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Research Article

Developing Sterilization and Lighting Systems for Sprouting Rooms Using Ozone and Optical Fibers

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Abstract: The increasing population has led to the widespread adoption of hydroponics. Hydroponic production of fresh green forage requires minimal space, does not use soil, and allows for rapid harvesting. A fully controlled sprouting room can yield a substantial amount of green fodder from a small area with less water consumption. This study aims to investigate the effectiveness of ozone on seed germination, seedling growth, and microbial sterilization during germinated barley processing. Additionally, the sterilization of the barley sprouting room was conducted using ultraviolet and infrared light, which provides optimal sprouting conditions.The study comprises three experimental variables: three levels of ozonized water (13, 26, and 39 mg L^{-1}) combined with three light sources (fluorescent, infrared, and ultraviolet) and three light duration times (8, 16, and 24 h). The measurements include shoot length, fresh yield weight, dry yield weight, conversion factor, chlorophyll content, N, P, K, crude protein, ash, and log reduction.The results indicated that the maximum values were observed when using ozonized water at 39 mg L^{-1}, Ultraviolet LED as a light source, and a sterilizing medium with a light duration time of 24 h. Conversely, the minimum values were observed when using ozonized water at 13 mg L^{-1}, fluorescent LEDs as a light source, and a light duration time of 8 h. Based on the findings, it is highly recommended to utilize the developed sprouting room throughout the year for the production of fresh forage.

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1. Introduction

By 2050, the world's population is expected to reach approximately 10 billion, with 66% residing in urban areas. This burgeoning population has led to an increasing demand for food and livestock feed (Ghorbel et al., 2021). Traditional farming methods are facing limitations in meeting the growing need for higher quality and quantity of crops (Grigas et al., 2019). As a result, hydroponic systems have emerged as a promising solution to address these challenges. In Egypt, the use of hydroponics to produce high-moisture content forage has garnered significant attention, especially in

arid desert regions where fodder scarcity is a concern (Mariyappillai et al., 2020). Furthermore, climate change has resulted in water shortages, prompting the adoption of hydroponic systems for sustainable organic animal forage production (Sharma et al., 2018). Hydroponics is a soilless method of cultivating plants, enabling the production of fresh green forage from various crops like maize, barley, oats, and cowpeas (Bakshi et al., 2017). Among these crops, barley grains (*Hordeum vulgare L.*) have long been utilized in animal feed (Adiban et al., 2021) and the malting industry (Madakemohekar et al., 2018). Barely sprouts provide animals with essential nutrients, including proteins, carbohydrates, minerals, vitamins, and water. They are grown without soil by germinating seeds in water or nutrients solutions, offering easily digestible and nutritious fodder for livestock. Sprouting facilitates year-round availability of green fodder, conserves water by minimizing evaporation, reduces labor, preserves soil for main crop cultivation, lowers reliance on chemical fertilizers, decreases meat and milk production costs, and ultimately boosts national income.

During sprouting, proteins are converted into essential amino acids, carbohydrates into sugars, and fats into essential fatty acids. Increased enzymatic activity further enhances these conversions, making sprouts easier to digest than dry seeds (Shit, 2019). The hydroponic green forage produced through sprouting takes a period of 8-15 days and requires a small land area (Mooney, 2005; Gebremedhin, 2015). Approximately 6 to 10 kg of fresh green sprouts can be generated from one kg of seeds throughout the year (Kruglyakov, 1989). The resulting forage comprises germinated seeds with intertwined white roots and green shoots, which are entirely consumed by livestock (seed, roots, and shoots). Barley seeds are preferred due to their affordability and widespread availability. The sprouts maintain a crude protein content of 16-17% with more than 85% in vitro digestibility, as well as high levels of vitamin E and beta-carotene, which promote animal fertility (Atlas, 2004). Hydroponic fodder production significantly reduces water usage. For instance, around 3 liters of water are needed to produce one kg of fresh hydroponic feed (Ramteke et al., 2019). Comparatively, 1.5-2 liters of water are required to produce one kg of hydroponically grown green barley forage, whereas conventional field conditions consume 73 liters per kg (Al-Karaki, 2010).

However, despite the benefits of hydroponic sprouting, there are challenges in maintaining the production, nutritional value, and economic yield of sprouts due to of microbiological infections and physiological deterioration (Randeniya and de Groot, 2015). To address these issues, low-dose irradiation and acidified sodium chlorite have been combined to achieve effective sterilization during germination (Nei et al., 2010). The utilization of UV radiation for air, surface, and material disinfection has gained popularity (Yang et al., 2020). The germicidal effect is caused by UV damage to the DNA or RNA of bacteria or viruses. Ozone (O_3) is an inorganic molecule composed of 3 oxygen atoms. It is an unstable gas and cannot be stored. Monroy et al. (2017) reported that ozone may trigger antioxidants that regulate hormone levels, particularly abscisic acid, to break seed dormancy and enhance germination.

The application of ozone technology has become vital for eliminating microbes, fungus, and viruses from various seeds instead of using chemical methods (Mohammad et al. 2019). Ozone sterilization technology has recently been successfully employed in the agricultural sector, either as an aqueous solution or as a gas (Loeb 2018). Abeli et al. (2017) Ozone sterilization technology has recently been successfully employed in the agricultural sector, either as an aqueous solution or as a gas. Vazquez-Ybarra et al. (2015) examined the effects of 0.53 and 59.40 mg L^{-1} ozone on lettuce (*Lactuca sativa* L.) plants in a hydroponic float system. After 10 weeks of growth, the shoot and root biomass, root length, and stem diameter of the plants treated with 2.66 and 3.96 mg L^{-1} significantly increased. Rodrigues et al. (2019) found that ozone exposure at a concentration of 25 g m^{-3} for 120 minutes had no effect on the physiological or biochemical processes of cultivar soybean seeds. According to Mohammad et al. (2019), ozonizing at 5 mg L^{-1} for 20 minutes had no negative effects on the size, color, or germination percentage of alfalfa sprouts. UV-C radiation, known for its high photochemical and biological activity, has been utilized to accelerate the germination of maize and sugar beet seedlings (Sadeghianfar et al., 2019). UV radiation is divided into three wavelength ranges: UV-A (320–390 nm), UV-B (280–320 nm), and UV-C (100–280 nm). Sadeghianfar et al. (2019) found that UV-C radiation had a significant effect on the photosynthesis of treated maize and sugar beet seedlings. Furthermore, UV-C irradiation was used to improve wheat seed germination and growth parameters (Rupiasih and Vidyasagar, 2016) and groundnut (Neelamegam and Sutha, 2015).

Different crops were subjected to infrared micro-spectroscopy to determine structural alterations in the xylem of the plants. Rico et al. (2015) reported a significant increase in root elongation in barley plants in the range of 1727–1760 cm^{-1}.

Hydroponics fodder production is considered a successful alternative method for sustainable livestock production (Ramteke et al., 2019). The chemical composition of hydroponically germinated barley fodder is affected by the harvesting duration, with the seventh day being the optimal harvest day for producing usable fodder (Akbağ et al., 2014). Chlorophyll is essential for photosynthesis (Konica Minolta, 2009) and serves as an accumulation point for rising nitrogen quantities in plants. Measuring chlorophyll content provides information about a plant's general health (Marsh, 2016). Sprouting under net houses recorded the highest values of physical characteristics and chemical analysis compared to the control room.

In Egypt, where the costs of lighting and sterilization of sprouting rooms are high and ineffective, the increase in construction fees for sprouting rooms poses an obstacle to the widespread application of such technology. There are few reports on the application of ozone, ultraviolet light, and infrared to reduce fungal infections in sprouted barley. The traditional method of sprouting rooms relies on ultraviolet tube bulbs distributed along the shelves, resulting in high construction costs and limited dissemination. Additionally, exposure to fungal infections producing toxic auxins adversely affects animal health when feeding on contaminated food. Regarding barley, various parameters such as shoot length, fresh yield weight, dry yield weight, conversion factor, chlorophyll content, N, P, K, crude protein, ash, and log reduction were evaluated. The sterilization efficiency of ozone, UV, and infrared on barley sprouts was assessed using colony-forming units (CFU). The development of this system has enabled the production of fresh forage almost year-round. The research also aims to design an optical device connected to a collection of optical fibers that will transmit light from a single source inside the sprouting room equally to all plants, while controlling the intensity and duration of lighting using a special electronic circuit. This will help reduce the number of bulbs and electrical capacity needed, ultimately lowering production costs.

The present study aims to explore the effect of different concentrations of ozonized water on seed germination and seedling growth. Additionally, the research aims to develop a multi-spectral light system and evaluate its effect on barley growth speed in sprouting rooms.

2. Material and Methods

2.1. Material

2.1.1. Overall structure of the sprouting room

This experiment was conducted in an environmentally controlled room with an average temperature of 23±1°C throughout the study period. The sprouting room was constructed using a steel stand (2 m length, 0.5 m width, and 2 m height) with two shelves placed 30 cm apart. Each shelf had a capacity of holding 12 Polystyrene trays (0.6 m length, 0.3 m width, 0.03 m height, and 0.18 m^2 area), which were obtained from the local market, as shown in Figure 1. The developed lighting system comprises optical fibers in the form of a flexible strip of LED bulbs known for their energy-saving properties. Three different types were employed to replace fluorescent lamps or tube UV lamps, as follows:

1. Fluorescent LED plant grow light strips with a full spectrum (4000K natural white).
2. Infrared flexible LED strip light (940nm, 72 watts).
3. Ultraviolet flexible LED strip light (405nm), all illustrated in Figure 1.

An electronic control circuit was designed to automate the timing of lighting and irrigation with ozone water. The timing circuit incorporates an Arduino Nano, programmed via a computer to logically manage the timing. A crystal LED screen has been added to display the timing programming commands for controlling the sprouting room. The electronic circuit can be programmed using the included control buttons. There is a pair of relays, one for operating the lighting circuit and the other for operating the developed ozone device, as demonstrated in Figure No. 3. The shelves were irrigated using water sprinklers attached to a motor, with a total irrigation rate was 500 ml per tray per day, ensuring adequate moisture for the seedlings. Tap water was used for irrigation without any additional nutrient solutions or additives, following the method suggested by Naik et al. (2015). The relative humidity was adjusted

to approximately 70% through air circulation. The hydroponic system occupied an area of 1 m^2 (Figure 1).

Raw barley grain samples (*Hordeum vulgare* L., "*Giza 128*") from the 2022 crops were purchased from the Barley Department, Agricultural Research Institute, Egypt, and stored at 5 °C until use. The grains were not subjected to any abrasion treatment. Before use, the viability of the barley grains was checked, and the germination percentage was found to be 95%.

The experiments were conducted during the winter season of 2023, from 15th December to 30th of March, at El-Serw Agricultural Research Station, Damietta, Egypt (latitude 31° 41' - 95° 73' N, and longitude 31° 81' - 40° 72' E). The minimum temperature ranged from 10.7 °C in January to 14.2 °C in March, while the maximum temperature ranged from 17.3 °C in January to 20.1 in March. The average nighttime temperatures in December, January, February, and March were 14.3, 12.3, 14, and 15.5 °C, respectively, while the daytime temperatures were 16.9, 14.9. 16.8 and 18.9 °C, respectively. A total of 1290 hours of sunshine duration were recorded, with the maximum average rainfall of 30.4 mm occurring in January. The average relative humidity during December, January, February, and March was 74%, 76%, 73%, and 66%, respectively.

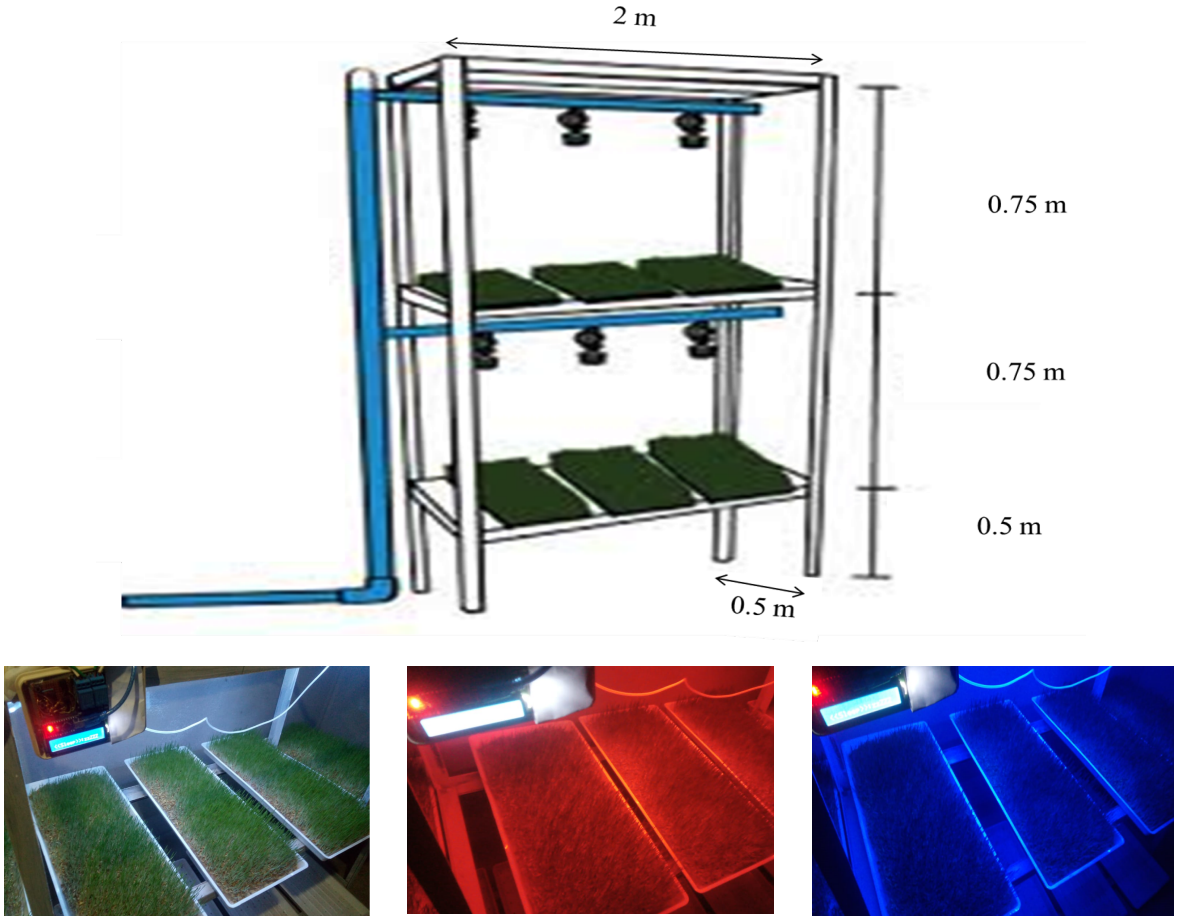
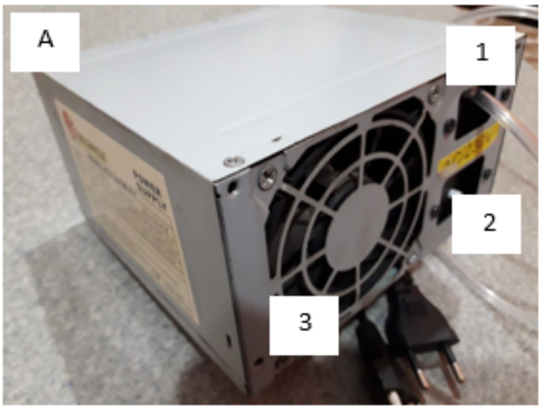


Figure 1. Barely sprouting room.

2.1.2. General description of the ozone device

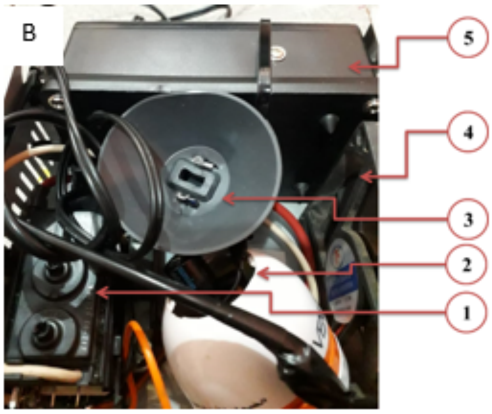
The locally manufactured ozone generation device is illustrated in Figure 2. The concept behind the ozone-generating device involves transforming the air or gas intended for treatment from an electrical insulator into a conductor of electricity, subsequently ionizing it through exposure to high electrical voltage. This process leads to the release of electrons, which gain acceleration due to the high electric voltage, resulting in collisions among them. These collisions break the bonds connecting the two atoms in an oxygen molecule, allowing one of the oxygen atoms to bond with a third atom, creating

ozone gas. This ozone gas offers numerous sterilization benefits. This device is capable of generating two types of ozone. Firstly, it produces ozone gas directly, which is then pumped through an air pump for sterilization purposes, as depicted in Figure 2B (No. 5). The ozone-directive pump is equipped with a switch that allows the user to choose between two speeds to control the amount of ozone gas emitted from the device. Secondly, the ozone generator generates dissolved ozone in the water through a hose connected to the water source, as shown in Figure 2A (No. 1 and 2). The core of the ozone generator includes a high-voltage transducer (Figure 2B, No. 1) that generates a significant electric charge, of up to 50,000 volts. This electric charge passes between two electrodes, the cathode, and the anode, resulting in the formation of an electric arc (Figure 2B, No. 3). This electric arc ionizes the oxygen and converts it into ozone. The device is further equipped with an electronic circuit to regulate the voltage entering the electrical transformer, as shown in Figure 2B (No. 2). Additionally, it incorporates an internal fan (Figure 2A, No. 3) to draw air from the surroundings and direct it internally into the device. Once the ozone gas is generated, an internal pump (Figure 2B, No. 5) functions to withdraw the ozone gas and direct it to the respective outlets, either as direct gas or dissolved in water. The device is further equipped with an electronic circuit to regulate the voltage entering the electrical transformer, as shown in Figure 2B (No. 2). Additionally, it incorporates an internal fan (Figure 2A, No. 3) to draw air from the surroundings and direct it internally into the device. Once the ozone gas is generated, an internal pump (Figure 2B, No. 5) functions to withdraw the ozone gas and direct it to the respective outlets, either as direct gas or dissolved in water. The device is further equipped with an electronic circuit to regulate the voltage entering the electrical transformer, as shown in Figure 2B (No. 2). Additionally, it incorporates an internal fan (Figure 2A, No. 3) to draw air from the surroundings and direct it internally into the device. Once the ozone gas is generated, an internal pump (Figure 2B, No. 5) functions to withdraw the ozone gas and direct it to the respective outlets, either as direct gas or dissolved in water.



A. Homemade ozone device

(1) Ozone outlet for water; (2) Ozone outlet directly; (3) Air suction fan



B. Components of the ozone device

(1) High-voltage transformer; (2) Electronic circuit; (3) Ozone generator cathodes; (4) Air suction fan; (5) Ozone pump

Figure 2. Homemade and Components of the ozone devices.

2.1.3. Generation of ozone treated water

A flask with a stopper and two holes was filled with irrigation water. One hole served as an inlet line for injecting ozone, while the other acted as an exit line to discharge any excess ozone gas. To achieve an aqueous ozone concentration of 13 mg L^{-1}, ozone gas was injected into the water for approximately 90 minutes. To prevent the dispersion of excess ozone into the air, it was passed through a second flask containing a 2% potassium iodide solution, following the method described by Mohammad et al. (2019). The ozone-treated water was prepared by bubbling the irrigation water with ozone gas for the required time to attain the desired concentration for each test. The spectrophotometric method, as outlined by Bader and Hoigne (1981), was used to determine the levels of ozone in the water used for treating barley grains and sprouts.

The following formula was employed to calculate the ozone concentration:

$$O_3 \text{ concentration } (mg\ L^{-1}) = 100\left(\frac{\Delta A}{f \times v \times b}\right) \rightarrow \quad (1)$$

Where ΔA is the difference in absorbance between the sample and blank solutions, b is the path length of the cell in the spectrophotometer (cm), v is the volume of the sample or blank (mL), and f is the factor corresponding to an aqueous ozone absorption coefficient of 0.42. The system was automated using a digital timer, as shown in Figure 3.

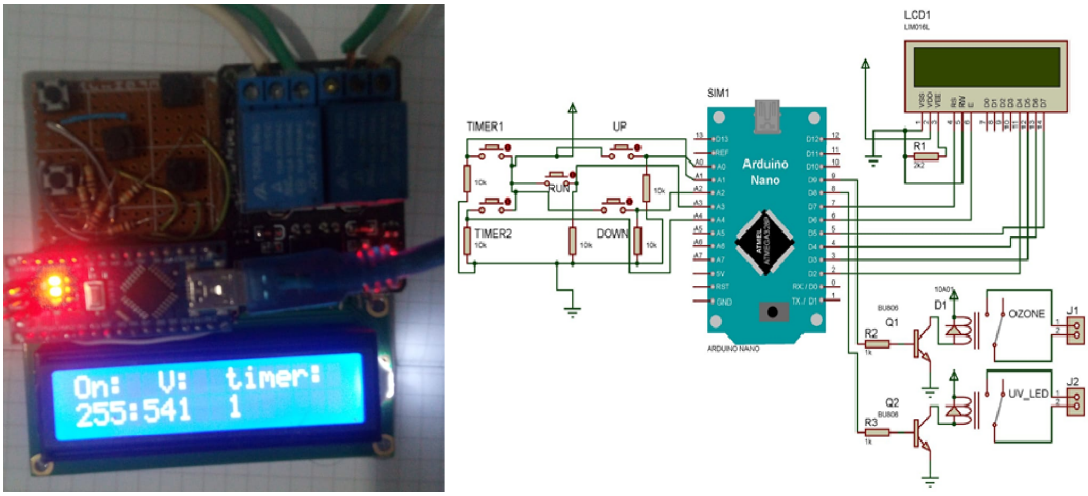


Figure 3. Digital timer circuit and its components.

2.2. Methods

At this stage and before seeding, about ten kg of barley seeds (*Hordeum vulgare L.*, *Giza 128*) were washed to eliminate floating materials (straw and immature seeds) for uniform growth. The seeds were then soaked in ozonated water for approximately 12 hours. After soaking, the seeds were placed in a wet gunny bag and incubated in a dark place to facilitate early germination. Trays were cleaned and disinfected using ozone gas generated from the ozone device, rather than undergoing chemical sterilization with a 0.1% sodium hypochlorite solution (5%), followed by a rinse with tap water. The soaked seeds were evenly spread out in the trays at a rate of 0.750 kg per tray with a thickness of 1.5-2 cm, following the sprouting method performed by Gebremedhin (2015). Small holes in the trays allowed excess water to drain, preventing water stagnation. A transparent plastic cover was placed over the trays to maintain seed moisture during germination. The germination process took 8 days to produce shoot sprouts.

2.2.1. Evaluated variables

Three variables were studied:

a) Ozone treated water (mg L^{-1}): Three levels of ozone treated water were used: 13, 26, and 39 mg L^{-1}.
b) Type of light source: Three light sources were utilized: fluorescent, infrared, and ultraviolet LED.
c) Light duration time (h): Three Light duration times were examined: 8, 16, and 24 h.

2.2.2. Measurements

2.2.2.1 Vegetative characteristics of barely sprout yield

After 8 days of seeding, the fresh fodder was harvested, and the following data were recorded: Shoot length (cm), total fresh yield (kg m^{-2}), total dry yield (kg m^{-2}), conversion factor [ratio of the produced yield to the initial planted seed weight (kg kg^{-1})], and chlorophyll content (mg m^{-2}). Chlorophyll content was measured using a nondestructive fluorometer analysis (Manetas et al., 1998). The CCM-300 chlorophyll meter, produced by OPTI-SCIENCES, Inc. (Hudson, NH), uses a

chlorophyll fluorescence ratio to measure the chlorophyll content within plant leaves (Opti-Sciences Inc., 2011).

2.2.2.2 Chemical, quality, and microbiological analysis of barely sprout yield

Fresh samples (leaves and roots) were collected, oven-dried at 70 °C in an air-forced oven for 48 hours, weighed, and stored for chemical analysis. Dried samples were turned into powder and digested in H_2SO_4 according to Allen (1974), and the nitrogen content was determined using the Kjeldahl method described by FAO (1980). The crude protein was calculated by multiplying N by 5.83 (Merrill and Watt 1955). According to AOAC (2000), samples were ignited in a muffle furnace (Protherm, PFL 110/10 MODEL) at 550 °C for four hours to determine the amount of ash. Potassium was determined using flame photometry (Ryan 1996).

The method described by Koide et al. (2009) was used to count the total number of bacteria on the barely sprouts. After germination (Day 7), all of the collected barely sprouts were weighed into 200 mL of sterile physiological saline solution. To eliminate the microbial cells from the sprouts' surface, the flask was forcefully shaken by hand. After that, 1 mL of the solution was diluted to 10 mL with sterile physiological saline solution. For the enumeration of the total bacterial count, 1 mL of the mixture was serially diluted (1:10) in a sterile saline solution. Then, 100 mL of the diluted solutions were evenly distributed in triplicate on LB plates and incubated at 37 °C to count the CFU of bacteria. Log colony-forming units per gram (log CFU g^{-1}) were used to express the microbial count. The log reduction, which was calculated using the following formula, was used to evaluate the sterilization ability of the new technique.

$$Log\ reduction = \log\left(\frac{CFU_{control}}{CFU_{treated}}\right) \quad (2)$$

2.2.2. Statistical analysis

The experiments were replicated three times. A completely randomized experimental study design was used for data analysis. The Costat Program (Oida, 1997) was used to determine the statistical significance of the variables under consideration based on the probability ($P<0.05$).

3. Results and discussion

3.1. Factors affecting some vegetative characteristics of barely sprout yield

The results presented in Figures 4 to 6 illustrate the relationship between ozonated water, the type of light source, and light duration time on various vegetative characteristics of sprouting barely fresh fodder, including shoot length, fresh yield weight, dry yield weight, conversion factor, and chlorophyll content. The findings indicate that fresh green fodder can be produced after 8 days of seeding in sprouting rooms from the tested barely crop (*Giza 128*). As the levels of ozone-treated water increased, there was an indirect increase in the vegetative characteristics of sprout yield.

Notably, the mean best values for vegetative characteristics were observed at an ozone-treated water level of 39 mg L^{-1}. Specifically, shoot length was 19.33±3.89 cm, fresh yield weight was 35.97±4.04 kg m^{-2}, dry yield weight was 7.60±1.06 kg m^{-2}, conversion factor was 8.63±0.97 kg kg^{-1}, and chlorophyll content was 44.58±2.09 mg m^{-2}. In contrast, the mean minimum values for vegetative characteristics were observed at an ozone-treated water level of 13 mg L^{-1}, with shoot length being 17.44±3.90 cm, fresh yield weight being 30.08±4.24 kg m^{-2}, dry yield weight being 6.44±1.50 kg m^{-2}, conversion factor being 7.20±1.02 kg kg^{-1}, and chlorophyll content being 35.96±2.41 mg m^{-2}.

Regarding the light source, the highest mean values of vegetative characteristics were observed when using Ultraviolet LED as a light source and sterilizing medium. Specifically, shoot length was 22.22±1.65 cm, fresh yield weight was 37.63±2.98 kg m^{-2}, dry yield weight was 8.21±0.77 kg m^{-2}, conversion factor was 9.03±0.71 kg kg^{-1}, and chlorophyll content was 42.56±3.66 mg m^{-2}. Conversely, the lowest mean values of vegetative characteristics were observed when using fluorescent lamps as a light source, with shoot length being 13.74±1.58 cm, fresh yield weight being 28.72±2.92 kg m^{-2}, dry

yield weight being 5.81±0.65 kg m^{-2}, conversion factor being 6.89±0.70 kg kg^{-1}, and chlorophyll content being 37.74±4.05 mg m^{-2}.

Furthermore, increasing the light duration time resulted in higher vegetative characteristics. The highest mean values were observed at a light duration time of 24 h, with shoot length being 19.37±3.55 cm, fresh yield weight being 34.69±4.79 kg m^{-2}, dry yield weight being 7.27±1.21 kg m^{-2}, conversion factor being 8.33±1.15 kg kg^{-1}, and chlorophyll content being 41.11±4.10 mg m^{-2}. In contrast, the minimum mean values were observed at a light duration time of 8 h, with shoot length being 17.37±4.00 cm, fresh yield weight being 32.06±4.69 kg m^{-2}, dry yield weight being 6.61±1.17 kg m^{-2}, conversion factor being 7.69±1.12 kg kg^{-1}, and chlorophyll content being 39.48±4.53 mg m^{-2}. The control group had significantly lower mean values for all vegetative characteristics, with shoot length being 10.40±1.30 cm, fresh yield weight being 20.56±2.67 kg m^{-2}, dry yield weight being 4.11±1.01 kg m^{-2}, conversion factor being 4.93±0.82 kg kg^{-1}, and chlorophyll content being 28.43±1.51 mg m^{-2}.

These results can be attributed to the beneficial effect of ozone-treated water as a nonchemical method for removing microbes, fungus, and viruses from sprouting barely. This is in line with previous research by Monroy et al. (2017) and Mohammad et al. (2019), which highlighted the regulatory role of ozone in hormone levels, particularly abscisic acid, leading to enhanced barley growth. Additionally, changes in ozone-treated barely sprouts may be attributed to the stress induced by certain doses of O_3, which triggered an adaptive response and improved the barley's resistance to environmental challenges.

The observed shoot length in barley is consistent with the findings of Al-Hashmi (2008). The values of total fresh yield align with previous reports by Shtaya (2004). Peer and Leeson (1985) have reported a negative relationship between dry matter content and fresh weight yield, which limits the consumption of green fodder by animals due to its low dry matter content.

The production conversion ratio, calculated as the quantity of new fodder produced per unit of seed utilized, ranged from 4 to 8 times the values reported by Morgan et al. (1993). Al-Ajmi et al. (2009) and Al-Hashmi (2008) also reported a ratio of 2.76 to 3 kg of green forage per kg of barely seed, which is lower than that reported by other researchers.

The higher production of chlorophyll has been linked to nitrogen accumulation and overall plant health (March, 2016). The higher level of chlorophyll found in these plants suggests that they were more vigorous and metabolically active.

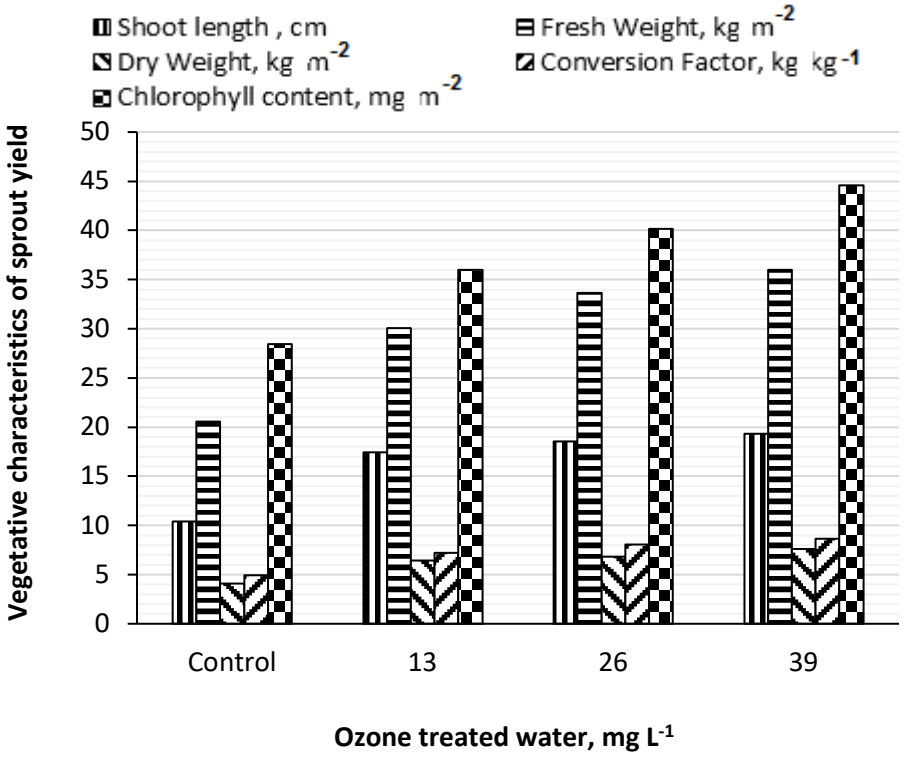


Figure 4. Effect of ozone treated water (mg L^{-1}) on vegetative characteristics of barely sprouts yield.

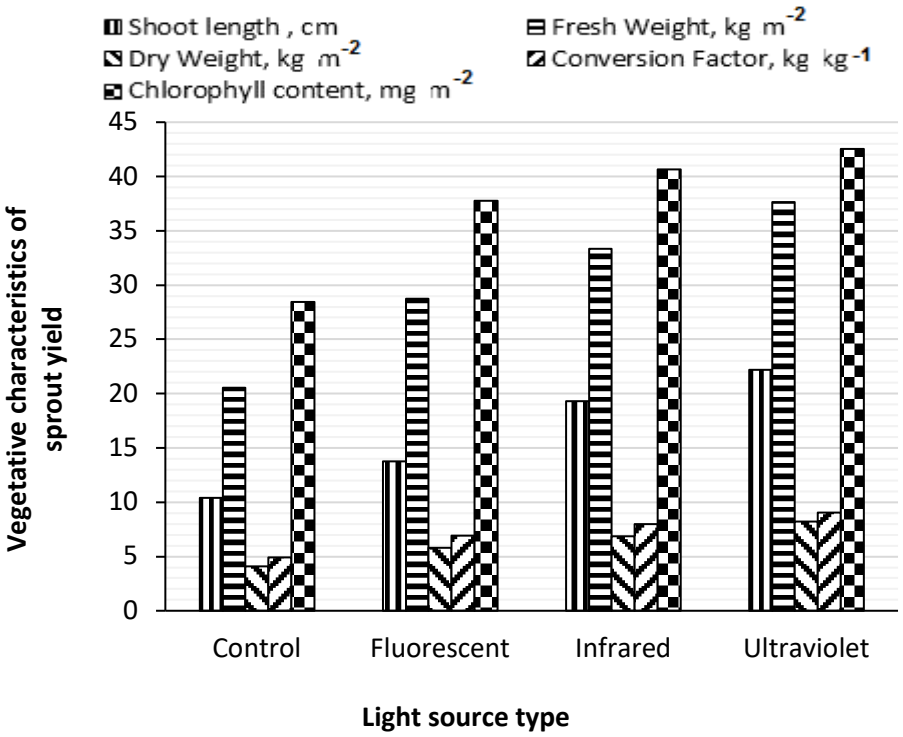


Figure 5. Effect of light source type on vegetative characteristics of barely sprout yield.

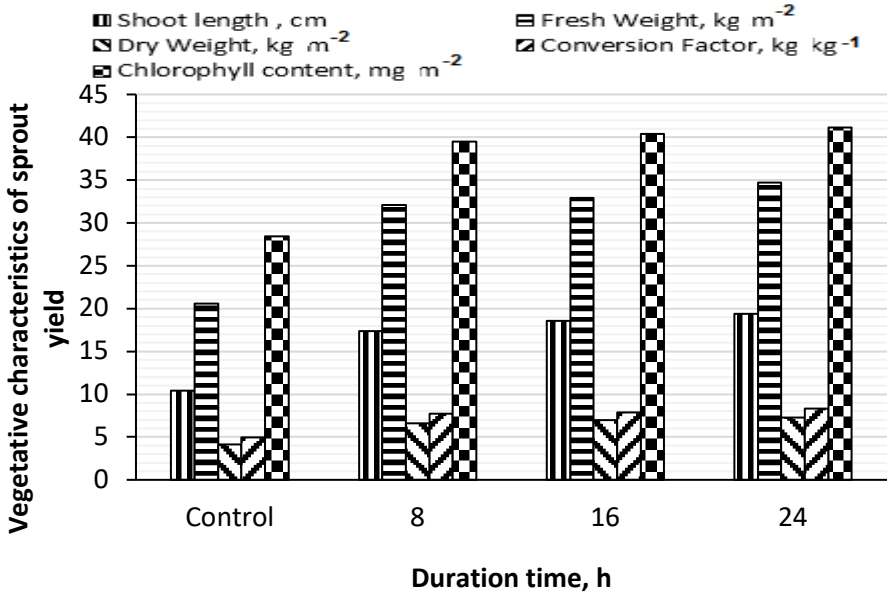


Figure 6. Effect of duration time (h) on vegetative characteristics of barely sprout yield.

3.2. Factors affecting chemical, quality and microbiological analysis of barely sprout yield

The results presented in Figures 7 to 9 demonstrate direct relationships between the tested factors (ozonated water, type of light source, and light duration time) and the measured chemical, quality, and microbiological analysis of sprouting barley yield. These findings have important implications for the optimization of fresh fodder production and the enhancement of the nutritional and microbiological quality of sprouting barley.

Regarding the chemical composition, the study revealed that the ozone-treated water significantly influenced the nutrient content of the sprouting barley. The most significant mean values of nitrogen (N), phosphorus (P), potassium (K), crude protein, ash, and log reduction were observed at an ozonated water level of 39 mg L^{-1}. The respective mean values were 3.20±0.04%, 0.77±0.03%, 2.05±0.06%, 18.64±0.24%, 3.43±0.19%, and 4.21±0.34. In contrast, the minimum mean values for these chemical parameters were observed at an ozone-treated water level of 13 mg L^{-1}, with the respective mean values being 2.98±0.13%, 0.54±0.05%, 1.39±0.25%, 17.39±0.74%, 2.22±0.31%, and 2.53±0.32. Additionally, the choice of the light source and light duration time also had a significant impact on the chemical composition of the sprouting barley. The highest mean values of N, P, K, crude protein, ash, and log reduction were obtained when using Ultraviolet LED as a light source and sterilizing medium. Conversely, the lowest mean values were observed when using fluorescent lamps as a light source. Moreover, increasing the light duration time was found to increase the mean values of N, P, K, crude protein, ash, and log reduction. The highest mean values were observed at a light duration time of 24 h, while the lowest mean values were observed at a light duration time of 8 h. These findings suggest that the combination of ozone-treated water, Ultraviolet LED as a light source, and longer light duration time contributes to the enhanced nutritional content of the sprouting barley, making it a more valuable feed source for livestock.

The study also compared the crude protein content obtained in this research with previous studies. The results were found to be comparable to the range reported by Sneath and McIntosh (2003) for the composition of sprouted barley, which ranged from 11.38% to 24%. This further validates the use of ozonated water and optimized light conditions to enhance the nutritional value of sprouting barley and ensure its suitability as a high-quality feed option.

In terms of microbiological analysis, the study demonstrated that non-treated sprouting barley had a significantly higher total number of bacteria. This highlights the importance of implementing effective measures to control microbial contamination during the sprouting process. Different authors have previously used chemical substances to treat microorganisms, but the study findings are consistent with Conceico et al. (2016) and Ferreira et al. (2016), who observed that such products may not completely remove the presence of microorganisms. The application of ozonated water in sprouting barley offers advantages over chemical treatments, as it significantly reduces the need for such chemicals, leading to a more environmentally friendly and sustainable approach to fresh fodder production.

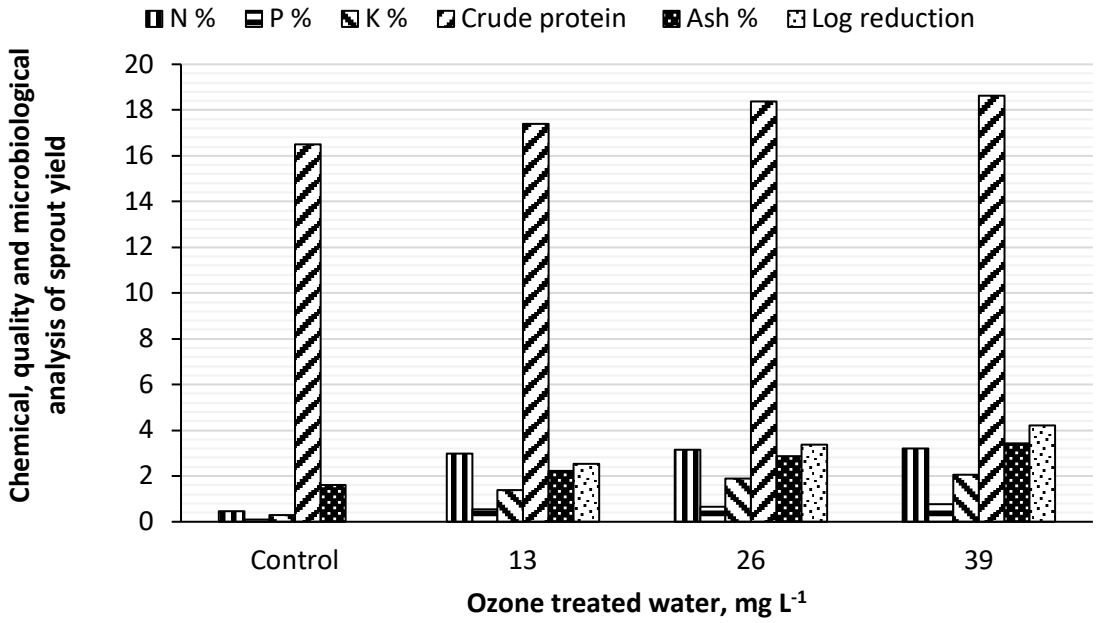


Figure 7. Effect of ozone treated water (mg L^{-1}) on chemical, quality and microbiological analysis of barely sprout yield.

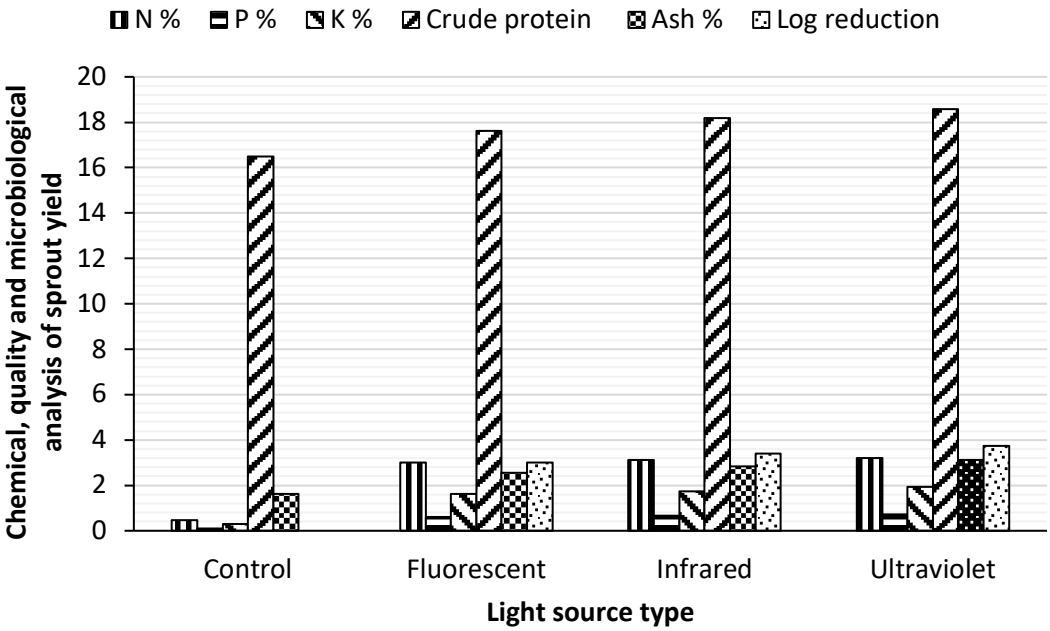


Figure 8. Effect of light source type on chemical, quality and microbiological analysis of barely sprout yield.

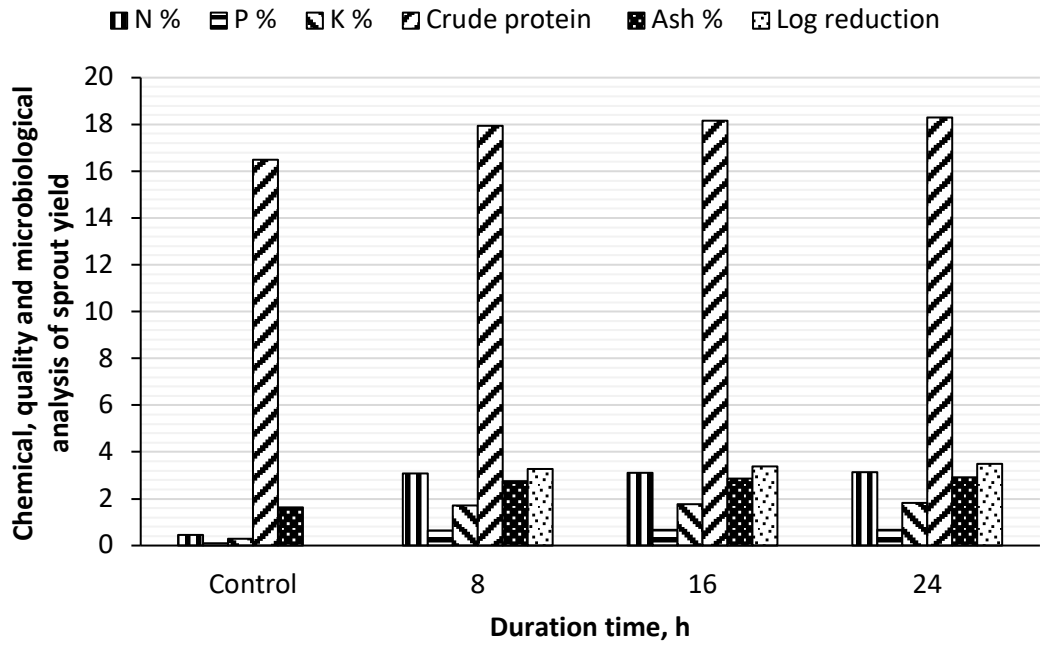


Figure 9. Effect of duration time (h) on chemical, quality and microbiological analysis of barely sprout yield.

The results presented in Table 1 and 2 provide valuable insights into the impact of various factors on the characteristics of barely sprout yield. The ANOVA analysis clearly demonstrates that different levels of ozone treated water have a significant effect on vegetative characteristics, as well as the chemical, quality, and microbiological analysis ($p < 0.001$). It is evident that the levels of ozonated water played a crucial role in influencing the growth and development of the sprouting barely. Specifically, the third level of ozonated water yielded the most favorable results, as indicated by the significant difference between the first level and both the second and third levels ($p < 0.05$). These

findings suggest that increasing the level of ozonated water positively correlates with improved vegetative characteristics of the sprouting barely, leading to enhanced yield and quality.

Moreover, the type of light source utilized in the sprouting process also exhibited a significant impact on the vegetative characteristics of barely sprout yield. Among the different light sources tested, the Ultraviolet LED emerged as the most effective in maximizing the vegetative characteristics. The results highlight the importance of choosing the appropriate light source for sprouting rooms, as it can significantly influence the growth and overall development of the sprouting barely.

Furthermore, the duration of light exposure demonstrated a noteworthy effect on all the measured parameters. The length of light exposure directly influenced the vegetative characteristics, with the third duration level producing the most favorable outcomes, while the first level yielded the least desirable results ($p < 0.05$). This finding emphasizes the importance of providing adequate light exposure to achieve optimal growth and yield of sprouting barely.

Table 1. Means and standard errors for some vegetative characteristics of sprout yield affected by studied factors

Factors	Level	Shoot length, cm	Fresh weight, kg m^{-2}	Dry weight, kg m^{-2}	Conversion factor	Chlorophyll content, mg m^{-2}
Control		10.40±1.30	20.56±2.67	4.11±1.01	4.93±0.82	28.43±1.51
Ozone treated water, mg L^{-1}	13	17.44±3.90^{a}	30.08±4.24^{a}	6.44±1.50^{b}	7.20±1.02^{b}	35.96±2.41^{b}
	26	18.52±3.76^{ab}	33.62±4.07^{ab}	6.85±0.83^{ab}	8.07±0.98^{ab}	40.15±2.55^{ab}
	39	19.33±3.89^{a}	35.97±4.04^{a}	7.60±1.06^{a}	8.63±0.97^{a}	44.58±2.09^{a}
p-value		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Type of light source	Fluorescent	13.74±1.58^{a}	28.72±2.92^{c}	5.81±0.65^{b}	6.89±0.70^{c}	37.74±4.05^{b}
	Infrared	19.33±1.57^{b}	33.31±3.24^{b}	6.86±0.87^{ab}	8.00±0.78^{b}	40.67±3.96$^{a b}$
	Ultraviolet	22.22±1.65^{a}	37.63±2.98^{a}	8.21±0.77^{a}	9.03±0.71^{a}	42.56±3.66^{a}
p-value		<0.0001	<0.0001	<0.0201	<0.0001	<0.0001
Light duration time, h	8	17.37±4.00^{b}	32.06±4.69^{b}	6.61±1.17^{b}	7.69±1.12^{c}	39.48±4.53^{b}
	16	18.56±3.95^{ab}	32.92±4.53^{ab}	7.00±1.30^{ab}	7.90±1.09^{ab}	40.37±4.37^{ab}
	24	19.37±3.55^{a}	34.69±4.79^{a}	7.27±1.21^{a}	8.33±1.15^{a}	41.11±4.10^{a}
p-value		0.0121	0.0131	0.0152	0.0116	0.0201

Table 2. Means and standard errors for chemical, quality and microbiological analysis of sprout yield affected by studied factors

Factors	Level	N %	P %	K %	Crude protein %	Ash %	Log reduction
Control		0.46±0.07	0.11±0.01	0.29±0.03	16.50±0.27	1.62±0.15	0.00±0.02
Ozone treated water, mg L^{-1}	13	2.98±0.13^{b}	0.54±0.05^{b}	1.39±0.25^{b}	17.39±0.74^{b}	2.22±0.3^{b}	2.53±0.32^{c}
	26	3.15±0.06^{ab}	0.67±0.05^{ab}	1.88±0.15^{ab}	18.37±0.37^{ab}	2.88±0.2^{ab}	3.38±0.29^{b}
	39	3.20±0.04^{a}	0.77±0.03^{a}	2.05±0.06^{a}	18.64±0.24^{a}	3.43±0.1^{a}	4.21±0.34^{a}
p-value		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Type of light source	Fluorescent	3.02±0.15^{b}	0.61±0.1^{b}	1.63±0.34^{b}	17.63±0.86^{b}	2.56±0.56^{b}	3.01±0.70^{b}
	Infrared	3.12±0.09^{ab}	0.66±0.1^{ab}	1.75±0.34^{ab}	18.19±0.50^{ab}	2.85±0.5^{ab}	3.39±0.72^{ab}
	Ultraviolet	3.19±0.07^{a}	0.71±0.0^{a}	1.94±0.24^{a}	18.59±0.39^{a}	3.12±0.46^{a}	3.74±0.70^{a}
p-value		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Light duration time, h	8	3.08±0.15^{b}	0.64±0.11^{b}	1.71±0.35^{b}	17.94±0.87^{b}	2.75±0.75^{b}	3.27±0.76^{b}
	16	3.11±0.12^{ab}	0.66±0.10^{ab}	1.78±0.33^{ab}	18.15±0.70^{ab}	2.86±0.55^{ab}	3.38±0.76^{ab}
	24	3.14±0.10^{a}	0.68±0.10^{a}	1.83±0.32^{a}	18.31±0.56^{a}	2.92±0.55^{a}	3.48±0.77^{a}
p-value		0.0112	0.0121	0.0107	0.0128	0.0171	0.0182

Conclusion

In conclusion, the application of ozonated water, type of light source, and light duration time had significant effects on the vegetative characteristics of sprouting barely fresh fodder. The results demonstrate the potential of ozone as an effective nonchemical method for improving sprout yield and enhancing barley growth. Additionally, the use of Ultraviolet LED as a light source and extending the light duration time contributed to increased vegetative characteristics. These findings provide valuable insights for optimizing the production of fresh fodder through hydroponics, promoting sustainable livestock

production and supporting animal health. Further research and optimization of the ozone and light treatment parameters are warranted to enhance the efficiency of the sprouting rooms and ensure consistent production of high-quality fresh forage.

Overall, the direct relationships observed between ozonated water, type of light source, light duration time, and the chemical, quality, and microbiological analysis of sprouting barley yield demonstrate the potential of this approach for optimizing fresh fodder production. The results provide valuable insights for the agricultural sector, particularly in the context of hydroponics fodder production. The use of ozonated water and the appropriate choice of light source and duration can significantly enhance the nutritional value and microbiological safety of sprouting barley, supporting sustainable livestock production and animal health. The study's findings contribute to the advancement of hydroponics technology and provide practical guidance for improving fodder production systems. Further research and development in this field can lead to more efficient and reliable methods for year-round fresh forage production, benefiting both farmers and the livestock industry as a whole.

In conclusion, the results indicate that the levels of ozonated water, type of light source, and light duration time all play vital roles in determining the vegetative characteristics and overall quality of barely sprout yield. The study suggests that the highest level of ozonated water, Ultraviolet LED as the light source, and the longest light duration time are the most favorable conditions for achieving the best results in terms of yield and quality. These findings provide valuable guidance for optimizing the production of fresh forage using hydroponics and can contribute to more sustainable and efficient livestock production practices. Additionally, the use of ozonated water offers environmental benefits by reducing the reliance on chemical treatments, which is a promising step towards eco-friendly agricultural practices.

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