Department of Genetics
University of Delhi South Campus
New Delhi- 110021

June 6th 2016

Limited Tender Enquiry for Mixed Transcriptome Sequencing of plant-fungal interaction

Sealed quotations are hereby invited from national and international firms/institutes with experience and expertise for **transcriptome sequencing of 48 plant (*Arabidopsis thaliana*) samples with data analysis** to be carried out as per specifications below. The sequencing and data analysis cost should be mentioned separately in the tender. **The tender is to be submitted in two parts, viz., technical and financial bids in separate sealed envelopes** and may be sent to the under signed within 21 days of advertisement of the tender on the university website.

mRNA sequencing:

- 200 bp sequencing libraries should be prepared using the TruSeq Stranded mRNA Library Prep Kit.
- Quantitative standards (spike-ins) should be used at the time of cDNA synthesis to calibrate quantification, sensitivity, coverage and linearity.
- 2 x 125 bp paired end sequencing should be performed using the Illumina HiSeq 2500/3000/4000 platform.
- Sequencing should be carried out using the Illumina SBS v4 Kit.
- Each sample should have a read depth of atleast 50 million raw reads with > 40 million reads after initial QC of > Q30 Phred score.
- For each sample > 95% of the leaf transcriptome should be covered and sequenced with atleast 15X coverage (with less than 5% clonal/duplicated reads)
- More than 4 GB cleaned data for each sample (adaptor and low quality sequence/reads removed) should be delivered. The proportion of clean data and raw data should be 90% or above.

- Summary of complete sequencing runs should be provided along with raw fastq files.

Downstream analysis:

1) **Read quality check and data filtering:** Detailed quality report before and after trimming should be provided. It should include base quality and sequence quality score distribution details, average base content and GC distribution in the reads, check for over-represented sequences, duplicate removal, adapter trimming details, read length details (percentage of read length distribution), parameters used for trimming and names and details of software used and other details of trimming/removal of low quality sequence/reads etc.

2) **Read alignment:** The paired end reads should be aligned to reference transcriptome of Arabidopsis (TAIR 10/11). Alignment should be done using standard tools. Details of parameters used for alignment and software details, read alignment statistics and quality metrics for each sample should be provided.

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3) **Details of target depth and raw depth:** Percentage of transcripts covered in each sample and details of transcripts not adequately covered and coverage depth details should be provided for each gene/sample.

4) **Extraction of fungal transcripts from infected samples:** Transcripts that do not map to the plant reference should be extracted, mapped on fungal reference genomes and annotated using fungal databases.

5) **Bioinformatic analysis (including but not limited to):**

a. Expression Quantification
b. Differential gene expression analysis (FPKM/RPKM of Arabidopsis genes)
c. DGE analysis of *in planta* expressed fungal genes.
d. GO enrichment analysis
e. Pathway and network analysis
f. Alternative splicing
g. Novel gene prediction

Sample Details: 48 RNA samples will be sent to the firm.

Data delivery deadline: Sample RNA QC report should be submitted within 7 days after receiving the sample. Raw FastQ files should be submitted as soon as the sequencing is carried out and before the data clean up and downstream analysis is undertaken. Raw FastQ file, Clean FastQ file and other quality and parameter files mentioned above (at 1) should be delivered within 30 days after sample QC completed. BAM file, filtered and unfiltered VCF files (individual sample's VCF and all samples together), quality files and final data analysis report of analyses mentioned in (5) should be delivered within 60 days after sample QC check is completed. Analysis should be satisfactory in all respect to the undersigned/principal investigator. Data generated should be kept confidential and avoid sharing/deposition.

Other conditions

- Firm/Institute should submit evidence of prior experience for mixed transcriptome sequencing in plant-pathogen interactions as publication in reputed journals and acknowledgements (minimum 5)
- List of clients/institutes where firm successfully completed mixed transcriptome sequencing projects should be submitted (min. 5).
- Firm should have an in-house sequencing facility and experienced personnel for data analysis, for which adequate proof (e.g. Installation certificate) should be provided (should not be on lease or tie up).
- Samples should be sequenced in the respective facilities in India and samples or data should not be outsourced to firms outside India at any stage.

Payment: Payment will be made only after the receipt of the complete data (with above criteria) in accordance with above mentioned requirements/criteria. **If data does not fulfill 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.**

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If firm/institute does not follow the above mentioned detailed criteria the University/Lab has the full right to cancel the agreement with the firm/institute 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 lowest quoted price, evidence of expertise in the field and technical specification mentioned in the tender.

The University reserves the right to accept or reject quotation without assigning any reason thereof.

Quotations may be sent to –

Dr. Jagreet Kaur
Department of Genetics
First floor, Biotech Centre
University of Delhi South Campus
Benito Juarez Road
New Delhi – 110021

Thanking you.

Yours sincerely,

Jagreet Kaur

Jagreet Kaur
Dr. JAGREET KAUR
Asst. Professor
Department of Genetics
University of Delhi South Campus
New Delhi-110021