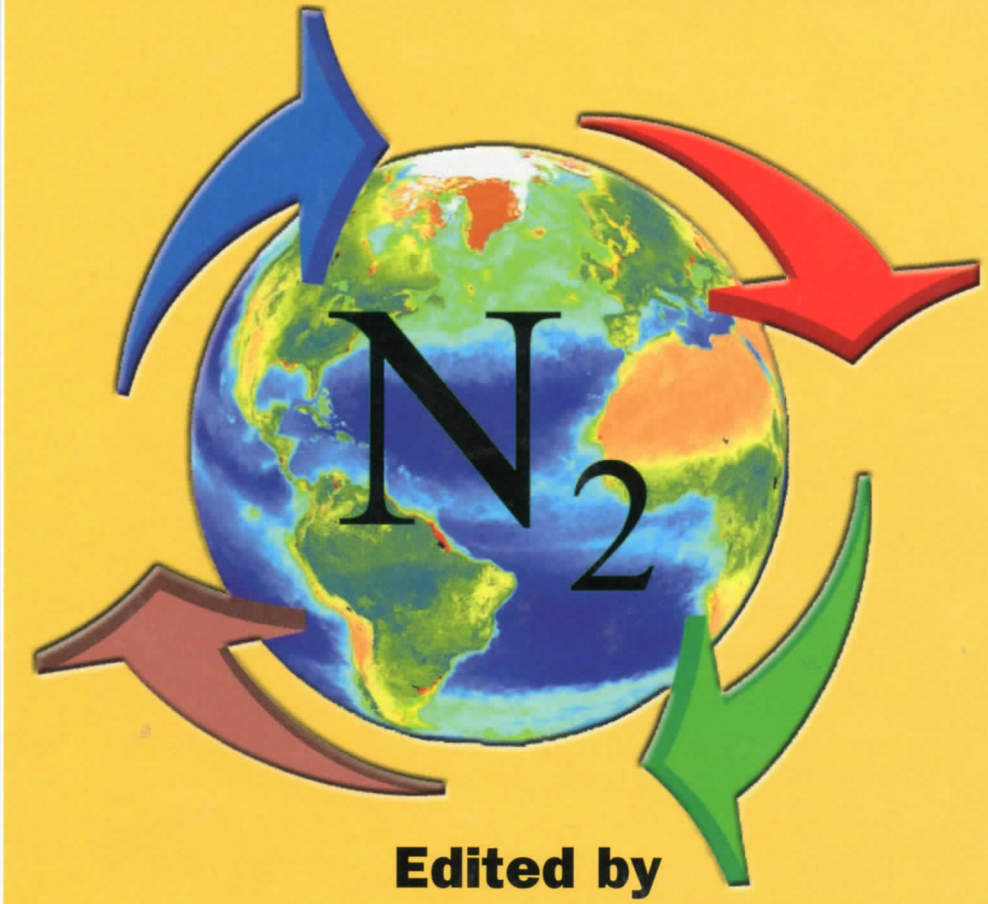
NITROGEN FIXATION

Global Perspectives



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CABI *Publishing*

NtcA ACTIVATES THE *nblA* GENE OF THE CYANOBACTERIUM *SYNECHOCOCCUS* sp. PCC 7942

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1. Introduction

NtcA is a global transcriptional regulator for nitrogen control present in every cyanobacteria tested (Herrero *et al.* 2001) and most of its targets are genes involved on nitrogen assimilation. Non-diazotrophic cyanobacteria degrade their light-harvesting antennae, the phycobilisomes, when exposed to a variety of stress conditions, including nitrogen starvation. This phenomenon, termed chlorosis or bleaching has been shown to be dependent of the response regulator NblR in *Synechococcus* sp. PCC 7942 (Schwarz, Grossman 1998). The expression of *nblA*, a key gene in degradation of phycobilisomes is induced during nitrogen starvation, although previous work failed to show the direct involvement of NtcA on *nblA* expression (Bhaya *et al.* 2000).

2. Results and Discussion

To throw some light on transcriptional regulation of *nblA* in response to nutrient starvation, we performed Northern and primer extension analyses in wild type, NblR- and NtcA- strains from *Synechococcus* sp. PCC 7942 and gel retardation assays with purified NblR and NtcA. Results show that (i) after nitrogen depletion, *nblA* transcript accumulation was impaired in NtcA- cells; (ii) *nblA* expression was still responsive to nitrogen in the absence of NblR; (iii) purified NtcA and NblR bind to the nblA promoter region; and (iv) the NtcA-mediated increase in *nblA* transcripts is not via NblR, since *nblR* transcript levels were not affected by *ntcA* inactivation or by the nutritional conditions tested. As a whole, these results demonstrate that NtcA directly activates nblA transcription under nitrogen starvation conditions.

Primer extension and sequence analysis also indicate that *nblA* transcription can initiate at several promoters. The most active one, P*nblA*-2, is indeed directly regulated by NtcA under nitrogen starvation conditions and constitutes a novel type of NtcA activated promoter, with putative NtcA binding sites centered at –68 and –100.5 from the transcription star point.

Our results indicate that *nblA* promoter region is a complex regulatory region and that both NtcA and NblR greatly stimulate transcription from P*nblA*-2 in response to nitrogen starvation and, in the case of NblR, also in response to other signals. Lack of viability of the NblR$^-$-NtcA$^-$ double mutant and other observations made in the course of this work anticipate additional regulatory connections between the NblR and NtcA regulons.

3. References

Bhaya *et al.* (2000) The Ecology of Cyanobacteria, pp. 397-442, Kluwer Academic Publishers, Dordrecht, The Netherlands

Herrero A *et al.* (2001) J. Bacteriol. 183, 411-425

Schwarz R, Grossman AR (1998) Proc. Natl. Acad. Sci. USA 95, 11008-11013