



## Development of Lettuce Plant in Spring and Autumn Period, Effects of Led Lightening on the Quantity of Mineral Substrates and Leaf Nitrate

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### Authors' contributions

This work was carried out in collaboration between both authors. Author SŞ designed the study, wrote the protocol and wrote the first draft of the manuscript. Authors SBA and SŞ reviewed the study design and all drafts of the manuscript. Author SBA managed the analyses of the study and performed the statistical analysis. Author SŞ managed the literature searches. Both authors read and approved the final manuscript.

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### ABSTRACT

**Aims:** This study was performed during early spring and winter period of 2015-2016 by utilizing soilless technique in a non-heated glasshouse that belongs to Gaziosmanpaşa University Faculty of Agriculture.

**Study Design:** Whereas Funly F1 lettuce species was used as vegetal materials, 2:1 ratio cocopeat and perlite mixture was used as cultivation environment. According to the experiment design, experiment coincidence parcels were performed as 3-recurrences. In the experiment, the effects of different colored LED lights (blue, yellow, red, blue + yellow, blue + red, yellow + red, blue + yellow + red) additional to sunlight were examined.

**Results:** SMD strip LEDs with different colors were used as light source. The light practice does not affect on the plant diameter, plant length, SÇKM, pH, titered acid, vitamin C and plant nutrient

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concentrations. Statistically significant difference occurred in the yield of spring and winter curly leaf head salad. There was also an increase at 1% importance level in the light practices compared to the control. In the experiment, red and red blue light combinations had an increase of 1% in the curly leaf head salads in the yield rate when it was compared to the control. Whereas the highest total plant head weight was 840 gr/piece in spring practice, and it was 732 gr/piece for the red light practices in winter practice. Compared to the control, the amount of plant leaf nitrate for the light practices resulted in a decrease at 1% importance level in the curly leaf head salad. When the results were compared with the control conditions, the lowest nitrate contents were obtained as 1764.5 mg NO<sub>3</sub><sup>-</sup>kg<sup>-1</sup> in spring practices, 1898.6 NO<sub>3</sub><sup>-</sup>kg<sup>-1</sup> in winter practices.

**Conclusion:** More amount of nitrate was observed on the leaves of curly leaf head salad in winter practice compared to the one in spring. The amount of leaf nitrate decreased in the light practices compared to the control. As a result, the red and blue light practices and their combinations improved the amount of yield and plant growth by reducing nitrate content.

*Keywords: Soilless culture; led light; curly lettuce.*

## 1. INTRODUCTION

Upon the increasing population ratio, the demand for food increases, and so the producers desire to harvest more products from their production fields. While using the agricultural fields to harvest more products at the maximum level, as a result of chemical fertilizers, pesticides, wrong cultural practices, or etc., the agricultural fields become inefficient. As a result of this, even though product growth is expected, it is resulted in product losses. This has led the producers to conduct new technological studies in agriculture. One of these agricultural practices is the soilless agriculture.

Soilless agriculture has such advantages as to produce in fields where the soil is not suitable for plant production, to increase the efficiency of water use, to provide the plants to be fed in a controlled way, to increase yield, to improve quality, to reduce the necessary labour force, to facilitate irrigation, and to provide facilities that can be made without the need for environment sterilization. Soilless culture has also such superiorities as the absence of problems such as soil fatigue, soil-borne diseases and pests, controlling the development of plants by examining the fertilizer and water, eliminating the soil-based factors that reduce the quality, and increasing yield [1]. The purpose of soilless agriculture is to provide the development of plants through the nutrient solution, to meet the nutrient and water requirements of the plants without causing stress, and to realize this in the most economical way.

Cocopeat is one of the cultivation environments that is widely used in soilless agriculture. Cocopeat is a fibrous organic substance obtained from coconut (*Cocos nucifera*) which is

a type of palm grown in tropical regions. It is made out of the shell of coconut fruits. Whereas the long fibres are used for various purposes (rope, wicker, basket, or etc.), the rest is composted into heaps, and the coconut turf is produced.

Coconut fibre powders including good physical features are suitable for being used as cultivation environment. Coconut fibre powders can be used because of such features as their high water holding capacity, easily-absorbable water content, sufficient air capacity, low volume content and nutrient element content, being used without any chemical treatment, and being mixed with peat physically and chemically.

Another positive feature of coconut fibre powders is the high content of lignin and cellulose. Therefore, it can maintain air and water balance for a long time. It contains about 20% air by volume, and the pH value is determinedly saved. On the other hand, the most important negative side of the cultivation environment is the use of sea water in processing the green shells of it. As a result, the salt content increases owing to Na and Cl (Anonymous, 2014).

Greenhouse cultivation carried out in our country is expanded and developed in Marmara Region, Aegean Region, and Mediterranean coastline. Dense cultivation fields are occurred in this distribution. The greenhouse cultivation is observed in the microclimate around Yalova on the northern and is concentrated around İzmir and Mugla in the west, around Antalya and Mersin in the south, and reaches to Samandag district of Hatay province. When the recent distribution of greenhouse in our country is numerically examined, it is concluded that approximately 65% of greenhouse fields is

located in Antalya, 21% in Mersin, 7% in Mugla, 2% in Izmir, and 1% in Istanbul.

The main harvest in greenhouse production is targeted in the summer period. The cultivation process is prolonged due to the insufficient temperature and frost since the crops grown at the beginning of autumn and spring period do not get enough light during these periods, and yield losses may occur. This situation causes the financial losses for farmers. It is possible to reduce the losses during these periods by taking some cultural measures. Making artificial lightening in the environments where the plants are grown and creating cultivation environments (organic and inorganic) that can hold water and mineral substances in the root part of the plant are among the cultural measures that can be taken.

All living creatures across the world benefit from the rays coming from the Sun. As a result of photosynthesis, the plants benefit from these rays by transforming the physical energy of the Sun into chemical food energy in organic substance. Lack or excess of light intensity has a significant effect on the metabolic functions in plant. If light is less than the necessary intensity, a series of metabolic changes follow the reduction of carbohydrate content in plants. In case of light deficiency, the shadow plants expand the leaf surfaces and slow down root growth by sending photosynthesis products less than the required ratio. The slowdown in root growth negatively affects the nutrient utilization of plants [2].

Light sources are divided into two as natural and artificial. Whereas the light from the Sun is stated as natural light, the light emitted from artificial lightening sources is defined as artificial light. Such light sources as fluorescent lamps, metal halogen lamps, and light emitting diodes (LEDs) are some examples of artificial lightening sources [3]. It is necessary in artificial lightening to know the total amount of natural light reaching the plant during the day and the duration of sunbathing. These parameters are important for both photosynthesis and photo-period lightening.

The positive effect of light on plant growth has brought about the use of additional light in greenhouse cultivation. Likewise, that the latest technological greenhouses have the ability to produce their own energy and to use this energy for lightening in the greenhouse has led to an increase in the light-plant development studies.

Samuoliene, et al. (2009), lettuce and onion 3-day before the harvest have made additional lighting. The researchers reported that the amount of leaf nitrate decreased by 44% and 65% with additional lighting. Macedo, et al. [4] in vitro *Alternanthera brasiliana* Kuntze plant red light reduces plant growth while the blue light leaf, plant number and leaf surface increased reported. Chen, et al. (2014) reported that the most appropriate light for the development of lettuce plant is 24-hour red-blue led lighting. Wojciechowska, et al. [5] applied LED lighting to lettuce plant in the middle of the day and in the evening. They reported that the amount of leaf nitrate decreased compared to the control.

When the light intensity decreases especially during winter months, additional lightening has significant effects on plant growth [6,7,8]. Since they have various advantages, the use of LED lights has become widespread in recent years for additional lightening to be carried out before sunrise and / or after sunset [9-13]. LED lightening also allows the plant to grow even in sunless hours [14-17].

According to the growth stages of the plants, the use of LED lights in different colours has started to become widespread in the greenhouse vegetable cultivation. For this purpose, red and blue LED light-emitting lamps are widely started to be used and sold [12]. LED lamps provide lightening without damaging the plants because they do not emit ultraviolet or infrared radiation and the system does not contain mercury and lead (Anonymous, 2011).

Many out-greenhouse factors such as cloudiness, air pollution, fog, various precipitation forms and regimes, high proportional humidity, pollution of greenhouse cover, reflection due to roof slope, type of cover, and radiation absorption cause the changes in the amount of PAR (Photosynthetic Active Radiation) and wavelength during the greenhouse cultivation period. In addition, such in-greenhouse factors as shade curtains used in greenhouses, metal building components used in greenhouse construction, ventilation fans, lightening units, and heating units lead the radiation energy entering into the greenhouse to be reached onto the leaves of the plant at different amounts [18,19]. Moreover, the light level in the greenhouse decreases by 35-75% depending on many factors such as solar radiation incidence angle, length of the day, sunshine duration, cloudiness, structural shading, plant density,

cover material, and pollution status [20]. Therefore, shading should be applied in plant cultivation in greenhouses through the help of complementary photosynthetic lightening (CPL), full artificial photosynthetic lightening (FAPL) or shade curtains [19]. However, most of the lightening-shading practices are carried out in the form of a uniform lightening practice by assuming that all plants in the cultivation environment need the same PAR amount at the same time [21]. Artificial lightening gives positive results in the regions with an average sunshine duration less than 4.5 hours (Argus, 2010).

The purpose of this research is to determine the effects of artificial lightening implemented on lettuce plant cultivated in spring and autumn period under the conditions of hydroponics in an unheated glass greenhouse on plant growth, mineral matter concentration, and nitrate content on the leaves of plants.

## 2. MATERIALS AND METHODS

### 2.1 Plant Material and Cultivation Conditions

This study examining the effects of additional LED lightening practices in curly leaf salad and pepper cultivation in solid environment culture on the yield quality and plant growth was conducted in a glass greenhouse located in Gaziosmanpasa University Faculty of Agriculture Implementation and Research in Tokat Central District during early spring and summer period in 2014-2015. The dimensions of the single-roofed greenhouse established in the east-west direction are 12 x 35 (420 m<sup>2</sup>) with a side height of 2.2 m and a roof height of 5 m. The practice was designed in randomized plot design with 3 replications [22].

As lettuce variety, Funnly F1 was used in the practices. A 2:1 mixture of cocopeat (coconut turf) and perlite, which are widely used in soilless agriculture, were used as plant cultivation environment.

A total of 72 pedestal rigid PVC pots with a depth of 18 cm, an internal width of 20 cm, an inner length of 71 cm, and a volume of approximately 25.6 litres were used in order to grow the plants. The pots were arranged in such a way that the distance between the rows is 1 m. 12 pots were arranged on the row, but 9 pots were put into the practice, and the other 3 pots were accepted as edge effect. The floor of greenhouse was cleaned and covered with pebbles so as to disconnect the pots from the soil. The blocked

cocopeats were saturated with water and mixed with perlite in a 2:1 ratio to obtain a homogeneous mortar. The pots were made ready for planting by filling the mortar into them. Drip irrigation system was applied to irrigate the plants. The drip irrigation system were operated from the tank containing 2 tons of nutrient solution through the electrotopomp system.

### 2.2 Light Practices

3 different LED light sources such as red, blue, and yellow were used in this research. 4 rows of LED lights were placed on each parcel. Accordingly, the total amount of LEDs used = number of parcels x number of LED rows in the parcel x length of parcel = 21 x 4 x 2.25 = 198 m. These LEDs were operated through an adapter and a timer.

The LED strip factors were investigated in this practice. The levels of LED colour factor discussed were blue LED, yellow LED, red LED, blue + yellow LED, blue + red LED, yellow + red LED, blue + yellow + red LED, and control.

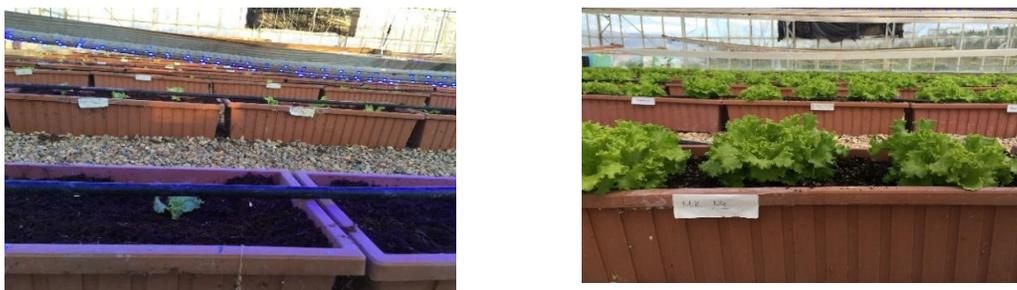
15 days after planting the seedling, LED strip was glued under transparent flexiglass strips, and these plates were hung on the roof with gut and placed 40 cm above the plants. The LEDs were operated between 7-24 hours. Empty transparent flexiglasses were placed on the control plots (Fig. 1).

### 2.3 Planting, Maintenance and Harvest Operations

First stage curly leaf salad seedlings cultivated in a ready-made turf were planted on March 16, 2015. Second stage curly leaf salad seedlings were planted as 3 plants in 1 pot and 9 plants in parcel on December 1, 2015. A total of 216 plants were grown in 24 parcels.

Hoagland nutrient solution containing all the necessary nutrients for plant growth (N 150 mg / l, P 60 mg / l, K 150 mg / l, Ca 150 mg / l, Mg 50 mg / l, Fe 5 mg / l, Mn 0,5 mg / l, Zn 0.05 mg / l, B 0.5 mg / l, Cu 0.03 mg / l, Mo 0.02 mg / l) were given to the plants by drip irrigation system. The electrotopomp was operated 3 times a day for 10 minutes (at 10, 13 and 16 o'clock) by using a 15-minute precision time clock. The fungal diseases in plants were fought by applying fungicide.

The first stage curly leaf salads were harvested on May 8, 2015 when they were in variety size,



**Fig. 1. Light applications to lettuce plant**

and the second stage curly leaf salads were harvested on February 25, 2016.

## 2.4 Chemical Analysis

**Vitamin C (mg/100 g):** The content of Vitamin C was determined through the spectrophotometric method.

**Water Soluble Dry Substance (WSDS) (%):** It was determined by measuring through hand refractometer from fruit juice obtained by shredding of fruits with solid juicer.

**pH:** It was measured with the help of pH meters. The plant samples taken separately for each parcel were grinded in a blender so as to obtain water, and the obtained plant juice was measured through pH meter.

**Titrateable acidity (%):** pH was calculated through the metric method. 1 ml of fruit juice was taken for each variety, and 50 ml of purified water was added into it. 1 or 2 drops of phenol phthalein were dropped to provide colour transformation. Then, the juice samples were titrated with 0.1 N NaOH until pH 8.1, and the ratio of spent sodium hydroxide was determined for this practice. The calculations were made in citric acid.

**Nitrate analysis:** According to Cataldo et al. [23], it was carried out through the spectrophotometric method.

**Amount of chlorophyll:** Chlorophyll in the leaves was measured through a chlorophyll meter during the harvest period.

**Light Intensity:** The light intensity of the practices was measured through a planimeter.

## 2.5 Statistical Analysis

(ANOVA) SPSS (Version 12.00; Chicago, IL, USA) statistical software was applied for

evaluation of the data obtained in the practice and analysis of variance. The comparison of the means was made according to Duncan test at  $P \leq 0.05$  level.

## 3. RESULTS AND DISCUSSION

### 3.1 Effect of Light Practices on Yield, Some Properties and Leaf Nitrate Content of Lettuce Plant

In this study in which different coloured LED lights and their combinations were used, yield and some properties of lettuce plant in spring period are given in Tables 1 and 2. Whereas the plant head weights were 804 g/pc in the control practice, the highest head weight was obtained in the red and yellow light practices. The head weight was reduced in the light combined with blue and red. Light practices were effective at 1% significance level. The effect of light practices on plant diameter and plant height of lettuce was not statistically significant. Plant diameter was 32.6 cm in yellow light practice and 32.5 cm in blue light practice. While the average plant height was 28.2 cm, the highest lettuce lengths were obtained in the control practice and mixture of blue and red.

The effect of light practices on the WSDS, titrateable acid, pH, vitamin C, and chlorophyll content of leaves of lettuce plant was not determined to be statistically significant. Whereas the amount of water soluble dry substance of the lettuce cultivated during the spring period was 5.4%, it was close to this amount in other practices. While the titrateable amount of lettuce plant was 1.74% on average, it was measured that the pH values were 6.02 on average and that vitamin C amount was 11.3 mg/100 g.

Whereas the average head yield of spring cultivation was 823 gr/pc, it was 703 gr/pc in

autumn cultivation. It is thought that this yield difference is due to growing time, temperature difference in greenhouse, and differences in lightening times. Due to the high light and temperature in the spring, it increased photosynthesis through biomass and vegetative components and therefore increased the growth.

In this study in which different coloured LED lights and their combinations were used, yield and some properties of lettuce plant in autumn cultivation are presented in Tables 3 and 4. Whereas the plant head weights were 650 g/pc in the control practice, the highest head weight was obtained in red and yellow-blue light practices. The weight of the head was decreased in light carried out in the control practice. Light practices were effective at 1% significance level. The effect of light practices on plant diameter and plant height of lettuce was not statistically significant. Plant diameter was measured as 33.2 cm in blue-red light practice, and it was 33.2 cm in blue-yellow-red light practice. While the average plant height was 26.4 cm, the highest

lettuce lengths were obtained in the control practice and mixture of blue and red.

The effect of light practices on the WSDS, titratable acid, pH, vitamin C, and chlorophyll content of leaves of lettuce plant was not determined as statistically significant. Whereas the amount of water soluble dry substance of lettuce cultivated in the spring period was 5.2%, it was close to this amount in other practices. While the titratable amount of lettuce plant was 1.73% on average, it was measured that the pH values were 5.69 on average and that the vitamin C amount was 12 mg / 100 g. Chlorophyll values of lettuce leaves were periodically examined, and the average is showed in Table 4.

### 3.2 Comparison of Spring and Autumn Cultivation in terms of Yield and Leaf Nitrate Amount

Numerous factors determine the amount of leaf nitrate. The most effective of these factors is light. The nitrate content of lettuce leaf cultivated

**Table 1. The effect of light practices in spring cultivation on head weight, plant diameter and height of lettuce**

Light practices	Plant head weight (g / piece)**	Plant diameter (cm)	Height of plant (cm)
Control	804 c	31.3	30.5
Blue	820 c	32.5	28.3
Yellow	828 c	32.6	27.6
Red	840 a	31.9	27.3
Blue + Red	836 b	32.6	30.7
Yellow + Blue	825 c	32.5	27.9
Red + Yellow	814 d	32.3	27.2
B + Y + R	820 c	31.9	26.5
Mean	823	32.2	28.2

The differences between the means in each column were determined through Duncan test. N.I: Not important; \* P <0.05; \*\* P <0.01 is important

**Table 2. The effect of light practices on WSDS, pH, titratable acid, vitamin C, chlorophyll content of lettuce leaves cultivated in Spring period**

Light practices	WSDS (%)	TA (%)	pH	Vitamin C (mg/100 g)
Control	5.4	1.64	6.05	10.6
Blue	5.8	1.75	5.93	11.6
Yellow	5.5	1.61	6.05	11.4
Red	5.4	1.64	6.06	12.1
Blue + Red	5.4	1.91	6.11	11.3
Yellow + Blue	5.4	1.68	6.04	11.5
Red + Yellow	4.9	1.73	5.98	11.2
B + Y + R	5.1	1.98	6.01	11.3
Mean	5.4	1.74	6.02	11.3

The differences between the means in each column were determined through Duncan test. N.I: Not important; \* P <0.05; \*\* P <0.01 is important

**Table 3. The effect of light practices in autumn cultivation on head weight, plant diameter and height of lettuce plant**

Light practices	Plant head weight (g / piece)**	Plant diameter (cm)	Height of plant (cm)
Control	650 c	32.3	27.3
Blue	670 bc	30.2	24.5
Yellow	720 b	32.2	26.5
Red	732 a	32.5	26.4
Blue + Red	723 b	33.2	28.6
Yellow + Blue	732 a	32.4	25.6
Red + Yellow	710 b	32.1	26.5
B + Y + R	690 bc	33.2	25.9
Mean	703	32.3	26.4

The differences between the means in each column were determined through Duncan test. N.I: Not important; \*  $P < 0.05$ ; \*\*  $P < 0.01$  is important

**Table 4. The effect of light practices in autumn cultivation on WSDS, pH, titratable acid, vitamin C, chlorophyll content of lettuce leaves**

Light practices	WSDS(%)	TA (%)	pH	Vitamin C (mg/100 g)
Control	4.9	1.73	5.75	11.5
Blue	5.2	1.78	5.68	12.1
Yellow	5.1	1.71	5.71	11.9
Red	5.1	1.69	5.72	11.8
Blue + Red	5.2	1.74	5.73	12.5
Yellow + Blue	5.2	1.67	5.66	11.9
Red + Yellow	5.4	1.74	5.64	11.9
B + Y + R	5.6	1.78	5.61	12.6
Mean	5.2	1.73	5.69	12.0

The differences between the means in each column were determined through Duncan test. N.I: Not important; \*  $P < 0.05$ ; \*\*  $P < 0.01$  is important

**Table 5. The effect of light practices on nitrate content of lettuce leaves in Spring and Autumn cultivation**

Light practices	Spring period nitrate content** mg NO <sub>3</sub> <sup>-</sup> kg <sup>-1</sup>	Autumn period nitrate content** mg NO <sub>3</sub> <sup>-</sup> kg <sup>-1</sup>
Control	1880.5 a	2008.8 a
Blue	1784.5 b	1910.8 b
Yellow	1794.8 b	1920.3 cd
Red	1772.5 d	1924.6 cd
Blue + Red	1775.8 d	1950.6 c
Yellow + Blue	1785.5 c	1898.6 d
Red + Yellow	1784.7 c	1954.6 c
B + Y + R	1764.5 d	1978.3 c
Mean	1792.8 B	1968.3 A

The differences between the means in each column were determined through Duncan test. N.I: Not important; \*  $P < 0.05$ ; \*\*  $P < 0.01$  is important

in spring and autumn is presented in Table 5. When the average of spring period and autumn period were examined, it was measured that the average lettuce leaves in autumn period was 1968.3 mg NO<sub>3</sub><sup>-</sup> kg<sup>-1</sup>, this amount was measured as 1792.8 mg NO<sub>3</sub><sup>-</sup> kg<sup>-1</sup> in spring period. There is a linear correlation between total biomass and nitrate content of the plant.

This result was acquired since it was tested through the light measurements that the plant received more light in the spring period. As the lightening time of the plant increased, nitrogenase enzyme increased, and leaf nitrate amount decreased. Some other studies have reported the same results in this direction. Different light practices varied in terms of quantity. In particular,

red light and its combinations significantly reduced the nitrate content. The lowest nitrate content in spring period was obtained in red light and in the combination of red and blue light. In autumn period, nitrate decreases were observed in the combination of yellow and blue light, yellow, and red light practices.

#### 4. CONCLUSION

In this research, it was observed that there was a yield difference between spring and autumn cultivation. The use of LED lights in crop production has increased the efficiency according to the control conditions. It is understood that the use of LED lights in agriculture and the necessity to study these kinds of studies in detail and the necessity with these and other studies. In winter (December, January, February, March) light intensity decreases, additional lighting is needed in greenhouse plant growing. In the studies conducted with led lighting, it has been reported that the effect of the lightings made during seedling period on growth and development is quite high [24,25]. This situation can be explained through the temperature difference and duration of lightening. On the other hand, in light practices, there was a difference in efficiency in practices where red light and its combinations were made according to control conditions. One of the main factors determining the nitrate content of plants is light. Nitrate contents in plants are influenced by light intensity, photoperiod and photoperiod. According to the researches, nitrate accumulation increased in low light conditions such as winter season and nitrate content decreased at high light intensities. Besides, significant decreases were observed in leaf nitrate contents compared to control conditions. Especially in the greenhouse conditions with cloudy autumn period and low duration of lightening, implementation of 7-24 light practices will cause the plant to carry out more biochemical activities. This also will lead to further development of vegetative components.

Even though technological innovations are offering a wide variety of possibilities and different solutions for growing plants almost everywhere, the energy demand is still the main limiting factor of these cultivation systems. For this reason, it is necessary to evaluate and consider the feasibility of these solutions in the proper context, thus economic and environmental studies will be needed to assay

the real sustainability of such innovative growing systems on a larger scale.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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