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The alternative sigma factor RpoH2 is required for salt tolerance in *Sinorhizobium* sp. strain BL3

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Abstract

The objectives of this investigation were to isolate the *rpoH2* gene encoding an alternative sigma factor from *Sinorhizobium* sp. BL3 and to determine its role in exopolysaccharide (EPS) synthesis, salt tolerance and symbiosis with *Phaseolus lathyroides*. The *rpoH2* gene of *Rhizobium* sp. strain TAL1145 is known to be required for EPS synthesis and effective nodulation of *Leucaena leucocephala*. Three overlapping cosmid clones containing the *rpoH2* gene of BL3 were isolated by complementing an *rpoH2* mutant of TAL1145 for EPS production. From one of these cosmids, *rpoH2* of BL3 was identified within a 3.0-kb fragment by subcloning and sequencing. The cloned *rpoH2* gene of BL3 restored both EPS production and nodulation defects of the TAL1145 *rpoH2* mutants. Three *rpoH2* mutants of BL3 were constructed by transposon-insertion mutagenesis. These mutants of BL3 grew normally in complete or minimal medium and were not defective in EPS synthesis, nodulation and nitrogen fixation, but they failed to grow in salt stress conditions. The mutants complemented with cloned *rpoH2* from either BL3 or TAL1145 showed higher levels of salt tolerance than BL3. The expression of *rpoH2* in BL3 started increasing during the exponential phase and reached the highest level in the mid-stationary phase. These results indicate that RpoH2 is required for salt tolerance in *Sinorhizobium* sp. BL3, and it may have additional roles during the stationary phase.

Keywords: Alternative sigma factor; *rpoH2*; *Sinorhizobium*; Exopolysaccharide; Salt tolerance