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## “Non-thermal techniques: Application in food industries” A review

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#### Abstract

Non-thermal technologies (NTP) are processing methods for achieving microbial inactivation without exposing foods to adverse effects of heat whilst extending product shelf life and retaining their fresh-like physical, nutritional, and sensory qualities. It consist the high hydrostatic pressure, pulsed electric fields, high-intensity ultrasound, ultraviolet light, pulsed light, ionizing radiation and oscillating magnetic fields have the ability to inactivate microorganisms to varying degree. Pulsed Light is also considered an emerging, non-thermal technology capable of reducing the microbial population on the surface of foods and food contact materials by using short and intense pulses of light in the Ultraviolet near Infrared (UV-NIR) range. Ultraviolet (UV) radiation is an established disinfection alternative used to produce drinking water. The benefits of UV treatment in comparison to other methods of disinfection are very clear no chemicals are used; it is a non-heat related process; lesser changes in color, flavor, odor, or pH and no residuals are left in the fluid stream. However, a potential problem of using short-wave UV light is that it can damage human eyes, and prolonged exposure can cause burns and skin cancer in humans which is a concern especially among industry workers. The application of UV light treatment for the decontamination of fresh fruits and vegetables surface. During the last decades, this technology has emerged as an industrially adopted method for food pasteurization of rare and cooked meat, fish and seafood, dairy and vegetable products, and ready-to-eat meals. This is an opportunity for those who would like to take advantage of NTP technologies to ensure safety and quality of food products.

**Keywords:** Non-thermal technique, HPP, UV, PEF, application, advantages

#### Introduction

The novel non-thermal methods may be useful in inactivating foodborne pathogens and spoilage microorganisms from a range of solid and liquid foods Montenegro *et al.*, 2002 [47]; Pereira and Vicente 2009 [54]; Song *et al.*, 2009 [65]; Ekem and Akan 2006 [20]; Purevdorj *et al.*, 2002 [56]; Critzer *et al.*, 2007 [17]; Deng *et al.* [18].

The term ‘Non-thermal processing’ is often used to designate technologies that are effective at ambient or sub lethal temperatures. High hydrostatic pressure, pulsed electric fields, high-intensity ultrasound, ultraviolet light, pulsed light, ionizing radiation and oscillating magnetic fields have the ability to inactivate microorganisms to varying degree. Non-thermal technologies (NTP) are processing methods for achieving microbial inactivation without exposing foods to adverse effects of heat whilst extending product shelf life and retaining their fresh-like physical, nutritional, and sensory qualities. High hydrostatic pressure, pulsed electric fields, high-intensity ultrasound, ultraviolet light, pulsed light, ionizing radiation and oscillating magnetic fields have the ability to inactivate microorganisms to varying degrees (Butz and Tauscher, 2002) [12]. Some of these treatments may involve heat due to the generation of internal energy (e.g. adiabatic heating and resistive heating during HHP and PEF, respectively), however, they are classified as non-thermal once, contrarily to thermal processing technologies, they can eliminate the use of high temperatures to kill the microorganisms, avoiding the deleterious effects of heat on flavor, color and nutritive value of foods. These novel technologies are still struggling with impairments to their full industrial application. For example, irradiation has a high potential and is probably one of the most versatile among the food preservation technologies. However, its development and commercialization has been hampered in the past by unfavorable public perceptions (Resurreccion, Galvez, Fletcher, and Misra, 1995). Ultraviolet (UV) radiation is an established disinfection alternative used to produce drinking water. More than 500 UV plants supplying drinking water operate in North America, and in Europe more than 2000 plants use this technology as a common disinfection technique for drinking water supplies. The benefits of UV treatment in comparison to other methods of disinfection are very clear:

no chemicals are used; it is a non-heat related process; lesser changes in color, flavor, odor, or pH; and no residuals are left in the fluid stream. However, a potential problem of using short-wave UV light is that it can damage human eyes, and prolonged exposure can cause burns and skin cancer in humans (Bintsis, Litopoulou Tzanetaki, and Robinson, 2000; Shama, 1999) <sup>[62]</sup>, which is a concern especially among industry workers. Pulsed Light is also considered an emerging, non-thermal technology capable of reducing the microbial population on the surface of foods and food contact materials by using short and intense pulses of light in the Ultraviolet near Infrared (UV–NIR) range. PL systems have relatively low operation costs and do not significantly contribute negatively to the environmental impact of the processes where it is included because it has the potential to eliminate microorganisms without the need for chemicals. Further, it does not produce volatile organic compounds (VOC) and generates only reduced amounts of solids wastes. However, the poor penetrating power of light (requires transparency and surface smoothness of the product to be treated) and high investment costs (€ 300,000– 800,000) are currently limiting PL applications. The most interesting application of PL systems could be in sterilizing films of packaging material as an alternative to hydrogen peroxide (Palmieri and Cacace, 2005). Efforts have been made to improve non-thermal devices (Forney and Pierson, 2004) with efficient microbial inactivation capabilities, without generating harmful compounds. Technologies which were designed for non-food purposes have now been modified for food applications and positive progress have been reported. Recently, a booming market for nutraceuticals and functional beverages in Asia as well as price cuts for vitamins, minerals and pharmaceutical products in China has led to the increased trends in food fortification (Frost and Sullivan, 2006) <sup>[24]</sup>. This is an opportunity for those who would like to take advantage of NTP technologies to ensure safety and quality of food products. The timing is perfect with the rise in health awareness among consumers. Like any other process, NTP can be combined with thermal or other processes as a hurdle technology or as a compliment to other processes. Combining non-thermal processes with conventional preservation methods enhances their antimicrobial effect so that lower process intensities can be used (Ross *et al.*, 2003) <sup>[59]</sup>. Regardless of the approach, mechanisms and specific targets, these technologies are tools for manufacturers to achieve the same objectives of quality and safety of manufactured food products. It is the purpose of this article to address the opportunities for NTP processing and to describe some of the applications currently researched. Can potential applications to be used within the food industry. Choi and Nielsen (2005) <sup>[13]</sup> demonstrated that UV pasteurized apple cider were superior in colour and overall sensory scores compared to thermally pasteurized apple cider.

Matak (2004) <sup>[43]</sup> studied the efficacy of UV light in inactivating *E. coli* K 12 for different fat percentages of milk at different temperatures (4°C and 20°C) using Cider Sure 3500 apparatus.

### **Kinds of non -thermal processing**

1. Ultraviolet
2. High Pressure Processing (HPP)
3. Pulsed Electrical Fields
4. Cold Plasma
5. Irradiation
6. Supercritical Carbon Dioxide

### **Ultraviolet**

UV irradiation is considered one of the effective means of disinfection, which excludes the necessity of heat to get rid of microorganisms (Sastry *et al.*, 2008). Fruits and vegetables are important components of a healthy and balanced diet. Their sufficient daily consumption could help prevent major diseases such as cardiovascular diseases and certain cancers. The risk involved with the consumption of fresh fruits and vegetables could be minimized either reducing or eliminating external surface contamination. Because simply washing of fresh fruits and vegetables with water may not remove pathogens and other spoilage microorganisms, other alternative processes were researched. UV systems are affordable as they require low initial investment and a lower operating cost of treatment (Yuan *et al.*, 2004). The simple washing of raw fruits and vegetables in hot water or water containing disinfectants removes a portion of the pathogenic and spoilage microorganisms, reductions of 10-fold to 100-fold could sometimes be achieved. Traditional disinfectants (chlorine, chlorine dioxide, bromine, iodine, disodium phosphate, sodium chlorite, sodium hypochlorite, quaternary ammonium compounds, acids, hydrogen peroxide, ozone, permanganate salts etc.) are partially effective in removing pathogens, each type of disinfectant varying in efficiency and in allowable maximum concentration. The efficiency of the treatment will strongly depend on the actual location of the bacterial contaminant as well as the composition, surface topography and transmissivity of the food (Allende *et al.*, 2006) <sup>[3]</sup>. Other attempts in reducing the number of microorganisms on the surface of fresh fruits and vegetables and extending the shelf life were modified atmosphere packaging, low temperature storage and the use of edible films. These treatments are selective in reducing the number of pathogens on the surface of fresh fruits and vegetables. Therefore, the use of non-selective treatments for the destruction of pathogens on the surface of fresh fruits and vegetables would be a better option. Such alternative processes are the irradiation of food and the use of germicidal ultraviolet light (UV-C). The aim of this paper was to review the available literature data and provide a general review of the application of UV light treatment for the decontamination of fresh fruits and vegetables surface.

### **UV light used to control fungal decay**

A number of studies showed that the pre-storage exposure of fruits and vegetables to UV light was effective in reducing the development of postharvest diseases: citrus fruits, kumquat, carrots, apple, strawberry, sweet cherry, mandarin, bell peppers, mango, blueberry, grapes, persimmon fruit. For instance, Baka *et al.*, (1999) <sup>[5]</sup>. Investigated the effect of pre storage exposure to shortwave ultraviolet (UV-C) light on the decay and quality of fresh strawberries. They exposed fresh strawberries to UV-C at doses of 0.25 and 1.0 kJ/m<sup>2</sup>. UV treated fruits were randomly placed in plastic mesh baskets and stored in the dark at 4°C or 13°C. The storehouse atmosphere was maintained at about 95% relative humidity by continuous ventilation with humidified air. The decay caused by *Botrytis cinerea* at both temperatures was controlled through UV-C treatment and the shelf-life of the fruits was extended by 4 to 5 d. UV treated fruits had a lower respiration rate, higher titratable acidity and anthocyanin content, and were firmer than the untreated fruits.

### **Ultraviolet spectrum**

Plants use sunlight for photosynthesis and, as a consequence,

are exposed to the ultraviolet (UV) radiation that is present in sunlight. UV radiation is divided into three segments: UV-A, UV-B and UV-C. The UV-C ( $\lambda = 200\text{-}280\text{ nm}$ ) radiation is absorbed by ozone in the upper and middle parts of atmosphere and, thus, is not present in sunlight at the earth's surface. UV radiation promotes photo-oxidative reactions in plants producing reactive oxygen species (ROS). The major ROS are singlet oxygen, hydrogen peroxide and hydroxyl radicals. The free radicals generated from UV radiation can target cell membranes, nucleic acids, cell walls and enzymes, inducing the acceleration of senescence. The effect of UV-C is directly lethal to microorganisms, hence the term "germicidal". However, the germicidal action of UV light is strongly dependent on the natural resistance to UV-C of the microorganisms. Shama (2005) <sup>[63]</sup> has shown that microorganisms differ greatly in the UV doses required for inactivation. Another important factor of survival is the surface on which microorganisms are attached. Gardner and Shama (2000) have shown that surface "topography" plays a major role in determining survival following exposure to UV-C. Microorganisms present on a surface that may be considered smooth are more susceptible to the effects of UV than the microorganisms present on a surface containing crevices inside which they might be shielded from the lethal effects of UV-C. The germicidal effect occurs over relatively short time that is essentially limited to the time of exposure of the microorganism to the UV source. The exposure times typically range from fractions of a second to perhaps tens of seconds. Ultraviolet light treatment of fruits and vegetables surface During the last two decades, the exposure of horticultural crops to non-ionizing artificial UV-C light (180280 nm with maximum at  $\lambda = 254\text{ nm}$ ) has been considered as an alternative to chemical fungicide in order to control postharvest diseases. Furthermore, researches were shown that UV-C is able to induce resistance of fruits and vegetables to postharvest storage rots and to delay the ripening process extending the shelf life of fruits and vegetables. Moreover, when used at optimum level, UV-C light induces an accumulation of phytoalexins that play an important role in the resistance to disease of many plant systems and activates genes encoding pathogenesis related proteins.

Treatment with UV-C light offers several advantages to food processors as it does not leave any residue in treated food, is easy to use and lethal to most types of microorganisms, and does not require extensive safety equipment to be implemented. However, more research is needed to optimize UV light use.

#### Decontamination of fresh fruits and vegetables with UV light

The use of non-ionizing, germicidal UV-C light could be effective for the decontamination of fruits and vegetables as a whole or as fresh cut products. UV-C affects several physiological processes in plant tissues and damages microbial DNA <sup>[47, 25, p. 69- 71]</sup>. Lado and Yousef (2002) <sup>[36]</sup> reported that UV-C light inhibited microbial growth through a very simple way: radiation generates hydroxyl radicals from water, which remove hydrogen atoms from DNA components, sugar and bases. UV light at 254 nm induces the formation of pyrimidine dimers which alter the DNA helix and block microbial cell replication.

#### High pressure processing

The extensive research on high pressure processing (HPP) has created new opportunities to improve the balance between the

safety and quality of current food products. During the last decades, this technology has emerged as an industrially adopted method for food pasteurization of rare and cooked meat, fish and seafood, dairy and vegetable products, and ready-to-eat meals (Wilson *et al.*, 2008). In this application, HP processing is essentially a non-thermal decontamination process, in which the food is typically subjected to pressures of 400 to 600 MPa at ambient or cooled temperature for 1 to 15 min. These conditions inactivate vegetative microorganisms, providing safety and prolonged shelf life to chill or high-acid (e.g. fruit juices, guacamole) foods (Patterson, 2005). Unfortunately, bacterial spores are extremely resistant to commercially attainable pressure levels, and therefore low-acid shelf stable products cannot be achieved by elevated pressure only (Black *et al.* 2007) <sup>[9]</sup>. To reach commercial sterility, an additional inactivating factor is necessary. Toshiaki Ohshima *et al.* (2004) carried out the study on high pressure processing of fish and fish products. High hydrostatic pressure has recently been applied in food processing, and several commercial fruit and vegetable products have already been put on sale. High hydrostatic pressure results in protein denaturation, resulting in inhibition of some inherent enzymatic activities and of the biogenic activity of some microorganisms. However, high pressure also accelerates lipid oxidation in muscle tissues. Application of pressures in the range of 350-550 MPa has been reported to result in 0.46-3.67 log reduction in *S. enterica* serovar Braenderup inoculated on diced and whole tomatoes (Maitland *et al.*, 2011) <sup>[41]</sup>. At the same time, HPP was reported not to have any significant effect on the quality parameters of orange juice such as titratable acid content, °Brix, viscosity, alcohol insoluble acids, color, ascorbic acid and  $\beta$ -carotene concentrations (Bull *et al.*, 2004) <sup>[11]</sup>.

#### Effects of high pressure processing on food

Cheftel and Culioli, (1997) studied the basic principles underlying the effects of high pressure on food constituents and quality attributes. Extensive investigations in the last decade have revealed the potential benefits of high pressure processing (100–800 MPa) for the preservation and modification of foods. Simultaneously, a few pressurized foods have become commercially available in Japan, Europe and the USA. Recent data concerning the following specific effects of high pressure on muscle and meat products are then reported and discussed: changes in muscle enzymes and meat proteolysis; modifications in muscle ultra-structure; effects on myofibrillar proteins; meat texture and pressure assisted tenderization processes; pressure-induced gelation and restructuring of minced meat; changes in myoglobin and meat colour; influence of pressure on lipid oxidation in muscle; high pressure inactivation of pathogenic and spoilage microorganisms in meat; combined high pressure-moderate temperature 'pasteurization' of meat products.

High-pressure processing (HPP) is a method of food processing where food is subjected to elevated pressures (up to 87,000 pounds per square inch or approximately 600 MPa), with or without the addition of heat, to achieve microbial inactivation or to alter the food attributes in order to achieve consumer-desired qualities. Unlike high-pressure homogenization where the food is exposed to high velocity, turbulence, and shear forces, during HPP the food is subjected to isotactic pressure treatment. High-pressure pasteurization treatments inactivate pathogenic and spoilage bacteria, yeasts, molds, and viruses. However, the treatment has limited efficacy against spores and enzymes. The extent of bacterial

inactivation also depends on the type of microorganism, food composition, pH, and water activity. High pressures at ambient or chilled temperatures have been employed for processing a number of liquid and semi-solid foods such as fruit juices, purees, smoothies, jellies, and guacamole. Several studies have evaluated the beneficial effects of pressure treatment over conventional treatment on preserving quality attributes of foods. However, high capital expenditure and limited throughput are some current limitations of this technology for fluid-food processing. With wider industrial adaptation and technology innovation, it is expected that the cost of equipment will come down.

This led to the development of a number of non-thermal preservation approaches including high pressure processing, wherein a lethal agent (such as pressure, high-voltage electric field, irradiation, ultrasound, etc.) other than heat is primarily used to make the food safe without adversely impacting product quality. Application of high pressure has shown the potential to preserve fresh-like attributes of various value-added liquid foods including juices, smoothies, puree, soups, gravies, etc. Depending on the intensity of pressure heat treatment, both pasteurization and sterilization effects are possible. Studied the preservation of fluid foods by high pressure processing including technology basics process equipment, and microbial efficacy. Combined pressure–heat effects on various food enzymes, product quality, and nutrient attributes were summarized. Daniel *et al.*, (2012) concluded that HP processed avocado paste is commercially available and is stable to the action of spoilage microorganisms during refrigerated storage. However, there are no prior reports on the effects of HP processing and storage time on the stability of health bioactive compounds present in avocados, particularly carotenoid profiles. Results of work indicated that the nutritional and nutraceutical values of avocado carotenoids were retained after commercial HP-processing of the paste and until the end of its sensory shelf-life.

The knowledge generated has scientific relevance for the advancement of non-thermal technologies, and is also considered relevant to consumers and the avocado industry because of the potential impact of the product on human health. HHP processing of avocado paste increased carotenoid content and Carotenoid content decreased during subsequent storage (40 days, 4 °C). Original carotenoid content was retained during product's shelf-life (20 days). Liesbeth Vervoort *et al.*, (2012) reported the comparative study on thermal versus high pressure processing (HPP) of carrots. To allow a fair comparison, the processing conditions were selected based on the principle of equivalence. Moreover, pilot- and industrial-scale equipment was opted for, supporting conditions close to industrial application. The overall impact on carrot quality was characterized by analyzing a wide range of quality attributes, including specific (micro)nutrients (carotenoids and sugars), process-induced contaminants (furfural and 5- hydroxyl methyl furfural), enzyme activities (pectin methyl esterase and peroxidase) and other relevant quality aspects (texture, dry matter content and colour). This study demonstrated that the potential benefit of HP over thermal processing of carrots is largely dependent on the processing intensity applied. Thermal sterilization affected carrot quality the most, while mild and severe thermal pasteurization, mild and severe HP pasteurization and HP sterilization resulted in a comparable overall quality. Industrial relevance: Vervoort *et al.*, (2012) concluded that the extensive nature of their investigation and the corresponding results can be considered of key importance for

further implementation of HP technology in the food industry, since a correct and complete assessment of process-induced changes is of major importance in the context of legislative aspects of novel processing technologies.

### Equipment of HPP

There are two methods of processing foods in high pressure vessels: in-container processing and bulk processing. Because foods reduce in volume at the very high pressures used in processing (for example, water reduces in volume by approximately 15% at 600 MPa), there is considerable stress and distortion to the package and the seal when in container processing is used. It is likely that conventional plastic and foil pouches will prove suitable and research is continuing on the optimum design of the package, seal integrity and other suitable packaging materials. A material handling for in-container processing is achieved using automatic equipment, similar to that used to load/unload batch retorts Bulk handling is simpler, requiring only pumps, pipes and valves. A representation of a commercial processing unit is shown in Fig. 1. Semi continuous processing of fruit juices at 4000-6000 l h<sup>-1</sup> using pressures of 400–500 MPa for 1–5 min at ambient temperature is used by one company in Japan, whereas another uses a similar process operating at 120–400 MPa followed by a short heat treatment before the juice is packaged. The process is highly energy efficient although at present the capital costs of equipment remain high. It is possible that such liquid foods could also be used as the pressurizing fluid by direct pumping with high pressure pumps. Such systems would reduce the capital cost of a pressure vessel and simplify materials handling. If liquids were also rapidly decompressed through a small orifice, the high velocity and turbulent flow would increase the shearing forces on micro-organisms and thus increase their rate of destruction (Earnshaw, 1992) [19].

### Effect of High Pressure processing on micro-organisms

Enzymes and food components Examples of the effect of high pressure treatments on micro-organisms are shown in Table 3. Germination of spores under high pressures is temperature dependent: near 0°C spores resist germination even at pressures of 1000 MPa, whereas at moderate temperatures, pressure induced germination can be achieved at 100 MPa. Germinated spores can be destroyed at a pressure of 600 MPa and a temperature of 50–70°C. However, these effects are not consistent and a combination of high pressure and moderate heating can have either synergistic or antagonistic effects on microbial growth, enzyme activity and chemical reactivity. For example high pressure can either make microorganisms more sensitive to heat or it can prevent their destruction at higher temperatures, depending on the type of micro-organism being studied. As fruit processing has so far been the main application of high pressure technology, many studies of enzyme inactivation are concerned with those enzymes that affect the quality of fruit products. For example polyphenol oxidase has been shown to resist pressures of up to 1200 MPa for 10 minutes before inactivation although it is more sensitive at higher pH levels. report differences in polyphenol oxidase sensitivity from different sources, with potato and mushroom polyphenol oxidase being very pressure stable (800–900 MPa required for inactivation), and more pressure sensitive enzymes from apricot, strawberry and grape (100, 400 and 600 MPa respectively). Pectin esterase is responsible for cloud destabilization in juices, gelation of fruit concentrates and loss of consistency in tomato products. It is

less resistant than polyphenol oxidase; its activity decreases above 300 MPa and it can be inactivated at pressures above 700 MPa at 45°C for 10 minutes. Orange pectin esterase is partially (90%) inactivated at 600 MPa at room temperature and does not reactivate during storage (Fellows, 2009). Reviews of high pressure processing on enzymes and microorganisms have been made by the effects on microbial and enzyme activity.

### Applications of HPP

A potential application of high pressure processing is the tenderization of meat. Processing at 103 MPa and 40–60°C for 2.5 min improves the eating quality of meat and reduces cooking losses. The extent of tenderization depends on all three factors involved: pressure, temperature and holding time. Commercially produced products include pressure-processed salted raw squid and fish sausages (Hayashi, 1995). Other possible applications are improved microbiological safety and elimination of cooked flavours from sterilized meats and pate'. These effects are reviewed by Starch molecules are similarly opened and partially degraded, to produce increased sweetness and susceptibility to amylase activity. Other research has found that the appearance, odour, texture and taste of soybeans and rice did not change after processing, whereas root vegetables, including potato and sweet potato, became softer, more pliable, and sweeter and more transparent. Fruit products are reported to retain the flavour, texture and colour of the fresh fruit. Other applications include tempering chocolate, where the high pressures transform cocoa butter into the stable crystal form, preservation of honey and other viscous liquids, sea foods, dairy products such as unpasteurized milk and mould ripened cheese (Fellows, 2009). When compared to thermal and chemical alternatives, high pressure (HHP) processing is the most effective non-thermal technology.

Daniel *et al.*, (2012) reported the effects of Processing (600 MPa, 3 min) and storage (40 days, 4 °C) on the stability of avocado paste carotenoids. Likewise, the effects of HHP processing and storage on hydrophilic and lipophilic oxygen radical absorbance capacities (ORAC) of the product were studied. Pressurization induced a significant increase (approx. 56%) in concentrations of total extractable carotenoids. Highest increases for individual carotenoids were observed for neoxanthin-b (513%), followed by  $\alpha$ -cryptoxanthin (312%),  $\alpha$ -carotene (284%),  $\beta$ -cryptoxanthin (220%),  $\beta$ -carotene (107%), and lutein (40%). Carotenoid levels declined during storage, but at the end of the product's sensory shelf-life were higher than those initially present in unprocessed avocado paste.

### Pulsed electrical field

Pulsed electric field (PEF) is another non-thermal technology that can be used to inactivate bacterial cells at ambient temperatures. The process involves placing the food material between two electrodes and passing pulses of high electric field (1-50 kV/cm) strengths. Since the pulses are applied for short durations (2 $\mu$ s to 1 ms) the negative impact on food quality due to heat processing is highly diminished (Barbosa-Cánovas *et al.*, 2001). Non-Thermal processing is a major technology that has been commonly used in the food industry to increase shelf life and maintain food safety with low processing costs. Compared with thermal pasteurization, non-thermal processing offers an advantage of low process temperatures which results in a better retention of flavours and nutrients (Vega-Mercado *et al.*, 1996). In recent years,

several technologies have been investigated that have the capability of inactivating microorganisms at lower temperatures than typically used in conventional heat treatments. The increasing consumer interest for high nutritious fresh-like food products, together with the search for environmentally friendly processing technologies, has aided in the development of emerging non-thermal technologies such as pulsed electric fields. Among all emerging non-thermal technologies, high intensity pulsed electric fields (PEF) is one of the most appealing technologies due to its short treatment times and reduced heating effects with respect to other technologies. PEF is commonly understood as a non-thermal food preservation technology that involves the discharge of high voltage electric pulses (up to 70 kV/cm) into the food product, which is placed between two electrodes for a few microseconds (Angersbach *et al.*, 2000). High intensity Pulsed electric field (PEF) processing involves the application of pulses of high voltage (typically 20 - 80 kV/cm) to foods placed between 2 electrodes. PEF treatment is conducted at ambient, sub-ambient, or slightly above ambient temperature for less than 1 s, and energy loss due to heating of foods is minimized. For food quality attributes, PEF technology is considered superior to traditional heat treatment of foods because it avoids or greatly reduces the detrimental changes of the sensory and physical properties of foods. The application of electric field results in cellular death due to generation of pores (electroporation) in the bacterial cell membrane without having an effect on enzymes or proteins present in foods (Wouters *et al.*, 2001) [70].

### Applications of pulsed electric field

PEF treatment has lethal effects on various vegetative bacteria, mold, and yeast. Efficacy of spore inactivation by PEF in combination with heat or other hurdles is a subject of current research. A series of short, high-voltage pulses breaks the cell membranes of vegetative microorganisms in liquid media by expanding existing pores (electroporation) or creating new ones. Pore formation is reversible or irreversible depending on factors such as the electric field intensity, the pulse duration, and number of pulses. The membranes of PEF-treated cells become permeable to small molecules; permeation causes swelling and eventual rupture of the cell membrane.

### Juice Processing

Higher number of pulses and electric field was reported to be a stronger factor for reducing the number of *S. Typhimurium* population in orange juice (Liang *et al.*, 2002) whereas in another study on melon and water melon juices, treatment time was found to be a more important factor (Mosqueda-Melgar *et al.*, 2007). Juice extraction from Chardonnay white grape using pulsed electric field with two pressure conditions was studied. A PEF treatment of 400 V/cm was applied. The PEF pre-treatment increased the juice yield by 67 - 75% compared to the control sample without any treatment (Grimi, 2009). Formation of pores across the membrane often leads to an enhanced extraction process since solvents can penetrate the biomass matrix more efficiently. In this study the influence of PEF treatment on the extraction of lipids from the microalgae *Auxano chlorella protothecoides* was investigated. Microalgae suspensions with a cell density of 50-100 g/l (dry mass) and a lipid content of the algae of 15- 20% (by weight) were treated in a flow cell connected to a transmission line pulse generator. Pulses of 1 $\mu$ s were applied at a constant

specific energy input. After PEF treatment the microalgae were removed immediately from the suspension by centrifugation. The algae-pellet was dried before a solid liquid extraction was performed using ethanol as organic solvent. In order to investigate the spontaneous release of intracellular compounds the supernatant was analyzed using High-Performance Liquid Chromatography (HPLC). Compared to the control samples the Dissolved Organic Carbon Content (DOC) of the supernatant was up to 4 times higher after PEF-treatment. However the HPLC analysis revealed that hardly any high molecular weight compounds and no hydrophobic substances, e.g. lipids, did leak out of the cells. The increase in DOC primarily was due to low molecular weight compounds, presumably monosaccharides and amino acids. The solid-liquid extraction experiments showed that the lipid-recovery from PEF treated microalgae was considerably higher compared to untreated cells. Although the EP did not lead to a spontaneous release of intracellular lipids into the surrounding liquid, it resulted in an improved lipid extraction from dehydrated microalgae.

#### **Aid in air drying**

PEF pre-treated samples showed a reduction of approximately 25 % of drying time compared with control samples. The total specific energy consumption was 3.0 kJ/kg and the temperature increase due to PEF treatment was less than 1°C.

#### **Effect of PEF on osmotic dehydration**

Rastogi *et al.*, (1999) investigated the effect of PEF on the osmotic dehydration of carrot. Carrot discs (2 cm of diameter, 1 cm of thickness) were pre-treated by PEF at different level with exponential pulses, 5 pulses and a total specific energy input in the range of 0,04 to 2,25 kJ/kg. The electric field strength was in the range between 0, 22 to 16 kV/cm and pulse duration between 378 up and 405µs. Both PEF-treated and untreated samples were osmotically dehydrated (immersion in 50° B sucrose solution at 40 °C for 5 h). PEF pre-treated samples showed decreased moisture content as well as an increased solid content during osmotic dehydration.

#### **Plant oil extraction**

Yield and quality of oils from plant origin has been studied by Guderjan *et al.*, (2005) and a modified processing scheme for production of maize germ oil with an increased amount of phytosterols and high oil yield was developed. Wet milled com, steeped for 48 h at 30, 40, and 50°C in water, was treated at field strength of 3.0kV/cm, 120 pulses. Subsequently, oil was separated by hexane extraction, pressing, and supercritical CO<sub>2</sub> extraction. At an electrical field strength of 3.0kV/cm and a steeping water temperature of 50 °C the oil yield could be increased by 27.8% for hexane.

#### **Meat and fish treatment**

The electro per-meabilization of cell membranes, leading to increase in mass transfer rates, can be utilized to enhance drying rates of cellular tissue. An increase in mass transfer rates, resulting in faster water transport to the product surface and therefore reduction of drying time after a pre-treatment will lead to drastic saving of energy and better utilization of production capacities during convective air drying. Taking into account the low energy input required for a PEF treatment of plant or animal tissue (2-20 kJ/kg), it is evident that there is a potential to reduce the total energy input for product drying.

#### **Cold Plasma**

The name plasma was first used who described this state of matter as near the electrodes, where there are sheaths containing very few electrons, the ionized gas contains ions and electrons in about equal numbers so that the resultant space charge is very small. We shall use the name plasma to describe this region containing balanced charges of ions and electrons. Later, the definition was broadened to define a state of matter in which a significant number of atom and/or molecules are electrically charged or ionized.

The pathogenic and spoilage microorganisms are problematic in the food industry due to their significant public health risks and economic impact (Stoica *et al.*, 2011; Afshari and Hosseini, 2014). Plasma is considered as a distinct state of matter due to its properties; it does not have a regular shape or volume and it can form filaments or beams under magnetic fields. Depending on the method of generation used, the plasma can display a broad spectrum of states ranging from extreme non-equilibrium to almost complete thermal equilibrium. Plasma can be found in the form of natural phenomena such as stars and lightning or man-made as in the production of fluorescent and neon lights, plasma television etc. The research areas of plasma technology is fast growing and has been particularly studied for its use on biomedical materials and devices, surface modification of textiles, removal of chemicals on surfaces of devices manufactured from heat sensitive materials, water sterilization and more recently wound healing and food decontamination. The most feasible way to create non-equilibrium plasmas is "pumping" of power selectively to electrons (Bardos and Barankova, 2010). Often the noble gases are employed for inducing plasmas, but this can increase the cost of treatments. Thus, for inducing plasma should be the use of ambient air (Misra *et al.*, 2014). Non-thermal atmospheric Plasma can be divided into two groups depending on the method of generation as non-thermal plasma (NTP), and thermal plasma (TP). NTP consists of gas molecules with moderate temperatures and electrons with higher temperatures whereas in TP the electrons and gas temperatures are several thousands of Kelvin and these species are found in equilibrium. NTP is also known as cold and non-equilibrium plasma with regards to the energy level, temperature and ionic density. An important aspect in the use of NTP for decontamination is the ability to be effective, without affecting the material being decontaminated. This is possible due to the weakly ionized nature of the cold plasma discharge. The antimicrobial efficacy of NTP has been related to the specific type of plasma technology used including; the power level used to generate the plasma, the gas mixture in the plasma emitter, the intensity and length of exposure, design of the system, flow rate and pressure. Cold atmospheric plasma has potential in the food manufacturing sector to inactivate microorganisms, thereby improving food safety. Growing demand for fresh produce poses the challenge to the food industry of supplying safe food with minimal processing. It is crucial that foods are supplied without any microbial contamination as many products are eaten raw. As a result, there is much interest in novel ways of preserving food and destroying microorganisms without affecting its quality. One such emerging technology that has shown promise is the use of cold atmospheric plasma (CAP) treatment. An overview of the cold plasma technology is presented with its potential applications in food processing sector. Plasma is considered as the fourth state of matter. The concept of the fourth state of matter results from the idea that phase transitions occur by

progressively providing energy to the matter, such as the one from the solid state to liquid up to the gas state. A further phase transition may be thought as the one from the gas state to plasma state, even if these states is reached gradually by providing more and more energy to the system. Plasma can be seen as a particular ionized gas, which retains some unique features which distinguish it from an (ideal) gas.

### Plasma science and technology

Plasma is ionized gas that consists of a large number of different species such as electrons, positive and negative ions, free radicals, and gas atoms, molecules in the ground or excited state and quanta of electromagnetic radiation (photons). It is considered to be the fourth state of matter in the world. It can be generated in the large range of temperature and pressure by means of coupling energy to gaseous medium. This energy can be mechanical, thermal, nuclear, radiant or carried by an electric current. These energies dissociate the gaseous molecules into collection of ions, electrons, charge – neutral gas molecules and other species. Depending on the type of energy supply and amount of energy transferred to the plasma, density and temperature of the electrons are changed. These lead Plasma to be distinguished into two groups, high temperature plasma and low temperature plasma. High temperature plasma implies that electron, ions and neutral species are in a thermal equilibrium state. Low temperature plasma is subdivided to thermal plasma, also called local thermodynamic equilibrium plasmas (LTE) and non-thermal plasma (NTP), also called non-local thermodynamic equilibrium plasmas (non-LTE). An equilibrium or near equality between electrons, ions and neutrals is the main characterization of thermal plasmas (TP). Frequently employed thermal plasma generating devices are those produced by plasma torches, and microwave devices. In generation of cold plasma most of the coupled electrical energy is channelled to electron component instead of heating entire gas stream so the temperature of heavy particle remains near the room temperature, these characteristics make it suitable to be used in processes which high temperature is not desirable.

Plasma is a term which refers to the fully ionized gas composed of various substances such photons and free electrons, along with atoms in excited state having a neutral charge. Plasma has a net charge of Plasma is a term which refers to the fully ionized gas composed of various substances such photons and free electrons, along with atoms in excited state having a neutral charge. Plasma has a net charge of due to its equal number of positive and negative ions (Kudra and Majumdar, 2009). Cold plasma is a novel non thermal food processing technology that uses energetic, reactive gases to inactivate contaminating microbes on meats, poultry, fruits, and vegetables. This flexible sanitizing method uses electricity and a carrier gas, such as air, oxygen, nitrogen, or helium; antimicrobial chemical agents are not required. The primary modes of action are due to UV light and reactive chemical products of the cold plasma ionization process. A wide array of cold plasma systems that operate at atmospheric pressures or in low pressure treatment chambers are under development.

### Benefits of cold plasma over other food safety technologies

Cold plasma can be used for decontamination of products where micro-organisms are externally located. Unlike light (e.g. ultraviolet light decontamination), plasma flows around objects which means- shadow effects do not occur ensuring

all parts of a product are treated. For products such as cut vegetables and fresh meat, there is no mild surface decontamination technology available currently; cold plasma could be used for this purpose. Cold plasma could also be used to disinfect surfaces before packaging or included as part of the packaging process. Energy consumption would be similar to existing UV systems and the treatment of foods would be highly cost-effective; the electronics and lifetime of plasma technologies are comparable to UV-C systems even with the additional need for a carrier gas. Atmospheric plasmas containing high levels of bactericidal molecules (>100 ppm ozone, nitric oxides, peroxides, etc.) are generated with minimal power under room temperature conditions in seconds to minutes, with little or no product heating. Atmospheric plasma technology (APT) requires a few hundred watts of power and a supply of compressed air or other gas; sometimes, a gas blend is used depending on the reactive gas species being generated. APT can generate bactericidal molecules very efficiently solely from air with product temperature increases less than 5 °C. This flexibility and unique processing capabilities are driving the technology into new markets and applications. Atmospheric plasma offers a number of advantages over existing food safety technologies, including the following: 1) it is a dry process; 2) it is readily adaptable to a food manufacturing environment; 3) it requires very little energy; 4) reactive gas species revert back to original gas within minutes to hours after treatment and 5) it requires short treatment times.

### Cold plasma as a novel food processing technology

Potential application in food NTP has been applied in the food industry including decontamination of raw agricultural products (Golden Delicious apple, lettuce, almond, mangoes, and melon), egg surface and real food system (cooked meat, cheese,). In one study on *E. coli* 12955 a non-pathogenic surrogate for *Salmonella* spp. inoculated onto almonds, Deng *et al.* reported a reduction of more than 4 log CFU/ml after 30 s treatment at 30 kV and 2000 Hz.

### Cold plasma used to kill bacteria on raw chicken

Pathogens such as *Campylobacter* and *Salmonella* contaminate over 70 percent of the raw chicken meat tested. Recent research from a food safety team at Pennsylvania's Drexel University made use of high-energy, low temperature plasma to eliminate unwanted bacteria while leaving the food basically unchanged.

### Cold plasma in food packaging

Whether labelling jam jars, printing on glass containers, or sealing liquid packaging, a key factor in the packaging industry is the ability to process materials reliably and at low cost. Pre-treatment with atmospheric pressure plasma makes it possible to process different materials and coatings that are sometimes very thin, for example, in the production of composite packaging. Where packs are processed at high speed and an adhesive bond is required, recesses in the area of the bonding surfaces usually have to be taken into account, especially in the case of high-gloss plastic-coated surfaces. By using Open-air plasma technology, such high-gloss gluing points are directly and selectively pre-treated inline so that reliable bonding is ensured. In labelling glass bottles, atmospheric-pressure plasma is employed for pre-treating glass. This allows the use of a universal and low-cost water-based adhesive. One of the most common applications of plasma in packaging is in the area of labelling. For advertising

stickers, information labels or tamper evidence, there's always one key requirement: the glue must be water based and the adhesive joint must not loosen by itself.

### **Irradiation**

It is a method of food preservation from time immemorial such as sun drying, pickling and fermentation. These methods were supplemented with more energy consuming techniques like refrigeration, freezing and canning; however had its merits and demerits. The quest was ever on for newer methods of food preservation with least change in sensory qualities. Preservation of food items is a prerequisite for food security. The seasonal nature of production and the long and unmanageable distances between the production and consumption centres and the rising gap between demand and supply have posed great challenges to conventional techniques of food preservation and thereby to food security. The hot and humid climate of the country is quite favourable to the growth of numerous insects and microorganisms which destroy stored crops and cause spoilage of food. In the changing scenario of world trade, switching over to radiation processing of food assumes great importance. Radiation processing can be used for disinfestations of pests and disease-causing organisms from a range of products including fruits and vegetables. Park *et al.*, (2010) reported lower total aerobic counts in gamma rays treated beef sausage patties as compared to electron beam treated samples. Reduction of 3.78 and 2.04 logs has been reported using electron beam irradiation (2 kGy) for *S. Typhimurium* inoculated in sliced ham (Song *et al.*, 2011) <sup>[64]</sup>. and powdered weaning foods (Hong *et al.*, 2008) <sup>[32]</sup>, respectively whereas (Martins *et al.*, 2004)<sup>[42]</sup>, reported a 4 log reduction in a cocktail of *Salmonella* strains using 1.7 kGy in watercress thereby showing the applicability of gamma radiation in salad vegetables.

### **Radiation as a preservation technique**

Radiation is one of the latest methods in food preservation. Radiation technique makes the food safer to eat by destroying bacteria which is very much similar to the process of pasteurization. In effect, radiation disrupts the biological processes that lead to decay and the ability to sprout. Being a cold process, radiation can be used to pasteurize and sterilize foods without causing changes in freshness and texture of food unlike heat. Further, unlike chemical fumigants, radiation does not leave any harmful toxic residues in food and is more effective and can be used to treat packaged commodities too. Countries including India guard against import of exotic insect pests by requiring a post-harvest disinfestations treatment of commodities that can carry pests. India will not be able to resist the flow of radiated foods to the country as a prohibitive policy would be difficult to justify on technical grounds and could be challenged under the WTO agreement as a technical barrier to trade. Above all, radiation technology for food preservation will be moving fast to the status of a 'wonder technology' to satisfy the sanitary and phyto sanitary requirements of importing countries. The Ministry of Food Processing Industries has a major role to play in ensuring food security by processing and preserving food items to be released in consuming areas and at lean periods. The ministry has a number of schemes to encourage entrepreneurs for setting up facilities for radiation processing of food in public as well as private sectors. The Ministry extends assistance for setting up facilities for food processing by using radiation technology to promote commercial use of

this technology in India. BARC will be the nodal agency for assistance and guidance on process technology while BARC/BRIT will provide information on availability, cost and possible alternatives of essential machinery including pollution control and disposal of radio isotopes. BARC can provide training to the staff on various aspects of operation and safety in India (Saylor, and, Jordan, 2000) <sup>[61]</sup>.

### **Radiation processing**

Food irradiation is a technology that can be safely used to reduce food losses due to deterioration and to control contamination causing illness and death. Food irradiation uses radiant energy electron beams, gamma rays or x-rays to rid food of harmful microorganisms, insects, fungi and other pests, and to retard spoilage. It does not make food radioactive. Irradiation kills pathogens and makes them incapable of reproduction. There are several processes that are collectively referred to as Food Irradiation. The object of each process is to kill or impair the breeding capacity of unwanted living organisms or to affect the product morphology in a beneficial way that will extend shelf-life. Each process has an optimal dose of ionizing energy (radiation) dependent on the desired effect. The dose of radiation is measured in grays (Gy). A gray is a unit of energy equivalent to 1 joule per kilogram. This unit of measure is based on the metric system. Thus, 1 kilogray (kGy) is equal to 1,000 Grays (Gy).

### **Gamma rays**

The rays used in food processing are obtained from large <sup>60</sup>Co radionuclide sources. This type of radiation is essentially mono energetic (<sup>60</sup>Co emits simultaneously two photons per disintegration with energies of 1.17 and 1.33 MeV). Using analytical techniques such as the point kernel or Monte Carlo methods, it is possible to compute the dose distribution in irradiated food products even when very complicated source geometries such as extended plaque sources are used. The resulting depth dose distribution in the food products usually resembles an approximately exponential curve. Irradiation from two sides (two sided irradiation), obtained either by turning the process load or by irradiation from two sides of a plaque source, is often used to increase the dose uniformity in the process load (Olivieira, *et al.*, 2000; Saylor, M.C., Jordan, 2000). Electrons emitted by accelerators have fairly narrow spectral energy limits (usually less than  $\pm 10\%$  of the nominal energy). The energy of the electrons reaching the product is further controlled by the bending magnets of the beam handling system, if applicable. The range of an electron in a medium is finite (unlike that for photons) and is closely related to its energy (Hayashi, 1998) <sup>[29]</sup>.

### **X rays**

Bremsstrahlung irradiator design principles are essentially the same as those for electron irradiators. An extended source of X rays is achieved by distributing the primary electron beam over a target (X ray converter) of sufficient size. In contrast to the radionuclide sources, which emit nearly mono energetic photons, bremsstrahlung (X ray) sources emit photons with a broad energy spectrum (Aikawa, Y, 2000 <sup>[2]</sup>; Cleland, M.R., Pageau, G.M., 1987). The effectiveness of processing of food by ionizing radiation depends on proper delivery of absorbed dose and its reliable measurement. For food destined for international trade, it is of the utmost importance that the dosimeter techniques used for dose determination are carried out accurately and that the process is monitored. Food packed



in crates or boxes is placed on conveyor belts and moved into the heart of the irradiator, where it is exposed to the radiation source. The productions of X-rays are energy intensive and inefficient as the out of total electrons, only about 4-6% is producing X-rays (Brewer, 2004) [10]. Electron beam irradiators can cleanse packaged food at the end of food-processing production lines. High energy waves pass through the food, exciting the electrons in both the food and any pests or pathogens. When the electrons absorb enough energy, they break away from their atoms, leaving positively charged centres behind. Irradiation disrupts the molecular structure; kills or reduces the number of bacteria and yeasts; delays the formation of mold; and sterilizes or kills parasites, insects, eggs and larvae. Levels of absorbed radiation are currently measured in kilo gray (kGy).

### Applications of irradiation

Irradiation has the potential to make meat and meat products safe by killing the various pathogens and parasites. This enhances the keeping quality and storage life of food by better maintaining the nutritive quality. This process is known as "electronic pasteurization" or "cold sterilization" (Crawford and Ruff, 1996) [16]. More than 90% of bacteria can be inactivate with increasing the shelf life of meat by use of lower dose irradiation (Lacroix *et al.*, 2000) [35]. Food preservation by irradiation technique provides consumers with wholesome and nutritious food items having improved hygiene and easy availability and quantity with increased storage life, convenience to transport. Applications at low dose levels (10 Gy 1 kGy) Sprouting of potatoes, onions, garlic, shallots, yams, etc. can be inhibited by irradiation in the dose range 20 150 Gy. Radiation affects the biological properties of such products in such a way that sprouting is appreciably inhibited or completely prevented. Physiological processes such as ripening of fruits can be delayed in the dose range 0.11 kGy. These processes are a consequence of enzymatic changes in the plant tissues. Insect disinfestations by radiation in the dose range 0.2 1 kGy is aimed at preventing losses caused by insect pests in stored grains, pulses, cereals, flour, coffee beans, spices, dried fruits, dried nuts, dried fishery products and other dried food products. A minimum absorbed dose of about 150 Gy can ensure quarantine security against various species of tephretid fruit flies in fresh fruits and vegetables, and a minimum dose of 300 Gy could prevent insects of other species from establishing in non-infested areas. In most cases irradiation either kills or inhibits further development of different life-cycle stages of insect pests. The inactivation of some pathogenic parasites of public health significance such as tapeworm and trichina in meat can be achieved at doses in the range 0.3 1 kGy. (i) Inhibition of sprouting 0.05 - 0.15 Potatoes, onions, garlic, root ginger, yam etc. (ii) Insect disinfestations and parasite disinfection 0.15 - 0.5 Cereals and pulses, fresh and dried fruits, dried fish and meat, fresh pork, etc. (iii) Delay of physiological processes (e.g. ripening) 0.25 - 1.0 fresh fruits and vegetables.

Osterholm and Norgan (2004) described three levels of irradiation viz. Low, Medium (pasteurization) and High. Low level dose up to 1 kGy kills insects and parasites (especially *Trichinella spiralis*) and inhibits the sprouting of potatoes. Medium level radiation dose range is 1-10 kGy inactivates the spoilage microorganism and food borne pathogens and extend the shelf life of meat and other food products. Sterilization of meat and meat products is treated with dose rate of more than 10 kGy dose of irradiation in high level. Applications at

medium dose levels (110 kGy) Radiation enhances the keeping quality of certain foods through a substantial reduction in the number of spoilage causing micro-organisms. Fresh meat and seafood, as well as vegetables and fruits, may be exposed to such treatments with doses ranging from about 1 to 10 kGy, depending on the product. This process of extending the shelf life is sometimes called randomization. Pasteurization of solid foods such as meat, poultry and sea foods by irradiation is a practical method for elimination of pathogenic organisms and micro-organisms except for viruses. It is achieved by the reduction of the number of specific viable non spore forming pathogenic micro-organisms such that none is detectable in the treated product by any standard method, for which doses range between 2 and 8 kGy. The product will usually continue to be refrigerated after the radiation treatment. This process of improving the hygienic quality of food by inactivation of food-borne pathogenic bacteria and parasites is sometimes called rededication. This medium dose application is very similar to heat pasteurization, and is hence also called radio pasteurization. (i) Extension of shelf-life 1.0 - 3.0 kGy fresh fish, strawberries, mushrooms etc. (ii) Elimination of spoilage and pathogenic microorganisms 1.0 - 7.0 kGy Fresh and frozen seafood, raw or frozen poultry and meat, etc. (iii) Improving technological properties of food 2.0 - 7.0 kGy Grapes (increasing juice yield), dehydrated vegetables (reduced cooking time), etc. Applications at high dose levels (10 100 kGy) Irradiation at doses of 10 30 kGy is an effective alternative to the chemical fumigant ethylene oxide for microbial decontamination of dried spices, herbs and other dried vegetable seasonings. This is achieved by reducing the total microbial load present in such products including pathogenic organisms.

### Supercritical carbon dioxide

A proper diet implies the daily consumption of fruits and vegetables (F&V); the quantity depends on several factors and can be estimated, knowing the nutrient composition of the food, using the dietary reference intakes (DRIs) tables or similar methods. Traditionally, F&V were consumed as fresh, after simple homemade operations: elimination of roots, seeds and other uneatable parts, washing, peeling, cutting, and others. However, in developed countries, the use of ready-to-use F&V products became very popular as consequence of lifestyle: the ready-to-eat minimally processed F&V (MPF)/fresh like F&V, are prepared at industrial scale, packed, stored at about 4 °C and distributed. It is worth to underline that the shelf life of these MPF is significantly higher than that of the corresponding fresh product after homemade preparation for eating. An alternative way to supply the body with the necessary nutrients is to drink the juice obtained from the F&V. In the most general sense, juice is defined as the extractable fluid contents of cells or tissue, while defines juice as "unfermented but fermentable, intended for direct consumption, obtained by mechanical process from sound, ripe plant material, preserved exclusively by physical means." Traditionally industrially made F&V juices are strongly concentrated and pasteurized in order to insure safety and to reduce handling and transportation costs. High pressure carbon dioxide (HPCD) is another upcoming treatment that is being extensively used as a non-thermal technique for food pasteurization. The process is not only environmentally friendly due to the non-toxic nature of carbon dioxide but also involves application of lower CO<sub>2</sub> pressure as compared to those employed for HPP. The use of lower pressures makes

this technique an energy-saving process. In addition, generation of carbonic acid ( $H_2CO_3$ ) in the water present in food products further results in a reduction in the pH of the food products enhancing the penetration of  $CO_2$  (Wei *et al.*, 1991). Studies involving the use of HPCD for the inactivation of *S. Typhimurium* (Kim *et al.*, 2007; Erkmén and Karaman, 2001; Erkmén 2000; Wei *et al.*, 1991) [69]. have clearly reported the microbial strain, pressure applied, pH of the medium, type of medium and temperature to be important factors for the inactivation.

Subcritical (gaseous or liquid)  $CO_2$  on the other hand, is  $CO_2$  at a temperature or pressure below its thermo dynamical critical point values. In the supercritical state,  $CO_2$  has low viscosity ( $3-7 \times 10^{-5}$  Ns/m<sup>2</sup>) and zero surface tension (McHugh and Krukónis, 1993) [44]. However, organoleptic, nutritional and functional properties of the F&V reconstituted are very far from that of the corresponding fresh squeezed. On the other hand, F&V juices are very perishable with a shelf life under refrigerated conditions of 2 to 4 days. Very recently the F&V market registered an unpredictable interest for “cold pressed” juices which are a kind of minimally processed beverages (MPB). Actually this product, characterized by a shelf life of about 3 or even 4 weeks under refrigerated conditions, is a single strength F&V juice pasteurized by a non-thermal technology which implies the use of very high hydrostatic pressure in the range from 400 to 1000 MPa (HHP). In the technical and scientific literature there are a lot of information regarding the application of HHP to a variety of food and beverages with special emphasis on the preservation of thermo-labile useful compounds. Anyway, although the results are often conflicting, HHP is usually considered better than thermal pasteurization and consequently became very popular in the food industry. However, it can only be applied in the batch mode using very costly apparatus while, for liquid foods and beverages, one is naturally led to imagine a continuous process, possibly under operating conditions less drastic. Generally, an increase in the pressure results in a proportionate increase in the microbial inactivation. As a consequence, at higher pressures, a shorter exposure is needed to inactivate the same level of microbial cells (Hong *et al.*, 1997; Hong and Pyun, 1999). Therefore, a number of non-thermal pasteurization technologies for the food and beverages industry were proposed and are currently under investigation/development. Among these, the one that uses supercritical carbon dioxide (HPCD) in a range of pressure between 10 to 40 MPa appears very interesting for continuous non thermal pasteurization of single strength F&V juices; aim to obtain a significant extended shelf life (ESL), while preserving most of the nutritional, sensorial, and functional properties of the raw extracts. The purpose of this research is to contribute to show the technical feasibility of HPCD for the industrial production of single strength, minimally processed, carrot juice (CJ) to be commercialized as safe and high quality fresh-like products. The first reason for focalize on CJ is the consequence of the “familiarity” of the authors with carrots and derived products with the obvious consequence that a very rich baggage of knowledge concerning cultivation, harvesting, composition, up-stream of the raw material, products derivable from it, technologies, nutritional and functional properties, and other information may be used for the purposes of this work. Moreover, carrot is one of the top ten vegetable cultivated in huge amount almost all around the world and is rich in bioactive compounds like carotenoids with appreciable level of several other functional component having significant health-promoting properties.

## Advantages and limitation of NTP

Techniques	Advantages	Limitations
Irradiation	(1) Effective for several foods (2) Many different sources available (Gamma rays, electron beam)	Limited public acceptance. The use of radiation dose up to 7 kilo Gray (KGY) has been sanctioned.
UV Radiation	No chemical are used Non heat related method	Long term exposure can be harmful to the industry workers.
HPP	Independent of the shape of food can be use for both solid and liquid sample.	Changes in quality of food have been observed.
PEF	Pulse applied for a short period so no generation of heat and less use of energy	Cannot be applied to foods which is not withstand high fields.
Supercritical carbon dioxide	It can be used in a batch or continuous process	It's not more successful for solids foods.

## Conclusion

The application of Non-thermal technologies holds potential for producing high quality and safe food products. It is likely that some of these technologies, according to their specificities, will find niche applications in the food industry (some of them already did), replacing or complementing conventional preservation technologies, through synergistic interactions (e.g. hurdle concept). Cold Plasma is now being investigated for application to foods as a sanitizing and/or conditioning step. The atmospheric cold plasma, by far, is one of the newest technologies used in food industry for microbial inactivation. Future studies should be directed towards assessment of the efficacy of cold plasma on the processing of different food products. Fresh fruits and vegetable are highly susceptible to microbial spoilage. This can be avoided with the application of surface treatments. The treatment of their surface has to be as gentle as possible for keeping the integrity and the freshness of fruits and vegetables. Minimal processing techniques such as ultraviolet (UV) light treatment meet these requirements. The use of UV-C light treatment proved to be effective at reducing microbial loads of pathogens on fresh fruits and vegetables.

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