Chapitre III

Impact des radiations UV-C appliquées en pré-récolte sur la sensibilité du fraisier à différents agents pathogènes et sur la qualité/conservation des fruits en post-récolte

I. Lien pré-post récolte

Ce chapitre est rédigé sous la forme d'un article scientifique, soumis dans le journal « Post-harvest Biology and Technology » (Figure 33).

Impact of UV-C radiation applied during strawberry cultivation on the strawberry sensitivity and quality of the fruit

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Figure 33 : **Article soumis traitant du le lien pré-post récolte**. Cet article va être soumis au journal Post-Harvest Biology and Technology.

1. Abstract

Applications of UV-C radiation have proven very efficient in reducing the development of diseases in many species including strawberry (*Fragaria x ananassa*). Several studies suggest that UV-C radiation is effective not only because of its disinfecting effect but also because it may stimulate plant defenses. This study involved to apply UV-C radiation during strawberry cultivation and to evaluate its impact on the plant growth, the quality of the produced fruits and the susceptibility to major plant diseases during the plant cultivation and during the fruit storage. UV-C treatments have an impact on the flowering initiation, on the fruit development and on its conservation. Flowering occurs earlier for UV-C treated plant and consequently more fruits are produced at harvest despite a slight

decrease in leaf area. UV-C treatments did not improve the shelf life but did not alter the physical integrity of strawberry fruits. A strong decrease in sensitivity of leaves to powdery mildew and fruits to *Rhizopus* was also noticed for the UV-C treated plants.

Keywords: strawberry, UV-C, pre-post-harvest, phytopathology, quality

2. Introduction

Strawberry is a very popular fruit appreciated by consumers because of its sensory characteristics. Moreover, strawberries are one of the richest source of natural antioxydants and thus it has a high beneficial power on human health (Hannum, 2004). However, many microorganisms, including fungi, are very damaging for plant crops and also for the storage of plant products in post-harvest. Yields can be largely affected by these fungal attacks and strawberries are subjected to rapid degradation affecting fruit flavor (Perkins-Veazie, 1995). Restrictions on the use of synthetic fungicides make it necessary to find alternative phytosanitary solutions wich aim more at stimulating plant defense or their organs rather than destroying infectious agent. Among these solutions, physical methods applied before or after harvest can improve the resistance of plants to pathogens and can increase the synthesis of vitamins, micronutrients and secondary metabolites of fruit (Poiroux-Gonord *et al.*, 2010). Among these physical methods, application of non-deleterious low doses of ultraviolet-C (*UV-C*) radiation on fruits and vegetables during cultivation or post-harvest creates a moderate oxidative stress in plants that can increase the resistance to phytopathogenic agents (Charles *et al.*, 2008; Ouhibi *et al.*, 2015a,b; Vasquez *et al.*, 2017) and improve the nutritional quality of the product (Mercier *et al.*, 2001). For instance, it can induce partial resistance to the plant pathogenic fungus *Botrytis cinerea* on carrots, lettuce and tomatoes in postharvest (Charles *et al.*, 2008; Mercier *et al.*, 1993a,b; Ouhibi *et al.*, 2015a,b).

Application of UV-C radiation on plant organs induces defense mechanisms, increasing for instance the chitinase content in mango and peach, or increasing key enzymes such as SOD (*Superoxide Dismutase*) or PAL (*Phenylalanine Ammonia-Lyase*) (El-Ghaouth *et al.*, 2003; Gonzalez-Aguilar *et al.*, 2007; Yang *et al.*, 2014). UV-C radiation applied in post-harvest also induces secondary metabolite modifications in mango, grapes and peaches compared to non-treated fruits (Freitas *et al.*, 2015; Gonzalez-Aguilar *et al.*, 2004; Gonzalez-Aliguar *et al.*, 2001). UV-C treatments applied in post-harvest on strawberries have shown an increase of chitinase, PAL and antioxidant compounds (Erkan *et al.*, 2008; Mohammadi *et al.*, 2012; Pombo *et al.*, 2011). In addition to these compounds, UV-C radiation on strawberry fruits in post-harvest also causes variations in the activation of genes involved in the firmness of fruit thus allowing an improvement in the shelf life and in the production of volatile compounds giving strongly aromatic fruits (Severo *et al.*, 2015). These volatile compounds thus bring a taste interest and also play a role in the defenses of plants against pathogen.

In post-harvest, UV-C treatments are widely studied. However, very few studies have been carried out to evaluate the link between the treatment of plants during its cultivation with UV-C radiation and the quality of fruits produced. In general, studies on UV-C treatments applied during plant growth are focused on their impact on the resistance of plant vegetative organs to pathogens (Obande *et al.*, 2011). However, it is important to verify that the improvement of plant resistance by UV-C treatments does not negatively impact the yield or the quality of fruits at the harvest.

In this work, we applied UV-C radiation (254 nm) on strawberry plants during their cultivation. The aim of this study was to evaluate the effect of these UV-C treatments (i) on the resistance of leaves and of fruits to the major plant pathogens of strawberry, (ii) on the vegetative growth and the yield of the plants and (iii) on the quality of strawberry fruits after harvest.

II. Material and methods

1. Plant material

The Candiss cultivar was created by the Ciref (*centre inter-régional de recherche et d'expérimentation de la fraise*). This strawberry cultivar is a non-remontant plant with very aromatic fruits easily recognizable thanks to its very characteristic conical shape and marked golden achenes. This experiment was carried out twice: one in 2017 and another one in 2018.

Strawberry plants (300 "trayplants", supplied by the Martaillac nursery, Sainte-Marthe, France) were placed in a greenhouse pot at the end of February 2017/2018 (INRA, Montfavet, France) in a mixture of substrate: 60 % compost of TS3 type (Code of Practice) with fine granulometry and 40 % of pine bark. Fertilization and irrigation were done daily by metering pumps and drip. All seedlings were randomized and were grown for 3 months (from planting to harvesting the first commercially mature fruits).

The fruits were harvested at the beginning of May 2017/2018 when they reached the stage of commercial maturity.

2. UV-C treatments

The device used during treatments is an UV-C enclosure with a luminous ceiling with 9 UV-C lamps (DSP UV-C tube, OSRAM HNL, 24 W). The measurement of spectrum (by an UV sensor, OSI UV-20 TO-8 photodiode) confirmed a major peak at 254 nm. Four plants were processed at the same time in the box at a distance of 40 cm from the UV-C lamps.

The dose rate was calculated with light intensity and exposure time. Light intensity measurements were performed with a radiometer positioned at 40 cm from the ceiling light.

The UV-C dose selected was 1.70 kJ/m² (*ie* a treatment time of 4 min 08 sec). Strawberry plants were treated once a week. Several mode of application were tested:

- Before flowering "Av" (1 x 1.70 kJ/m² = 1.70 kJ/m²),
- After flowering "Ap" (5 x 1.70 kJ/m² = 8.50 kJ/m²),
- During all the growth "Pdt" (6 x 1.70 kJ/m² = 10.20 kJ/m²).

Strawberry plants without any UV-C treatment were the control group "T". There were 47 strawberry plants per modality. Following each irradiation, the treated plants were kept in the dark during 6 hours.

3. Chlorophylle a fluorescence

In order to characterize the impacts of UV-C treatments on photosystems, chlorophyll a fluorescence was measured and parameters of the OJIP method were calculated (Annexe 2; Sirbet and Govindge, 2011). The objective was to identify the frequency of application of UV-C that was not deleterious for the plant.

The chlorophyll a fluorescence was measured each harvest day, with a fluorimeter (Pocket-PEA). All measurements were made every morning on non-senescent and fully developed leaves. A highlight pulse (3000 µmol/m²/s) was sent on leaves after they underwent a dark adaptation for 30 min with clamps placed on the leaf limb. This period of dark adaptation allowed the electron acceptor of photosystem II (*PSII*) to be re-oxidized gradually until all of PSII reaction centers are able to redo photochemistry. This allows for the quantification of the flow of electrons that takes place in the photosynthetic machinery.

4. Plant growth and production phase

In order to characterize the strawberry plant growth, the number of leaves before the first UV-C treatment (*ie* 15 days after planting the trayplants) and at harvest (after 8 weeks of growth) was counted at harvest, leaf area was also measured.

For the fruit production phase, counting of buds, flowers and green/turning/mature fruits was carried out each week (from flowering to harvest day, which gives an enumeration of 6 consecutive weeks). These data gave information about the impact of UV-C treatments on flowering, on fruit production and on plant yield at harvest.

5. Fungus material and pathological tests on leaves and fruits

Botrytis cinerea (strain Bc1) was used in this study to artificially inoculate leaves and fruits. For pathological tests on leaves, strains were grown 3 days on PDA medium (*Potato Dextrose Agar*, 39 g/L, Sigma-Aldrich) and at 21°C (16 h of day and 8 h in dark). Mycelial plug of 0.5 cm diameter were used as inoculum and deposited on the central veins of leaves. Lesions are monitored for one week by taking pictures and measuring lesion areas with ImageJ software. The area under the disease progress curve (*AUDPC*) was calculated to determine the level of sensitivity of the strawberry plants.

Fruits were inoculated by a spore suspension of Bc1 at 10^6 spores/mL. 10 µL was spot inoculated into wounds on the epidermal surface. The inoculated fruits were stored in plastic boxes at 21°C and 60 % of relative humidity. The disease development was estimated by counting the number of contaminated fruits and by taking pictures for 5 days after inoculation and calculated the fruit lesion area by using ImageJ software.

A natural powdery mildew infection occurred during the fruit production in the greenhouse a few days before mature fruits were harvested. As a result, the total number of leaves and the number of leaves with visible symptoms of powdery mildew were counted and the percentage of infected leaves per plant was computed. In addition, a natural infection with *B. cinerea* and with *Rhizopus* sp. occurred during the postharvest storage of fruits. Therefore, the number of fruits affected by these fungi was counted and the percentage of infected fruits was computed.

6. Quality analysis of fruits during postharvest storage

Firmness and color of fruits were estimated after harvest (D0) and after 2 and 4 days of storage at 21°C.

Fruit firmness was carried out on the domed part of strawberry thanks to a penetration probe (5 mm diameter). The force required for the probe to penetrate the fruit was measured by a Penefel texture analyser (Setop Giraud-Technologie, France). Firmness was reported as force in newtons (N).

Fruit color was measured thanks to a chromameter (CR-400, Minolta). The apparatus was calibrated with a white reference plate and the parameters L*, a* and b* were measured.

In order to evaluate consumer preferences, taste tests were carried out on harvested strawberry fruits. A panel of 30 consumers tasted strawberry fruits from plants undergoing the various UV-C treatments tested. Consumers tasted blind and had to indicate their preferences and taste criteria (such as acidity, sweetness or texture in the mouth).

7. Statistics

All statistical analyzes were performed with the software Statistica. Firstly, analysis of data normality were performed using Shapiro test. If the data were normal, an analysis of variance (*ANOVA*) was performed. In the case of significant effect of the test factor, a comparison of mean were made thanks to Duncan test or Newman-Keuls test (*NKT*). On the contrary, if data were not Gaussian, non-parametric tests were used such as Kruskal-Wallis test. For each test, a constraint of p-value < 0.05 was provided.

III. Results

1. Impact of UV-C treatments on strawberry plant development

In order to evaluate the effect of UV-C treatment on the vegetative growth of the strawberry plants, the number of leaves before the first UV-C treatment and at harvest, 8 weeks after plantation, was counted. Plant growth was homogeneous before the first UV-C treatment (Figure 34-A). At harvest, no significant difference in the number of leaves was observed. However, leaf area was significantly smaller in the plants treated after flowering (Ap and Pdt) compared to the non-treated plants (T) and the plants treated once before flowering (Av) (Figure 34-B).

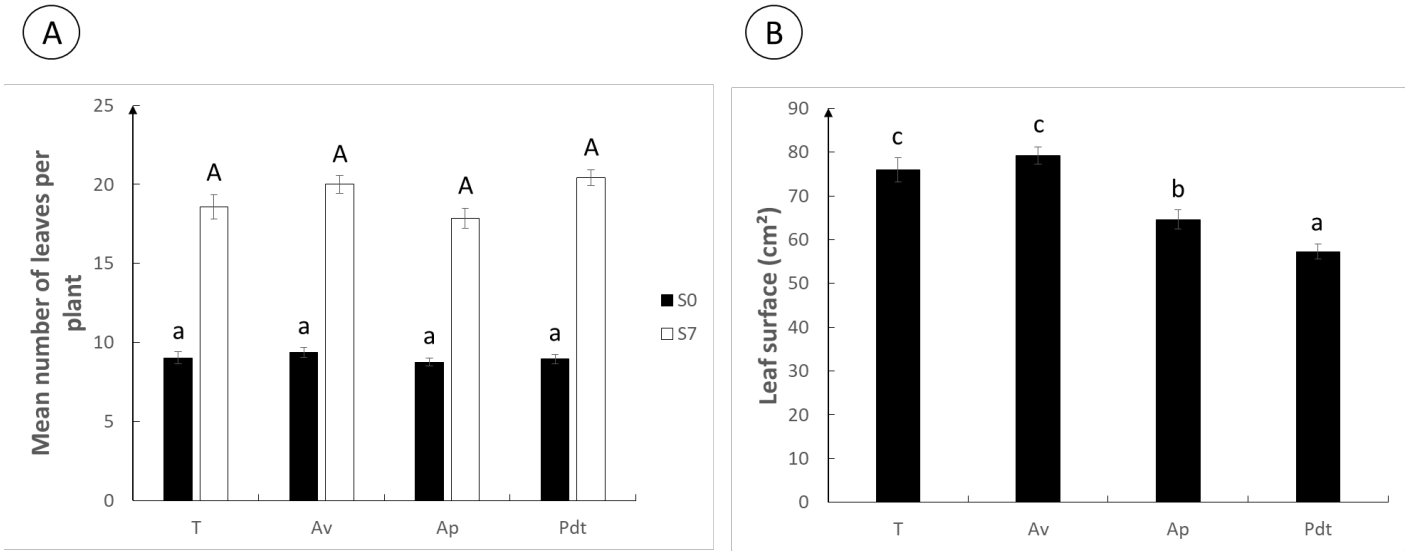

Figure 34: **Number of leaves per plant before UV-C treatments (S0) and after UV-C treatments, at harvest (S7), and measurement of leaf area at harvest**. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment realized after flowering and "Pdt" corresponds to UV-C treatment realized throughout the crop. (A) Count of leaves was done few hours before the first UV-C treatments, after 15 days of plant growth (S0) and at harvest (S7), after the successive UV-C treatments. Lower case letters indicate significant differences identified between modalities just before the first UV-C treatment (standard errors, Newman-Keuls, $p < 0.05$). The letters in upper case indicate significant differences identified between modalities at the harvest day, after the application of the totality of UV-C treatment (standard errors, Newman-Keuls, $p < 0.05$). (B) Leaf surface was measured at harvest day. Lower case letters indicate significant differences identified between modalities (standard errors, NKT, p-value < 0.05).

The fluorescence of chlorophyll a was measured in order to verify that UV-C treatments are not harmful for the proper functioning of the plant's photosystem (Figure 35). Here, we show that the successive application of UV-C doses dit not cause any major damage to the photosynthetic apparatus of strawberry plants. Looking more closely, we can observe that the measured parameters associated with the fluorescence of chlorophyll a increased for plants treated before flowering (Av) and decreased for plants treated after flowering (Ap) or during the cultivation (Pdt), compared to the non-treated group (T).

AUFC	Av	Ap	Pdt
Area	**21.63**	-16.86	-11.90
F0	0.95	**-11.64**	**-10.57**
Fm	-0.18	**-13.01**	**-15.81**
Fv	-0.40	**-13.28**	**-16.83**
F0 / Fm	1.56	1.71	**6.38**
Fv / Fm	-0.30	-0.33	**-1.24**
Fv / F0	-1.35	-1.83	**-7.15**
Sm	**14.85**	-4.82	5.53
N	10.91	-10.79	-0.22
ABS / RC	**-5.15**	**-6.08**	**-4.09**
Di0 / RC	-3.69	-4.06	1.77
TR0 / RC	**-5.43**	**-6.48**	**-5.24**
Et0 / RC	**-7.58**	**-7.09**	**-4.89**
Re0 / RC	**-4.16**	**-10.54**	**-4.83**
Pi total	7.08	-1.60	-1.52
(1-Vi) / (1-Vj)	**3.23**	**-3.48**	0.20
1 - (F4/Fm)	**-2.97**	-0.78	-0.90
Vk / Vj	**-9.66**	**-7.60**	**-4.29**

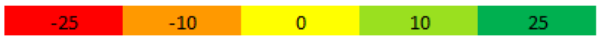

Figure 35: **Relative fluorescence of chlorophyll a of strawberry plant at the harvest day**. The group "Av" corresponds to UV-C treatment before flowering, the group "Ap" corresponds to UV-C treatment after flowering and the group "Pdt" corresponds to UV-C treatment during all cultivation phase. Values represented AUFC (*Area Under Fluorescence Curve*) values: data represent difference percentages between each group treated with UV-C and the control group having no undergone any UV-C treatment. A color scale is provided, ranging from red (negative effect of UV-C radiation) to green (positive effect of UV-C radiation). Values in bold and underlined mean significant difference (NKT, p-value < 0.05).

2. Impact of UV-C treatments on strawberry fruit development

The number of flowers was significantly higher for strawberry plants that have received UV-C treatment (1.5 to 2 flowers per plant depending on the treatment), compared to the plants that have not been treated with UV-C (0.25 flowers per plant for the control) (Figure 36-A). Conversely, at the same time, the number of buds was significantly lower in the UV-C treated plants compared to the non-treated plants. It suggests that buds have turned into flowers for the treated plants (Figure 36-B).

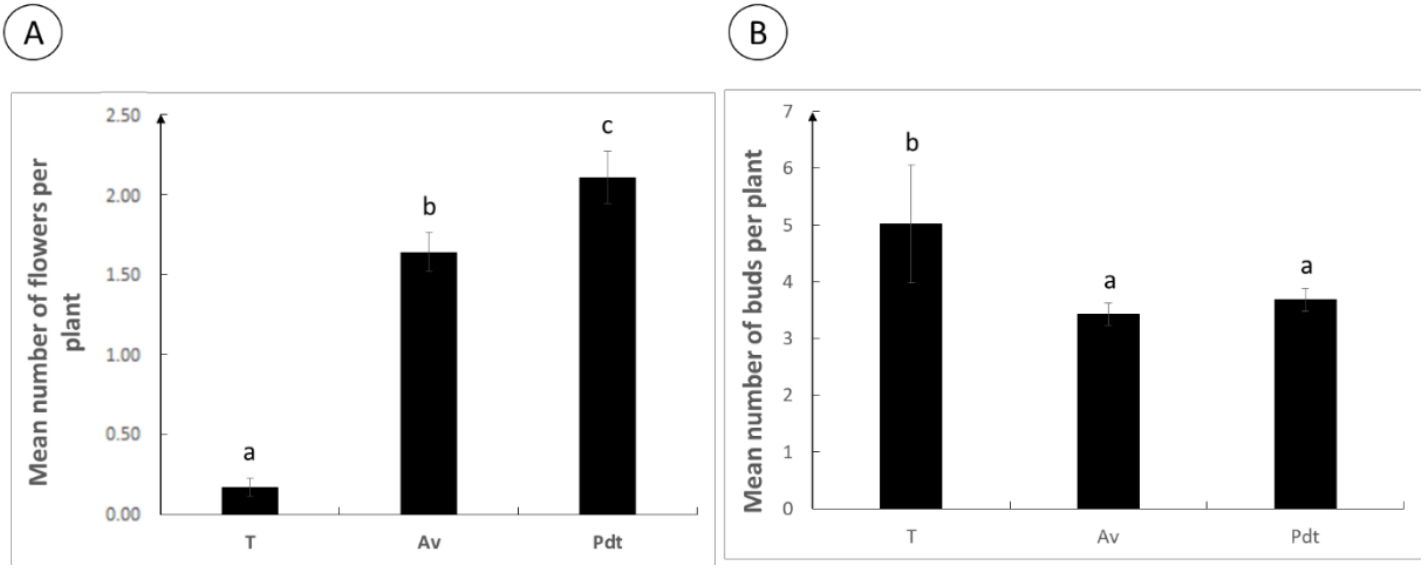

Figure 36: **Number of flowers and buds per plant on non-treated and on UV-C treated plants**. Strawberry plants were treated after 15 days of cultivation with a UV-C dose of 1.70 kJ/m². "T" corresponds to the control group without UV-C treatment, "Av" corresponds to UV-C treatment realized before flowering and "Pdt" corresponds to UV-C treatment applied during all the cultivation. One week after the first UV-C treatment, the number of flowers (A) and the number of buds (B) per strawberry plant were counted. Lower case letters indicate significant differences between different modalities tested (standard errors, NKT, p-value < 0.05).

Subsequently, green fruits were counted each week until harvest (Figure 37). The number of green fruits per plant was greater in plants that have received UV-C treatments (Av and Pdt modalities) compared to non-treated plants (T), up to 4 weeks after the end of flowering. From 5 weeks after the end of flowering, the number of green fruits produced per plant is similar in all modalities.

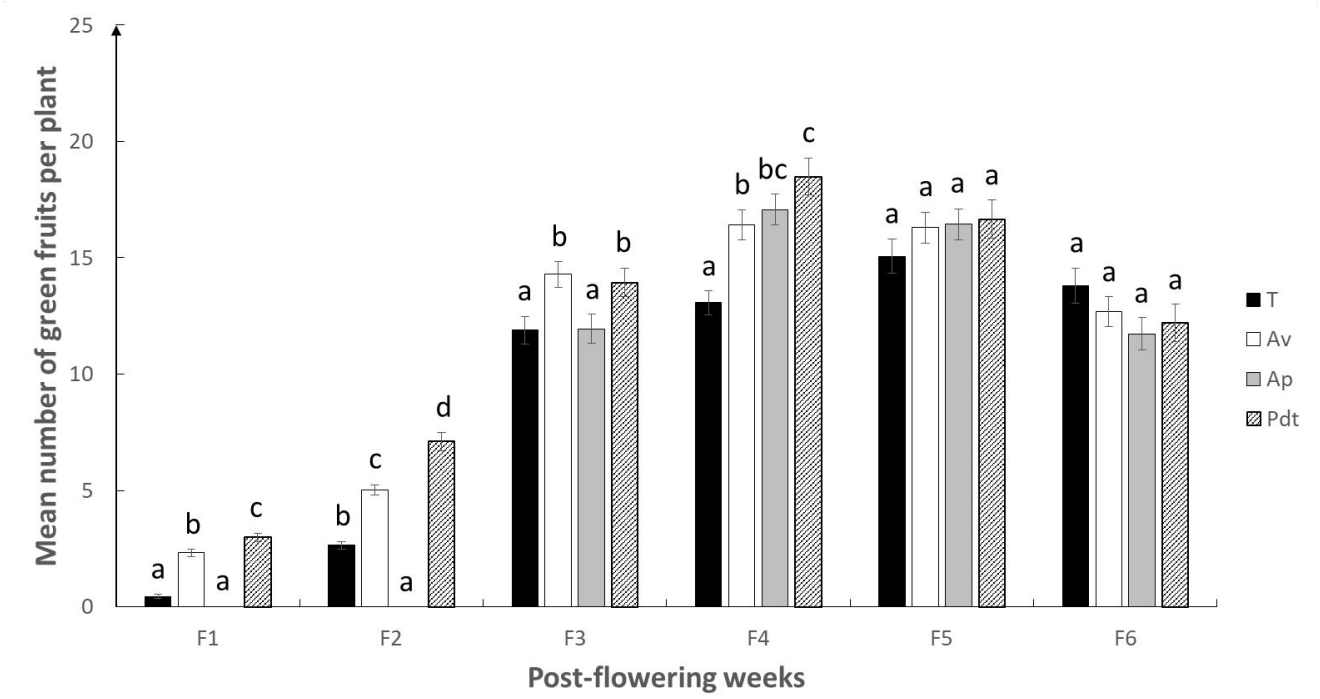

Figure 37: **Mean number of green fruits produced per strawberry plant during the cultivation**. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment applied after flowering and "Pdt" corresponds to UV-C treatment applied during all cultivation phase. Count was made during 6 weeks just after flowering (post-flowering weeks = F). Lower case letters indicate significant differences between different modalities tested (standard errors, NKT, p-value < 0.05).

Yield was estimated by counting the number of ripe fruits per plant at the day of harvest (Figure 38). The amount of ripe fruits was significantly higher for

the plants treated with UV-C before flowering (Av) or during all the cultivation process (Pdt) compared to the non-treated plants (T). The number of mature fruits produced is even twice as large as on the control group. Moreover, UV-C treatment realized before flowering (Av) increased significantly the weight of fruit compared to the non-treated plants (T) and the plants treated after flowering (Ap) (Figure 39). It suggests that the treatment of strawberry plants before flowering has an effect on the development of their fruits.

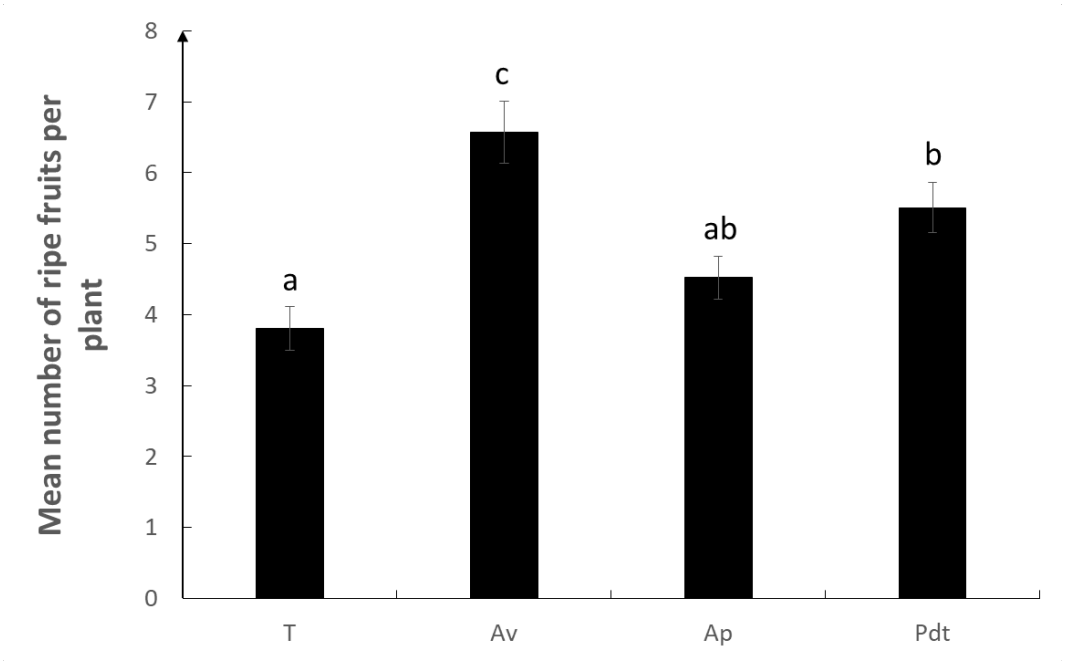

Figure 38: **Number of ripped fruits per strawberry plant at harvest**. The harvest day was carried out one week after the last UV-C treatment with UV-C dose of 1.70 kJ/m². "T" corresponds to the control group (without UV-C treatment). "Av" corresponds to UV-C treatment realized before flowering and strawberry plants belonging this modality received only one UV-C dose (1.70 kJ/m²). "Ap" corresponds to UV-C treatment applied after flowering and strawberry plants belonging to this modality received 5 doses of UV-C radiations (in total 8.50 kJ/m²). "Pdt" corresponds to UV-C treatment applied during all the cultivation and strawberry plants belonging to this modality received 6 doses of UV-C radiations (in total 10.20 kJ/m²). Lower case letters indicate significant differences between different modalities tested (standard errors, NKT, p-value < 0.05).

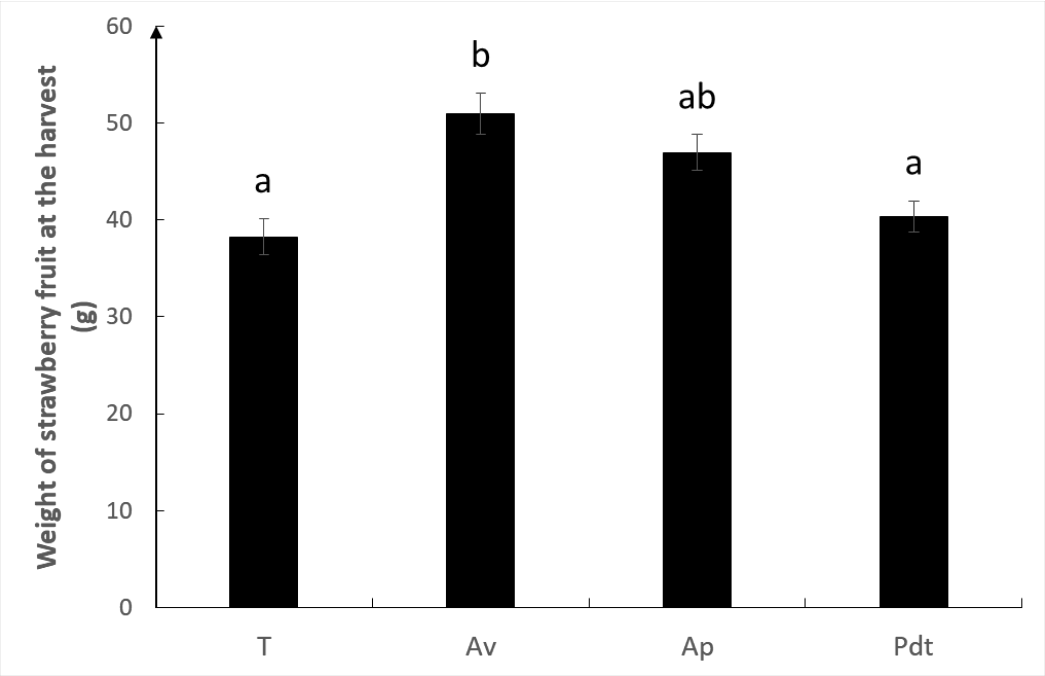

Figure 39: **Weight of strawberry fruit at harvest**. The harvest day was carried out one week after the last UV-C treatment. "T" corresponds to the control group (without UV-C treatment). "Av" corresponds to UV-C treatment realized before flowering and strawberry plants belonging this modality received only one UV-C dose (1.70 kJ/m²). "Ap" corresponds to UV-C treatment applied after flowering and strawberry plants belonging to this modality received 5 doses of UV-C radiations (in total 8.50 kJ/m²). "Pdt" corresponds to UV-C treatment applied during all cultivation phase and strawberry plants belonging to this modality received 6 doses of UV-C radiations (in total 10.20 kJ/m²). Lower case letters indicate significant differences between different modalities tested (standard errors, NKT, p-value < 0.05).

3. Impact of UV-C treatments on strawberry sensitivity (leaves and fruits) to *B. cinerea*

Pathogenicity tests of *B. cinerea* on detached leaves of strawberry collected at harvest revealed a significant reduction in the susceptibility to the plant pathogen (19 %) for the plants that have been treated with UV-C after flowering (Ap). For plants treated before flowering, the sensitivity against *B. cinerea* significantly increased by 25 % compared to the non-treated control plants (Figure 40).

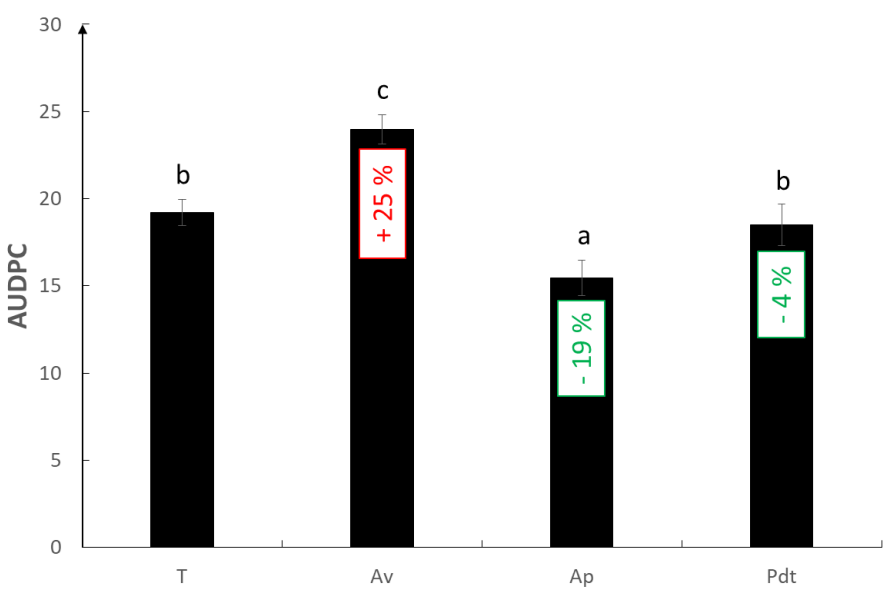

Figure 40: **Susceptibility of strawberry leaves to *B. cinerea***. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment realized after flowering and "Pdt" corresponds to UV-C treatment applied during all the cultivation. Inoculations were made after UV-C treatments by deposing a mycelial plug of *B. cinerea* on the main vein of detached leaves. Surface of necrosis was measured daily for 6 days in order to calculate the AUDPC. Percentages indicated in red or green correspond to percentage of protection of strawberry plants against *B. cinerea* compared to the control group. Lower case letters indicate significant differences identified between modalities (standard errors, NKT, p-value < 0.01).

The test of sensitivity of strawberry fruits to *B. cinerea* revealed no difference between fruits collected from UV-C treated or from non-treated plants (Figure 41).

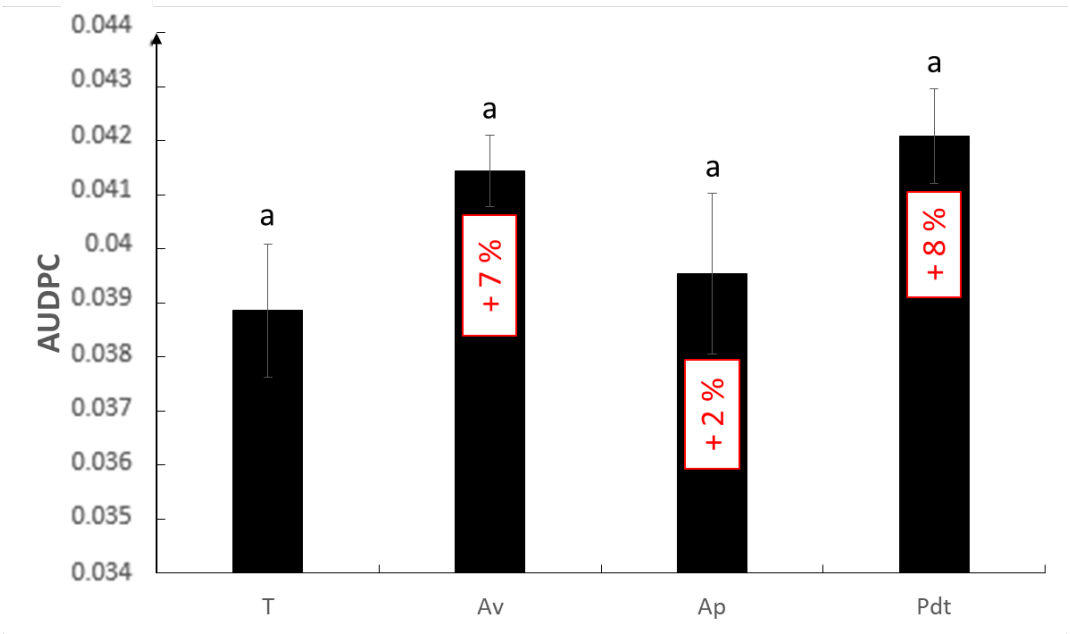

Figure 41: **Susceptibility of strawberry fruits to *B. cinerea***. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment applied after flowering and "Pdt" corresponds to UV-C treatment applied during all the cultivation process. All strawberry fruits were harvested at commercial maturity and inoculation was carried out by depositing a drop of spore suspension of *B. cinerea* dosed at 10^6 spore/mL, after wounding the tip of the fruit. Measurement of necrosis width was done daily for 4 days in order to calculate AUDPC. Percentages indicated in red correspond to sensitivity rate of strawberry fruits compared to the control group. Lower case letters indicate significant differences identified between modalities (standard errors, NKT, p-value = 0.05).

4. Impact of UV-C treatments on strawberry sensitivity (leaves and fruits) to natural infections

Natural and spontaneous powdery mildew infection (*Podosphaera aphanis*) occurred a few days before harvest. We evaluated the level of strawberry resistance against this biotroph by counting attacked leaves (Figure 42). UV-C treatment, whatever the time of application significantly decreased by 51 %, 59 % and 75 % the susceptibility of strawberry plants against the powdery mildew in Av, Ap and Pdt treatments respectively.

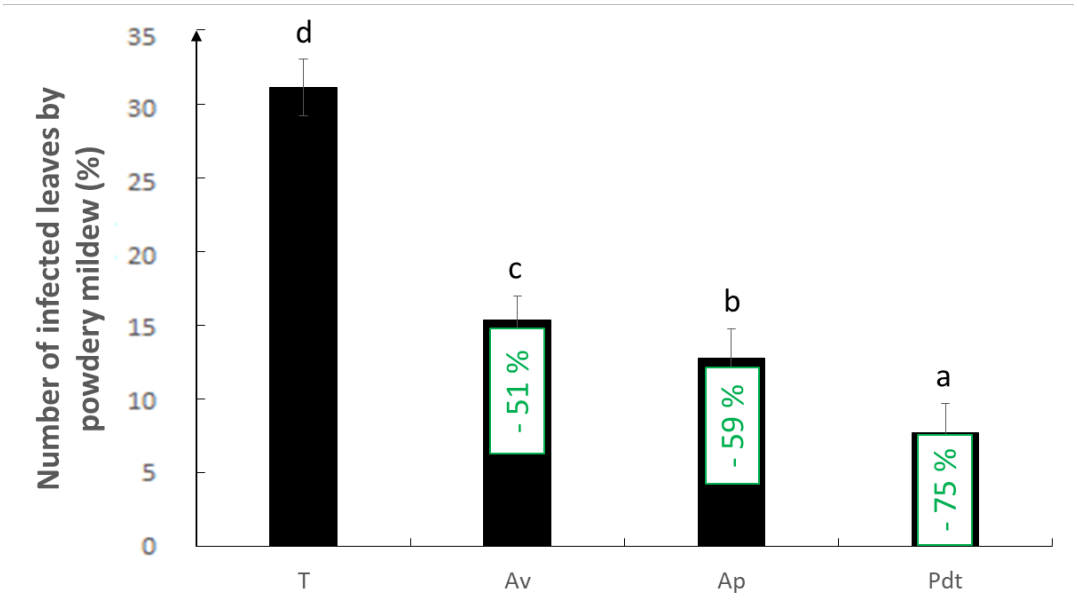

Figure 42: **Susceptibility of strawberry leaves to powdery mildew**. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment before flowering, "Ap" corresponds to UV-C treatment after flowering and "Pdt" corresponds to UV-C treatment during all cultivation phase. Amount of leaves with symptoms was estimated by appearance of powdery and whitish leaf spots. Lower case letters indicate significant differences identified between modalities (standard errors, NKT, p-value < 0.0001).

At the harvest, some strawberry fruits were infected naturally by *B. cinerea* (Figure 43). Strawberry plants treated with UV-C had less infected strawberry fruits, suggesting that UV-C treatments have an impact on the sensitivity of strawberry fruit at the harvest.

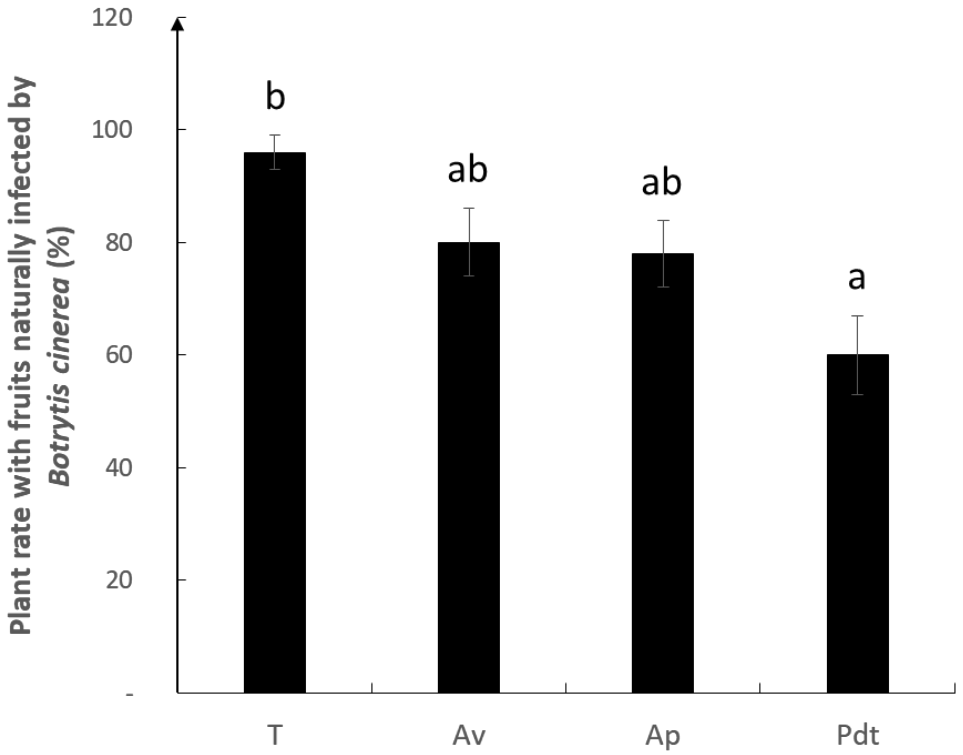

Figure 43: **Percentage of plants with fruits naturally infected by *Botrytis cinerea***. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment realized after flowering and "Pdt" corresponds to UV-C treatment applied during all the cultivation process. The presence or absence of *B. cinerea* on strawberry fruit was evaluated and the percentage of infected fruits was calculated. Lower case letters indicate significant differences identified between modalities (standard errors, Khi² test, p-value < 0.05).

Moreover, during the storage at 21°C, strawberry fruits were also naturally infected by *Rhizopus* sp. (Figure 44).

The results showed a decrease in the sensitivity of strawberry fruits to *Rhizopus* when UV-C radiation were applied before flowering and during the course of plant cultivation.

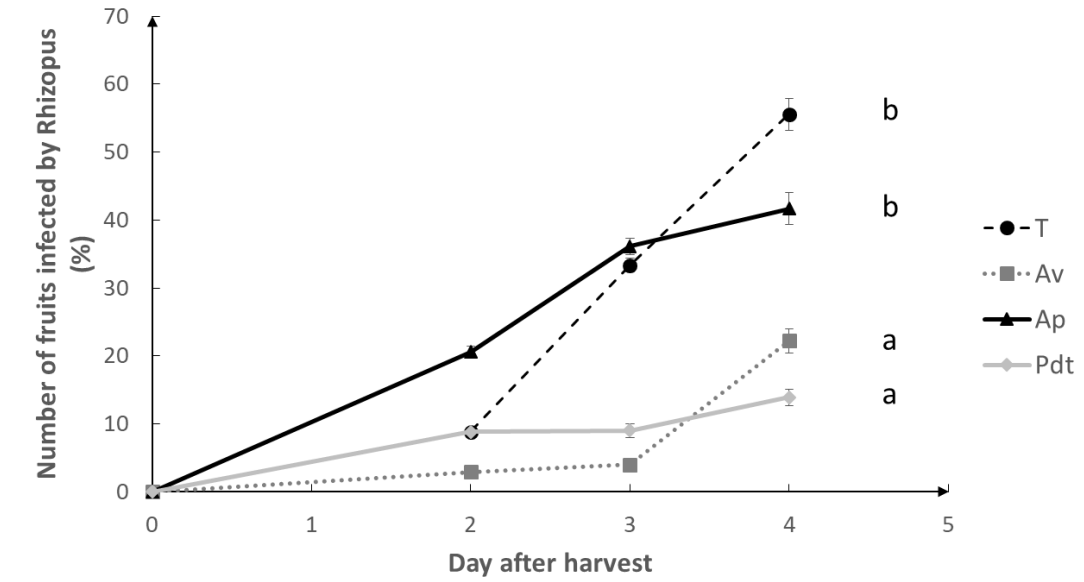

Figure 44: **Susceptibility of strawberry fruits to *Rhizopus* sp. during fruit storage at 21°C**. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment realized after flowering and "Pdt" corresponds to UV-C treatment applied during all the cultivation process. All ripe fruits were harvested and then stored for 5 days at 21 °C in the dark. A follow-up of fruits infected with *Rhizopus* sp. was carried out every day and the percentage of infected fruits was calculated. Lower case letters indicate significant differences identified between modalities (standard errors, NKT, p-value < 0.05).

5. Impact of UV-C treatments on fruit quality

The color and the firmness of strawberry fruits were analyzed (Figure 45). During the storage, all samples (control and treated) showed the same behavior: strawberry became less bright (L* values decreased, Figure 45-A) and lose their intense red color (a* and b* values decreased, Figure 45-A). A significant difference was observed for the b* value, showing that plants treated after flowering and during all the cultivation phase induced fruits with a more brown color.

The firmness lightly decreased during the storage for all the samples (Figure 45-B).

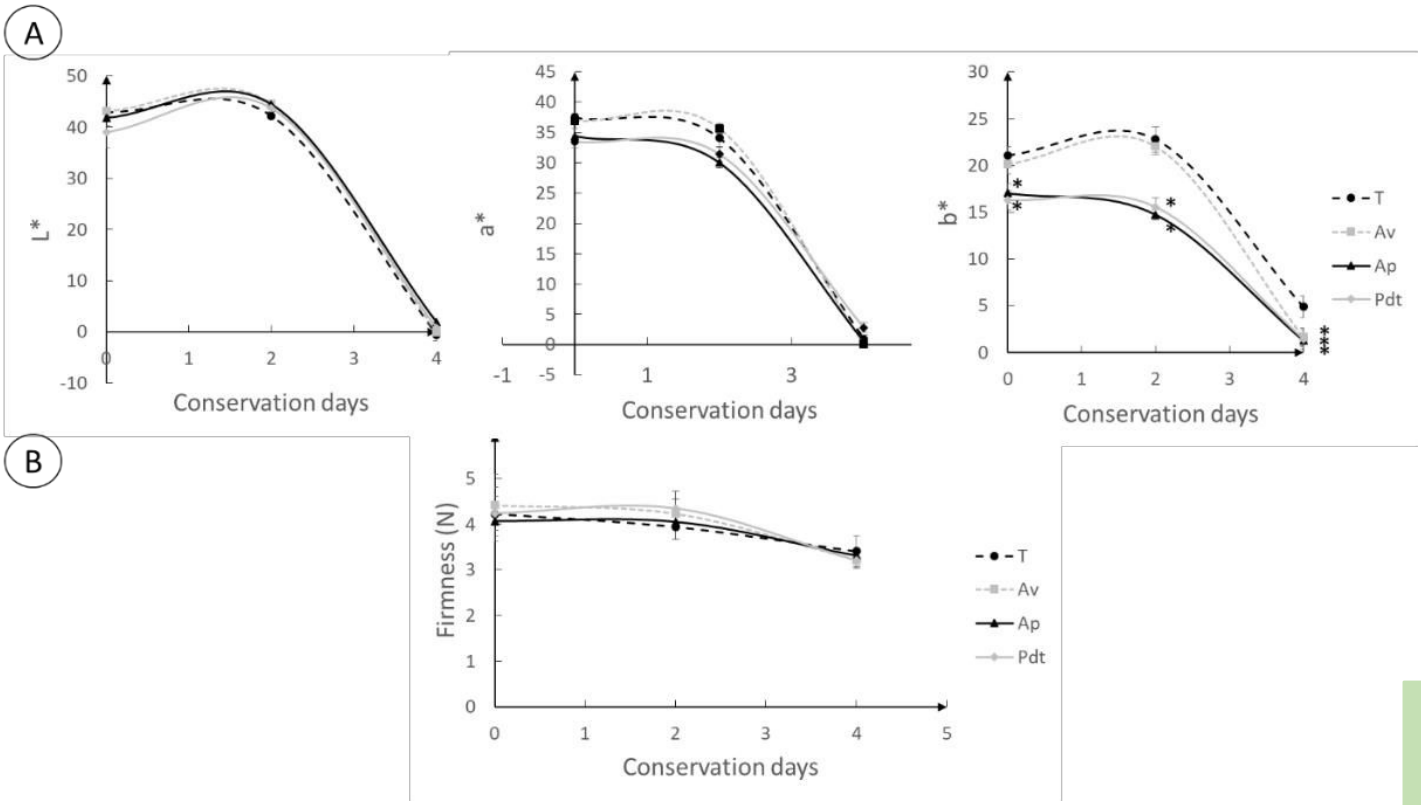

Figure 45: **Evolution of color and firmness of strawberry stored at 21 °C during 4 days**. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment realized after flowering and "Pdt" corresponds to UV-C treatment applied during cultivation. (A) Color measurements were carried out with a colorimeter. Values L*a*b* have been reported. The (*) shows statistical differences between modalities (NKT, p-value < 0.05). (B) Firmness measurements were carried out thanks to penetration probe. No statistical difference was found (standard errors, NKT, p-value < 0.05).

Sensory evaluation was carried out by conducting blind tests (Figure 46). The panel appreciated in the same way both control (47 %) and all UV-C treated fruits (53 %).

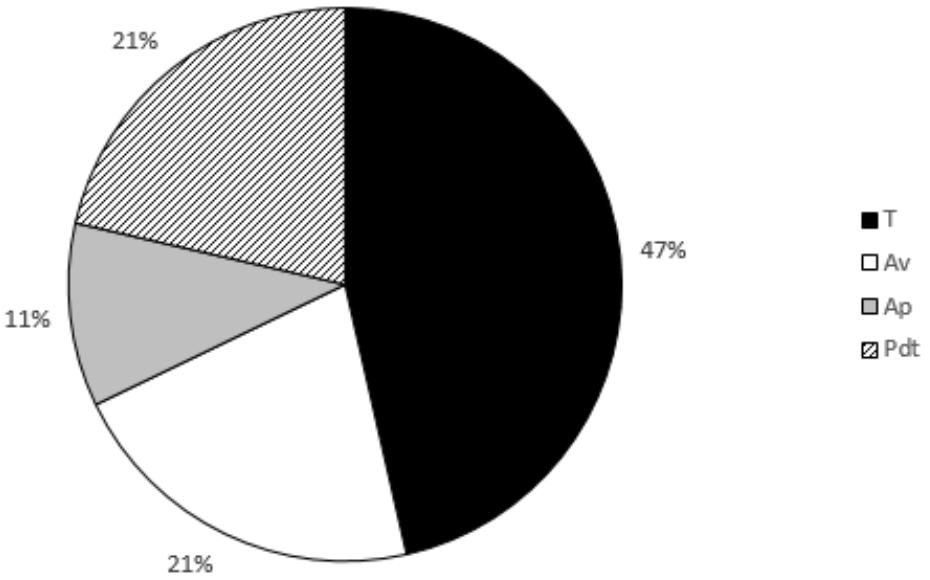

Figure 46: **Consumer preferences for strawberry fruits**. Sensory tests were conducted one day after harvest of all ripe fruits. A panel of 30 consumers tasted fruits by blind tests. "T" corresponds to the control group (without UV-C treatment), "Av" corresponds to UV-C treatment realized before flowering, "Ap" corresponds to UV-C treatment realized after flowering and "Pdt" corresponds to UV-C treatment applied during cultivation.

IV. Discussion

This study investigated the impact of UV-C treatment applied during the cultivation of strawberry on the plant growth, the sensitivity of leaves and fruits to major plant pathogens and the fruit quality after harvest.

One striking result concerns the rate of flowering of strawberry plants between non-treated and UV-C treated plants. Flowering occured earlier when strawberry plants receive UV-C treatments. This fact is consistent with two studies conducted by Darras *et al.* (2012 and 2015), which demonstrated that the application of short UV-C radiation improved flowering and even increased the biomass of geranium plants. Early flowering was also observed in strawberry treated by low-dose of UV-C in pre-harvest and this could be related to change in phytohormone profile (Xu *et al.*, 2017). There is a potential link between fruits yield, quality parameters and phytohormonal changes in preharvest UV-C strawberry (Xu *et al.*, 2017). If flowering happens earlier, the production cycle can be done faster as shown in our study. Thus, larger amounts of ripe fruit at harvest, especially for the strawberry plants treated before flowering, have been observed. Indeed, strawberry fruits continue their maturation process so the green fruits will give turning fruits. Strawberry fruit amount decreasing slightly suggests that a new initiation of flowering took place giving a new cycle of ripening of fruits produced. These results confirm that UV-C radiation can induce a high irradiance response that encourages plant growth or development of fruit (Taiz and Zeiger, 1998).

Crop quality is correlated with the accumulation of direct or indirect solar radiation, which is absorbed by the leaves and is dependent on the total area and number of leaves (Marcelis *et al.*, 1998). In this study, we did not highlight significant differences in the number of leaves per plant between non-treated and UV-C treated plants.

A slight decrease in leaf area was observed for plants treated with UV-C. This result was in line with the work of Darras *et al.* (2012). It suggests that UV-C

radiation was powerful enough to give maximum benefits and plant does not need to expand its leaf area in order to capture more light radiation.

In addition, the analysis of chlorophyll a fluorescence makes it possible to apprehend the potential damage caused by repeated and successive UV-C treatments. And in this study, we demonstrated that successive UV-C dose of 1.70 kJ/m² did not damage the photosynthesis pathway. But, hese same data may indicate that the repetition of UV-C treatments may improve the desired effect of UV-C radiation, that is to say the stimulation of plant defenses. Indeed, we saw that plants treated during the cultivation (Pdt) were less sensitive than the group treated before flowering (Av) having received a single dose of UV-C. The solution would be to find a threshold application to improve plant resistance without damaging the photosynthetic system of plants.

Leaves and fruits of strawberry are very sensitive to pathogens causing considerable damage. A large number of previous studies have demonstrated that UV-C radiation applied in postharvest elicits plant defense responses (Terry and Joyce, 2004). This study investigated the impact of pre-harvest UV-C treatment on the susceptibility of strawberry plants and fruits. During the growth, UV-C treatment applied after flowering significantly decreased by 19 % the susceptibility of the strawberry leaves to *B. cinerea*. Plants treated with UV-C radiations greatly reduced the susceptibility of strawberry plants to powdery mildew (from 51 to 75 % of protection). These results demonstrated that UV-C treatments induced both a direct germicidal effect on the fungus and also an hormetic effect. Indeed, the decrease of susceptibility in strawberry plants treated by UV-C after flowering and during all the cultivation phase indicated that UV-C treatments had a direct germicidal effect on the fungus. The decrease in the susceptibility of strawberry plants that have received UV-C only before flowering demonstrated the hormetic response of the plant, it means the stimulation of a beneficial plant response.

Plants treated with UV-C had significantly less naturally infected fruits to *Rhizopus* sp. and *B. cinerea*, demonstrating a potential link between pre- and post-harvest. However, after inoculation, we could not observe the effect of UV-C

treatments on the development of *B. cinerea*. The inoculation method of the fruit could be too invasive so that *B. cinerea* grows too quickly on strawberry fruits, and we could not identify potential differences in susceptibility. Indeed, due to the high sugar content and the fragile flesh, strawberry fruits are a good vector for contamination.

The sensory and physical quality of strawberry fruits were followed after harvest and during 4 days of storage at 21 °C. Color and firmness are widely used to monitor post-harvest fruit quality and are very well known to be involved in complex maturation processes and therefore in many physiological mechanisms (Gunness *et al.*, 2009). UV-C treatments did not alter the physical integrity of strawberry fruits, and did not improve the shelf life. In this study, there was no difference of firmness change between fruits control and from plants treated by UV-C (from 1.7 to 10.20 kJ/m²). This result is in contradiction with a previous study (Xie *et al.*, 2016) that showed that UV-C treatments (3.6 kJ/m²) improved the firmness of post-harvest strawberry fruits after preharvest treatment. They also indicated that cultivar and season played a more important role in influencing fruit quality than pre-harvest UV-C treatment. When treatment are applied after flowering and during all the cultivation phase, strawberries were browner. Other studies have shown that UV-C radiation applied to plants during their cultivation had an impact on the color of post-harvest strawberry fruits. Xie *et al.* (2016), have observed a significantly higher value for a*, the redness. In the case of strawberry fruits, this parameters generally indicates an increase in the anthocyanin content and is therefore a marker of the progress of ripening of fruit. However, Xie *et al.* (2015) didn't observe any effect of pre-harvest UV-C on anthocyanin content in strawberry fruits. Obande *et al.* (2011) demonstrated a delay in the red color of tomato fruit when UV-C treatments are applied to crops emphasizing the contrasted results for color.

We also conducted blind tests with a panel of 30 consumers to estimate the sensorial quality of fruits. This study highlighted that UV-C treatment during pre-harvest did not change the taste of fruits compared to the control.

To conclude, pre-harvest UV-C treatments had a significant effect on growth and quality crop of strawberry plants and reduce the natural infections of pathogens such as powdery mildew on leaves. The impact on fruit quality on post-harvest was not significant but there was a significant reduction of natural infection, such as *Rhizopus* sp. on fruits in post-harvest.

However, it is difficult to find only one UV-C treatment which can be optimal for all parameters measured such as resistance of plant or fruit, or fruit quality. Indeed, if we look at plant growth and fruit production, it is better to apply UV-C treatments before flowering to increase flowering and plant yield. If we look at the results obtained on the pathological tests with *B. cinerea*, it is better to apply UV-C after flowering to reduce the susceptibility of leaves.

Besides, UV-C treatments applied in pre-harvest seem to be promising in terms of crop quality but further evaluation are needed to find optimal UV-C treatments that can have also an impact on strawberry fruits.