A Microarray for Measuring Medicago Expression

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ABSTRACT

Medicago truncatula is an ideal model organism for legumes. A simple diploid genome, rapid growth, and prolific seed production continue to make M truncatula the choice for basic and complex legume studies. Recently, the Medicago truncatula Sequencing Consortium, an international collaboration of Medicago researchers, released Mt4, the latest version of the Medicago truncatula genome. Mt4 represents the most comprehensive assembly of Medicago to date built *de novo* from a whole genome shotgun assembly. In addition to an updated genome assembly, transcript resolution improved with increased transcript placement on the genome. The updated M truncatula genome provided an opportunity to develop a microarray to capture updates made by the consortium. In addition to designing the array against the reference cultivar A17, probes for an alternative cultivar, R108, were also included. Moreover, a closely related legume, Medicago sativa (alfalfa), is represented on the high density array also. The presence of polyploid alfalfa as part of the array design is integral to facilitate research of this extremely important forage crop. This updated microarray will have probes for all 3 organisms which, in addition to surveying the latest transcriptome content, will facilitate cross-species transcriptome analysis as well as assist in *de novo* transcript detection for lesser annotated genomes. Each organism has roughly 60k gene models represented in this microarray in addition to a variety of unmapped and novel sequences that have yet to be mapped to the genome. Consequently the array is an optimum tool to further characterize Medicago truncatula and sativa transcriptomes.

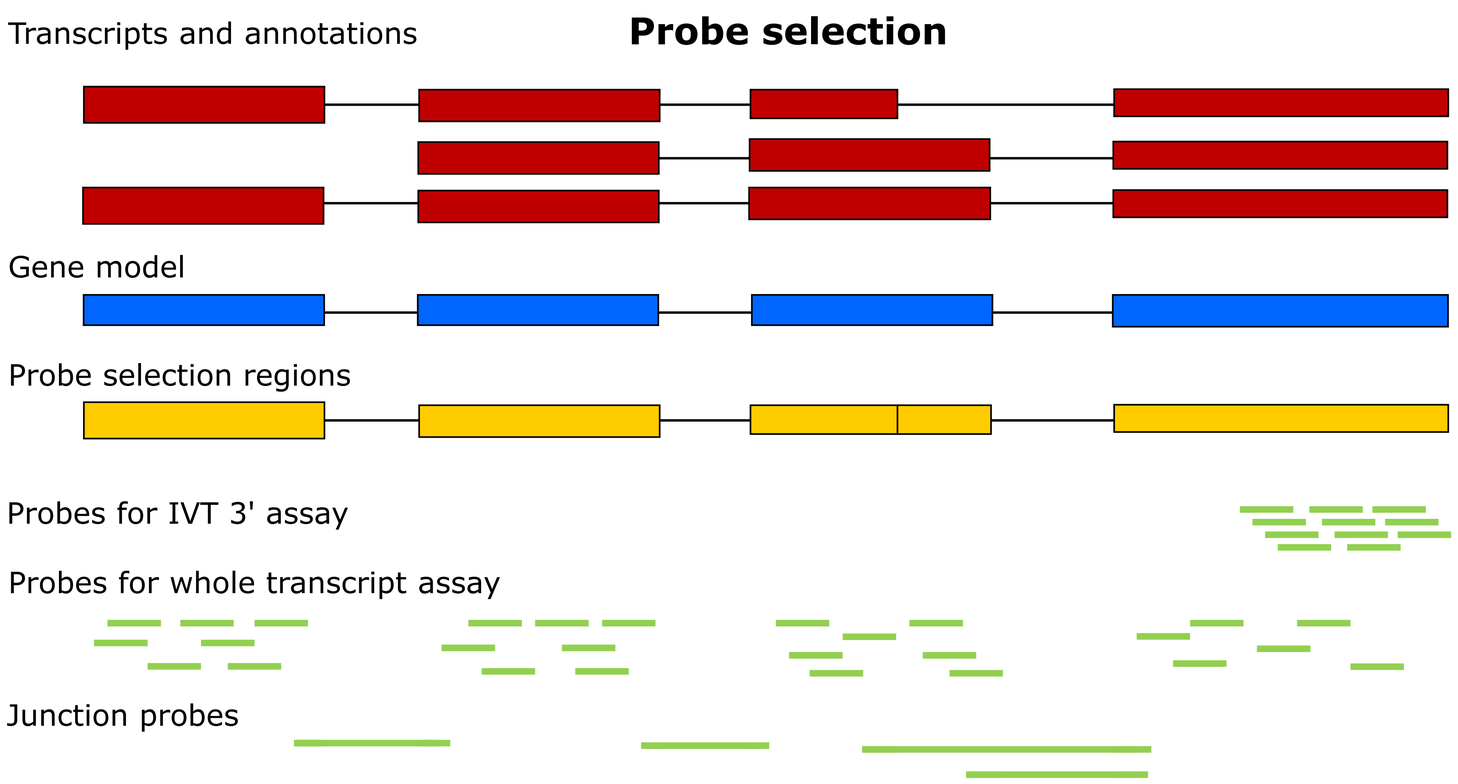

Figure 1: Transcripts and annotations are consolidated into gene models from which probe selection regions are calculated. Probes are then selected from the probe selection regions.

Probe selection

The figure on the left shows the comparison of probes selected for the classical IVT 3'-based amplification assay and a whole-transcript (WT) amplification assay. Whereas the probes for an IVT design are restricted to the 3' end and therefore cannot differentiate between isoforms with the same 3' end, probes for a WT design allows for a more comprehensive view of the transcriptome and the ability to differentiate isoforms. The inclusion of junction probes increases the confidence of alt-splice detection.

Organism	Genome	Annotation source	Genes	Probe sets	Probes / probes per gene	Junction probes
Medicago truncatula A17	Mt4 by MTSC	JCVI	50k genes 69k transcripts 298k exons	65k	3.6M probes 30 median per gene 45 mean per gene	Yes
Medicago truncatula R108	R108 v0.9 by the Medicago Hapmap Project	Medicago Hapmap Project	68k genes 83k transcripts 221k exons	95k	2.3M probes 25 median per gene 24 mean per gene	No
Medicago sativa	Draft by Noble Institute	Noble Institute	54k genes 71k transcripts 172k exons	90k	2.1M probes 25 median per gene 24 mean per gene	No

References

[1] Cold Spring Harb. Protoc.; 2008; doi:10.1101/pdb.emo105

[2] *Fusarium* Comparative Sequencing Project, Broad Institute of Harvard and MIT (http://www.broadinstitute.org/)