

Department of Genetics
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Tender Enquiry for Next Generation Sequencing

UDSC/GEN/BKT/NGS/04/2021

April 16, 2021

E-tender is invited from national firms/institutes with experience and expertise to perform

- 1) Whole exome sequencing; and
- 2) Whole genome sequencing of human samples;
- 3) Whole Metagenome Sequencing; and
- 4) 16S rRNA (V3-V4) Sequencing from Human stool and tissue samples
as per specifications below.

Technical and financial bids should be submitted separately through CPP Portal (tender available on CPP Portal) on or before May 6, 2021.

Please Note: Only vendors with the specified sequencing platforms available in their respective facilities in INDIA need apply

1) Whole Exome Sequencing (without downstream data analysis):

Technical specifications:

- Whole exome sequencing should be performed using the Illumina HiSeq X Ten or NovaSeq 6000 platforms
- Genomic DNA will be provided by the client
- Exome enrichment should be performed using Agilent SureSelect Human All Exon V5/V6/V8/ V5+UTRs /V6+UTRs /V8+UTRs as per order. Cost for all of these may be quoted separately
- Each sample should be sequenced to at least 100fold (100X) mean on target depth with 101/150-bp paired-end sequencing method
- For each sample >99% target regions should be captured and sequenced with minimum 1X, and >98% of the target region should be captured and sequenced with minimum 10X. The target depth calculation should be done on per base basis
- Minimum 8 GB clean data per sample (after removal of adapters and low-quality sequence/reads, Phred quality score (Q score) <20 and reads with <70bp) should be delivered
- >90-95% of total data should be >Q30 Phred score



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- >99% of the sequence/reads (after the removal of PCR duplicates and low-quality sequences) should be aligned to the human reference genome (hg19). Alignment should be performed using BWA and alignment statistics calculation should be based on Qualimap and Samtools flagstat
- Summary of complete sequencing runs should be provided

Sample Details: A total of 100-400 samples will be sequenced in this project. It will be the responsibility of the Service provider to collect the samples from the lab and transport to the sequencing centre under proper conditions. The client will not provide any payment for the transportation of the samples.

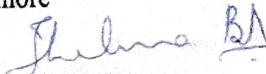
Data delivery deadline: Sample DNA QC analysis should be done at no extra cost and report should be submitted within 5 days after receiving the samples. Raw FastQ file, Clean FastQ file and sequencing details should be reach the client in hard disk(s) within 45 days after sample QC is completed.

Please Note: If the client observes that the data is not up to the standard mentioned above, the service provider should accept it or disprove the claim by the client within three days using the same data delivered by the service provider and software used by the client using default parameters. If not done within three days data provided will be considered as bad quality and below mentioned conditions will automatically apply.

2) Whole Genome Sequencing (without downstream data analysis):

Technical specifications:

- Whole genome sequencing should be performed using the Illumina HiSeq X Ten/NovaSeq6000 platform. Preference will be given to NovaSeq 6000 using S4 flow cell and Reagent Kits v1.5
- Genomic DNA will be provided by the client
- PCR free library preparation kit, preferably Illumina DNA PCR-Free Prep or Illumina TruSeq DNA PCR-Free should be used
- DNA input for library preparation must be quantified using the Qubit fluorometric quantitation of Thermo and the reading should be provided to the client. Similarly final concentration and volume of DNA library prior and post normalization should also be provided
- Normalization calculation of DNA library should be based on a Q-PCR reading in triplicates and not based on other fluorometric methods
- Sequencing should be done using 350bp paired end sequencing at an average coverage >30X with data output of 115 GB per sample. (Average base pair coverage should be between 30X and 40X, samples with coverage rate <30X will be considered as failure)
- At least 95% of the target region (hg19) should be covered with 10X or more


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- Minimum 90 GB of clean data per sample to be obtained after removal of adapter and low-quality sequence/reads
- >80-85% of total clean data should be >Q30 Phred score. Sequence analysis viewer (SAV) report must be provided for all runs to support the Q30 pass % rate
- DNA insert size report using a bioanalyzer of the final DNA library must be provided along with the Q30 report of every sequencing run
- Overall duplicates should be less than 15%

**Sample details: A total of 50-100 samples will be provided in total in this project.
All other conditions remain same as for WES.**


Data analysis cost for both WES and WGS may be quoted separately and time frame should be provided for the same.

Other mandatory conditions

- 1) Vendors should submit evidence of prior experience for human whole exome/genome sequencing
- 2) Vendors should have done whole exome sequencing of a minimum of 3000 and whole genome sequencing of 100 samples to date. Complete contact details of the clients for whom the above mentioned work has been done should be provided. The firm should also submit a satisfaction report from clients (PIs) (of already completed projects with minimum of 50 samples at one time) certifying both the quality of data and keeping to time frame of the order. Appropriate web link/documentation to support this claim should preferably accompany the proposal
- 3) List of clients/institutes where vendors successfully completed whole exome/genome sequencing projects in last one year should be submitted
- 4) Vendors should provide evidence of technical expertise and experience in this area for completion of this work.

Other terms:

1. The complete work should be done by the vendor within India
2. The vendor should provide a facility certificate to testify the possession of an Illumina HiSeq X Ten/NovaSeq 6000 platforms
3. Organization should be involved in providing services for the last two years
4. Copy of the purchase order (which they received from other organization) should be provided
5. The vendor should specify the GST tax in the financial bid
6. CAP or Equivalent accreditation for Human Whole Genome Sequencing is preferred


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Payment: Payment by bank transfer will be made only after the receipt of the complete data in accordance with criteria already mentioned in the tender document. If data does not fulfil the above detailed criteria, the data will not be accepted and payment will not be made. If timely delivery of data is not done, penalty will be applied. **In case of late delivery of data 1% of total cost will be deducted for delay of every day and delay of more than 25 days will lead to the cancellation of the order. If vendor does not follow the above mentioned detailed criteria the client has the full right to cancel the agreement with the vendor at any time. In that case no payment would be made.**

The selection of the firm for the above mentioned service will be based on a two bid system. Commercial bids of only those vendors who clear the technical specifications will be opened. The client shall not be under any obligation to accept the lowest or any other offer received in response to this tender notice and shall be free to reject any or all offers. Furthermore, the client is not obliged to meet and have any discussion with any vendor.

The qualified vendor should provide a bank guarantee of 30% of the total cost and should sign an agreement within five working days after receiving the work order, after which the samples will be handed over. This guarantee money will be returned immediately after successful completion of the work. If the vendor does not complete the project on time with above mentioned conditions the order will be cancelled and bank guarantee will not be returned. If the vendor cancels the order due to their own internal problems the bank guarantee will not be returned.

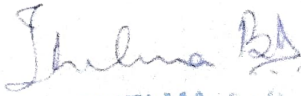
Compliance sheet mentioning each of the points specified in the tender should be submitted along with the technical bid.

3. Metagenome Sequencing:

Technical specifications:

Complete DNA sequencing using whole metagenome extracted from Human stool or other tissue samples should be performed using Illumina HiSeq X Ten/NovaSeq 6000 platform

- Genomic DNA will be provided by the client
- Chemistry: 250bp x 2 paired end reads
- Data Size: 3-5GB clean data, with 10 GB raw data per run/sample, preferably run with flow cell type -SP
- Raw data should be provided in FastQ format
- Data analysis should include:
 - a) Library QC and filtering of reads
 - b) Assembling reads into contigs
 - c) Mapping, annotation and OTU generation
 - d) Taxonomic and functional annotation of OTUs with standard software


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Detailed methodology:

1. Whole genome library preparation and sequencing
2. Deep sequencing of the samples using appropriate platform for complete metagenome analysis for microbial diversity (alpha & beta), abundance & functional prediction using optimized and published pipelines
3. Metagenome analysis with suitable server for both assembled and unassembled data and validation of unassembled microbes
4. Metagenomic sequences should be annotated using evidence based annotation approach for all sequences against protein databases
5. Genes prediction should be tabulated and classified into functional categories from individual genes to cellular processes
6. Relative abundance for each gene should be calculated
7. Functional analysis of bacteria along with identification of pathways)

Sample details: A total of 100-400 samples will be provided in this project.

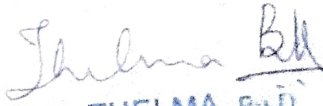
4. 16S rRNA (V3-V4) Sequencing:

Technical specifications:

- Genomic DNA will be provided by the client
- Amplification and sequencing of 16S region (V3/V4) should be done with Illumina MiSeq platform
- Each sample should be sequenced with 2x250 paired end reads sequencing depth to deliver at least 0.5M data.
- Detection sensitivity should be around 1:1000
- Primary QC and analysis until OTU generation (using closed reference and *de novo*) should be performed and files should be provided
- Detailed technical note should be provided for methodology used and Bioinformatic analysis (co-occurrence network, picrust analysis etc.)
- Cost for downstream analysis should be mentioned separately

Sample details: A total of 100-1000 samples will be provided in this project.

Please note: All other terms as mentioned above will apply for these projects also.


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