Sequence of the Rhizobium leguminosarum biovar phaseoli syrM gene

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Rhizobium forms nitrogen-fixing nodules in symbiosis with its leguminous host plant. In *R. meliloti*, the symbiont of alfalfa (Medicago sativa), the symbiotic regulatory gene syrM is involved in the coordinated expression of genes controlling nodulation (nod) and exopolysaccharide (exo) synthesis (1). SyrM activates the expression of the regulatory nod gene nodD3 (1). In turn, NodD3 stimulates transcription of syrM (2). NodD and SyrM proteins belong to the LysR family of transcriptional regulatory proteins (3). When carried on a multicopy plasmid, nodD3 and syrM allow high levels of nodABC gene expression in the absence of plant inducer (1). In addition syrM stimulates the expression of exoA, exoB and exoF (1). syrM-dependent exo gene expression is mediated by syrA (1). So far, a syrM gene has been identified only in R. meliloti (2, 4).

Rhizobium leguminosarum biovar phaseoli, the symbiont of common beans (Phaseolus vulgaris), possesses three nodD genes (5). While transcription of nodD2 and nodD3 is constitutive, nodD1 is inducible in the presence of bean exudates (6). Here we report the sequence of the R.l. by. phaseoli syrM gene.

Using a R.l. by. viciae nifAB probe (7), we have previously identified a *nifB*-like gene in R.l. bv. phaseoli CNPAF512 (unpublished results). DNA sequence analysis of a 1.8 kb SalI-HindIII fragment, located downstream of this nifB-like gene, indicated the presence of two open reading frames (ORF) coding for polypeptides of 171 and 336 amino acids respectively. The polypeptide encoded by the largest ORF, designated as R. l. bv. phaseoli SyrM, has a calculated molecular mass of 37,800 Da and displays homology to R. meliloti SyrM and to various NodD proteins (Figure 1). R. l. bv. phaseoli and R. meliloti SyrM proteins display 55% identity and 68% similarity (Figure 1). Amino acid similarity between R. l. bv. phaseoli SyrM and different NodD proteins is between 33% (Azorhizobium caulinodans NodD) and 41% (R. l. bv. phaseoli NodD3). The conserved amino acids between NodD and SyrM proteins are found predominantly in their amino terminus (underlined in Figure 1). This region contains a helix-turn-helix motif, characteristic of DNA-binding proteins (3).

The ORF encoding the 171 amino acids protein (19,187 Da) is located upstream from syrM and reads in the same direction. This ORF displays 81% similarity to the product of a partially sequenced unidentified ORF downstream of the Bradyrhizobium japonicum nodIJ genes (8). The function of this protein is unknown. No significant homology to any protein sequence in the Swiss-Prot data base was detected.

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 - IN FMKRNVEKEVTDQLAYRRKMLQDWRGERLTGHQI---NLASIDLNLLV MQQPTWKRPHRAKFAGVSDAAQQRQMPNLASIDLNLLV Rp Rm 47 38
 - ALEALLEYRNVTHAGQHIGRSQPAMSRALGRLRGLFNDDLLVRSSTGLIP DLEALLQYRHITQAAQHVGRSQPAMSRALSRLRGMLKDDLLVAGSRGLVL Rp Rm 97 88
 - TPQGEHLAQRLPSALRTIRENVTSRSVISKENGRGATLAIPDHQALAVLP TPLAECLTQHLPSVLDAIRQH-HNLSLAPAQRRMKVTHANPDHQAVVLLP ±** ±.**** ± .*** * . .** Rp Rm 147
 - RLLPWLRERAPHLDTLACLPFDRAVRGLEQGDIDLAVGHIDVQLPGYFRR HLLPRLHERAPHLDIVTDPLLGGALGLLEQGEIDVVVQQMGAAPLGYLRR 197 187
 - SLYTDRFACLLRHGHPALAQEWTIDNFATLRHAAISTDSPOHFGPIYDHL RLYADSFTCVLRHNHPALAQEWTIEAFAALRHVAIASEPDELFGQIYDRL Rp Rm 247
 - PNLRADRS-PILFSSVLTAAVVASATDLVLLVPRRVATQVSAMLPLRVVD TKLGLQRGDPMVVSTVLTAAVLIAATDSVLVVPSRVATRVAAMLSLAVIP Rp Rm 296 287

Figure 1. Amino acid sequence alignment of the SyrM proteins of R.l. bv. phaseoli (Rp) and R. meliloti (Rm, ref. 3). Identical amino acid residues are marked by asterisks and positions with conservative substitutions (S-T-A; L-V-I-M; K-R; D-E; Q-N; F-Y-W) are indicated with dots. Identical amino acids conserved in the NodD proteins of R. meliloti (NodD1, NodD2, NodD3), R. l. bv. phaseoli (NodD1, NodD2, NodD3), R.l. bv. viciae, R.l. bv. trifolii, B. japonicum (NodD1, NodD2), A. caulinodans and B. japonicum sp. ANU289 and in both SyrM proteins are underlined.

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PPLEPAPYKVMLIWHERCHHDPQHKWLRKEVAAALETGAD PPVELRPYEVALIWHERCHRDPEHRMLRGEIAAAAST-AG Rp Rm 336 326