

**OPTIMIZATION OF THE EXPLANT STERILIZATION PROTOCOL FOR
ESTABLISHING ASEPTIC AND VIABLE IN VITRO CULTURES
OF *Salvia korolkowii* L.**

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The genus *Salvia* L. (family Lamiaceae Martinov) comprises more than 1,000 species distributed across temperate, subtropical, and tropical regions. Its representatives are valuable medicinal and aromatic plants rich in bioactive secondary metabolites, including diterpenoids, tanshinones, triterpenes, phenolic acids, and flavonoids. Among Asian species, *Salvia korolkowii* Regel & Schmalh. is of particular scientific interest as a rare, narrow-range endemic of the Western Tian-Shan. Its natural populations have a restricted distribution, low population numbers, and high vulnerability to anthropogenic and climatic factors (Sennikov and Turdiboyev). Therefore, establishing *S. korolkowii* in vitro is an important biotechnological approach for ex situ conservation and the sustainable production of bioactive compounds.

The aim of the study was to optimize sterilization conditions and establish an effective protocol for introducing *Salvia korolkowii* L. into in vitro culture for subsequent callus induction.

Leaf and stem explants of *S. korolkowii* were used as the initial plant material. Surface sterilization was performed using a 15% sodium hypochlorite (NaOCl) solution supplemented with Tween-80. To determine the optimal exposure duration, three treatment periods were tested: 10, 12, and 15 min. Each treatment was performed in four flasks containing five explants each. Following sterilization, the explants were rinsed repeatedly with sterile water and transferred onto nutrient medium for subsequent establishment in in vitro culture. The selection of 15% NaOCl supplemented with Tween-80 was based on a published protocol for *S. officinalis*, in which sterilization was performed for 15 min.

Sterilization efficiency was evaluated based on the proportion of sterile explants. After 10 min of treatment, 65% of the explants remained sterile. Increasing the exposure duration to 12 min increased this proportion to 84%. However, further extension of the treatment to 15 min reduced the value to 71%. Thus, among the tested sterilization regimes, the 12-min treatment was the most effective. The results obtained demonstrate that increasing the sterilization duration does not result in a linear increase in sterilization efficiency; after reaching the optimal exposure time, further prolongation of treatment may be associated with reduced viability of the plant material.

Therefore, for leaf and stem explants of *S. korolkowii*, the 12-min treatment with 15% NaOCl supplemented with Tween-80 was the most effective among the tested regimes, resulting in 84% sterile explants. This treatment can be used as an optimized initial step in the development of a protocol for establishing *S. korolkowii* callus cultures in vitro.

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