

**Differences in development *in vitro* of *Cannabis sativa* L.  
(variety K290) under the influence of various mineral  
components of nutrients, vitamins  
and plant growth regulators**

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## **Abstract:**

*In vitro* plant cultures of *Cannabis sativa* may have a valuable contribution to the production of prospective genotypes as high-quality propagation material for further breeding work towards new applications of hemp in fields of economy and medicine. The key to this study is to optimize the micropropagation process of the selected Russian variety K290 *C. sativa*, which has the appropriate level of cannabinoids.

**The aim of the present** work was to comparative phenotypic study in terms of assessing the effect of the type of medium with different mineral salt content, i.e. Schenk-Hildebrandt medium (SH) and Gamborg medium (B-5), addition of vitamins and IAA auxin on growth of shoot with leaves during induction and development of adventitious roots.

In total, 12 variants were compared, taking into account the presence of vitamins and two concentrations of auxin IAA (0.5 and 1.0 mg/L). Nodal and apical fragments of plants obtained from seeds were selected as explants. The experiment was performed in triplicate.

**Our results show** that in terms of shoot length and number of leaves, the SH with the addition of IAA at a concentration of 1.0 mg/L without vitamins is the most effective. It was also found that IAA at a concentration of 0.5 mg/L in SH stimulated the formation of adventitious shoots ( $n = 3.21$  after 3 weeks). The average number of roots was 4.65 per shoot on the SH with 0.5 mg/L.

**Keywords:** *Cannabis sativa*; plant *in vitro* culture; auxin, Schenk-Hildebrandt medium; Gamborg medium

# Results and Discussion

## Schenk-Hildebrandt medium (SH)

1. SH medium with the addition of IAA at a concentration of 1.0 mg/L without vitamins was the most effective in terms of shoot length and number of leaves.
2. Auxin IAA at a concentration of 0.5 mg/L in SH stimulated the formation of adventitious shoots ( $n = 3.21$  after 3 weeks).
3. The average number of roots was 4.65 per shoot on the SH with 0.5 mg/L
4. After 6 weeks, the number of leaves and their size had doubled.
5. Did not observed the callus induction, malformation and necrosis of leaves and shoots.

# Results and Discussion

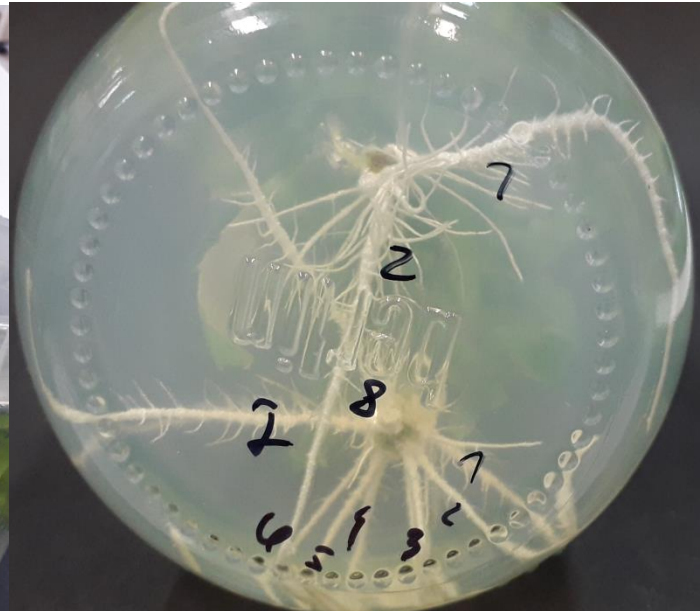
## Gamborg medium (B-5)

1. Among the variants of the B-5 medium, the most effective was B-5 with the addition of IAA at a concentration of 1.0 mg / L with vitamins - in stimulating the development of shoots along the length and number of leaves.
2. Auxin IAA at a concentration of 1.0 mg/L in B-5 medium without vitamins stimulated the formation of adventitious shoots (n = 1.3 after 3 weeks).
3. In contrast to SH medium, no adventitious roots were observed on the B-5 medium over a period of 7 days.
4. Did not observed the callus induction, malformation and necrosis of leaves and shoots.

# Summary of results

Lp	Variants of the medium	Shoot length (cm)	Number of leaves	Number of adventitious shoots
		(average $\pm$ SD)		
1	SH – control, without vitamins and auxin	1,9 $\pm$ 0,79	3,7 $\pm$ 1,7	1,0 $\pm$ 0,0
2	SH, without vitamins , with IAA 0,5 mg/L	4,53 $\pm$ 1,63	4,61 $\pm$ 2,02	3,2 $\pm$ 2,82
3	SH, without vitamins, with IAA 1,0 mg/L	7,55 $\pm$ 3,1	8,2 $\pm$ 3,15	1,6 $\pm$ 1,35
	SH – K, plus vitamins, without auxin	5,08 $\pm$ 1,43	5,2 $\pm$ 2,39	1,0 $\pm$ 0,0
5	SH, plus vitamins, with IAA 0,5 mg/L	2,4 $\pm$ 0,6	3,2 $\pm$ 1,09	1,0 $\pm$ 0,0
6	SH, plus vitamins, with IAA 1,0 mg/L	6,9 $\pm$ 2,88	8 $\pm$ 2,31	1,3 $\pm$ 0,48
7	B-5 – control, without vitamins and auxin	5,62 $\pm$ 4,42	4,5 $\pm$ 2,68	1,23 $\pm$ 0,43
8	B-5, without vitamins , with IAA 0,5 mg/L	1,92 $\pm$ 0,47	2,4 $\pm$ 0,89	1,0 $\pm$ 0,0
9	B-5, without vitamins, with IAA 1,0 mg/L	4,375 $\pm$ 1,69	6,7 $\pm$ 1,56	1,3 $\pm$ 0,94
10	B-5 – K, plus vitamins, without auxin	4,98 $\pm$ 3,71	4,73 $\pm$ 2,21	1,06 $\pm$ 0,26
11	B-5, plus vitamins, with IAA 0,5 mg/L	2,53 $\pm$ 1,78	3,33 $\pm$ 2,31	1,0 $\pm$ 0,0
12	B-5, plus vitamins, with IAA 1,0 mg/L	5,625 $\pm$ 2,23	6,9 $\pm$ 2,99	1,2 $\pm$ 0,63

*Cannabis sativa* growing on SH medium with IAA at 0,5 mg/L



## Conclusions

It can be concluded that a higher content of potassium ions as well as cobalt, copper, magnesium, molybdenum ions and a lower content of calcium, zinc, boron, manganese and iron ions in the SH medium (compared to Murashige Skoog medium) positively influenced the development of shoots and leaves.



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*in vitro* culture  
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