

Methylome Analysis during Soybean Seed Development

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WHAT QUESTIONS ARE BEING ADDRESSED BY THE GOLDBERG LAB?

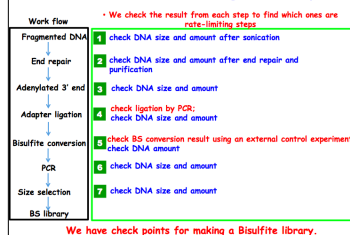
Why Study Soybean?

- Second major US crop
- Total crop value \$32 billion (46% value exported)
- Major food source
- Important biofuel source (biodiesel~20% of US soybean oil production)
- Excellent model plant (transformation, knockdowns, genetics)
- Genome sequenced (2009)
- Available seed gene expression data
- Major funding source (NSF)

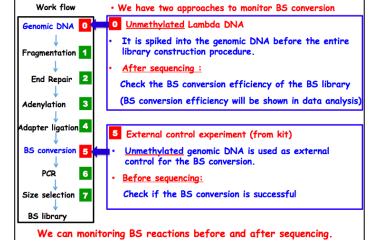
- What is the spectrum of genes that are active in seed compartments of different origin (embryo, endosperm, and seed coat) at different developmental stages?
- What microRNAs are present in specific tissues and compartments during seed development, and what are their targets?
- What are the epigenetic changes that occur in specific compartments and tissues during differentiation and maturation?
- What are the correlations between the methylome, microRNA, and compartment- and tissue-specific gene expression patterns during seed development?
- What are the regulatory processes required to regulate genes during soybean seed development?

HOW DO WE MAKE A BISULFITE LIBRARY?

What is the work flow for Making a BS Library?

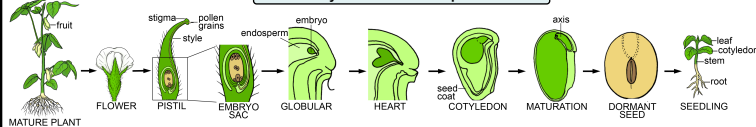


How Do We Monitor Successful BS Conversion?

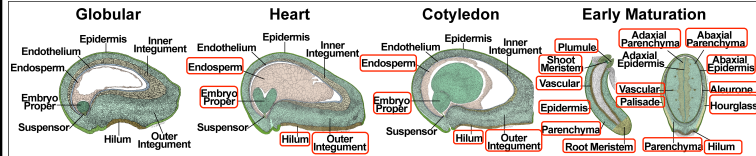


WHAT SEED COMPARTMENTS, REGIONS AND TISSUES ARE BEING STUDIED?

Soybean Seed Development

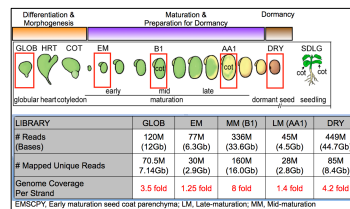


22 Different Soybean Seed Compartments, Regions and Tissues Are Being Studied

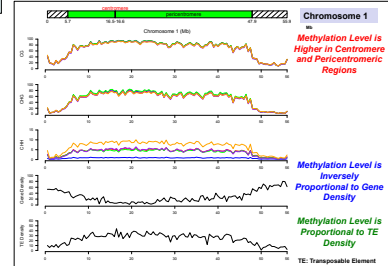


WHAT ARE THE SEQUENCING RESULTS?

The sequence number of BS-DNA-Seq

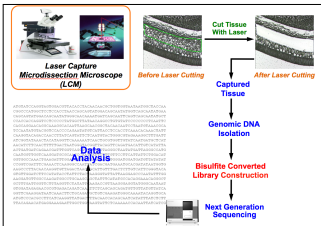


Overall Picture of DNA Methylation Profiles During Seed Development

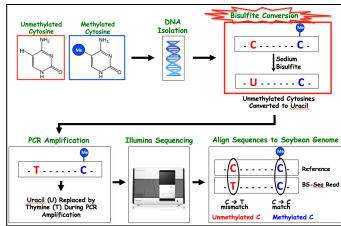


WHAT ARE THE APPROACHES?

The work flow in our study



Using Bisulfite Sequencing (BS-Seq) to study DNA Methylation



WHAT TISSUE/COMPARTMENT/SEED HAVE BEEN EARLY SEQUENCED?

The Tissue/compartment/whole seed that have been sequenced

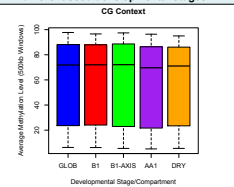
Global Stage	Heart Stage	Cotyledon Stage	Early Maturation Stage	Dry Seed
Whole Seed	Whole Seed	Whole Seed	Whole Seed	Whole Seed
Embryos	Embryo	Root Meristem Region	Parenchyma	Palisade
Endosperm	Endosperm	Shoot Meristem Region	Vascular Tissue	Hilum
Hilum	Hilum	Parenchyma	Epidermis	Parenchyma
Outer Integument	Outer Integument	Epidermis	Parenchyma	Hilum
		Plumule	Vascular Tissue	

DNA isolated from LCM captured tissue

Tissue	Seed Coat Parenchyma	Seed Coat Hilum	Seed Coat Palisade	Seed Coat Hilum	Cotyledon Parenchyma
Total # of LCM captures (20x)	4,509	803	1,181	453	1,025
Total capturing time	133 hr	22 hr	29 hr	17 hr	65 hr
Total DNA isolated from LCM captured tissues	980 mg	25 mg	436 mg	300 mg	120 mg

The most time-consuming work in this study is the LCM work. This table shows the total number of LCM captures, and how much did we work to get these result.

DNA methylation Profiles for the soybean genome in different seed developmental stages



DNA Methylation Profiles for genes

