------------------------------|--|--|--|--|--|
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| Sample | Application/ Library Prep Method | Minimum required amount | Recommended amount | Minimum Concentration | Minimum Volume |
|--|---|---|--|-------------------------------------|-------------------|
| Genomic DNA | de novol resequencing | 100-200 ng* | 500 ng | 5 ng/μl | 20 µl |
| Genomic DNA | <i>de novol</i> resequencing PCR-free | 1-2 μg* | 3 µg | 20 ng/μl | 20 µl |
| Plasmids, cDNA, amplicons | de novol resequencing | 100-200 ng* | 300 ng | 20 ng/μl | 10 µl |
| Genomic DNA | Exome Enrichment | <i>200</i> ng | 500 ng | 25 ng/µl | 10 µl |
| Genomic DNA | Mate Pair | 4 μg | 5 μg | 20 ng/µl | 20 µl |
| Genomic DNA | 16S/ITS | 50 ng | 200 ng | 5 ng/µl | 10 µl |
| Genomic DNA | Microsatellite | 1 μg | 4 μg | 20 ng/µl | 20 µl |
| Genomic DNA, plasmids, amplicons, cDNA | Nextera XT/ Agilent SureSelect QXT | <i>1/50</i> ng | 50/200 ng | 2/25 ng/μl | 10 µl |
| ChIPseq | ChIPseq | <i>10</i> ng | 100 ng | 1 ng/µl | 10 µl |
| Total RNA | stranded RNA-seq poly A+ | <i>100</i> ng | 1-2 µg | 20 ng/μl | 10 µl |
| mRNA | RNA-seq/ stranded RNA-seq | <i>10</i> ng | 200 ng | 1 ng/μl | 10 µl |
| rRNA depleted total RNA | RNA-seq/ stranded RNA-seq | minimum amount of total RNA before depletion = 100 ng, delivered Volume must be 5 µl | maximum amount of total RNA before depletion = 4 μg, delivered Volume must be 5 μl | delivered Volume must be 5 µl | 5 μΙ |
| Total RNA | small RNA | 1 μg | 2 μg | 200 ng/μl | 10 µl |
| Prepared small RNA | small RNA | 5 μl small RNA isolated from 1 μg total RNA | 5 μl small RNA isolated from 1 - 10 μg total RNA | | 5 μΙ |
| Ready to load libraries | | 10 nM (10 μl) | 10 nM (10 µl) | | 10 µl |

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All samples have to be provided in nuclease free water or buffer (10 mM Tris-HCl, pH 8.5). **No EDTA!** Do not use Parafilm to seal your tubes.

All samples should be free of contaminants that could inhibit enzymatic reactions. Precipitation with salt and EtOH is not suitable. Please use clean kit based preparation of samples. We recommend to use fluorometric-based methods for quantification (Qubit or PicoGreen) and 260/230 + 260/280 ratios to identify possible contaminants.

RNA samples should have an RNA Integrity Number (RIN) value greater or equal to 7. RNA sample shipment strictly has to occur on dry ice and should undergo DNase I treatment prior to submission.

For ready to load libraries, delivery of Bioanalyzer data or data from comparable technique is mandatory. As alternative StarSEQ can offer this service (see below).

Custom sequencing primer (read 1, index, read 2) must be delivered as 100 µM dilution. For microsatellite PCR assay development we need at least the gDNA from 16 unrelated individuals (500 ng/sample).

Genomic DNA (hmw) samples and RNA samples should not be degraded and are required to have the following OD ratios:

260/280 = 1.8 - 2.0260/230 = 1.7 - 2.2

The use of too little or degraded starting material (RNA/DNA) can result in low yield or failure of the library preparation. Library and amplicon preparation from low quality samples will induce a bias and may result in a failed library preparation. Preparing libraries from low quality samples is on customers risk and will be charged.

For custom or ready-to-load libraries do not use any indices beginning with two G bases (e.g. do not use Illumina index no. D504, N718, N705, RPI24, R724, R711, NR011, ND011). Custom libraries must be larger than 250 bp and less than 800 bp. The fragment range of the ready-to-load libraries is not allowed to exceed 400 bp.

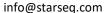
If the amount, concentration and/or quality of the starting material does not meet the requirements (see above) for the library preparation, StarSEQ will contact you to discuss how to proceed. If possible, StarSEQ will recommend additional pre-processing steps in order to optimize the sample quality. Size selection will be mandatory if sample quality or sample contamination affect enzymatic tagmentation or fragmentation resulting in large fragments. If additional services are requested, the following prices per sample apply:

- 1. RNA / DNA concentrating, € 25 / sample
- 2. RNA / DNA purification, € 40 / sample
- 3. RNase treatment, € 30 / sample
- 4. DNase treatment, € 30 / sample
- 5. Quality check of (replacement) samples on Bioanalyzer or Qiaxcel, € 20 / sample
- 6. Quantification of (replacement) samples on Qubit, € 10 / sample
- 7. Size selection, € 50
- 8. Run preparation with custom primers (read1, index 1, read 2) € 80 / sample
- 9. Normalization and pooling of 96 ready to load amplicon based libraries (e.g. 16S or ITS) € 300

The customer provided material is not infectious and not hazardous. The Data amount estimation relies on specifications of Illumina and may vary depending on the nature of starting material. For quality control and verification, StarSEQ will spike in up to 3% of PhiX to all runs. StarSEQ will give no guarantee for sequencing of custom/customer prepared libraries. Failed

^{*} dependent on desired size range of library

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runs from poor custom libraries will be charged. For ready to load libraries, the customer will provide Bioanalyzer data or similar and sample names with index sequences as excel sheet. Indexing strategy must be discussed prior to project. StarSEQ will give no guarantee for successful library preparation from rRNA depleted material, all libraries will be charged if library preparation fail. StarSEQ will give no guarantee for successful library preparation from samples that did not fit our requirements. All libraries will be charged if library preparation fail due to missed sample requirements. For difficult samples like amplicons and low diversity samples (e.g. 16S, ITS) it is necessary to co-load up to 30% PhiX on the lane to provide the necessary diversity in the composition of the bases. In such cases the estimated data output is reduced in proportion to the content of PhiX. According to Illumina specifications overall Q30 values for low diversity samples are expected to be not higher than 70%. Placing an order is possible only via our sample submission form.

All conditions defined in the sample submission form apply to this quotation. The sample submission form has to be filled in completely. Failed library preparation or sequencing attempts due to lack of information will be charged.

Turnaround time will be determined when StarSEQ has received the samples. Depending on the nature of starting material and according to Illumina specifications the data output can vary 5-10%. For difficult samples like amplicons, low quality or low diversity samples, it is possible that the specifications cannot be reached.

Please note, that your samples will be stored under the appropriate conditions for 3 months after analysis. Hereafter the samples will be deleted if no other arrangement is made regarding shipment or storage of the samples. Please create one or more backups immediately after receiving the data. StarSEQ store the data for a maximum of 6 months, after which it will be deleted.

For Research Use Only – not for any clinical, diagnostic or therapeutic use in humans or animals.

With below Signature I confirm above conditions and sample requirements (page 1-5) and order legally binding the quotation noted in this document. I also confirm that the provided material is not infectious and not hazardous.

Placing of order and offer acceptance:

| Name (capitals): | | | |
|------------------|--|---|--|
| | | | |
| | | | |
| | | | |
| Signature/Date: | | / | |