

Enhanced Antioxidant Capacity of Fresh Blueberries by Pulsed Light Treatment

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Abstract

As a novel technology for food safety risk mitigation, pulsed light (PL) has been shown effective in surface decontamination of fresh blueberries in literature. However, little is known about the effects of PL on the antioxidant capacity and quality characteristics of fresh blueberries. Fresh blueberries from a local farm were treated with PL for 30, 60, 90 and 120 s. Results show that PL exposure enhanced the antioxidant activity (ORAC) and total phenolic content of fresh blueberries 50 and 48% respectively, relative to the control. Pulsed light also significantly increased the total anthocyanin contents, which may be due to the upregulation of Phenylalanine Ammonium Lyase (PAL) enzymes. There was no significant difference ($P \leq 0.05$) in the soluble solids, pH, titratable acidity, firmness, color and mass of the fresh blueberries within 120 s PL exposure. In conclusion, PL illumination enhanced the antioxidant capacity of fresh blueberries while maintaining other quality characteristics.

Keywords: nonthermal, phytochemical, anthocyanins, phenylalanine ammonium lyase, phenolics, flavonoids

1. Introduction

Blueberries (*Vaccinium corymbosum*) are a rich source of phenolic compounds, including quercetin, procyanidins and flavonoids (Sellappan et al., 2002). Anthocyanins are the major water-soluble flavonoids in blueberries, giving the red, purple and blue color to many fruits and vegetables (Espín et al., 2007). Additionally they are considered biologically active compounds exhibiting a wide range of health benefits, e.g., antioxidant (Cao et al., 1997), antifungal (Benkeblia, 2004), and anti-carcinogenic properties (Ames, 1983). It is well established that free radicals play an important role in the etiology of many diseases, such as cancer, vascular and neurodegenerative disease (Jacob, 1995). Espín et al. (2007) reported that anthocyanins and anthocyanin-rich berries or plant extracts exhibited a wide range of potential health benefits in both human and animal studies. These benefits are attributed to their free radical scavenging properties and their unique capacity in absorbing oxygen radicals, and chelating metal ions which are strong oxidizing agents of deoxyribonucleic acid (DNA) (Halliwell, 1995).

Among other fresh fruits, blueberries are regarded as the agricultural commodity rating the highest in antioxidant capacity, which warrants their potential health benefits and status as a functional food (U.S. Highbush Blueberry Council, 2011). It has been estimated that U.S. per capita fresh blueberry consumption would increase 65% from estimated 0.756 kg in 2008 to 1.25 kg by 2015 (U.S. Highbush Blueberry Council, 2009). This predicted increase in consumption is also accompanied with a demand for blueberry products with minimal changes in quality, nutritional profile and enhanced shelf life.

Pulsed light (PL) (100-1100 nm) is a novel food processing technology, which has shown effectiveness in decontaminating food surfaces, packaging materials and small berries and fruits including blueberries. To illustrate, Bialka and Demirci (2007) reported that maximum reductions in pathogenic bacteria such as *E. coli* O157: H7 and *Salmonella* were 4.3 and 2.9 Log₁₀ CFU/g respectively, in fresh blueberries using PL. Additionally, Krishnamurthy et al. (2008), have reported on the effects of continuous ultraviolet (UV) light (100-400 nm), primarily UV-C (100-290 nm), which has been predominantly used for its germicidal properties. However, the effects of PL on fresh blueberries antioxidant properties, is little known. The efficacy of PL can be attributed to its photothermal, photochemical and photophysical effects. We hypothesized that similar to UV light, PL exposure

might have enhancing effects on the antioxidant potency of fresh blueberries, besides its preservation on fruit quality. The most studied enzymes associated with secondary metabolite synthesis (anthocyanins and polyphenolics) in plants are PALs. Synthesis of these metabolites catalyzed by PAL is achieved through several pathways (Pentose Phosphate, Shikimate, Phenylpropanoid and Flavonoid), which may function synergistically (Cao et al., 2010) to counteract the damaging effects external stressors on cell components such as UV light. Therefore, the objective of this study was to investigate the effects of PL on the total antioxidant activity, polyphenolic contents, PAL enzyme, physiochemical attributes (pH, TA, SS), and quality (color, texture) of fresh blueberries.

2. Method

2.1 Sample Preparation

Early season (May - June) highbush blueberries were handpicked from a local farm (Gainesville, FL, USA) at commercial maturity stage from trees within the same grove. The fruits were stored at 4 °C for 1 day, and prior to PL treatment, they were sorted to remove impurities, washed and left to dry at ambient temperature (\approx 20-25 °C) for 1 day.

2.2 Pulsed Light Treatment

Blueberries (20 ± 1 g) were placed in 70 mL aluminum dishes. Samples were transferred to the Xenon PL system model LH840 LMP HSG (Xenon Corporation, Wilmington, MA, USA) and treated at 3 pulses/s for 30, 60, 90 and 120 s at a distance of 13 cm from the lamp in stationary mode. The initial and final surface temperature of the samples was measured using a handheld infrared thermometer (Omega OS423-LS, Omega Technologies, Stamford, CT, USA).

2.3 Antioxidant Capacity

The blueberry samples were macerated using a homogenizer and extracted with a solvent mixture (20 mL) of acetone/water/acetic acid (70:29.9:0.1, v/v/v), followed by sonication (Zenith ultrasonic bath, 25 kHz, 1350 W, Zenith Manufacturing and Chemical Corp, Norwood, NJ, USA) for 30 min. The extracts were centrifuged (12,000 G [10,000 rpm], 45 min, 4 °C) and the supernatant was collected for oxygen radical absorbance capacity (ORAC) analysis as described by (Huang et al., 2002). Fifty μ L ORAC phosphate buffer (PB) (75 mM ORAC-PB) and samples were added to a 96-well black plate (Fisher Scientific, Pittsburgh, PA, USA), followed by addition of 100 μ L fluorescein (20 mM) solution. The mixture was incubated (37 °C, 10 min) before adding the peroxy radical generator 2,2'-azobis (2-amidinopropane) dihydrochloride (140 mM AAPH). The rate of fluorescence decay (485 nm excitation and 530 nm emissions for 1 min intervals for 40 min) was monitored by using a microplate reader (Spectra Max Gemini XPS Molecular Devices, Sunnyvale, CA, USA). The rate of florescent decay was then calculated using the area under the fluorescent decay using the Trolox standard curve. Antioxidant capacities were expressed as μ mol trolox equivalents (TE)/g extracted samples.

2.4 Phenylalanine Ammonia Lyase (PAL) Enzyme

Under minimal light conditions, enzyme extraction was performed using untreated and PL treated blueberry samples (4 g) added to polyvinylpyrrolidone (PVP) (0.4 g) and homogenized into 16 ml of ice-cold borate buffer (50 mM borate; 400 μ L/L β -mercaptoethanol; pH 8.5) (Velazquez et al., 2011). The homogenates were subsequently centrifuged (12,000 G [10,000 rpm], 30 min, 4 °C), and supernatants maintained at 4 °C and promptly assayed. To a 96 well clear plate, 235 μ L borate buffer, 35 μ L L-phenylalanine substrate solution (100 mM) and 80 μ L of PAL extract were added. Using a microplate reader (Synergy HT, Biotek Instruments INC, Vermont, USA) spectrometric readings (290 nm) were taken before and after 1 h of incubation (40 °C) versus a reagent blank. The PAL activity (mg of t-cinnamic acid/ h) was calculated using cinnamic acid as a standard (0.02-0.10 mg/mL). L-Phenylalanine substrate solution was prepared in nanopure water before each assay.

2.5 Total Polyphenolics

Polyphenolic compounds were extracted as described by Kim *et al.* (2003). The sample mixture (20 g sample, 80% of 200 ml methanol, v/v) was sonicated for 30 min and centrifuged (12,000 G, 50 min, 4 °C). The solvent was evaporated using a Büchi Rotavapor 2025 (Gardner Denver Thomas, Inc., Niles, IL, USA) at 40 °C and the phenolic extracts were stored at -20 °C until further analysis. To a 96 well clear plate (Fisher Scientific, Pittsburgh, PA, USA), 12.5 μ L of 2 M Folin-Ciocalteu phenol reagent was added to 50 μ L of deionized distilled water (ddH₂O) and 12.5 μ L of sample. After 5 min, 7% sodium carbonate (Na₂CO₃) solution (125 μ L) was added to the mixture and incubated (90 min, 25 °C). Sample absorbance was measured at 750 nm using a microplate reader. A standard curve was developed using gallic acid and the concentration was expressed as mg gallic acid equivalents (GAE)/g fruit on a wet basis.

2.6 Total Flavonoids

A standard colorimetric assay (Kim et al., 2003) with slight modifications was used to quantify total flavonoid content. 25 μ L of the sample along with 125 μ L of ddH₂O was added to a 96 well clear plate. Subsequently, 7.5 μ L of 5% sodium nitrate (NaNO₂) was added to the mixture and allowed to stand for 5 min. Fifteen microliters of 10% aluminum chloride (AlCl₃) was added to the mixture and incubated at ambient temperature for an additional 5 min. Following that, 50 μ L of sodium hydroxide (1M, NaOH) was added to the mixture and immediately diluted by the addition of 27.5 μ L of ddH₂O. The absorbance of the mixture was measured at a wavelength of 510 nm against a reagent blank and compared to a catechin standard using a microplate reader. The total flavonoids was calculated as mg of catechin equivalents (CE)/g of fruit.

2.7 Total Anthocyanins

Two buffer systems (pH differential method) composed of potassium chloride (KCl) (pH 1.0, 0.025 M) and sodium acetate (NaC₂H₃O₂) (pH 4.5, 0.4 M) were used to perform anthocyanin analysis as described by Benvenuti et al. (2004). An aliquot of the blueberry extracts were simultaneously diluted (1:10) and adjusted to pH 1.0 and pH 4.5 using the respective buffers, and incubated at ambient temperature for 20 min. Absorbance was measured at each pH using a UV/VIS spectrophotometer (Beckman Coulter, Du 730, Life Sciences UV/VIS, Lawrence, KS, USA) at 510 and 700 nm respectively. Results were calculated using equations 1 and 2 below and expressed as mg cyanidin-3-glucoside (predominant anthocyanin) per g fruit using the corresponding MW (molecular weight [449.2]) and ϵ (molar absorptivity [26 900]). The acronyms A (absorbance) and DF (dilution factor) also denote the elements used in the equations below (Sellappan et al., 2002).

$$A = (A_{510 \text{ nm}} - A_{700 \text{ nm}})_{\text{pH}1.0} - (A_{510 \text{ nm}} - A_{700 \text{ nm}})_{\text{pH}4.5} \quad (1)$$

$$\text{Anthocyanins} = A \times \text{MW} \times \text{DF} \times 1000 / (\epsilon \times 1) \quad (2)$$

2.8 Color Analysis

The blueberry color was measured using a machine vision system (Nikon D200 digital camera housed in a light box [42.5 cm (W) \times 61.0 cm (L) \times 78.1 cm (H)] (Wallat 2002). The camera (focal light, 35 mm; polarization, 18.44 mm) was controlled by the LensEye software (Engineering and Cybersolutions Inc. Gainesville, FL, USA) and calibrated with a standard blue tile (L: 58.24; a*: -4.74; b*: -42.44) (Labsphere, North Sutton, NH, USA).

2.9 Texture Analysis

A texture analyzer (TA.XT Plus, Texture Technologies Corporation, Scarsdale, NY, USA) was utilized to evaluate the firmness of the blueberries by the compression test. The firmness was measured at the surface of the horizontally aligned blueberry to a total distance of 5 mm, using a TA-212 5/16" diameter probe (radius of curvature 13/64") and subjecting 50 kg of force a speed of 2.0 mm/s.

2.10 Soluble Solids (SS), pH and Titratable Acidity (TA)

Soluble solids were determined using a digital refractometer (Leica Mark II Abbe Refractometer, Buffalo, NY, USA) and expressed as Brix°. The sample pH was measured using a pH meter (Fisher Scientific Accumet® Basic AB15/157, Pittsburgh, PA, USA). For TA, 10 g of blueberry samples was macerated into 50 mL of ddH₂O and titrated with sodium hydroxide (NaOH, 0.1 M) until a pH of 8.2 was obtained. The TA results were reported as % equivalent weight of malic acid/g fruit.

2.11 Statistical Analysis

The data obtained was analyzed using a statistical analysis system (SAS 9.1). Analysis of variance (one-way ANOVA) was performed and the significant differences in the means were separated using the Tukey's studentized range test. The data was tabulated as an average of triplicates \pm standard deviation, and the level of significance was determined at $P \leq 0.05$.

3. Results and Discussion

3.1 Antioxidant Capacity

No significant differences ($P \leq 0.05$) were observed in ORAC values between control and PL treatments of 60 and 90 s. However, significant differences ($P \leq 0.05$) existed between control 30 and 120 s (Table 1). There was nearly a 75% increase in ORAC values (30 s) relative to control. Similarly, Wang et al. (2009) evaluated the antioxidant capacity and individual flavonoid compounds in blueberries exposed to UV-C illumination for 1, 5, 10 and 15 min. They found an increase in ORAC values compared to the control. Their values ranged from a low of 40.4 ± 3.2 (Control) to a high of 59.6 ± 2.0 μ mol TE/g (UV-C illumination at 6.45 kJ m⁻² which was lower than

our reported values. Our results indicated that PL is also capable of increasing the antioxidant content of blueberries, which was achieved at much shorter exposure times than those of UV-C illumination.

Table 1. Antioxidant capacity of pulsed light treated blueberries expressed as the oxygen radical absorbance capacity (ORAC)

Treatment	$\mu\text{mol Trolox eq/g fresh weight}$
Control	119.2 \pm 4.82 b
PL 30 s	131.7 \pm 14.7 a
PL 60 s	121.4 \pm 14.7 b
PL 90 s	117.4 \pm 5.14 b
PL 120 s	97.9 \pm 4.14 c

Means (in columns) with the same letter are not significantly different according to the Tukey's studentized Range Test $P \leq 0.05$. Data are expressed as mean \pm standard deviation (SD).

3.2 Phenylalanine Ammonium Lyase (PAL)

In our study, the stimulatory effect of PL on PAL activity was investigated. The highest response was observed at a treatment time of 120 s (Figure 1). This may account for the increase in the antioxidant capacity and anthocyanin content of the blueberries as observed in our study. Previous research (Dixon & Paiva, 1995) indicates that PAL are photo induced by intense or UV light as a defensive response to attenuate its intensity which may damage photosynthetic cells. In this experiment, reduced enzymatic activity was observed at 90s. While an increase was expected as observed in blueberries treated with PL for 30 and 60s, the disparity in the enzyme activity could have been attributed to the enzymes biosynthesis pathways where tyrosine instead of phenylalanine could have been the main substrate resulting in another end product mainly coumaric acid instead of cinnamic acid (Cao et al., 2010).

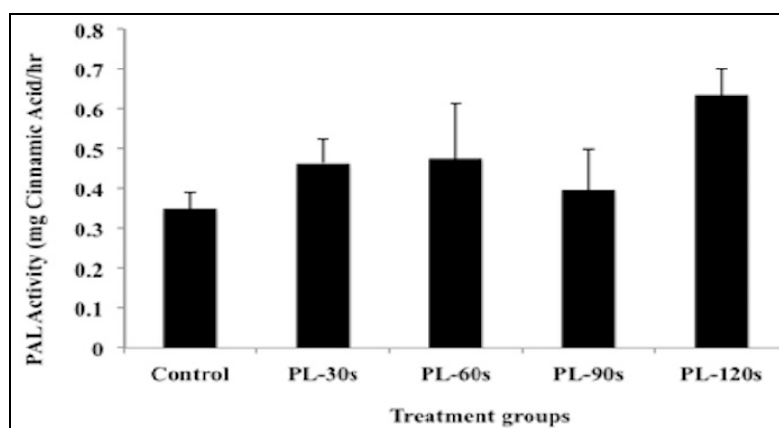


Figure 1. Stimulatory Effect of PL illumination on PAL activity

3.3 Total Phenolics

There was no significant difference ($P \leq 0.05$) in total phenolics between PL 30 s and control. A significant increase ($P \leq 0.05$) was found for PL 60 and 90 s compared to control (Table 2). The highest percentage increase relative to control was 48% at PL 90 s, indicating that PL had a significant ($P \leq 0.05$) enhancing effect on total phenolics within 90 s exposure. However, a decrease in total phenolics was observed at PL 120 s, which had a significantly ($P \leq 0.05$) lower value than PL 90 s, but comparable with control. Phenolic decrease in our experiments may be attributed to increased polyphenoloxidase (PPO) activity, which may have resulted in oxidation of these compounds as suggested by Moreno et al. (2007). According to Agarwal (2007), enzymes such as PPO function as antioxidants against oxidative stress, induced by the UV-B spectrum of PL, which may have upregulated their activity resulting in the oxidation of phenols to quinones. However, the fact that the total

phenolics at 120 s was not significantly different ($P \leq 0.05$) from control, suggested that PL did not have a degradative effect on the total phenolic content. Similar trends were observed by Wang et al. (2009). They found an increased level of total phenolics proportionate to the time of UV-C exposure. They reported a low of 3.12 ± 0.06 (control) to a high of 4.97 ± 0.09 mg GAE/g at the energy strength of 2.15 kJ m^{-2} .

3.4 Total Flavonoids

The flavonoid content significantly ($P \leq 0.05$) increased in all PL treated samples after 30 s, 60 s, 90 s and 120 s of PUV exposure as compared to the control (Table 2). Our values ranged from a low of 0.147 ± 0.01 mg/CE/g fruit (Control) to a high of 0.197 ± 0.02 mg/CE/g fruit (PUV 30s). However, although there was an increase in flavonoid content, there were no significant ($P \leq 0.05$) differences observed among the samples treated with PUV at 60s, 90s and 120s, showing that exposure after 30s did not significantly increase flavonoid content. The main underlying mechanism for this phenomenon was explained by Wang et al. (2009). They mentioned that at high doses of UV light might result in too much stress resulting in injury consequently inhibiting further flavonoid synthesis.

3.5 Total Anthocyanins

It was observed that PL stimulated an increase in blueberry anthocyanins, which ranged from a low of $0.738 \text{ mg/g} \pm 0.27$ (control) to a high of $0.962 \text{ mg/g} \pm 0.24$ (PL 120 s) (Table 2). Significant differences ($P \leq 0.05$) were observed in all PL treated samples relative to control, however, no significant differences were observed among the samples PL treated for 30, 60 and 90 s. As alluded to previously, PAL enzymes are associated with the biosynthesis of anthocyanins. This provides direct evidence as to why there was an increase in the anthocyanin content of blueberries as observed in our study. According to researchers (Heredia & Cineros-Zevallos, 2009), the upregulation of polyphenolic compounds such as anthocyanins may be a major defense mechanism inducible in blueberries due to PL illumination as an external stressor.

Table 2. Effect of pulsed light on total phenolics, flavonoids and anthocyanin content in blueberries

Treatment	Phenolics (mg/GAE/g Fruit)	Flavonoids (mg/CE/g Fruit)	Anthocyanins (mg/g Fruit)
Control	2.08 ± 0.84 a	0.147 ± 0.01 c	0.738 ± 0.28 c
PUV 30 s	2.46 ± 1.42 a	0.196 ± 0.02 a	0.916 ± 0.16 b
PUV 60 s	3.05 ± 0.22 b	0.180 ± 0.01 b	0.882 ± 0.11 b
PUV 90 s	3.07 ± 0.47 b	0.183 ± 0.02 b	0.851 ± 0.13 b
PUV 120 s	2.24 ± 0.44 a	0.190 ± 0.02 b	0.962 ± 0.24 a

Means (in columns) with the same letter are not significantly different according to the Tukey's studentized Range Test $P \leq 0.05$. Data are expressed as mean \pm standard deviation (SD).

3.6 Soluble Solids

Although the mean SS in blueberries ranged from a high of $11.67^\circ \text{Brix} \pm 0.04$ at PL 30 s to a low of $10.86^\circ \text{Brix} \pm 0.04$ at PL 60 s (Table 3), there was no significant difference ($P \leq 0.05$) of SS among all PL treatment groups compared to control. This may suggest that PL did not have an effect on SS metabolism, which may also vary according to blueberry cultivar. Perkins-Veazie et al. (2008) also reported that exposure to UV-C light did not affect SS content, but slight variations may be attributed to cultivar and storage conditions.

3.7 pH

A stable pH is very important to blueberries during processing and post-processing handling, as its hue and color is highly dependent on the pH change. This is because anthocyanin pigments may undergo reversible transformations with the change of pH (Tomas-Barberan & Espin, 2001). Table 3 shows the average pH values after the blueberries were PL treated compared to control. The pH value ranged from a low of 3.07 ± 0.02 at PL 90 s to a high of 3.21 ± 0.04 at PL 120 s. However, in general there was no significant difference ($P \leq 0.05$) in pH value among the PL treated blueberries and no difference either compared to control. This indicates that PL exposure up to 120 s did not have an effect on the pH of fresh blueberries. A similar trend was observed in a study performed by Perkins-Veazie et al. (2008). They reported that after the UV-C exposure (1-15 min, 8 cm from UV lamp), there was a slight increase in blueberry pH values, but there was no significant effect ($P \leq 0.05$).

3.8 Titratable Acidity

The TA values in Table 3 ranged between $0.40\% \pm 0.13$ at PL 120 s and $0.47\% \pm 0.15$ in the control. Our results were comparable with those of Perkins-Veazie et al. (2008), who reported 0.44 and 0.54% malic acid equivalents in the Collins and Bluecrop variety, respectively, after UV-C treatments. Our results showed no significant effect ($P \leq 0.05$) of PL on TA even after exposure for 120 s, which may imply that PL (within 120 s) had no degradative effects.

3.9 Texture Analysis

As an important rheological parameter for textural quality, firmness has been shown in literature as a standard trait to reflect the quality of fresh fruits and vegetables. As shown in Table 3, the firmness values of PL treated vs. untreated (control) blueberries showed no significant difference ($P \leq 0.05$). The firmness values ranged from a low 8.82 ± 0.05 Newtons (N) in blueberries PL treated for 120 s to a high of 10.29 ± 0.04 N in untreated blueberries (control). Our results suggest that PL treatment in the time range tested did not have an effect on the firmness of the blueberries. In contrast, a study by Silva et al. (2005) investigating the variations in the physiochemical (pectin, pH, TA, fiber, skin toughness) observed differences in the firmness in several cultivars of blueberries. They found that the texture ranged from a high of 7.28 N to a low of 3.58 N in the Climax and Jersey cultivars, respectively. Their values were lower than those reported in our study, which may be attributed to varietal differences.

Table 3. Physiochemical properties of pulsed light treated blueberries.

Treatment	pH	Titrateable acidity (% malic acid)	Soluble solids ($^{\circ}$ Brix)	Firmness (Newtons)
Control	3.16 ± 0.06 a	0.48 ± 0.15 a	11.6 ± 0.5 a	10.30 ± 0.03 a
PL 30 s	3.21 ± 0.04 a	0.41 ± 0.02 a	11.7 ± 0.5 a	8.92 ± 0.04 a
PL 60 s	3.13 ± 0.04 a	0.41 ± 0.11 a	10.9 ± 0.2 a	9.32 ± 0.05 a
PL 90 s	3.07 ± 0.02 a	0.42 ± 0.10 a	10.9 ± 0.1 a	9.51 ± 0.03 a
PL 120 s	3.21 ± 0.03 a	0.40 ± 0.13 a	11.4 ± 0.4 a	8.83 ± 0.05 a

Means (in columns) with same letter not significantly different according to the Tukey's Studentized Range Test $P \leq 0.05$. Data is expressed as mean \pm standard deviation (SD).

3.10 Color Analysis

Significant differences ($P \leq 0.05$) were observed among all treatment groups in the degree of lightness (L^*), greenness (a^*), and blueness (b^*) values compared to the control (Table 4). The L^* values indicated that the blueberries darkened in color after prolonged PL exposures. No significant differences ($P \leq 0.05$) were observed between those treated for 30 s and 60 s relative to control. However, those treated for 90 and 120 s ($L^* = 17.23 \pm 1.94$, 15.50 ± 1.75 respectively), were darkest in color as compared to control ($L^* = 20.51 \pm 1.93$) and other treatment groups. Overall, our results showed that PL decreased b^* values (i.e., more dark blue) of the samples as compared to the control. A similar result was observed by Moreno et al. (2007) who investigated the effects of ionizing radiation (0, 1.1, 1.6 and 3.2 kGy) on blueberry color. With a higher dosage of irradiation, there was a decrease in the b^* values resulting in darker fruit. They hypothesized that the darkening of the fruit color in the treated blueberries may be attributed to co-pigmentation where anthocyanins form complexes with flavonoid compounds, causing an increase in color intensity. As previously mentioned, other associative mechanisms during treatment could be the activation of enzymes (PAL) in the pentose phosphate, shikimate, phenylprenoid and flavonoid pathways, associated with the synthesis of anthocyanins that could act as UV screens to reduce the damaging effects of UV on the genetic materials in plant tissues (Tomas-Barberan & Espin, 2001). Our results showed that there was an increase in PAL enzyme activity, indicated by the accumulation of cinnamic acid after PL treatments (Figure 1). These results suggest that exposure to PL might have resulted in the darkening of the fruits.

Table 4. Effect of pulsed light treatment on blueberry color

Treatment	Lightness (L-)	Blueness (-b)	Greenness (-a)
Control	20.51±1.93 a	-7.29±0.93 c	-5.25±0.30 b
PL 30 s	20.40±1.56 a	-6.76±0.39 c	-5.46±0.29 b
PL 60 s	19.38±2.54 a	-6.12±1.12 b,c	-5.36±0.16 b
PL 90 s	17.23±1.94 b	-5.04±1.02 a,b	-5.30±0.17 b
PL 120 s	15.50±1.75 b	-4.35±0.51 a	-4.52±0.69 a

Means (in columns) with same letter not significantly different according to the Tukey's Studentized Range Test $P \leq 0.05$. Data are expressed as mean \pm standard deviation (SD).

3.11 Temperature Rise

Novel technologies such as PL have been used as an alternative to thermal processing of foods (Oms-Oliu et al., 2010). It is well known that foods treated with high temperatures are susceptible to thermal degradation, which may result in undesirable changes to their organoleptic and physiochemical properties. For PL, when the exposure time is short (e.g., seconds), temperature rise during the treatment is low, because the photothermal effect of PL is minimal; however, prolonged exposure (e.g., minutes) would result in temperature increase of the product (Shriver et al., 2011; Yang et al., 2011) as the photothermal effect is intensified and is attributed to the infrared portion of the PL spectra.

According to Table 5, the surface temperature ranged from a low of $22.5^{\circ}\text{C} \pm 0.7$ in untreated (control) blueberries to a high of $35.5^{\circ}\text{C} \pm 1.2$ in PL treated blueberries for 120 s. It is noted that there was a few seconds delay before the temperature reading, when the sample was removed from the treatment chamber upon completion of the PL treatment, so the instantaneous surface temperature could possibly be a bit higher. It was observed that the surface temperature was not significantly different ($P \leq 0.05$) among control, PL 30 and 60, but significantly different after 90 s (Table 5).

Temperature rise to between 45°C and 54°C has been reported to improve sensory and nutritional quality of some horticulture products like tomato (Lurie & others, 2006; Rajchl et al., 2009) or low-temperature storage quality (Vlachonassios et al., 2001) without damage to the product. In contrast, the recorded surface temperature of blueberries after 120 s PL exposure generally did not exceed 54°C , so there should not be any negative impact of the temperature rise on the blueberry quality, although no corresponding tests were conducted to verify such an inference in this study.

3.12 Weight Loss

Pulsed light treatment up to 120 s was found to cause no weight loss of the blueberries (Table 5). The initial mass was 20 ± 1 g for each of the samples, and no mass changes were detected on the balance (accurate to 1 g) for PL 30 to 120 s. Since the evaporation of water from the fruit to the environment is the predominant cause for weight loss during processing (Yang et al., 2010; Duan et al., 2011), the temperature of the product needs to be high enough to initiate the phase change of water from liquid to vapor, before it could escape from the food matrix. As mentioned earlier, the temperature rise of the fresh blueberries after PL treatments up to 120 s was not significant in this study. This explains why no mass changes were recorded in this study.

Table 5. Effect of pulsed light on the surface temperature and mass of fresh blueberries. The initial mass (control) was set to 20 g with triplicate samples

Treatment	Temperature ($^{\circ}\text{C}$)
Control	22.5±0.7 b
PL 30 s	23.0±0.4 b
PL 60 s	25.1±0.3 b
PL 90 s	32.7±0.7 a
PL 120 s	35.5±1.2 a

Means (in columns) with same letter not significantly different according to the Tukey's Studentized Range Test $P \leq 0.05$. Data are expressed as mean \pm standard deviation (SD).

4. Conclusion

Pulsed light exposure up to 120 s had an enhancing effect on the ORAC and total phenolic content of fresh blueberries, without degradative effects on other quality characteristics. The ORAC values and total phenolic content increased by nearly 75% and 48%, respectively, relative to control. Anthocyanin production can be easily elicited and peaked within 30 s PL exposure and then level off for longer exposures up to 120 s. There was no significant difference ($P \leq 0.05$) in the SS, pH, TA, texture, color, and weight loss of the fresh blueberries up to 120 s PL exposure compared to control. The highest temperature rise was within 35.5 °C at 120 s PL exposure. The increase antioxidant capacity and increase in polyphenolics may be attributed to the stimulatory effect of PL on PAL.

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