

Sinorhizobium Meliloti

Rhizobium-Legume Interaction

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Outline

- Sinorhizobium Meliloti overview
- Features of the genome
 - genetic overview
 - pSym megaplasmids
- Nitrogen fixation and metabolism
- Transport functions
- Regulatory proteins
- Bacterial adhesion elements
- 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

General Features of *S. Meliloti* 1021 genome

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.

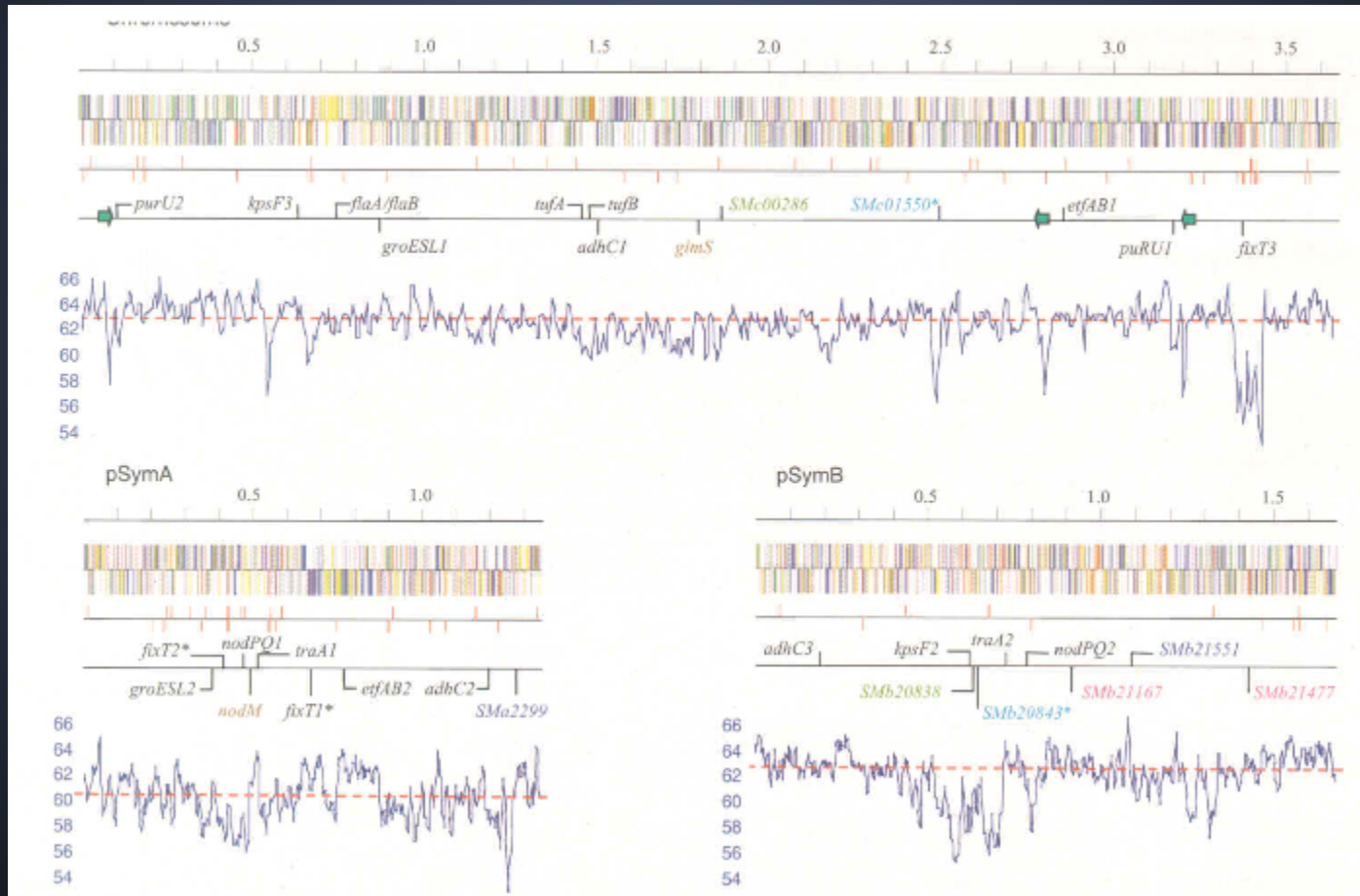
S. meliloti genome is fully sequenced

- 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

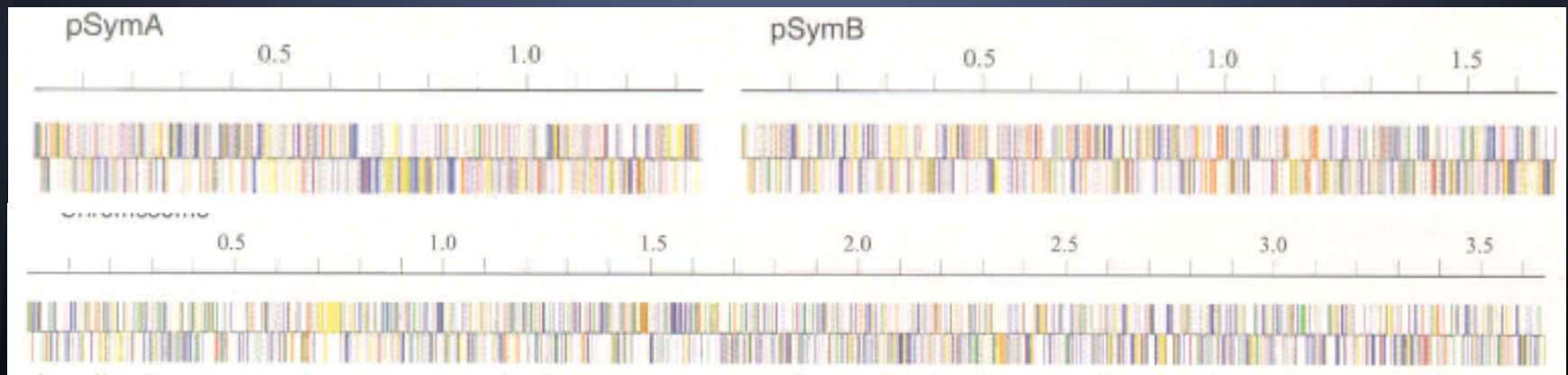
- Found in highly diverse species
- Contain the ABC transporter genes
- pSymA
 - May be transferable
 - No essential genes
 - Plasmid like
- pSymB
 - non-transferable
 - contains essential genes
 - Chromosomal like

Linear Representation of Genome



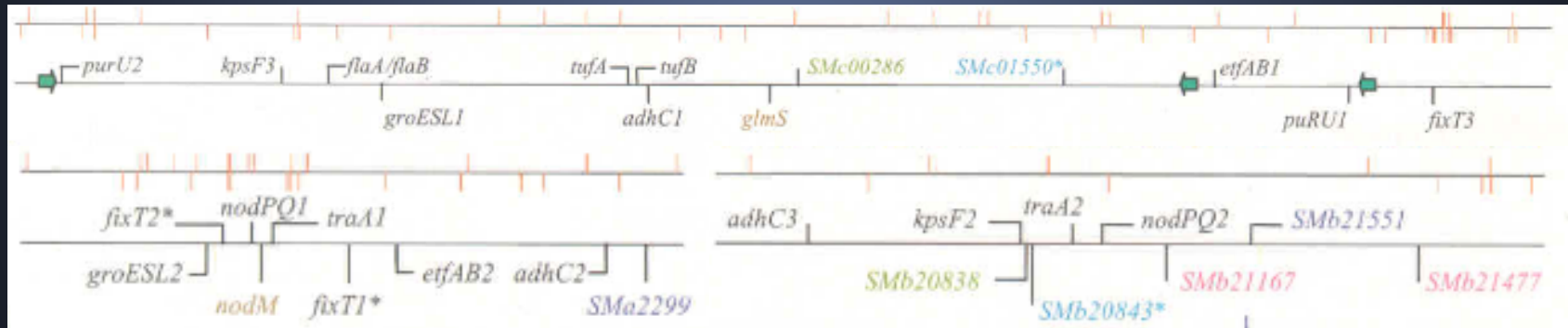
First and Second line, Fig. 1

- Top line derived from coordinates relative to the genome sequence on Web 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



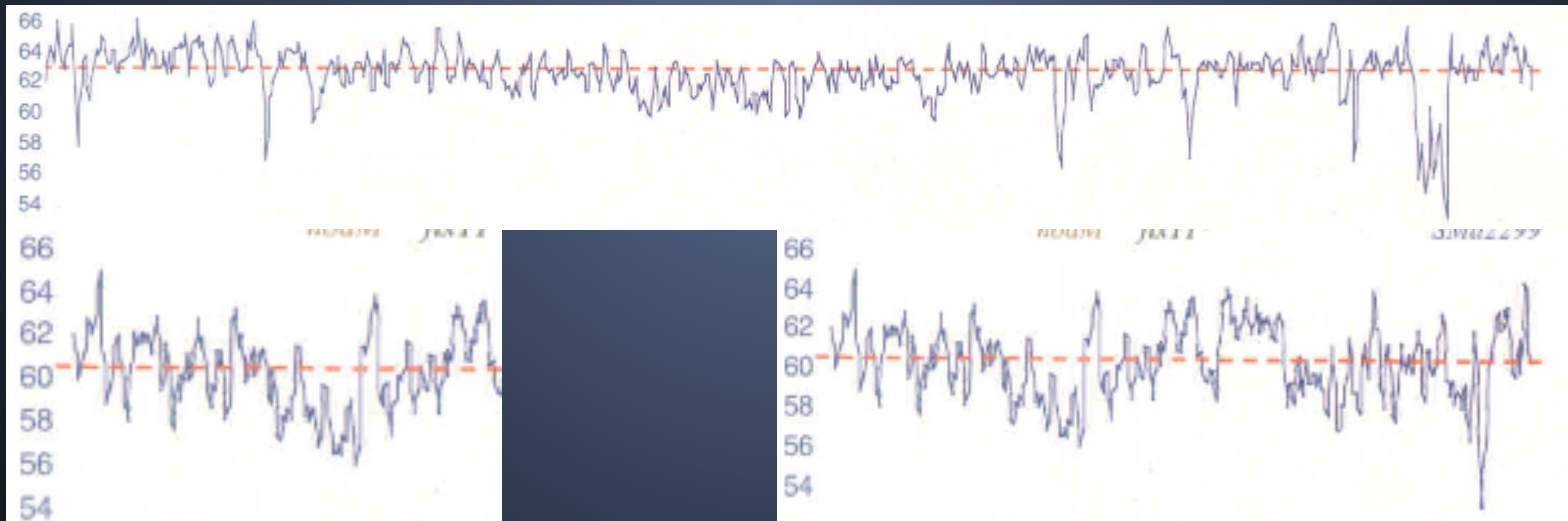
Third and Fourth line, Fig. 1

- Distribution of phage-related sequences
- Recently duplicated genes



Fifth Line, Fig. 1

- GC% variation along the replicons
- Dotted line is the mean

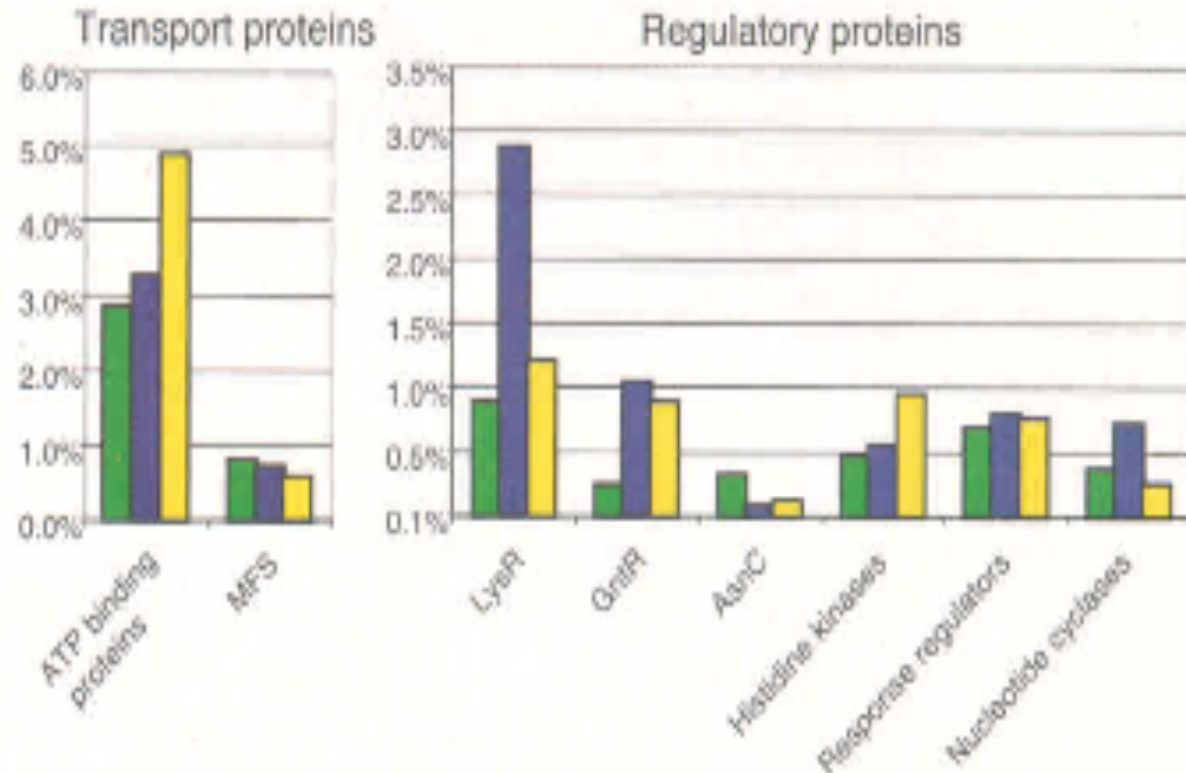


Nitrogen Fixation and Metabolism

- 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

Transportation and Regulatory Protein Frequency

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

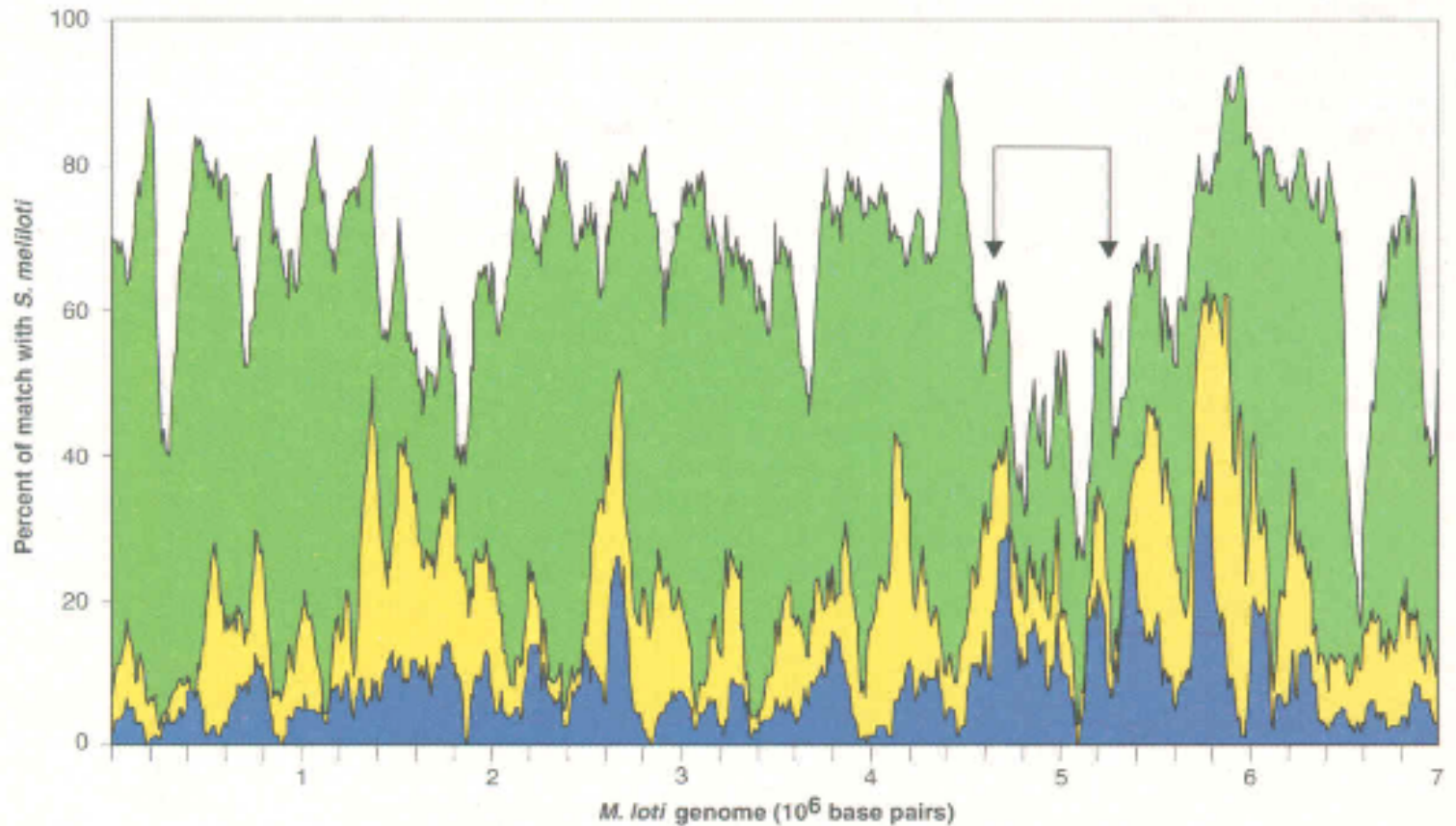
- 8.7% of genes
- most are LysR family
- high abundance on pSymA
- GntR regulators → more on megaplastids
- AsnC regulators → more on chromosome
- no Serine-Threonine kinases found
- only 7 regulatory families in *S.meliloti* make up Quorum sensing system

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

Comparison between *M. loti* and *S. meliloti*

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

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

- 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](#)