# DETECTION OF *nifH* GENE IN INDOLE ACETIC ACID PRODUCING BACTERIA

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**Introduction.** Production of indole acetic acid (IAA) and nitrogen fixation by rhizobacteria aid to promote plant growth. The genus *Azospirillum* is widely known by its capacity for nitrogen fixation and production of substances such as auxins, gibberellins and cytokinins that enhance plant growth (1). Furthermore, *Bacillus* strains have been reported to promote plant growth by the same mechanism described above (2). This study aims to determine the potential of *Azospirillum* spp and *Bacillus* spp. in the production of IAA and nitrogen fixation by the presence of nitrogenase gene (*nif*H).

**Methods.** We used three strains of *Azospirillum* spp. and five strains of *Bacillus* spp. isolated in the Yucatan Peninsula, Mexico. The production of IAA was performed by colorimetric reaction Salkowski. For amplification of the gene coding for the nitrogenase (*nif*H), extraction was conducted using DNA Wizard ® Genomic DNA Purification Kit (Promega). NifH gene amplification by PCR was performed according to Shime-Hattori *et al*. (2011) (3).

**Results.** All the strains produced IAA, with values that ranged from 3.34 to 10.96 $\mu gmL^{-1}$. *Azospirillum* spp. showed higher values for the production of IAA (Table 1), while *Bacillus* spp, showed lower capacity to produce IAA. In literature, however, *Bacillus* spp, has been shown to be good producers of IAA, with values from 0.87 to 15.16 $\mu gmL^{-1}$ (2). The variation is likely due to the genetic diversity of species of the genus *Bacillus*.
As for the presence of the nitrogenase gene, all strains of *Azospirillum* spp. showed presence of *nif*H gene by PCR amplification. In contrast, only the strain RF8 of *Bacillus* spp. showed the presence of such gene. *Azospirillum* species are well known nitrogen-fixing bacteria, while in the case of *Bacillus* spp. few studies have documented the presence of *nif*H gene, which in turn suggests that these strains might have the capacity for nitrogen fixation (Ding et al., 2005) (4).
Amplification of *nif*H gene in RF8 and other strains of *Bacillus* spp. will be confirmed using specific primers for the gene *nif*H in *Bacillus* spp.

**Table 1.** Production of indole acetic acid and gene amplification *nif*H

| Isolate | Bacterium | IAA ($\mu gmL^{-1}$) | *nifH* |
|---|---|---|---|
| P10 | *Azospirillum* sp. | 10.94±2.1 | + |
| P13 | *Azospirillum* sp. | 8.07±0.24 | + |
| P22 | *Azospirillum* sp. | 8.94±0.53 | + |
| MT2 | *Bacillus subtilis* | 4.12±0.3 | - |
| CK36 | *Bacillus subtilis* | 6.08±0.89 | - |
| RF5 | *Bacillus* sp. | 5.92±0.42 | - |
| RF15 | *Bacillus* sp. | 5.28±0.27 | - |
| RF8 | *Bacillus* sp. | 3.34±0.03 | + |

IAA. Production of indole acetic acid, + *nif*H gene amplification.

**Conclusions.** All bacterial strains produced IAA. The amplification of *nif*H gene was observed in all *Azospirillum* spp. strains and only in strain RF8 of *Bacillus* sp.

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**References.**

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