

DOI: <https://doi.org/10.23670/IRJ.2023.138.30>**EFFECT OF PEA SEED PRETREATMENT WITH MELATONIN AND GERMINATION SUBSTRATES ON RESPIRATORY ACTIVITY AND EMBRYO GROWTH AFTER SHORT-TERM DEHYDRATION OF SWELLING SEEDS**

Research article

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**Abstract**

We studied the effect of substrates for seed germination and pea (*Pisum sativum* L) seedlings cultivation – distilled water and Hoagland's solution ½ – on embryo respiration and growth of pea seedlings after pretreatment of seeds with melatonin under control conditions and under unfavorable conditions – seed dehydration. Distilled water was considered as a substrate weakening the physiological state of seeds and seedlings, and Hoagland's solution ½ – as a substrate, providing them more fully with mineral compounds. It was shown that soaking seeds and growing seedlings in distilled water negatively affects the respiratory activity of embryos, reduces the rate of seed germination, and also negatively affects the result of seed pretreatment with melatonin. At the same time, seed resistance to short-term dehydration decreases, which is ultimately expressed in the reduction of seedling growth activity. The results obtained showed that the effect of pretreatment of seeds with melatonin largely depends on the physiological state of the forming seedlings. It is assumed that the negative effect of this hormone on plant seedlings weakened under the influence of unfavorable environmental factors may be due to insufficient supply of their metabolites, including metal ions and other mineral elements, for accelerated development of seedlings.

**Keywords:** *Pisum sativum* L, embryo respiration, seedling growth, substrates, melatonin, dehydration.**ВЛИЯНИЕ ПРЕДОБРАБОТКИ СЕМЯН ГОРОХА МЕЛАТОНИНОМ И СУБСТРАТОВ ПРОРАЩИВАНИЯ НА ДЫХАТЕЛЬНУЮ АКТИВНОСТЬ И РОСТ ЗАРОДЫШЕЙ ПОСЛЕ КРАТКОСРОЧНОГО ОБЕЗВОЖИВАНИЯ НАБУХАЮЩИХ СЕМЯН**

Научная статья

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**Аннотация**

В настоящей работе изучено влияние субстратов для проращивания семян и выращивания проростков гороха (*Pisum sativum* L) – дистиллированной воды и раствора Хогланда ½ – на дыхание зародышей и рост проростков гороха после предварительной обработки семян мелатонином в контрольных условиях и в условиях неблагоприятного фактора – обезвоживания семян. Дистиллированная вода рассматривалась как субстрат, ослабляющий физиологическое состояние семян и проростков, а р-р Хогланда ½ – как субстрат, более полно обеспечивающий их минеральными соединениями. Показано, что замачивание семян и выращивание проростков в дистиллированной воде отрицательно влияет на дыхательную активность зародышей, снижает скорость прорастания семян, а также негативно сказывается на результате предварительной обработки семян мелатонином. При этом снижается устойчивость семян к кратковременному обезвоживанию, что в конечном итоге выражается в снижении ростовой активности проростков. Полученные результаты показали, что эффект предварительной обработки семян мелатонином во многом зависит от физиологического состояния формирующихся проростков. Предполагается, что отрицательное действие этого гормона на проростки растений, ослабленные под действием неблагоприятных факторов внешней среды, может быть связано с недостаточным обеспечением их метаболитами, в том числе ионами металлов и другими минеральными элементами для ускоренного развития проростков.

**Ключевые слова:** *Pisum sativum* L, дыхание зародышей, рост проростков, субстрат, мелатонин, дегидратация.**Introduction**

In our previous work, it was shown that melatonin treatment of lupin seedlings under water deficit conditions had opposite effects on hypocotyl and root growth, the level of POL in the tissues of these organs, and respiration of mitochondria isolated from them; in particular, it acted as an antioxidant in the first case and as a prooxidant in the second [1]. Roots experienced a much stronger water deficit. At the same time, mitochondrial functional activity was reduced in roots, including inhibition of respiratory substrate oxidation, as well as decreased activity of the alternative electron transport pathway in the ETP. As a result, the balance of POLs in roots was disturbed in the direction of increasing their content in cells. To explain the obtained

results, we assumed that melatonin may have an opposite effect on plant objects and/or individual plant organs in different functional states. However, since roots and epicotyls were compared in the above work, the effect of tissue-specificity of melatonin action was not excluded. Therefore, in the present work we studied the effect of substrates for seed germination and pea (*Pisum sativum* L) seedlings growth – distilled water and Hoagland's ½ solution – on germ respiration and growth of pea seedlings after pretreatment of seeds with melatonin under control conditions and under the unfavourable factor of seed dehydration. Treatment of plants with hormones and growth stimulants increases the yield and quality of agricultural products [2], [3]. Seed pretreatment is particularly effective, probably because the effect occurs during seed swelling and germination, the period most critical for plant growth and development [4], [5]. As was shown in [3] on maize seed embryos, it is during the early period of swelling that the systems that ensure high seed performance are laid down. The most important of them is the respiratory system, which in the absence of photosynthesis is the only source of energy metabolites. Oxygen uptake was recorded from the first minutes of swelling of separated lupin embryos [6]; in seeds, the rate of respiratory system establishment depended on the rate of water supply to the seeds [7], [8]. In parallel, the storage tissue of the seed awakens and there is an intensive outflow of carbohydrates for the synthesis of protein compounds that support the formation of new structures and protective compounds in the embryo tissues [3].

Intensification of metabolism and contact with air oxygen cause an increase in ROS levels in tissues [9]. Normally, this ROS surge is under strict control of the antioxidant system and is not accompanied by oxidative stress [10], but if this control is disturbed, the seed dies.

The hormone melatonin [11] is widely known as a stimulant and regulator of plant growth and development [13], which interacts with other hormones [3], particularly under dehydration conditions [13]. Pretreatment with melatonin has been shown to be a stimulatory factor that improves seed quality. The ability of melatonin to enhance germination and increase the rate of seed germination has been shown in numerous experiments [14], [15], [16]. Melatonin is able to neutralise oxidative stress occurring under unfavorable conditions for plants by regulating physiological processes under stress conditions, in particular, by regulating the activity of antioxidant systems [17]. Under conditions of salt stress, melatonin is able to regulate ion homeostasis by directly regulating the operation of ion channels and maintaining a reduced content of Na and K ions in the cytoplasm [18]. The dose effect of melatonin is known [19], [20], [21]. Thus, it was shown on *Arabidopsis thaliana* that only in low doses the drug can cause plant growth stimulation [18]. In addition to dose-dependence, melatonin can exhibit pro-oxidant properties in some cases, which requires understanding and studying the mechanism of its action. We have previously shown that the same dose of melatonin can act as a prooxidant or as an antioxidant, as we assume, depending on the organ's resistance to unfavorable effects [22]. To date, the literature has paid insufficient attention to the plant state, which is the target of melatonin action. The only paper we found that compared the state of plants grown on deionised water and Hoagland's solution dates back to 1982 [23]. The inhibitory effect of deionised water on mitosis in the root tips of pea seedlings was detected 3 days after the onset of seed swelling, and a significant increase in the organ content of ISC was observed in the roots. The roots were more sensitive to the effect than the epicotyl. These results indicate that under the action of Hoagland's solution physiological changes in seedling metabolism occurred, which were absent in seedlings grown on water. In the present work, we investigated the effect of melatonin pretreatment of pea seeds and their dehydration on the respiratory activity of embryos, oxidative stress and subsequent growth of seedlings depending on the medium of seed swelling and seedling growth: distilled water or Hoagland's solution ½.

## Research methods and principles

### 2.1. Research object

The object of the research are presented by embryos, isolated from 18 hour-old swelling seeds and 5-day-old seedlings of Pea seeds (*Pisum sativum* L) variety "Nemchinovsky 100" obtained at the Institute of FGBNU "Federal Scientific Centre "Nemchinovka". Seeds were surface sterilised with 5% calcium hypochlorite for 5 min, washed three times with sterile distilled water and soaked for 1 h in water or in 0.1 µM melatonin solution, then transferred to filter paper moistened with distilled water (group 1) or ½ Hoagland's solution (group 2). After 14 h of germination, a part of seeds of both variants was transferred to dry filter paper for 4 h, after which samples were taken to examine embryos of control and experimental seeds, and a part was left to germinate for 5 days in the dark on distilled water or ½ Hoagland's solution. We used short-term exposure under dehydration conditions (4 h), considering these conditions to be less traumatic than exposures from 12 h to several days used by other authors [24], [25], and the sampling time coincided with the period of active water saturation of swelling embryos, which made them, we believe, more sensitive to dehydration. Germination was defined as the emergence of embryos from the seed coat. In seeds of the first group, by the time of sampling, the embryos had swollen but had not broken through the seed coat, while in seeds of the second group, the embryos were actively emerging from the seed coat and were in contact with the external environment. The most developed embryos were selected for the study.

### 2.2. Oxygen uptake by embryos

Oxygen uptake by embryos was measured amperometrically using the electrode designed by Sholts and Ostrovsky (1975). A reaction medium (1 cm<sup>3</sup>) contained 20 mM HEPES buffer (pH 7.4), To determine the activity of cytochrome (V<sub>cyt</sub>) and alternative (V<sub>alt</sub>) pathways of mitochondrial oxidation, as well as the rate of residual respiration (V<sub>res</sub>), we used specific inhibitors of terminal oxidases – KCN and salicylhydroxamic acid (SHAM), inhibiting the activity of cytochrome oxidase (COX) and alternative oxidase (AOX), respectively. The optimal final concentrations of KCN and SHAM, which are, respectively: 1 mM and 10 mM, were selected by titration in preliminary experiments. In embryos, V<sub>cyt</sub> activity was determined by the sensitivity of respiration to KCN in the presence of SHAM, and V<sub>alt</sub> activity was determined by the sensitivity of tissue respiration to SHAM in the presence of cyanide. Residual respiration (V<sub>res</sub>) was determined by the rate of tissue oxygen uptake in the presence of inhibitors of cytochrome and alternative pathways [26].

### 2.3. The content of TBARS

The content of TBARS (thiobarbituric acid reactive substances) in embryonic tissues was determined according to the method of Hodges et al. [27]. Tissue suspension (1 g) was homogenised in 25 ml of water-ethanol solution in the proportion of

20/80, then the samples were centrifuged at 3000 g for 10 min. Aliquots of 1 ml each were introduced into tubes containing 20% TBA (- TBA solution) or containing 65% TBA (+ TBA solution) in addition to TCA. The samples were incubated in a water bath at 95°C for 30 min, immediately cooled and centrifuged for 10 min at 3000 g. The staining intensity of the supernatant was determined on a Genesis 10uv spectrophotometer at three wavelengths (440, 532 and 600 nm). TBARS content was determined according to the following formula:

$$1) [(Abs\ 532_{+TBA}) - (Abs\ 600_{+TBA})] - [(Abs\ 532_{-TBA}) - (Abs\ 600_{-TBA})] = A$$

$$2) [(Abs\ 440_{+TBA} - Abs\ 600_{+TBA}) \cdot 0.0571] = B$$

$$3) MDA\ equivalents\ (nmol\ /ml) = [(A-B) / 157\ 000] \cdot 10^6$$

#### 2.4. Water deficit of embryo tissues

Water deficit of embryo tissues was determined according to the method of Schmit and Diesengremel [28] using the following formula:

$$\text{Water deficit} = \frac{\text{Turgescent weight} - \text{initial weight}}{\text{Turgescent weight} - \text{dry weight}} \cdot 100$$

The size of epicotyls of 5-day-old seedlings was determined using a ruler with a nonius.

#### 2.5. Statistical Analysis

The arithmetic mean values of the obtained values and their standard errors are presented in the tables. The paper presents data of experiments with 3-5-fold biological repetition. Statistically significant differences between the mean values were determined using Tukey test in ANOVA programme, the results marked with different letters are statistically significant at  $P \leq 0.05$ .

#### Results

The average weight of 1 embryo from seeds of group 1 by the time of sampling – 18 h of seed soaking – was  $0.010 \pm 0.0022$  g, from seeds of group 2 –  $0.013 \pm 0.0024$  g. Obviously, the negative effect of water as a substrate on seed germination manifested itself already after 18 h of seed soaking.

##### 3.1. Water deficit of embryos

Under initial water deficit of embryo tissues 8-9%, under dehydration conditions it increased by 11.6% in embryos of group 1 (seeds germinated on distilled water) and by 8.7% in embryos of group 2 (seeds germinated on Hoagland ½ solution). Pretreatment with melatonin in group 2 increased water deficit in the control variant by 24%, making it equal to the dehydration+melatonin variant, the same in both groups of embryos (Table 1). The increase in water deficit under the action of melatonin in the control variant may indicate a greater saturation of tissues with metabolites.

Table 1 - Effect of seed dehydration and melatonin treatment on water deficit of embryo tissues of group 1 (1) and group 2 (2) 18-hour-old pea seeds grown on water or Hoagland 1/2 medium

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Variant/germination medium	Water deficit (%)	
	Water (group 1)	Hoagland's medium ( group 2)
Control	$8.68 \pm 1.11^a$	$8.09 \pm 1.32 a$
+ melatonin	$9.37 \pm 1.28 a$	$21.02 \pm 1.22 b$
Dehydration	$20.25 \pm 1.13^b$	$16.82 \pm 1.13 c$
Dehydration + melatonin	$21.59 \pm 1.22 b$	$22.21 \pm 1.2 b$

Note: the table shows the arithmetic mean values and their standard errors. Statistically significant differences between mean values were determined using Tukey test in ANOVA programme, results marked with different letters are statistically significant at  $P \leq 0.05$ . Water deficit of embryo tissues germinated on water or Hoagland's medium ½ (%)

Variant/germination medium Water deficit (%) Water (group 1) Hoagland's medium ( group 2) Control  $8.68 \pm 1.11^a$   $8.09 \pm 1.32 a$  + melatonin  $9.37 \pm 1.28 a$   $21.02 \pm 1.22 b$  Dehydration  $20.25 \pm 1.13^b$   $16.82 \pm 1.13 c$  Dehydration + melatonin  $21.59 \pm 1.22 b$   $22.21 \pm 1.2 b$

##### 3.2. Respiration rate

In embryos of group 1 under the influence of dehydration and seed treatment with melatonin, a decrease in respiration rate (Vt), activity of cytochrome and alternative pathways of mitochondrial oxidation was observed. On the contrary, the combined effect of melatonin pretreatment and dehydration caused an increase in respiratory activity in group 2 embryos due to V cyt and Vres (Table 2).

Table 2 - Effect of seed dehydration and melatonin treatment on embryo respiration of group 1 (1) and group 2 (2) of 18-hour-old pea seeds grown on water or Hoagland 1/2 medium

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Germination medium	Respiration rate (nmol O/(min g dry w))							
	Water (group 1)				Hoagland's medium (group 2)			
Variant / respiration pathway	V <sub>t</sub>	V <sub>cyt</sub>	V <sub>alt</sub>	V <sub>res</sub>	V <sub>t</sub>	V <sub>cyt</sub>	V <sub>alt</sub>	V <sub>res</sub>
Control	5806 ± 170 a	3574 ± 170 h	1562 ± 125 m	671 ± 150 t	5869 ± 160 a	3304 ± 150 i	917 ± 140 n	1648 ± 140 y
+ melatonin	4515 ± 150 b	3347 ± 150 h	291 ± 120 o	873 ± 140 x	6881 ± 150 e	4583 ± 150 j	557 ± 150 p	1741 ± 140 y
Dehydration	7275 ± 210 c	4710 ± 150 b	1070 ± 120 r	1495 ± 130 z	4161 ± 150 f	2602 ± 150 k	1214 ± 130 r	346 ± 110 z-1
Dehydration + melatonin	3769 ± 150 d	2734 ± 140 k	281 ± 140 s	754 ± 130 z-3	8675 ± 160 g	5398 ± 130 l	1158 ± 140 r	2120 ± 140 z-2

Note: the table shows the arithmetic mean values and their standard errors. Statistically significant differences between mean values were determined using Tukey test in ANOVA programme, results marked with different letters are statistically significant at  $P \leq 0.05$ . Water deficit of embryo tissues germinated on water or Hoagland's medium 1/2 (%)

### 3.3. The TBARS content

The TBARS content in control tissues of group 2 embryos was 4 times higher than in group 1 embryos, but no differences were found during dehydration (Table 3). These values indicate that the more vigorously growing embryos of group 2 had an increase in oxidative activity caused by stimulation of cellular metabolism by contact with air oxygen, as noted by many authors [3]. Dehydration increased TBARS content in group 1 embryos by 63%, whereas it decreased 2.4-fold in group 2 embryos, which can be explained by dehydration-related inhibition of metabolism, in particular inhibition of respiration, observed in group 2 embryos. Pretreatment with melatonin reduced TBARS content in all cases.

Table 3 - Effect of seed dehydration and melatonin treatment on TBARS content in tissues of seed embryos of group 1 (1) and group 2 (2) 18-hour-old pea seeds grown on water or Hoagland 1/2 medium

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Variant/germination medium	TBARS content (nmol/g dry w)	
	Water (group 1)	Hoagland's medium (group 2)
Control	176 ± 11a	704 ± 13 c
+ melatonin	151 ± 10 a	258 ± 9b
Dehydration	287 ± 10 b	297 ± 8 b
Dehydration + melatonin	186 ± 11 a	235 ± 7 d

Note: the table shows the arithmetic mean values and their standard errors. Statistically significant differences between mean values were determined using Tukey test in ANOVA programme, results marked with different letters are statistically significant at  $P \leq 0.05$ . Water deficit of embryo tissues germinated on water or Hoagland's medium 1/2 (%)

### 3.4. Morphometric indices

Morphometric indices of epicotyl length and weight of seedlings grown from seeds of group 2 decreased under dehydration conditions and significantly increased under melatonin pretreatment, both in control and dehydration conditions, whereas in seedlings from seeds of group 1 there was only a tendency to increase epicotyl size and seedling weight under the influence of melatonin pretreatment, as the differences were not significant (Table 4). In control seedlings of group 1, dehydration reduced epicotyl length more markedly than in variant 2.

Table 4 - Effect of seed dehydration and melatonin treatment on morphometric indices of 5-day- old seedlings of group 1 (1) and group 2 (2), grown on water or Hoagland 1/2 medium

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Germination medium	Water (group 1)			
Variant/subject	Control	+ melatonin	Dehydration	Dehydrtion + melatonin.
Epicotyl, length mm	41.7875 ± 1.3557 a	42.5375 ± 1.515 a	32.7778 ± 2.1561 b	35.6456 ± 2.0600 b
Sprout, weight g	0.4648 ± 0.0092 g	0.4696 ± 0.0076 g	0.4746 ± 0.0095 g	0.4763 ± 0.0073 g
Germination medium	Hogland's medium ½ (group 2)			
Variant/subject	Control	+ melatonin	Dehydration	Dehydrtion + melatonin
Epicotyl, length mm	59.3 ± 1.81 c	66.2 ± 1.78 d	57 ± 2.2569 e	69.4375 ± 1.097 f
Sprout, weight g	0.5699 ± 0.0075 h	0.5941 ± 0.0071i	0.5584 ± 0.0082 j	0.6034 ± 0.0063 k

Note: The table shows the arithmetic mean values and their standard errors. Statistically significant differences between mean values were determined using Tukey test in ANOVA program, results marked with different letters are statistically significant at  $P \leq 0.05$ . Water deficit of embryo tissues germinated on water or Hogland's medium ½ (%)

## DISCUSSION

Of all the systems formed in the developing embryo, the most important for the further condition of the seed is the main energy-supporting process, respiration, which provides the embryo tissues with high-energy substrates already from the first minutes of seed soaking, depending on the rate of water penetration into the seed [4], [29], [30]. The role of mitochondria for seed recovery after exposure to unfavorable conditions was reported by Wang et al. [24]: only in the case of complete restoration of mitochondrial structure and function, seed recovery after the action of an unfavorable factor is possible. The results of the present work showed that even short-term dehydration inhibited energetically more efficient respiration through the cytochrome pathway in seeds of group 2, which in this case can be considered as a positive factor, since inhibition of oxidation through the cytochrome pathway of respiration seems to protect plants from excessive expenditure of energy and metabolites under conditions of inadequate water supply. Pretreatment with melatonin relieved this inhibition, possibly because melatonin promoted an increase in tissue mineral content in the case of Hoagland's medium, markedly activating seedling growth (Table 4). At the same time, melatonin showed itself as an antioxidant, reducing the level of lipid peroxidation in both control and dehydrated embryos, which can be judged by the reduction of TBARS level in tissues (Table 3).

In group 1 embryos, dehydration stimulated energetically more efficient cytochrome respiration, which in the absence of water supply can be dangerous because it leads to excessive carbohydrate and energy expenditure. Pretreatment with melatonin reduced the rate of Vcyt but decreased the maximal Valt activity (Table 2). It is known that the alternative pathway of mitochondrial oxidation catalysed by alternative oxidase may play an important regulatory role by eliminating the imbalance between carbohydrate supply and demand [31]. As shown in Sieger et al. [32], under conditions of P and N deficiency in the growth medium of tobacco cells, Valt activity increases significantly. It is turned on to enhance its regulatory function as a factor modulating growth in response to nutrient availability and plays a specific role in maintaining cellular redox and carbon balance [31]. It has been shown that in the absence of such a mechanism, an imbalance between growth and nutrient supply can develop. Based on these data, it can be assumed that melatonin, by reducing Valt activity in seedlings of group 1 under dehydration conditions, negatively affected the balance of nutrients in seedlings of this group, whereas the maintenance of Valt activity level under the action of melatonin in seedlings of group 2 was a factor contributing to the rational utilisation of plant nutrients, as well as a factor of antioxidant nature. The latter aspect is also very important for the normal development of seedlings, because, as we noted above, in embryos exposed to air oxygen, there is an increase in the level of AFCs in tissues due to metabolic activation and excess oxygen [9]. Indeed, under the influence of melatonin pretreatment, the TBARS level in tissues decreased in seed embryos of group 2 both in control and after dehydration, whereas in seed embryos of group 1 it decreased only under dehydration conditions (Table 3). Also, melatonin effectively influenced the parameters of growth and weight of seedlings, simultaneously increased or decreased the activity of the alternative pathway respiration of embryos, as well as decreased the level of TBARS content in embryo tissue both under dehydration and control conditions, but only when plants were grown on Hoagland's solution. It becomes evident that the significant increase in respiration rate of group 1 seedlings under dehydration was a negative factor, requiring excessive expenditure of energy and metabolites. Although melatonin was shown to partially neutralize the increase in respiration rate in the embryos of this group under dehydration conditions, this inhibition did not compensate for the substrate wasting under unfavorable conditions, but, on the contrary, lowered the energy potential of the embryo tissues, since, as we assume, the lack of ions necessary for further active growth supplied to the embryos in the case of cultivation on Hoagland's medium was decisive.

The results obtained indicate a positive effect of melatonin on respiratory metabolism, epicotyl growth and embryo weight under the conditions of using Hoagland's medium rather than distilled water. The ideas about melatonin regulation of tissue ion

homeostasis seem to be supported by these results [33], [34]. This leads us to suggest that, indeed, under conditions of cultivation on Hoagland medium, melatonin is more fully expressed as a regulator of metabolism under unfavorable conditions. It should be borne in mind that, as Kolodziejczyk et al. [4] showed, a pool of proteins protecting membranes and cytoplasmic proteins from the effects of unfavorable factors, as well as proteins of transport and energy metabolism, is formed already in the first hours of embryo swelling. At the same time, in *Zea mays* seeds under hydropriming conditions, and especially when melatonin 50  $\mu$ M was added to the seed pretreatment medium, the synthesis of a pool of proteins related to protective proteins (Lea proteins, HSP), a number of antioxidant, detoxifying and chaperone proteins, transporter proteins, and energy metabolism proteins was additionally stimulated in 1-day-old seed embryos [4]. Interestingly, in the absence of direct stress effects, seed embryos are enriched at the early stages of swelling by a system that protects plants from possible unfavorable effects. At the same time, it was shown that the mentioned protective proteins protecting from the action of unfavorable factors – heat, dehydration – are preserved in the tissues of pea seedlings for 3 days [35], [36]. These data confirm the effectiveness of early seed treatment with stimulants, in this case melatonin, but also draw our attention to the fact that the implementation of the program laid down by such treatment requires an increased content of proteins of different spectrum of action, from proteins of energy metabolism, to transporters and protective proteins that allow to survive adverse effects, as well as a rise in the level of ROS, arising at the moment of germinal roots emergence from the seed coat and associated contact with air oxygen, and a significant increase in the level of ROS, which occurs at the moment of the germinal roots exit from the seed coat and the associated contact with air oxygen and significant metabolic stimulation [3]. Such a program may be more successful in the presence of additional supply of mineral compounds contained in Hoagland's solution, as shown in our work. The failure of melatonin action in some cases may be associated not only with poorly selected melatonin concentration, but also with insufficient reserve of metabolites, including metal ions and other mineral elements for accelerated development of seedlings, as well as for mitochondrial biogenesis during seed germination.

### Conclusion

Thus, in addition to the known concentration dependence of melatonin's effect on plant viability mentioned earlier (see INTRODUCTION), there is obviously a dependence of the hormone's action on the readiness for efficient realisation of all currently available resources especially at the early stage of growth and development of the plant organism. In this regard, it can be assumed that under the conditions we used, melatonin had a negative effect on weakened seedlings, but supported more energetically and substrate-provided plants. The mechanism of action of the hormone, including its effect on the transport of ions and other mineral substances in plants under different conditions, remains to be revealed. Great prospects here may belong to studies in the field of respiratory metabolism, since it is obvious that melatonin actively interferes with the work of components of the mitochondrial respiratory chain that provide energy for the processes of transport and accumulation of substances in plants. On the other hand, as our (and not only our) studies have shown, the respiratory chain is highly sensitive to external influences. Therefore, it is possible to consider the noted interactions as one of the mechanisms of melatonin influence on plants. It is very early – already after 18 h of seed swelling allows us to evaluate the effectiveness of melatonin's effects. Exerting the influence of phytohormones, including using the priming method, on swelling seeds, it is possible to obtain an effect on the growth and development of seedlings, as well as on their resistance to unfavorable environmental factors. This allows us to consider phytomelatonin as one of the promising and multifaceted stimulants, one of the features of the effect on plants of which have been presented in this paper.

### Конфликт интересов

Не указан.

### Рецензия

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### Conflict of Interest

None declared.

### Review

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