

Sinorhizobium Meliloti

Rhizobium-Legume Interaction

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Outline

- Overview
- Features of the genome
 - genetic overview
 - pSym megaplasmids
- transport functions
- regulatory proteins
- bacterial adhesion elements
- nodule formation
- nitrogen fixation and metabolism
- symbiosis
- Comparison of genes to other rhizobium
- conclusion

Rhizobia: aerobic gram-negative soil bacteria

- ◆ When plants are deprived of nitrogen, rhizobia are capable of establishing nitrogen-fixing symbiosis.
- ◆ Rhizobia infect roots to form root nodules. The bacteria then undergo nitrogen-fixation within the cell cytoplasm.
- ◆ Rhizobia and plant exchange signals during this process. The rhizobia continue to provide nitrogen to plant in exchange for plant carbon compounds
- ◆ Evolution in legumes has shown the ability to obtain nitrogen from rhizobia while growing in poor soils

Sinorhizobium meliloti:

rhizobia symbiont to alfalfa

- Legume symbiont
- Creates nodules in the roots that convert Dinitrogen to ammonia
- Aerobic but lives in low Oxygen
- Affects stable agriculture and ecosystem function → nitrogen fixation
- How Rhizobia work
 - infect roots
 - induce nodules
 - bacteria and plant exchange signals throughout nodule development
 - create a close metabolic exchange of bacterial fixed nitrogen for plant carbon compounds

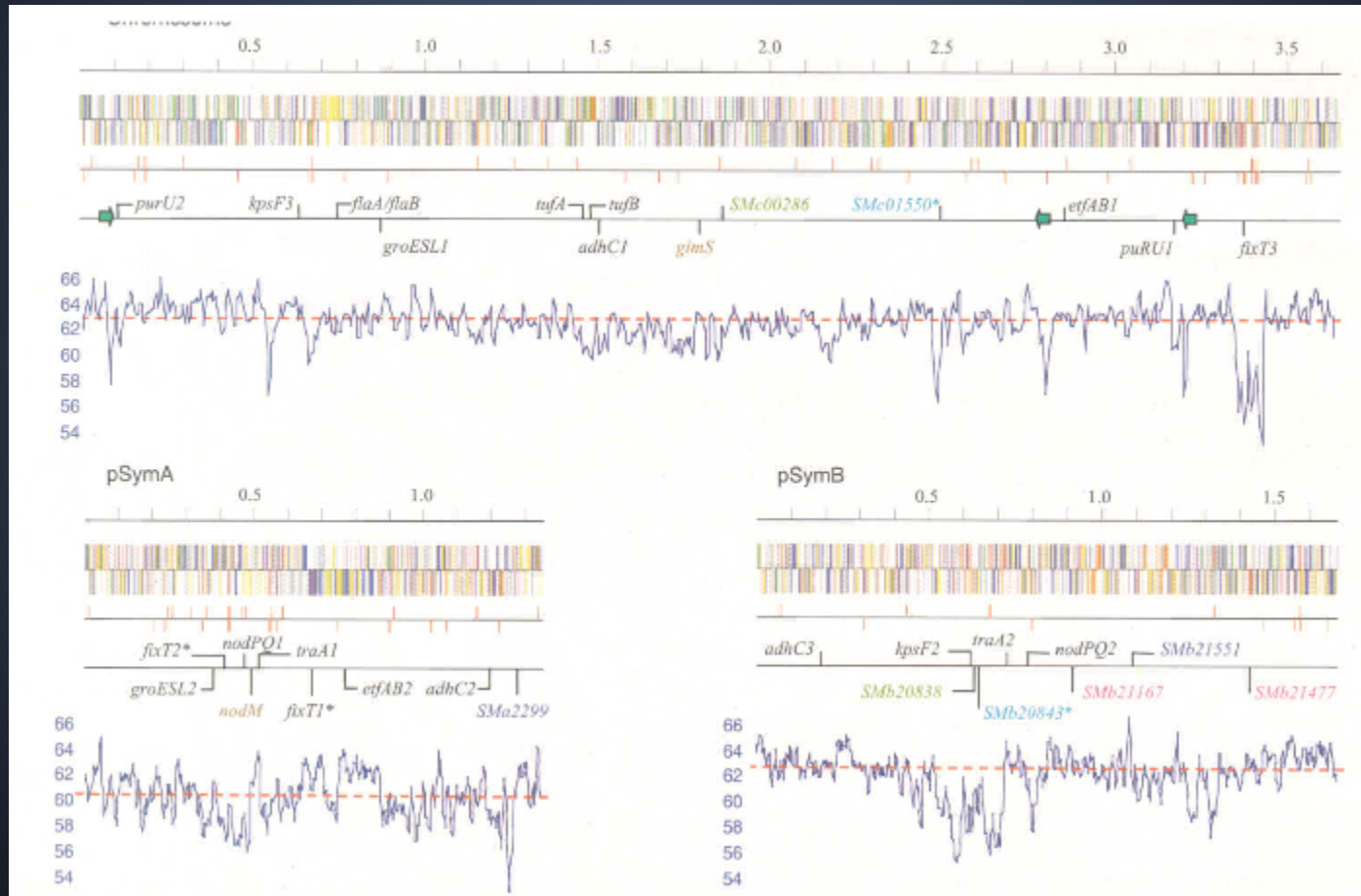
S. meliloti genome is fully sequenced

- 3 Replicons
 - 3.65 mb Chromosome
 - 1.35 mb pSymA megaplasmid
 - 1.68 mb pSymB megaplasmid
- megaplasmids act like larger plasmids
- pSymB provides extended metabolic pathways
- pSymA provides factors included in node formation
- 42% of genes found in 582 paralogous families
 - Genome → no recent size constraints → more adaptive organism
- 2% of genes are Insertion sequences
 - most near pSymA near genes used for symbiosis
 - shows Symbiotic elements prone to change

pSym Megaplasמידs - Genomic information not found in the chromosome that contains 100kb or more of information

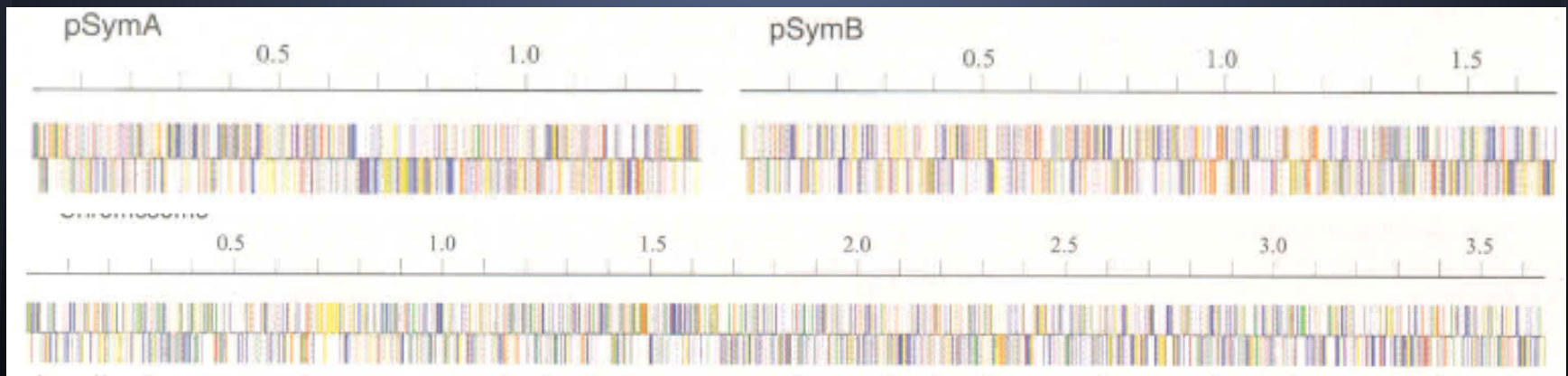
- Found in highly diverse species
- Contain the ABC transporter genes
- pSymA
 - May be transferable
 - No essential genes
 - Plasmid like
- pSymB
 - non-transferable
 - contains essential genes
 - Chromosomal like

Figure 1



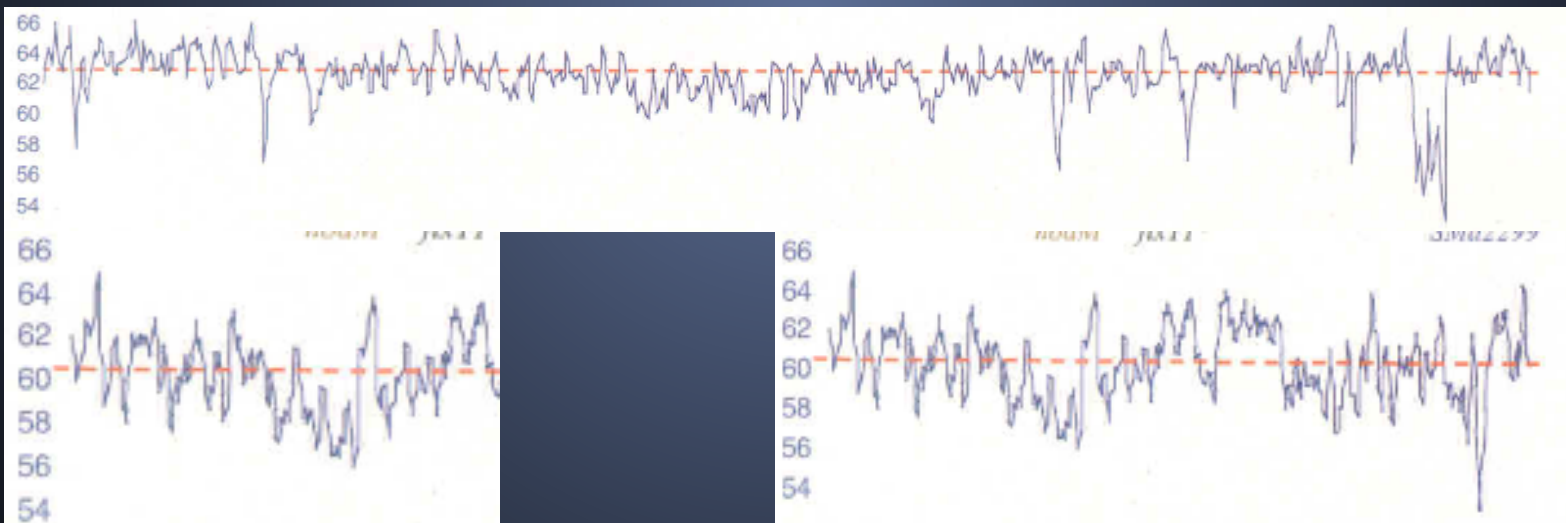
First and Second line, Fig. 1

- Top line derived from coordinates relative to the genome sequence on Wed site.
- Blue = Small molecule metabolism
- Green = Macromolecule metabolism
- Orange = Structural elements
- Yellow = Cell processes
- Red = Elements of external origin
- Pink = regulators that are not classified
- Gray- Unknown/Hypothetical



Third and Fourth line, Fig. 1

Fifth Line, Fig. 1

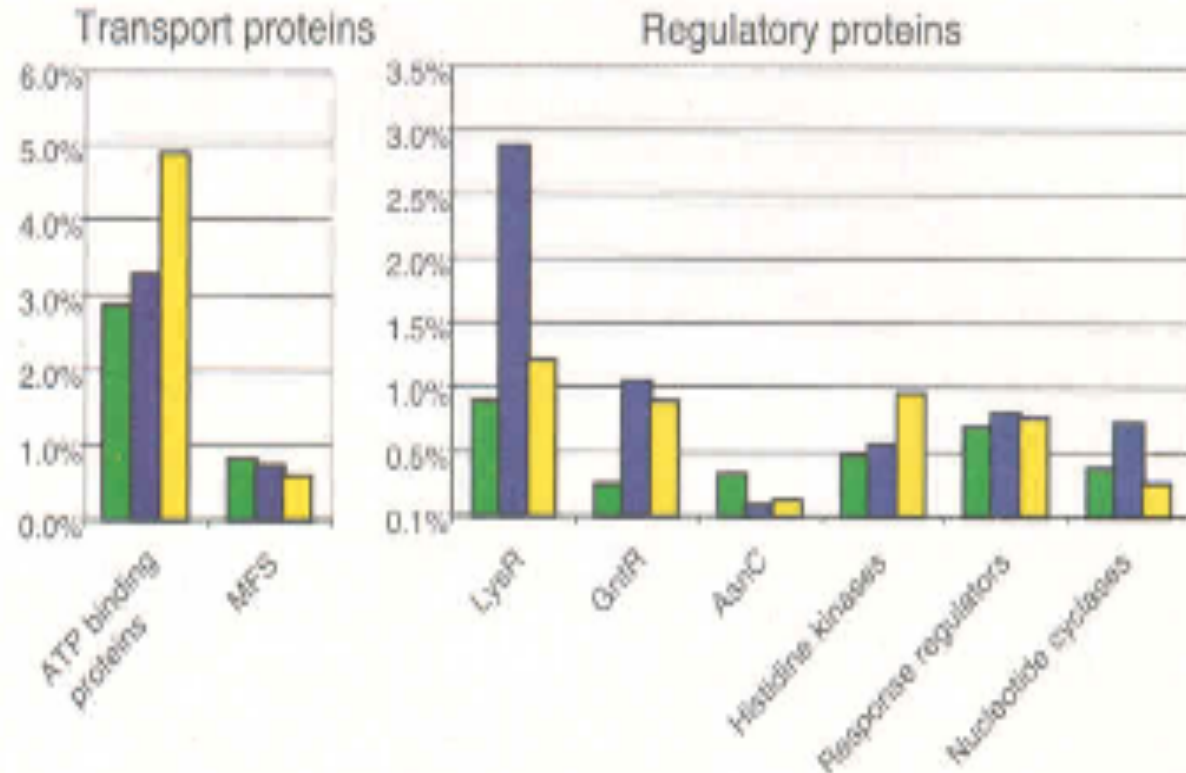


Nitrogen Fixation and Metabolism

- Large relative portion located on pSymA
- No homologous genes
- Factors allowing *S. Meliloti* to live in low oxygen environment
 - Cytochrome c oxidase
 - NADH-ubiquinone gene clusters (pSymA + Chromosome)
- Nitrogen fixation → high energy expenditure
- lives in low Oxygen environments

Figure 2

Fig. 2. Distribution of transport and regulatory proteins by replicon. pSymA is blue, pSymB is yellow, chromosome is green.



Transport functions

- Makes up the largest class of genes (~12%)
- Mainly ABC Transport proteins
- Abundance is high in pSymB (17.4%)
-

Regulatory proteins

- 8.7% of genes
- most are LysR family
- high abundance on pSymA
- GntR regulators → more on megaplastids
- AsnC regulators → more on chromosome
- no Serine-Threonine kinases found
- only 7 regulatory families in *S.meliloti* make up Quorum sensing system

Table 1

Table 1. General features of the *S. meliloti* strain 1021 genome.

Feature	Chromosome	pSymA	pSymB	Genome
Length (bp)	3,654,135	1,354,226	1,683,333	6,691,694
G + C ratio	62.7%	60.4%	62.4%	62.1%
Protein-coding regions	85.8%	83.2%	88.6%	85.9%
Transfer RNAs	51	2	1	54
tmRNA*	1	0	0	1
Ribosomal RNA operons	3	0	0	3
Protein-coding genes	3341	1293	1570	6204
Average length of protein-coding genes (pb)	938	871	950	927
Genes with functional assignment	59%	56.5%	64.4%	59.7%
Orphan genes (% of total protein-coding genes)	5%	11.5%	12.3	8.2%
Regulatory genes (% of total protein-coding genes)	7.2%	10.4%	10.5%	8.7%
Insertional and phage sequences (% of replicon size)	2.2%	3.6%	0.9%	2.2%
RIME elements	185	6	27	218
Palindromes A, B, and C	253	0	5	258

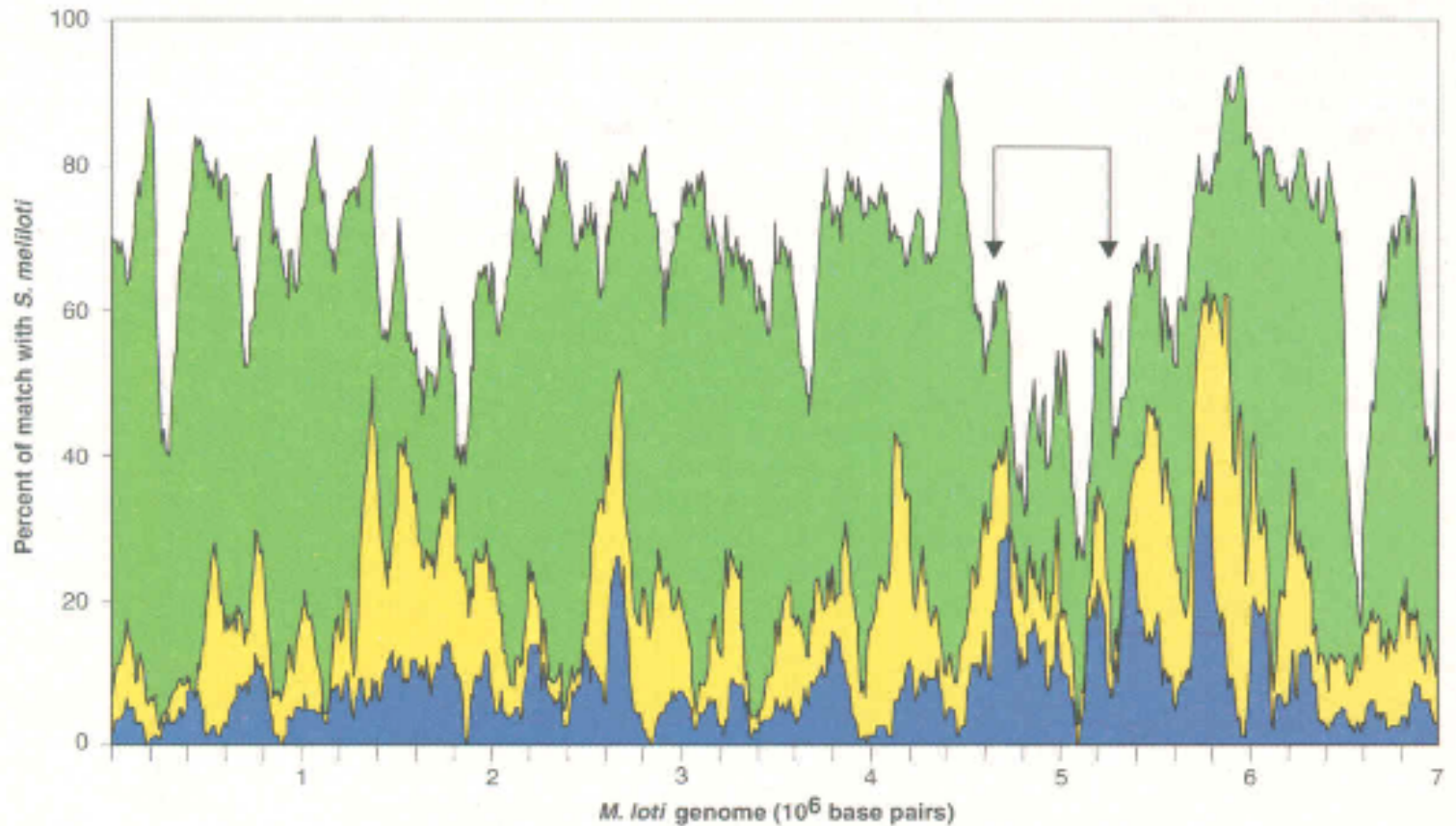
*tmRNA derives its name from the presence of two separate domains, one that functions as a tRNA, and another that serves as an mRNA.

Bacterial Adhesion mechanisms

- Adhesion to root hairs not well understood
- 1 putative adhesin, 2 agglutinin-like genes, 1 ABC transporter
 - all similar to attachment genes of *A.tumefaciens*
- Two new pilus systems thought to be homologous and located on chromosome and pSymA

Figure 3

Fig. 3. Comparison of *M. loti* and *S. meliloti* predicted proteins. The *M. loti* genome from bp 1 to 7 Mb is distributed along the x axis. In any given window along the x axis, the proportion within that window that has a significant match [see (21)] in the *S. meliloti* genome is displayed, and the color indicates the location of the match: blue for pSymA, yellow for pSymB, and green for the chromosome. White represents the proportion that has no global match to *S. meliloti*. Arrows indicate the *M. loti* symbiotic island.



Sinorhizobium meliloti vs. *M. loti*

- 35% of *M. loti* genes had no ortholog in *S. meliloti*
- Megaplasmid genes in *S. meliloti* were spread out in the *M. loti*
- Symbiotic plasmid genes of *M. loti* have no orthologs in *S. meliloti*
 - exceptions: nitrogen-fixing and nodule forming genes

Database

information available [here](#)

Determination of *S. meliloti* 1021 shows that it has a composite structure consisting of three unique replicons of different architecture and service features.

- Complete genome sequence of *Sinorhizobium meliloti* allows the study of...
 - i. rhizobium-legume symbiosis
 - ii. nitrogen-fixation mechanisms of the bacteria.

- This is consistent with the hypothesis that both megaplasmids were inherited from an ancestor with a single chromosome.
- Acquisition of pSymB has increased the metabolic capacity considerably.
- Acquisition of pSymA also led to ability to nodulate, colonize low-oxygen environment of nodules, and metabolize nitrogen compounds under various chemical forms.
- Complete genome sequence allows for opportunities to study the rhizobium-legume symbiosis and the nitrogen-fixation mechanisms of these bacteria.

References

Domínguez-Ferreras, A., Pérez-Arnedo, R., Becker, A., Olivares, J., Soto, M.J., Sanjuán, J. (2006) Transcriptome Profiling Reveals the Importance of Plasmid pSymB for Osmoadaptation of *Sinorhizobium meliloti* *Journal of Bacteriology* 188:7617-7625

Galibert, F., Finan, T.M., Long, S., Puhler, A., et al. (2001) The composite genome of the legume symbiont *Sinorhizobium meliloti* *Science* 293:668-672

[Link to Database](#)