

Morphological-anatomical changes in somatic wheat calli in vitro under the effect of bacterial lipopolysaccharide.

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We examined the effect of the lipopolysaccharide (LPS) of the associative, plant-growth-promoting bacterium *Azospirillum brasilense* Sp245 on the morphogenetic parameters of somatic tissues of spring wheat *in vitro*. For this purpose, we used a genetic model including two near-isogenic lines of wheat cultivar Saratovskaya 29 that differed in the *RhtB1c* gene and had contrasting embryogenic capacities. The work was conducted in accordance with the classic scenario of cellular transformations in a culture *in vitro* of immature (14-day-old) wheat embryos by way of indirect somatic embryogenesis. The control was a Linsmaier–Skoog medium containing 2 mg/L of 2,4-dichlorophenoxyacetic acid (2,4-D). In the experimental treatments, the standard medium, after being autoclaved, received 10 µg/mL of LPS isolated from the bacterial outer membrane. The resultant calli and regenerated plants were analyzed by morphometric and morphological-anatomical analysis. On day 30 of cultivation, analysis of the calli showed that in all experimental treatments, there was active callusing of somatic cells of the immature embryos. The callusing effectiveness was high (close to 100%) and did not depend on the genotype or the presence of LPS in the nutrient medium.

Yet in both lines, the formation of morphogenic calli in the control was less than 20% of the number of explants used. On the whole, the addition of LPS to the medium increased the activity of morphogenetic processes. Particularly noticeable was an increase in the yield of calli with meristematic activity loci in the tall line in the presence of 10 µg/mL of LPS in the nutrient medium. Consequently, the morphogenic potential of the calli of the lines under study leveled off. The yield of regenerated plants was influenced profoundly by the explant genotype. The regeneration capacity of the dwarf line was greater than that of the sister line, a fact evident in the control plants. In the experimental treatments, the yield of regenerated plants increased in the tall line under the effect of LPS. Morphological-anatomical analysis confirmed the positive effect of the *RhtB1c* gene at all stages of tissue culturing *in vitro*. LPS was found to promote the secondary differentiation of callus cells and the formation of morphogenesis loci as early as day 7 of embryo culturing, regardless of the genotype. Meristematic activity was observed mostly in the nodal plate, vascular bundles of the scutellum, and coleoptile of the embryos.

On the basis of this data, we established that the LPS of the associative bacterium *A. brasilense* Sp245 promotes the secondary differentiation and the regeneration capacity of wheat callus cells, thereby improving the effectiveness of culturing of genotypes with low embryogenic potential. A nutrient-medium concentration of LPS of 10 µg/mL has the greatest physiological activity toward callus cells. These results make possible a deeper understanding of the mechanisms of morphogenesis in tissue culture and can be used to increase the embryogenic potential of plant objects, specifically wheat, in an *in vitro* culture.