

## Effects of High Temperature on Survival and Relative Performance of Bean Nodulating *Rhizobium* strains

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High soil temperatures in tropical areas is a major problem for biological nitrogen fixation in legumes crops. Common bean (*Phaseolus vulgaris* L.) is generally considered to be very sensitive (Piha and Munns, 1987) although it is an important crop in tropical countries. High temperatures may affect the survival and establishment (Karanja and Wood, 1988) and change symbiotic properties in rhizobia (Toro and Olivares, 1986). Bean nodulating *Rhizobium* strains are considered to be particularly sensitive, because in these strains high temperature can cause plasmid deletions (Trevors, 1986) and genomic rearrangements (Soberon-Chavés *et al.*, 1988), changing symbiotic properties. Therefore, the potential benefit of this legume - *Rhizobium* association may not be fully exploited when high soil temperatures become limiting.

This study was aimed to evaluate the effects of heat on bacterial survival and the rate of symbiotic nitrogen fixation in efficient *Rhizobium tropici* and *R. leguminosarum* bv. *phaseoli* strains, isolated from Cerrado soils. The native strains (with different levels of heat tolerance) and commercial strains for each one of the species were inoculated in Cerrado soil and incubated in a shaker at 45°C for 4 hours. Viable cells were counted before and after temperature stress and then inoculated, in the same number, in Leonard jars with aseptically cultivated beans (cv. aporé).

The survival capacity, nodulation and nitrogen fixation of the studied strains, were affected differently by temperature. The survival capacity, determined by the number of viable cells after stress temperature, showed that this characteristic was typical of each strain and is independent of its thermotolerance. For example, tolerant strains, Br322 and SLP1.3 (growing T.max. 39°C) kept the same cell number after exposition to high temperature while FJ2.2 (T.max also 39°C) decreased drastically to 10<sup>3</sup> cell/ml (Table 1). In relation to symbiotic properties, two strains (SLB3.12 and SLP4.9) lost their capacity to nodulate and two strains (FJ2.21 - *R. tropici* and SLA1.5 - *R. leguminosarum* bv. *phaseoli*) the nitrogen fixation levels decreased significantly after thermal shock (Table 1). The last two strains showed high thermotolerance (T.max. 39°C) but low survival capacity compared to all other tested strains. This can be a limiting factor for the utilisation of these strains in tropical regions. Besides affecting the cell survival capacity, high temperature can change the *Rhizobium* genome (Soberon Chaves *et al.*, 1986). The effect of high temperature on the genome modification is being analysed.

**Table 1 - Survival capacities of *R. tropici* and *R. leguminosarum* bv. *phaseoli* strains, with different temperature tolerance, as well as their capacities of nitrogen fixation in common bean after thermal shock at 45°C for 4 hours (mean of 3 replicates).**

| <i>Rhizobium</i><br>strain                     | Growing<br>T.O.max. °C | Viable cell number/ml |                   | Plant dry weigh<br>(g/plant) |        | Total plant N<br>(mg/plant) |      |
|--|------------------------|-----------------------|-------------------|------------------------------|--------|-----------------------------|------|
|  |                        | A                     | B                 | C                            | D      | E                           | F    |
| <i>R. tropici</i>                              |                        |                       |                   |                              |        |                             |      |
| BR322  | 39                     | 8x10 <sup>8</sup>     | 8x10 <sup>8</sup> | 0.527                        | 0.498  | 220                         | 198  |
| FJ2.2  | 39                     | 4x10 <sup>8</sup>     | 1x10 <sup>5</sup> | 0.450                        | 0.446  | 229                         | 270  |
| SLBR3.12                                       | 39                     | 11x10 <sup>8</sup>    | 4x10 <sup>6</sup> | 0.418                        |        | no nodulation               |      |
| SLP4.9   | 38                     | 15x10 <sup>8</sup>    | 3x10 <sup>6</sup> | 0.520                        |        | no nodulation               |      |
| FJ2.21   | 36                     | 4x10 <sup>8</sup>     | 6x10 <sup>4</sup> | 0.739*                       | 0.399* | 286*                        | 143* |
| SLA2.2   | 36                     | 4x10 <sup>8</sup>     | 1x10 <sup>7</sup> | 0.375                        | 0.422  | 239                         | 270  |
| SLA3.2   | 36                     | 3x10 <sup>9</sup>     | 1x10 <sup>7</sup> | 0.522                        | 0.511  | 270                         | 254  |
| <i>R. leguminosarum</i><br>bv. <i>phaseoli</i> |                        |                       |                   |                              |        |                             |      |
| SLP2.10  | 39                     | 2x10 <sup>9</sup>     | 2x10 <sup>8</sup> | 0.382                        | 0.387  | 249                         | 278  |
| SLP5.8   | 39                     | 8x10 <sup>8</sup>     | 6x10 <sup>6</sup> | 0.394                        | 0.393  | 273                         | 263  |
| SLA1.5   | 39                     | 2x10 <sup>8</sup>     | 2x10 <sup>5</sup> | 0.393*                       | 0.283* | 314*                        | 201* |
| SLP1.3   | 39                     | 2x10 <sup>8</sup>     | 2x10 <sup>8</sup> | 0.485                        | 0.465  | 218                         | 204  |
| SLP24.1  | 38                     | 9x10 <sup>8</sup>     | 1x10 <sup>7</sup> | 0.449                        | 0.504  | 253                         | 265  |
| BR10.026                                       | 38                     | 4x10 <sup>8</sup>     | 3x10 <sup>6</sup> | 0.330                        | 0.304  | 265                         | 253  |
| BR10.028                                       | 36                     | 7x10 <sup>8</sup>     | 2x10 <sup>5</sup> | 0.375                        | 0.373  | 292                         | 245  |
| SLP4.4   | 36                     | 9x10 <sup>8</sup>     | 6x10 <sup>6</sup> | 0.361                        | 0.356  | 293                         | 288  |

A) cell number before thermal shock;

B) cell number after thermal shock;

C) plant dry weight inoculated with control strain;

D) plant dry weight inoculated with strain thermal shock submitted;

E) N total plant inoculated with control strain;

F) Total plant N inoculated with strain thermal shock submitted.

\* Indicated significant differences (Duncan, 5% of probability) in plant dry weight and total plant N.

## References

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