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Nutritional and Physicochemical Effects of Photoperiod and Germination Time in Oat Sprouts as Food

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ABSTRACT

The present work aims to explore the impact of the photoperiod and germination time on the quality of oat sprouts for food consumption. To that purpose, oat (*Avena sativa* L.) sprouts were germinated from seeds at two different photoperiods (12 and 24 h of exposure to light) throughout nine days. Samples of germinated oat sprouts were analyzed for changes in nutritional and physicochemical composition, *viz*. vitamin C (ascorbic acid), carotenoids, vitamin B3 (niacin), plumule height, dry-matter, dietery fibers,

total oil and total protein content. When comparing with the original oat seeds, significant increase in the content of vitamins, carotenoids and dietery fibers, and a decrease in dry matter was observed in germinated oat sprouts over the time. Results showed that both the photoperiod and germination time had a significant impact on the analyzed parameters. The germinating regime of 12 h light per day for 9 days proved to be the most appropriate for obtaining oat sprouts with the highest value of carotenoids and vitamin B3 and the second highest value of vitamin C. The germinating regime of 12 h light per day for 9 days also the highest mean plumule height response was obtained from treatment 3 with an average of 7.1 cm.

Keywords: Oat sprout; oat shoot; nutritional parameter; vitamin; carotenoid, protein

INTRODUCTION

Cereal grains are among the earliest food resources for humankind and have become an inseparable part of human diet worldwide, being the raw material for the production of a wide variety of cereal products. In recent years, the use of seed sprouts has become widespread in the diet of the western population due to the increasing interest of modern consumers to minimum processed and natural food, as well as the rising adherence to vegetarian and vegan diets in pursuit of a healthy lifestyle.

The nutritional quality of sprouts is well acknowledged since they present a good source of macronutrients, such as dietary fibre, proteins and vitamins, and have low calorie content. Several nutritionally beneficial effects occur

during seed germination - bioavailability of the major nutrient groups increases, while the activities of toxins and enzyme inhibitors decrease. Germination is also fundamental to increase seeds digestibility [1,2,3]. Furthermore, sprouts are recognized by their high antioxidant capacity, reducing the negative effects of free radicals [4, 5,6, 7,8]. Some studies have reported that the amount of crude protein increases along with germination [8,9,10,11] while others observed the opposite trend [12,13,14,15].

Seed sprouts are used in many different ways in human nutrition. They can be consumed fresh or after storage under refrigeration or freezing conditions. Sprout production mostly resorts to the plant families of Gramineae and Leguminosae or Fabaceae. Legumes and cereals are high in fiber and protein content, and low in fat [3,16].

Recent studies have demonstrated the benefits of consumption of oat (and other cereals) regarding reducing the risk from cardiovascular diseases by lowering serum low-density lipoprotein (LDL) cholesterol levels in humans. Several studies have claimed the positive effects of oat against heart disease risk factors such as diabetes, obesity and hypertension [4,5,18,19]. Particularly, cereal sprouts have received considerable attention in some countries such as Japan due to their nutritional and health attributes when compared to the corresponding cereal seeds [9, 20,21].

Various germination strategies can be adopted to improve the nutritional composition of sprouts including temperature, salinity, photoperiod and germination time [3, 17]. Among cereals, oats are characterized by high

protein and lipid content [22,23,24,25]. The nutritional value of oat sprouts may vary according to their composition in bioactive compounds, minerals and other nutrients. In addition, the type and varieties of seeds, processing conditions (e.g. time, temperature, light duration and relative humidity) and storage conditions have impact on the final quality of sprouts [16,26,27,28]. Especially light and light duration are very important for germination and growth and therefore sprout quality. Light is a critical abiotic stimulus for plants, shaping growth and development both directly and indirectly over large scales. Besides being the energy source for photosynthesis, light is perceived as a signal by photoreceptors, regulating growth, differentiation and metabolic processes. The literature states that the minimum light exposure time for sprout cultivation is 12 hours [29,30,31]. Therefore, in this study, light exposure times of 12 and 24 hours were also determined.

Overall, germination is a cost-effective and environmentally safe approach for cultivation of seeds and obtaining sprouts in a short period of time. Sprouts are rich sources of health-promoting phytochemicals compared to seeds and their nutritional composition could be further improved by elucidating the effect of significant factors and their interaction [22,32,33]. However, the variety of sprouts for human consumption on the market is still quite limited [34].

Based on the valuable nutritional characteristics of oats and their health benefits, it was selected as the raw material for the present study, which aimed to explore the impact of the light hours and germination time during

the production of oat sprouts for food consumption in order to identify the most favorable conditions for obtaining a high-quality product [35,36]

MATERIALS AND METHODS

Materials

Oat (*Avena sativa* L.) seeds of Kirklar variety were provided by Trakya Agricultural Research Institute Edirne (Edirne, Turkey).

The oat seeds were harvested in September of 2019 and stored until further use in light-protective plastic bags under controlled conditions in storage room at 25ºC and 80% relative humidity. Stored oat seeds were protected from light. The experiments with stored oat seeds (approximately 120 days of storage) were undertaken between June and September of 2019 at the Laboratory of Çanakkale Onsekiz Mart University (Çanakkale, Turkey).

All chemicals were purchased from Merck (Darmstadt, Germany) and were of analytical grade.

Experimental design and sampling

The experimental design and conditions used in this study are presented in Table 1. Oat seeds were subjected to six different treatments (T1 to T6) using two different photoperiod (12 and 24 light hours per day) and analyzed every 3 days throughout a total germination time of 9 days. Oat seeds at the beginning of the germination processes (T0) were also analyzed. A factorial design in randomized blocks with 3 analytical repetitions was employed. Applications are explained below.

T0 : control

T1 ; Half 3 days, 12-hour light

T2 : Half 6 days, 12-hour light

T3: Half 9 days, 12-hour light

T4: Complete 3 days, 24-hour light

T5: Complete 6 days, 24-hour light

T6 : Complete 9 days, 24-hour light

Germination conditions

Prior to the germination assays, oat seeds were subjected to a manual soaking in 500 ml of sterilized distilled water for 30 minutes. The supernatant was discharged and the soaked seeds were sterilized by keeping them in a 2000 ppm aqueous solution of calcium hypochlorite [$Ca(OCl)_2$] for 5 minutes [36,37] and then washed with sterilized distilled water using a ratio of 500 ml water per 20 seeds. The 20 seeds were carefully arranged on the filter papers within each petri dish. Additionally, all petri dishes were encased within closed transparent plastic bags to mitigate evaporation.

In each treatment (T0 to T6), twenty soaked and sterilized oat seeds were chosen at random and placed in standard Petri dishes (60mm indiameter,15mm height) mm-ϕ (Sigma-Aldrich). Seed germination was carried out in a plant growth chamber (Nucleon-NBB/Europe, Germany) throughout 9 days at controlled temperature of (20±1)°C and without regulation of the relative humidity. A visible white light source was used with light intensity of 17000 lux. A timer coupled to the power source of the lamps was used to allow the photoperiods (light/dark) of 12 h per day to change in

treatments T1 to T3 (Table 1). A visible white light source was used with light intensity of 17000 lux. A timer coupled to the power source of the lamps was used to allow the photoperiods (light) of 24 h per day to change in treatments T4,T5 and T6 (Table 1). A volume of *ca*. 50 mL sterilized distilled water was added to the Petri dishes twice a day. Samples were taken after 3, 6 and 9 days of germination and subjected to physicochemical analyses. The same analyses were performed for the oat seeds without any treatment (T0).

Physicochemical analyses

The height of plumule, defined as height, in centimeters, was measured with a ruler from the epicotyl to the growing point of ten randomly chosen sprouted seeds, for each treatment(T0-T6).

Dry-matter (drying to constant weight at 100ºC, measured gr) of the oat seeds and sprouts was determined using the official standard gravimetric method according to American Association of Cereal Chemists [33]. For each treatment, after their fresh weight were determined dry weights of the sprouts were determined following 48 hours drying period at 100ºC. Total fiber content was determined using the standard enzymatic method as described in Association of Official Analytical Chemists (AOAC) 991.43 [29]. It used 80% ethanol and some enzymes (protease and amylglucosidase). Total protein (Nx5.7) was determined according to AACC standard method 46-12.01[21]. The protein mass fraction (Kjeldahl method) was determined by Kjeldahl distillation unit. Total oil was determined according to AACC 30.26-

01by Soxhlet extraction with petroleum eter [21]. After extraction the oil was determined by weighing.

Dry-matter, fiber, total oil and total proteins were expressed as weight percentage on a dry basis (%, w/w).

Vitamin C content was analysed according to the method described by Pearson (1, 25) with minor changes. Vitamin C content was determined spectrophotometrically at 520 nm by using a standard curve according to the procedure described by Pearson. Total carotenoids were determined using the AOAC spectrophotometric method described by Lichtenthaler [16,24]. 5 gr frozen sprouts grounded and then they extracted with hexane (16). Total carotenoids content was determined by the spectrophotometer UV -120 IV Shimadzu, using the wavelength 517nm [16]. Niacin content was analysed using a method AACC 86-80 [33]. Total niacin was determined by HPLC-UV: C18 column (using the wavelength of 264 nm). Vitamins C and carotenoids were expressed as milligrams per 100 grams of sample on a dry basis (mg/100g).

All experiments were done in 3 repetitions.

Statistical Analysis

Descriptive statistics for each dependent variable – *i.e*. the nutritional (vitamins C, niacin and carotenoids) and physicochemical (plumule length, dry matter, edible fiber, total oil and total protein) parameters were calculated. The data were first tested for constant variance assumption using the Levene's test, and it was found that the variance was constant across the

7 groups, *i.e*. no treatment (T0) and 6 treatments (T1 to T6). Consequently, factorial analysis of variance (ANOVA) was performed for each output (Table 2), *i.e*. distinct treatments of photoperiod (12 and 24 h light per day) and germination time of 3, 6 and 9 days. In order to understand the different effect, parameter eta-squared (η^2) was calculated for both half and complete light conditions after performing two-way ANOVA (Table 2 qnd supplemantery file). Tukey-HSD test for the effect of day on half and complete photoperiod treatment data was performed separately to compare the means and indicate significant differences (Table 2 and supplemantery file). An α-value of 0.05 was used as reference for the statistical analyses, thus whenever P-values were equal or below 0.05 the means were considered statistically different. All statistical procedures were performed in R studio [26, 34].

RESULTS AND DISCUSSION

The profiles of the vitamin and carotenoid content during germination of oat seeds under the two different photoperiod conditions are presented in Figure 1, and the profiles of the remaining analyzed physicochemical parameters are shown in Figure 2.

Descriptive statistics for each dependent variable (vitamin C, carotenoids, niacin, plumule height, dry-matter, edible fiber, total oil, and total protein) were calculated (Table 2). The results from Levene’s test proved that the variance was constant across the seven groups. The statistical significance in all compared combinations based on the above experimental results and

using the 2-way ANOVA, is presented in Tables 1 and Table 2. (available as also S1- S3 “Supplementary material”).

Vitamin C, carotenoid and Niacin content in oat seeds and sprouts

Vitamin C is present in plants as a result of its biosynthesis reactivation during germination (1, 27). Oat sprouts showed a significant increase in vitamin C when compared to seeds (Table 2, Figure 1). The highest value of vitamin C concentration was obtained in Treatment 2 (Table 1) with an average of 351 mg/100 g (Table 2, Figure 1). Nutritional composition of common oat (*Avena sativa* L.) variety Auron and naked oat (*Avena nuda* L.) variety Abel were examined by Gabrovska et al., [12] after 4, 6 and 8 days of germination. Their highest vitamin C content was detected after 6 days of germination for both varieties and it was above 500 mg/100 g (1). No vitamin C was detected in buckwheat dry seeds compared to 6 days germinated buckwheat sprouts with vitamin C ranging from 50 to 60 mg/100 g. Dry wheat grains (*Triticum aestium*) showed similar vitamin C range after 5 days of germination (5.81 *versus* 5.18 mg/100 g [4]. Vitamin C content of cowpea sprouts after only 1 day of germination increased 4.38 times compared to the seeds [27]. The vitamin C content differences observed between our results and those found in the literature are mainly due to plant species specificity and the different germination periods applied.

A significant increase in carotenoid content was also observed in oat sprouts compared to the dry grains (Table 2, Figure 1). The highest value of carotenoids concentration was obtained for Treatment 3 (Table 1) with an

average of 490 mg/100 g (Table 2, Figure 1). Some researchers examined the physicochemical composition of two oat varieties after 4, 6 and 8 days of germination [15]. Like our results, they detected the highest carotenoid content after 8 days of germination for both varieties with approximate amounts above 500 mg/100 g. In another study, after 7 days of germination in dark conditions, buckwheat sprouts exposed to salinity stress generated carotenoid content of about 50 mg/100 g, which was twice as high than the control [17]. Like the comparison of vitamin content, differences in carotenoid content observed in our study compared to other research reports can be mainly attributed to plant species specificity.

Niacin content (vitamin B3) in oat sprouts also showed a significant increase compared to the dry grains (Table 2, Figure 1). The highest value of niacin concentration was obtained in Treatment 3 (Table 1) with an average of 375 mg/100 g (Table 2, Figure 1). Some researchers found the highest niacin concentration after 8 days of germination for both varieties of tested oat with approximate content of 350-400 mg/100 g[12]. Another study performed with dry wheat grains (*Triticum aestium*) showed lower niacin content (72.8 mg/100 g) in 5-day germinated sprouts [3]. The differences observed in niacin concentration between our findings and those in the literature were mainly attributed to plant species followed by photoperiod application and germination time.

Our results showed significant increases in the amounts of vitamins (C and niacin) and carotenoids in oat sprouts (treatments T1 to T6) compared to the

dry grain (T0) in both light conditions, *i.e*. 12 and 24 light hours per day (Table 2, Figure 1). These findings were also demonstrated in Table 2 when comparing the great averages per type of treatment, *i.e*. light hours per day and germination days, as well as maximum and minimum observed mean values.

Effect of photoperiods during germination time on vitamin C, carotenoid and niacin content

Results presented in Table 2 and Figure 1 display a significant decrease in the amount of vitamin C between 6 and 9 days of sprouting, from *ca*. 351 to 300 mg/100g and from *ca*. 210 to 179 mg/100g when using a photoperiod of 12 and 24 h per day, respectively. These findings are in agreement with those observed in previous studies [20,28,19] where the amount of vitamin C was demonstrated to increase 3.35 folds with the advent of the seed sprout germination. On the other hand, the amount of niacin and carotenoids (Table 2, Figure 1) increased continuously over the 9 days of germination in both photoperiod conditions. The great averages tabulated in Table 2 make it apparent that unlike vitamin C, niacin and carotenoids are higher when a photoperiod of 12 h is used. In addition, data in Table 2 highlights the accumulation of vitamins and carotenoids throughout the entire period of 9-day-old sprout germination.

The observed decrease in vitamin C between 6 and 9 days may be attributed to the metabolic activity of seeds during the germination and growth processes, but it can also be a consequence of environmental

oxidation including the result of photo-oxidation. The amounts of vitamin C in germinated seeds are fairly high (Figure 1, Table 2), emphasizing the importance of oat sprouts as a significant source of this nutrient. It is reported that the amount of water-soluble vitamin C in seeds germinated for 150-162 h (approximately 6 days) can provide 10% of the recommended human daily intake, which is around 50 mg [20].

Niacin (vitamin B3) is a key nutrient well known for its health attributes, and niacin deficiency is the cause of a disease – pellagra [20]. Oat sprouts prove to be a valuable source of niacin, since increase of niacin during sprouting is observed [7]. Our results showed considerable amounts of niacin in the oat sprouts obtained by the different treatments (Figure 1, Table 2). When comparing the effect of time of germination on the content of vitamins and carotenoids (Table 2, Figure 1), for both photoperiods applied it was observed that 6 days of germination is a preferable period to reach the highest values of vitamin C, while 9 days of germination result in highest amounts of niacin and carotenoids. A significant increase in the vitamin content in grains due to sprouting was also reported in many other studies [38].

Effect of photoperiods on vitamin C, carotenoid and niacin content

When comparing the samples with the same time of germination but subjected to different photoperiods (*i.e.* T1-T4, T2-T5 and T3-T6) (Table 2, Figure 1) it is apparent that the use of a daily photoperiod of 12 h is preferable than the use of continuous light to reach higher values of vitamin C and

carotenoids. However, no significant effect of light time was observed for vitamin B3 (niacin) content. These conclusions are based on the comparison of the great averages for 12 and 24 h light per day (Table 2).

Physicochemical characteristics of oat seeds and sprouts

Results from the observations of several physicochemical parameters, *viz*. plumule height, dry-matter, edible fiber, total oil and total protein content after different treatments (T1 to T6) of oat seeds are presented in Table 2 and Figure 2. The increase of the plumule height and the decrease of dry-matter result from the sprouting process as a consequence of water addition twice a day (see Materials and Methods) to trigger and develop the sprouting process. On the other hand, a noticeable increase of the total edible fiber was observed as the germination progressed. Regarding the total oils (lipids) and total proteins, the results disclosed some slight variations but with a general trend to remain quantitatively stable. Nevertheless, in comparison with the raw seeds (T0), the results of total oils and proteins showed some tendency to a slight decrease and increase, respectively, as a result of the metabolic activity during germination.

Effect of photoperiods on the physicochemical parameters over time

Table 2 and Figure 2 revealed significant differences in the plumule height during germination in both conditions of photoperiod, particularly between days 3 and 9. However, the differences in plumule height between each consecutive three days of germination were not always significantly different under both photoperiods. In terms of plumule height, a growth of *ca*. 37%

over the 9 days of germination were observed. The highest plumule height of 7.1 cm was obtained from Treatment 3 (T3, 12-hours/9 days) (Table 2, Figure 1). In comparison, plumule in various cowpea genotypes reached a maximum length (chiefly, 2 25-3.32 cm) after 4 days of germination [27].

With the advancing germination period, dry-matter decreases due to respiration (30) which was also apparent in our findings. As shown in Table 2, no significant differences in dry matter content were observed in consecutive sampling days (T1-T2 and T2-T3, or T4-T5 and T5-T6) but for the whole period of 9 days of germination the dry-matter decreased *ca*. 50% (Figure 2).

Oat sprouts showed significant increase in edible fiber composition compared to the dry seeds (Table 2, Figure 2). The highest concentration of edible fiber was obtained from treatment 6 (24-hours/9 days) with an average of 35% (w/w). In another study, the highest dietary fiber was detected after 8 days of germination with 26.8% for naked oat Abel and 35.1% for common oat Auron[28]. Additionally, significant increase in crude fiber content was observed for sprouts of various cowpea genotypes compared to seeds [27]. Some researchers reported an increase of the total dietary fiber content throughout germination carried out both with and without daylight [19,31,32].

Our analysis for total edible fiber showed an increase over the germination time (Figure 2 and Table 2). However, between consecutive samplings of 3

days of germination (*i.e.* 1 to 3, 3 to 6 and 6 to 9 d) the differences were generally not statistically different.

Decrease in oil content could be due to degradation of stored oil to provide energy and trigger catabolic activities as protein synthesis during germination [27]. Dry grains showed significantly high oil content (9.4%, w/w) compared to the oat sprouts (Table 2, Figure 2). The highest oil content was detected in dry grains (compared germination time) with 9.6% (w/w) for naked oat Abel and 8.4%, w/w for common oat Auron by researchers [14]. Significant decrease in oil content was also observed for sprouts of various cowpea genotypes compared to seeds [17,27].

The increase of protein concentration could be related to dry-matter loss (especially carbohydrates) through respiration during germination [17,27]as it is also supported by our findings. Oat sprouts showed significant increase in protein content compared to the dry seeds (Table 2, Figure 2). The highest total protein response was obtained from Treatment 2 (Table 1) with an average of 14% (w/w) (Table 2, Figure 2). Other researchers reported the highest protein concentration after 6 days of germination for naked oat Abel and 4 days of germination for common oat Auron with about 15% (w/w)(28). Cowpea sprouts showed 9-12% protein content increase after 1 day germination period compared to seeds [27].

Two-way ANOVA showed that the main effects for day and light factors were highly significant for all responses (p-value < 0.001). The light effect was ineffective only on total oil content (p-value >0.05) (Table 2 and Tables

S1-S3). The interaction effect for light and day inputs were highly significant for vitamin C, carotenoids, niacin, plumule height, edible fiber, and total protein outputs (p-value <0.001) and less significant for total protein (p-value <0.05), and with no effect on total oil (p-value >0.05) (Tables S1 and S3). Eta-squared (η^2) analysis was conducted to define the effect of germination time on the tested responses. For 12-hours photoperiod 99.9% of the vitamin C variance was explained by the germination time variable.

Eta squared is the proportion of the total variability in an outcome that is related to the membership of various groups defined by an input. Partial eta squared is the variance explained by a specific factor where the other independent variables main and interaction effects are excluded [8]. The day factor had a very high effect size on the tested responses under both 12-hours and 24-hours photoperiods with partial eta squared above 99% for most of the outputs. Under 12 hours of light, 99.9% of the vitamin C, carotenoids, niacin, plumule height and dry-matter, as well as 99.6% of the edible fiber and 98.4% of the total oil variances were explained by the germination time (days) as a factor (p-value < 0.001). The germination time input also accounted for 65.9% of the total protein output variability under 12 hours of light (p-value < 0.05) (Tables S1 and S3). Under 24 hours of light, 99.9% of the vitamin C, carotenoids, niacin, plumule height and total oil, as well as 99.7% of the dry-matter and 99.8% of the edible fiber variances were also attributed to the germination time factor (p-value < 0.001). The total protein

variability of 78.4% was explained by the germination day input under 24-hours light (p-value < 0.05) (Table 2).

Effect of photoperiods on physicochemical parameters

Under 12-hour light conditions, all of the germination time mean difference pairwise comparisons were highly significant except for total protein content (Table 2), where only the mean difference between days 9-6 was statistically significant (p-value < 0.05). These results are compatible with the findings in previous studies [20,28].

When applying 24-hour light treatment, almost all of the germination time mean difference pairwise comparisons were highly significant (p-value < 0.001) (Table 2). For total protein content, the mean difference between days 9-6 was insignificant (p-value = 0.68), and the mean difference between the other day groups was significant at p-value < 0.05. These results are also in agreement with data reported by other researchers [1,20,28].

When comparing the total edible fiber with the same days of germination but at different photoperiods, Figure 2 and Table 2 revealed that sprouts with 24h light have significantly higher values. Such results are also apparent when analyzing the great averages in Table 2 and may be attributed to the effect of light on tissue aging.

Finally, when comparing total oil and protein contents in sprouts with the same days of germination but under different photoperiods, Figure 2 and Table 2 show a substantial effect of light in these parameters. However, it

must be stressed that when comparing with oat seeds, the amount of total oil shows slight decrease, opposed to increase of proteins.

Main effects and their interaction

The ANOVA factorial analysis allowed to verify that the two main factors (hours of light and time of germination) as well as their interaction were highly significant for all the parameters studied except dry matter content.

CONCLUSIONS

The present study compared the levels of nutritional and physicochemical parameters of oat sprouts resulting from different germination conditions, *viz*. time of germination (up to 9 days) and type of photoperiod (12 h and 24 h). The results revealed that both factors had a major impact on the quality of the final oat sprouts for food consumption. The continuous use of light during germination of oat seeds proved to be less appropriate than the alternation between day and night (*i.e*. the use of a photoperiod) in the natural catabolism of plant vitamins and carotenoids. Furthermore, the combination of a photoperiod of 12 h and a germination time of 9 days resulted in highest amounts of niacin and carotenoids, whereas the highest values of vitamin C were observed after 6 days of germination. The highest edible fiber content was obtained under 24-hours light after 9 days of germination. Dry matter and total oil were highest in dry seeds compared to sprouts. Germination time had a very high effect on the nutritional composition of oat under both light conditions with partial eta squared above 99% for the outputs. The use of a photoperiod (12 hours of light) and

germination time between 6 and 9 days are recommended to maximize the nutritional quality of oat sprouts.

The current research highlighted the importance of germination as a bio-processing approach in food science and technology to produce sprouts with improved nutrition characteristics as increasingly demanded by contemporary consumers.

Supplementary materials: Figure S1: Graphical representation of Table 1 summarizing a) vitamin C, b) carotenoid, c) niacin, d) plumule height, e) dry matter, f) edible fiber, g) total oil, and h) total protein dependent variables. The red dashed line indicates the treatments with highest mean response.; Table S1: Two-way ANOVA of vitamin C, carotenoid, niacin, plumule height, dry-matter, edible fiber, total oil and total protein responses; Table S2: Eta-squared (η^2) of germination time (day) factor under 12- and 24-hours photoperiods for vitamin C, carotenoid, niacin, plumule height, dry-matter, edible fiber, total oil and total protein responses; Table S3: Tukey-HSD comparing (pairwise day groups) on 12- and 24-hours light conditions for vitamin C, carotenoid, niacin, plumule height, dry-matter, edible fiber, total oil and total protein responses.

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Author contributions

Gül Ebru Orhun was actively engaged in performing analysis, data collection,writing and revision of the manuscript. Meleksen Akın, actively participated in performing analysis and revision of the manuscript. Eda Orhun and J.M. Rocha was actively involved in work design, interpretation, drafting of the article, and final approval of the version to be published. V.Gotcheva and Elena Barkiene was actively involved conceptualism and editing.

Declarations

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Data Availability Statement: The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

Consent to Participate declaration: Not applicable

Ethical approval: Not applicable

Plant guidelines Statement: The plant materials were collected from licensed commercial sources in Türkiye, originating from Trakya Agricultural Research Institute, and were grown under standard cultivation practices. No permits were required for this non-endangered material; all procedures complied with institutional and national guidelines. We confirm that the use of the plant material complied with all relevant local and national guidelines.

Permissions to collect the plants/plant parts: Oat (*Avena sativa* L.) seeds of Kirklar variety were provided by Trakya Agricultural Research Institute Edirne (Edirne, Turkey).

Competing interests: The authors declare no competing interests

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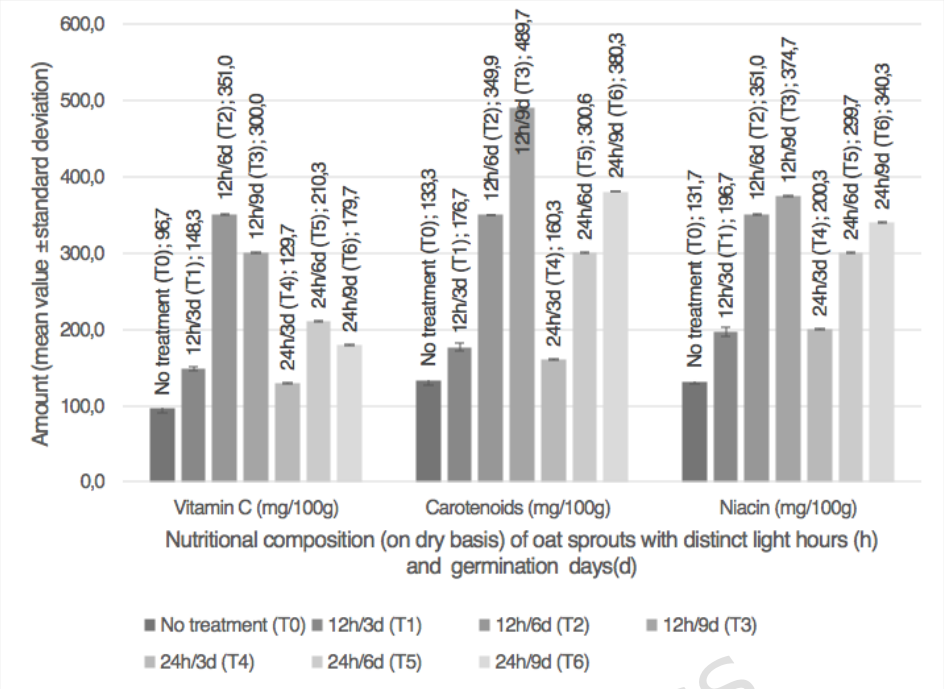

Figure 1. Nutritional composition of oat sprouts (dry basis) obtained after six different treatments (T1-T6) differing by photoperiod/light (12 and 24 h of light per day) and time of germination (3, 6 and 9 days)

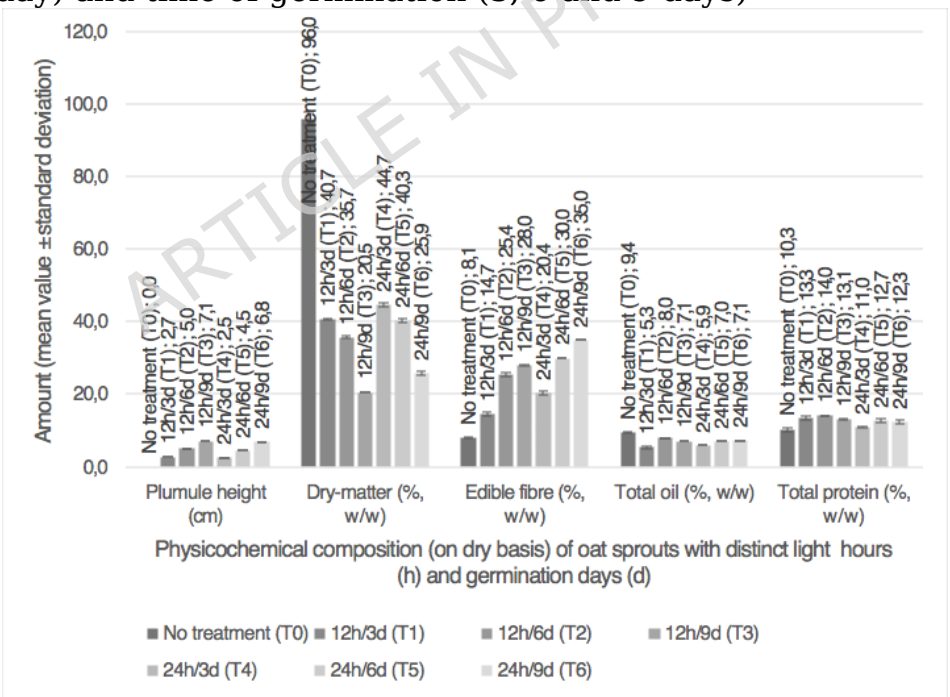

Figure 2. Physicochemical composition of oat sprouts (dry basis), obtained after six different treatments (T1-T6) differing by photoperiod/light (12 and 24 h of light per day) and time of germination (3, 6 and 9 days)

Table 1. Experimental design and conditions of the study

Independent variables / Factors

Designation of the treatment	Photoperiod (light hours per day) (h)	Days of germination (d)	Dependent variables / Observations
No treatment (T0)	0	0	Plumule height, Dry-matter, Edible fiber, Total oil, Total protein, Vitamin C (Ascorbic acid), Carotenoids, Vitamin B3 (Niacin)
12h/3d (T1)	12	3	
12h/6d (T2)	12	6	
12h/9d (T3)	12	9	
24h/3d (T4)	24	3	
24h/6d (T5)	24	6	
24h/9d (T6)	24	9	

Vitamin C				**Carotenoid**			
Factor	**d.f.**	**F**	**P**	**Factor**	**d.f.**	**F**	**P**
Day	3	5371	<0.001	Day	3	10130.3	<0.001
Light	1	6129	<0.001	Light	1	1574.9	<0.001
Day*Light	2	1004	<0.001	Day*Light	2	343.5	<0.001
Niacin				**Plumule height**			
Factor	**d.f.**	**F**	**P**	**Factor**	**d.f.**	**F**	**P**
Day	3	8080.9	<0.001	Day	3	249726.1	<0.001
Light	1	534.9	<0.001	Light	1	3756.1	<0.001
Day*Light	2	189.2	<0.001	Day*Light	2	369.6	<0.001
Dry matter				**Edible fiber**			
Factor	**d.f.**	**F**	**P**	**Factor**	**d.f.**	**F**	**P**
Day	3	6503.69	<0.001	Day	3	3474.22	<0.001
Light	1	168.93	<0.001	Light	1	1109.74	<0.001
Day*Light	2	0.91	**0.425**	Day*Light	2	16.59	<0.001
Total oil				**Total protein**			
Factor	**d.f.**	**F**	**P**	**Factor**	**d.f.**	**F**	**P**
Day	3	612.327	<0.001	Day	3	33.011	<0.001

Light	1	4.446	**0.053**	Light	1	52.007	<0.001
Day*Light	2	58.764	<0.001	Day*Light	2	5.124	**<0.05**

Table 2. Two-way ANOVA of vitamin C, carotenoid, niacin, plumule height, dry matter, edible fibre, total oil, and total protein responses. Factors (Day, Light) and their interactions (Day*Light); d.f., degrees of freedom; F value for comparison with the critical value for significance; P level of significance (p-value).