International Journal of Current Engineering and Technology
E-ISSN 2277 – 4106, P-ISSN 2347 - 5161

Available at http://inpressco.com/category/ijcet

Review Article

Intense Pulse Light in Food Technology-A Review

Ravi Shankar[Å*], U Kaushik [Å] and Shayeeb A Bhat[Å]

[Å]Department of Food Process Engineering, Vaugh School of Agriculture Engineering and Technology, Sam Higgin Bottom Institute of Agriculture, Technology and Sciences-Deemed University, P.O Naini, Allahabad, U.P-211007, India

Accepted 27 May 2014, Available online 05 July 2014, **Vol.4, No.4 (Aug 2014)**

Abstract

This Review Article deals with the principle, working of Pulse Light system, its mechanism of action in inactivation of microorganisms, its applications in food processing and preservation, factor affecting its effectiveness, its nutritional and toxicological aspects; and the research needed in this area for its complete and diversified application.

Keywords: *PLC, Pulse Light, Voltage, Contamination, Toxicological, inactivation.*

1. Introduction

The technique of pulsed light food processing was developed as a non-thermal food processing technique, that involves discharge of high voltage electric pulses (upto 70 Kilovolt/cm) into the food product placed between two electrodes for few seconds (Angersbach *et al*., 2000). It is one of the emerging technologies which are used for the replacement of traditional thermal pasteurization among non thermal processes (Heinz *et al*., 2002). It is a decontamination technique which aims at reducing the pests, spoilage microorganisms and pathogens from food without much effect on its quality (Bank *et al*., 1990). It is recognized by several names in scientific literature i.e., Pulsed ultraviolet light (Sharma and Demirci, 2003), high intensity broad-spectrum pulsed light (Roberts and Hope, 2003), Pulsed light (Rowan *et al*., 1999) and pulsed white light (Marquenie *et al*., 2003).

The pulsed light processing can be described as a sterilization or decontamination technique used mainly to inactivate surface micro-organisms on foods, packaging material and equipments. This technique uses light energy in concentrated form and exposes the substrate to intense short bursts of light (pulses). Typically for food processing about one to twenty flashes per second are applied. Ultraviolet light, broad spectrum white light and near infrared light can be used for pulsed light processing (Green *et al*., 2005). Ultraviolet-C treatment for preserving food was discovered in 1930s (Artes and Allende, 2005).

1.1 Principle

It is the non thermal method of food preservation that involves the generation of pulsed light with gradually increasing from low to high energy and then releasing the highly concentrated energy as broad spectrum bursts, to ensure microbial decontamination on the surface of foods and packaging foods. Within fraction of second, the electromagnetic energy gets stored in the capacitor and is then released in the form of light within a billionth of a second, which results in power amplification and minimum additional energy consumption (Green et al., 2005). The inactivation efficiacy of pulsed light depends upon intensity (measured in Joule/cm-2) and the number of pulses delivered. The flow chart of pulsed electric field is shown in Figure 1.

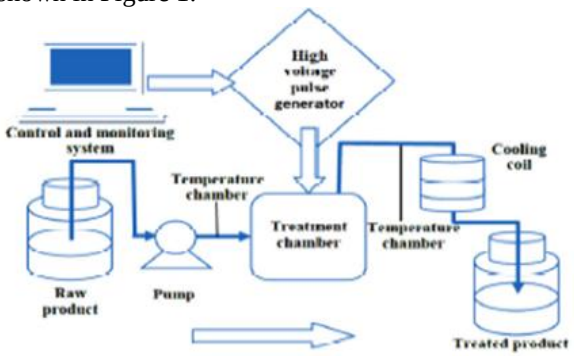

Fig. 1 Flow chart of a PEF food processing system with basic component (Maged *et al*., 2012)

- Pulsed light is produced using engineering technologies that **multiply power many fold**
- Accumulating electrical energy in an energy storage capacitor over relatively long times (a fraction of a s) and releasing this storage energy to do work in a much shorter time (millionths or thousandths of a s) magnifies the power applied
- The result is **a very high power** during the duty cycle, with the expenditure of only **moderate power consumption** (Dunn and others 1995)
- The pulsed light process developed by PurePulse Technologies Inc. utilizes flashes of intense broad-

*Corresponding author: **Ravi Shankar** is M.Tech Food Process Engineering student

spectrum pulsed light (BSPL) to sterilize pharmaceuticals, medical devices, packaging, and water

- The system delivers a spectrum **20,000 times** more intense than **sunlight** at the earth's surface
- The intense flashes of light are less than 1 millisecond in duration (PurePulse Technologies Inc. 1999)
- A different system using pulsed UV light for microbial control has been patented in the United States with patent pending in Chile (Lagunas-Solar and Pyne 1994)
- This technology uses monochromatic excimer lamps at a wavelength of 247 nm rather than the xenon flash tubes
- This technology is in commercial production in Chile and is being used on grapes exported to the United States

Pulsed light (PL) is a technique to decontaminate surfaces by killing microorganisms using short time pulses of an intense broad spectrum, rich in UV-C light. UV-C is the portion of the electromagnetic spectrum corresponding to the band between 200 and 280 nm. PL is produced using technologies that multiply the power manyfold. Power is magnified by storing electricity in a capacitor over relatively long times (fractions of a second) and releasing it in a short time (millionths or thousandths of a second). The emitted light flash has a high peak power and consists of wavelengths from 200 to 1100 nm (Dunn, Bushnell, Ott, & Clark, 1997; Dunn, Ott, & Clark, 1995). The technique used to produce flashes originates, besides high peak power, a greater relative production of light with shorter bactericidal wavelengths (MacGregor *et al*., 1998). This technique has received several names in the scientific literature: pulsed UV light (Sharma & Demirci, 2003), high intensity broad-spectrum pulsed light (Roberts & Hope, 2003), pulsed light (Rowan *et al*., 1999) and pulsed white light (Marquenie, Geeraerd, *et al*., 2003). Barbosa-Canovas, Schaffner, Pierson, and Zhang (2000) reviewed the literature on PL some years ago, warning that most results presented in their report should be confirmed by independent researchers. Nowadays, a higher amount of independently originated data exists that justifies an updated review. According to Wekhof (2000), the first works on disinfection with flash lamps were performed in the late 1970s in Japan, and the first patent dates from 1984 (Hiramoto, 1984). Bank, John, Schmehl, and Dracht (1990) seems to be the first work published in the scientific literature on the application of PL to inactivate microorganisms. By using a UV-C light source of 40 Wat maximum peak power, a 6e7 log decrease in viable cell numbers was achieved. Additional information on this work was published also in Bank (1992).

The technique of UV-C treatment to preserve foods was discovered in the 1930s (Arte´s & Allende, 2005). PL is a modified and claimed improved version of delivering UV-C to bodies. The classical UV-C treatment works in a continuous mode, called continuous-wave (CW) UV light. Inactivation of microorganisms with CW UV systems is achieved by using low-pressure mercury lamps designed to produce energy at 254 nm (monochromatic light), called germicidal light (Bintsis, Litopoulou-Tzanetaki, & Robinson, 2000). More recently, medium-pressure UV lamps have been used because of their much higher germicidal UV power per unit length. Medium-pressure UV lamps emit a polychromatic output, including germicidal wavelengths from 200 to 300 nm (Bolton & Linden, 2003). Another possibility for UV-C treatments is the use of excimer lasers, which can emit pulsed light at 248 nm (Crisosto, Seguel, & Michailides, 1998). PL works with Xenon lamps that can produce flashes several times per second. The following units are commonly used to characterize a PL treatment.

- Fluence rate: is measured in Watt/meter2 (W/m2) and is the energy received from the lamp by the sample per unit area per second.
- Fluence: is measured in Joule/meter2 (J/m2) and is the energy received from the lamp by the sample per unit area during the treatment.
- Dose: used sometimes as a synonym of fluence.
- Exposure time: length in time (seconds) of the treatment.
- Pulse width: time interval (fractions of seconds) during which energy is delivered.
- Pulse-repetition-rate (prr): number of pulses per second (Hertz [Hz]) or commonly expressed as pps (pulses per second).
- Peak power: is measured in Watt (W) and is pulse energy divided by the pulse duration.

Formal definitions can be found in IUPAC (1996). Proper determination of the fluence received by the treated body is the most important factor in characterizing a PL treatment; however, it is sometimes neglected or improperly reported. The same problem also exists in the literature on CW UV treatments (Bolton & Linden, 2003; Hijnen, Beerendonk, & Medema, 2006). Fluence determination can be complex, requiring a good knowledge of light properties. The assistance of experts is sometimes necessary. Researchers with scarce physics background are encouraged to look for advice before planning experiments involving light pulses. Precaution should be taken in reporting the energy received by the sample, which is substantially different from the energy delivered by the light source. Since research on PL is relatively scarce, especially in food applications, no selection was done in this review regarding the accuracy of fluence determinations. Recommendations on fluence determinations for future research can be found in Bolton and Linden (2003), Jin, Mofidi, and Linden (2006), and Ryer (1997).

1.2 Pulsed light devices

The pioneer company producing PL equipment for disinfection was Purepulse Technologies Inc. (San Diego, California), a subsidiary of Xenon Corp., which commercialized the PureBright system. Applications included water purification systems and virus inactivation systems for biopharmaceutical manufacturers. References by Dunn *et al*. (1995, 1997) correspond to the early efforts of this company, which is no longer active, to promote this novel technology. A brief history of the evolution of the pioneer companies related to PL can be found in Wekhof (2000).

As far as we know, there are nowadays two commercial companies producing disinfection systems based on PL. One is SteriBeam Systems from Germany, the other is Xenon Corporation from USA. References by Kaack and Lyager (2007), Wekhof (2000), and Wekhof *et al*. (2001) are associated with SteriBeam, while results reported by Demirci (Hillegas & Demirci, 2003; Jun *et al*., 2003; Krishnamurthy *et al*., 2004; Ozer & Demirci, 2006; Sharma & Demirci, 2003) were obtained with a Xenon Corp. device, mainly the model SteriPulse -XL 3000. Information regarding devices for industrial applications can be found at the websites of these companies. The basic benchtop equipment for laboratory studies is composed of a treatment chamber and a control module (Fig. 2). The treatment chamber is built of stainless steel.

It has a shelf to hold the samples (microorganisms on agar in Petri dishes, or food samples), which can be displaced vertically, allowing to regulate the distance between the target and light source. The light source is a linear Xenon flash lamp located at the top centre of the chamber, inside the lamp housing. A basic benchtop equipment was used in the works by Go´mez-Lo´pez *et al*. (2005a, 2005b), Hillegas and Demirci (2003), Krishnamurthy *et al*. (2004), Lammertyn, De Ketelaere, Marquenie, Molenberghs, and Nicola€ı (2003), Marquenie, Geeraerd, *et al*. (2003), Marquenie, Michiels, Van Impe, Schrevens, and Nicola (2003), Ozer and Demirci (2006), Sharma and Demirci (2003), Takeshita *et al*. (2003) and Wuytack *et al*. (2003) Experimental units used by Jun *et al*. (2003) and Kaack and Lyager (2007) were equipped with two lamps, and that used by Jun *et al*. (2003) had also a blower. By using a blower, a filtered air stream can flow around the lamp serving two functions: dissipating the heat generated by the lamp, and avoiding the accumulation of high levels of toxic ozone produced by the shortest wavelengths. Anderson *et al*. (2000), MacGregor *et al*. (1998), and Rowan *et al*. (1999) used a benchtop experimental facility where two inoculated Petri dishes inclined 45^0 received equivalent doses. Experimental units with more complicated configurations have also been used. Fine and Gervais (2004) used a fluidized bed to mix powders and to increase particle exposure, and Huffman, Slifko, Salisbury, and Rose (2000) plumbed three PureBright units to treat water continuously. Most of the cited references provide schematic representations of the experimental units.

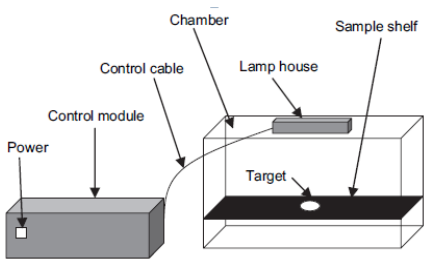

Fig. 2 Schematic representation of a bench top experimental unit of pulse light

A control cable connects the light source with the control module, in which the electric current is modulated to produce a specific prr, pulse width and peak power. The control module has a switch to start the flashing period and a timer to control the exposure time. High peak power is produced by pulse power energization techniques. Information related to this kind of technique can be found in Anderson et al. (2000), Ghasemi, Macgregor, Anderson, and Lamont (2003) and Hancock et al. (2004).

2. Mechanism of microbial inactivation

The lethality of Pulsed Light may be attributed to its rich broad spectrum ultraviolet content, its short duration, high peak power and the ability to regulate the pulse duration and frequency output of flash lamps (Dunn *et al*., 1995., Takeshita *et al*., 2003). As a substantial portion of the Pulsed light spectrum covers ultraviolet light, it is considered that ultraviolet plays a vital role in the microbial cell inactivation. It was also found that that there is no killing effect if a filter is used to remove ultraviolet (UV) wavelength region lower than 320 nm (Takeshita *et al*., 2003). The ultraviolet spectrum comprises of three wave ranges: Long-wave ultraviolet -A (320-400 nm), Medium-wave ultraviolet -B (280-320 nm) and Short-wave ultraviolet -C (200-280 nm).

Mechanisms that have been proposed to explain the lethality of pulsed light treatment are related to ultraviolet (UV) part of the spectrum which include photochemical and photothermal effect (Anderson *et al*., 2000; Takeshita *et al*., 2003; Wuytack *et al*., 2003).

The lethal effect of pulsed light can be due to photochemical or photothermal mechanism or both may exist simultaneously. However their relative importance depends on the fluence and target microorganism. The lethal effect of pulsed light was explained by most of the authors on the basis of photochemical mechanism e.g., the inactivation

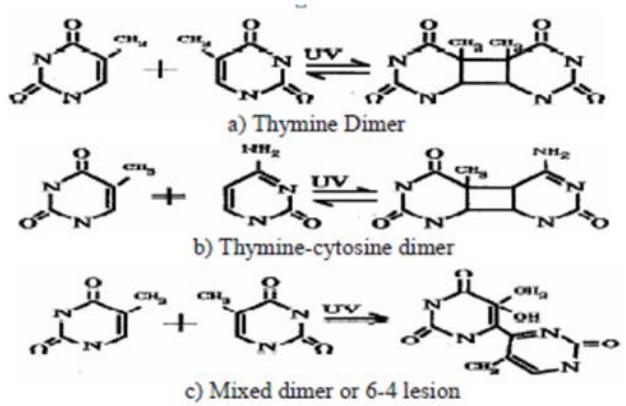

Fig. 3 Formation of Thymine Dimer, Thymine-cytosine dimer and mixed dimer (Setlow *et al*., 1966)

achieved by (Rowan *et al*., 1999) was associated with less than 1oC rise in temperature concluded that the lethality can be attributed to the photochemical action of the shorter ultraviolet wavelengths.

The primary target cell of pulsed light in photochemical mechanism is nucleic acid as DNA is the target cell for these ultraviolet wavelengths (Chang *et al*., 1985; Miller *et al*., 1999). Ultraviolet light absorbed by the

conjugated carbon-carbon double bonds in proteins and nucleic acids induces the antimicrobial effect as it changes the DNA and RNA structures. The bactericidal effect is attributed to the high energy short wave ultraviolet-C range. In the ultraviolet-C range of 250-260 nm, alterations in DNA take place due to pyrimidine dimers mainly thymine dimers (Mitchell *et al*., 1992; Giese and Darby, 2000). Ultraviolet irradiation usually generates thymine dimers in large quantity, cytosine dimers in low quantity and mixed dimers at an intermediate level as shown in Figure 2 (Setlow *et al*., 1966). These dimers inhibit the formation of new DNA chains in the process of cell replication resulting in the chologenic death of affected microorganisms by ultraviolet (Bolton and Linden, 2003). The ultraviolet-C treatment of bacterial spores may result in the formation of spore photo-product 5-thyminyl-5, 6-dihydrothymine and in single-strand breaks, double-strand breaks and cyclobutane pyrimidine dimers (Slieman and Nicholson, 2000). It was also found by experiments that enzymatic repair of DNA does not occur after damaged by pulsed light.

The lethal effect of Pulsed light can also be due to photothermal effect. Wekhof (2000) proposed that with a fluence exceeding 0.5 Joule/cm2, the disinfection is achieved through a rupture of bacteria during their temporary overheating caused by absorption of all ultraviolet light from a flash lamp. This hypothesis become evident by (Wekhof *et al*., 2001) when they showed electron-microscope photographs of flashed Aspergillus niger spores presenting severe deformation and rupture. The ruptured top of spore become evident of an escape of an overheated content of the spore, which became empty after such an internal ‘‘explosion’’ and ‘‘evacuation’’ of its content took place during the light pulse.

Other effects on the cells include, collapse of cell structure, enlargement of vacuoles as found in some microbial studies (Proctor, 2011) as showed by flashed yeast cells. Antimicrobial effects are also manifested due to changes in ion flow, increased cell membrane permeability and depolarization of cell membrane (Ohlsson and Bengtsson, 2002). As Pulsed light causes cell membrane damage, it could be considered as a technique for sterilization (Takeshita *et al*., 2003, Bialka *et al*., 2008).

2.1 Photosensitization: another application of light to inactivate microorganisms

Photosensitization is another technique that might be useful to decontaminate food surfaces by using light. It has been proposed as a milder alternative to the emerging non-thermal technologies for food preservation. Photosensitization has been defined by Luksˇiene (2005) as a treatment involving a photoactive compound that accumulates in microorganisms followed by illumination with visible light. The combination of photosensitizer and light, in the presence of oxygen results in the destruction of microorganisms. After the work by McDonald, Curry, Clevenger, Brazos, *et al*. (2000) described before, it is foreseeable that this definition can be expanded to include the UV part of the spectrum. The primary field of application of photosensitization has been photodynamic cancer treatment. Haematoporphyrin, sodium chlorophyllin, riboflavin and psoralen are examples of photodynamic active plant food constituents that could be used as photosensitizers for foods (Kreitner *et al*., 2001).

Luksˇiene (2005) published a comprehensive review on photosensitization encompassing history, photosensitizers, light sources, and mechanism of microbial inactivation. As an example of photosensitization, Kreitner *et al*. (2001) inactivated as much as 3.9 log CFU/ml of S. aureus cells after incubation for 1 h with haematoporphyrin, followed by illumination for 1 h. To date, its potential application in food preservation has only been tested for the inactivation of bacteria, yeasts (Kreitner *et al*., 2001), and fungal food contaminants in vitro (Luksˇiene, Pe_ciulyte, Jurkoniene, & Puras, 2005; Luksˇiene, Pe_ciulyte, & Lugauskas, 2004). Therefore, more research is necessary to evaluate its future in food preservation.

2.2 Susceptibility of microorganisms

Anderson, Rowan, MacGregor, Fouracre, and Farish (2000) and Rowan *et al*. (1999) reported the following trend of susceptibility in decreasing order: Gram-negative bacteria, Gram-positive bacteria and fungal spores. The colour of the spores can play a significant role in fungal spore susceptibility. Aspergillus niger spores are more resistant than Fusarium culmorum spores, which could be because the pigment of the A. niger spores absorbs more in the UV-C region than that of F. culmorum spores, protecting the spore against UV (Anderson *et al*., 2000). In contrast, Go´mez- Lo´pez, Devlieghere, Bonduelle, and Debevere (2005a) did not observe any sensitivity pattern among different groups of microorganisms, after studying 27 bacterial, yeast and mould species.

2.3 Inactivation curve

The shape of the inactivation curve for microbial inactivation by CW UV light is sigmoid. The initial plateau is due to an injury phase. Once the maximum amount of injury has been surpassed, minimal additional UVexposure would be lethal for microorganisms and survivor numbers would rapidly decline (Barbosa-Canovas *et al*., 2000). The end of the curve has a tailing phase that has received several explanations, which have been summarized by Yaun, Summer, Eifert, and Marcy (2003): lack of homogeneous population, multi-hit phenomena, presence of suspended solids, the use of multiple strains that may vary in their susceptibility to UV-C, varying abilities of cells to repair DNA mutations, and the shading effect that may have been produced by the edge of the Petri dishes used in some experiments. Another possible explanation was given by McDonald, Curry, Clevenger, Brazos, *et al*. (2000) when explaining the tailing of the inactivation curve of Bacillus subtilis treated with PL: the probability of exposing a biological element to the requisite conditions for lethality is reduced with decreasing population density. For PL treatments, the

shape of the inactivation curve of conidia of Botrytis cinerea and Monilia fructigena was also found to be sigmoid (Marquenie, Geeraerd, *et al*., 2003). Anderson *et al*. (2000) and MacGregor *et al*. (1998) reported that the higher the number of pulses the higher the lethal effect. It can be observed in their results that the microbial population as a function of the number of pulses keeps constant until a certain point beyond which the inactivation starts. The same observation was reported by Fine and Gervais (2004) on the viability of S. cerevisiae cells dried on a quartz plate, which suggested a threshold level of energy for total destruction. These findings are in line with the sigmoid pattern discussed before for CW UV lamps. However, complete inactivation of microorganisms and absence of tailing have also been reported (Krishnamurthy, Demirci, & Irudayaraj, 2004; Otaki *et al*., 2003; Wang *et al*., 2005), although the effect of the detection limit of the enumeration method should be better assessed.

2.4 Peak power dependence

Some research has been conducted to investigate if PL sources really yield improved microbial inactivation rates in comparison with CW UV light sources, as claimed by early literature coming from private sources. A peak power effect would not be in line with the BunseneRoscoe law (Sommer, Haider, Cabaj, Heidenreich, & Kundi, 1996). As a general rule in photochemical processes, the principle of equi-effectivity of the product of fluence rate and exposure time is valid. This principle is known as the Bunsene Roscoe reciprocity law. It asserts that for the effectiveness of radiation it does not matter whether the fluence is reached with high fluence rate and short exposure time or with low fluence rate and long exposure time. An exception to this principle has been found for CW UV light treatment (Sommer *et al*., 1996). The diversity of findings does not allow setting a definitive conclusion on the validity of this principle for microbial inactivation by PL, although results point towards possible peak power dependence. According to McDonald, Curry, and Hancock (2002) several theories predict a more rapid kill of vegetative cells with PL. The most probable theory postulates that the high photon flux emanating from a pulsed source simply overwhelms the cellular repair mechanisms before repair can be completed.

Rice and Ewell (2001) examined the peak power dependence in the UV inactivation of bacterial spores by comparing the output of a high-peak-power UV source at 248 nm from an excimer laser to a low-power CW UV source (254 nm) used to inactivate B. subtilis spores. The two UV sources differed by eight orders of magnitude in peak power. Results showed no discernible peak power effect. Therefore, it appears that the total number of photons delivered is the important parameter and not the number of photons delivered per unit time (peak power). The results agree with the principle of BunseneRoscoe. A similar conclusion can be derived from the work of Otaki *et al*. (2003), who compared the inactivation of three strains of E. coli and two coliphage types by a low-pressure UV lamp emitting at 254 nm and a broad-spectrum flash lamp. Working at the same germicidal dose, no differences were found between both lamp sources. Moreover, Wang *et al*. (2005) concluded that the germicidal efficiency obtained with a Xenon flash lamp used to inactivate E. coli shows no obvious difference to the published data on inactivation using CW UV low-pressure mercury lamps at the same wavelength.

However, a single research group working with B. subtilis spores has reported conflicting results and conclusions. Two articles concluded that given the same fluence, no differences in the inactivation efficacy were observed when comparing the results obtained with a CWUV lamp producing 3.9 mW/cm2 on the target to those obtained with two kinds of flash lamps producing in excess of 60 W/cm2 on the target, in the fluence range 0e200 mJ/cm2 (Hancock, Curry, McDonald, & Altgilbers, 2004; McDonald *et al*., 2002). On the other hand, other articles concluded that pulsed UV light exhibits a minor improvement over that of the CW UV source over the fluence range 0e80 mJ/cm2 (McDonald, Curry, Clevenger, Brazos, *et al*., 2000) or that PL significantly outperforms CW UV light in aqueous suspensions and on surfaces (McDonald, Curry, Clevenger, Unklesbay, *et al*., 2000). Results reported by Takeshita *et al*. (2003), also supported a violation of the principle of BunseneRoscoe. The authors compared the effect of peak power on S. cerevisiae cells, using 4655 and 2473 kW. Their results revealed that under high-peak-power conditions, the killing effect and concentration of eluted protein were higher than under low-peak-power conditions. Furthermore, the photothermal effect is not in agreement with the BunseneRoscoe law. It seems that under certain extreme conditions, PL causes different kinds of damage than CW UV.

2.5 Photoreactivation

Photoreactivation means the reversal of ultraviolet damage in bacteria by illumination with visible light (Cleaver, 2003). It is a well known phenomenon in the CW UV treatment field. It is catalysed by the enzyme photolyase, which uses light energy to split UV-induced cyclobutane dimers in damaged DNA through a radical mechanism. ‘‘Photolyase is a flavoprotein and contains two noncovalently bound chromophores. One chromophore is the fully reduced flavin- adenine dinucleotide (FADH_), the catalytic cofactor that carries out the repair function upon excitation by either direct photon absorption or resonance energy transfer from the second chromophore (methenyltetrahydrofolate or deazaflavin) that harvests sunlight and enhances the repair efficiency. The excited flavin cofactor transfers an electron to the cyclobutane pyrimidine dimer to generate a charge separated radical pair. The anionic ring of the dimer is split, and the excess electron returns to the flavin radical to restore the catalytically competent FADH form and close the catalytic photocycle’’ (Kao, Saxena, Wang, Sancar, & Zhong, 2005).

In the PL research field, Rowan *et al*. (1999) wrapped their samples with aluminium foil after PL treatment as a precaution to avoid photoreactivation. Otaki *et al*. (2003)

found photoreactivation after a PL treatment, being the photoreactivation rate slower than after a CW UV treatment. The photoreactivation suppression was assumed to have been due to the difference in wavelength. The wider wavelength light of the pulsed Xenon lamp was considered to have some effects on photoreactivation; for example, shorter wavelengths damage some kinds of enzyme related to this process. Evidence of photoreactivation in flashed cells has also been given by Go´mez-Lo´pez *et al*. (2005a). However, future research is needed to better quantify this phenomenon.

There are two other repair mechanisms for UV damage that might reactivate PL treated cells. One is the dark repair mechanism, which does not require light as photoreactivation does. The other is specifically related to spores. Spores can repair themselves from the spore photoproduct by the common excision repair system, or the spore photoproduct specific repair system (Setlow, 1992).

3. Factors determining the efficacy of a PL treatment

The most important factor determining the effect of PL is the fluence incident on the sample. The energy emitted by the flash lamp is different from the energy incident on the sample. Factors such as distance from light source to target, and propagation vehicle (air, water, fruit juice) affect the level of energy than ultimately reaches the target. The inactivation efficacy of PL is higher when treated samples are closer to the lamp (Hillegas & Demirci, 2003; Ozer & Demirci, 2006). An equation to describe the effect of distance taking into account both the photochemical and the photothermal effects was described by Go´mez- Lo´pez *et al*. (2005a). The effect of distance was modeled by Sharma and Demirci (2003) for inactivation of E. coli O157:H7 on inoculated alfalfa seeds, and by Jun *et al*. (2003) for A. niger spores in corn meal.

Furthermore, tests on microbial inactivation are generally performed by placing the sample directly below the lamp. The effect of the placement of samples at other positions inside the treatment chamber was studied by Go´mez- Lo´pez *et al*. (2005a). The authors demonstrated that when a group of samples is placed at a short vertical distance from the lamp, those located directly below the lamp will be decontaminated while the rest will undergo almost no decontamination. When the vertical distance is increased, the decontamination will be less intense in those samples located directly below the lamp but the rest of the samples will be also decontaminated. Sample thickness is another limiting factor for microbial inactivation with PL. Due to the restricted penetrability of the UV light, overlapping opaque samples shield surfaces from decontamination and also light is attenuated during the treatment of fluid samples. That was observed by Sharma and Demirci (2003) for alfalfa seeds and by Hillegas and Demirci (2003) for honey. The decontamination efficacy decreases at high contamination levels, which is also related to light attenuation. At high population densities, microorganisms overlap each other. Therefore, microorganisms placed in the upper layers will become inactivated, but will shadow the rest from the light (Go´mez-Lo´pez *et al*., 2005a).

Food composition also affects the efficacy of the decontamination by PL. Go´mez-Lo´pez *et al*. (2005b) treated Photobacterium phosphoreum, L. monocytogenes and Candida lambica inoculated onto surfaces of agars supplemented with several food components. The results demonstrated that proteins and oil decreased the decontaminant efficacy of PL, whereas when water or starch was added to the agar, no particular trends were observed. Roberts and Hope (2003) also found that the addition of protein to a buffered saline solution decreased the efficacy of virus inactivation. Therefore, high protein and fat containing food products have little potential to be efficiently treated by PL. Vegetables, on the other hand, could therefore be suitable for PL treatment. With regard to the long term applicability of PL, the possible development of resistant strains should be taken into account. However, Marquenie, Geeraerd, *et al*. (2003) observed no development of resistance in fungi. This was also found by Go´mez-Lo´pez *et al*. (2005a) in the case of L. monocytogenes.

3.1 Critical Process Factors

- Light characteristics (wavelength, intensity, duration and number of the pulses)
- packaging and food attributes (type, color and transparency)
- For fluid food, transparency and depth of the fluid column become critical factor
- The lethality of the pulsed light increases with increasing light intensity or fluence (PurePulse Technologies Inc. 1999)
- Formulation of a model for dose-response is not currently possible

3.2 Factors affecting the microbial inactivation by pulsed light

3.2.1 Type of micro-organism

Optical properties of cells, for example their degree of scattering and absorption of light are important. The incident beam of light undergoes refraction due to difference in the optical density between the substrate and the surrounding air. There are also some micro-organisms resistant to pulsed light (Ethan, 2009; Rajkovic *et al*., 2010; Manzocco *et al*., 2011; Uysal and Kirca, 2011).

3.2.2 Interaction between light and the substrate or between light and the microbial cells (micro-organism)

This factor is very important from the point of view of the efficacy of the pulsed light treatment. The composition of the medium and the wavelength of the incident light decide the reflection, refraction, scattering and absorbing of the light, the refraction and reflection of light being vital for surface treatments. For transparent and coloured

food materials, refraction is particularly relevant, whereas for opaque food materials, reflection is the prevailing phenomenon. Specular or diffused reflection can occur depending on the smoothness or roughness of the surface of the material respectively. For smooth surfaces, the incident light bounces on the surface and comes out at the same angle as the incident beam, with the same spectral distribution of energy, which is termed as specular reflection. For rough surfaces, light travels through the outer layers of the material, where the incident light is partly absorbed, this phenomenon is called diffuse reflection. The absorption at different wavelengths is different and hence the resulting spectral distribution of incident and the diffused light coming out is in all directions (Duran and Calvo, 2002), and reflection of light can tend to decrease the efficiency of the pulsed light treatment. For translucent materials, some part of the incident light interacts with the internal structures and causes multiple internal reflections, redirections which result into scattering. In the case of biological tissues, absorption and scattering are the most relevant types of light–substrate interaction (Cheong et al., 1990).

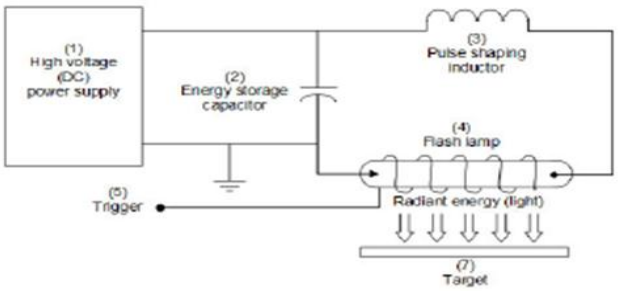

Fig. 4 Functional diagram of a high-intensity pulsed-light system. (Adapted from Xenon Crop., 2005)

3.2.3 The distance from the light source

As the distance from light source and depth of the substrate increases, the absorption and scattering diminishes. This is because the light intensity decreases as it travels through the substrate. The quantitatively distribution of light dose inside a substrate is described by the term Optical penetration depth, which represents the distance over which light decreases in fluence rate to 37% of its initial value. The optical penetration varies with wavelength, with shorter wavelengths providing deeper penetration into the food than longer wavelengths (Dagerskog and Osterstrom, 1979).

3.2.4 Design of pulsed light system

Pulsed-light equipment may vary from manufacturer to manufacturer. The system of pulsed light consists of several common components as shown in Figure 4) A high voltage power supply: provides electrical power to the storage capacitor 2) A storage capacitor: which stores electrical energy for the flash lamp 3) A pulse-forming network: determines the pulse shape and spectrum characteristics 4) The gas discharge flash lamp 5) A trigger signal: which initiates discharging of the electrical energy to the flash lamp, which is the key element of a pulsed light unit.

The flash lamp is the important element of any Pulsed light unit that converts 45% to 50% of the input electrical energy to pulsed radiant energy (Xenon Corp., 2005). This is filled with an inert gas such as xenon or krypton. Xenon is mostly preferred because of its higher conversion efficiency and also because it is a gas of choice for most of the microbial inactivation applications. The envelope, the seals and the electrodes are the main structural components of the flash lamp, the envelope being a jacket that contains the filling gas and also surrounds the electrodes. Figure 4. Functional diagram of a high-intensity pulsed-light system. (Adapted from Xenon Corp., 2005) to the radiations that are emitted by the lamp, be impervious to the filling gas as well as air, must be able to withstand high temperatures and thermal shocks and have mechanical strength. The envelopes are typically made out of clear fused quartz, called suprasil, of about 1 millimetre thickness. Metallic electrodes protrude into each end of the envelope and are connected to the capacitor which is charged to a high voltage. The electrodes provide electric current into the gas. The lifetime of the lamp is determined by the cathode and is hence an important component. The operational requirements decide the material of making of the electrode. The duty of the cathode is to provide unsputtered and adequate amount of electrons, because sputtering, caused due to hot spots created during peak power supply, may lead to corrosion of the cathode material. This in turn would reduce the lifetime of the cathode. The anode should have sufficient mass or surface area to sustain the loading of power caused by the electron bombardment from the electric arc. The whole assembly of the flash lamp needs to be sealed. Commonly used seals include, solder seals, rod seals and ribbon seals.

The gas in the flash lamp undergoes ionisation when subjected to a high voltage, high-current electrical pulse and plasma formation takes place near the anode by the electrons travelling towards it. A very large current pulse formation occurs and this is sent through the ionized gas, exciting the electrons surrounding the gas atoms, causing them to jump to higher energy levels. The electrons while jumping back to their lower energy levels, release quanta of energy producing photons. Overheating problems are encountered during this operation and hence cooling devices are to be provided for long lamp life and undeviating operation. Cooling fans can be used to serve the purpose.

Other pulsed light sources are explored such as solid state marx-generator for pulsing an ultraviolet lamp in microbial inactivation applications, static discharge lamps with spectral outputs similar to flash lamps and sparker technology which generates a light and sonic sound pulse (Proctor, 2011). Adjustable one or more flash lamp units, a power unit and a high voltage connection that allows a high electric pulse transfer are used to produce the pulsed light. The current passing through the gas chamber of the flash lamp unit emits a short intense burst of light. There are numerous patented equipments and are designed to control unique treatment and are product specific (Green *et al*., 2005). The high current discharge through gas filled

flashlights results in millisecond flashes of broad spectrum white light, about 20,000 times more intense than sunlight. Conversion efficiency of electricity to light is about 50%. The spectral distribution is 25% ultraviolet, 45% visible light and 30% infrared. The rate of flashes is 1-20 flashes/sec, a few flashes being generally sufficient for the pasteurizing or sterilizing treatment. This means that the treatment time is very short and throughput is high. Depending on the application, wavelengths that would adversely affect food flavour or quality are filtered off.

The flashlights are arranged in arrays, adapted to the particular application, be it the continuous sterilisation of packaging film in aseptic processing, the sterilisation on-line of transparent liquids or the surface pasteurisation of solid foods in plastic packaging. Most plastic packaging materials transmit broadband light well, exceptions being Polyethylene terephthalate (PET), polycarbonate, polystyrene and polyvinyl chloride (PVC). For complex surfaces, such as those of foods like meat and fish, it will be difficult to illuminate or reach all parts of the surface to obtain a sterilising effect (Ohlsson and Bengtsson, 2002). The type of equipment for food preservation depends on some factors such as ozone build-up, surface area of food product and dimensions of each treatment unit and desired degree of decontamination. A cooling unit facility maybe required in the case of a food under treatment is temperature sensitive (Green *et al*., 2005).

Pulsed light systems can be of either batch or continuous type depending on the usage. In the case of batch processing, such as those developed by Xenon Corp. (Waltham, MA), the packets are placed in a chamber with lamps located along the walls of the chamber. The simplest designs include a single lamp located above the sample and an adjustable tray to hold the samples. More complex designs may incorporate up to eight lamps within a chamber along with a quartz stand to hold the sample and allow a 360o exposure and treatment. In the case of continuous processing, the packaged or unpacked products are placed on conveyor belts, on spool bars and in tunnels and then passed through chamber with lamps (Proctor, 2011). An in-line treatment system is possible with such an assembly. Experiments with continuous pulsed light have been performed. These were for milk decontamination (Krishnamurthy *et al*., 2007) and for various fruit juices (Palgan *et al*., 2011; Pataro *et al*., 2011).

For all existing pulsed-light systems, a control system is used to automate the process and control the rate of pulsing. Optical sensors can be installed to record the output of the entire unit. The newest generation of SteriPulseTM - XLR systems sold by Xenon Corp. are equipped with a LiteMark light monitor. This system contains a photoelectric detector module installed in the treatment chamber that senses the light intensity from each flash, which is scattered sideways in the lamp housing window, and relates it to the side-scattered intensity produced by a new lamp. This enables the operator to monitor in real time the performance of a lamp system and to make decisions regarding the lamp replacements prior to the lamp output reaching a predetermined minimal level.

4. Applications

4.1 Pulsed light treatment given to eggs for surface decontamination

Eggs and egg-based products were frequently associated with salmonellosis outbreaks caused by Salmonella Enteritidis in the United States of America (U.S.A.), as well as in the European Union (E.U.) (Braden, 2006; EFSA, 2007). This is a potential consequence of the high frequency at which Salmonella Enteritidis colonizes the ovaries of laying hens (Gantois *et al*., 2008)

Eggs were treated with pulsed light of flashes of 2.1 Joule/cm2 and 10.5 Joule/cm2. Exposure to 2.1 Joule/cm2 leads to death of Salmonella cells (5 log colony-forming units (CFU) per egg shell) on the egg surface with slight increase in temperature. Increase to 10.5 Joule/cm2 did not cause penetration of Salmonella cells to the egg contents from the shell. No adverse effect on quality of egg albumin was observed. No effect on sensory and functional properties (Lasagabster *et al*., 2011). Another study for inactivation of Salmonella enteritidis was performed that involved usage of pulsed light treatment of 0.5 Joule/cm2 to 0.7 Joule/cm2. Treatment of 0.5 Joule/cm2 gave an inactivation of 6.7 log colony-forming units (CFU)/cm2 on noble agar. Different results were obtained based on the state of the cuticle. In case of unwashed eggs, the highest decontamination of 3.6 log colony-forming units (CFU)/egg was observed and for washed eggs, highest decontamination of 1.8 log colony-forming units (CFU)/egg was observed. The integrity of the cuticle is maintained and hence this technique could be used in egg processing (Hierro *et al*., 2009).

4.2 Shelf-life extension and inactivation of Listeria monocytogenes on ready to eat cooked meat products using pulsed light

Listeria monocytogenes is responsible for severe foodborne disease outbreak. Processed meats are well documented to be a potential vehicle for human listeriosis. Vacuum packaged ham and bologna slices were artificially inoculated with Listeria monocytogenes and then treated with pulsed light with fluences of 0.7, 2.1, 4.2 and 8.4 Joule/cm2. It was found to reduce the microbial load of ham by 1.78 colony-forming units (CFU)/cm2 and of bologna slices by 1.11 colony-forming units (CFU)/cm2 (Hierro *et al*., 2011). The lower inactivation obtained on bologna could be explained by the distinct microstructural features of both products. It is well known that the surface topography greatly influences the efficacy of pulsed light treatment (Woodling and Moraru, 2005). It tripled the shelf-life of ham as compared to conventional ready-to-eat (RTE) products. 2.1 Joule/cm2 adversely affected the sensory quality of bologna slices (Hierro *et al*., 2011).

4.3 Pulsed light treatment for decontamination of chicken from food pathogens

High-power pulsed light of 1,000 pulses, treatment duration 200 seconds and total ultraviolet light dose 5.4

Joule/cm2 was found to reduce viability of Salmonella typhimurium and Listeria monocytogenes inoculated on the surface of chicken by 2-2.4 log10 (N/N0) colony-forming units (CFU)/ml (Paskeviciute *et al*., 2011). Also, the total aerobic mesophiles on the surface of meat were diminished by 2 log10 (N/N0) colony-forming units (CFU)/milliliter. Nonthermal conditions were maintained throughout (<42oC). The intensity of lipid peroxidation in control and treated chicken samples differed in 0.16 milligram (mg) malondialdehyde per kilogram of chicken meat. Organoleptic properties of treated chicken did not detect any changes of raw chicken, chicken broth or cooked chicken meat when it was treated under nonthermal conditions in comparison with control (Paskeviciute *et al*., 2011).

4.4 Pulsed light treatment for freshly cut mushroom

Pulsed light treatment for freshly cut mushroom Fresh slices of mushrooms were exposed to pulsed light treatment by flashing at 4.8, 12 and 28 Joules/cm2 and it was found to increase the shelf life by 2-3 days in comparison to untreated samples. The native microflora reduction ranged from 0.6-2.2 log after 15 days of refrigeration. 12 and 28 Joule/cm-2 treatment affected the texture due to thermal damage by treatment. It induced enzymatic browning due to increase in polyphenoloxidase activity. Some phenolic compounds and vitamin C content were found to be reduced. But 4.8 Joule/cm2 increased shelf-life without affecting the texture and antioxidant properties (Oms-Oliu *et al*., 2010).

4.5 Continuous flow pulsed light system for bacterial inactivation in fruit juices and milk

Apple juice (pH of 3.49) and orange juice (pH of 3.78) were inoculated with gram positive (Listeria innocua 11288) and gram negative (Escherichia coli DH5-α) bacteria. These were then subjected to continuous pulsed light system. Xenon-flash lamp emitting light in the range of 100-110 nanometre (nm) and with the flashes at constant frequency of 3 Heartz and lasting for 360 microseconds (μs) was used. It was concluded that the lethal effect of pulsed light processing depends on the type of microorganism and the absorption properties of the liquid food. With treatment of 4 Joule/cm2, the microbial load reductions in apple and orange juices for Escherichia coli were 4.00 and 2.90 Log-cycle respectively and for Listeria innocua were 2.98 and 0.93 Log-cycles respectively (Pataro *et al*., 2011).

Continuous flow pulsed light technique was also used for inactivation of Staphylococcus aureus in milk and has a potential in treatment of milk pathogens (Krishnamurthy *et al*., 2007) carried out measurements of the temperature increase during the pulsed light treatment of milk in a continuous flow system. Milk temperature increased up to 38°C, depending on the residence time as well as the distance of the product from the light source. This temperature increase caused a fouling effect as well as possible changes in milk quality.

4.6 Decontamination of food powders using pulsed ultraviolet (UV) light

Food powders were decontaminated using pulsed ultraviolet (UV) light and the treatment parameters were optimized. 58 Joule/cm2 of pulsed light was required for Saccharomyces cerevisiae decontamination and reducing the microbial load by 7 log. It was found that the thermal effect rather than the Ultraviolet (UV) effect of pulsed light worked for decontamination of coloured powders (Fine and Gervais, 2004).

4.7 Decontamination of packaging material

Paper-polyethylene was artificially inoculated with spores such as Cladosporium herbarum, Aspergillus niger, Aspergillus repens and Aspergillus cinnamomeus and then exposed to pulsed light with fluence ranging from 0.244 to 0.977 Joule/cm2. The highest level of inactivation of 2.7 log reduction was achieved. The colour of the spores affected their resistance to pulsed light. Different spores required different fluences for their inactivation (Turtoi and Nicolau, 2007).

4.8 Application on food processing equipment

Pulsed ultraviolet (UV) light treatment was studied for its applicability in decontamination of the stainless steel surface contacting meat from Listeria monocytogenes and Escherichia coli O157:H7. A four lamp batch scale apparatus which generated 3 Joule/cm2 with an input voltage of 3000 Volts was used. The study was performed on stainless steel slicing knife. The type of meat product in contact with the treatment surface and the time between contamination and intense pulse treatment decide the effectiveness of the microbial inactivation. When the knife surface was in contact with meat product containing lower fat and protein content and the time between contamination and treatment was 60 seconds, highest effectiveness of inactivation of 6.5 log colony forming units (CFU)/side of knife was achieved. It was also observed that even though the number of flashes was increased to compensate for the extended time between contamination and treatment, the lost effectiveness of microbial inactivation could not be restored (Rajkovic *et al*., 2010).

4.9 Effects of PL on foods of vegetable origin

The inactivation of microorganisms naturally present on vegetable surfaces by PL has been demonstrated by Hoornstra, de Jong, and Notermans (2002). The authors treated five vegetables, namely white cabbage, leek, paprika, carrots and kale with two pulses of wide spectrum PL that amounted to a fluence of 0.30 J/cm2. The reduction in aerobic count at the surface of the vegetables varied from 1.6 log CFU/cm2 for carrots to >2.6 log CFU/cm2 for paprika. No significant increase in the inactivation was observed after using a third pulse. Moreover, no adverse effects on sensorial quality were observed after treating the vegetables with three pulses

and storage at 7 and 20 0C for up to 7 days with one exception, iceberg lettuce showed some discoloration after 48 h at a storage temperature of 20 0C. Their own calculations demonstrated that a reduction of 2 log CFU/cm2 increases the shelf-life at 7 0C of cut vegetables by about 4 days, which is remarkable given a treatment of only 0.4 ms effective duration. The efficacy of PL to inactivate vegetable spoilage microflora was again proved afterwards. Mesophilic aerobic counts were reduced between 0.56 and 2.04 log CFU/g after treating with more than 2000 pulses several minimally processed vegetables (spinach, radicchio, lettuce, cabbage, carrot, green bell pepper) and soybean sprouts (Go´mez- Lo´pez *et al*., 2005b). Counts of S. cerevisiae cells inoculated onto carrot slices were reduced by more than 4 log CFU/g after 24 pulses.

The inactivation of microorganisms present on food surfaces does not necessarily result in an extension of shelf-life. PL treatment failed to prolong the shelf-life of shredded Iceberg lettuce and shredded white cabbage, stored under modified atmosphere packaging at 7 0C. For Iceberg lettuce, a 0.46 log reduction was achieved in psychrotroph counts. However, no-flashed and flashed samples were rejected at the third day of storage due to excessively high psychrotroph counts and bad sensory quality. For white cabbage, a 0.54 log reduction was achieved in psychrotroph counts. But after 2 days of storage, no-flashed and flashed samples had the same psychrotroph counts; therefore, the benefit of the decontamination was readily lost (Go´mez-Lo´pez *et al*., 2005b). It should be mentioned that the failure in shelf-life prolongation does not necessarily imply the inefficacy of PL, but can also be a consequence of the storage conditions.

Regarding fruits, Marquenie, Geeraerd, *et al*. (2003) reported a maximal inactivation of 3 and 4 log units for conidia of B. cinerea and M. fructigena in vitro, which are fungi responsible for important economical losses during postharvest storage and transport of strawberries and sweet cherries. PL however had no effect on strawberries or on the development of B. cinerea inoculated on strawberries. The treatment did not induce resistance against fungal infection (Marquenie, Michiels, *et al*., 2003). This could be due to insufficient dose or inadequate exposure. PL has also been tested to decontaminate food powders and seeds. Jun *et al*. (2003) reported a 4.96 log reduction of A. niger spores inoculated in corn meal, but Fine and Gervais (2004) achieved less than 1 log reduction in S. cerevisiae cells inoculated onto wheat flour and black peppers. For seeds, Sharma and Demirci (2003) showed >4 log reduction in E. coli O157:H7 inoculated on alfalfa seeds. The colour of foods can be modified by PL. Fine and Gervais (2004) compared the effects of light pulses on the inactivation of S. cerevisiae on black pepper and wheat flour. Colorimetric results indicated a rapid modification of product colour well before the decontamination threshold was reached and this was clearly more rapid for black pepper than for wheat flour. This colour modification was attributed to overheating combined with oxidation. The difference in colour modification between wheat flour and black peppers was explained by the difference in initial colour, dark products absorb more light energy than pale products. On the other hand, Lammertyn *et al*. (2003) found that PL treatment did not significantly reduce strawberry sepal quality decay rate during storage for 10 days at 12 0C. PL can affect the enzymatic activity of flashed vegetables as well as cause physiological changes also observed due to application of CW UV. Since proteins have a strong absorption at about 280 nm (Hollo´sy, 2002), broadspectrum pulsed light could be useful to inactivate enzymes and as such inhibit adverse effects due to enzymatic activity. Dunn *et al*. (1989) claimed that by using two to five flashes of light at a fluence of 3 J/cm2, it is possible to inhibit potato slice browning. The polyphenol oxidase extract recovered from the treated slices exhibited less activity than that from the untreated slices. PL can also increase the respiration rate of vegetables. Go´mez-Lo´pez *et al*. (2005b) reported an 80% increase in the respiration rate of shredded Iceberg lettuce, but no significant effects on the respiration rate of shredded white cabbage treated under the same conditions.

4.10 Continuous flow pulsed light system for bacterial inactivation in fruit juices and milk

Apple juice (pH of 3.49) and orange juice (pH of 3.78) were inoculated with gram positive (Listeria innocua 11288) and gram negative (Escherichia coli DH5-α) bacteria. These were then subjected to continuous pulsed light system. Xenon-flash lamp emitting light in the range of 100-110 nanometre (nm) and with the flashes at constant frequency of 3 Heartz and lasting for 360 microseconds (μs) was used. It was concluded that the lethal effect of pulsed light processing depends on the type of microorganism and the absorption properties of the liquid food. With treatment of 4 Joule/cm2, the microbial load reductions in apple and orange juices for Escherichia coli were 4.00 and 2.90 Log-cycle respectively and for Listeria innocua were 2.98 and 0.93 Log-cycles respectively (Pataro *et al*., 2011).

Continuous flow pulsed light technique was also used for inactivation of Staphylococcus aureus in milk and has a potential in treatment of milk pathogens (Krishnamurthy *et al*., 2007) carried out measurements of the temperature increase during the pulsed light treatment of milk in a continuous flow system. Milk temperature increased up to 38°C, depending on the residence time as well as the distance of the product from the light source. This temperature increase caused a fouling effect as well as possible changes in milk quality.

4.11 Decontamination of other foods

Besides its application on foods of vegetable origin, PL has been tested for efficacy to inactivate microorganisms in a very limited number of other foods. As much as 0.97 log reduction was obtained for spores of Clostridium sporogenes inoculated in honey. Also in this case, the longer the treatment time and the shorter the distance between sample and lamp, the higher the inactivation, but also the higher the temperature. When the depth of the honey was decreased the spore kill increased. It appeared

that the UV light has a limited penetration in the honey. The heat generated does not appear to have a synergistic effect on the inactivation of C. sporogenes in honey (Hillegas & Demirci, 2003). The inactivation of E. coli O157:H7 and L. monocytogenes inoculated on raw salmon fillets by pulsed UV light was studied by Ozer and Demirci (2006), using a broadspectrum lamp that generated 5.6 J/cm2 per pulse at the lamp surface and three pulses per second. Two different surfaces were treated, the skin side and the muscle side. For E. coli O157:H7, the maximum log reduction was 1.09 on the muscle side and 0.86 on the skin side, while for L. monocytogenes, the maximum log reduction was 0.74 on the muscle side and 1.02 on the skin side. Some fish fillets were overheated at shorter distances and longer treatment times. The higher inactivation on the skin side was attributed by the authors to a higher temperature in comparison with the muscle side.

CW UV light has been used for meat treatments, but oxidation reactions (notably of lipids) resulted in significant changes. PL systems should effectively limit oxidation reactions because of the short pulse duration and the halflife of p-bonds, which prevent efficient coupling with dissolved or free oxygen (Fine & Gervais, 2004). Other applications are described in the US patent of Dunn *et al*. (1989), such as disinfection of curds of dry cottage cheese, hard crusted white bread rolls, cake, packaging materials, and shrimps.

4.12 Mitigation of allergen using pulsed ultraviolet light

Peanut allergy is a severe Immunoglobin E mediated reactions with food. Peanut allergy can be prevented by complete avoidance. But pulsed ultraviolet light treatment of peanut extracts and peanut butter showed to deactivate Ara h 2, the most potent allergic protein present in peanut. Protein band intensity for Ara h1, Ara h2, Ara h3 reduced at energy levels ranging from 111.6 – 223.2 Joule/cm2 (Yang *et al*., 2011).

Pulsed ultraviolet light treatment of soy extracts has found to decrease the levels of soy allergens (glycinin, β-conglycinin). But clinical data are needed for development of products from such treated soy extracts (Yang *et al*., 2010).

5. Pulsed light field technology in combination with other non-thermal processing technologies

Pulsed light technology in combination with other non-thermal processing technologies was experimented on a blend of apple and cranberry juice and the efficacy of the combination of technologies was determined on the basis of quality attributes such as odour and flavour. The non-thermal technologies studied were, ultra-violet light (5.3 Joule/cm2), high intensity pulsed light (3.3 Joule/cm2), pulsed electric field processing (34 kilovolt/cm, 18 Hertz, 93 microsecond) and manothermosonication (5 bar, 43oC, 750 Watt, 20 kilohertz). A blend of apple and cranberry juice in the ratio of 90:10 (volume/volume) was taken and stored at -20oC pre- and post processing. The juice was filtered through 425 micrometre (μm) steel sieve and then processed. The above mentioned processes were paired, their combinations were analysed. A light based technology (ultra violet or high intensity light pulses) in combination with pulsed electric field or manothermosonication was applied. It was concluded that ultraviolet and pulsed electric field combination or high intensity light pulses and pulsed electric field combination was found to maintain product quality better than any combination with manothermosonication under the applied conditions which lead to adverse effects on product quality (Caminiti *et al*., 2011a). High intensity light pulses in combination with pulsed electric field were used to inactivate Escherichia coli in apple juice. The optimum combination was obtained and sensory analysis was performed as well for quality effects. The optimum combination did not affect the quality (Caminiti *et al*., 2011b).

High intensity light pulses in combination with thermosonication were experimented for inactivation of Escherichia coli in orange juice could be developed as hurdle technology. Individual as well as combination of the techniques were studied and inactivation from 2.5 to 3.93 log colony forming units (cfu) ml-1 was achieved (Munoz *et al*., 2011). Using naturally occurring antimicrobial substances in combination with the novel techniques of processing such as pulsed light processing can provide new avenues in controlling pathogenic bacteria and thus improve safety and quality of food (Galvez *et al*., 2010).

6. Advantages and disadvantages

The intensity of light, that lasts for only a second, is 20,000 times brighter than sunlight, but there is no thermal effect, so quality and nutrient content are retained (Brown, 2008). The xenon-flash lamps used in pulsed light treatment are more eco-friendly than the mercury vapour lamps used in ultraviolet (UV) treatment (Gomez-Lopez *et al*., 2007). Pulsed white light is not strictly a non-thermal, but the thermal action, due to its very short duration, it doesn't show much adverse effect on the nutrients (Ohlsson and Bengtsson, 2002).

A possible problem of this preservation method is that folds or fissures in the food may protect microbes from being exposed to the pulsed light (Brown, 2008). There might be some strains of micro-organisms which are resistant to the pulsed light treatment, for example Listeria monocytogenes (Caminiti *et al*., 2011a). This technique for decontamination of micro-organisms is useful mostly in case of liquid foods and surface of solid foods and hence limiting its application.

The short pulse width and high doses of the pulsed UV source may provide some practical advantages over CW UV sources in those situations where rapid disinfection is required (Wang *et al*., 2005). For example, Rice and Ewell (2001), in the aforementioned experiment, needed 3 h to deliver 104 J/m2 using a CW UV lamp and 40 s to deliver the same total fluence using a laser with a repetition rate of 10 Hz. Other advantages of PL treatment are the lack of residual compounds, and the absence of applying

chemicals that can cause ecological problems and/or are potentially harmful to humans. Xenon flash lamps are also more environment friendly than CW UV lamps because they do not use mercury. Sample heating is perhaps the most important limiting factor of PL for practical applications. Heat can originate from the absorption of light by the food or by lamp heating. When studying the inactivation of A. niger spores on corn meal, Jun, Irudayaraj, Demirci, and Geiser (2003) found that some experimental factor settings resulted in sample temperatures of 120 _C, which might have been due to the large amount of heat generated by the lamps, even though their device had a cooling system. Heating also limited the treatment of alfalfa seeds (Sharma & Demirci, 2003), grated carrots (Go´mez-Lo´pez, Devlieghere, Bonduelle, & Debevere, 2005b), and raw salmon fillets (Ozer & Demirci, 2006). It was also demonstrated to occur in honey (Hillegas & Demirci, 2003) and agar (Go´mez-Lo´pez *et al*., 2005a). Another disadvantage of PL treatments is the possibility of shadowing occurring when microorganisms readily absorb the rays, as in the case of A. niger, and are present one upon another. This makes the organisms in the lower layers very hard to destroy in contrast to those in the upper layer (Hiramoto, 1984), although the use of relatively high peak powers can overcome the shadowing effect. In order for a PL treatment to inactivate microorganisms, contact between photons and microorganisms should occur. Therefore, any body between the light source and the microorganism that absorbs light will impair the disinfection process. This restriction is different when flashing solid foods, and when flashing fluid foods or microorganism suspensions.

For the decontamination of solid foods, the situation can be divided into three cases. The first and most important case is that food components absorb light. Therefore, opaque solid foods can only be decontaminated superficially. The most important implication of this fact is a food safety concern. It has been demonstrated that pathogenic microorganisms can be internalised in produce tissues (Beuchat, 2006). PL cannot inactivate those microorganisms because the light will be absorbed at the surface, and the more opaque and thicker the food item, the lower the inactivation below the surface. This drawback should not be overestimated since the superficial character of PL treatment is also common with washing solutions such as chlorinated water and its substitutes, applied to decontaminate raw fruits and vegetables.

The superficial absorption of light should be regarded in view of how deep the light can penetrate into the food, i.e. the superficial character of the PL decontamination process should not be considered limited to an infinitesimal superficial layer of the food because some degree of inactivation can occur below it. The inactivation of microorganisms below the surface was demonstrated and modelled by Gardner and Shama (2000) for CW UV. Although it has been claimed that PL has a big penetration power, independent experiments have not been reported. The second case is that the entire surface of the food piece should be flashed in order to achieve the decontamination of its whole surface, where irregularities of the food surface complicate the achievement of the goal. The other case is that food pieces can shadow each other when treated together. Both cases require engineering solutions that need sometimes equipment with radically new designs (Gardner & Shama, 2000). Regarding fluid foods and microorganism suspensions, the liquid will absorb light depending on its absorption coefficient and depth. The challenge consists in promoting the flow of the fluid in an adequate way to drive microorganisms close to the light source in order to achieve a uniform exposure. Coping with this problem also requires engineering solutions, and a possibility has been offered by Forney, Pierson, and Giorges (2005) for CW UV applications. PL is safe to apply but some precautions have to be taken to avoid exposure of workers to light and to evacuate the ozone generated by the shorter UV wavelengths.

7 .Nutritional and toxicological aspects

Neither the effect of PL on nutritional components of vegetables nor the potential formation of toxic by-products has been studied yet. Since the wavelengths used for PL are too long to cause ionisation of small molecules and are in the nonionising portion of the electromagnetic spectrum (Dunn *et al*., 1995), the formation of radioactive by-products is not expected.

PL treatment of foods has been approved by the FDA (1996) under the code 21CFR179.41. According to Dunn *et al*. (1997), in assessing the safety of foods treated with all forms of radiation, the agency considers changes in chemical composition of the food that may be induced by the proposed treatment, including any potential changes in nutrient levels. The legal status of PL in the European Union has a different approach, since the legislation is not technology oriented but food and food ingredient oriented. This technology would fall in the scope of regulation 258/97 on novel foods and novel food ingredients, article 1, item f (European Union, 1997). Among other categories, this legislation applies to foods and food ingredients to which a production process not currently used has been applied, and evaluates possible changes in nutritional value, metabolism and level of undesirable substances. The EU therefore would not approve PL technology as such, but specific foods and food ingredients treated with PL.

It has been proved that CW UV treatment can increase the concentration of phytochemicals in fruits. Cantos, Espı´n, and Toma´s-Barbera´n (2001) found that UV treatment can increase by more than 10-fold the levels of resveratrol in grapes. Given the similarities between both techniques, PL might also have the same effect.

8. Research Needs

- Identification of critical process factors and their effect on microbial inactivation.
- Suitability of the technology for solid foods and non-clear liquids where penetration depth is critical.
- Potential formation of unpalatable and toxic by-products.
- Resistance of common pathogens or surrogate organisms to pulsed light treatments.

- Differences between this technology and that of the more conventional UV (254 nm) light treatment.
- Mechanisms of microbial inactivation to determine whether they are significantly different from those proposed for UV light.
- Understanding of the mechanism and quantification of the benefit attributed to the pulse effect

Conclusions

The use of Pulse Light can be a boom for food processing and preservation knowing the complete physical and bio-chemical aspects of its complete system and operation and its effects.

Acknowledgments

We are appreciative of the SHIATS University for its continuous support in the development of important technologies for the future use. The effort of higher authorities to promote the technologies has been very valuable in the promotion of new technologies. A special thanks goes to the dean and head of department for believing in our dream to develop new technologies.

References

Anderson, J. G., Rowan, N. J., MacGregor, S. J., Fouracre, R. A., & Farish, O. (2000). Inactivation of food-borne enteropathogenicbacteria and spoilage fungi using pulsed-light. IEEE Transactions on Plasma Science, 28(1), 83–88. doi:10.1109/27.842870.

Baka, M., Mercier, J., Corcruff, R., Castaigne, F., & Arul, J. (1999). Photochemical treatment to improve storability of fresh strawberries. Journal of Food Science, 64, 1068–1072. doi:10.1111/ j.1365-2621.1999.tb12284.x.

Bialka, K. I., Demirci, A., & Purl, V. M. (2008). Modelling the inactivation of Escherichia coli O157:H7 and Salmonella entericaon raspberries and strawberries resulting form exposure to ozone or pulsed UV-light. Journal of Food Engineering, 85(3), 444–449. doi:10.1016/j.jfoodeng.2007.08.007.

Bintsis, T., Litopoulou-Tzanetaki, E., & Robinson, R. (2000). Existing and potential applications of ultraviolet light in the food industry— A critical review. Journal of the Science of Food and Agriculture, 80, 637–645.

Cantos, E., Espin, J. C., & Tomas-Barberan, F. A. (2002). Postharvest stilbene enrichment of red and white table grape varieties using UVC irradiation pulses. Journal of Agricultural and Food Chemistry, 50, 6322–6329. doi:10.1021/jf020562x.

Chung, S. Y., Yang, W., & Krishnamurthy, K. (2008). Effects of pulsed UV-light on peanut allergens in extracts and liquid peanut butter. Journal of Food Science, 73(5), 400–404. doi:10.1111/ j.1750-3841.2008.00784.x.

Demirci, A. (2002). Novel processing technologies for food safety. Association of Food Drug Officials, 64(4), 1–8.

Dhallewin, G., Schirra, M., Manueddu, E., Piga, A., & Ben-Yehoshua, S. (1999). Scoparone and scopoletin accumulation and ultraviolet- C induced resistance to postharvest decay in oranges as influenced by harvest date. Journal of the American Society for Horticultural Science, 124, 702–707.

McDonald, K. F., Curry, R. D., & Hancock, P. J. (2002). Comparison of pulsed and CW ultraviolet light sources to inactivate bacterial spores on surfaces. IEEE Transactions on Plasma Science, 30, 1986e1989.

Mitchell, D. L., Jen, J., & Cleaver, J. E. (1992). Sequence specificity of cyclobutane pyrimidine dimers in DNA treated with solar (ultraviolet B) radiation. Nucleic Acids Research, 20, 225e229.

Otaki, M., Okuda, A., Tajima, K., Iwasaki, T., Kinoshita, S., & Ohgaki, S. (2003). Inactivation differences of microorganisms by low pressure UV and pulsed xenon lamps. Water Science and Technology, 47, 185e190.

Ozer, N. P., & Demirci, A. (2006). Inactivation of Escherichia coli O157:H7 and Listeria monocytogenes inoculated on raw salmon fillets by pulsed UV-light treatment. International Journal of Food Science and Technology, 41, 354e360.

Rice, J. K., & Ewell, M. (2001). Examination of peak power dependence in the UV inactivation of bacterial spores. Applied and Environmental Microbiology, 67, 5830e5832.

Roberts, P., & Hope, A. (2003). Virus inactivation by high intensity broad spectrum pulsed light. Journal of Virological Methods, 110, 61e65.

Rowan, N. J., MacGregor, S. J., Anderson, J. G., Fouracre, R. A., McIlvaney, L., & Farish, O. (1999). Pulsed-light inactivation of food-related microorganisms. Applied and Environmental Microbiology, 65, 1312e1315.

Ryer, A. D. (1997). The light measurement handbook. Newburyport, USA: International Light, Inc. Setlow, P. (1992). I will survive: protecting and repairing spore DNA. Journal of Bacteriology, 174, 2737e2741.

Sharma, R. R., & Demirci, A. (2003). Inactivation of Escherichia coli O157:H7 on inoculated alfalfa seeds with pulsed ultraviolet light and response surface modeling. Journal of Food Science, 68, 1448e1453.

Slieman, T. A., & Nicholson, W. L. (2000). Artificial and solar UV radiation induces strand breaks and cyclobutane dimers in Bacillus subtilis spore DNA. Applied and Environmental Microbiology, 66, 199e205.

Sommer, R., Haider, T., Cabaj, A., Heidenreich, E., & Kundi, M (1996). Increased inactivation of Saccharomyces cerevisiae by protraction of UV irradiation. Applied and Environmental Microbiology, 62, 1977e1983.

Takeshita, K., Shibato, J., Sameshima, T., Fukunaga, S., Isobe, S., Arihara, K., *et al*. (2003). Damage of yeasts induced by pulsed light irradiation. International Journal of FoodMicrobiology, 85, 151e158.

Takeshita, K., Shibato, J., Sameshima, T., Fukunaga, S., Isobe, S. and Arihara, K. 2003. Damage of yeast cells induced by pulsed light irradiation. International Journal of Food Microbiology 85 : 151-158.

Turtoi, M. and Nicolau, A. 2007. Intense light pulse treatment as alternative method for mould spores destruction on paper–polyethylene packaging material. Journal of Food Engineering 83: 47-53.

Uysal, P. C. and Kirca, T. A. 2011. Effect of Ultravoilet-C light on anthocyanin content and other quality parameters of pomegranate juice. Journal of Food Composition and Analysis 24 (6) :790-795.

Wekhof, A. 2000. Disinfection with flash lamps. PDA Journal of Pharmaceutical Science and Technology 54: 264-276.

Wekhof, A., Trompeter, F. J. and Franken, O. 2001. Pulse Ultravoilet disintegration (PUVD): a new sterilisation mechanism for packaging and broad medical hospital applications. The first international conference on ultraviolet technologies. Washington D.C. USA.

Woodling, S. E. and Moraru, C. I. 2005. Influence of surface topography on the effectiveness of pulsed light treatment for the inactivation of *Listeria innocua* on stainless-steel surface. Journal of Food Science 70: 345–351.

Wuytack, E. Y., Phuong, L. D. T., Aertsen, A., Reyns, K. M. F., Marquenie, D., Ketelaere, D. B., Masschalck, B., Van Opstal, I., Diels, A. M. J. and Michiels, C. W. 2003. Comparison of

sublethal injury induced in *Salmonella enterica* serovar Typhimurium by heat and by different nonthermal treatments. Journal of Food Protection 66: 31-37.

Xenon Corp. 2005. AN-104 application note. Achieving faster cure time with pulsed ultraviolet, available online at *http://www.globalspec.com*.

Authors' Biography

Ravi Shankar- AMIE, AMIMI, AMIAEI, M.Tech. (4th sem.) Food Process Engineering, Department of Food Process Engineering, Vaugh School of Agriculture Engineering and Technology, Sam HigginBottom Institute of Agriculture, Technology and Sciences, SHIATS -Deemed University P.O Naini, Allahabad, U.P-211007, India B.E in Food Technology, SLIET, Longowal, Sangrur, Punjab, India.

U Kaushik- M.Tech. (4th sem.) Food Process Engineering, Department of Food Process Engineering, Vaugh School of Agriculture Engineering and Technology, Sam HigginBottom Institute of Agriculture, Technology and Sciences, SHIATS-Deemed University

Shayeeb A Bhat- M.Tech. (4th sem.) Food Chain Management, Department of Food Process Engineering, Vaugh School of Agriculture Engineering and Technology, Sam HigginBottom Institute of Agriculture, Technology and Sciences, SHIATS-Deemed University