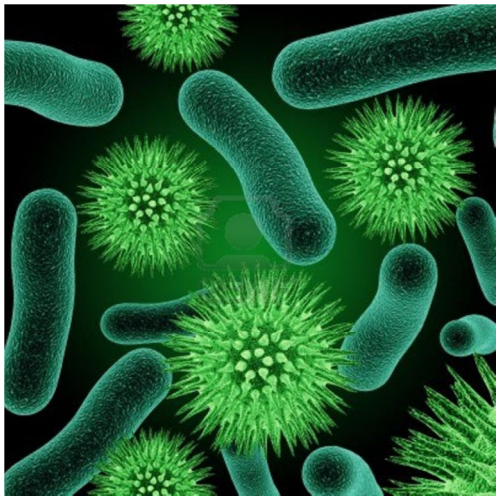


# SCIENTIFIC PAPER ABSTRACTS

## PULSED LIGHT RESEARCH

## Table of Contents

**Introduction** ..... 3

**1-Inactivation of Escherichia coli O157:H7 and Listeria monocytogenes in bio films by pulsed ultraviolet light** ..... 4

**2- Efficacy of Pulsed UV-Light Treatment on Wastewater, Effluent Disinfection and Suspended Solid Reduction** ..... 4

**3-Pulsed-light inactivation of Pathogenic and Spoilage Bacteria on Cheese Surface** ..... 5

**4-Pulsed Light Treatments for Food Preservation. A Review** ..... 5

**5-Pulsed Ultraviolet Light** ..... 5

**6-Milk Pasteurization by Pulsed UV-light Treatment** ..... 6

**7-*Staphylococcus aureus* Inactivation Using Pulsed UV light for Cont. Milk Treatment** ..... 6

**8-Effect of Spectral Range in Surface Inactivation of *Listeria innocua* Using Broad-Spectrum Pulsed Light** ..... 6

**9-Modeling the inactivation of *Escherichia coli* O157:H7 and *Salmonella enterica* on raspberries and strawberries resulting from exposure to ozone or pulsed UV-light** ..... 7

**10-Comparative disinfection efficiency of pulsed and continuous-wave UV irradiation technologies** 7

**11-Effects of Pulsed UV-Light on Peanut Allergens in Extracts and Liquid Peanut Butter** ..... 7

**12-Decontamination of Chicken Frankfurters with Pulsed UV-Light** ..... 8

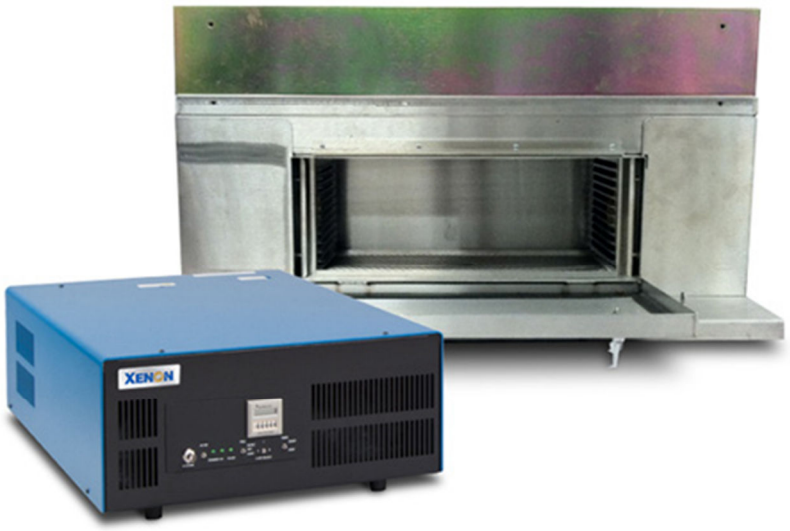
**13-Pulsed UV Light Inactivation of Salmonella Enteritidis on Eggshells and Its Effects on Egg Quality** 8

**14-Effect of Pulsed Light on Safety and Quality of Fresh Egg Pasta** ..... 9

## Introduction

XENON Corporation's model RS-3000C[1] Pulsed Light Sterilization system shown below has been utilized in a range of University research studies investigating the efficacy of pulsed ultraviolet (UV) light as a means of decontaminating food surfaces, water, wastewater, air and food contact surfaces. These scientific studies included a range of microorganisms that impact on human health and safety: bacteria, yeasts, molds, spores and viruses. Included within this range of work was the microbial inactivation on food surfaces such as cheese, strawberries, peanut butter, clover honey, chicken frankfurters, milk, ready-to-eat meats, fruits, vegetables and alpha seeds.

The selected abstracts listed in this paper are taken from published research papers covering a period of 2004 thru 2015. Many of these reports offer details on treatment conditions such as pulse energy, pulse duration, number of pulses, pulse rate and target volume, that control achieving microbial reduction on surfaces in the range of 1 to 6 $\log_{10}$. These studies offer evidence of achieving inactivation of pathogens using high energy pulsed light.




Model RS-3000 Sterilization Lab System

[1] *Model RS-3000C has been renamed model Z-1000.*

## 1-Inactivation of Escherichia coli O157:H7 and Listeria monocytogenes in bio films by pulsed ultraviolet light

Nedra L. Montgomery and Pratik Banerjee. 2015

**Abstract:** The inactivation of biofilms formed by pathogenic bacteria on ready-to-eat and minimally processed fruits and vegetables by nonthermal processing methods is critical to ensure food safety. Pulsed ultraviolet (PUV) light has shown promise in the surface decontamination of liquid, powdered, and solid foods. In this study, the antimicrobial efficacy of PUV light treatment on nascent biofilms formed by *Escherichia coli* O157:H7 and *Listeria monocytogenes* on the surfaces of food packaging materials, such as low-density polyethylene (LDPE), and fresh produce, such as lettuce (*Lactuca sativa*) leaves, was investigated. The formation of biofilms on Romaine lettuce leaves and LDPE films was confirmed by crystal violet and Alcian blue staining methods. Inactivation of cells in the bio lm was determined by standard plating procedures, and by a luminescence-based bacterial cell viability assay. Upon PUV treatment of 10 s at two different light source to sample distances (4.5 and 8.8 cm), viable cell counts of L. monocytogenes and E. coli O157:H7 in biofilms on the lettuce surface were reduced by 0.6–2.2 log CFU mL−1 and 1.1–3.8 log CFU mL−1, respectively. On the LDPE surface, the efficiency of inactivation of biofilm-encased cells was slightly higher. The maximum values for microbial reduction on LDPE were 2.7 log CFU mL−1 and 3.9 log CFU mL−1 for L. monocytogenes and E. coli O157:H7, respectively. Increasing the duration of PUV light exposure resulted in a significant ($P < 0.05$) reduction in biofilm formation by both organisms. The results also revealed that PUV treatment was more effective at reducing *E. coli* biofilms compared with *Listeria* biofilms. A moderate increase in temperature (~7–15°C) was observed for both test materials.

## 2- Efficacy of Pulsed UV-Light Treatment on Wastewater, Effluent Disinfection and Suspended Solid Reduction

Gulsad Uslu; Ali Demirci; and John M Regan. 2015

**Abstract**: Disinfection of municipal wastewater effluents is a critical component of water pollution control. To achieve this, novel alternative disinfection technologies have been getting attention recently such as pulsed ultraviolet (UV) light, which can be used to inactivate microorganisms in a short time. Therefore, the research reported in this paper was undertaken to determine the efficacy of pulsed UV light for inactivation of *Escherichia coli* and *Bacillus subtilis* spores, in synthetic and real municipal wastewater effluent. The results with synthetic municipal wastewater effluent demonstrated that complete inactivation was obtained with an 8-cm sample distance, 30-mL sample volume, and 15-s time combination for *E. coli* [8.53 $\log_{10}$ colony-forming units (CFUs)/mL reduction], whereas a 15-mL sample volume (with the same sample distance and treatment time) was required for *B. subtilis* (7.57 $\log_{10}$ CFU/mL reduction) at total energy dose of 10.9 J/cm$^2$. A response surface model was developed to predict the inactivation for E. coli and B. subtilis spores. Using sterilized real wastewater effluent with the same experimental conditions of an 8-cm distance and 30-mL volume for *E. coli*, and 8-cm distance and 15-mL volume for *B. subtilis*, complete inactivation required the same 15-s treatment for E. coli, but 20-s treatment for *B. subtilis*. In addition, a 2.5, 5, 10, 15, and 25% (volume/volume) *E. coli* or *B. subtilis* inoculum was added to the synthetic municipal wastewater effluent, and treated by pulsed UV light for 5–45 s. Results showed that pulsed UV treatment at the optimum conditions increased suspended solids removal by 26.5 and 21.45%, for *E. coli* and *B. subtilis*, respectively. Overall, the results of the research reported in this paper clearly demonstrated the complete inactivation of vegetative cells or spores in municipal wastewater effluents, and further demonstrated the reduction of suspended solids, suggesting that pulsed UV light has the potential to be used for disinfection of municipal wastewater effluent.

## 3-Pulsed-light inactivation of pathogenic and spoilage bacteria on cheese surface

J. Proulx, L. C. Hsu, B. M. Miller, G. Sullivan, K. Paradis, and C. I. Moraru. 23 May 2013

**Abstract:** Cheese products are susceptible to post processing cross-contamination by bacterial surface contamination during slicing, handling, or packaging, which can lead to food safety issues and significant losses due to spoilage. This study examined the effectiveness of pulsed-light (PL) treatment on the inactivation of the spoilage microorganism *Pseudomonas fluorescens*, the nonenterohemorrhagic *Escherichia coli* ATCC 25922 (nonpathogenic surrogate of Escherichia coli O157:H7), and *Listeria innocua* (nonpathogenic surrogate of Listeria monocytogenes) on cheese surface. The effects of inoculum level and cheese surface topography and the presence of clear polyethylene packaging were evaluated in a full factorial experimental design. The challenge microorganisms were grown to early stationary phase and subsequently diluted to reach initial inoculum levels of either 5 or 7 log cfu/slice. White Cheddar and process cheeses were cut into 2.5 x 5 cm slices, which were spot-inoculated with 100 µL of bacterial suspension. Inoculated cheese samples were exposed to PL doses of 1.02 to 12.29 J/cm2. Recovered survivors were enumerated by standard plate counting or the most probable number technique, as appropriate. The PL treatments were performed in triplicate and data were analyzed using a general linear model. *Listeria innocua* was the least sensitive to PL treatment, with a maximum inactivation level of 3.37 ± 0.2 log, followed by P. fluorescens, with a maximum inactivation of 3.74 ± 0.8 log. *Escherichia coli* was the most sensitive to PL, with a maximum reduction of 5.41 ± 0.1 log. All PL inactivation curves were nonlinear, and inactivation reached a plateau after 3 pulses (3.07 J/cm$^2$). The PL treatments through UV-transparent packaging and without packaging consistently resulted in similar inactivation levels. This study demonstrates that PL has strong potential for decontamination of the cheese surface.

## 4-Pulsed Light Treatments for Food Preservation. A Review

Gemma Oms-Oliu, Olga Martín-Belloso and Robert Soliva-Fortuny. 10 Sept. 2008

**Abstract**: Consumers demand high-quality processed foods with minimal changes in nutritional and sensory properties. Nonthermal methods are considered to keep food quality attributes better than traditional thermal processing. Pulsed light (PL) is an emerging nonthermal technology for decontamination of food surfaces and food packages, consisting of short time high-peak pulses of broad spectrum white light. It is considered an alternative to continuous ultraviolet light treatments for solid and liquid foods. This paper provides a general review of the principles, mechanisms of microbial inactivation, and applications of PL treatments on foods. Critical process parameters that are needed to be optimized for a better efficiency of PL treatments are also discussed. PL has considerable potential to be implemented in the food industry. However, technological problems need to be solved in order to avoid food overheating as well as to achieve better penetration and treatment homogeneity. In addition, a more extensive research is needed to understand how PL affects quality food attributes.

## 5-Pulsed Ultraviolet Light

A. Demirci and L. Panico. 8 August 2008

**Abstract**: Pulsed Ultraviolet (UV)-Light is an emerging processing technology, which has a potential to decontaminate food products. The light generated by pulsed UV lamps consists of a continuum broadband spectrum from deep UV to the infrared, especially rich in UV range below 400 nm, which is germicidal. In pulsed UV-light system, UV-light is pulsed several times per second and each pulse lasts between 100 ns and 2 ms. The pulsed UV-light has a modest energy input which can yield high peak power dissipation. Many researchers have demonstrated the effectiveness of pulsed UV-light on microbial loads on food surfaces. In this paper, various applications of pulsed UV-light treatment of foods found in the literature as well as future research needs will be discussed.

## 6-Milk Pasteurization by Pulsed UV-light Treatment

Kathiravan Krishnamurthy, Ali Demirci and Joseph Irudayaraj. 4 August 2004

**Abstract**: *Staphylococcus aur*eus is a pathogen of concern in milk and milk products. Pulsed UV-light is a novel technology that can be used to inactivate this pathogen in a very short time. Efficacy of pulsed UV-light was investigated for inactivation of *S. aur*eus in milk. A surface response model was used to design the experiments. 12, 30, and 48 ml of cell suspension in milk was treated under pulsed UV-light for 30, 105, and 180 seconds. 0.1 ml of treated and untreated samples was spiral plated on Baird-Parker agar and incubated at 37°C for 24 h. The colonies were then enumerated and $log_{10}$ reduction was calculated. The $log_{10}$ reduction obtained varied from 0.16 to 8.55 $log_{10}$ CFU/ml demonstrating the ability of pulsed UV-light to inactivate *Staphylococcus aureus*. The effect of treatment time, time*distance interaction, and time*volume interaction were found to be significant ($p<0.05$). Maximum $log_{10}$ reductions were obtained for (i) 8 cm sample distance from UV-strobe, 30 ml sample volume, and 180 sec treatment time combination and (ii) 10.5 cm sample distance from UV-strobe, 12 ml sample volume, and 180 sec treatment time combination.

## 7-*Staphylococcus aureus* Inactivation Using Pulsed UV light for Continuous Milk Treatment

Kathiravan Krishnamurthy, Ali Demirci and Joseph M. Irudayaraj. 20 July 2005

**Abstract**: *Staphylococcus aur*eus is a pathogen frequently associated with milk. Pulsed UV-light is a novel technology which can be used for inactivation of this pathogen in milk in a short time. Pulsed UV-light damages DNA of the bacteria by forming thymine dimmers which leads to bacterial death. This study investigated the efficacy of the pulsed UV-light system for continuous milk treatment for inactivation of *S. aureus.* The effect of sample distance from the UV-light source, number of passes, and flow rate were investigated. A response surface method was used for design and analysis of the experiments. Milk was treated at 5, 8, or 11 cm distance from UV-light strobe at 20, 30, or 40 ml/min flow rate and treated up to three times by recirculation of milk to find the effect of number of passes on inactivation of pathogenic microorganisms. $Log_{10}$ reductions varied from 0.55 to 7.26 $log_{10}$ CFU/ml. Complete inactivation was obtained in two cases and in most conditions, growth was not observed following an enrichment procedure. This research demonstrates that pulsed UV-light has a potential to be used for inactivation of *S. aureus* in milk.

## 8-Effect of Spectral Range in Surface Inactivation of *Listeria innocua* Using Broad-Spectrum Pulsed Light

Sarah E. Woodling and Carmen I. Moraru 1 December 2006

**Abstract:** Pulsed light (PL) treatment is an alternative to traditional thermal treatment that has the potential to achieve several log- cycle reductions in the concentration of microorganisms. One issue that is still debated is related to what specifically causes cell death after PL treatments. The main objective of this work was to elucidate which portions of the PL range are responsible for bacterial inactivation. Stainless steel coupons with controlled surface properties were inoculated with a known concentration of *Listeria innocua* in the stationary growth phase and treated with 1 to 12 pulses of light at a pulse rate of 3 pulses per second and a pulse width of 360 μs. The effects of the full spectrum (180 to 1,100 nm) were compared with the effects obtained when only certain regions of UV, visible, and near-infrared light were used. The effectiveness of the treatments was determined in parallel by the standard plate count and most-probable-number techniques. At a fluence of about 6 $J/cm^2$, the full-spectrum PL treatment resulted in a 4.08-log reduction of *L. innocua* on a Mill finish surface, the removal of 200 nm diminished the reduction to only 1.64 log, and total elimination of UV light resulted in no lethal effects on *L. innocua*. Overwhelmingly, the portions of the PL spectrum responsible for bacterial death are the UV-B and UV-C spectral ranges (300 nm), with some death taking place during exposure to UV-A radiation (300 400 nm) and no observable death upon exposure to visible and near-infrared light (400 nm). This work provides additional supporting evidence that cell death in PL treatment is due to exposure to UV light. Additionally,

it was shown that even a minor modification of the light path or the UV light spectrum in PL treatments can have a significant negative impact on the treatment intensity and effectiveness.

## 9-Modeling the inactivation of *Escherichia coli* O157:H7 and *Salmonella enterica* on raspberries and strawberries resulting from exposure to ozone or pulsed UV-light

Katherine L. Bialka, Ali Demirci, and Virendra M. Puri. 17 August 2007

**Abstract:** Inactivation data for *Escherichia coli* O157:H7 and *Salmonella enterica* on raspberries and strawberries resulting from treatment with gaseous ozone, aqueous ozone, or pulsed UV-light were used to construct inactivation models; a log-linear model (based on first-order kinetics) and a Weibull model were developed. Initial analysis indicated that survival curves were non-linear and that the log-linear model failed to accurately estimate the inactivations in most instances. The Weibull model more accurately estimated the inactivation and the concavity exhibited in the survival curves. Validation of the Weibull model produced correlation coefficients of 0.83–0.99 and slopes of 0.76–1.26. The results presented in this study indicated that first-order kinetics are not suitable for the estimation of microbial inactivation on berries treated with ozone or pulsed UV-light, but that the Weibull model can be successfully used to estimate the reductions of *E*. coli O157:H7 and *Salmonella enterica* on raspberries and strawberries.

## 10-Comparative disinfection efficiency of pulsed and continuous-wave UV irradiation technologies

Zuzana Bohrerovaa, Hilla Shemerb, Robert Lantisc, Christopher A. Impellitteri and Karl G. Lindene. 29 March 2008

**Abstract:** Pulsed UV (PUV) is a novel UV irradiation system that is a non-mercury lamp-based alternative to currently used continuous-wave systems for water disinfection. PUV polychromatic irradiation disinfection efficiency was compared to that from continuous wave monochromatic low-pressure (LP) and polychromatic medium-pressure (MP) UV systems, using two types of actinometry (ferrioxalate and iodide–iodate) and an absolute spectral emission method for fluence measurement. All three methods were in good agreement. Once accurate and reliable methods for fluence measurement were established, the inactivation of *Escherichia coli* and pathogen surrogates' phage T4 and T7 were investigated under each technology. Inactivation was significantly faster using PUV irradiation compared to LP or MP UV lamps at equivalent fluence levels. A significant fraction of the enhanced PUV inactivation efficiency was due to wavelengths greater than 295 nm.

## 11-Effects of Pulsed UV-Light on Peanut Allergens in Extracts and Liquid Peanut Butter

S.Y. Chung, W. Yang, and K. Krishnamurthy. 26 March 2008

**Abstract**: Pulsed ultraviolet (PUV) light, a nonthermal technology, was used to treat both the peanut extracts and liquid peanut butter. The objective was to determine if such treatment would lead to a reduction in the allergenic properties of the peanut extract and butter. Peanut samples were PUV treated using a Xenon RS-3000C under the following conditions: 3 pulses/s, 14.6 cm from the central axis of the lamp, 4 min (extract) or 3 min (liquid peanut butter). After the treatment, the peanut samples were centrifuged and the supernatants analyzed by SDS-PAGE and competitive inhibition enzyme-linked immunosorbent assay (ciELISA). For comparison, boiling treatments were also performed. SDS-PAGE showed that while boiling treatment had little effect on the peanut allergens, PUV-light treated samples displayed a reduced solubility or level of peanut allergens (63 kDa). Solubility of another allergen (18 to 20 kDa) was unaffected. Insoluble aggregates formed were responsible for the reduced level of allergens in PUV-light-treated samples. ciELISA showed that untreated samples exhibited an IgE binding 7-fold higher than the PUV-treated samples. It was concluded that PUV light was effective in reducing IgE

binding of peanut extracts and liquid peanut butter. The current study provides an approach to the development of a possibly less allergenic peanut product. However, the reduction in actual allergenicity needs to be confirmed by clinical studies.

## 12-Decontamination of Chicken Frankfurters with Pulsed UV-Light

Nene M. Keklik, Ali Demirci and Virendra M. Puri. 24 June 2009

**Abstract**: The effectiveness of pulsed UV-light on the microbial load and quality of unpackaged and vacuum-packaged chicken frankfurters was investigated. Samples inoculated with *Listeria monocytogenes Scott A* on top surfaces were treated with pulsed UV-light for 5, 15, 30, 45, and 60 seconds at 5, 8, and 13 cm distance from the quartz window in a pulsed UV-light chamber. 2 Log reductions (CFU/cm$^2$) of unpackaged samples were between 0.3 and 1.9 after 5-s treatment at 13 cm (mild) and 60-s treatment at 5 cm (extreme), respectively. Log reductions (CFU/cm2) on vacuum-packaged samples ranged from 0.1 to 1.9 after mild and extreme treatments, respectively. The extent of lipid peroxidation and color were determined by thiobarbituric acid reactive substances (TBARS) test and CIELAB color method, respectively. Lipid peroxidation of samples did not change significantly ($p>0.05$) after mild and moderate (30-s treatment at 8 cm) treatments. Significant differences ($p<0.05$) in color parameters were observed after treatments of both unpackaged and vacuum-packaged samples. Packaging material was also analyzed for elastic modulus as a mechanical property. The elastic modulus did not change significantly ($p>0.05$) after mild treatment. Overall, this study demonstrated that pulsed UV-light has a potential to decontaminate ready-to-eat (RTE) poultry-based food products.

## 13-Pulsed UV Light Inactivation of Salmonella Enteritidis on Eggshells and Its Effects on Egg Quality

Nene M. Keklik, Ali Demirci, Paul H. Patterson, and Virendra M. Puri. 2 Dec 2009

**Abstract:** The majority of Salmonella Enteritidis outbreaks have been related to the consumption of raw or undercooked eggs or egg containing foods. Therefore, the U.S. Department of Agriculture mandates egg washing for all graded eggs by use of a detergent solution and sanitizer. These agencies and the egg industry have been investigating alternative decontamination techniques, which could better serve the public, minimize costs, and benefit both the public and the industry. Pulsed UV light is an emerging technology that is used to inactivate microorganisms quickly. In this study, the effectiveness of pulsed UV light was evaluated for the decontamination of eggshells. Eggs inoculated with Salmonella Enteritidis on the top surface at the equator were treated with pulsed UV light 1 to 30 s, at a distance of 9.5 and 14.5 cm from the UV lamp in a laboratory-scale, pulsed UV light chamber. Three eggs were used per treatment in each repetition, except for quality measurements, which involved six eggs per treatment in each repetition. A maximum log reduction of 5.3 CFU/cm2 was obtained after a 20-s treatment at 9.5 cm below the UV lamp at a total dose of 23.6 $\pm$ 0.1 J/cm2, without any visual damage to the egg. After a 30-s treatment at 9.5 and 14.5 cm, the temperature of eggshell surfaces increased by 16.3 and 13.3uC, respectively. Energy usage increased up to 35.3 $\pm$0.1 and 24.8 $\pm$0.1 J/cm2, after 30-s treatments at 9.5 and 14.5 cm, respectively. The effect of pulsed UV light treatments on egg quality was also evaluated. Pulsed UV-light treatments for 3, 10, and 20 s at either 9.5 or 14.5 cm did not change the albumen height, eggshell strength, or cuticle presence significantly (P. 0.05). This study demonstrated that pulsed UV light has potential to decontaminate eggshell surfaces.

## 14-Effect of Pulsed Light on Safety and Quality of Fresh Egg Pasta

Manzocco, L., Maifreni, M., Anese M., Munari, M., Bartolomeoli, I., Zanardi, S., Suman, M., and Nicoli, M.C., 20 May 2013

**Abstract**: This study investigated the effect of pulsed light (up to 26.25 J/cm$^2$) on the inactivation of Salmonella enterica and on the eventual occurrence of undesirable changes in the quality of fresh egg pasta just after preparation and during storage at 4 °C. When S. enterica was inoculated on egg pasta surface, a light dose of 0.70 J/cm$^2$ sufficed to lower counts by 2.5 log units while 3.50 J/cm$^2$ were required for a 3.3 log unit reduction (below detection limit). For *S. enterica* inoculated in the dough, a light dose of 3.50 J/cm$^2$ lowered counts by only 1.0 $\log_{10}$ unit while 17.50 J/cm$^2$ were required for a 3.3 log unit reduction, due to the limited light penetration through egg pasta. At a dose of 1.75 J/cm$^2$, pulsed light induced no significant changes in egg pasta appearance, oxidation state and sensory properties. At higher doses, off-flavor formation was detected. Independently of the dose applied, pulsed light did not induce furan formation and promoted an increase in the oxidative stability of egg pasta lipids as well as pigment bleaching during storage. The latter was attributed to the formation of photo-induced non-enzymatic browning products.

www.xenoncorp.com
**37 Upton Drive**
**Wilmington, MA 01887-1018**
**USA**
**Telephone: +1 978-661-9033**
**Toll Free (USA): +1 800-936-6695**
**Fax: +1 978-661-9055**



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