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Effects of LED Lights and New Long-Term-Release Fertilizers on Lettuce Growth: A Contribution for Sustainable Horticulture

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Abstract: The horticulture sector has been directed by European guidelines to improve its practices related to environmental sustainability. Moreover, the practice of horticulture in urban areas is increasing since it provides fresh products that are locally produced. At the same time, horticulture needs to implement circular economy approaches and energy-efficient models. Therefore, to address these issues, this study investigated the effects of an integrated fertilizer-box-based cultivation system equipped with LED lights and coated porous inorganic materials (C-PIMs), which was applied as fertilizer, on *Lactuca sativa* L. growth. Two different types of lightweight aggregates were formulated considering agri-food and post-consumer waste, and they were enriched with potassium and phosphorus. Involving waste in the process was part of their valorization in the circular economy. Using PIMs as fertilizers enabled the controlled release of nutrients over time. The tests were carried out in controlled conditions using two LED lighting systems capable of changing their light spectrum according to the growth phases of the plants. The effects of two different lighting schemes on the growth of lettuce plants, in combination with different amounts of aggregates, were studied. The results showed that increasing the amount of C-PIMs statistically improved the lettuce growth in terms of dry biomass production (+60% and +34% for two different types of PIM application) when the plants were exposed to the first LED scheme (LED-1). Plant height and leaf areas significantly increased when exposed to the second LED scheme (LED-2), in combination with the presence of C-PIMs in the soil. The analysis of the heavy metal contents in the lettuce leaves and the soil at the end of the test revealed that these elements remained significantly below the legislated thresholds. The experimental achievements of this study identified a new approach to improve the environmental sustainability of horticulture, especially in an urban/domestic context.

Keywords: aggregates; byproducts; LED; lettuce; long-term release fertilizer

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1. Introduction

Lettuce (*Lactuca sativa* L.), and its numerous cultivars, is an important dietary component, providing significant amounts of fiber, minerals, and other compounds that are important for health, as well as polyunsaturated fatty acids (PUFAs), vitamins, and bioactive compounds in variable amounts among the lettuce types [1]. It is a crop species widely cultivated as a fresh-cut vegetable and the most popular ingredient in salads. China is the greatest producer of lettuce in the world, while U.S. and Western Europe cultivate 22% and 13% of all lettuce globally, respectively [2]. In Italy, lettuce cultivation in greenhouses covered a total area of about 5000 ha [3], ranking lettuce as the second most cultivated species after tomato. Due to its common use in everyday life and its current intense production, lettuce was chosen as the subject for this research.

The practice of horticulture in urban areas is increasing, as it provides locally produced fresh products [4,5]. It has several advantages, such as being able to cultivate fresh food anywhere, including on window ledges and balconies; in greenhouses and vertical farms; and on green roofs and other modern infrastructure [6]. Furthermore, horticultural activities provide numerous benefits, including low-cost food production [7]. They have many positive effects for practitioners, as well, such as providing entertainment; improving wellbeing and mental health; and enhancing biodiversity in the environment [8–10] This is particularly significant in the light of the rapid growth of global urban populations, and this trend is expected to continue [11,12]. The United Nations has forecasted that by 2050, 68% of the global population is likely to live in cities [11]. However, it is essential to ensure this is sustainable from an environmental perspective by following the principles of reusable byproducts, circularity, and energy conservation [13,14]. Therefore, a new technological approach for crop cultivation is required to ensure the ease and sustainability of urban horticulture. New horticultural practices have included soilless systems, hydroponic cultures, and substrates that integrate both organic (e.g., peat moss) and inorganic (e.g., vermiculite, perlite, aggregates of lightweight expanded clay) components [15]. These systems limit the exploitation of the soil and conserve energy and water for post-harvesting operations. However, peat moss is the principal organic component of growing substrates, and as a non-renewable resource, its increased application places peat-land ecosystems at risk of exploitation and increases carbon dioxide mobilization [13,16]. For these reasons, finding alternatives to traditional peat, with a focus on reusable, recyclable materials, has become both urgent and necessary. One of the strategies to reduce the environmental impact of peat use in horticulture has been its partial replacement with other soilless solutions [17]. Among them, porous inorganic materials (PIMs) have been used widely as a substrate due to their lightweight, large pores and their enhanced drainage characteristics [18]. In a circular economy, PIMs can be produced from the byproducts of other industrial process, which is in agreement with Sustainable Development Goal 12 (i.e., responsible consumption and production). In this view, industrial and agricultural waste can be used in the formulation of aggregates with several potential applications, including as physical support structures and chemical substitutes for the pot-cultivation of plants. For this research, the aggregates were obtained in the form of a core-shell specimen by formulating an NPK supply system of spent coffee ground, bone ash, and defatted biomass from black soldier fly prepupae. These lightweight materials were designed and customized to provide nutrients over a period of time [18–20]

Horticultural activities can be carried out indoors, for example, in kitchen gardens, with many advantages. First, there is no seasonality or extreme weather conditions to hinder crop growth, and it allows for the efficient use of resources, such as water and fertilizers, that can be applied according to the crop's specific needs. Furthermore, it prevents both water loss and pollution due to nutrients released into the environment [21]. However, indoor horticulture requires a controlled environment and artificial lighting, which could lead to waste due to the high amount of energy required. In this context, light-emitting diodes (LED) were particularly attractive due to their energy efficiency, long operating life, and tunable wavelength emissions [22]. Artificial lights operate consistently, independently of the adverse climatic events throughout the year. However, the interactions between plant species and some environmental factors, many of which may affect indoor horticulture systems, are poorly understood. For example, a plant's explore to light must be regulated both in its intensity and its wavelength composition. Light intensity, expressed as photosynthetic photon flux density (PPFD), has to be tailored in order to optimize energy use and to ensure crop yield [23]. On the other hand, light wavelengths influence plant physiology and metabolite production during their life cycle. Regarding lettuce cultivars, blue (440–460 nm) and red (630–660 nm) are the most used wavelengths for the indoor growth of this species, and this is occasionally coupled with white light [24,25]. One method to enhance the effect of red and blue on lettuce growth has been to alternate the two wavelengths [26] or to delay the application of one, in respect to the other [27].

Another approach has been to add other wavelengths to the red and blue in order to simulate sunlight [28]. For example, the green (520–530 nm) wavelength did not appear to have a significant impact on morphological traits but, instead, affected the content of the metabolites in the lettuce [29]. Far-red (730–745 nm) wavelengths influenced the growth of different lettuce cultivars, and the addition of far-red to red or red-and-blue light improved the growth of lettuce according to the fresh weight and leaf area of the plant [30]. However, the effects of far-red emissions on organic compounds have been shown to vary [31], as its addition enhanced antioxidant phenolic compounds while lowering the relative specific chlorophyll content [32,33].

This study developed an integrative technological application to enhance the sustainability of horticultural practices, even those conducted indoors. For this purpose, two different types of aggregates were applied as fertilizers, along with two different LED-lighting schemes, and their results were evaluated according to the lettuce growth.

The first lighting scheme applied in this study was based on blue and red wavelengths in a 50/50 ratio. The second one combined different strategies to enhance lettuce growth, namely the alternation of blue and red lighting, along with green, far-red, and white lighting. To further enhance the novelty of this work, the second lighting scheme was also characterized by an initial stage that employed only red and far-red spectra to promote the germination of lettuce seeds.

The chemical analyses, carried out both on the substrate and the lettuce leaves, monitored the release of the elements in order to prevent the bioaccumulation of toxic substances in the leaves.

2. Materials and Methods

2.1. Plant Material, Growing Media, and Lighting Conditions

The influence of porous inorganic materials (PIMs), coated with long-term fertilizers (C-PIMs), and two different lighting conditions were tested on baby leaf lettuce *Lactuca sativa* L. cultivar Chiara (ISI Sementi S.p.A, Fidenza, Italy). The choice of this species was due to its features, such as a medium-to-short growing cycle (20–28 days), small size, and high tolerance to tip burn [15]. These characteristics ensured its cultivation was suitable both indoor and outdoor applications, as well as in pots, vertical green walls, etc.

The substrate used for germination and growth tests was prepared by mixing agriperlite (Agrilit 3, Perlite Italiana Srl, Italy, particle size 2–5.6 mm, density 90 kg/m^3) with a universal mold (UNICO, AL.FE Srl, Pomponesco MN, Italy), which was obtained from a mixture in different granulometry percentages of blond and brown peats, organic C (23%), organic N (0.4%), and organic matter (46%). Agriperlite and mold were mixed at a rate of 3:1, respectively, in order to reduce the exploitation of natural resources, as well the environmental impact linked with peat extraction [13]. Lettuce cultivation was carried out in plastic pots (about 500 cm^3 volume); after the pots had been filled with the substrate, 50 mL of tap water was added, and 5 seeds per pot were seeded. The pots were irrigated with the same amount of water every 3 days for the entire duration of any test run, i.e., 28 days. Pots were placed in trays in a growth chamber at 24 ± 1.5 °C and a range of 65–75% of relative humidity, under day-long conditions (16/8 h light/dark photoperiod), at 55 cm under the light panel, and this distance was the same for all testing durations. Two different lighting conditions were applied, LED-1 and LED-2. Details on the lighting conditions are provided in Table 1.

Two different types of C-PIMs were applied, APNUT and APV50. The hidden part of both C-PIMs was obtained by powder sintering clay, pumice scraps (provided by Europomice srl, Milan, Italy), and spent coffee grounds, at 1000 °C, as already tested in [18]. In addition to spent coffee grounds, other types of agro-industrial waste were considered and valorized in the composition of each aggregate, following a circular economy approach. Spent coffee grounds in this study acted as a pouring agent, while animal bonemeal ash and K_2CO_3, which were added as-is or vitrified with pumice scraps, acted as a source of phosphorus and potassium. Afterwards, an N-rich bio-coating (defatted biomass of black soldier fly prepupae) was applied [18]. Among these materials, pumice scraps, spent coffee grounds, and animal bonemeal ash were recovery raw materials that could be valorized in order to avoid their potential management and disposal costs. In detail, spent coffee grounds were a post-consumer byproduct that are typically treated as organic waste. The pumice scraps, obtained from a pumice extraction operation, were very fine and could not, therefore, be sold commercially. If these remain in a quarry and exceed the recovery plans, they are placed in authorized areas with relative charges. Finally, the animal bonemeal, obtained from a meat processing plant, was typically sold as fertilizer. The meal ash was specially created by an industrial partner to improve the valorization as an inorganic concentrate of phosphate compounds. Pumice waste, spent coffee grounds, and animal bonemeal ash accounted for 41.7 wt%, 10 wt%, and 14 wt%, respectively, in both PIM formulations.

Table 1. Description of the two lighting schemes used for the growth tests performed on lettuce.

Time of Application (days)	Hours of Application (h d^{-1})	Wavelengths (PPFD%) Blue (450 nm)	Green (521 nm)	Hyper-Red (660 nm)	Far-Red (730 nm)	White 2700 K
			LED-1			
1–28	1–16	50.0%		50.0%	0%	0%
			LED-2			
1–7	1–16			66.6%	33.3%	0%
8–28	1–8	59.3%	14.0%	0.0%	7.3%	19.3%
	9–16		14.0%	59.3%	7.3%	19.3%

The sources of nutrients were introduced in the C-PIMs, as they were in a ceramic matrix (APNUT) or embedded in fertilizer glass (APV50) in order to verify the effect of the amorphous structure for the sustained release of nutrients. The organic coating was composed of a biomass obtained from black soldier fly larvae (BSFL) that had been reared on waste of vegetable origin and used as a source of nitrogen [11]. The formulation of the coating had been designed in a previous study [19]. The organic coating was developed using the defatted BSFL biomass, glycerol, and water. The removal of the lipidic fraction from BSFL was necessary to maximize the N-content of the biomass. The optimization of the coating considered two different methods for lipid fraction removal and aimed to maximize the content of the BSFL biomass. The final coating formulation was 89 wt% defatted BSFL and 11 wt% water, eliminating the presence of glycerol.

The two lighting schemes applied in this study were characterized by the same photoperiod and PPFD, equal to 16 h/day and 150 µmol m^{-2} s^{-1}, respectively. The resulting daily light interval (DLI) was 8.64 mol $m^{-2}d^{-1}$. One Phytofy® RL (OSRAM GmbH, Munich, Germany) LED module, designed for horticulture applications, acted as the only light source. The characteristics and differences are shown in Table 1. The LED-1 lighting scheme was composed of only hyper-red and -blue wavelengths in a 50/50 proportion, while LED-2 was composed of green, far-red, and white lights, and it was evaluated to verify its effect on lettuce growth, as suggested in [24,34,35]. The LED-1 scheme was constant throughout all the 28 days of the growth test, while LED-2 was divided into two main parts. The first 7 days of LED-2 used only hyper-red and far-red wavelengths in a 2:1 proportion to promote a rapid germination of lettuce seeds, as suggested in [28,36]. The second part of LED-2 used all the wavelengths, while alternating the blue and hyper-red (Table 1). Furthermore, the alternation of these two wavelengths on lettuce plants has been reported to promote their growth [26,27].

Two repeated growth tests were designed for each lighting condition (LED-1 and LED-2). The conditions for each test involved the two types of fertilizers (APV50 and APNUT) that were applied to the substrates in two different quantities, 3 and 6 granules, named APV50_3, APNUT_3 and APV50_6, APNUT_6, respectively. Then, four groups, in addition to the controls (to which no C-PIMs were added, named APV50_0 and APNUT_0), were determined for each lighting condition. Upon the conclusion of our repetitive testing, the six pots were analyzed for each condition, including controls, considering that five seeds had been sown in each pot. Upon the conclusion of growth test, each lettuce plant was considered as a sample for each condition. However, germinated seedlings that did not develop further were not considered in the agronomic evaluations.

2.2. Morphological and Agronomic Parameters

The following morphological and agronomic parameters were recorded at the end of each test: shoot fresh weight (SFW) using an analytical scale (Kern, d = 0.1 mg), shoot dry weight (SDW), and roots dry weight (RDW). Dry weight was measured after desiccation in a stove (Argo Lab TCN 50) at 65 °C for a minimum of 48 h. The height (H) of the lettuce shoots and roots were measured with a caliper. Germinability rate (GI, %) was evaluated by the number of shoots on the total of planted seeds, according to the following formula [18]:

$$GI = (N^{\circ}\ shoots\ / N^{\circ}\ total\ seeds) \times 100$$

The leaf area index was evaluated for two plants in each pot and the three largest leaves of each plant. The image of the leaves was captured using a Canon EOS 1100D at a fixed position, height, and lighting condition for each capture. The images were then analyzed using ImageJ software (version 1.52, NIH, Bethesda, MD, USA). For each plant, the total area of the three leaves was considered, and the average value of the plant area for each pot was calculated.

Moreover, the fraction of biomass to root (FTR) and SDW/H ratio were calculated. The first parameter indicated the percentage of total dry plant biomass that was allocated in the roots, while the rate between shoot dry weight and height represented a further evaluation of the plant development.

The total pigments content was also assessed. At least two leaf samples from each pot and for each test were analyzed. Approximately 0.5 g of each sample was weighed with an analytical scale and immediately frozen at −20 °C; samples were then ground with a mortar, using 2 mL of acetone 80% and 0.1 g of sand, according to Lichtenthaler's method [37]. The extract was inserted into tubes, vortexed for homogenization, and then shaken for 30 min in a dark room (BioSan mini shaker). Then, the tubes were centrifugated at 5000 rpm for 5 min (Thermo Scientific, SL 8R), and the supernatant was recovered. Two addition extractions were implement to extract as much pigment as possible. The absorbance was measured with a UV–vis spectrophotometer (Jasco, V730). The equations used to determine chlorophyll a and b, and total carotenoid concentration were based on the specific absorption coefficients in acetone 80%, according to [37].

2.3. Soil and Leaves Chemical Analysis

To evaluate the release of elements into the soil by the fertilizers, samples of substrate were analyzed before and after testing. In detail, total organic C, total N, assimilable P, exchangeable K, Ca, Mg, Na, and assimilable Fe were considered; furthermore, nitrates (NO_3), Al, Cr, Pb, Cu, and Si were analyzed to verify the levels as required by legislation. All the analyses were conducted by an external chemical laboratory that applied certified methods, according to Italian [38] and other European legislation (UNI EN 16174, 2012 Met. A, UNI EN16170, 2016). Silicon was extracted by acid attack and according to UNI EN16170, 2016; Fe was determined according to a certified method [39]. The chemical analysis of soil was performed before plant growth (t0); on soil without C-PIMs after plant growth and exposed to LED 1 (t0LED-1); and exposed to LED-2 (t0LED-2). Moreover, the analysis was performed on each test condition after lettuce growth on soil with 3 and 6 APV50 aggregates and with 3 and with 6 APNUT aggregates, both exposed to LED-1 and LED-2 lights.

Moreover, lettuce leaves were analyzed to assess the uptake of nutrients from the substrate and to monitor the bioaccumulation of toxic elements. Analyses were carried out by an external laboratory that applied certified methods: Aluminum, chrome, lead, and copper were extracted through acid attack and analyzed in agreement with UNI EN ISO 11885-2009. Silicon was extracted through alkaline attack according to the UNI EN ISO 11885-2009 method, in agreement with ISTISAN 1996/34 [40]. Organic carbon content was determined according to ANPA (2001) [41], and the nitrates (NO_3) were analyzed in agreement with EN 12014-2 2018.

2.4. Statistical Analysis

The growth tests were designed to provide four repetitions for every condition at the end of the growth tests. Every repetition corresponded to a pot with 5 seeds of lettuce, a total of 48 pots, to guarantee sufficient replicates for the analyses of the results, including the exclusion of lettuce plants that germinated but did not undergo further development. The design of the experiment ensured statistical relevance in order to elaborate the results. The experimental data were statistically analyzed using the Student's *t*-test and an analysis of variance (ANOVA), with a confidence interval of 95% (p-value < 0.05). We also implemented the Siegel–Tukey test in order to compare different conditions and observe any significant differences among the repeated sample tests.

3. Results

The addition of the APV50 and APNUT C-PIMs under the two different LED lighting conditions had a significant impact on the growth of baby lettuce, as compared to the plants grown without fertilizers. Regarding the shoot dry weight (SDW), our results indicated, as shown in Figure 1, that the highest number of C-PIMs in the substrate corresponded to the highest biomass production under most conditions. Both the APV50 and APNUT C-PIMS had significant results on plant growth when exposed to LED-1 and with 6 granules in the substrate. APV50 had a similar positive effect, with 3 or 6 granules, as the dry biomass increased by 63% and 60%, respectively. The same effect was not observed with LED-2, and only the 6 granules of APV50 correlated to a significant increase (+30%) in SDW. On the 28th day after sowing, the growth of the baby lettuce with the APNUT fertilizer was significantly higher under LED-1, and only in the samples with 6 granules (+34%).

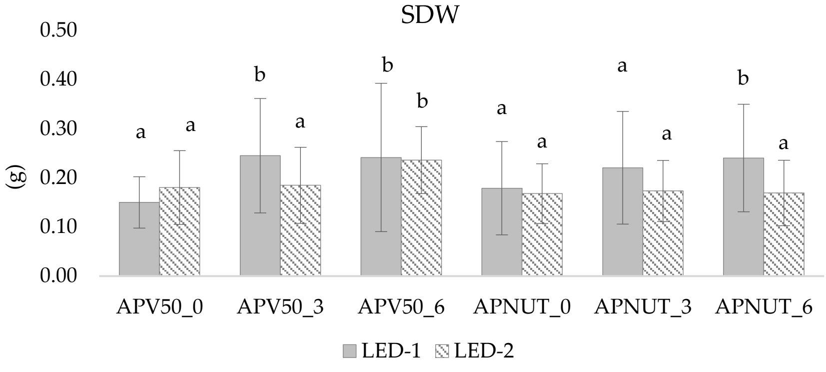

Figure 1. Shoot dry weight (SDW) of lettuce plants exposed to LED-1 and LED-2 lighting conditions. Statistically significant ($p < 0.05$) differences in the values are marked with different letters.

The root dry weight (RDW), however, had no significant difference among the fertilized and controls samples, under both lighting conditions (data not shown).

The germinability rates (GI) are reported in Table 2. The percentages were consistently high (approximately 90%, on average), and there were no significant differences among the different conditions.

Table 2. Mean germinability rates (%) ± standard deviation (SD). All condition and all lighting conditions are considered.

LED-1					
APV50_0	**APV50_3**	**APV50_6**	**APNUT_0**	**APNUT_3**	**APNUT_6**
93.3 ± 11.5	86.7 ± 10.3	88.0 ± 11.0	86.7 ± 11.5	93.3 ± 16.3	84.0 ± 6.7
LED-2					
APV50_0	**APV50_3**	**APV50_6**	**APNUT_0**	**APNUT_3**	**APNUT_6**
100.0 ± 0.0	91.4 ± 22.7	88.6 ± 10.7	90.0 ± 11.5	97.1 ± 7.6	97.1 ± 7.6

The different lighting schemes produced different effects on plant height, as shown in Figure 2. While the greatest amount of biomass was obtained with LED-1, LED-2 appeared to stimulate the plant height more than LED-1, as shown in Table 3. The mean height of plants grown under LED-2 was consistently higher than the heights of plants grown under LED-1 (Table 3). Moreover, the height of the plants grown with the addition of the aggregates was greater, as compared to the respective controls exposed to the same type of light. In the case of LED-1, the samples with 3 granules as well as those with 6 granules showed a significant increase in plant height (p-value < 0.05). In the case of LED-2, the increase in height, as compared to the relative controls, was significant only in those samples with 6 granules of APV50, while for APNUT, it was consistent for both quantities of fertilizers added.

Figure 2. Comparison of control and treated samples: (**a**,**b**) represent control pots with exposure to LED-1 and LED-2, respectively; (**c**,**d**) represent APV50_6 pots with exposure to LED-1 and LED-2, respectively.

Table 3. Mean value of the heights of lettuce plants grown under LED-1 and LED-2 conditions, and different numbers of C-PIMs granules. S.D.: Standard deviation. Means followed by different letters significantly differ at $p < 0.05$.

LED-1	**1_APV50_0**	**1_APV50_3**	**1_APV50_6**	**1_APNUT_0**	**1_APNUT_3**	**1_APNUT_6**
Height (cm) ±S.D.	4.2^{a} ±1.0	5.6^{c} ±0.9	5.9^{c} ±0.8	3.7^{a} ±1.0	6.1^{c} ±1.0	6.7^{c} ±1.3
LED-2	**2_APV50_0**	**2_APV50_3**	**2_APV50_6**	**2_APNUT_0**	**2_APNUT_3**	**2_APNUT_6**
Height (cm) ±S.D.	7.5^{b} ±1.4	8.5^{bd} ±1.0	9.0^{d} ±1.1	6.6^{bc} ±0.9	8.1^{d} ±1.1	8.2^{d} ±1.2

However, the analysis of the SDW/H rates highlighted that both 3 and 6 granules of fertilizer induced an increase in plant height, regardless of the lighting (LED-1, LED-2) or the amount of C-PIMs (APV50, APNUT) applied (Figure 3).

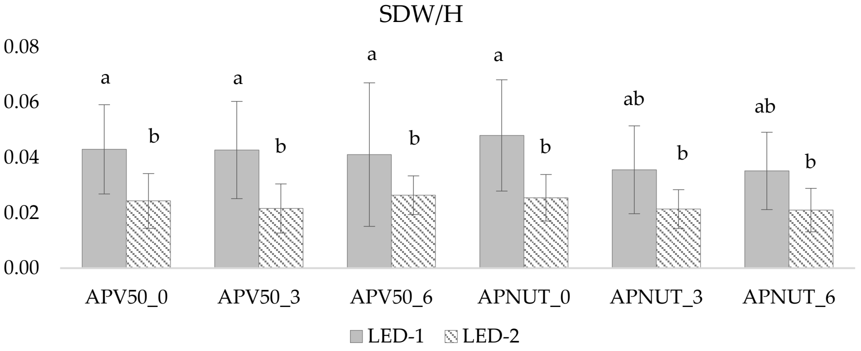

Figure 3. Rates between shoot dry weight and height in lettuce with different levels and types of fertilizer and exposed to LED-1 and LED-2 lighting conditions. Statistically significant ($p < 0.05$) differences in the values are marked with different letters.

The analysis of the leaf area provided interesting results in terms of productivity: On average, the plants under LED-2 conditions developed wider leaves than those grown under LED-1 (p-value < 0.05). Furthermore, the addition of 3 or 6 C-PIM granules was correlated with a greater leaf area, as compared to the control plants, under each studied condition (Figure 4). An example is shown in Figure 5, where the leaves of two control plants exposed to different lighting conditions were compared. In detail, the mean of the leaf area obtained under LED-1 was 15.9 cm^2 and 17.1 cm^2 for the APV50 and APNUT samples, respectively, while the mean area obtained in the control samples was 10.3 cm^2 and 10.0 cm^2, respectively. The values increased with LED-2 exposure: The mean results were 21.1 cm^2 and 20.8 cm^2 for APV50 and APNUT, respectively, while the controls were 16.8 cm^2 and 14.8 cm^2, respectively.

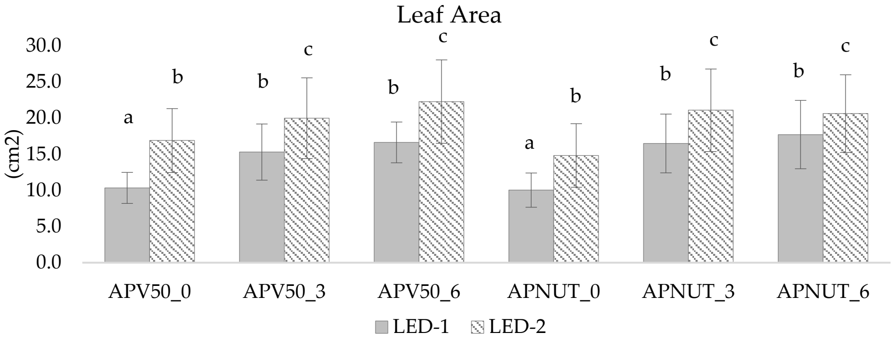

Figure 4. Mean leaf area in lettuce with and without fertilization, using both types of C-PIMs, under LED-1 and LED-2 lighting conditions. Statistically significant ($p < 0.05$) differences are marked with different letters.

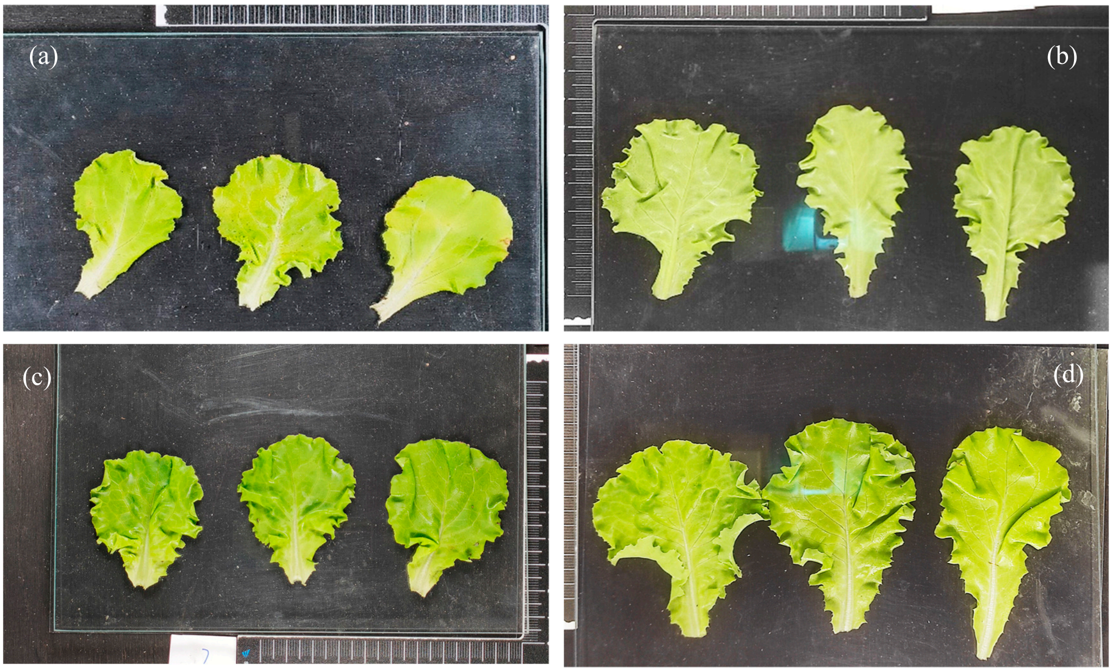

Figure 5. Leaves used for leaf area index: (**a**) and (**b**) represent control leaf samples from test with exposure to LED-1 and LED-2, respectively; (**c**) and (**d**) represent APV50_6 leaf samples from test with exposure to LED-1 and LED-2, respectively.

The quantification of the pigments did not provide any significant outcome. The chlorophyll *a* and *b* and the carotenoids content remained unchanged, regardless of the lighting conditions or the type or amount of C-PIMs (Figure 6).

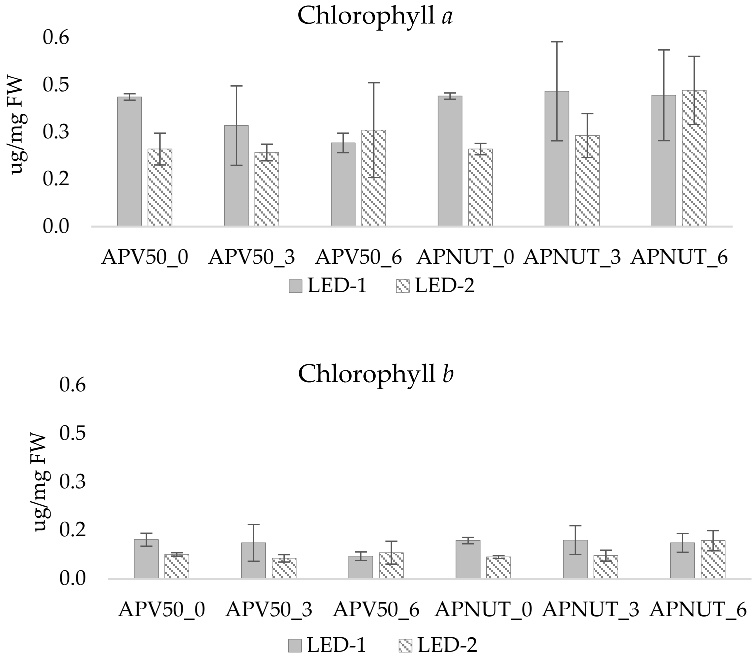

Figure 6. Mean pigment amounts (µg/mg of fresh weight) extracted from baby lettuce leaves. Chlorophyll *a*, *b*, and total carotenoids content is shown. Values obtained through testing of samples exposed to LED-1 were compared with those exposed to LED-2. Not all results showed significant differences.

The data analysis of the chemical elements before and after plant growth had significant results. First, the amount of K and NO_3 in the soil consistently decreased during the plant's growth (Table 4), in all the analyzed samples. The Si content was reduced in the soil exposed to LED-1 but was much higher in all substrates exposed to LED-2. A similar trend was observed for the K content. The P content had slightly decreased in the substrates exposed to LED-1.

Table 4. Chemical analysis of substrate before (and without C-PIMs (t0)) and at the end of lettuce growth under different experimental conditions: Phosphorous, potassium, nitrates and silicon contents are reported.

		P (mg/kg)	**K (mg/kg)**	**Si (mg/kg)**	**NO_3 (mg/kg)**
	t0	320	1254	1381	85.7
LED-1	t0LED-1	160	305	19.2	42.9
	tAPV50_3	169	208	94	55.8
	tAPV50_6	208	244	111	46.8
	tAPNUT_3	201	168	117	65.8
	tAPNUT_6	288	254	286	39.6
LED-2	t0LED-2	226	615	117	264
	tAPV50_3	431	547	534	174
	tAPV50_6	280	540	655	227
	tAPNUT_3	351	517	466	224
	tAPNUT_6	288	519	381	260

The presence of chemical elements and heavy metals in the lettuce leaves is shown in Table 5. Chrome was consistently <1 mg/kg; lead was <0.3 mg/kg; and copper was consistently <3.6 mg/kg, while aluminum content was consistently <4.0 mg/kg of fresh biomass. The amount of nitrates was consistently less than the legislated thresholds of <500 mg/kg of fresh biomass, as defined by European Regulation 1258/2011 [42].

Table 5. Amounts of elements and heavy metals in lettuce leaves exposed to LED-1 and LED-2 (mg/kg). Mean and standard deviation of at least two replicate samples are presented.

		LED-1					
		APV50_0	**APV50_3**	**APV50_6**	**APNUT_0**	**APNUT_3**	**APNUT_6**
%	C	4.2 ± 0.1	4.4 ± 1.5	3.2 ± 0.3	5.2 ± 1.1	3.9 ± 1.3	3.3 ± 0.9
mg/kg	NO_3	<200	<200	<200	<200	<200	<200
mg/kg	Al	3.1 ± 0.8	2.7 ± 1.0	2.3 ± 0.7	3.2 ± 0.0	2.7 ± 0.2	2.3 ± 0.3
mg/kg	Si	73.0 ± 46.6	76.3 ± 22.3	61.6 ± 20.7	74.3 ± 1.1	62.6 ± 6.2	53.0 ± 18.8
mg/kg	Cr	<1.0	<1.0	<1.0	<1.0	<1.0	<1.0
mg/kg	Pb	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
mg/kg	Cu	<1.0	<1.0	<1.2	<3.6	<1.0	1.3
		LED-2					
		APV50_0	**APV50_3**	**APV50_6**	**APNUT_0**	**APNUT_3**	**APNUT_6**
%	C	3.5 ± 0.2	3.1 ± 0.3	3.1 ± 0.4	3.1 ± 0.3	2.8 ± 0.3	2.9 ± 0.3
mg/kg	NO_3	<200	<200	<200	<200	<200	<200
mg/kg	Al	1.5 ± 0.8	1.1 ± 0.1	1.0 ± 0.2	0.9 ± 0.4	1.7 ± 0.9	1.2 ± 0.1
mg/kg	Si	44.5 ± 8.8	56.9 ± 9.8	64.5 ± 17.5	56.9 ± 8.1	72.9 ± 25.1	58.0 ± 22.6
mg/kg	Cr	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5
mg/kg	Pb	<0.3	<0.3	<0.3	<0.3	<0.3	<0.3
mg/kg	Cu	<1.4	<1.0	<1.0	<2.1	<1.0	<1.0

4. Discussion

This work investigated the effects of multiple environmental factors on indoor horticulture, including the exposure to two different lighting conditions and the addition of nutrient-enriched aggregates in the substrate. Each of these factors was studied, elaborated, and then applied in order to optimize plant growth by following the principles of a circular economy [43]. The replacement of 2/3 of the peat normally as the substrate with agriperlite in indoor cultivation represented the first step towards reducing the environmental impact of the peat extraction process. Peatlands are rich in soil-based carbon, carbon dioxide, and atmospheric methane. Since peat has a very slow regrowth rate, it is classified as a solid fossil by the Intergovernmental Panel on Climate Change (IPCC) and greenhouse gas (GHG) emissions, comparable to that of fossil fuels [44]. For these reasons, replacing peat in horticulture applications is essential to mitigate the negative impact. At the same time, the use of organic waste and byproducts (e.g., spent coffee grounds and animal bonemeal ash) to formulate fertilizing aggregates is a sustainable alternative to synthetic chemical fertilizers [45]. The use of LED lighting reduces the energy demand of indoor cultivation, which is an increasingly popular practice in urban areas.

The increase in dry biomass obtained in *Lactuca sativa* growing in substrates enriched with C-PIMs indicated the positive effects of these experimental fertilizers on the plants, and our results agreed with previously reported preliminary results obtained using only six aggregates of APNUT and APV50 to grow lettuce in a simplified system [11]. Other parameters related to the growth promotion of the fertilizers, including the leaf area and plant height, were recorded in this study.

The application of six APV50 aggregates was consistently correlated with increases in shoot dry weight, under both LED schemes. The results were not the same with APNUT, with no significant weight improvements in the samples exposed to LED-2. The main biomass production by lettuce was achieved when exposed to LED-1. These results were consistent with data on the soil chemical content. Two of the main nutrients of the substrate (K and NO_3) drastically decreased in the substrate of the samples exposed to LED-1, as compared to those exposed to LED-2. These findings suggested an improvement in the use of nutrients by plants when exposed to LED-1, which achieved the highest dry biomass production with a corresponding adsorption of nutrients from the soil. LED-2 exposure obtained the best performance, as compared to LED-1, in terms of plant height and leaf area, which were positively correlated with both C-PIMs. Therefore, the higher values obtained in substrates with APV50 aggregates that were exposed LED-1 indicated that this lighting scheme enhanced the biomass growth, while LED-2 promoted plant height. The differences are shown in Figures 2 and 5, in which the APV50 control plants grown under LED-1 and LED-2 are compared.

These differences were likely due to the different lighting schemes, since light is a critical factor that drives photosynthesis and regulates plant morphology, physiology, and phytochemical content [28]. Red and blue LED lighting also have a significant function in driving photosynthesis; however, plants in nature have adapted to utilize a wide spectrum of light in order to control photo-morphogenesis [24]. Several previous studies described the behavior of lettuce grown under different LED wavelengths, specifically identifying their effects on different agronomic parameters [46–48]. It is well known that red light induces the hypocotyl elongation and the cotyledon expansion in seedlings. Previous findings have emphasized the height of lettuce plants could be significantly increased with supplemental far-red treatments [34], as applied through LED-2 exposure. The LED-2 scheme contained a consistent percentage of far-red in the first growth week and maintained this wavelength (at different percentages) until the end of the test. Moreover, the supplemental white light (contained in LED-2) could be strategically used to enhance agronomic growth parameters (e.g., leaf area) in baby leaf lettuce, as previously described in [34]. Importantly, the elemental compositions of the lettuce leaves for all the analyzed samples in each test were largely compliant with the legislated thresholds. The European Regulation 1881/2006 [49] and its successive amendments (2021/1317) defined all legislated limits that had to be observed to consider a vegetable suitable for sale and human consumption. In detail, the contents of heavy metals were below the legislated limits defined in [42]. The nitrate contents were consistently below the legislated thresholds defined in European Regulation 1258/2011 [42].

5. Conclusions

In recent years, new applications in horticulture have involved cultivation systems for urban use. A major challenge for these crop-cultivation systems is to increase their sustainability and efficiency in terms of resource demand. This study investigated the effects of two different LED lighting schemes with two types of lightweight aggregates, derived from agri-food waste and applied as sustained-release fertilizers, on the growth of *Lactuca sativa*.

The results of the study demonstrated that the use of C-PIMs had satisfactory effects on the lettuce growth and increased the production of the biomass, as compared to the controls, thought increasing the number of C-PIMs applied to the soil did not consistently increase all agronomic parameters. The application of the first LED lighting scheme, based on the combination of blue and red wavelengths at a 50/50 ratio, demonstrated good performance in biomass production, while the second LED scheme, which added green, far-red, and white wavelengths, had better results in height and leaf-area development.

The circularity of resource use in this study provided an example of the valorization of agri-food waste products in the horticultural field, specifically in urban/domestic contexts. Our results highlighted the excellent potential of these new types of fertilizers for soilless cultivations, using and recycling organic and inorganic materials. Future studies could also consider the economic evaluation of the implemented system using a life-cycle-assessment approach. Further investigations are needed to verify that the proposed system is also effective for other plant species of horticultural or floricultural interest, but the current results indicated our system should be scalable for sustainable cultivation and production with a significantly lower environmental impact.

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Data Availability Statement: The data presented in this study are available upon request from the corresponding author. The data are not publicly available due to founding regulations.

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