

The Genome Sequencer FLX System

Plant and Agricultural Biotechnology

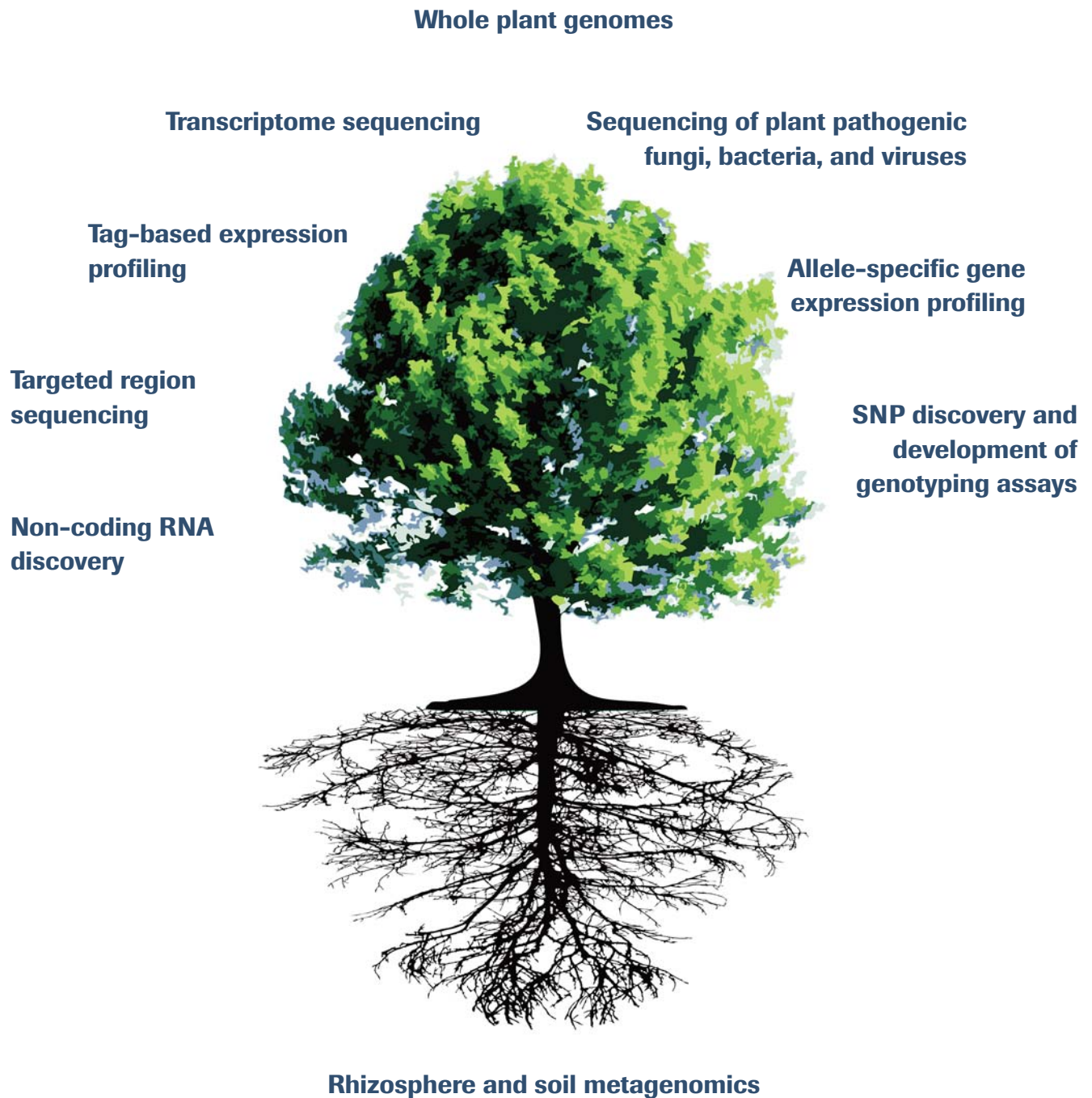


Plant Research and the Genome Sequencer FLX System

Plant genomics in your hands

Traditionally, the complexity and size of plant genomes has limited the ability of plant researchers to obtain comprehensive sequence information. With conventional Sanger sequencing technology, many plant sequencing projects have been unaffordable or technically unfeasible, confining projects to large-scale sequencing centers.

The Genome Sequencer FLX System, with its long, highly accurate reads, puts affordable high throughput sequencing in the hands of the entire plant research community. Learn how 454 Sequencing systems can help you answer your research questions.



Whole plant genome de novo sequencing – Replacing conventional capillary sequencing

- Obtain results equivalent to 6x Sanger oversampling, but significantly faster and at a fraction of the cost.
- Use GS FLX Titanium series reagents for rapid whole genome shotgun or BAC sequencing.
- Combine long single reads (>400 bp) and long-tag paired end reads (>100 bp each) at different insert lengths (3-20 kb) for improved *de novo* assembly.
- Analyze regions that cannot be sequenced with conventional capillary technology due to cloning bias.

Targeted region sequencing – Fast, comprehensive, and straightforward

- Correlate phenotypic traits with genomic variations such as SNPs, insertions, deletions, and structural variations.
- Combine NimbleGen's Sequence Capture arrays and 454 Sequencing to analyze up to eight different 1 MB regions in a single run.
- Sequence PCR fragments to analyze smaller target regions or utilize ultra-deep sequencing to find genetic variants on a population level.

Transcriptome sequencing – A quick and affordable focus on the coding genome

- Assemble whole plant transcriptomes using shotgun sequencing to get the complete picture of alternative splice variants and genomic variations (SNPs, insertions, deletions, structural variations).
- Perform an unbiased transcriptome survey with one run of shotgun reads (1 million, >400 bp reads).
- Obtain gene expression profiles using methods such as SuperSAGE ditag sequencing.
- Only long read lengths ensure comprehensive assembly, gene annotation, and accurate calling of similar alleles.

Rhizosphere and soil metagenomics – An unprecedented picture of diversity with long specific reads

- Obtain a comprehensive view of the diversity and metabolic profile of the rhizosphere with 1 million, >400 bp reads.
- Quantify microbial abundance, discover novel genes, and establish the signature profile of an environment.

Plant pathogenic viruses, fungi, and bacteria – Best in class assembly, lower finishing costs

- Perform rapid whole genome shotgun sequencing of fungi, bacteria, and viruses; sequence as many as 10 bacteria or 96 viruses in a single GS FLX Titanium run.
- Perform superior assemblies with long single and paired end reads (one contig per genome goal), and minimize expensive downstream finishing costs.

A versatile tool – Use the Genome Sequencer FLX System to identify and analyze non-coding RNAs, develop genotyping assays, and much more.

The Genome Sequencer FLX Bioinformatic Tools

1. **De Novo Assembler:** Fast and accurate *de novo* assembly (BAC-to-BAC or whole genome shotgun)
2. **Reference Mapper:** Accurate assembly and comparison with a given reference sequence
3. **Amplicon Variant Analyzer:** Identification and quantification of known and novel DNA variants (*e.g.*, rare alleles) by Ultra Deep Sequencing coverage of targeted regions of interest
4. **454 FusionPrimer design software and custom primers from IDT:**
<http://www.idtdna.com/Catalog/fusionprimers/Page1.aspx>

Selected References

Whole plant genome *de novo* sequencing

Velasco R *et al.* (2007) A high quality draft consensus sequence of the genome of a heterozygous grapevine variety. *PLoS ONE*. **2**(12):e1326.

Plant transcriptome analysis

Bräutigam A *et al.* (2008) Low-coverage massively parallel pyrosequencing of cDNAs enables proteomics in non-model species: comparison of a species-specific database generated by pyrosequencing with databases from related species for proteome analysis of pea chloroplast envelopes. *J Biotechnol*. **136**(1-2):44-53.

Expression profiling

Eveland AL *et al.* (2008) Transcript profiling by 3'-untranslated region sequencing resolves expression of gene families. *Plant Physiol*. **146**(1):32-44.

Rhizosphere and soil metagenomics

Roesch LF *et al.* (2008) Pyrosequencing enumerates and contrasts soil microbial diversity. *ISME J*. **1**(4):283-90.

Plant pathogenic viruses, fungi, and bacteria

Katiyar-Agarwal S *et al.* (2007) A novel class of bacteria-induced small RNAs in *Arabidopsis*. *Genes Dev*. **21**(23):3123-34.

Non-coding RNAs

Pandey SP *et al.* (2008) Herbivory-induced changes in the small-RNA transcriptome and phytohormone signaling in *Nicotiana attenuata*. *PNAS*. **105**(12):4559-64.

More references can be found at www.454.com

“We have extensively tested the use of the Genome Sequencer FLX System in our BAC-by-BAC sequencing approach of the potato and tomato genomes. We were thrilled with the results, which were comparable to or sometimes better than conventional sequencing. We obtained high-quality and entirely unbiased coverage of the BACs quickly and cheaply. We were also very impressed by the power and quality of the software that comes with the platform, which made the assembly of the sequences a straightforward and robust procedure. We are convinced that the Roche 454 GS FLX System will enable us to complete our sequencing effort in these genomes ahead of schedule.”



— Dr. Roeland van Ham,
Cluster Leader Bioinformatics,
Plant Research International

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