



ORIGINAL ARTICLE

Regulation of Potato Morphogenetic Processes *in vitro* by Hormonal and Light Actions

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ABSTRACT: The research on how phytohormones and light affect the morphogenetic responses of potatoes in culture conditions is presented in this article. Researchers looked at how growth promoters affect the regulation of potato morphogenesis in *in vitro* cultures of different potato varieties. They also looked at how light quality affects this regulation. Research has been conducted on Kazakh selection potatoes, which are commonly grown in Northern and Central Kazakhstan. In the course of the research, various variants of phytohormones and light quality were used for the growing and developing of micro-gears *in vitro*. The light quality has a different action on the potatoes' speed of growth. For example, rhizogenesis for the Tamyr variety has increased because of red light use. For the Aksor, Orbitaa, and Nerli varieties the best results were got in case the white light has been used. The concentration of the phytohormones has an influence on the potatoes growing. So, the most effective leafiness of shoots of all considered potato varieties has been gotten by auxin use in the concentration of 0.1 and 0.5 μ M.

INTRODUCTION

Potatoes became a dietary staple all over the world. It is an ideal crop especially for the mountainous regions because of its hardiness [1]. If we analyze the carbohydrate food source in the world range, has to be noted that roots and tubers have the third of the leader position. And potatoes take nearly half of all root crops consumed [2]. This agricultural culture contributes key nutrients to the diet: such as vitamin C, dietary fiber, and potassium [3]. One more positive effect is that potatoes have a more favorable overall nutrient-to-price ratio compared to many other vegetables. So, the potatoes became one of the most important staples worldwide

[4–6].

We have to note, potatoes are really popular in Kazakhstan like other the world. The average consumption of potatoes per capita in Kazakhstan is 120–130 kg per year per person, i.e., potatoes for Kazakhstanis are still the "second bread" and is one of the main branches of crop production, which determines the country's food security [7–10].

World Potato Congress (February 2020) gave data that Kazakhstan has a place in the top 25 of the countries that grow potatoes at the 20 points with the 3.807 million tons per a year (Table 1).

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Table 1. World's 25 largest individual producers of potatoes (million tons) (Source: FAOSTAT).

	2018	% Change	2017	2016	2015	2014	2013	2012
World	368.2471	-1.5	373.8486	357.0363	365.8195	369.9665	365.0875	361.0033
China	90.321	2	88.536	84.987	82.683	84.212	85.931	84.404
India	48.529	-0.2	48.605	43.77	48.009	46.395	45.344	41.483
Ukraine	22.504	1.3	22.208	21.75	20.839	23.693	22.259	23.25
Russian Fed.	22.395	3.2	21.708	22.463	33.646	31.501	30.199	29.533
USA	20.607	0.8	20.453	20.426	20.013	20.057	19.715	21.091
Bangladesh	9.744	-4.6	10.216	9.474	9.254	8.95	8.603	8.205
Germany	8.921	-23.9	11.72	10.772	10.37	11.607	9.67	10.666
France	7.871	-26.4	8.547	6.955	7.12	8.085	6.957	6.376
Poland	7.478	-18.5	9.172	8.872	6.314	7.689	7.29	9.092
Netherlands	6.03	-18.4	7.392	6.534	6.652	7.1	6.577	6.766
Belarus	5.865	-8.6	6.415	5.986	5.995	6.28	5.911	6.911
Canada	5.791	-3	5.97	5.92	5.875	5.643	5.733	5.642
Iran	5.321	4.3	5.102	4.995	5.141	4.989	4.598	5.069
Peru	5.121	7.2	4.776	4.4	4.705	4.705	4.571	4.475
UK	5.028	-19.1	6.218	5.373	5.588	5.911	5.685	4.553
Egypt	4.896	1.1	4.841	4.113	4.955	4.611	4.265	4.758
Algeria	4.653	1	4.606	4.783	4.54	4.674	4.887	4.219
Pakistan	4.592	19.2	3.853	3.978	4	2.901	3.802	3.393
Turkey	4.55	-5.2	4.8	4.75	4.76	4.166	3.955	4.822
Kazakhstan	3.807	7.2	3.551	3.546	3.521	3.411	3.344	3.126
Brazil	3.688	0.9	3.657	3.851	3.868	3.69	3.554	3.732
Colombia	3.108	-16.2	3.707	3.034	2.582	2.158	2.129	1.847
Nepal	3.088	14.8	2.691	2.806	2.586	2.818	2.69	2.584
Belgium	3.045	-31.1	4.417	3.403	3.69	4.381	3.428	2.93

Bureau of the national statistics of the Republic of Kazakhstan (2020) gave data that Kazakhstan has manufactured 4 million tons of potatoes per year and 359600 tons have been exported. However, potatoes' import was at the level of 45800 tones. So, the Kazakh companies provided the Kazakh citizens' demands in potatoes in 108.5% (95 kg per a citizen in a year).

This crop has widespread in Kazakhstan and potato growing is one of the main branches of crop production. This crop determines the food security of the country. The official data gives us an understanding that 20% of elite potato seeds are growing in Kazakhstan. The rest of the elite seed potatoes are imported from abroad. Has been noted, the poor quality of seed material became the main cause of low potato yield in Kazakhstan [11]. Moreover, potatoes are prone to degeneration due to the accumulation of phytopathogens with vegetative propagation, so the cultivation of disease-resistant

varieties and effective seed production technologies are of paramount importance [12,13].

Modern potato growing provides for the production of initial seed potatoes based on biotechnological methods of health improvement, microclonal reproduction and subsequent methods of accelerated reproduction of healthy plants. Potato growth and morphogenesis *in vitro* are very sensitive to the action of phytohormones and physical factors of cultivation [14]. There is no doubt that the conditions of growth and development of test tube plants have an effect on the nature of physiological processes. The study of the peculiarities of potato physiology is important for the creation and optimization of the technology of clonal micropropagation of planting material.

Obtained virus-free potato plants have to be improved to increase the multiplication factor and the rate of plant recovery after microreplication. The purpose of this work was to study the effect of exogenous hormones and light

of different spectral compositions on potato morphogenesis in culture *in vitro*.

MATERIAL AND METHODS

The research methods that have been conducted in our research were based on the classic biotechnologies work methods with the cultures of the isolated tissues and plant organs. All conducted procedures during the study have been considered and approved by the Ethical comity of the Abai Kazakh National pedagogical university.

Research design

We have taken 4 potato varieties (Aksor, Nerli, Tamyr, Orbita) as the research objects. All these varieties are included in the State Register of the Republic of Kazakhstan. Fragments of internodes obtained by cutting donor plants were used as explants, placed on agarized nutrient media with a mineral composition to induct the potatoes' morphogenesis [15,16].

Microcuttings with 1–2 axillary buds and tops of microshoots were used as explants. Rooting of micro-plants was carried out on an agarized nutrient medium according to Murashige and Skoog (MS), supplemented

for the induction of rhizogenesis of α -naphthylacetic (NAA), β -indolylbutyric (IBA), β -indolylpropionic (IPA) in various concentrations. Explants were cultivated under the following conditions: at a temperature of 23-26°C, light mode 16/8 (day/night), illumination of 5000 lux and humidity of 50-60%. The duration of the passage was 20 days [17].

Figure 1 demonstrates the preparation process of the MS nutrient medium. And different concentrations of growth regulators (0.1, 0.5, 1.5, 3 and 5 microns of NAA, IBA and IPA) have been added in it.

Morphogenetic processes were induced under controlled conditions: at a temperature of 23-26°C, light mode 16/8 (day / night), illumination of 5000 lux and humidity of 50-60%. The duration of the passage was 20 days.

The plants were illuminated using special light-emitting diode (LED) lamps designed for growing plants. LED A60 6 W E27 and LS670 120 SMD IP65 12 W lamps of Gauss and Feron brands were used as experimental lighting sources in tissue culture. Lamp power is 6 W, diameter is 60 mm, length is 110 mm. Gauss LED A-60 6 W E27 lamps emit white spectrum light (white light – WL), Feron LS670 12 W - red-blue lights (RBL) (Figure 2).

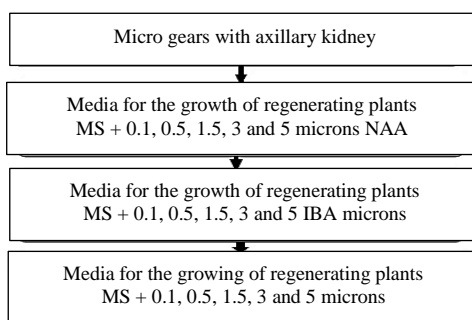


Figure 1. Scheme of morphogenesis *in vitro* potato varieties.

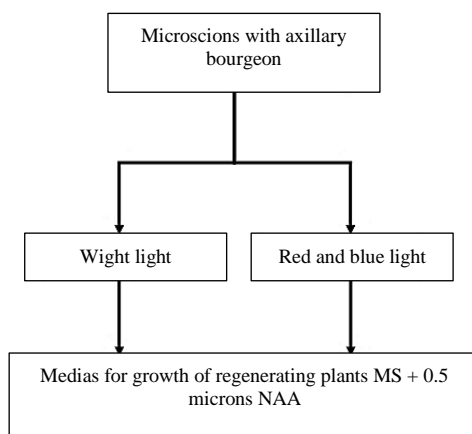


Figure 2. Scheme of studying the effect of light quality on morphogenesis *in vitro* culture of potato varieties Axor, Taimyr.

The indicators were recorded on the 20th day after planting explants on the nutrient medium: the number of roots (pcs/exp), the length of the roots (cm), the length of the shoot (cm), the number of leaves (pcs/exp).

RESULTS

The main task in obtaining virus-free potato plants is to increase the coefficient of their reproduction and the rate of regeneration of micro clones. So, it becomes necessary to optimize the growing conditions *in vitro* [18].

It's known, that light is one of the most important growth-regulating environmental factors for plants, and above all its spectral composition. Plant life processes are closely dependent on the intensity and spectral composition of light. Light of different spectral compositions regulates

growth and development, as well as photosynthetic processes and plant productivity, both *in vivo* and *in vitro* [19, 20].

In the course of the study, the effects of various concentrations were studied (0.1, 0.5, 1.5, 3, 5 microns) of α -naphthylacetic (NAA), β -indolyl butyric (IBA), β -indolylpropionic (IPA) acids on the rhizogenesis of four potato varieties in culture *in vitro*.

Analysis of the effect of the concentration of NAA (0.1, 0.5, 1.5, 3, 5 microns) on the rhizogenesis of potato varieties *in vitro* showed that after 20 days on a nutrient medium with 0.5 microns of NAA, the maximum number of roots (24.1 ± 4.32 pcs/exp) was obtained in the Tamyр variety (Figure 3).

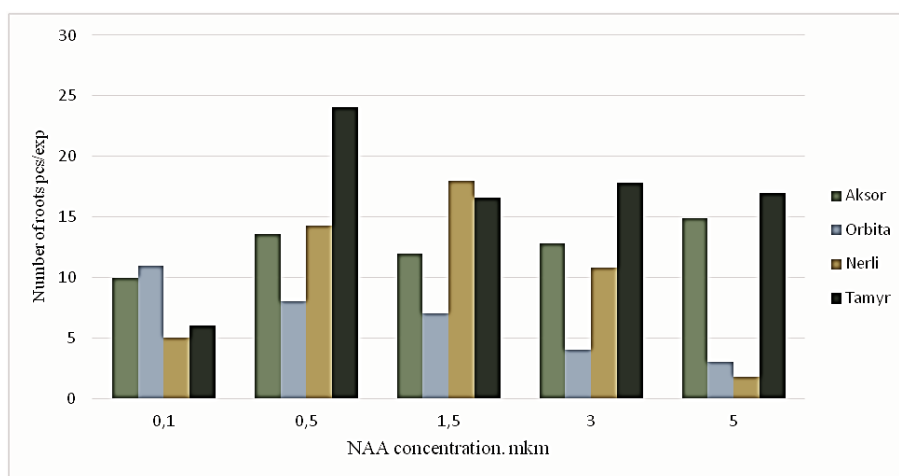


Figure 3. The effect of the concentration of α -naphthylacetic acid on the rooting of various potato varieties in culture *in vitro*.

The study of the length of the formed roots in the Tamyр variety showed that in the variant with 0.5 microns of NAA there were the longest roots (3.3 ± 1.2 cm). When using a concentration of 1.5 microns of NAA, similar

results were revealed. But higher concentrations of auxin (3.0 and 5.0 microns) inhibited the growth of the formed roots (Figure 4).

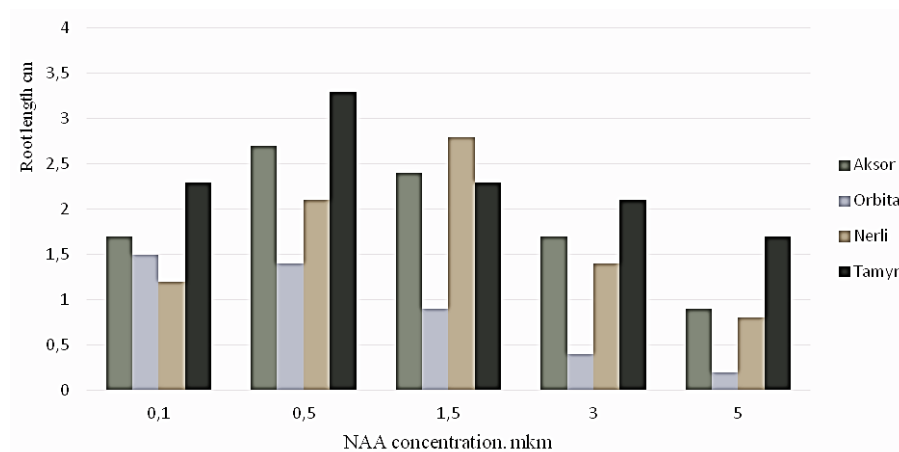


Figure 4. The effect of the concentration of α -naphthylacetic acid on the root length of various potato varieties in culture *in vitro*.

The highest shoots were formed in the Orbitaa variety on a nutrient medium with 0.5 microns of NUC (3.95 ± 0.53 cm). This indicator is 1.5 cm less compared to Tamyr when adding 1.5 microns of auxin to the medium. The smallest shoot length was characteristic of the Orbitaa

variety in the experiment variant with 1.5 microns of NAA, which is 1.2 cm longer than that of the Nerli variety with 5 microns of NAA (Figure 5).

The leafiest shoots were observed in the Tamyr variety in the variant with 1.5 microns of NAA (Figure 6).

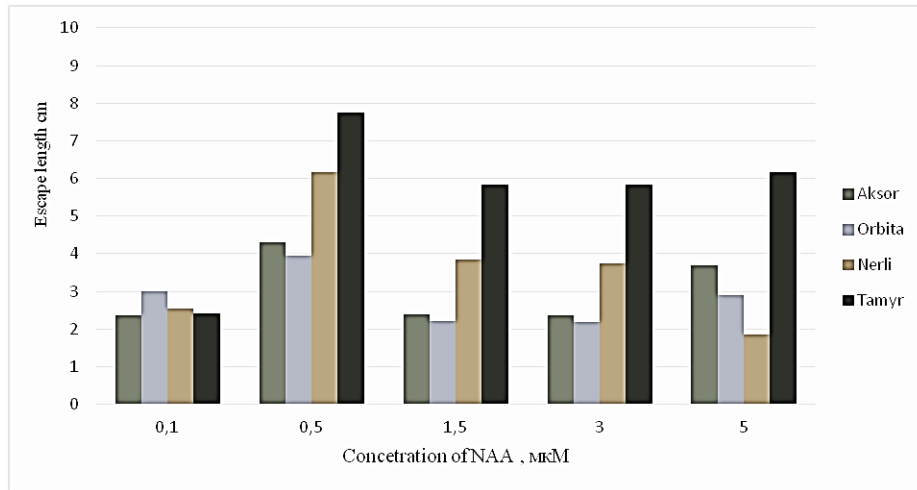


Figure 5. The effect of the concentration of α -naphthylacetic acid on the shoot length of potato varieties (*Solanum tuberosum L.*) in culture *in vitro*.

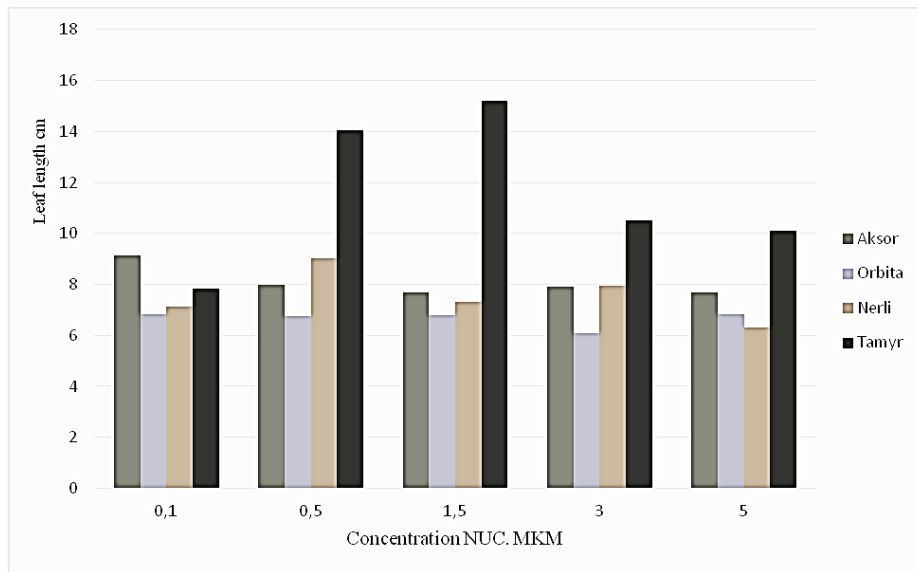


Figure 6. The effect of the concentration of α -naphthylacetic acid on the number of leaves of potato varieties (*Solanum tuberosum L.*) in culture *in vitro*.

Higher concentrations of NAA (3.0 and 5.0 microns) reduced its leafiness by almost 1.5 times. The lowest leafiness of shoots was fixed for the Orbita variety in the variant with 3 microns of NAA. On media with 0.1, 3 and 5 microns of auxin, the average number of leaves was about 7.56 ± 1.85 pcs/exp; 7.14 ± 1.32 pcs/exp; 7.22 ± 1.18 pcs/exp.

The effect of the concentration of β -indolylbutyric acid on the rooting of potato varieties β -indolylbutyric acid (IBA)

It is a phytohormone from the auxin class and is part of root formation stimulants. The use of IBA as a stimulator of rhizogenesis showed that the maximum number of roots (33.2 ± 1.05 pcs/exp) was observed in Tamyr cultivar on a nutrient medium with 3.0 microns. A high concentration of auxin (5.0 microns) caused inhibition of rhizogenesis in all potato varieties studied. The low

concentration of IBA did not sufficiently induce root formation. Thus, the average number of roots in mini-

plants of all potato varieties on a medium with 0.5 microns of IBA was 11.9 ± 1.51 pcs/expl (Figure 7).

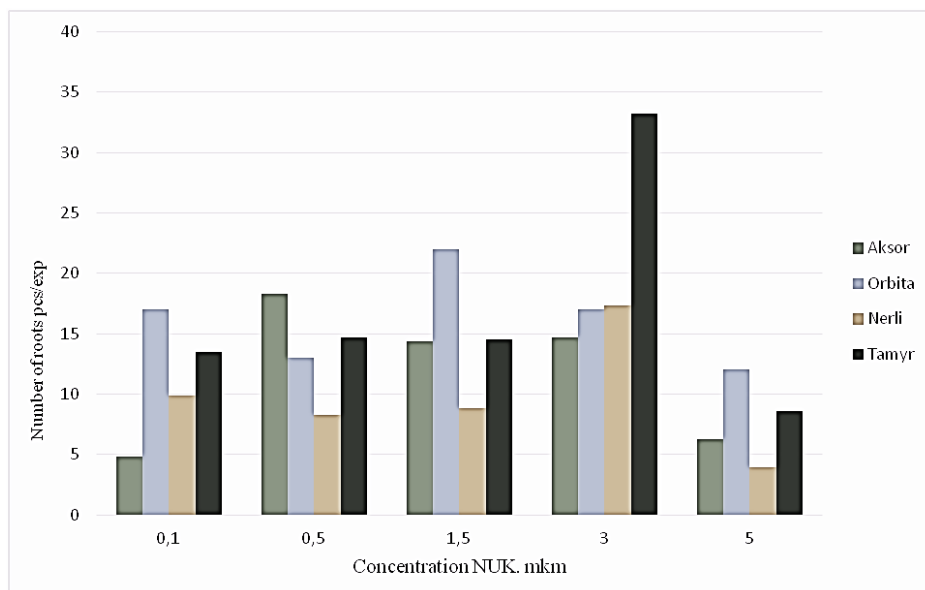


Figure 7. The effect of the concentration of β -indolylbutyric acid on the rooting of potato varieties (*Solanum tuberosum L.*) in culture *in vitro*.

Analysis of the length of the formed roots revealed that the Aksor variety in the concentration variant with 0.1 microns of IBA had the longest roots (6.06 ± 0.14 cm). An increase in the auxin concentration to 0.5 microns contributed to the active growth of the roots of all potato varieties (3.92 ± 0.46 cm) (Figure 8).

When using a low concentration of auxin (0.1 microns), the shoot length was maximum, on average for varieties - 7.31 ± 0.22 cm. The most optimal was the concentration of IBA equal to 3 microns, since there was no strong pulling of potato shoots on it (Figure 9).

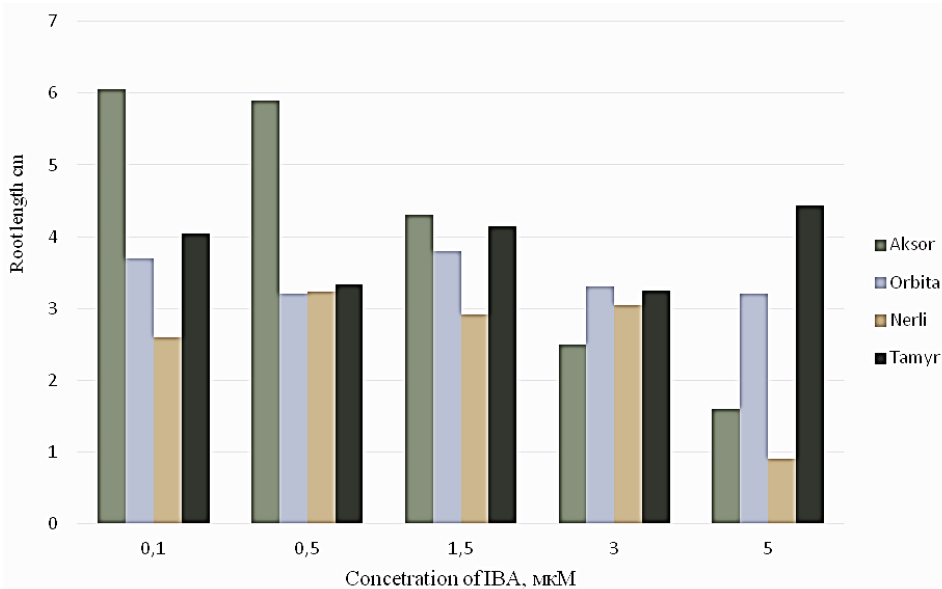


Figure 8. Effect of the concentration of β -indolylbutyric acid on the root length of potato varieties (*Solanum tuberosum L.*) in culture *in vitro*.

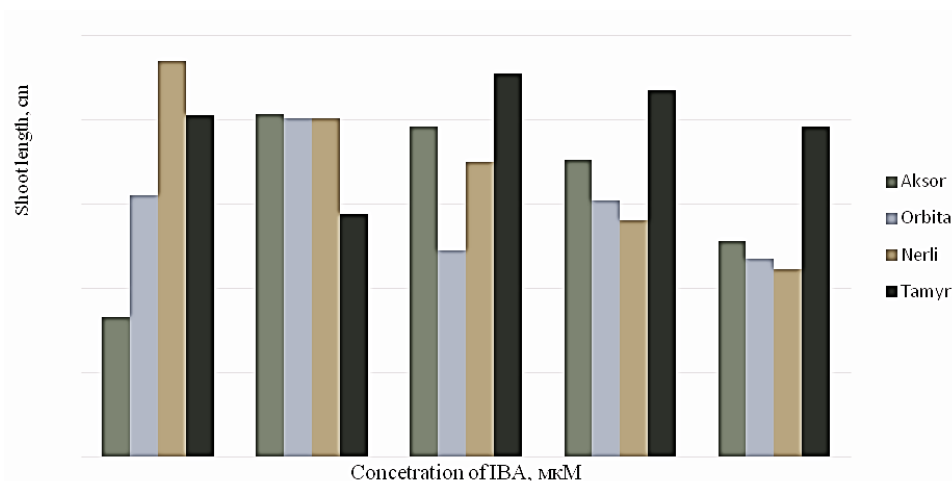


Figure 9. Effect of the concentration of β -indolylbutyric acid on the shoot length of potato varieties (*Solanum tuberosum L.*) in culture *in vitro*.

The most leafy shoots were observed in the Tamyр variety in the variant with 3 microns of IBA. A higher concentration of IBA (5.0 microns) reduced its leafiness

by 1.5 times. On average, auxin concentrations of 0.5 and 1.5 microns caused good leafiness of shoots of all studied varieties (Figure 10).

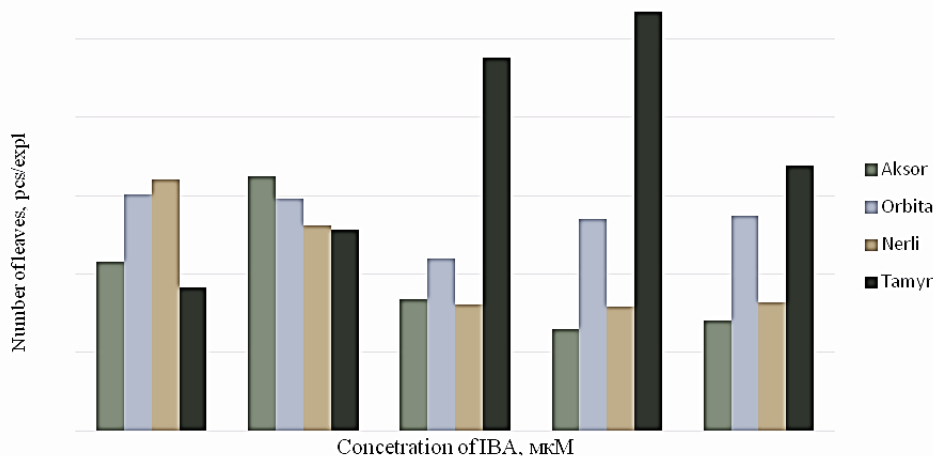


Figure 10. Effect of the concentration of β -indolylbutyric acid (IBA) on the number of leaves of potato varieties (*Solanum tuberosum L.*) in culture *in vitro*.

A low auxin concentration (0.1 microns) was optimal for the Orbitaa and Nerli varieties. The concentration of 0.1 microns of IBA for the Aksor variety turned out to be optimal, since the greatest leafing of shoots was noted here (10.7 ± 0.35 pcs/expl), compared with other studied concentration variants.

The effect of the concentration of β -indolylpropionic acid (IPA) on the rooting of potato varieties

The highest percentage of rooted shoots was obtained after 20 days of cultivation on MS medium supplemented with 0.1; 0.5; 1.5; 3.0 and 5.0 microns of β -indolylpropionic acid (IPA) in the Aksor and Orbita varieties (Figure 11).

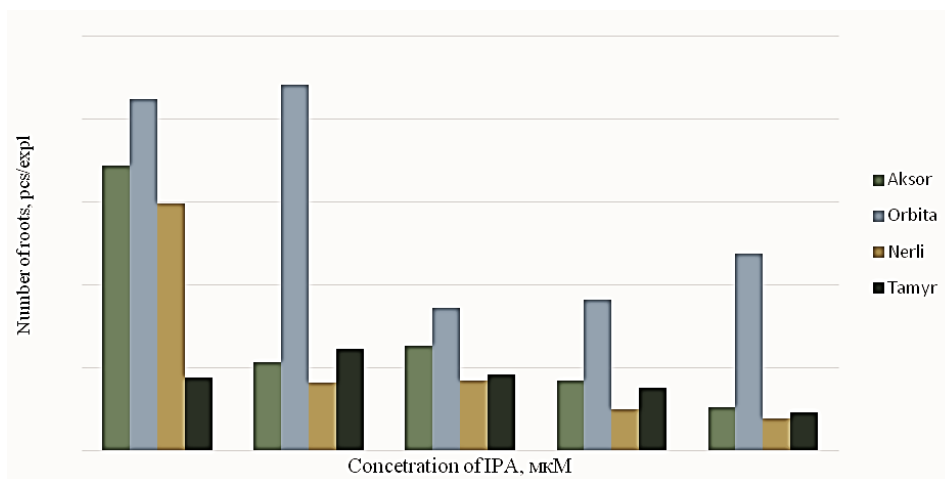


Figure 11. The effect of the concentration of β-indolylpropionic acid on the rooting of various potato varieties *in vitro* culture.

The low concentration of IPA (0.1 microns) contributed to the active root formation of three potato varieties - Aksor (17.2±2.21 pcs./exp.), Orbita (21.2±4.52 pcs./exp.). Nerli (14.9±2.35 pcs/exp). A higher concentration of auxin (0.5 microns) weakly induced rhizogenesis in all potato varieties except Orbita (22.5±3.91 pcs/expl). In the variant with 3.0 and 5.0 microns, it caused inhibition of root formation in all the studied varieties.

Analysis of the length of the formed roots revealed that the Orbitaa variety in the variant with 1.5 microns of IPA

had the longest roots (3.98±0.24 cm). Higher concentrations of auxin (1.5; 3.0; 5.0 microns) contributed less effectively to the growth of the roots of all potato varieties studied. The low auxin concentration (0.1 microns) weakly stimulated an increase in root length in all potato varieties (Figure 12).

When using a low concentration of IPA (0.1 microns), the shoot length was maximum, on average for varieties - 4.74 ±0.58 cm (Figure 13).

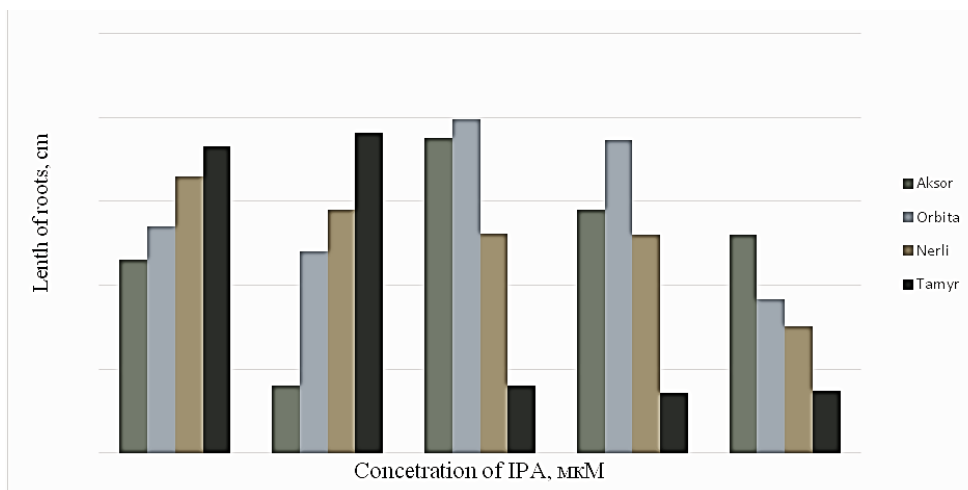


Figure 12. Effect of the concentration of β-indolylpropionic acid on the root length of potato varieties (*Solanum tuberosum L.*) *in vitro*.

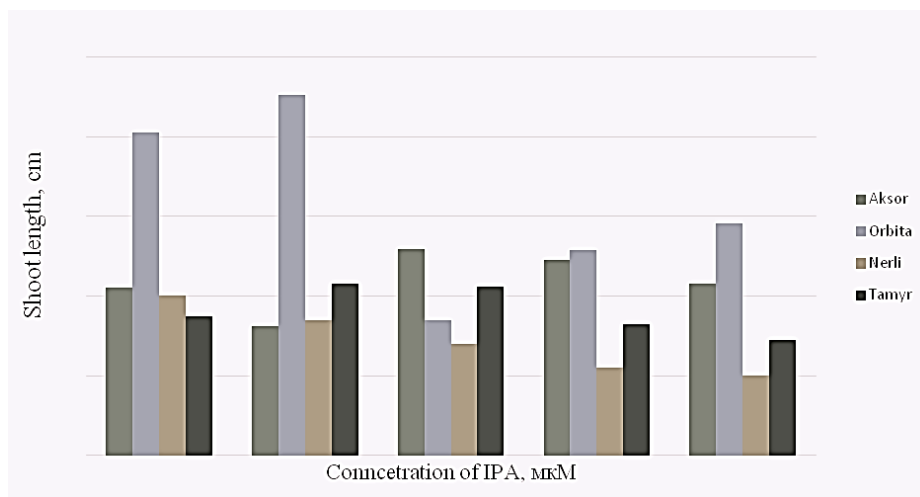


Figure 13. Effect of the concentration of β-indolylpropionic acid on the shoot length of potato varieties (*Solanum tuberosum L.*) *in vitro*.

The shoot length was minimal on the MS medium supplemented with 5 microns of IPA (3.22±0.25 cm). The maximum shoot length at concentrations of 0.1 and 0.5 microns of IPA was observed in the Orbitaa variety

(8.45±0.15 cm).

In the variant with 0.5 microns and IPA, the maximum number of leaves was noted (11.35=1.14 pcs/expl) (Figure 14).

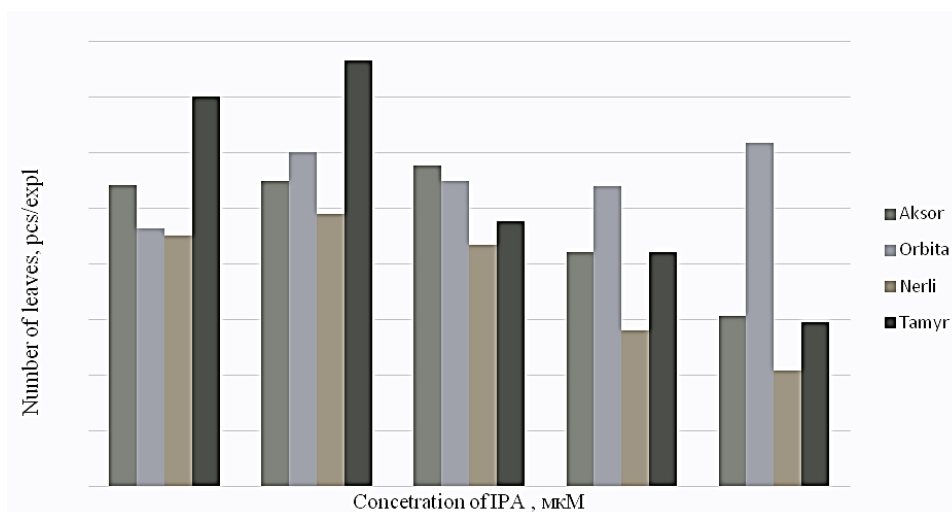


Figure 14. Effect of the concentration of β-indolylpropionic acid on the number of leaves of potato varieties (*Solanum tuberosum L.*) *in vitro* culture.

The leafiness of all studied potato varieties decreased at higher concentrations of IPA (1.5, 3.0, and 5.0 microns). The best leafiness of the shoots of all the studied varieties was caused by auxin concentrations of 0.1 and 0.5 microns.

The influence of light in controlling *in vitro* potato morphogenesis

In laboratories, fluorescent lamps are used as a light source. The optimal illumination values for herbaceous plants are values up to 1000 lux. Illumination below 300 lux or above 3000-10000 lux suppresses the growth and development of plants. The spectral composition of light

and its quality, the intensity of illumination will affect the physiological development of the plant, and seasonal and daily dynamics are also of considerable importance [21]. Light of different spectral composition regulates the growth, development, photosynthetic processes and productivity of potatoes *in vitro* [22,23].

To study the effect of the spectral composition of light for growing potato regenerants, we used white, combined (red and blue) LED panels. The efficiency of LED lamps is justified by their monochromatic radiation. The phytoactive part of the spectral composition of the light was selected directly for the cultivated plant. LED phytolamps have a wavelength that is necessary to illuminate plants, blue and red spectrum. Depending on

the ratio of blue and red colors, the effect of the LED lamp on the microclones of plants varies from their power. LEDs make it possible to adjust the spectrum of the luminous flux and are characterized by high light output, the duration of the working resource [24].

The study of the effect of light quality on the induction of rhizogenesis of potato root varieties showed that Aksor and Tamyр varieties formed 15.6-24.3 pcs/root exploit.

When the WL was illuminated, an insignificant increase in root formation was noted in the Aksor variety (by 3-7 pcs/exp), and in the Tamyр variety, 4.1 pcs/exp of the root was formed when the RBL was illuminated. A significant induction of rhizogenesis in the Nerli variety was noted in the variant with RBL - 37.6 ± 4.1 versus 25.6 ± 2.4 pcs/expl of roots on RBL (Figure 15).

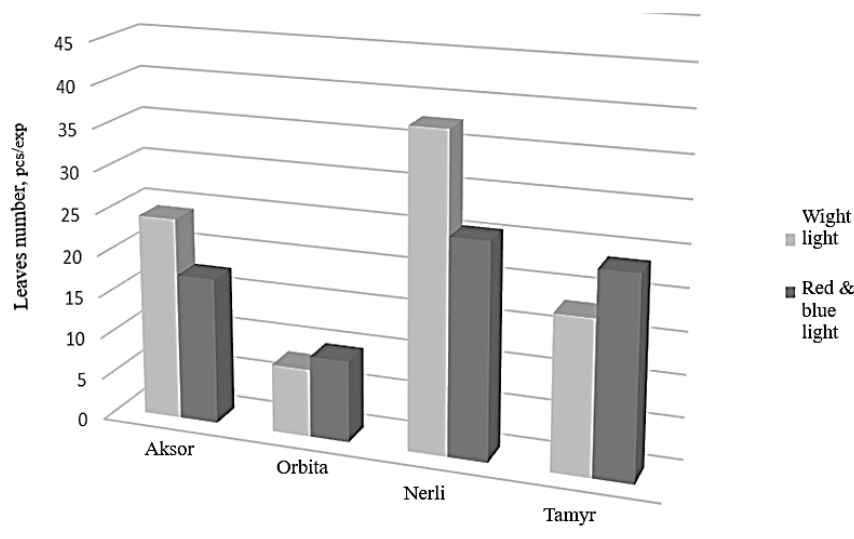


Figure 15. The effect of light quality on the induction of rhizogenesis in regenerating plants of different potato varieties in *in vitro* culture.

When irradiating plants of the Orbita CS variety, an insignificant increase in the number of roots was observed compared to the control (BS).

Similar results were recorded for other morphological features (number of leaves and shoot height). The Tamyр variety formed almost 2 times more leaves (18.8 ± 2.1 pcs/exp on WL and 22.9 ± 2.2 pcs/exp on RBL) compared to other studied varieties (8.5 ± 0.8 pcs/exp in Nerli and 12.6 ± 1.3 pcs/exp leaves in Aksor variants with RBL).

As an inducer of rhizogenesis, it is more effective to use red light for the Tamyр variety and white light for the Aksor, Orbita, Nerli varieties. The quality of light of LED lamps (white and red-blue), in the potato varieties studied by us, does not affect the length of the roots formed.

DISCUSSION

The Tamyр type of potatoes produced the most roots after 20 days on a nutritional medium with 0.5 microns of NAA, according to research on the impact of NAA concentration on rhizogenesis of different potato varieties

in vitro. The results were in agreement with [25]. The concentration of IBA at 3 microns was the best since potato shoots did not pull strongly against it, similar results were obtained [26]. The highest shoot length for the tested types was 4.74 ± 0.58 cm while employing a low concentration of IPA (0.1 microns). On the MS medium with the addition of 5 microns of IPA, the shoot length was minimal. These results were in line with other studies [3, 27]. The Aksor and Tamyр types created 15.6-24.3 pcs/root exploit, according to research on the influence of light quality on the induction of rhizogenesis of potato root varieties. Red light is more successful as a rhizogenesis inducer for the Tamyр variety compared to white light, Aksor, Orbita, and Nerli varieties. Similar results were recorded [12,19 and 27].

CONCLUSIONS

The morphogenetic reactions of potatoes grown in culture conditions are significantly influenced by phytohormones and light. Phytohormones are plant growth regulators that regulate various physiological and developmental processes, including root initiation, shoot development,

and cell division. Light, on the other hand, plays a key role in regulating photomorphogenesis, which affects plant architecture, growth, and development. To summarize, the relationship between phytohormones, light, and the morphogenetic reactions of potatoes in culture conditions is intricate and influenced by multiple elements. A thorough understanding of the effects of phytohormones and light on potato tissue culture is important for optimizing the culture conditions and achieving successful results. By controlling these factors, it is possible to promote root initiation, shoot development, and the formation of lateral buds, leading to the development of healthy and vigorous potato tissue culture plants. The results of the conducted studies can be applied in optimizing the cultivation regime of potato microclones *in vitro*, as well as in conditions of hydroponic cultivation in order to successfully adapt plants *in vivo*, shorten the growing season and enhance the production process to maximize the yield of healthy mini-tubers.

Conflict of interests

Regarding the publication of this article, the authors declare that there are no conflicts of interest.

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