Chapitre I

Impact des radiations UV-C sur la sensibilité de trois variétés de fraisiers (*Fragaria x ananassa*) contre *Botrytis cinerea*

I. Modèle végétal : le fraisier

Le fraisier appartient à la famille des Rosacées. *Fragaria x ananassa* est une espèce hybride interspécifique octoploïde à laquelle appartiennent toutes les variétés actuellement cultivées. La création de cet hybride est due à un explorateur français nommé Amédée-François Frézier (1682 - 1773) qui a importé du Chili 5 plantes de *F. chiloensis* pour sa grande taille des fruits et des plantes de *F. virginiana* (identifiés en Amérique du Nord). Au début des années 1700, le croisement entre *F. virginiana* (mâle) avec *F. chiloensis* (femelle) en France a conduit à la production d'hybrides qui allaient être connus sous le nom d'ananas, progéniteurs du fraisier moderne, *Fragaria x ananassa Duch*.

Les fraisiers sont des plantes vivaces qui se propagent *via* des stolons (Figure 11). Les feuilles sont trifoliées et se développent à partir de la « couronne » (tige réduite dans le cœur de la plante). Les fleurs sont généralement blanches. Il existe quelques espèces à fleurs roses telles que la variété Toscana .Les fleurs possèdent 25 à 30 étamines jaunes et 50 à 500 pistils surélevés sur un réceptacle jaune conique. La fleur est habituellement hermaphrodite, et n'a donc pas besoin de pollinisation croisée.

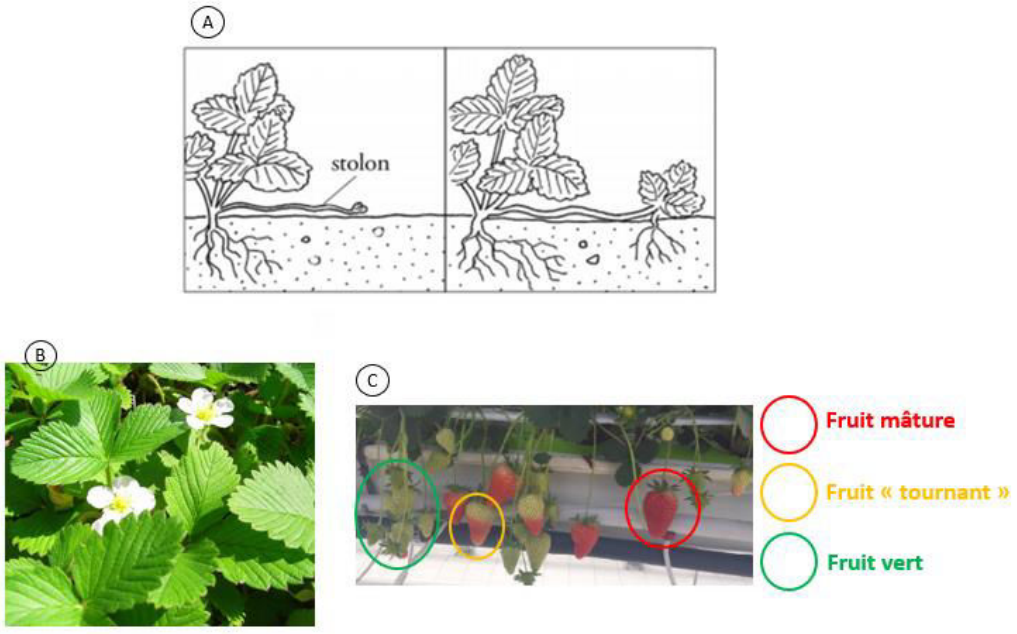


Figure 11 : **Le fraisier**. Le fraisier fait partie de la famille de Rosacées. A) Mode de reproduction asexué du fraisier (phase végétative) : développement de stolons. B) Phase reproductive du fraisier : apparition des fleurs. C) Fructification du fraisier : développement des fraises avec un différentiel de maturation (images personnelles, Pathologie Végétale, INRA PACA Avignon).

Le climat joue un rôle fondamental dans la croissance du fraisier puisque selon la saison, l'état de la plante diffère (Risser et Navatel, 1997). En effet, en été, les plants ont plutôt une croissance végétative. Lorsque l'automne arrive, les températures diminuent et cela initie la floraison et c'est au printemps que la phase de fructification commence.

Ce premier chapitre de thèse est rédigé sous forme d'articles : le premier de méthodologie (Annexe 1) et le second de résultats.

II. Impact of UV-C radiation on the sensitivity of three strawberry plant cultivars (*Fragaria x ananassa*) against *Botrytis cinerea*

Cette partie est présentée sous la forme d'un article de résultats accepté dans le journal Scientia horticulturae en 2018 (Figure 12).

Scientia Horticulturae 240 (2018) 603–613

Contents lists available at ScienceDirect

Scientia Horticulturae

journal homepage: www.elsevier.com/locate/scihorti

Impact of UV-C radiation on the sensitivity of three strawberry plant cultivars (*Fragaria x ananassa*) against *Botrytis cinerea*

M. Forges[a,b,*], H. Vàsquez[a], F. Charles[a], D. Chabane Sari[c], L. Urban[a], Y. Lizzi[a], M. Bardin[b], J. Aarrouf[a,*]

[a] UMR 95 Qualisud, Université d'Avignon et des Pays de Vaucluse, 84916, Avignon, France
[b] Pathologie Végétale, INRA, 84140 Montfavet, France
[c] Institut d'Electronique et des Systèmes, Université de Montpellier, 34090 Montpellier, France

Figure 12 : **Article accepté dans un journal scientifique concernant la pré-culture**. Cet article a été soumis en 2018 dans le journal Scientia Horticulturae.

1. Abstract

Several studies suggest that UV-C radiation, known for its disinfecting effect, may also stimulate plant defenses. The objective of this study is to reduce the sensitivity of strawberry plants (*Fragaria x ananassa*) to *Botrytis cinerea* by application of non-deleterious doses of highly energetic UV-C light (254 nm) on leaves. Preliminary tests were carried out on strawberry plants: Cirafine, Charlotte and Candiss, to optimize the doses of UV-C to apply on plants and to test the sensitivity of these three cultivars to *B. cinerea*. The three cultivars showed

different levels of susceptibility to *B. cinerea*: Cirafine was the most resistant followed by Charlotte and Candiss being the most sensitive. These observations were supported by histological examination and phenol levels in the leaves that indicated deeper penetration of *B. cinerea* into Candiss. Nine variations of treatments were applied to the plants, which were composed of varying UV-C doses and differing application frequencies. The treatment of UV-C applied at 0.85 and 1.70 kJ/m², four times every second day (p-value = 0.05), were shown to have a significant increase, around 25 %, in the protection of Candiss against Bc1 strain of *B. cinerea*. Our observations show that exposing strawberry plants (Candiss) to low repeated doses of UV-C could improve their resistance against gray mold, while avoiding any apparent negative effects to the plants.

Keywords: UV-C radiations, strawberry, *Botrytis cinerea*, plant defense

2. Introduction

The cultivated strawberry *Fragaria x ananassa* is one of the most important fruit crops worldwide. It is ranked first within berry crops with a worldwide fruit yield of 4.1 million tons per year (Flachowsky *et al.*, 2011). Strawberries are produced in more than 70 countries and organic fruit production is becoming increasingly important (Wilbois *et al.*, 2012).

Pathogen development on host plants, especially on plants producing edible fruits, has a major impact on agricultural production giving rise at once economical issue and phytosanitary issue. Several diseases can be particularly damaging for strawberry production, and among these, gray mold is a major concern for growers. Gray mold is an airborne disease, caused by the necrotrophic fungus *Botrytis cinerea*. The disease is difficult to control because the fungus can infect the plant as well as the fruits (Williamson *et al.*, 2007). *B. cinerea* can also cause partial or total destruction of the host plant and its products like fruit, in pre- or post-harvest (Gullino, 1992). The most common and effective strategy to control gray mold is

the application of fungicides. Unfortunately, the frequent use of fungicides can lead to the development of pathogen resistance (Fillinger and Walker, 2015). In addition, the excessive use of fungicides can have a negative impact on the environment as well as on human health, requiring the development of environmentally-friendly alternatives.

Therefore, there is a growing interest in alternative methods that could enabled the stimulation of plants defense mechanisms, based on the use of biotic or abiotic factors (Conrath, 2009). Unlike chemical products, abiotic mechanisms for pathogen control, such as light radiation, have been sparsely studied. Outside the visible light, infra-red light (*IR*) or ultraviolet light (*UV*) have specific properties which are exploited in agricultural sector and food industries. UV light corresponds to electromagnetic irradiations produced by the sun or by an artificial source. Three types of UV irradiations, corresponding to three ranges of wavelength, are distinguished according to their energy and biological activities. UV-A (315–400 nm) that are less energetic represent 95% of UV irradiations that arrive on earth surface, UV-B (280–315) that are moderately energetic are largely stopped by the ozone layer and UV-C (100–280 nm) that are the most energetic but they are integrally absorbed by the ozone layer.

Previous studies have shown that light is an important regulator of plant-pathogen interactions (Demkura and Ballare, 2012; Jenkins, 2009; Magerøy *et al.*, 2010). In response to a sudden exposure to high light intensity or to UV light, plants respond by producing reactive oxygen species (*ROS*) that play a role in plant defenses against pests and diseases. Similarly the rapid accumulation of ROS at the pathogen attack site is toxic to pathogens directly (Lamb and Dixon, 1997) and can trigger signaling pathways that are responsible for the activation of other defense mechanisms (Dat *et al.*, 2000; Grant and Loake, 2000). Part of these resistances can be attributed to changes in plant tissue metabolites induced by UV-B radiations, which include accumulation of protective phenolic compounds and enhancement of jasmonic acid dependent defense responses (Ballare *et al.*, 1996; Demkura *et al.*, 2010; Foggo *et al.*, 2007; Izaguirre *et al.*, 2003, 2007; Kuhlmann and Müller, 2009; Mazza *et al.*, 1999; Rousseaux *et al.*, 1998). Indeed, UV-B radiations stimulates

transcription of defense genes, including those encoding for phenylalanine ammonia-lyase (*PAL*) and chalcone synthase, two key-enzymes controlling the synthesis of defence-related phenolic compounds, as well as pathogenesis-related proteins such as chitinase and β-1,3-glucanase (El Ghaouth *et al.*, 2003; Bonomelli *et al.*, 2004; Borie *et al.*, 2004).

There is however a major problem associated with UV-B light: it is generally effective only when delivered over rather extensive periods of time because of its lower photon energy, typically several hours or days. It is often difficult to consider exploiting UV-B light in practical terms. Hence the idea to using UV-C light which is capable to supply large amounts of energy in a very short period of time. The lethal effect of UV-C light has been exploited successfully to control postharvest diseases, thus extending shelf-life of fruits and vegetables (Liu *et al.*, 2011; Maharaj *et al.*, 1999; Mercier *et al.*, 2001; Mercier *et al.*, 1993a; Siddiqui *et al.*, 2011). Previous studies have also defined hormetic doses that can stimulate plant defense without entailing negative side effects on the stored plant organs like fruits (Charles *et al.*, 2008a-d; Charles *et al.*, 2009; Ouhibi *et al.*, 2015a,b; Pataro *et al.*, 2015; Pinheiro *et al.*, 2015; Sari *et al.*, 2016; Mohamed *et al.*, 2017). Numerous studies have tested different UV-C doses in order to find hormetic dose and doses were in the range of 0.125–9 kJ/m² (Pombo *et al.*, 2011; Ouhibi *et al.*, 2015a,b; Vasquez *et al.*, 2017). Moreover, UV-C radiation stimulates production of pathogenesis-related proteins that play a significant role in plant defense such as chitinase and β-1,3-glucanase in strawberry leaves infected by *Colletotrichum acutatum* or in fruit infected by *B. cinerea* (Casado-Díaz *et al.*, 2006; Jin *et al.*, 2017). If a large number of studies have shown that UV-C radiation elicits defense responses in fruits, few studies have worked on treating plants during their growth. Treating *Arabidopsis*, *Pelargonium* or *Lettuce* plants during their growth with UV-C induces reduction of infection by *Hyaloperono sporaparasitica* and *B. cinerea* (Kunz *et al.*, 2008; Darras *et al.*, 2015; Vasquez *et al.*, 2017). Xie *et al.* (2016) have shown that UV-C treatment applied in pre-harvest caused no leaf damage at the cumulative dose of 3.6 kJ/m², and fruits tend to be firmer and in some

case redder. Unfortunately, there is still a lack of literature on crops destined for human consumption (Vasquez *et al.*, 2017).

The objective of this study was to research the impact of non-damaging doses of UV-C radiation in strawberry plant on the sensitivity to *B. cinerea*. It was carried out on three strawberry cultivars (Cirafine, Charlotte and Candiss) and two strain of *B. cinerea* (Bc1 and Bc21). We proceeded in two steps. First, we tested the basal level of sensitivity of the three strawberry cultivars against *B. cinerea* by evaluating lesion development of the pathogen (histological experiment) and by measuring phenols content. Secondly, we tested the effect of single and repeated non-deleterious doses of UV-C radiation on lesion development of *B. cinerea* on leaves.

3. Material and methods

a. Plant material

Three cultivars of strawberry plants were used: Cirafine, Charlotte and Candiss. These cultivars have been developed and were provided by CIREF (*Centre interrégional de recherche et d'expérimentation de la fraise*, Dourville, France) as "frigo" plant and all have red fruits. Candiss corresponds to non-remontant plants, Charlotte and Cirafine are remontant plants.

Strawberry plants were transplanted in pots containing a horticulture compost mix (TS4 type, Klasmann and Deimann) in a glasshouse. The plants were fertilized with a standard commercial nutrient solution (Soluveg Parme, NPK 16-6-27 + 3 MgO + OE, Angibaud Derome) with a drip irrigation system (one dripper per pot) at a frequency adapted to the climatic demand. For each repetition, plants were randomly distributed and were grown for 2 months before treatment.

Three batches of plants were produced in 2016 to provide independent repetitions of the whole study. The first repetition (R1) was carried out in spring (from January to April 2016) with mean temperatures of 22.5 °C during the day and

13.5 °C during the night throughout the period of plant growth. The second repetition (R2) was carried out in summer (from April to June 2016) with mean temperatures of 27.0 °C during the day and 14.9 °C during the night. The third repetition (R3) was carried out in autumn (from August to October 2016) with mean temperatures of 27.2 °C during the day and 16.7 °C during the night.

b. UV-C treatment of plants

A closed box having a ceiling light with 9 UV-C lamps (DSP tube UV-C, OSRAM HNL, 24 W) of 254 nm (Figure 13) was used to treat strawberry plants with UV-C radiation (Pascal *et al.*, 2018). Four plants were processed at the same time in the box at a distance of 40 cm from the UV-C lamps (Pascal *et al.*, 2018). Several doses and application frequencies were tested during a week (Ouhibi *et al.*, 2015a,b ; Vasquez *et al.*, 2017):

- a single application of UV-C at 0.40, 0.85 or 1.70 kJ/m²,
- a double application of UV-C at an interval of two days between each application at either 0.40 + 0.60 kJ/m² or 0.85 + 1.30 kJ/m²,
- four successive applications of UV-C, every two days, at 0.40, 0.85 or 1.70 kJ/m².

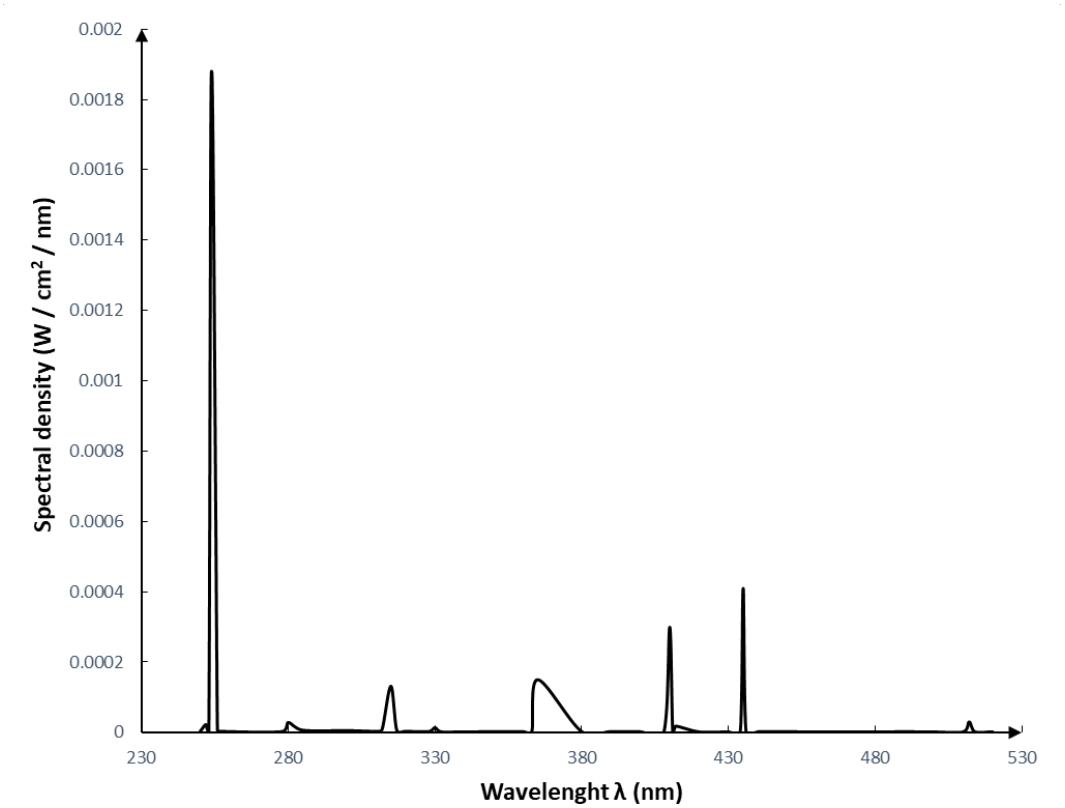


Figure 13: **Spectral density of UV-C lamps, DSP tube UV-C, OSRAM HNL, 24 W**. The test window for testing the homogeneity of the emitted radiation had the following dimensions: 310 x 260 nm. Radiation is homogenous over the entire emissive surface of the lamp. Only the significant peaks that carry 99 % of the radiated optical power were reported. UV-C peaks at 252 and 254 nm carried out 64 % of the power, peaks at 280 nm and 315 nm carried out 5.7 % of the power, peak at 365 nm carried out 5 % of the power. The reminder was transported by the visible part of radiation emitted by the lamp: 25.3 % (purple / blue / green).

The calculated UV-C dose applied on the plants depended on the fluence of the UV lamps and on the exposure time (Houghton *et al.*, 2001). Light intensity measurements were performed with a radiometer positioned at 40 cm from the ceiling light and time of plant exposure to UV-C was calculated through the measurement of light intensity at a given time. The duration of UV-C radiation required was 49 sec to obtain a final dose of 0.40 kJ/m², 1 min and 13 sec to obtain 0.60 kJ/m², 1 min and 44 sec to obtain 0.85 kJ/m², 2 min and 39 sec to obtain 1.30

kJ/m² and 3 min and 28 sec to obtain 1.70 kJ/m². Strawberry plants without any UV-C treatments were used as control. To avoid the photo-reactivation of white light (Mercier *et al.*, 2001), plants were placed in the dark for 15 hours after each UV-C treatment.

c. Chlorophyll a fluorescence

In order to characterize the impacts of UV-C treatment on the plant photosystem, the chlorophyll a fluorescence was measured (Annexe 2; Stirbet and Govindjee, 2011). The parameters of chlorophyll a fluorescence derived from measurements were made with a fluorometer (Pocket-PEA). A light pulse was sent on leaves after they underwent a dark adaptation for 30 min with clamps placed on the leaf limb. All measurements were made every morning on non-senescent and fully developed leaves. This period of dark adaptation enables the electron acceptor of PSII to be re-oxidized gradually until all of PSII reaction centers are able to redo photochemistry (Stirbet and Govindjee, 2011). This allows for the quantification of the flow of electrons that takes place in the photosynthetic machinery. The measurements were performed by a 1 sec induction period in which the leaves were illuminated at a light intensity of 3000 mol/m²/s.

Recorded fluorescence parameters were (Annexe 2):

- the minimal fluorescence when all reaction centers are open (F0 correspond to the *first reliable fluorescence value* after the onset of actinic illumination),
- S_m, assumed to be proportional to the number of reduction and oxidation of one Q_A molecule (corresponding to *primary quinone acceptors*) until F_m is reached, and therefore, related to the number of electron carriers per electron transport chain as well as an indicator of the size of the plastoquinone pool (Yordanov *et al.*, 2008). The plastoquinone pool decreased due to stress (Bishop, 1961; Shavit and Avron, 1963),

- the maximum quantum yield of PSII (Fv/Fm, where Fm is the *maximal fluorescence* value and Fv = Fm – F0, corresponding to the *maximal variable chlorophyll fluorescence*), and it is widely used to assess stress in plants,
- the maximum primary yield of photochemistry of the PSII (Fv/F0). This parameter is an indicator of the energy trapping probability (Krause and Weis, 1991).
- the performance index (*PI*) of Strasser *et al.*, (2000), which is a multi-parametric expression of three independent steps contributing to photosynthesis, namely RC/ABS, Fv/Fo and (1-Vj)/Vj. RC/ABS is an indicator of the size of the chlorophyll antenna serving reaction center whereas (1-Vj)/Vj is an indicator of the performance due to the conversion of excitation energy to photosynthetic electron transport. PI is considered more sensitive and discriminating than the stress indicator Fv/Fm (Thach *et al.*, 2007),
- Vk/Vj correspond to the time plot of the values of relative variable fluorescence where the K step occurs in OKJIP transient where step K corresponds to a threshold around 300 µs which appears only in certain conditions of stress (like heat).

Fluorescence measurements were made on 25 leaves per treatment after each UV-C treatment (after 12 h). The area under the curve of induction of the fluorescence of chlorophyll a (*AUFC*) was computed to investigate changes due to UV-C treatment in the photosynthetic machinery.

d. Determination of soluble and related phenolic compounds in non-treated leaves

To determine the amount of soluble-phenolic compounds, five samples of non-treated strawberry leaves per cultivar were ground using a mortar and pestle cooled in liquid nitrogen. For each sample, 1 g of fresh material was supplemented with 20 ml ethanol 70 % and then crushed again for 1 min with an Ultraturrax. After stirring in the dark for 30 min, the sample was filtered on a Buchner funnel under vacuum and the filtrate was made up to 50 ml.

Assays were made by Folin-Ciocalteau method described by Swain and Hillis with modifications according to Marigo (1973), where the color development is proportional to phenols content. The amount of phenolic-related compounds was performed with respect to a gallic acid standard curve measured at 760 nm.

Thanks to residues from previous extractions, phenolic-related compounds were extracted. Two consecutive extractions with 70 % methanol followed by an additional extraction with 80 % acetone and 3 other extractions at the end with pure acetone were done. Each extraction step was performed with 6 mL of solvent and the supernatant was systematically removed. The final residue was dried at room temperature and stored at -20 °C.

Related phenol compounds were extracted from 40 mg of dried-leaves in which 5 ml of sodium hydroxide 4 N previously bubbled in liquid nitrogen was added. The extract was then incubated 1 hour at 121 °C and the pH adjusted to 2 with hydrochloric acid 8 N and centrifuged at 5000 g for 5 min and at 4 °C. The pellet was discarded and the supernatant was stored at 4 °C in the dark until assays of phenolic-related compounds were realized.

e. PAL activity

PAL activity was assessed on the leaves of the three cultivars of strawberry plants. PAL activity was measured as described by Siriphanich and Kader (1985). The extraction was done from 200 mg of dry matter (freeze-dried leaves). The extract was mixed with 4 mL of borate buffer 1 M (pH 8.6) containing 5 mM of 2-mercaptoethanol and 0.40 mM of polyvinylpyrrolidone. Then the mixture was centrifuged at 12000 g for 20 min at 4 °C. The supernatant containing PAL was collected.

To quantify PAL activity, the reaction mixture consisted of the enzyme extract (0.5 ml) and borate buffer 0.1 M (pH 8.6) containing 5 mM of 2-mercaptoethanol and 0.1 M of phenylalanine. After 1 h of incubation at 30 °C, 400 µL of 28 % trichloroacetic acid was added to the mixture to precipitate proteins. After spending 5 min room temperature, the mixture was centrifuged at 12000 g for 20 min at 4 °C. PAL activity was measured by spectrometry at 290 nm to detect cinnamic acid as a product and expressed as nmole of cinnamic acid per hour per mg fresh weight.

f. Production of B. cinerea and leaf inoculation

Two strains of *B. cinerea* were used in this study: strain Bc1 that had a high level of aggressiveness on strawberry leaves, and strain Bc21 that had a medium level of aggressiveness (Bardin *et al.*, 2013). Strains were grown 3 days on PDA (*Potato Dextrose Agar*, 39 g/L, Sigma-Aldrich) in a growth chamber (21°C, 14 hours of photoperiod at 114 $\mu mol.s^{-1}.m^{-2}$) and mycelial plug of 5 mm diameter taken from the growing margin of the culture were used as inoculum.

Tests on detached strawberry leaves were performed 48 h after the last UV-C treatment. Two leaves per plant were collected from 5 plants and the 10 detached leaves were placed on moistened filter paper in 5 transparent polystyrene boxes (2 leaves / box) and inoculated with the mycelium plug in the center of the leaf.

Following inoculation the detached leaves were placed in a growth chamber (21°C, 14 hours of photoperiod at 114 µmol.s^{-1}.m^{-2}). Detached leaves were photographed every day from day 3 to day 6 after inoculation and the lesion areas were assessed with "Image J" software (US National Institutes of Health, Bethesda, MD, USA). The tests were performed in triplicates (R1 / R2 / R3).

The area under the disease progress curve (*AUDPC*) was calculated to determine the level of sensitivity of the strawberry plants. To compare the protection induced in the leaves by the different UV-C treatments, a protection index was computed as follows:

$$\text{Protection (\%)} = 100 \times (AUDPC_{untreated} - AUDPC_{UV\text{-}C\ treated}) / AUDPC_{untreated}$$

g. Histological examination: penetration zone of the pathogen in leaves

Thirty detached leaves of each cultivar of strawberry plants were inoculated with Bc21, by depositing mycelial plugs (5 mm in diameter) excised from the growing margin of a three day-old PDA cultures, on the main vein and its vicinity of the detached leaves. Leave fragments of about 1 cm² were taken from the infected areas of the leaves 24 h after inoculation using a sterile razor blade. Fragments were immediately immersed in a mixture of paraformaldehyde (4 %), glutaraldehyde (1 %) and caffeine (1 %) supplemented with a drop of tween 80 for 24 h to stabilize the leaf cells. During stabilization, vacuum was applied several times in order to facilitate penetration of the fixing agent into tissue. Samples were then rinsed thoroughly with distilled water and gradually dehydrated by a graded ethanol series (25, 50, 70, 85, 95 and 100 %). Leaf fragments were then impregnated into a mixture (1:1) of absolute ethanol and synthetic resin (Technovit 7100 kit Kulzer) overnight and then in pure resin for 24 h. Then, they were included in the same resin to which a curing agent was added to initiate polymerization at room temperature. After polymerization, blocks were cut with an automatic retraction microtome (Supercut 2065 Jung-Kulzer, Leica) equipped with a blade, and specimens of 3 µm of thickness were made sequentially. Resin tapes containing section samples were

then carefully layered. Specimens were placed on microscope slides and blades and then dried on a hot plate before being subjected to a reactive double staining Schiff/Naphthol Blue Black. Double staining has the advantage of coloring polysaccharide products in pink and protein in blue. Stained sections were mounted between slide and coverslip in Acrytol before being observed using a light microscope (Leica DM 2000) equipped with a digital camera (DFX 100). The penetration zone of *B. cinerea* in each leaf was examined by microscope and rated as follows: 1 = presence of mycelium of *B. cinerea* in the cuticle; 2 = presence of mycelium under epidermis; and 3 = presence of mycelium in the parenchyma. An average rate of penetration was then calculated by the examination of 30 samples per strawberry cultivar.

h. Statistical analysis

All statistical analyzes were performed with Statistica software (Statsoft Inc., Tulsa, USA). Firstly, analysis of data normality was performed using Shapiro-Wilks normality test. If the data was normal (p-value > 0.05), an analysis of variance (*ANOVA*) was then performed. In the case of significant effect of the tested factor, a comparison of mean was realized thanks to Duncan test or Newman-Keuls test (*NKT*). However, if the data was not found to be Gaussian (Shapiro-Wilks normality test, p-value < 0.05), a non-parametric analysis of variance (Kruskal-Wallis test) was performed.

4. Results

a. Sensitivity level of three strawberry cultivars to B. cinerea

The three cultivars of strawberry were inoculated with *B. cinerea* Bc21 in three separate replicate experiments, R1, R2 and R3. A significant difference in the

sensitivity of the three strawberry cultivars to *B. cinerea* was observed for experiments R1 and R3 (ANOVA on AUDPC values, p-value < 0.1; Table 2). Candiss appeared to be the most sensitive cultivar, whereas Cirafine was the least sensitive.

Table 2: **Sensitivity level of the three strawberry cultivars Cirafine, Charlotte and Candiss to the strain Bc21 of *B. cinerea***. Pathological tests were performed on detached leaves. The values represent the average AUDPC calculated between the 3rd and the 6th day after inoculation. To test a varietal effect on the sensitivity to *B. cinerea*, ANOVA was performed for each independent repetition of the test. Different letters within a column show significant differences between the three varieties of strawberry plants for each independent repetition (NKT, p-value < 0.05).

	Bc21		
	R1	R2	R3
Cirafine	11.1 b	22.6 a	12.7 b
Charlotte	13.6 ab	23.7 a	15.4 ab
Candiss	18.3 a	29.9 a	20.0 a
P =	0.04	0.06	0.02

To understand differences of sensitivity of strawberry cultivars, a histological study was conducted and the level of penetration of *B. cinerea* on leaves was determined 24 h after inoculation on or in the vicinity of the main rib of leaflets. Histological observations confirmed differences in the sensitivity of the three cultivars. Penetration of *B. cinerea*, at main rib or its vicinity, was deeper in Candiss which was the most sensitive cultivar than in Charlotte and Cirafine (Figure 14-A). In Cirafine leaves, *B. cinerea* was under the cuticle and under the epiderma for Charlotte leaves. The leaf anatomy was, however, comparable between strawberry cultivars (Figure 14-B); and an upper epidermis, a palisade parenchyma, spongy parenchyma and a lower epidermis were observed for all cultivars.

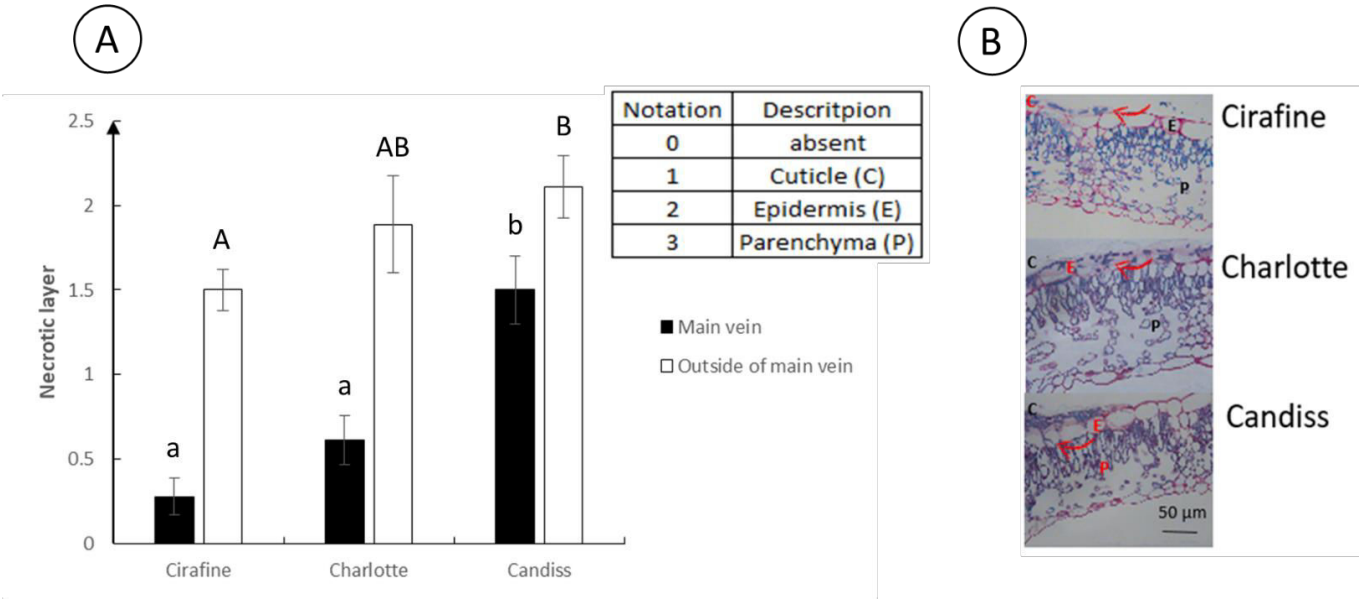


Figure 14: **Penetration of *B. cinerea* in different cell layers of leaves of three cultivars of strawberry plant (Cirafine, Charlotte, Candiss)**. Histological study was conducted 24 h after inoculation with Bc21 on detached leaves. Mycelial plugs were placed on either the main rib or on the leaf limb. A) Bc21 penetration level in cell layers of leaves either at the central rib or on the leaf limb by histology method. Lowercase letters show significant differences between the three varieties of strawberry plants when leaves were inoculated on the main vein (standard errors). Uppercase letters indicate significant differences between the three varieties of strawberry plants when leaves were inoculated outside of the main vein (standard errors). B) Pictures taken by light microscopy. Penetration of Bc21 in cell layers of strawberry leaves which were inoculated on the leaf limbs.

To explain differences of penetration by *B. cinerea* in leaves, we hypothesized the differences in the resistance of the plants could be attributable to the phenolic compounds reinforcing the cell wall. Quantitative analysis of phenolic-related compounds in leaves revealed that Candiss had a slightly higher concentration of phenolic compounds than Cirafine before inoculation by *B. cinerea* (Figure 15). Unlike the other cultivars, the concentration of phenolic

compounds increased significantly in Cirafine 24 h and 48 h after inoculation (Figure 15).

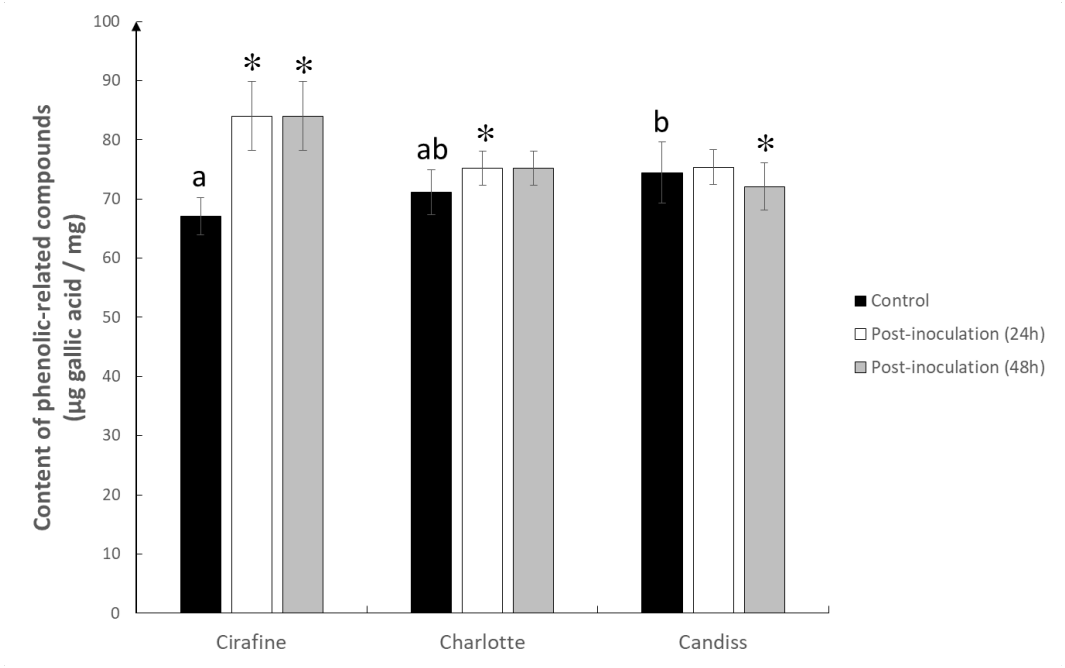


Figure 15**: Content of phenolic-related compounds in leaves of three cultivars of strawberry plants (Cirafine, Charlotte, Candiss) before and after inoculation with *B. cinerea***. Detached leaves of the three varieties of strawberry plants were inoculated with mycelial plugs of *B. cinerea* (Bc21 strain). The non-necrotic portion of each leaves were recovered in order to dose phenolic compounds, at 24 h and 48 h post inoculation. Lowercase letters show significant differences between the three controls (NKT, p-value < 0.05). The (*) correspond to significant differences between the control group and the inoculated group in each strawberry cultivar (standard errors, NKT, p-value < 0.05).

b. Impact of UV-C on sensitivity level of strawberry plants against B. cinerea

Strawberry plants from the three cultivars Cirafine, Charlotte and Candiss were treated with various UV-C doses with different timing of application. After

receiving a single or double application of UV-C (at 48 h intervals), an overall increase in the susceptibility of Cirafine to *B. cinerea* and a slight decrease in the susceptibility of Candiss at the highest UV-C doses were observed (Figures 16-A and 16-B). These results were generally similar regardless of the *B. cinerea* strain tested (Bc1 or Bc21).

When strawberry plants received four successive UV-C treatments at various doses every two days, an increase in the susceptibility of Cirafine to both strains of *B. cinerea* and a decrease in the susceptibility of Charlotte (3 % to 18 %) and Candiss (15 % to 27 %) was observed (Figure 16-C). This decrease in susceptibility was statistically significant only for Candiss treated with UV-C radiation at 0.85 kJ/m^2 and 1.70 kJ/m^2 and inoculated with Bc1.

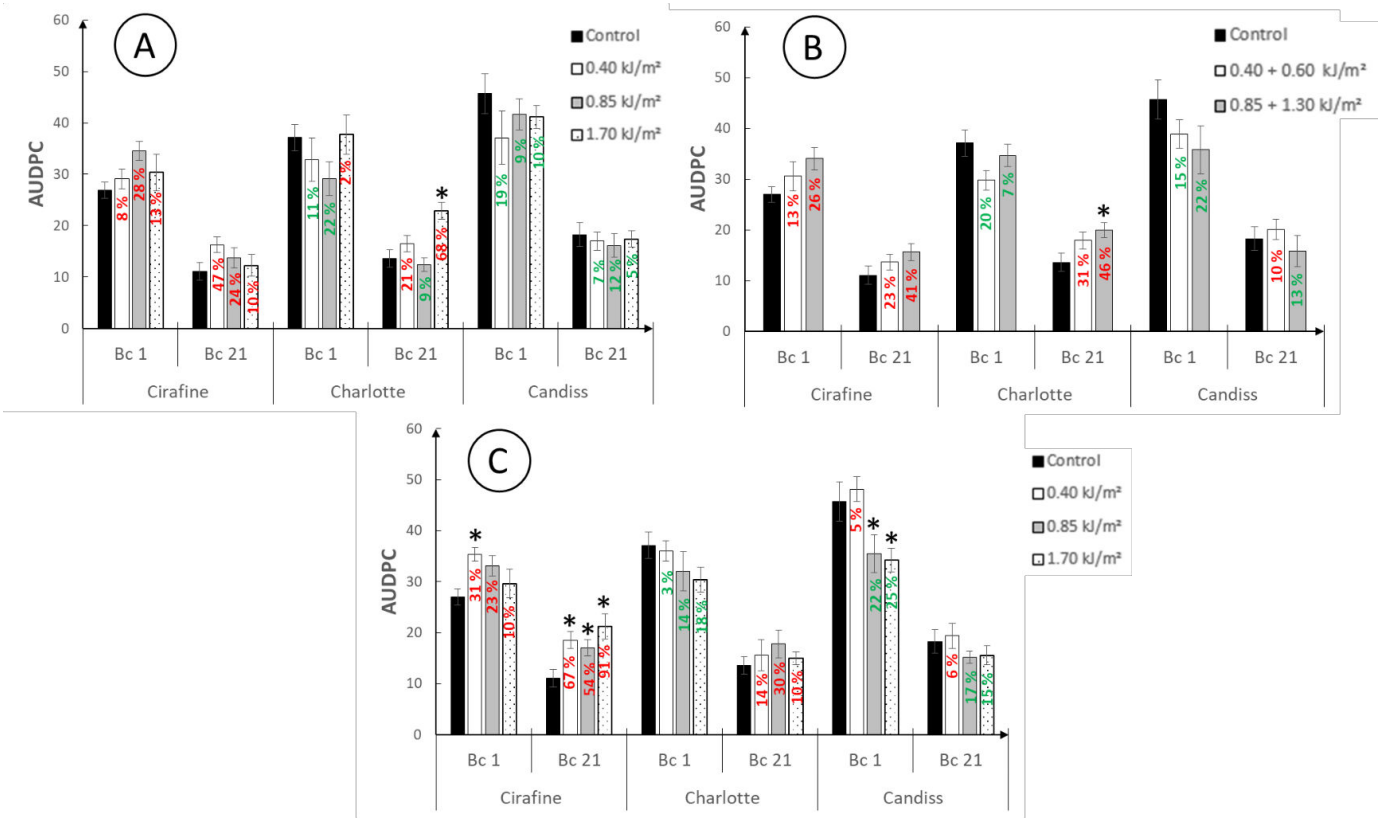


Figure 16: **Sensitivity level of strawberry plants to *B. cinerea* after different UV-C treatments**. Three strawberry cultivars (Cirafine, Charlotte, Candiss) and two strains of *B. cinerea* (Bc1 and Bc21) were tested. The protection rate is calculated according to the difference of sensitivity between the control and plants which are treated by UV-C radiation: when the protection rate is in red it means that leaves are more sensitive and in contrast, when the protection rate is in green it means that leaves are less sensitive than the control group. A) Sensitivity of strawberry plants to Bc1 and Bc21 after a single application of UV-C radiation. B) Sensitivity of strawberry plants to Bc1 and Bc21 after a double application of UV-C radiation. C) Sensitivity of strawberry plants to Bc1 and Bc21 after four UV-C applications (each two days), named 4 x UV-C dose. The (*) show significant differences between the inoculated leaves and control (standard errors, NKT, p-value < 0.05).

After successive exposures to UV-C radiation, Charlotte and Cirafine had the strongest oxidation symptoms on both the top (Figure 17-A) and the lower (Figure 17-B) surfaces of leaves while Candiss had no symptoms (Figure 17-C). In order to see the antioxidant composition of leaves for the three strawberry cultivars, measurements of soluble phenolics were made on leaves before UV-C treatment. Larger amount of soluble phenolic compounds were found in Candiss leaves compared to other cultivars (Figure 18-A). The presence of a higher amount of phenolic compounds in Candiss leaves correlated to increased PAL activity than in Cirafine and Charlotte (Figure 18-B).

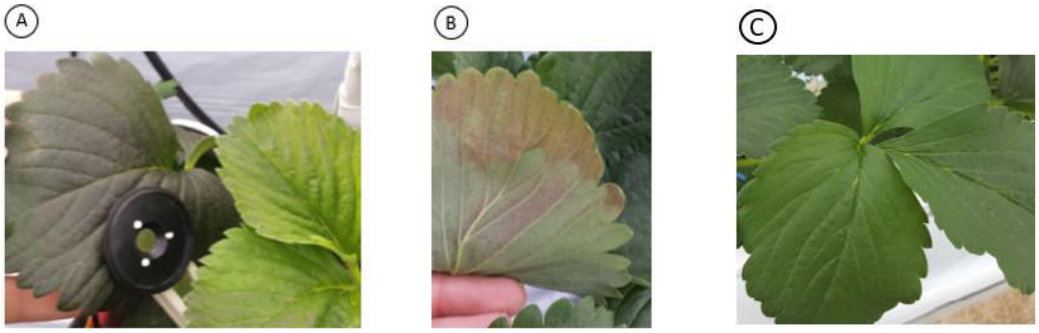


Figure 17: **Foliar symptoms after UV-C treatment**. Leaf symptoms are visible: darkening (color "lawyer"). A) Top side of strawberry leaf (Cirafine and Charlotte). B) Bottom of strawberry leaf (Cirafine and Charlotte). C) Strawberry leaf without symptoms (Candiss).

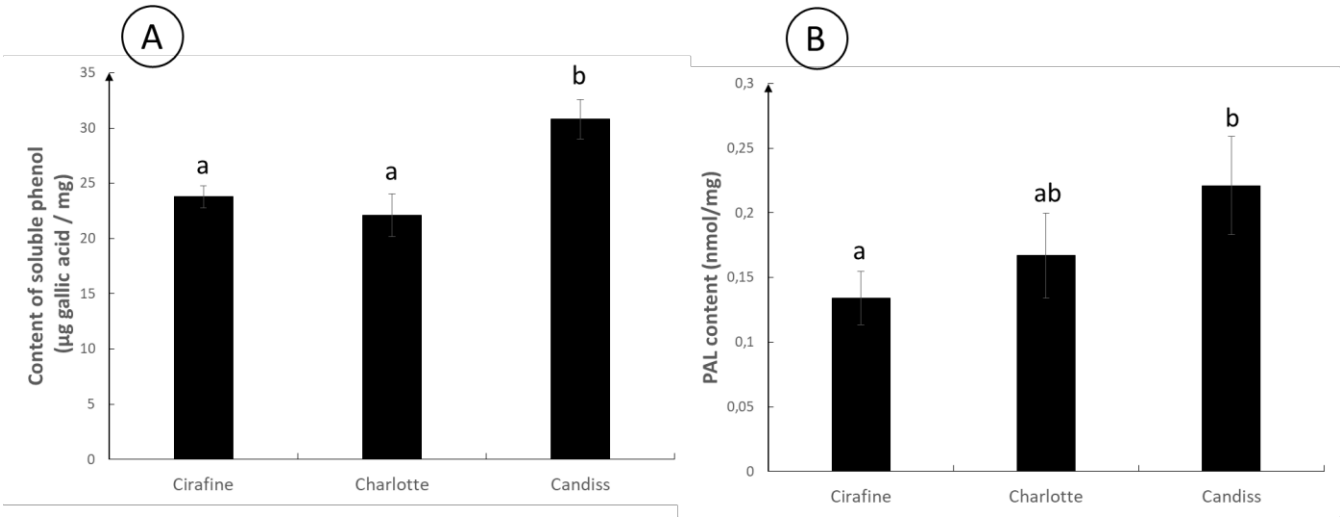


Figure 18**: Soluble phenolic content and PAL activity in leaves of three cultivars of strawberry plants (Cirafine, Charlotte, Candiss)**. Measurements were carried out before plants receive different UV-C treatment. A) Amount of soluble phenolic compounds in strawberry leaves. B) Activity of PAL in strawberry leaves. Lowercase letters show significant differences between the three varieties of strawberry plants (standard errors, NKT, p-value < 0.05).

Two additional experiments were carried out from April to June 2016 (R2) and from August to October 2016 (R3) to evaluate the level of sensitivity of Candiss that has undergone four successive UV-C treatments against *B. cinerea* (Bc21). These experiments showed similar trends than the first experiment (R1), performed from January to March 2016 (Figure 19). The decrease of the sensitivity level of the plant against Bc1 was shown in Figure 19. Except for the R2 experiment, the reduction in sensitivity against Bc1 after treatment with UV-C radiations was statistically significant (p-value < 0.05).

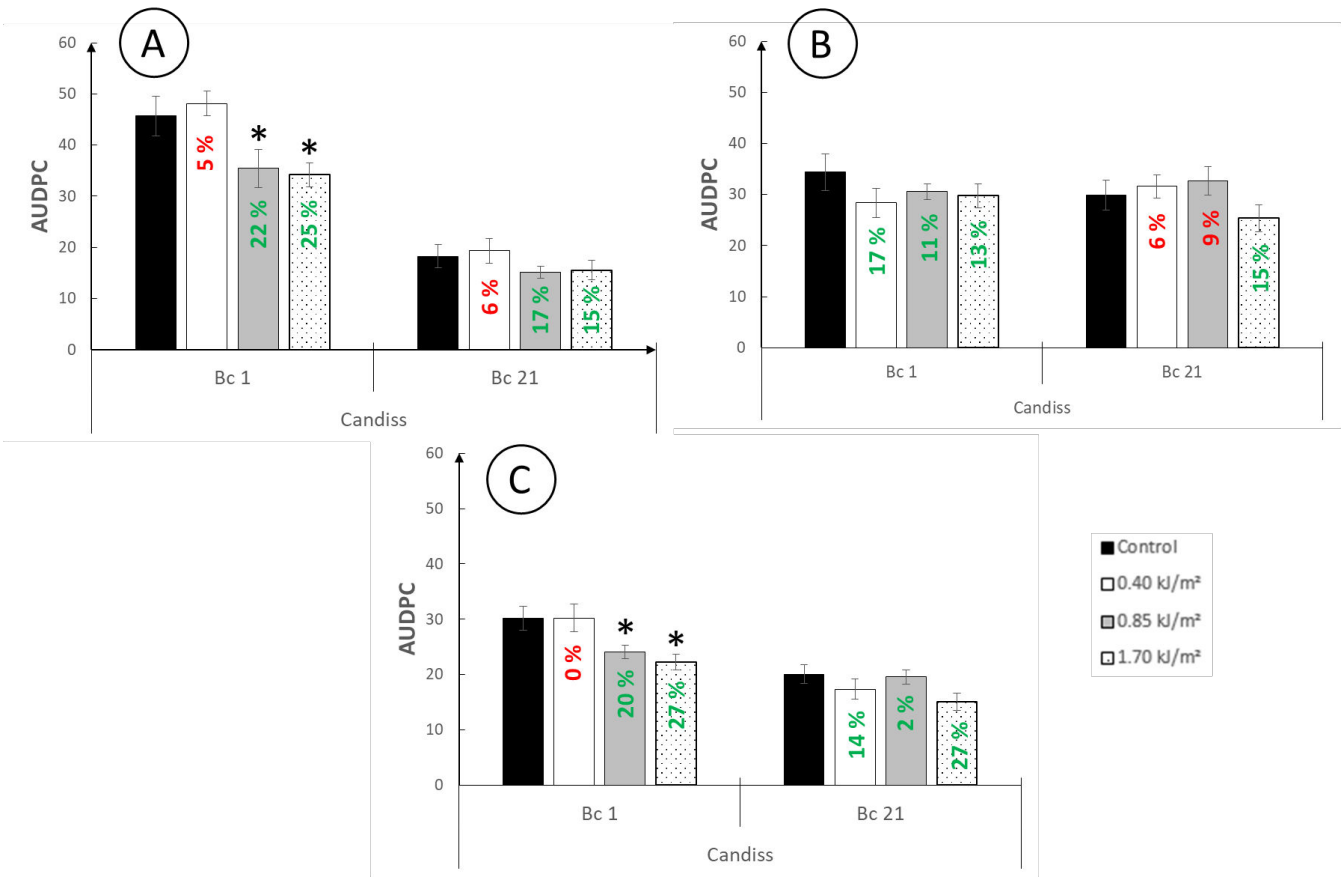


Figure 19: **Sensitivity level of Candiss with two strains of *B. cinerea* after UV-C treatments**. Plants were treated with UV-C radiation at different doses each two days during one week. Pathological tests are performed on separate leaves and inoculation was done by filing mycelial implants on the main rib of each leaves. Sensitivity level is correlated with symptom development and therefore disease development: calculating the AUDPC. Plants grew on different seasons (three experimental repetitions): A) Spring (R1). B) Summer (R2). C) Autumn (R3). The (*) shows significant differences between the inoculated leaves and control (standard errors, NKT, p-value < 0.05).

c. Impact of UV-C on chlorophyll fluorescence parameters

Chlorophyll fluorescence parameters (Annexe 2) were derived from the analysis of maximal fluorescence induction curves with the objective of assessing whether UV-C treatments had damaging effects on leaves or disturbing effects on energy or electron fluxes in or around photosystem PSII.

Some evidence for damaging effects were observed on Charlotte at high doses of UV-C treatments (Figures 20-B and 20-C) as shown by the decrease in the normalized area of the fluorescence induction curve (Sm). In the 0.85 + 1.30 kJ/m^2 UV-C treatment, the decrease in Sm was accompanied by a substantial increase in all parameters of energy or electron fluxes (x/RC) expressed on an active PSII reaction center basis (Figure 20-B). The Vk/Vj ratio decreased in Candiss at 4 x 0.40 kJ/m^2, at 4 x 0.85 kJ/m^2 and at 4 x 1.70 kJ/m^2 (Figure 20-C) and in Cirafine at 0.85 kJ/m^2 and at 4 x 0.40 kJ/m^2 (Figures 20-A and 20-C).

Other evidence for damaging effects were found in Cirafine at 0.40 + 0.60 kJ/m^2 and at 0.85 + 1.30 kJ/m^2, as assessed by substantial decreases in both Sm and Vk/Vj (Figure 20-B), and in Cirafine at 0.85 kJ/m^2 and at 4 x 0.40 kJ/m^2, as assessed by decreases in Vk/Vj. Damaging effects in Cirafine were accompanied by changes in energy and electron transport in PSII which were especially marked at 0.85 kJ/m^2 although PI was not negatively affected (Figure 20-A).

Decreases in the PI (Srivastava and Strasser, 1999; Stirbet and Govindjee, 2011; Strasser and Srivastava, 1995; Strasser *et al.*, 2004) were observed in Charlotte at 4 x 0.40 kJ/m^2, at 4 x 0.85 kJ/m^2 and at 4 x 1.70 kJ/m^2 (Figure 20-C) and in Cirafine at 0.40 + 0.60 kJ/m^2 and at 0.85 + 1.30 kJ/m^2 (Figure 20-B). For Charlotte at the 4 x 0.85 kJ/m^2 and the 4 x 1.70 kJ/m^2 treatments, the decrease could be attributable to the decrease in Fv/F0 (Figure 20-C), which is an indicator of trapping probability. For Cirafine, at the 0.40 + 0.60 kJ/m^2 and the 0.85 + 1.30 kJ/m^2 treatments, the decrease in PI could be attributable to the decrease in $(1 - V_J)/ V_J$,

which is an indicator of the performance of conversion of excitation energy to photosynthetic electron transport (Figure 20-B).

The damaging effects of the 4 x 0.40, the 4 x 0.85 and the 4 x 1.70 kJ/m^2 treatments in Candiss, as assessed by the observed decreases in Vk/Vj (Figure 21-C or Figure 21-A), appear to be very dependent on the season since they were absent in the summer (Figure 21-B) as well as in the autumn (Figure 21-C). Similarly, the negative spring effects of these treatments on the electron transport flux from Q_A to Q_B per PSII, Et_0/RC, on the electron transport flux until PSI, RE_0/RC, or on the efficiency of electron transfer from Q_B until PSI acceptors, $(1 – V_I)/(1 - V_J)$, were not apparent in the summer as well as autumn. There were even positive effects observed in plants resulting from treatments in the autumn, notably for the 4 x 0.40 kJ/m^2 dose, on $(1 – V_J)/ V_J$ and therefore, on PI; on RE_0/RC as well as on $(1 – V_I)/(1 - V_J)$ (Figure 21-C).

A

AUFC	Candiss			Charlotte			Cirafine		
	0.40 kJ/²	0.85 kJ/²	1.70 kJ/²	0.40 kJ/²	0.85 kJ/²	1.70 kJ/²	0.40 kJ/²	0.85 kJ/²	1.70 kJ/²
Fv / Fm	3.20	-1.02	-1.96	1.06	-2.68	-0.65	-0.34	2.02	-0.20
F0	0.19	2.22	5.99	-8.41	0.30	-8.07	6.23	-2.51	4.77
Vk / Vj	-12.71	-0.48	-8.91	10.86	**33.71**	12.46	3.18	**-21.94**	-8.66
Sm	8.14	-10.03	11.58	-11.24	-13.18	-19.86	3.50	**26.30**	13.58
ABS / RC	-7.19	6.14	5.93	-8.33	15.18	1.41	1.58	-20.72	-7.29
DiO / RC	-12.14	11.73	52.21	-33.22	10.00	-11.87	2.11	-30.01	-4.81
TrO / RC	-6.09	4.90	-4.31	-0.33	16.85	5.68	1.47	**-18.73**	-7.82
EtO / RC	-9.18	2.08	-8.45	5.87	**25.92**	9.67	3.27	**-19.26**	-7.11
ReO / RC	-13.61	2.32	0.77	7.56	38.24	11.22	4.07	-27.01	-2.81
Pi total	-2.49	-12.52	10.44	21.45	2.60	-2.61	4.81	11.82	12.56
Fv / F0	9.20	-5.12	2.86	-2.60	-14.34	-6.12	-1.93	11.31	0.37
(1-Vj) / Vj	-8.52	-5.48	-3.27	18.51	**23.40**	12.94	4.38	-4.99	-1.63
(1-Vi) / (1-Vj)	-3.32	0.40	8.20	5.65	9.96	1.85	2.73	-7.15	4.51

B

AUFC	Candiss		Charlotte		Cirafine	
	0.40 + 0.60 kJ/m²	0.85 + 1.30 kJ/m²	0.40 + 0.60 kJ/m²	0.85 + 1.30 kJ/m²	0.40 + 0.60 kJ/m²	0.85 + 1.30 kJ/m²
Fv / Fm	0.75	0.12	0.35	-3.35	-0.51	-0.15
F0	-1.23	-1.67	-4.98	-1.75	2.96	7.43
Vk / Vj	-3.15	-3.23	0.91	24.23	**-9.18**	**-8.75**
Sm	7.97	4.28	-13.35	**-21.63**	**6.97**	**5.61**
ABS / RC	-5.86	0.28	-6.32	**25.51**	**-0.42**	**-1.73**
DiO / RC	-8.82	1.10	-22.45	**47.01**	2.07	-0.84
TrO / RC	-5.24	0.11	-2.10	**19.88**	**-0.92**	**-1.91**
EtO / RC	-3.96	-1.34	-0.48	**21.48**	**-4.92**	**-4.99**
ReO / RC	-4.70	-0.41	-6.47	**27.19**	**-3.93**	**-4.64**
Pi total	9.22	-4.47	-8.65	-19.80	**-9.46**	**-7.81**
Fv / F0	4.73	1.42	-2.30	-15.81	-2.57	-0.44
(1-Vj) / Vj	2.41	-5.66	4.19	7.13	**-11.34**	**-8.92**
(1-Vi) / (1-Vj)	-1.34	0.47	-4.63	3.84	**2.94**	**1.36**

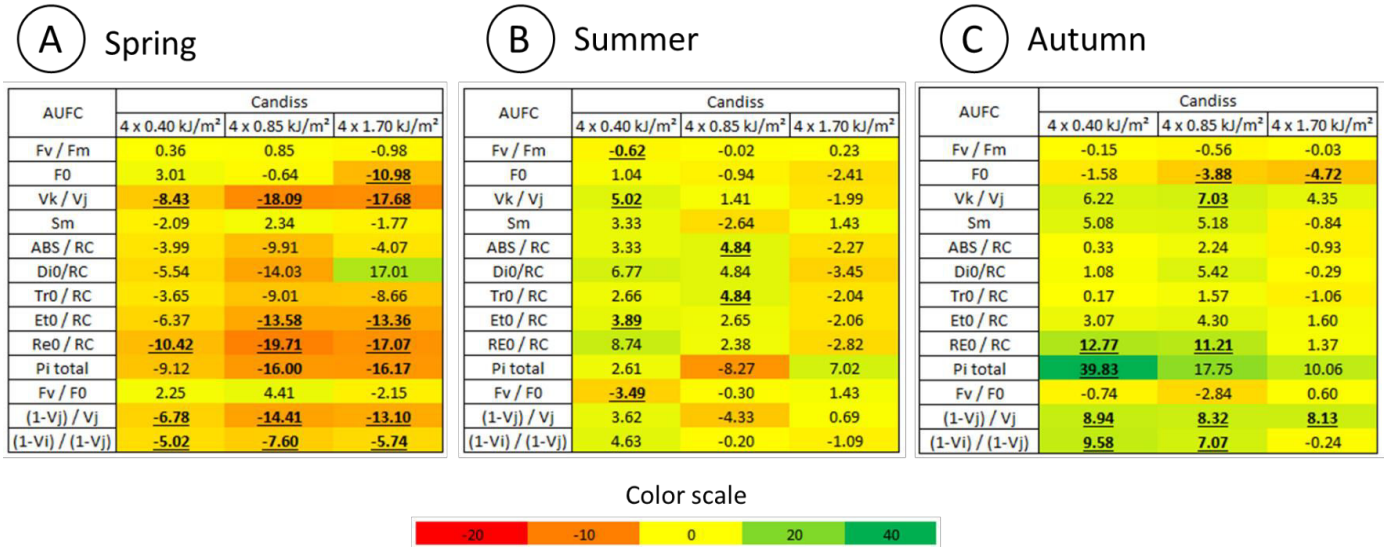
Color scale

-20 0 25 55

C

AUFC	Candiss			Charlotte			Cirafine		
	4 x 0.40 kJ/m²	4 x 0.85 kJ/m²	4 x 1.70 kJ/m²	4 x 0.40 kJ/m²	4 x 0.85 kJ/m²	4 x 1.70 kJ/m²	4 x 0.40 kJ/m²	4 x 0.85 kJ/m²	4 x 1.70 kJ/m²
Fv / Fm	0.36	0.85	-0.98	-0.19	**-2.49**	**-3.28**	0.74	-0.14	-0.88
F0	3.01	-0.64	**-10.98**	-4.15	**-11.01**	**-15.70**	**-3.83**	**-6.23**	**-11.95**
Vk / Vj	**-8.43**	**-18.09**	**-17.68**	-9.83	4.87	2.41	**-13.06**	-7.83	-3.80
Sm	-2.09	2.34	-1.77	-9.68	**-15.74**	**-13.11**	4.34	1.19	-3.76
ABS / RC	-3.99	-9.91	-4.07	-4.39	10.61	2.31	-8.51	-1.63	0.30
DiO/RC	-5.54	-14.03	17.01	-13.29	13.72	5.18	-10.98	1.86	6.30
TrO / RC	-3.65	-9.01	-8.66	-2.33	**9.90**	6.18	-8.01	-2.34	-0.92
EtO / RC	-6.37	**-13.58**	**-13.36**	-8.16	5.77	1.58	-10.93	-5.40	-2.25
ReO / RC	**-10.42**	**-19.71**	**-17.07**	-17.75	3.99	37.59	-15.94	-7.46	-4.82
Pi total	-9.12	**-16.00**	**-16.17**	**-25.90**	**-29.05**	**-21.55**	-10.69	-14.23	-15.10
Fv / F0	2.25	4.41	-2.15	-3.59	**-13.90**	**-14.23**	1.03	-3.09	**-7.42**
(1-Vj) / Vj	**-6.78**	**-14.41**	**-13.10**	-12.01	-6.94	-4.02	-8.80	-7.71	-4.10
(1-Vi) / (1-Vj)	**-5.02**	**-7.60**	**-5.74**	**-9.15**	-0.79	8.48	-5.32	-2.21	-1.16

Figure 20: **Parameters of chlorophyll fluorescence a (OJIP method) of leaves of three cultivars of strawberry plants (Cirafine, Charlotte, Candiss) after UV-C applications**. Plants were treated with UV-C at different doses and different application frequencies. Fluorescence of chlorophyll a were measured on 25 leaves per treatment modality. Data represent the ratio between values of AUFC obtained on UV-C treated plants and on untreated plants, in percentage. A color scale was applied, ranging from red (strong negative effect of UV-C) to green (strong positive effect of UV-C). A) Single application of UV-C radiation. B) Double application of UV-C radiation. C) Four UV-C applications (4 x UV-C) every two days. The values being marked in bold and are underlined represent a significant difference between plants treated with UV-C and controls plants (NKT, p-value < 0.05).



A Spring	Candiss		
AUFC	4 x 0.40 kJ/m²	4 x 0.85 kJ/m²	4 x 1.70 kJ/m²
Fv / Fm	0.36	0.85	-0.98
F0	3.01	-0.64	**-10.98**
Vk / Vj	**-8.43**	**-18.09**	**-17.68**
Sm	-2.09	2.34	-1.77
ABS / RC	-3.99	-9.91	-4.07
Di0/RC	-5.54	-14.03	17.01
Tr0 / RC	-3.65	-9.01	-8.66
Et0 / RC	-6.37	**-13.58**	**-13.36**
Re0 / RC	**-10.42**	**-19.71**	**-17.07**
Pi total	-9.12	**-16.00**	**-16.17**
Fv / F0	2.25	4.41	-2.15
(1-Vj) / Vj	**-6.78**	**-14.41**	**-13.10**
(1-Vi) / (1-Vj)	**-5.02**	**-7.60**	**-5.74**

B Summer	Candiss		
AUFC	4 x 0.40 kJ/m²	4 x 0.85 kJ/m²	4 x 1.70 kJ/m²
Fv / Fm	**-0.62**	-0.02	0.23
F0	1.04	-0.94	-2.41
Vk / Vj	**5.02**	1.41	-1.99
Sm	3.33	-2.64	1.43
ABS / RC	3.33	**4.84**	-2.27
Di0/RC	6.77	4.84	-3.45
Tr0 / RC	2.66	**4.84**	-2.04
Et0 / RC	**3.89**	2.65	-2.06
RE0 / RC	8.74	2.38	-2.82
Pi total	2.61	-8.27	7.02
Fv / F0	**-3.49**	-0.30	1.43
(1-Vj) / Vj	3.62	-4.33	0.69
(1-Vi) / (1-Vj)	4.63	-0.20	-1.09

C Autumn	Candiss		
AUFC	4 x 0.40 kJ/m²	4 x 0.85 kJ/m²	4 x 1.70 kJ/m²
Fv / Fm	-0.15	-0.56	-0.03
F0	-1.58	**-3.88**	**-4.72**
Vk / Vj	6.22	**7.03**	4.35
Sm	5.08	5.18	-0.84
ABS / RC	0.33	2.24	-0.93
Di0/RC	1.08	5.42	-0.29
Tr0 / RC	0.17	1.57	-1.06
Et0 / RC	3.07	4.30	1.60
RE0 / RC	**12.77**	**11.21**	1.37
Pi total	**39.83**	17.75	10.06
Fv / F0	-0.74	-2.84	0.60
(1-Vj) / Vj	**8.94**	**8.32**	**8.13**
(1-Vi) / (1-Vj)	**9.58**	**7.07**	-0.24

Figure 21**: Effect of season on Chlorophyll fluorescence a of Candiss leaves after 4x UV-C applications**. Plants were treated with UV-C radiation at different doses each two days during one week and at different seasons (three experimental repetitions). Fluorescence of chlorophyll a was measured on 25 leaves per UV-C treatment. Here are represented the values of AUFC: data represent the percentage difference between treated plants to UV-C radiation and untreated plants (corresponding to our control group). A color scale was applied, ranging from red (strong negative effect of UV-C treatment on the physiological state of treated plants) to green (strong positive effect of UV-C treatment on the physiological state of treated plants) to highlight important differences. A) Spring (R1). B) Summer (R2). C) Autumn (R3). The values being marked in bold and are underlined represent a significant difference between plants treated with UV-C and controls plants (NKT, p-value < 0.05).

5. Discussion

Our results suggest that the reaction level of strawberry plants to UV-C depends on its basic resistance level to *B. cinerea*. The three strawberry cultivars used in this work (Cirafine, Charlotte and Candiss) had different susceptibility

levels to *B. cinerea*. We demonstrated first that Cirafine was the most resistant while Candiss was the most sensitive and Charlotte had a moderate resistance. These findings are supported by the observation that penetration of *B. cinerea* was deeper in Candiss with concomitant higher phenolic level in the leaves after inoculation. We showed that the level of phenols linked to cell wall increased by 15 % in Cirafine leaves 24 h and 48 h after inoculation contrary to Charlotte and Candiss, which were most susceptible to *B. cinerea*. In the host-*B. cinerea* systems, structural barriers of the epidermis serve as a line of defense (Jarvis, 1980; Elad, 1997).

In addition, the current study highlights that the treatment of strawberry plants with UV-C radiation during cultivation can modify their level of sensitivity to *B. cinerea*. Thus application of UV-C tends to reduce, almost systematically, the level of *B. cinerea* infection on Candiss, the most sensitive cultivar. However, this beneficial effect is only significant at the doses of 0.85 kJ/m² and 1.70 kJ/m² applied on strawberry plants every two days for one week. On the contrary, the application of UV-C radiation on Cirafine tends to increase its sensitivity to *B. cinerea* even though this effect was not always significant. Cirafine leaves seemed less protected against UV-C radiation than Candiss leaves. This hypothesis seems plausible in our work because we found larger amounts of soluble phenolic compounds and PAL activity in Candiss leaves compared to Cirafine leaves after UV-C treatment. It is known that soluble phenolic compounds absorb UV radiation thereby reducing the adverse effects they can have on the physiological status of plants. Phenolic compounds play a major role in photoprotection of plants due to their UV screening and antioxidant properties (Agati *et al.*, 2013; Carbonell-Bejerano *et al.*, 2014; Park and Kim, 2015; Rivera-Pastrana *et al.*, 2014; Tiecher *et al.*, 2013). In the case of Charlotte, the UV-C effect was intermediate between Candiss and Cirafine.

These results suggest that UV-C treatment applied on strawberry plants should be defined for each cultivar in order to know if the UV-C treatment is beneficial (increase in resistance level) or deleterious (increase sensitivity level). Previous studies have shown that the optimum dose of UV-C to achieve beneficial effects in postharvest crops is dependent on the plant species (Mercier *et al.*, 1993b;

Baka *et al.*, 1999; Maharaj *et al.*, 1999; Mercier *et al.*, 2001). We have demonstrated in this study that the effect of UV-C applied on the plant is dependent on the cultivar within a plant species. To our knowledge this is the only study that shows the impact of the plant cultivar on the efficacy of UV-C treatment. Two cultivars of *Pelargonium x hortorum* have been tested in a previous study, where UV-C treatment reduced the sensitivity to *B. cinerea* by 47 % with both cultivars (Darras *et al.*, 2015). Several authors have observed a biochemical production of secondary metabolites via the shikimic pathway is strongly enhanced by UV-C irradiation (Charles *et al.*, 2008d) and mainly related to significant increases in PAL activity (El-Ghaouth *et al.*, 2003; Lemoine *et al.*, 2007; Pombo *et al.*, 2011; Vasquez *et al.*, 2017) and/or other defense related-enzymes such as SOD, catalase (*CAT*), peroxidase (*POD*), polyphenol oxidase (*PPO*) and ascorbate peroxidase (*APX*) (Kim *et al.*, 2007; Tang *et al.*, 2010; Rai *et al.*, 2011; Darras *et al.*, 2012; Vasquez *et al.*, 2017). It is clear that the synthesis of phenolic compounds plays a role in plant resistance to pathogens.

Chlorophyll fluorescence-based indicators of damage and PI data show that with the exception of the 0.85 kJ/m^2 treatment for Cirafine, single dose of UV-C in the 0.40 to 1.70 kJ/m^2 range was not harmful and generally speaking not even disturbing. The picture becomes different for the 0.40 + 0.60 and the 0.85 + 1.30 kJ/m^2 doses. Whereas Candiss shows no sign of damage or downregulation, Charlotte at 0.85 + 1.30 kJ/m^2 and Cirafine at both doses seemed to have consequences. Even though not always significant, the UV-C effect was increasingly damaging with 4 x 0.40, 4 x 0.85 and 4 x 1.70 kJ/m^2 doses, which resulted in decreases in either Vk/Vj or Sm in all cultivars, and impact negatively PI. It must be emphasized, however, that the negative effects of these doses are strongly season-dependent. Season has a marked effect on plants which can lead to differences in behavior when attacking a pathogen or when a light stress is present like UV-C radiation. Plants grow under different climatic conditions depending on the season, which can cause physiological changes such as the structure of the leaf taking for example the thickness of the cuticle acting as a physical barrier slowing more or less the installation of the pathogen at the infection site. Negative effects

were not observed with the treatments either in the summer or in the autumn, but they exerted even positive effects, especially with the 4 x 0.40 kJ/m^2 dose. The ratio Vk/Vj decreased in Candiss and in Cirafine. It suggested that these treatments resulted in either a decrease in the functional antenna size (Yusuf *et al.*, 2010) or OEC (*oxygen evolving complex*) inactivation/damage (Kalachanis and Manetas, 2010). Season is a major factor to influence plant physiology. The light intensity is a parameter to be taken into account because it is a regulator of photosynthesis and it is strongly linked to the fluorescence of chlorophyll a.

During this work, one of major challenges was to find a dose that was both effective against the targeted pathogen and harmless for plants. Consequently, we have applied a range of dose of UV-C radiation based on previous results obtained on various plant species (Casati and Walbot, 2004, Frohnmeyer and Staiger, 2003; Vasquez *et al.*, 2017). However, a wider range of treatment modalities (doses x frequencies of application) deserve to be evaluated in future studies to identify potentially beneficial treatment modes for all strawberry cultivars. Moreover, Xie *et al.* (2015) had shown that strawberry cultivar and growing season played a more important role than UV-C treatment concerning fruit phenolic metabolism. Therefore, it is most likely that the variations in the sun radiation affected growth and physiological parameters. Our study raises fundamental points to be taken into consideration during treatments using UV-C: there are differences since UV-C treatments seem more effective in decreasing susceptibility of Candiss towards Bc1 in both spring and autumn, whereas susceptibility to damaging effects of UV-C treatments seems to be maximal in the spring, and progressively decreasing thereafter.

6. Acknowledgments

This paper was supported by the Federative Structure Tersys. The authors thank Ciref to give us strawberry plants. The authors thank Joël Béraud and Michel Pascal for their help in greenhouse.